

Appendices

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APPENDIX A. MORRO BAY NATIONAL ESTUARY PROGRAM
BACTERIA MONITORING PROTOCOLS

Morro Bay Volunteer Monitoring Program Bacteria Monitoring Protocols

Total Coliform and *E. coli* Monitoring Protocol

Background:

- Pathogens, consisting of viruses, bacteria and protozoans, can cause disease.
- Sources include fecal contamination from sewage, leaking septic systems, urban runoff, boat and marina waste, and waste from pets, farm animals and wildlife.
- Direct testing is very expensive and impractical since pathogens are rarely found in water bodies. Also, there are no existing regulatory standards for all of the individual pathogens.
- Instead, monitor for pathogens by testing for “indicator” species. Their presence indicates that fecal contamination may have occurred.
- Four common indicators: total coliforms, fecal coliforms, *E. coli*, and enterococci. These bacteria are prevalent in the intestines and feces of warm-blooded animals. The indicators themselves are usually not pathogenic.

Types of Indicators:

- Coliform bacteria live in the lower intestines of warm-blooded animals and may constitute as much as 50% of fecal waste. Not pathogenic themselves typically, but their presence indicates sewage contamination, perhaps accompanied by disease-causing pathogens.
- Total Coliforms are a group of closely related bacterial genera that all share the ability to ferment lactose. Total coliform standards exist for recreational or shellfish waters, although some species are found naturally in plant material or soil. More useful for testing drinking water where plant or soil material would be of concern.
- Fecal coliforms: Subgroup of total coliform. Widely used to test recreational waters and shellfish waters. Generally considered the best available indicators of contamination at this time. The current regulations are written for this indicator.
- *Escherichia coli*: Occurs only in the feces of warm-blooded mammals, making it a good fecal-specific indicator. Can provide better correlation with swimming-associated gastrointestinal illness than fecal coliforms. However not widely used, despite EPA recommendations, due to continuity with previous data and cheaper analytical costs. This indicator is valid in marine and freshwater, although there is no regulatory standard for comparison in marine waters. Our program continues to monitor for *E. coli* in marine waters because it is more closely comparable to fecal coliform data than is Enterococcus data. Since shellfish regulations and the current Basin Plan standard are for fecal coliform, this *E. coli* monitoring will continue until the regulatory standards are changed.
- Enterococci: Group of bacteria found primarily in the intestinal tract of warm-blooded animals. Unrelated to coliforms, they are a subgroup of the fecal streptococci group. But expensive to monitor, despite EPA recommendations that it is a better indicator of harmful recreational contact in salt water environments. This indicator is valid in marine and freshwater, although there is no regulatory standard for comparison at this time. Our program monitors for Enterococcus in marine samples and at a tidally-influenced creek.

Background on Colilert-18 Tests:

- These tests are for freshwater or marine waters.
- Based on the Most Probable Number (MPN) technique, which uses multiple tubes with broth that are inoculated with the sample at different dilutions. After incubation, the tubes show a positive or negative reaction for the target organism. The number of positive tubes corresponds to a statistical probability that the sample contained a certain number of bacteria.
- Kits are approved by EPA for monitoring drinking water.
- Based on a color change in the wells after incubation, total coliform and *E. coli* can be detected in freshwater samples. Only *E. coli* can be detected in marine samples.

Cost: Approximately \$10/test.

Procedure for Sample Collection

Equipment:

- 120-mL sealed sterile sample jars containing sodium thiosulfate (2 per site for creeks, 1 per site for bay))
- 250-mL sterile Nalgene sampling jars (1 per site for creeks)
- Disposable vinyl gloves
- Data collection sheet
- Pen
- Clipboard
- Sharpie pen for labeling sample containers
- Ice chest
- Ice packs
- Tide book

Bay Sites (land-based):

- Cuesta Inlet (CIN)
- Baywood Park (BAY)
- Pasadena Point (PAS)
- Windy Cove (WIN)
- Public Boat Ramp at Tidelands Park (TID)
- State Park Marina (SPM)
- Coleman Park (COL)
- Sharks Inlet (SIN)
- Los Osos Creek (GS1)
- [NOTE: Although GS1 is a site on Los Osos Creek, due to the amount of tidal influence, it is treated as a bay site for monitoring purposes.]

Creek Sites:

- Chorro Creek above the Ecological Reserve (UCR)
- Chorro Creek at Chorro Creek Road (CCC)
- Chorro Creek at South Bay Boulevard Bridge (TWB)
- Pennington Creek on Cal Poly property (CPN)
- Pennington Creek on Rancho El Chorro property (PEN)
- Dairy Creek, middle (DAM)

- San Luisito Creek, lower (SLU)
- San Bernardo Creek (SBC)

Sample Collection in Creeks:

- Fill out data collection form with date, sampler names, weather in past 24 hours, current weather.
- Record the three letter site code and the time the sample was taken. In the notes section, note things such as presence of animals and birds, abundant leaf matter, any strange debris, any unusual smells, etc.
- Label two sterile 120 mL sample bottles with site ID.
- Wear gloves to protect both you and the sample.
- Site should be free of algal blooms, surface debris, oil slicks and congregations of water fowl. Avoid agitating the bottom sediment. Avoid splashing water. Choose site with good mixing and no stagnant pools.
- If entering the water to take the sample, be careful to not stir up sediment. Face upstream to take the sample, so that you do not affect the sample.
- Submerge sterile 250 mL bottle underwater and remove cap from sample bottle, being careful not to touch the rim or inside the lid. Try to collect the sample 12” underwater or at 1/3 of the depth below the surface.
- Fill the sample bottle with water, with the bottle facing into the current.
- Fill the bottle to the shoulder, leaving headspace in the container. Cap bottle.
- Shake bottle 25 times in one-foot arc.
- Pour 100 mL into each of the two labeled 120 mL bottles.
- Store in the ice chest.
- Are your next site, **do not reuse the large bottle**. You must use a new sterile 250-mL bottle at each site.
- At the lab, process your samples as usual, **using one jar for the undiluted sample and one jar for the 1:10 sample at each site**.
- Rinse out the large 250-mL bottles in the sink and leave them in the Dirty Glassware bin to be reautoclaved along with the rest of your glassware.
- Change gloves at each site.
- Ideally, samples will arrive at the lab within six hours of collection and are tested within the next two hours. They must be maintained at 4 degrees C or lower. If they can't be tested within eight hours, they must be tested within 24 hours of collection.

Sample Collection in Bay and Los Osos Creek (GS1):

- Fill out data collection form with date, sampler names, weather in past 24 hours, current weather, time of high tide and height of high tide. There is a tide book in the equipment room. For GS1, collect near a low tide (2 feet or below).
- Record the three letter site code and the time the sample was taken. In the notes section, note things such as presence of animals and birds, abundant leaf matter, any strange debris, any unusual smells, etc.
- Use sterile sample bottles, labeled with site ID.
- Wear gloves to protect both you and the sample.
- Site should be free of algal blooms, surface debris, oil slicks and congregations of water fowl. Avoid agitating the bottom sediment. Avoid splashing water. Choose site with good mixing and no stagnant pools.
- If entering the water to take the sample, be careful to not stir up sediment. Face upstream to take the sample, so that you do not affect the sample.

- Submerge bottle underwater and remove cap from sample bottle, being careful not to touch the rim or inside the lid. Try to collect the sample 12” underwater or at 1/3 of the depth below the surface.
- Fill the sample bottle with water, with the bottle facing into the current.
- Fill the bottle to the shoulder, leaving headspace in the container. Cap bottle.
- Store in the ice chest.
- Change gloves at each site.
- If sampling on a beach, collect it on an incoming wave.
- For bay sites, sample on a high tide to avoid getting stuck in the mud.
- Ideally, samples will arrive at the lab within six hours of collection and are tested within the next two hours. They must be maintained at 4 degrees C or lower. If they can’t be tested within eight hours of collection, they must be tested within 24 hours of collection.

Procedure for Sample Analysis at the Morro Bay-Cayucos Wastewater Treatment Plant

Directions to the lab:

From Hwy 1, exit at the Hwy 41 exit and head west, toward Morro Bay High School. Pass the high school on your right. Pass the first gate on your left (the city maintenance yard). The second gate on the left is unlabeled – this is the WWTP. Turn left into the gate and take the first right. There is a small parking area on the right and the building to your right contains the WWTP operations center and lab. If you have more than one car, please park outside the facility and walk in.

Lab hours are as follows:

Monday – Thursday: 8 a.m. to 3:30 p.m.

But you must be out of the lab a half an hour before closing time.

Equipment:

- Sterile distilled water
- Sterile pipette tips
- Pipetter
- Disposable gloves
- Safety glasses
- Colilert-18 reagent packet
- Quanti-Tray/2000 foil tray
- Incubator
- Quanti-Tray sealer and rubber insert

1. Upon arrival at the lab, notify plant staff that you are at the facility and are going to be doing lab analysis for the VMP. It is crucial to let someone know you are there, in case there is a plant emergency. Plant personnel will be notified in advance of the date and approximate time of VMP use of the lab.
2. Wash your hands, using the anti-bacterial soap.
3. Put on disposable gloves and safety glasses.
4. Turn on the Quanti-Tray sealer (power switch on back, right hand side) so it has time to pre-heat. The amber light goes on. The sealer is ready when the green light also goes on. **Make sure there is nothing behind the sealer that would block trays from**

- exiting after they are sealed.**
5. Collect your sterile bottles.
 6. For bay water quality samples:
 - a. First you must dilute the sample 1:10 due to interference from marine samples.
 - b. Shake bottle 25 times in a one-foot arc.
 - c. Put sterile tip onto pipetter. Dial pipetter to read 10 mL. Use a pipetter to remove 10 mL from the sample container. Be careful to not tip the pipette once you have filled it – you must keep the pipette upright so that the sample does not contaminate the paper plug in the pipetter.
 - d. Add sample to a sterile plastic bottle.
 - e. Add Colilert-18 reagent.
 - f. Add 90 mL of sterile distilled water from squeeze bottle labeled ‘sterile distilled water’ into graduated cylinder. Add to the 10 mL of sample in the plastic bottle.
 - g. Swirl to dissolve powder. (Avoid shaking vigorously, which adds bubbles and foam to the sample.)
 - h. If sample is foamy, let sit for a few minutes to let foam settle.
 - i. Sample is ready to add to Quanti-Tray.
 7. For undiluted freshwater samples:
 - a. Shake bottle 25 times in one-foot arc to mix. Open sample container. Pour out or pipette out excess sample so that have a 100 mL sample remaining. Suggest pouring into sterile jar in case you accidentally pour out too much.
 - b. Add Colilert-18 reagent.
 - c. Re-cap container.
 - d. Swirl to dissolve reagent.
 - e. If sample is foamy, let sit for a few minutes to let foam settle.
 - f. Sample is ready to add to Quanti-Tray.
 8. If a 1:10 dilution is required for freshwater samples, see Step 6.
 9. If a 1:100 dilution is required:
 - a. Invert sample bottle 25 times to mix. Open sample container.
 - b. Insert clean, sterile tip into pipetter. Dial pipetter to read ‘1 mL.’
 - c. Place 1 mL of sample in sterile plastic bottle.
 - d. Add 99 mL of sterile distilled water from squeeze bottle into plastic bottle using the graduated cylinder.
 - e. Add Colilert-18 reagent and swirl to dissolve.
 - f. If sample is foamy, let sit for a few minutes to let foam settle.
 - g. Sample is ready to add to Quanti-Tray.
 10. Must use new sterile equipment (flasks, pipettes, plastic containers, etc.) for each site and each dilution.
 11. If a blue flash occurs upon addition of Colilert-18 reagent, the sample is considered invalid due to excessive chlorine and testing should be discontinued.
 12. To put sample in Quanti-Tray:
 - a. Look over the tray and make sure it has no tears or wrinkles in the seals.
 - b. First label the back of the Quanti-Tray with a Sharpie marker. Mark the site code, date, time of testing and dilution. **Label the back of the tray as ‘E. coli’ if you are also running Enterococcus samples to avoid confusion.**
 - c. Use one hand to hold the Quanti-Tray upright with the well side facing the palm.
 - d. Squeeze the upper part of the Quanti-Tray so that the tray bends toward the palm.
 - e. Gently pull foil tab to separate foil from the tray. **Avoid touching the inside of the foil or tray.**

- f. Pour the sample directly into the tray, avoiding contact with the foil. Tap the small wells 2-3 times to release any air bubbles. Allow foam to settle.
13. To seal the Tray:
 - a. Wait until both the amber and green lights are lit, indicating sealer is pre-heated.
 - b. Check behind the sealer to ensure that nothing is blocking the slot where trays exit the sealer.
 - c. Place the empty sealer rubber insert on the input shelf with the large cutout facing away from the sealer.
 - d. Place the tray filled with sample onto the rubber insert, making sure that the tray is properly seated on the rubber insert, with each well of the tray in its corresponding hole in the rubber insert.
 - e. Slide the insert into the sealer until the motor grabs it and draws it into the sealer.
 - f. In approximately 5 seconds, the tray will be sealed and partially ejected from the rear of the sealer. Remove the rubber insert and tray from the rear of the sealer.
 - g. If you wish to reverse the motor drawing the rubber tray into the sealer (if a misaligned tray is accidentally fed into the sealer), press and hold the reverse button on the front of the sealer. However, do NOT reverse the motor once the rubber insert has been drawn fully into the sealer.
14. When done sealing all trays, turn the sealer off.
15. As you finish sealing each tray, stack in the incubator.
16. The incubator does not need to be pre-heated before the samples are incubated. The incubator temperature should be between 34.5 and 37 degrees C during the incubation period.
17. On the data sheet, note the time the samples were put into the incubator.
18. Triple rinse all autoclavable bottles with hot tap water. Do not use soap. Once washed, place them in the bin that is labeled 'DIRTY'. Put caps on the bottles but don't tighten.
19. Place the pipette tips in the autoclavable tray and place in bin.
20. Refill empty squeeze bottles with distilled water from our large carboy sitting on the incubator. Put the tops on loosely, and do not put foil over the tip. Put those in the bin to be autoclaved. Be sure to refill them so there's enough sterile distilled water for the next volunteer! the graduated cylinder in the 'DIRTY' bin. Be sure to cover the top with foil, located in the drawer.
21. Lab personnel will sterilize the equipment before your next visit to the lab and place them under the counter.
22. Discard the sample collection bottles in the recycle bin and the caps in the garbage. These are not re-usable.
23. Wipe down the bench top.
24. Dispose of gloves in the trash. Wash hands with anti-bacterial soap.
25. Let plant personnel know that you are leaving.

QA Procedure for Creek Sites

Occasionally we will request that you collect a split sample for QA purposes. Both samples will be analyzed by the VMP for comparison to precision criteria.

To test precision, we will collect a single sample from the site that is *later* split between two separate bottles, rather than *initially* collecting two separate samples in separate bottles.

Two separate samples, despite being collected simultaneously, can have very different concentrations of bacteria because bacteria are not dispersed equally throughout the water column. By collecting one sample, mixing it, then splitting that sample into two bottles, we hope to increase the likelihood that both the samples will meet our precision criteria.

- We have designated autoclavable 250 ml bottles to use for collecting samples that will be split for QA purposes.
- For each site that you need to collect a QA sample from, bring a QA bottle along with three IDEXX bottles for creek sites (two for your samples, one for the QA sample).
- Collect a sample from your site using one of the QA bottles in the same manner with which you collected your samples using the IDEXX bottles.
 1. Wear gloves to prevent contamination from your hands
 2. Submerge bottle and remove cap, do not touch the rim or inside the lid
 3. Fill the sample bottle with water, with the bottle facing into the current
 4. Leave some headspace and cap bottle.
- Split the sample. Wearing clean gloves, open the IDEXX bottles, and then shake the QA bottle 25 times in a one-foot arc to mix the sample well.
- Label the three IDEXX bottles with the site code.
- If you run diluted samples you can pour 100 ml of the sample into each of the IDEXX bottles.

Precision Test

- If you run an undiluted sample along with a diluted sample, fill one IDEXX bottles to the 100 ml mark for your undiluted sample, fill another bottle to the 100 ml line for the QA replicate sample, and then pour the remaining sample into a third bottle for the diluted sample. This should allow 100 ml for your undiluted sample, 100 ml for the replicate sample, and nearly 100 ml for the diluted sample.
- The bottle that goes to QA should not always be the first, or always the second, bottle that the sample is poured into. So, vary which bottle you fill first.
- To proceed with the lab test, begin by shaking the IDEXX bottle with your sample in it to dissolve all the granules that neutralize any chlorine that may be present in the sample, and to ensure a well mixed sample. Then, run the test as usual.
- Run two of the bottles as undiluted samples.
- Run one of the bottles as a 1:10 dilution sample.
- The results from the two undiluted sample will be compared to determine if they meet our precision criteria.
- **Accuracy Test for E. coli**

1. Staff will provide a 100 mL water sample of known E. coli concentration.
2. Add a packet of Colilert-18 to the 100 mL sample.
3. Swirl to dissolve.
4. Seal in tray and incubate.

Sterility Test

- Run blank samples for Total coliform and E. coli:
 1. Use a clean graduated cylinder to measure 100 mL of sterile distilled into a mixing bottle.
 2. Add a packet of Colilert-18 to the water.
 3. Swirl to dissolve.
 4. Seal in tray and incubate.

QA Procedure for Bay Sites

Occasionally we will request that you collect a split sample for QA purposes. Both samples will be analyzed by the VMP for comparison to precision criteria.

To test precision, we will collect a single sample from the site that is *later* split between two separate bottles, rather than *initially* collecting two separate samples in separate bottles.

Two separate samples, despite being collected simultaneously, can have very different concentrations of bacteria because bacteria are not dispersed equally throughout the water column. By collecting one sample, mixing it, then splitting that sample into two bottles, we hope to increase the likelihood that both the samples will meet our precision criteria.

- We have designated autoclavable 250 ml bottles to use for collecting samples that will be split for QA purposes.
- For each site that you need to collect a QA sample from, bring a QA bottle along with two of the IDEXX bottles (one for your sample, one for the QA sample) for bay sites.
- Collect a sample from your site using one of the QA bottles in the same manner with which you collected your samples using the IDEXX bottles.
 1. Wear gloves to prevent contamination from your hands
 2. Submerge bottle and remove cap, do not touch the rim or inside the lid
 3. Fill the sample bottle with water, with the bottle facing into the current
 4. Leave some headspace and cap bottle.
- Split the sample. Wearing clean gloves, open the IDEXX bottles, and then shake the QA bottle 25 times in a one-foot arc to mix the sample well.
- Label the two IDEXX bottles with the site code.

- Pour 100 ml of the sample into each of the IDEXX bottles.
- To proceed with the lab test, begin by shaking the IDEXX bottle 25 times in a one-foot arc to ensure a well-mixed sample.
- Use one of the bottles to run two Enterococcus trays, with 1:10 dilution.
- Use one of the bottles to run two E. coli trays, with 1:10 dilution.
- The results from the two samples will be compared to determine if they meet our precision criteria.

Accuracy Test for E. coli

1. Staff will provide a sample of known E. coli concentration.
2. Add a packet of Colilert-18 to the 100 mL sample.
3. Swirl to dissolve.
4. Seal in tray and incubate.

Accuracy Test for Enterococcus

1. Staff will provide a 100 mL water sample of known E. coli concentration.
2. Add a packet of Enterolert to the 100 mL sample.
3. Swirl to dissolve.
4. Seal in tray and incubate.

Sterility Test

- Run blank samples for *E. coli* and for Enterococcus:
 1. Use a clean graduated cylinder to measure 100 mL of sterile distilled water into a mixing bottle.
 2. Add a packet of Enterolert to the water.
 3. Swirl to dissolve.
 4. Seal in tray and incubate.
 5. Repeat procedure using Colilert-18 to create a total coliform and *E. coli* blank.

Reading the Sample Trays

The trays must incubate between 18 and 22 hours at 34.5 to 37 degrees C before reading. **If you read the trays later than the 22-hour incubation time period, the results are not valid.**

There are a total of 49 large wells and 48 small wells per tray. The large, rectangular well at the top of the tray is counted as a large well.

Fill out the bottom section of the data collection form with your name, the incubator temperature and the date and time the results were read.

For total coliform results (for freshwater only):

1. Pull out the color comparator from the drawer.
2. Face the doorway to the lab, with your back to the room, and hold the tray and comparator out at arms-length and below eye level. This seems to facilitate detection of the color change.
3. Count the number of large wells (including the large rectangular well at the top of the tray) and small wells that are **more** yellow than the comparator tray. These wells are positive for total coliforms. All wells that are less yellow than the comparator are negative for total coliforms.
4. Record these values on the data sheet.
5. Use the MPN chart and based on the number of large and small wells that changed color, record the MPN value on the data sheet.
6. If your sample was diluted, then you must multiply the MPN value by the dilution factor (i.e., multiply by a factor of 10 if diluted original sample 1:10) to get the MPN value for the original, undiluted sample.
7. Return the color comparator to its red bag in the drawer. Do NOT autoclave it!

If a bacterium other than coliform bacteria were present in the sample and caused a color change other than yellow or fluorescing, do not record these as positive wells.

This gives an MPN value for total coliform. Note that the test kits can't be used to monitor for total coliforms in marine samples.

For *E. coli* results (both freshwater and marine samples):

1. Put the tray into the UV cabinet and plug in the UV light. Push the white switch and look through the viewing screen on the cabinet.
2. Count the number of large and small wells that are both fluorescing AND yellow. These are positive wells for *E. coli*. Record on the data sheet. All wells that fluoresce more than the comparator **and** are yellow are positive for *E. coli*.
3. If it is difficult to distinguish whether the well is both yellow and fluorescing when it is in the cabinet, use a Sharpie to make each of the fluorescing wells while it is in the cabinet. Then pull the tray out of the cabinet and verify that all of those fluorescing wells also have the yellow color change. Only the wells with BOTH fluorescence and a yellow color change are positive for *E. coli*.
4. Look up the MPN value from the chart and record on the data sheet.
5. If your sample was diluted (all marine samples are diluted to at least 1:10), then you must multiply the MPN value by the dilution factor (i.e., multiply by a factor of 10 if diluted original sample 1:10) to get the MPN value for the original, undiluted sample.
6. Be sure to turn off the UV light when you are done.

If a well has fluorescence of any color (i.e., a bright blue) **and** the well has a yellow color change under ambient light, then it is positive for *E. coli*. If it is fluorescing in any color and has **not** undergone a yellow color change, then it is not positive for *E. coli*.

This gives the MPN value for *E. coli*.

Once the readings are complete:

1. The trays must be autoclaved before they can be thrown out. Mark the back 'Destroy' and double bag it in **two** autoclave bags and place in the 'To be autoclaved' bin. Bags are located in the drawers under our lab space. Lab personnel will autoclave and then dispose of the tray. Do not fold the bags closed since steam must be able to escape during autoclaving.
2. Don't turn off the incubator since someone else may be coming in to use it.

Positives observed before 18 hours are valid, and negatives after 22 hours are valid.

Suggested Dilutions for Bacteria Testing:

Creek Sites: Due to variance in *E. coli* and total coliform levels, undiluted and 1:10 dilutions are run at each creek site.

Bay Sites: All sites are tested with a 1:10 dilution.

Enterococcus Monitoring Protocol

Background:

- Enterococci: Group of bacteria found primarily in the intestinal tract of warm-blooded animals. Unrelated to coliforms, they are a subgroup of the fecal streptococci group. Enterococcus is less sensitive to salt than *E. coli* and is thus considered a better indicator in marine environments.

Background on Enterolert Tests:

- These tests are valid for freshwater or marine waters. We will continue to monitor total coliforms and *E. coli* in freshwater. We will monitor *E. coli* as well as enterococcus in marine waters. The reason for continuing to monitor *E. coli* in the bay is that regulatory agencies use the levels of the coliform family of bacteria (which includes *E. coli*) to determine if the shellfish that grow in the water are safe for consumption. Since we are collecting data that may be used to assess the safety of bay waters for shellfish growing, we need to continue collecting *E. coli* data.
- Based on the Most Probable Number (MPN) technique, which uses multiple tubes with broth that are inoculated with the sample at different dilutions. After incubation, the tubes show a positive or negative reaction for the target organism. The number of positive tubes corresponds to a statistical probability that the sample contained a certain number of bacteria.
- Based on a color change in the wells after incubation, enterococcus can be detected in samples.
- Cost: Approximately \$10/test.

Notes on Sample Collection

Sample collection procedure is the same as for Colilert-18 testing. Since only 10 mL of sample are needed for each test, you only need to collect one sample jar at each site in order to conduct both enterococcus and *E. coli* testing.

We will conduct enterococcus monitoring at the following sites: Bay

Sites:

- Cuesta Inlet (CIN)
- Baywood Park (BAY)
- Pasadena Point (PAS)
- Sharks Inlet (SIN)
- Windy Cove (WIN)
- Public Boat Ramp at Tidelands Park (TID)
- State Park Marina (SPM)
- Coleman Park (COL)
- Los Osos Creek near Turri Road, downstream of SYB (GS1)
- [NOTE: Although GS1 is a site on Los Osos Creek, due to the amount of tidal influence, this site is treated as a bay site for monitoring purposes.]

Notes on Sample Analysis

Enterolert samples are processed identically to Colilert-18 samples (same process, same Quanti-trays, etc.). The only differences are as follows:

- A different packet of reagent is used. The Colilert-18 reagent packets are labeled in red, and the Enterolert reagent packets are labeled in purple.
- The samples incubate at 41 +/- 0.5 degrees C (versus 34.5 to 37 degrees C for Colilert-18)
- The incubation time is 24 to 28 hours (versus 18 to 22 hours for Colilert-18)

Some information for the lab analysis:

- All samples must be diluted 1:10.
- **Label the back of each tray with either “*E. coli*” or “Entero” to help keep them straight.** Once the reagent is added to the water sample, it is difficult to distinguish whether you have a Colilert-18 or an Enterolert sample. The Enterolert samples appear to be a slightly darker shade of yellow than the Colilert-18 samples.
- Be careful to place your enterococcus samples in the incubator labeled ‘Enterolert’. The incubator for the *E. coli* samples is labeled ‘Colilert-18.’ The enterococcus incubator is set for 41 +/- 0.5 degrees C, and the *E. coli* incubator is set for 34.5 to 37 degrees C.

Reading the Enterolert Trays

The trays must incubate between **24 and 28 hours** at 41 +/- 0.5 degrees C before reading. **If you read the trays later than the 28 hour incubation time period, the results are not valid.**

Fill out the bottom section of the data collection form with your name, the incubator temperature and the date and time the results were read.

For enterococcus results:

1. Put the tray into the UV cabinet and plug in the UV light. Push the white switch and look through the viewing screen on the cabinet.
2. Count the number of large and small wells that are fluorescing blue. These wells are positive for enterococcus. Any amount of fluorescing is considered to be a positive result. Record on the data sheet. If the wells fluoresce a color other than blue, do not record these as positive wells.
3. Look up the MPN value from the chart (the same chart as for the Colilert-18 results) and record on the data sheet.
4. Multiply the result by the dilution factor. All samples are diluted at least 1:10.
5. Be sure to turn off and unplug the UV light when you are done.
6. Once the readings are complete, treat them the same way as the Colilert-18 trays. Double bag in autoclave bags and put in the 'To be autoclaved' bin.

Morro Bay National Estuary Program Bacteria Monitoring

Date of Sample Collection:
Name of Sample Collector:
Name of Sample Analyst:
Rainfall in past week:

High Tide Height:
High Tide Time:
Weather, past 24 hours:
Weather currently:

Site	Collection Time	Analyte	Dilution Factor	# Large Wells	# Small Wells	MPN/100 mL	Notes
		Total coliforms					
		<i>E.coli</i>					
		Enterococcus					
		Total coliforms					
		<i>E.coli</i>					
		Enterococcus					
		Total coliforms					
		<i>E.coli</i>					
		Enterococcus					
		Total coliforms					
		<i>E.coli</i>					
		Enterococcus					

Date of analysis:	ColiIert-18	Enterolert
Time incubation started:		
Incubator temp when started:		
Name of sample reader:		
Date samples read:		
Time samples read:		
Incubator temp when read:		

Estuary Program Office: 805-772-3834
 Morro Bay Police: 805-772-6225
 Morro Bay Harbor Patrol: 805-772-6254
 Emergencies: 911

Confirm well count: _____	Date: _____
Entered by: _____	Date: _____
QA'd by: _____	Date: _____

APPENDIX B. MORRO BAY NATIONAL ESTUARY PROGRAM
WATER QUALITY MONITORING PROTOCOLS AND FIELD GUIDE

Water Quality Monitoring Protocols and Field Guide



Updated November 2019

Field Hazards and Precautions

Poison Oak and Stinging Nettle are two noxious plants that grow alongside many creeks throughout the watershed. If you need to move your sampling location upstream or downstream slightly to avoid contact with these plants please do so!

If you do brush up against poison oak, wash the affected area with Technu and Cala gel which are available in our office.

If you brush up against stinging nettle, it is best to soak the affected area in cold water and apply a Benadryl gel to block the antihistamine response.



Poison Oak:

Identify by shiny 'leaves of three' and lack of thorns on branches. Leaves turn red and orange seasonally.



Stinging Nettle:

Leaves have jagged edges, and small grainy flowers hanging from where leaves meet the stem.

Field Hazards and Precautions

Ticks are often found in cool riparian areas during the spring and summer. They will climb onto clothing and then onto skin where they bite and attach. They are known to transmit diseases, and should be avoided and removed immediately.

Rattlesnakes are also occasionally encountered in the watershed. They can be identified for the dark diamond pattern and characteristic rattle. If you see a rattlesnake, leave the area immediately.



Two common varieties of ticks in the Morro Bay area.



Rattlesnakes: Identify by diamond patterning and rattle at the tail.

Emergency Phone Numbers and Contacts

EMERGENCY: 911

Morro Bay Police: (805) 772-6225

Morro Bay Fire: (805) 772-6242

County Sheriff: (805) 781-4550

California Highway Patrol: (805) 593-3333

National Estuary Program Office: (805) 772-3834

Ann Kitajima: Extension 14

Karissa Willits: Extension 15

Karissa Willits's Cell Phone: (831) 334-9656

Wildlife:

Marine Mammal Rescue Center: (415) 289-7327
Injured or sick marine mammals.

Pacific Wildlife Care: (805) 543-9453
Injured or orphaned wildlife (fawns, birds of prey, other animals)

Gate Codes for Site Access

Chorro Creek Ecological Reserve (CCER) Combination Lock: 2855, 6012

Dairy Creek: 3834

The MBNEP has its own lock on this gate. It has 'MBNEP' etched on it. The combination is 3834. If you have trouble with access, contact Lasca Gaylord, Parks Supervisor: 805-440-3422.

Pennington Creek Gate: 1901

The MBNEP has access to one of Cal Poly's locks. It is a combination lock that is usually chained to two other locks. Please leave this lock and gate as you found it – if it was locked, lock it after you walk through. Be sure you lock it so that all three locks can open the gate. Cars are not allowed upstream of UPN when the road is wet. If you have any questions about if you can drive, please call Karissa or walk.

San Bernardo Creek Automatic Star Gate:

Do not drive through. Park on San Bernardo Road outside of the gate and walk in to the site.

Walters Creek, Gun Clubs: 1920

The gun club site should only be accessed on Tuesdays when the range is shutdown.

Site Access Permission

The staff and volunteers of the Morro Bay National Estuary Program have landowner permission to monitor at sites located throughout the Morro Bay watershed. Permission has been granted at the following specific sites:

Site Code	Site Description	Landowner Name
UCR	Chorro Creek on Cal Poly property, near the vineyard operations	Cal Poly (Kevin Piper)
APN	Upstream of the Beef Unit	Cal Poly (Aaron Lazanoff)
UPN	At the Beef Unit corrals near the creek	Cal Poly (Aaron Lazanoff)
CPN	Pennington Creek at the bridge on Cal Poly property	Cal Poly (Aaron Lazanoff)
SBE	San Bernardo Creek at the frontage road near Hwy 1	John Jones
CLV	Los Osos Creek, Clark Valley Road private crossing.	William Word
CER	At the road crossing on the Chorro Creek Ecological Reserve	California Department of Fish and Wildlife
CAN	At the road crossing on Canet Road	Within the rightofway of the county-maintained bridge
TWB	At the S. Bay Blvd. bridge	State Parks
SYB	On Turri Rd. near S. Bay Blvd.	State Parks
SLU	On Adobe Rd. at the bridge crossing	Pete Pedersen and Bruce Badrigian
PEN	At the amphitheater on Rancho El Chorro Outdoor School	Rancho El Chorro Outdoor School
DAL, DAM, DAU	DAL is at the bottom of the park, DAM near the dog park, and DAU at the border of National Guard and Parks property.	San Luis Obispo County Parks (Lasca Gaylord)
CHO	Downstream from the Highway 1 bridge	Camp San Luis Obispo (Sergeant Major Wilson)
COO	At the first bridge on the Coon Creek trail	State Parks
TUR	At the Turri Road bridge near Los Osos Valley Road	Within the right of way of the county-maintained
CCC	At the Chorro Creek Road crossing	Coastal San Luis Resource Conservation District

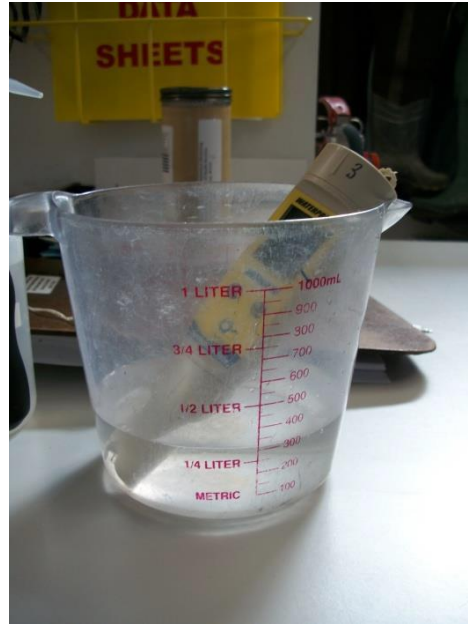
If you have any questions regarding landowner permissions or the monitoring, please contact Ann Kitajima at the National Estuary Program at 805-772-3834 or ank@mbnep.org.

Water Quality Monitoring for the Morro Bay Volunteer Monitoring Program

Where to Take Measurements:

- Optimum location: center of creek at 1/3 of water depth
- Try to sample in part of creek with mixing, do not sample in stagnant pools.
- Don't stir up water when taking measurements.
- Face upstream when taking sample – stay downstream of the equipment/sample.

Measuring pH with the pH Sensor



- Fill the measuring cup halfway with a water sample from the creek
- Press the ON/OFF button to switch the sensor ON and remove the cap from the end of the electrode.
- Place the electrode into the measuring cup. Wait for temperature reading to stabilize. This may take a few minutes. **It should come within 1 degree of the temperature reading from the DO Meter.**
- Record the temperature and pH readings on the datasheet.
- Press the ON/OFF button to switch the sensor OFF.
- Rinse the electrode with **TAP WATER** from the small squirt bottle. Squirt a small amount into the yellow sponge in the electrode cap.
- Replace the electrode cap on the end of the sensor, and place the sensor back in the case.

Dissolved Oxygen, Temperature, and Conductivity with YSI Model 2030

These YSI meters can measure DO, temperature, salinity, conductivity and specific conductance. We will be using the meters to measure DO, temperature and specific conductance, which is a temperature adjusted conductivity value. Some meters also display salinity on them. If you are taking measurements in the creek you can typically ignore the salinity value.

Instructions for use:

1. Turn on the meter before leaving the office, or upon arrive to the site, by holding down the **button on the upper right**. This button has a white *, and a green power symbol above it. You will need to hold down the power button for about 3 seconds for it to turn on.



Power
Button

2. This meter requires 10-15 minutes to stabilize before you can take readings. Turn the meter on before you leave the MBNEP office, or as soon as you reach your monitoring spot. When the Dissolved Oxygen, % saturated, is reading close to 100%, the meter is warmed up and ready to be used.

3. Remove the grey sleeve from the probe. Place the entire probe in the water. The entire probe, **up to the cable**, must be submerged to ensure that all necessary instrumentation in contact with the water.
4. Give the probe a quick shake to dislodge any air bubbles, and then gently move the probe while measuring. The instrument requires a minimum of 0.5ft/sec of flow across the probe for an accurate reading to be taken. NOTE: It gives a better measurement if you hold the probe rather than letting it hang in the water.
5. All analytes are displayed on the same screen. The order of the analytes might vary from meter to meter, so check the units. The units on the meter should match the units on the datasheet.
 - a. Dissolved oxygen concentration: DO mg/L
 - b. Dissolved Oxygen percent saturation: DO % L
 - c. Specific Conductance: spc uS/cm
 - d. *Salinity (sal ppt) – some meters only*
 - e. Temperature: °C



6. Most analytes are temperature dependent, so the temperature must be stabilized in order for the other readings to be recorded. Once the values have stabilized, record them on the datasheet.
 - a. Unlike dissolved oxygen, conductivity values stabilize very well. If the numbers continue to jump around once the temperature has stabilized, check that the probe is fully submerged in water and shake the probe to dislodge any air bubbles. If that doesn't work, turn the meter off and then back on (this should be done as a last resort, as you will need to wait for the D.O. to warm up again. NOTE: Conductivity should be a whole number (i.e., not decimals). If the number keeps jumping around or there are decimals in the value, then it is usually an indication that there is an air bubble or the probe is not fully submerged.

7. When finished monitoring
 - a. Rinse the probe thoroughly with D.I. water
 - b. Insert the probe into the grey sleeve
 - c. Leave the meter on as you travel to the next monitoring site, or turn it off when done sampling. To turn the meter off, hold down the power button until the screen goes blank.

Calibration of YSI 2030 Meter for Dissolved Oxygen

You should not need to do this. But, if the % saturation value for dissolved oxygen is not close to 100% after warming up the meter for 15 minutes, then the meter needs to be recalibrated.

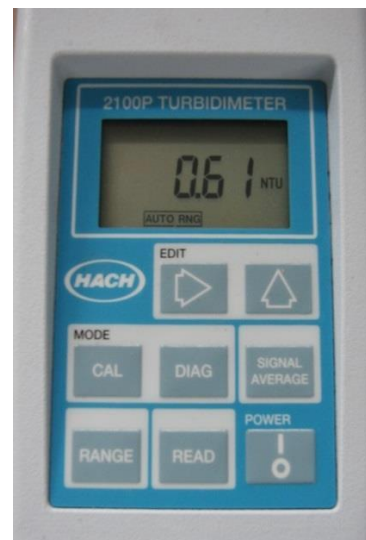
1. Ensure that the probe is inserted into the chamber on the side of the meter and that the sponge in the chamber is damp. You can add a small amount of DI water to the sponge if it does not appear to be damp.
2. Once the dissolved oxygen has stabilized, hold the “Cal” button for 3 seconds. Using the UP or DOWN arrow buttons, highlight Dissolved Oxygen and press enter. The Pro2030 will indicate “Calibrating %DO” on the display. The instrument will automatically calibrate the sensor to the current barometric pressure.
3. “Calibration Successful” will display for a few seconds to indicate a successful calibration and then the meter will return to the Run screen.
4. If the calibration is not successful, an error message will display on the screen. Press CAL to exit the error message and return to the Run screen. This is likely because the dissolved oxygen has not stabilized yet. Wait for a few more minutes for it to stabilize and try again.
5. If it still does not work, please try calling the NEP office at 805-772-3834.

Turbidity with HACH 2100P Portable Turbidimeter

- Triple rinse the syringe with creek water.
- Use syringe to fill glass sample cell to the mark (white band/arrow). Handle cell by the top, not the sides.



- If outside of cell gets wet, wipe off cell with a Kim-wipe – do not use your shirt or a paper towel since it could scratch the cell.
- Place a small drop of silicone on the cell then rub in with a Kim-wipe to reduce the effect of scratches in the glass on the reading.
- Turn instrument on.
- At the bottom of the screen it should say ‘Auto Rng’. If it doesn’t, press the RANGE button until the words appear.
- Insert sample cell with diamond on cell lined up with the notch on meter.



- Close lid.
- Press READ.
- Record value after light bulb icon turns off. Units in NTU.
- Turn instrument off.
- Empty cell. Rinse cell and syringe with DI. Leave cap off so cell can air dry.

Collecting Nutrient QA Sample

- Collect equipment: sample container, cooler and ice packs. They are all available in the back of the storeroom near our refrigerator.
- For each Ag Monitoring Site, you will need one 8 oz. plastic bottle without preservative and one 8 oz. plastic bottle with H₂SO₄ preservative.
- Label sample container as follows:
 - Client: Morro Bay National Estuary Program
 - Location: Code for your site
 - Project: MBNEP
 - Sampler: Your name
 - Date
 - Time [NOTE: It's very important to note the time because we need to get the sample analyzed by the lab before the holding time is exceeded.]
 - Sample#: Code for site



- At site, wade into stream at sampling location, taking care to not stir up water. Stay downstream of the area where you will be collecting the sample.
- For the non-acidified bottle: Open the container underwater at 1/3 the depth and fill. Dump out the water. Repeat two additional times so that the bottle has been triple rinsed.
- Fill the measuring cup and dump out. Repeat two additional times so that the measuring cup has been triple rinsed.
- Use the measuring cup to fill the sample bottle. Leave air space at the top.
- Use the measuring cup to fill the Whirl-pak.
- For the acidified bottle: Open the container underwater at 1/3 the depth and fill. Leave air space at the top.
- Cap the container.
- Store in cooler and return to NEP office.
- Put sample bottle in the refrigerator. Return the ice packs to the freezer.

- Let VMP staff know that you've left samples in the fridge.
- Samples will be picked up by the lab courier such that the appropriate hold time will be met.

Flow Measurement Protocol with Flo-Mate

- Choose a point on the creek with the following characteristics:
 - Wadeable (less than 3 feet deep) and not moving so swiftly as to be unsafe;
 - Lacks obstructions within 15 ft up or downstream of site;
 - Depth greater than 0.2 feet;
 - Bottom of the creek should be relatively flat and even;
 - Bottom of the creek should be solid so you won't sink;
 - Site at the beginning of a riffle, not a pool.



- Extend the tape measure or stadia rod across the section of the creek to be measured. Place the 0 end of the tape at the point where water is present on the left bank (when facing downstream). Make sure you use the side of the tape that measures feet in tenths, not inches. The tape must be held in place firmly during all measurements and not moved.
- You should move rocks/sticks/vegetation out of the way in the section upstream of your transect in order to achieve uniform and perpendicular flow. Try to avoid a transect that has eddying. However, only make these changes before you begin taking measurements.
- Measure the width of the creek bed with water. Record in the 'Total Width of Stream' field on the data collection form.
- Record the Equipment ID number for the meter.
- Divide the total length into **equal** segments following these guidelines:
 - If your site is < 5 feet wide – take at least 6 readings with positive velocities
 - If your site is > 5 feet wide – take at least 12 readings with positive velocities
 - Don't measure at segments smaller than 0.25ft
 - You should have a "Zero depth, zero velocity" measurement at each end of the transect that represents the wetted edge (where the water meets the side of the channel).

- If the creek length is not easily divided into segments of even length, it is fine to have shorter or longer segments at either end, but all other segments should be of equal length.
 - o For example, if the creek is 12.3' wide, you'll take measurements at the following points on the tape measure: 0', 1', 2', 3', 4', 5', 6', 7', 8', 9', 10', 11', 12' and 12.3'.
- Begin by sliding the sensor onto the mounting shaft of the top-setting rod and securing by tightening the thumbscrew. Be sure to mount the sensor so that the cable is coming straight up. Turn only until snug, do not over tighten.
- **Take your first depth and velocity reading at the 0 point on the tape measure, which is the start of the wetted width.** Don't skip that measurement, even if it looks like the water isn't really moving. Place the top-setting rod in the water so that the sensor faces upstream, directly into the current. Stand downstream of the meter and tape so that you do not interfere with the flow. Do not stand directly behind the flow meter or you might create eddies – try to stand off to the side. Make sure the rod sits flat on the bottom, stands upright, and that there are no rocks, sticks, algae, etc. interfering with the sensor.
- Take the first reading at the edge of the wetted width, which is the 0 point on the measuring tape.
- Under 'Distance from Bank', record the distance from the point on the left bank where the water begins to the point where the flow measurement is being taken.
- Visually measure the depth of the creek using the graduation lines on the hexagonal rod. **Lines are measured in tenths of feet.** Single line = 0.1 feet, double lines = 0.5 feet, triple lines = 1.0 feet. Record the depth on the data collection sheet.
- Adjust the rod so the sensor is raised above the bottom of the channel by setting the gauge on the top of the rod to the depth of the water. To set the sensor at the correct depth, press the trigger to slide the smaller rod up or down. The smaller rod has graduations marked in feet starting with '0' for depths less than 1 foot. For example, if the creek depth is 1 foot, move the small rod so that the 1 foot graduation lines up with the "0" on the large rod. If the creek depth is 1.4 feet, raise the small rod so the 1 foot graduation is aligned with the "4" on the large rod.
- Be sure the electrodes on the sensors are submerged. They are two shaded areas on the front of the sensor. If they are not submerged, there is insufficient depth to take a reading and you can record '0' flow in the fps column on the datasheet. (Typically less than 0.2 feet.) Make sure the electrodes on the sensor are facing directly into the flow.
- Once the rod is set, press the 'ON/C' button on the box to start your first measurement.



- The meter will take readings for 30 seconds as indicated by the 'Period ■■■■...' counter. You will see the reading fluctuate as the meter calculates an average. After 30 seconds, an average value in feet per second (ft/s) will be displayed on the readout. Record this value in the fps column of the datasheet. The counter will start over and give an average value after 30 more seconds. Note: If the sensor gives a negative number, record '0'.
- Repeat measurements across the channel. At each segment, measure the depth, set the rod, press the 'ON/C' button, wait thirty seconds, then record the ft/s value.

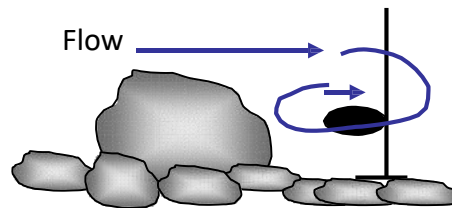
- Take the last reading at the edge of the wetted width.
- When you are done, turn off the meter by pressing the 'OFF' button.
- Disconnect the sensor from the rod, coil the cable, and place in mesh pocket.
- If a Low Battery warning ('Low Bat') is displayed on the screen, please notify the VMP staff so we can change the batteries.
- Use the 'Notes/Field Observations' portion of the datasheet to record any changes at the site (i.e., algal growth, wildlife, etc.). If you have had to move the site due to obstructions, note that on the datasheet.
- Report any problems with equipment to VMP staff.

Additional Notes on Measuring Flow

Ideally, flow is measured in an area that has a uniform shape and smooth boundary. Obviously a natural stream channel has neither. Nevertheless, you can try to increase the accuracy of your flow measurements by placing your transect at a site that does not have vegetation or rocks that sit higher in the water column than the sensor. These obstructions create swirls and eddies that disrupt a uniform flow.

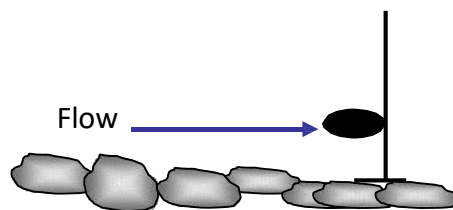
Obstructed Flow

When there are eddies from an obstruction, some of the flow is actually heading in the upstream direction, passing by the sensor from the back to the front. This is when the sensor reports a negative number.



Unobstructed Flow

You should move rocks, vegetation, or even your transect in order to minimize these disruptions and so the flow passes the sensor from the front to the back. Makes these adjustments along the transect BEFORE you start collecting measurements.



Because the FLO-MATE clearly indicates if the flow is not coming directly from an upstream direction, we can record the flow in these areas as 0 and make an effort to relocate the transect, ultimately making our measurements more accurate.

It will probably be impossible to find a location for your transect where there are not any disruptions to your flow, particularly near the edges of the channel where there is more friction and drag. Just do your best to select a transect site that has minimal disturbances.

Equipment Malfunctions and Common Mistakes

DO Meter:

- **Won't Stabilize:**

This meter can jump back and forth between a few values for a long time. Pick a mid-range value of those that are being repeatedly displayed and record on datasheet.

Make sure that the sensor is fully submerged in the water.

- **Displays LO BAT or will not turn on:**

You will not be able to collect data. Be sure to notify staff of the problem so that it can be corrected.

FLOW Mate:

- **Meter blinks on and turns off immediately:**

This meter takes size DD batteries, and has to be unscrewed in order to replace them. You do not need to attempt this replacement in the field. Notify the office when you return so that staff can fix the meter and make the measurements later in the day.

- **Meter displays negative numbers:**

This may be due to very low depth (less than 0.2) so the meter is unable to take a good reading. (If this is the case, record 0 on the datasheet.)

If the meter is entirely submerged, check for obstacles in front of the bulb (rocks, large tree roots.) These large obstacles may be creating turbulence near the meter and resulting negative numbers. You can often correct this problem by raising the bulb higher up in the water and re-starting the reading.

Quality Assurance Instructions

- The data for QA is recorded on a separate datasheet, the 'Replicate and QA Datasheet.' This form will be provided for you and is always on blue paper to distinguish it from the regular datasheets.
- **Collect replicate readings with the meters:**
 - **Dissolved Oxygen and Temperature:**
After your initial readings have been recorded, remove the probe. Wait 10 seconds, then place the probe back into the water and allow it to stabilize. Record these readings.
 - **Turbidity:**
After the initial reading has been recorded on the datasheet, remove the vial from the chamber and invert it a few times. Place the vial back into the meter and press 'READ.' The result is your replicate value.
 - **Conductivity:**
Remove the probe from the water and allow the reading to drop back to zero. Place back into the water and record the reading.
 - **pH:**
After your initial readings have been recorded, remove the probe. Wait 10 seconds, then place the probe back into the water and allow it to stabilize. Record these readings.
- **Nutrient Blanks:**
Run the test as normal, but use DI water rather than sample water.
- **Sample for QA**
Please record the samplers name, sample ID and time on the datasheet. For instructions on collecting the sample see the Collecting a Nutrient QA Sample and Agricultural Monitoring Sample portion of the protocol.
- **Flow Measurement Replicates:**
Randomly select three points in the creek with high velocity where you are measuring flow. After the first measurement, re-start the meter to count for another 30 second period. Record the second velocity, as well as the distance from the bank on the Replicate Datasheet.

Protocol for Bi-Monthly Nutrient Monitoring

Monitoring to be conducted by staff only

Overview: On a bi-monthly basis, ten sites throughout the watershed are monitored in a brief time period (approximately a four-hour period). Samples are collected and sent to a certified laboratory for analysis.

Equipment:

- DO meter
- Sample bottles, one per site
- Cooler & ice packs
- Datasheets
- Clipboard
- Camera
- Whiteboard & dry erase pen

- Sites: Dairy Creek, middle (DAM), Pennington Creek, upper (UPN), San Luisito Creek, lower (SLU), San Bernardo Creek, lower (SBE), Chorro Creek, upper (CHO), Chorro Creek, middle (UCR), Chorro Creek, lower (TWB), Los Osos Creek, Clark Valley (CLV), Warden Creek, middle (TUR), Los Osos Creek, at Turri Road and South Bay Blvd (GS1) ***Sample at a low tide***

Protocol:

- Monitoring is conducted bi-monthly.
- Notify landowners as appropriate.
- Upon arrival at site, collect readings of DO (mg/L), DO (% saturation), and water temperature. Record observations of algal coverage.
- Collect a sample:
 - o Label the bottle with date, time and site code.
 - o Remove the cap.
 - o Select a sampling spot within a riffle.
 - o Fill and empty the bottle three times.
 - o Fill the bottle and replace cap.
 - o Store in cooler.
- Photodocument the algal conditions:
 - o Write the site name, date and time on the white board. Take a picture of it.
 - o Take a picture upstream of the sample collection point, at the sample collection point, and downstream of the sample collection point.
 - o Estimate the percent cover of algae, both filamentous and microalgae. Select and record a percent coverage score: 0 = Absent (0%), 1 = Sparse (<10%), 2 = Moderate (10 – 40%), 3 = Heavy (40 – 75%), 4 = Very Heavy (>75%)
 - o Estimate the percent cover of floating mats. Select and record a percent coverage score: 0 = Absent (0%), 1 = Sparse (<10%), 2 = Moderate (10 – 40%), 3 = Heavy (40 – 75%), 4 = Very Heavy (>75%)
 - o Record the size of the area assessed.
 - o Note the presence or absence of filamentous algae and macroalgae.
- Upon return to the office, store samples in the fridge. Complete the Chain of Custody for the lab.

Water Quality Monitoring Data for the Morro Bay Volunteer Monitoring Program

Date:	Time:	Site ID:
Team names:		
Current weather:		Rainfall in past week:

Analyte	Equipment ID	Results
Water Temperature	DOE-MBV- <input type="text"/>	°C
Dissolved Oxygen <small>(concentration)</small>	DOE-MBV- <input type="text"/>	mg/L
Dissolved Oxygen <small>(% saturation)</small>	DOE-MBV- <input type="text"/>	%
Conductivity <small>(temperature corrected)</small>	DOE-MBV- <input type="text"/>	
Turbidity	TUN-MBV- <input type="text"/>	NTU
pH Electrode <small>Temperature & pH</small>	PHEL-MBV- <input type="text"/>	pH °C
Nutrient Sample	Time collected:	

Notes/Field Observations
Algae Present? YES or NO
Approx. % Algae Cover: <input type="text"/>
Trash/Litter Present? YES or NO
Oily Sheen or scum? YES or NO
Odor detected? YES or NO
Fish present? YES or NO
Other Notes? See backside for low flow

<p>Flow Volume Measurement</p> <p><small>If your site is < 5' wide, take 6 readings. If your site is > 5' wide, take at least 12 readings.</small></p>

Total Width of Stream: Equipment ID: FLO-MBV- If flow not taken, please fill out back form

Distance from Bank (feet)	Depth (feet)	Velocity (feet/second)
1)		
2)		
3)		
4)		
5)		
6)		
7)		
8)		
9)		
10)		
11)		
12)		
13)		
14)		
15)		
16)		
17)		

Distance from Bank (feet)	Depth (feet)	Velocity (feet/second)
18)		
19)		
20)		
21)		
22)		
23)		
24)		
25)		
26)		
27)		
28)		
29)		
30)		
31)		
32)		
33)		
34)		

Water Quality Monitoring Nutrient Analysis

Reason flow not measured (circle or write in) Creek connected; water level too low to measure YES or NO Creek stagnant/disconnected YES or NO Other (equipment failure, time etc). Please explain

Date Collected	
Time Collected	
Collected By	
Site Code	

Date Analyzed	
Time Analyzed	
Analysis Conducted By	
Did you collect an <u>extra</u> bottle for the lab?	Yes / No

Temperature at time of analysis (°C)	
---	--

Orthophosphates: Meter ID # COL-MBV-1

Orthophosphate as PO ₄ (mg/L)	X		=		
Meter Reading		Dilution Factor		Result	

* Place whirlpak in black refrigerator when done. Do NOT discard excess sample. Dilution necessary if meter exceeds 2.50mg/L

To be completed by staff:

Analyte	Dilution Factor	Uncorrected Result (mg/L)	Reagent Correction Factor (mg/L)	Date of RCF Analysis	Corrected Nutrient Result (mg/L)
Orthophosphates as PO ₄					
Orthophosphates as P					

Morro Bay NEP's Volunteer Monitoring Program Creek Nutrient Monitoring Protocol

This protocol describes the procedure for processing nutrient samples for creek water quality monitoring.

Samples are collected at the creek by the volunteers. They return to the office with the sample and conduct the analysis at the office.

This protocol is for all creek water quality sites.

Field Sample Collection

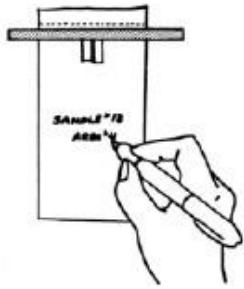
Equipment:

- Clean Whirl-pak bag, 1 per site
- Gloves
- Sharpie
- Pen
- Datasheet
- Clipboard
- Sampling pole (if necessary)

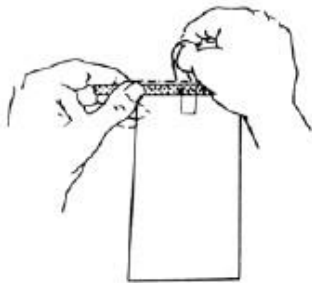
Sample Collection Procedure:

1. Collect the sample when you first arrive at the site.
2. Label the Whirl-pak with the site code, date of collection and time of collection.
3. Wear gloves for collecting the sample.
4. Be careful to not stir up sediment
5. Fill the Whirl-pak bag as follows:

INSTRUCTION SHEET FOR **WHIRL-PAK** SAMPLE BAGS



1. Label the bag with sample information if necessary.



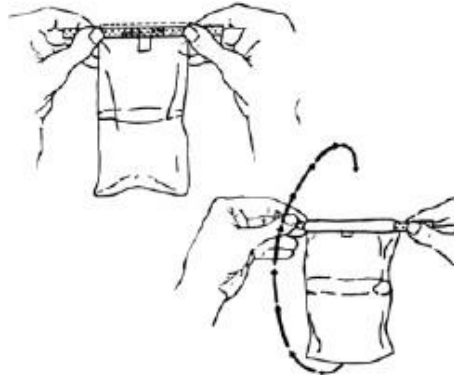
2. Tear off the top of the bag along the perforation.



3. Use pull tabs on each side to open the bag. Sometimes a little pull on the bottom of the bag helps open it completely.



4. Put sample, liquid or solid, into bag. Leave enough space at the top for closing and mixing if needed.



5. Pull the ends of the wire to close the bag. Holding the bag by the wire ends, whirl the bag three complete revolutions to form a leakproof seal. Whirling the bag will form the tightest seal. Larger bags can be closed by "folding" the tab over as tightly as possible.



6. Bend the wire ends over onto the bag to complete the closing.

Note from VMP Staff: We recommend folding the bag down, rather than whirling it. Otherwise you might end up wearing your sample.

6. Do not store the sample on ice. Bring sample to the office to conduct the analysis.

Note: Occasionally we will request that you collect a quality assurance sample to be sent to a laboratory for analysis. If that is the case, you must also bring with you an 8 oz. bottle from BC Laboratories, one per site, as well as a cooler and ice packs. Collect the QA samples in the same manner as the other samples and store in a cooler with ice packs.

In-Office Sample Analysis

Return to the office with the sample. All equipment for analysis is contained in the bin in the white cupboards. The bin is labeled “Nutrient Analysis Equipment.”

Measure Sample Temperature

1. Remove IR thermometer from its black case.
2. Point thermometer at Whirl-pak bag.
3. Press and hold grey button. You’ll see a red laser light which indicates where the thermometer is measuring.
4. Temperature should be between 18 and 25°C before conducting analysis.
5. If sample is too cold, place in incubator located under table with monitor for about five minutes and then re-measure the temperature.

Sample Analysis for Orthophosphates

Equipment:

- HACH DR/890 meter with glass cuvet and black plastic cap
- HACH orthophosphate reagent
- Kim Wipes
- DI water
- Pipetter labeled “PHOSPHATE”
- Pipette tips
- Timer
- Scissors
- PO4 Waste Container

Glass cuvet



Sample Analysis Procedure

1. Reuse the same gloves and pipette tip from the nitrate analysis.
2. Record the meter ID# on the datasheet.
3. Triple rinse the glass cuvet and plastic cap with DI water.
4. Turn on meter by pressing the **EXIT** button on the upper right hand corner of keypad.
5. Upon startup, “79 PO4” should be displayed on the bottom left hand corner of the screen.



6. If “PRGM 79” is not displayed, press the PRGM button. The display will show: “PRGM?” Then enter “79” and push the ENTER button on the bottom right hand corner of the keypad. “79 PO4” should now appear on the bottom left hand corner of the display.
7. Using the pipette labeled “PHOSPHATE”, pipette 5 mL of sample water into the cuvet twice for a total of 10 mL.
8. Cap the cuvet. Clean the outside of the cuvet with a Kim Wipe.

9. Insert the cuvet into the meter with the diamond shape on the cuvet facing forward.



10. Place the lid on the meter.



11. Press the **ZERO** button on the bottom left corner of the keypad. The display should now read “0.00 mg/L”.
12. Remove the cuvet from the meter and remove the cap. Use scissors to open silver reagent packet and add powder to the cuvet.
13. Cap the cuvet.
14. Swirl vigorously for 30 seconds and re-clean with a Kim Wipe.

15. Place the cuvet back into the meter with the diamond shape on the cuvet facing forward and place the lid back onto the meter.
16. Press **TIMER** and then **ENTER** which are both on the bottom right hand corner of the meter keypad. This starts the timer for the two minute reaction time.
17. Wait for the two minute reaction to complete.
18. After the timer finishes beeping, push the **READ** button on the bottom of the keypad. (Note: If the READ button is pushed before the timer finishes beeping, you will get a zero reading.)
19. The screen will then show the result.
 - a. If the value is less than 2.50 mg/L: Record the value on the datasheet.
 - b. If the value is greater than 2.50 mg/L: Sample requires dilution. Store sample in the fridge and notify staff.
20. Clean up:
 - a. Empty the glass cuvet in the waste container labeled 'PO4 Waste'.
 - b. Triple rinse the cuvet and cap with DI water and discard into PO4 Waste container.
21. Put the used pipette tip in the bin labeled 'Dirty Glassware'.
22. Put the water sample in the fridge.
23. Return equipment to bin and store on shelf.
24. Leave datasheet by sign-in sheet.

**Procedure for Dilutions of Orthophosphate Samples
To be Conducted by VMP Staff Only**

If orthophosphate results are greater than 2.50 mg/L, then the sample requires a dilution to obtain an accurate result. VMP staff check the sample fridge at the start of each day to determine if samples are present that require further dilutions.

Equipment:

- HACH DR/890 meter with glass cuvetts, black plastic caps and meter cap
- Orthophosphate reagent made by HACH (Phos Ver 3)
- Kim Wipes
- DI water in a bottle
- Adjustable pipetter
- Pipette tip
- Timer
- Scissors
- PO4 Waste Container

Sample Analysis Procedure:

1. Put on gloves.
2. Record the date and time of analysis on the datasheet.
3. Triple rinse the glass cuvet and plastic cap with DI water.
4. Triple rinse the pipette tip, inside and out.
5. Place a pipette tip on the pipetter.
6. Dial the pipetter to 5 mL.
7. Dispense 5 mL of DI water from the bottle into the glass cuvet.
8. Dispense 5 mL of sample water into the glass cuvet.
9. Clean the outside of the cuvet with a Kim Wipe.
10. Process sample as usual.

APPENDIX C. MORRO BAY NATIONAL ESTUARY PROGRAM
MINISONDE MS5 CALIBRATION AND DEPLOYMENT PROTOCOLS

Morro Bay National Estuary Program

HACH Minisonde MS5 Calibration and Deployment Protocols

The HACH MS5 sonde probe measures dissolved oxygen (DO), turbidity, conductivity, pH and temperature. Each parameter except temperature requires regular calibration and maintenance before and after deployment.

Storage

The MS5 needs to be stored wet to maintain the integrity of the probes. If storing the probe for longer than 24 hours, follow the protocol below.

1. Clean off the sensors using a diluted Simple Green solution and a toothbrush. Rinse off all of the soap with deionized water.
2. Wash and rinse the calibration cup, and remove all debris.
3. Fill the calibration cup up between a third to halfway full with pH 7.0 buffer or tap water. (NOTE: The probe should NOT be stored with deionized water as the pH is unstable and algae may begin to grow in the water.)
4. Screw the cap on tightly.
5. Remove batteries from battery chamber if the MS5 will not be used within the next 10 days.

Deployment

The Minisonde can be deployed either horizontal or vertical to the flow. It is important that all of the probes stay submerged throughout the deployment period.

1. The MS5 should travel to and from sites with the clear plastic calibration cup attached.
2. At the field site, remove the calibration cup and screw on the weighted black vented cap.
3. Use the chisel to remove the cap from the 4" PVC housing
4. Place the MS5 into the housing and align the venting holes of the weighted cap. Align the paint on the PCV cap to the housing and use the chisel to replace the cap on the housing.
5. Thread one cable loop through the U lock, and thread the U lock through the housing.
6. Position the MS5 in the water so that all of the vents are fully submerged. The MS5 can be deployed vertically or horizontally in the water column.
7. Gently shake the MS5 housing underwater to release any air bubbles that may be trapped in the housing and near the sensors. Secure the cable around a secure anchor such as a piling or tree trunk. Lock the other cable loop into the U lock.

Retrieval and Cleaning MS5s

The MS5 should travel to and from sites with the clear plastic calibration cup attached. Remember to bring : keys, chisel, and clear calibration cup to the site when retrieving an MS5.

1. Unlock the cable and U lock and remove the MS5 from the PVC housing.

2. Remove the weighted sensor guard and remove any debris that may have become attached to the sensors.
3. Fill the calibration cup at least halfway with creek water. Alternatively, bring a 1-gallon carboy of tap or deionized water to the site to fill the calibration cup.
4. At the lab, fill a beaker with diluted Simple Green soap and warm water.
5. Gently scrub the sensors with the diluted solution using a soft-bristled brush (toothbrush) to remove any films or foulants. *Hard scrubbing will damage probe coatings. Organic solvents like acetone or methanol will also damage the sensors.*
6. Be sure to very carefully clean the turbidity probe following marine deployment to prevent rusting.
7. Rinse the sensors with clean tap water.
8. Replace the calibration cup and wash the outside of the MS5 and weighted sensor caps with diluted Simple Green Solution.
9. Rinse black PVC housings with hot tap water and scrub with stiff bottle brushes to remove any debris that may have accumulated inside them.
10. Allow housings to dry fully before re-deploying in the field.

Power

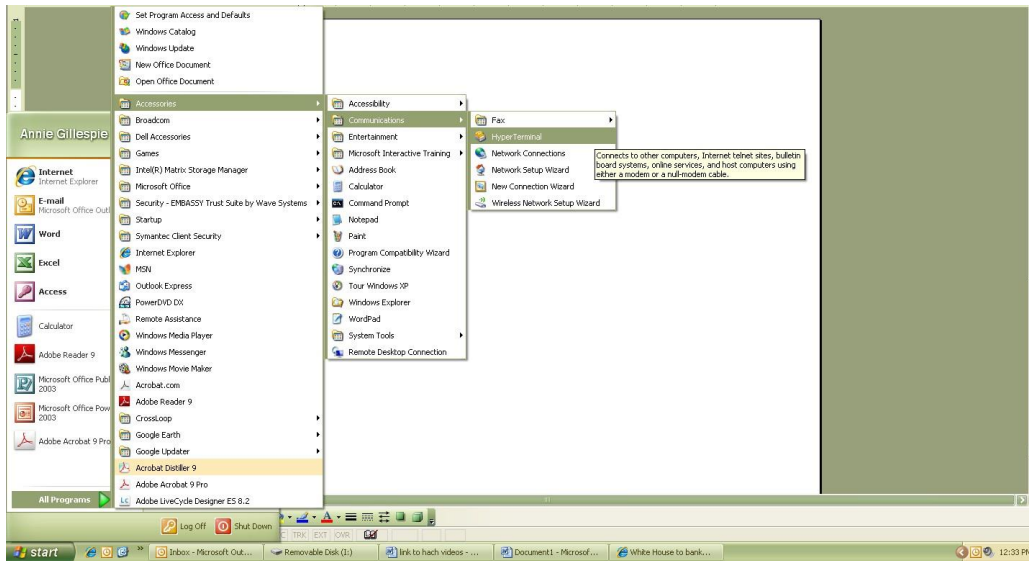
The MS5 sonde requires (8) internal AA batteries for power. The MS5 will display the remaining voltage of the batteries when connected through Hyperterminal. To replace the batteries:

1. Unscrew the white cap below the connection prongs.
2. Remove the black bands holding the batteries in place
3. Replace the batteries, carefully observing the polarity markings.
4. Replace the black bands into the indented rings.
5. Coat the battery sleeve O-rings sparingly with silicone grease if needed.
6. Screw the white cap back on to the sensor, taking care not to overtighten. If the cap is overtightened, it may crack and allow water into the battery compartment!
7. Batteries should be removed from the MS5 if it will not be used within 10 days.

MS5 Connection to Computer

The Water Science Lab computer is configured to connect to the MS5 through a program called Hyperterminal. Plug the MS5 into the connection cable before opening Hyperterminal.

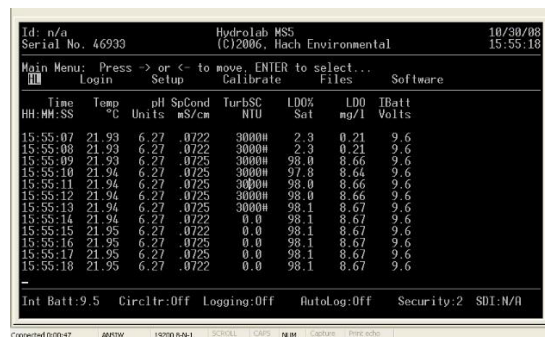
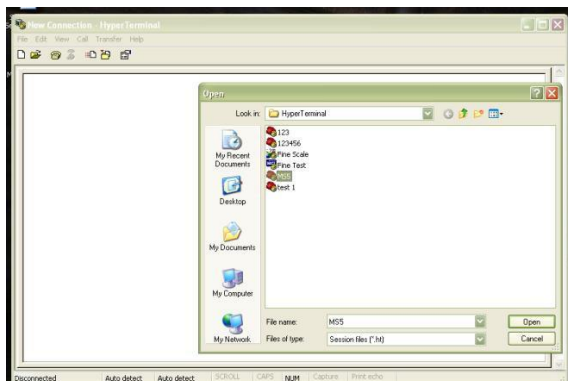
1. Open the Start Menu
2. Select “**Accessories**”
3. Select ‘**Communications**’
4. Open ‘**Hyperterminal**’



5. Hyperterminal defaults to creating a new connection, close the popup box, and select the ‘open file’ icon to open an existing connection file.
6. The connection settings to the Minsonde are saved as a connection named MS5. If the connection is not found, create a new connection with the following settings:

Connect Using: COM 1 Bits Per Second: 19200
Data Bits: 8
Parity: None
Stop Bits: 1
Flow Control: X on X off

3. When connected, the screen will turn black or potentially turn blank. If data does not start streaming automatically, press the space bar. The MS5 should beep and the screen should go black and begin streaming data from the MS5.
5. If the connection is not established and the black screen does not appear, disconnect the MS5 cable, and restart the connection process.



Calibration Procedures

Calibration must be done within 24-48 hours prior to each sampling event as well as within 24-48 hours after each sampling event.

For all parameters except dissolved oxygen and temperature, conduct a two-point calibration. The goal is to calibrate to known standards that will bracket the values you expect the equipment to encounter in the field.

Prior to calibration with each solution, ALWAYS rinse all probes and calibration cup thoroughly with a small amount of standard solution you will be using to calibrate.

Calibration Supplies

Calibration Access Database

Calibration cord to connect to computer

De-ionized water

(2) pH Buffer solutions (Usually 7.0 and 10.0)

(2) Turbidity standards (Usually deionized water 0 NTU and 20.00 NTU)

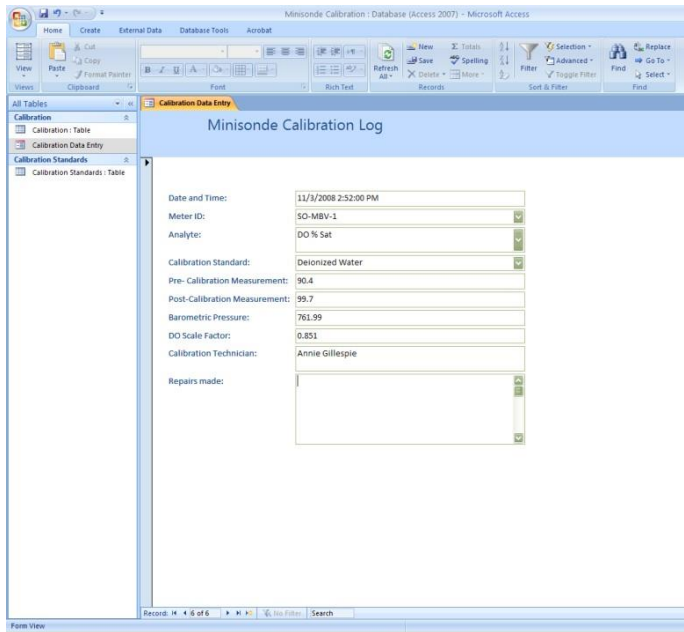
(2) Conductivity Standards (Usually 447 uS and 1413 uS for creeks)

Lint free, microfiber drying cloth.

Tap water

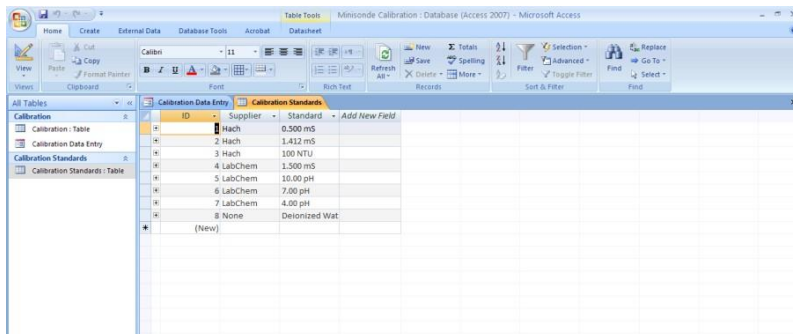
The calibration log is maintained electronically in an Access database. All calibration and repairs should be recorded in this log.

1. Open the Access database file on the desktop named '**Minisonde Calibration**'.
2. On the left menu bar double click the row for '**Calibration Data Entry**' to open the data entry form seen below. Look for the FORM, not the TABLE. Create a new record and fill in all of the rows for each calibration solution used.
3. Fill in the 'Repairs Made' box when physical maintenance is performed on any of the probes. Repairs include changing out the wiper on the turbidity probe, replacing the LDO cap, or maintaining the pH probe.



Logging in New Calibration Standards

1. All calibration standards should appear in the drop down list on the electronic calibration form
2. Calibration standards not in the list need to be entered into the Calibration Standards table. Once they are entered in this table, they will appear in the drop down list on the main calibration form.



pH Calibration

1. Rinse the calibration cup and sensor with DI water.
2. Pour a small amount (~50ml) of pH 7.0 solution into the calibration cup.
3. Screw on the calibration cup and invert to rinse the probes in pH 7.0 solution. Empty the rinse solution into the sink.
4. Fill the cup with ~100 ml of fresh 7.0 pH buffer solution. Invert the MS5 so that the solution covers the measurement probes.

5. Gently tap the calibration cup to release bubbles that are around the probes. Watch the recording values stabilize in the scrolling dataframe. When a full frame of stable values has appeared, record “pre cal” value in the calibration log.
6. Calibrate the pH probe to 7.00 using the using arrow keys.
 - a. Arrow to select ‘**Calibrate**’ and press <Enter>.
 - b. Arrow to select ‘**pH**’ and press <Enter>.
 - c. Arrow to select ‘**pH units**’ and press <Enter>.
 - d. Type in ‘**7.00**’ and press <Enter>.
7. This will return Hyperterminal to the main screen with scrolling values. When a full frame of stable values has appeared, record the “post cal” value in the calibration log (which should be 7.00).
8. Discard the pH 7.00 solution, or place in a clean beaker if calibrating a second MS5.
9. Rinse the calibration cup and probes with DI water.
10. Create a new record to calibrate using pH solution 10.0
11. Repeat calibration steps 1 through 9 using pH 10.0 buffer solution.

Conductivity Calibration

1. Rinse the calibration cup and probes with DI water.
2. Pour a small amount (~50ml) of conductivity solution into the calibration cup.
3. Screw on the calibration cup and invert to rinse the probes in calibration solution. Empty the rinse solution into the sink.
4. Fill the cup with ~100 ml of fresh conductivity solution. Invert the MS5 so that the solution covers the measurement probes.
5. Gently tap the calibration cup to release bubbles that are around the probes.
6. Watch the recording values stabilize in the scrolling dataframe. When a full frame of stable values has appeared, record “pre cal” value in the calibration log.
7. To Calibrate conductivity using the arrow keys:
 - a. Arrow to select ‘**Calibrate**’ and press <Enter>.
 - b. Arrow to select ‘**Cond**’ and press <Enter>.
 - c. Arrow to select ‘**Cond uS/cm**’ and press <Enter>.
 - d. Type in value ‘**447**’ or ‘**1413**’ and press <Enter>.
8. This will return Hyperterminal to the main screen with scrolling values. When a full frame of stable values has appeared, record the “post cal” value in the calibration log (which should be very near to the calibration standard value. Repeat steps 1 through 9 with a second conductivity standard with a calibration standard other than DI water to complete the two point calibration.
 - a. For marine deployment, use 12,880uS or 15,000 uS/cm calibration standard as the low calibration point. Use 100,000 uS/cm calibration standard as the high point.
 - b. For creek deployment use 447 uS/cm as the low point and 1413 uS/cm calibration standard for the high point.

Dissolved Oxygen Calibration

The LDO sensor compensates for the temperature of the water, so it is important that the water used for calibration maintain a constant temperature. Use the squirt bottles of DI water that have been stored in the cupboard at room temperature.

Calibrating the Dissolved Oxygen percent saturation requires an up-to-date, localized measurement of barometric pressure. Look up barometric pressure on one of two websites: the DCPD Forecast from Diablo Canyon, or the National Weather Service forecast for San Luis Obispo or Morro Bay. Both websites are bookmarked on the Sed Lab computer.

Determine the current local barometric pressure in either millibars or in/Hg. Convert the value to millimeters Hg via the Online Conversion website.

(Both weather websites, and Online Conversion are bookmarked in the Favorites Bar on the Sed Lab computer.) Record the converted barometric pressure measurement for use in calibration.

(Range is generally 740-780 mm/Hg)

1. Remove a bottle of DI water from the cupboard above the counter.
2. Pour 500ml of DI water into a clean 1liter nalgene bottle.
3. Cap the Nalgene bottle and vigorously shake the bottle for at least 60 seconds to aerate the water.
4. Stand the MS5 so that the sensors are pointed upwards with storage cup attached.
5. Fill the storage cup to the bottom of the cap threads.
6. Place the storage cap on upside-down. Do not fully screw the cap on.
7. To calibrate Dissolved Oxygen using the arrow keys:
 - a. Arrow to select '**Calibrate**' and press <Enter>.
 - b. Arrow to select '**LDO**' and press <Enter>.
 - c. Arrow to select '**LDO % SAT**' and press <Enter>.
 - d. Once the readings have stabilized, enter the current barometric pressure (as mm/Hg) and press <Enter>

Verify the accuracy of calibration by checking the Scale Factor.

1. Using the arrow keys, highlight '**Login**' and press <Enter>.
2. Highlight '**Level 3**', and press <Enter>.
3. Type in the password: **Hydrolab** and press <Enter>. The password is case sensitive.
4. Select '**Setup**' and press <Enter>.
5. Select '**Sensors**' and press <Enter>.
6. Select '**LDO**' and press <Enter>.
7. Select '**LDO % SAT**' and press <Enter>.
8. Read the scale factor, record in the calibration database. A valid calibration will produce a scale factor between 0.5 and 1.5. Values outside of this range may indicate a problem with the sensor or electronics.
- 9.
10. DO NOT type in a new value for the scale factor!
11. Press <Esc> to exit once you have verified the scale factor.
12. Record the barometric pressure used for calibration in the calibration database.

Turbidity Calibration

The turbidity sensor uses scattered light to report the concentration of suspended particles in water, so it is important to clean the instrument thoroughly and carefully before calibration and deployment. Use a soft brush and mild soap to remove dirt and debris from all of the sensors and inside the calibration cup. Rinse the storage cup and sensors with DI water and dry with lint free towels.

The turbidity probe can be calibrated using several standards. Ideally, the standards used for calibration should bracket the conditions expected during deployment. If environmental conditions are generally very clear, use DI water (0 NTU) and a calibration standard of either 20NTU or 40 NTU. If you are expecting higher turbidity during deployment, calibrate using 0 NTU, 100 NTU and 200 NTU. The high end calibration point should be a value higher than the highest value anticipated at the deployment site.

StablCal turbidity solutions are very expensive, so it is preferable to use only the smallest volume needed to conduct the calibration.

Low Point Calibration for Turbidity using DI water

1. Rinse the probes with DI water to remove any residual calibration standards.
2. Slowly fill the calibration cup with DI water.
 - a. Avoid creating bubbles when filling the cup. Bubbles can affect turbidity readings/
3. Arrow to select '**Calibrate**' and press <Enter>.
4. Arrow to select '**Turbidity**' and press <Enter>.
5. Arrow to select '**CleanRev**' and press <Enter>.
6. The next screen will ask for test rotations, type in '**2**' and press <Enter>. This activates the automatic wiper that will clean off the turbidity probe.
7. The display screen will show **3000#** for the turbidity reading until the probe has completed wiping the sensor. Proceed with step 8 when the turbidity probe begins generating numeric measurements.
8. Record the pre-calibration reading for turbidity in the calibration log.
9. Arrow to select '**Calibrate**' and press <Enter>.
10. Arrow to select '**Turbidity**' and press <Enter>.
11. Arrow to select "**TurbSC: NTU**" and press <Enter>.
12. The next screen will ask for a calibration point. Type in '**1.0**' and press <Enter>.
13. Enter the calibration standard value at the next screen. Type in '**0.0**' and press <Enter>.
14. Record the post-calibration reading for turbidity in the calibration log.

Turbidity calibration using StablCal standards

1. Gently swirl or invert the bottle of 100 NTU or 400 NTU StablCal standard for two to three minutes to mix the suspension.
 - a. Do not shake the bottle, which can suspend air bubbles in the solution and changes the turbidity.
2. Make sure that the probes are clean and dry, so that they will not drip contaminants into the StablCal standards.

3. Use a small syringe to draw 20 mL of calibration standard.
4. Use the syringe to rinse ONLY the turbidity probe with the calibration standard.
5. Slowly fill the calibration cup with a small amount of turbidity standard. The sensors should be pointing downwards for this calibration, with only the turbidity probe immersed in the solution.
6. Arrow to select '**Calibrate**' and press <Enter>.
7. Arrow to select '**Turbidity**' and press <Enter>.
8. Arrow to select '**CleanRev**' and press <Enter>.
9. The next screen will ask for test rotations, type in '**2**' and press <Enter>. This activates the automatic wiper that will clean off the turbidity probe.
10. The display screen will show **3000#** for the turbidity reading until the probe has completed wiping the sensor. Proceed with step 12 when the turbidity probe begins generating numeric measurements.
11. Record the pre-calibration reading for turbidity in the calibration log.
12. Arrow to select '**Calibrate**' and press <Enter>.
13. Arrow to select '**Turbidity**' and press <Enter>.
14. Arrow to select '**TurbSC: NTU**' and press <Enter>.
15. The next screen will ask for a calibration point. Type in '**2.0**' and press <Enter>
16. Enter the calibration standard value at the next screen and press <Enter>.
17. Record the post-calibration reading for turbidity in the calibration log.

If all plastic calibration cups are in use during calibration, trimmed plastic bottles can be used to calibrate the turbidity probe. Secure the probe (oriented downward) using the three pronged stand. The sensor requires an unobstructed view of the water to the depth of one inch. Pour one inch of calibration standard into the trimmed plastic cup, and carefully position the sensor using the three pronged stand.

Calibration standards that have been dispensed from the purchased solution bottle can be re-used for up to 2 weeks if stored correctly. Once calibration is completed, pour remaining calibration solution into a new, clean storage bottle. Cap the bottle, and label with the date and time it was dispensed, and with information identifying the StablCal standard. Used and expired solutions should be collected in the Turbidity Waste storage container for disposal at the Landfill.

New Logging File Set up

Before creating a new file for the monitoring period, it is likely that an old file will need to be deleted to create memory space. Using the black Hyperterminal interface, use the arrow keys to select the following options:

1. Arrow to select '**Files**' and press <Enter>.
2. Arrow to select '**Delete**' and press <Enter>.
3. Type in number (1 through 4) corresponding to the file you want to delete and press <Enter>.
4. At the prompt, type '**y**' for yes and press <Enter> to delete the file.
5. Hyperterminal will tell you file was successfully deleted and direct back to the mainframe menu.

To create a new file for your monitoring period:

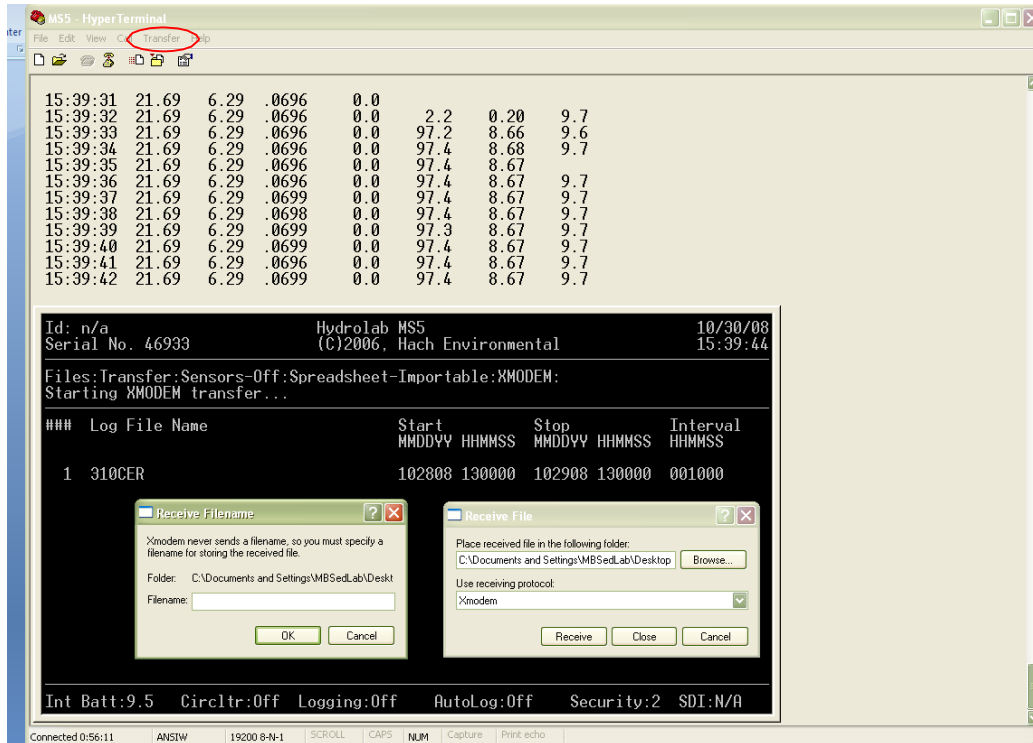
1. Arrow to select '**Files**' and press <Enter>.
2. Arrow to select '**Create**' and press <Enter>.
3. Name your file with three letter site name and 6-digit start date (the date that logging will begin).
4. Enter the following fields as they appear using the appropriate formats:
 - Start date- January 6, 2013 would appear: 010613
 - Start time- 6:30PM would appear:183000
 - End date- January 15, 2013 would appear: 011513
 - End time- 3:45 PM would appear as: 154500
 - Logging Interval (how often you want the meter to record readings)- 30 minutes would appear as: 003000, one hour as: 010000
 - Sensor warm up time should be set to two minutes: 000200
 - The MS5 sensors are NOT configured with circulators, this prompt is a remnant from prior equipment configurations. Enter 30 seconds at this prompt: 000030
 - Enable Audio: 'n'

For example, if you want to start on August 20, 2013 at 12:15PM and log every 15 minutes with a 2 minute sensor warm up and a 30 second minute circulator warm up until 5:45 PM on August 23, 2013, the series of numbers would be as follows:

Start Date: 082013
 Start Time: 121500
 End Date: 082313
 End Time: 174500
 Logging Interval: 150000
 Sensor Warmup: 000200
 Circulator Warmup: 000030
 Enable Audio: n

To download a file that has finished logging:Select '**Files**' and press <Enter>.

1. Arrow to select '**Transfer**' and press <Enter>.
2. Arrow to select '**Sensors Off**' and press <Enter>.
3. Arrow to select '**Spread Sheet Importable**' and press <Enter>.
4. Arrow to select '**XModem**' and press <Enter>.
5. Type in the number corresponding to the file (1 through 4) you want to download and press enter.
6. Go to '**Transfer**' on the Hyperterminal menu bar (outside the black mainframe- within Hyperterminal) in the drop down menu select '**Receive**'.

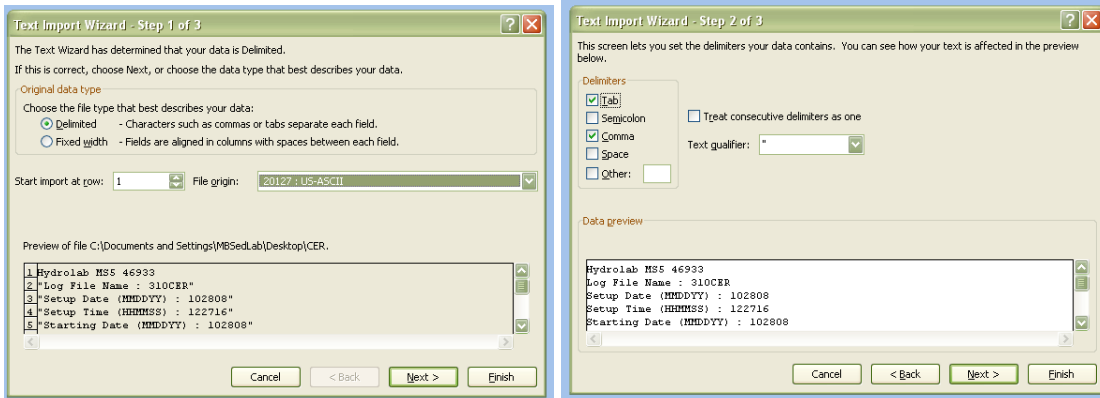


7. In the “Receive File” pop up box browse to the location for the file you are going to download.
8. Double check that the receiving protocol is “XModem”.
9. In the “Receive Filename” pop up box enter a name for the file. The downloaded csv file being created should be given the same name as the logfile that was created for the MS5. (As shown above, it would be 310CER)
10. Click “Receive” and “OK” where prompted, and the file will begin to download as packets.

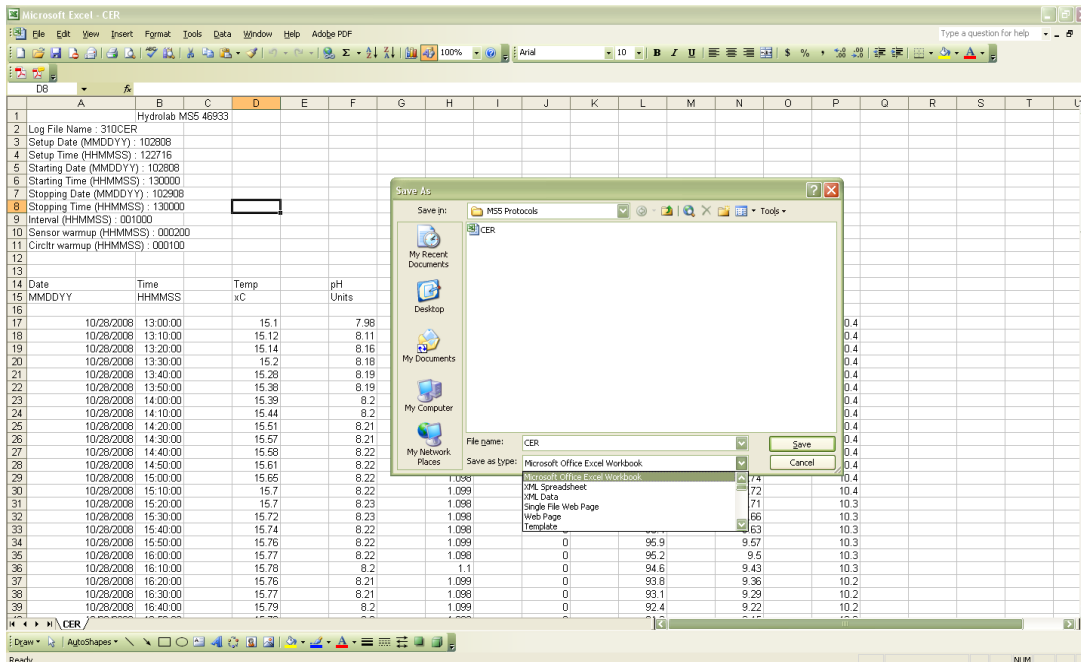
Importing Data to Excel The data file from the download needs to be imported into Excel for analysis. It is important to follow these steps in order to create an Excel workbook file.

1. Open Microsoft Excel.
2. Select “File”.
3. Select “Open”.
4. In the pop up box change the file type to include **All Files** since the imported file is not an excel workbook format file.
5. Select the file that you downloaded.
6. Select “OK”.
7. A window will appear with importing options.
 - Select the language option **US-ASCII**.
 - Check that the file is “**delimited**”.
 - Press “**Next**”.
8. In the next Text Import window:
 - Check the box for “**Tab Delimiters**”.
 - Check the box for “**Comma Delimiters**”.

- Press “Next”.



9. In the third window press “Finish”.
10. Data should appear in a columnar arrangement in a new Excel workbook.
11. To save the data file in an Excel workbook format:
 - Select “Save As”.
 - Enter a file name.
 - Open the drop down menu for “Save As Type”.
 - Select “Microsoft Office Excel Workbook”.



APPENDIX D. MORRO BAY NATIONAL ESTUARY PROGRAM
DISSOLVED OXYGEN IN THE BAY
(“DAWN PATROL”) PROTOCOL

Morro Bay Volunteer Monitoring Program Dissolved Oxygen in the Bay ("Dawn Patrol") Protocol

NEP Provided Equipment

- DO meter with safety leash
- Temp/Salinity meter with safety leash
- Clipboard
- Laminated data sheet and grease pencil
- DI water to rinse probes
- Headlamp
- Kayak
- Paddles
- Lifejacket
- Anchor

Recommended Personal Equipment

Heavy nylon or water repellent jacket and pants (rubberized clothing or wetsuits can get hot) under layers of fleece. Use neoprene or rubber gloves and booties and wool cap or bandana to cover head.

Preferred optimal tides for monitoring lowest level of oxygen in the bay would be at minimum tides. The tricky part is to account for sufficient tides to avoid getting stuck! General amount of time necessary for collection is 30 minutes.

MINIMUM TIDES NECESSARY TO GET IN AND OUT:

At least 3ft+ for an incoming tide
At least 4ft + for an outgoing tide

PROCEDURE

1. Choose a day that has an early morning high tide. You'll need three+ feet or more of water when you're taking your measurements. Field work should be conducted within two hours after sunrise. Choose a tide that allows enough water to get back safely without getting stuck in the mud (see above).
2. The most efficient route for upper bay sites (ATP, STP, LO2 and PSP), is to check ATP (Tidelands Park boat launch) from dock then drive to STP (State Marina) and launch there to get other three sites. The three backbay sites (CSI, SHI, and CHI) can be accessed by launching from Cuesta Inlet.
3. Before setting out, turn on the DO meter to give it time to warm up. It's a good idea to turn on the meter as you start out so it has adequate time to stabilize before you start taking measurements.

4. Use the safety leashes to clip the meters to either yourself or your kayak. We've lost many meters at the bottom of the bay!
5. For safety, and to help fill out the data sheets, use the headlamps. Don't forget to wear a lifejacket.
6. On the data sheet record the date, time, dawn time, sunrise time, height of tide, whether it is in or out going, and the current weather (especially wind, clear, foggy or rain).
7. Paddle to the site. Approximate your location based on landmarks.
8. Measure the DO at the site.
 - a. Remove the probe from the chamber.
 - b. Hit the Mode button so that the units shown for the reading is '%'
 - c. Place probe in the water just below the surface and move the probe slowly. You want to move the probe at an even pace, about 2 feet per second, either back and forth or in a circle. Alternatively, you could use faster and shorter strokes in an up and down motion. You'll know when you're moving the probe correctly because the meter reading will stabilize.
 - d. Wait until the meter reading stabilizes.
 - e. Record DO value at the surface (or a depth value of '0') in %.
 - f. Hit the Mode button to switch the units to mg/L. Wait for the reading to stabilize and record the value.
 - g. Record the temperature.
9. Measure salinity at the site.
 - a. Insert the probe just below the surface of the water and bob it up and down so that all air bubbles are forced out of the holes in the probe. The probe must be submerged in the water beneath the vent holes on the top of the probe. If not adequately submerged, the water won't come in contact with the probe and the reading will be inaccurate.
 - b. Hit the Mode button until the units for the reading are 'ppt.'
 - c. Wait for the reading to stabilize. (The range message must complete.)
 - d. Record the salinity value on the datasheet.
 - e. Rinse the probe with DI water and return to the storage chamber on the side of the meter.
10. Paddle to the next site and continue monitoring.
11. Upon completion of data collection, return equipment and datasheet to the NEP office. Let NEP staff know if there are any problems with the equipment.

Dawn Patrol	Date:	Time:	Sunrise:
	Samplers:		Weather:
	Tide (height & time): Incoming / Outgoing (circle)		Equipment ID:

Site	Time	Readings at the Surface				Description
		Salinity (ppt)	Temp (C)	DO (mg/L)	DO, %	
CSI						1/2 way between sign board on shore at boat launch & the other side of inlet
SHI						mouth of shark's inlet - end of golf course and across from last houses
CHI						next to 'no hunting' sign, water stick marker, south of private pier, north of cuesta inlet mouth
PSP						across from big brown house up on point, water marker to the west
LO2						midway between red tube bouy and black water marker 100 yds apart
SPM						directly out from museum of natural history
ATP						just off boat tie-up at tidelands

APPENDIX E. MORRO BAY MONITORING PROGRAM
STREAM PROFILING PROTOCOL

Morro Bay Volunteer Monitoring Program

Stream Profiling Protocol

Equipment List

1. Tripod
2. Level
3. Stadia Rod
4. Survey Tapes (100' and 200')
5. Clipboard, datasheet, pencil
6. Digital Camera
7. GPS (if needed)
8. Rebar, PVC Pipe (if needed)
9. Hammer (if needed)
10. Gloves, Machete (if needed)
11. Tecnu and Alcohol Wipes
12. Pants, Long Sleeves, Boots, [Water](#)

Main Steps

1. Locate Site
2. Define Cross Section
3. Measure Bankfull Width
4. Measure Bankfull Depth
5. Measure Floodplain Width
6. Set Up the Level
7. Measure Water Surface Slope
(Option 1)
8. Conduct Cross Section
Survey
9. Measure Water Surface Slope
(Option 2)
10. Take Digital Photos
11. Clean-Up

Procedure

1. Locate Site

- GPS points, maps, photos, and descriptions should illustrate site locations. Additionally, sites should be marked with orange flagging and PVC pipes that stick out of the ground about 2 feet on each side of the stream (Figure 1). These pipes sit over rebar stakes that also mark the cross section.
- The cross section number should be written on the pipe, the flagging, and a brass tag that is tied to one of the rebar stakes. If only one pipe/rebar can be found, focus your search for the other stake at an angle that is perpendicular to the channel (Figure 2), and use a plot of the cross section from previous surveys to measure the distance from the known stake to where the unknown stake should be located (Figure 3).



Figure 1 - Stake

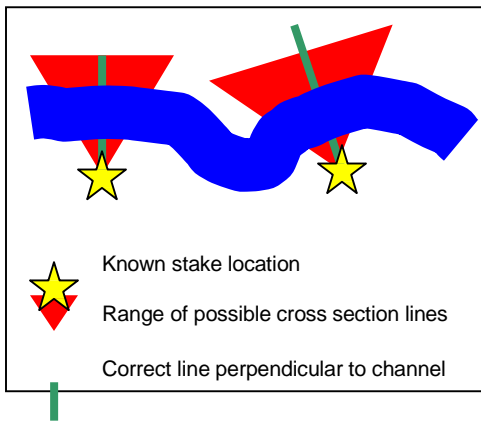


Figure 2 – Cross sections perpendicular to channel

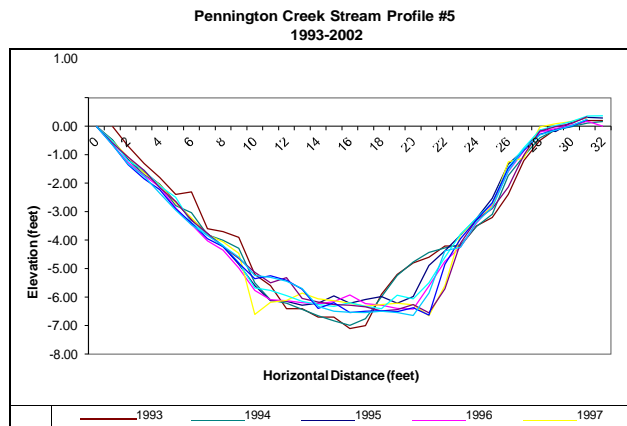


Figure 3 – Cross section plot showing 32 feet between stakes

- On data form, fill out **Creek Name, Site ID, Date,** and names of **Samplers** (Figure 4).

Creek: Chorro (upper)	Site ID: CU #5	Date: 7/1/03	Samplers: Sandy Smith, Dan Scott
-----------------------	----------------	--------------	----------------------------------

Figure 4 – Example of general survey information

2. Define Cross Section

- Remove PVC pipes to reveal rebar benchmarks, then attach the measuring tape’s metal hook to the rebar benchmark on the left bank (determine left bank by looking downstream) (Figure 5), stretch the tape tight across to the rebar benchmark on the right bank, and fasten by wrapping the tape around the rebar then knotting.

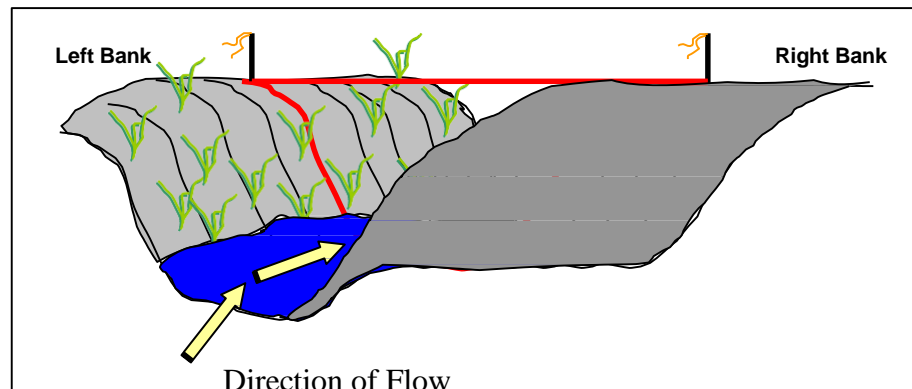


Figure 5 – The left and right banks are determined by facing downstream

3. Measure Bankfull Width

- Imagine you were standing in the stream during a large rainstorm and the channel was full of water from bank to bank. How deep would the water be? Looking both upstream and downstream try to identify combinations of clues on both banks that indicate a high water (“bankfull”) line. Note that this is not the flood line, but rather a line of flow that occurs approximately every couple years from extra large storms. Things to look for: a) obvious scour/stain lines, b) breaks in slope, c) changes in vegetation, d) changes in deposits, e) undercut banks, and f) the tops of point bars (Figure 6).

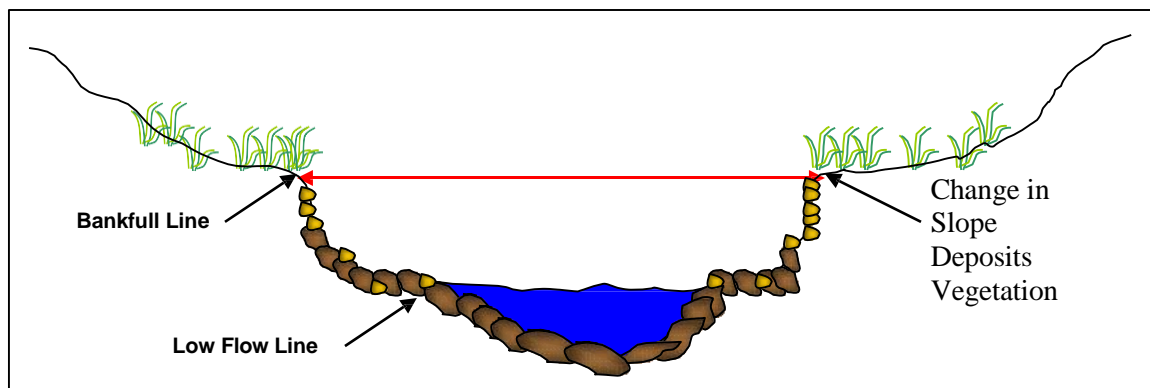


Figure 6 – Cross section depicting bankfull line

- Directly under the first tape you secured, stretch a second tape from the bankfull line on one bank, across the channel to the bankfull line on the other bank (Figure 7). This is your **Bankfull Width** — record it on the data sheet.

4. Measure Bankfull Depth

- While keeping the tape stretched across the bankfull channel, now imagine there was only a thin trickle of water in the stream. It would follow a path that wound through the deepest part of the channel bottom; this path is called the thalweg.
- To find the thalweg, use the stadia rod to locate the deepest point in the channel. Once this is established, measure the distance from the channel bottom up to the tape that represents a bankfull flow (Figure 7). This is your **Bankfull Depth** — record it on the data sheet.

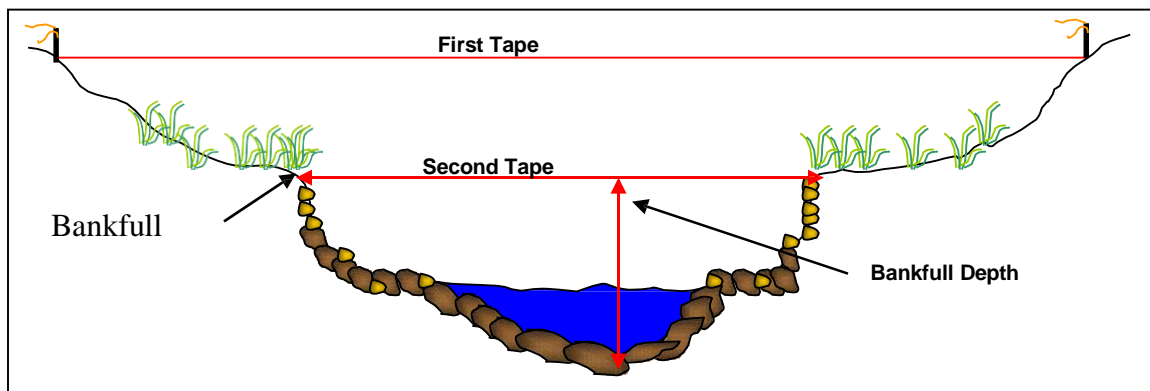


Figure 7 – Locating bankfull width and bankfull depth

5. Measure Floodplain Width

- The width of the floodplain is measured at a height above the thalweg equal to 2 times the bankfull depth (Figure 8).

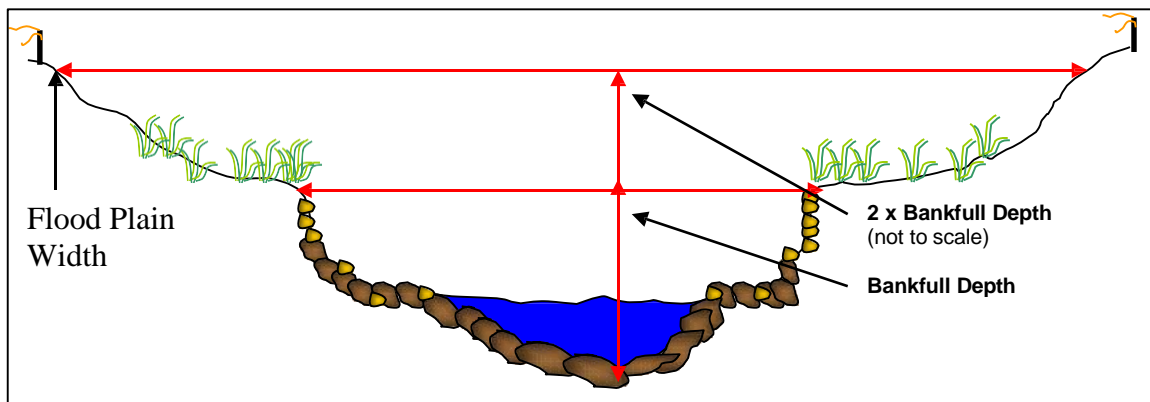


Figure 8 – Locating floodplain width

- While holding the rod in the thalweg, lift the tape to a height above the thalweg equal to 2 times the bankfull depth (e.g. if bankfull depth equals 2 feet, raise the tape to 4 feet) (Figure 9A), then raise both ends of the tape level and extend so the tape reaches both banks (Figure 9B). This is your **Floodplain Width** — record it on the data sheet.

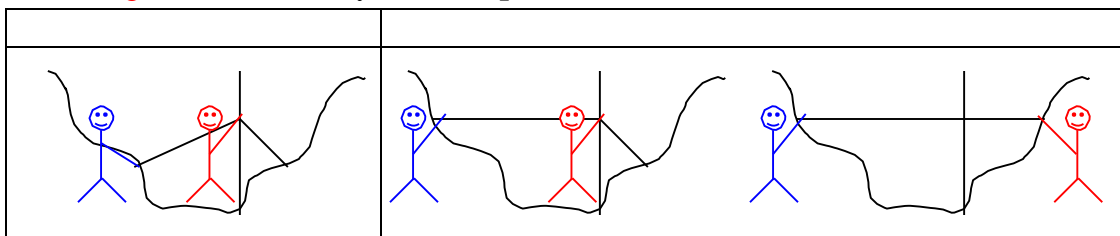


Figure 9 – Measuring floodplain width with two people

6. Set Up the Level

- Ideally, you want to be able to survey the cross section without moving the level, so try to locate the level so that it is *above* the entire line of measuring tape, although not above the extent the rod will extend, and not somewhere blocked by trees or other vegetation.
- When setting up the tripod, set it up in sturdy ground with the legs of the tripod spread sufficiently to keep it steady. Attach the level to the screw in the middle of the base on top of the tripod, tighten, and then adjust level until the bubble is centered in the middle of the circle.

7. Measure Water Surface Slope (Option 1)

- **Note:** Ideally the distance between the two readings would include at least two riffle pool sequences (Figure 10). If this cannot be achieved from the location on the bank where the level is set to read the cross section (e.g. brush obscures line of sight, creek bends, etc.), wait until you've completed surveying the cross section then move the level to a location that allows a better view both up and downstream - this may be in the channel itself rather than on the banks (**Option 2**).
- Locate the furthest riffle upstream of the cross section that is within your line of sight. Measure, along the centerline of the bankfull channel*, the distance from the cross section to the beginning of that riffle (Figure 10). This is your **Upstream Distance** — record it on the data sheet.
- At the beginning of the riffle, have one person hold the stadia rod right *on top of the water's surface* (use a rock or the toe of your boot to set it at this level), while the other

* This can be interpreted as the centerline of the creek if water was filled from bank to bank, which may be different than the centerline of the creek during the low flow summer months (Figure 11).

person takes a reading of the stadia rod with the level (Figure 10). This is your **Upstream Reading** — record it on the data sheet.

- Similarly, locate the furthest riffle downstream of the cross section that is within your line of sight, and repeat the same steps to measure your **Downstream Distance** and **Downstream Reading** (Figure 10) — record those on the data sheet.

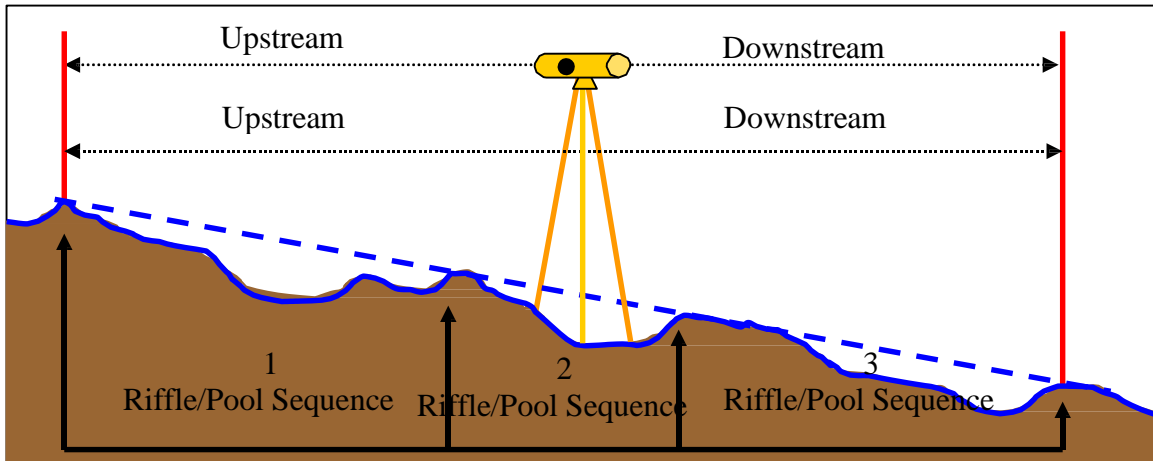


Figure 10 – Measuring slope

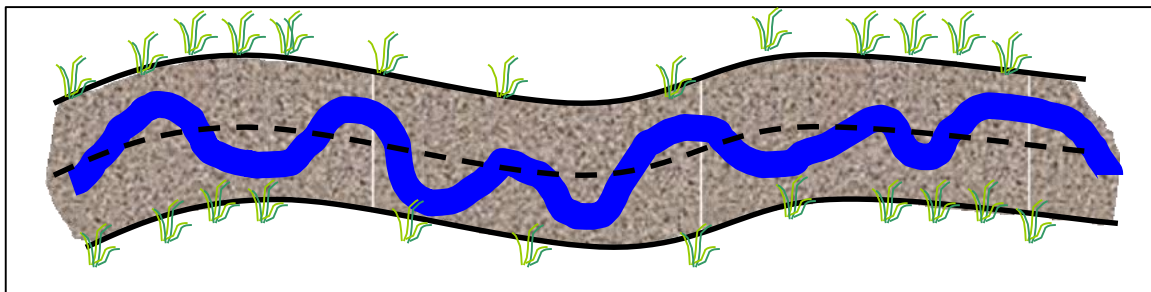


Figure 11 – Dotted line represents centerline of the bankfull channel; blue line represents a summer low flow channel

8. Conduct Cross Section Survey

- Place the rod at 0 feet (should be at the left-bank benchmark) then use the level to view where the cross hairs intersect the rod (Figure 12). This is your **Stadia Reading** — record it on the data sheet.

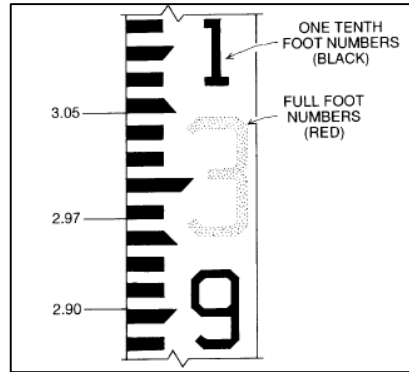


Figure 12 – Reading stadia rod

- Continue, taking a reading every horizontal foot, until you reach the right-bank benchmark (Figure 13).

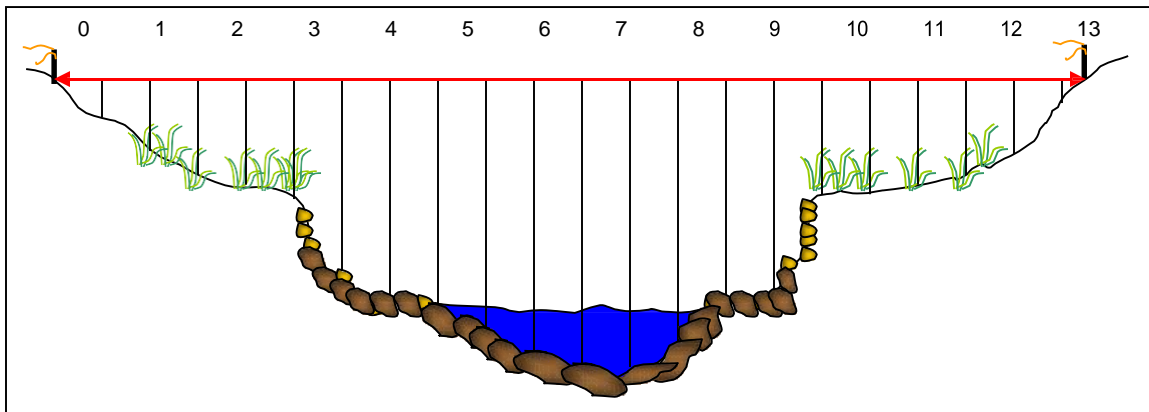


Figure 13 – Survey the cross section by taking a reading at every foot

- **Note:** It is very important that the rod be held vertically (Figure 14).

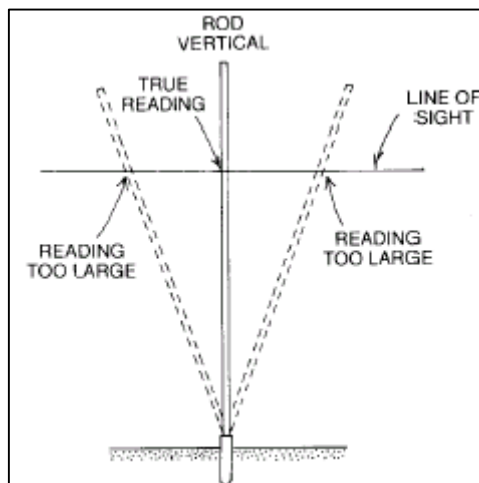


Figure 14 – Leaning the rod results in incorrect readings

9. Measure Water Surface Slope (Option 2)

- If your view upstream and downstream of the cross section was impaired from your location on top of the banks, relocate the level at this time, then take the water surface slope measurements.

10. Take Digital Photos

- Before removing the tape that stretches between the rebar, replace the PVC markers then take four digital photos to document site: 1) standing in the channel facing upstream 2) standing in the channel facing downstream 3) standing on the left bank facing the right bank, and 4) standing on the right bank facing the left bank (Figure 15).
- If possible, include the tape and markers in the view to clarify the location of the cross section within the photos. Also, if you feel that any additional photos are needed to clearly portray the cross section, feel free to take more than the primary four.

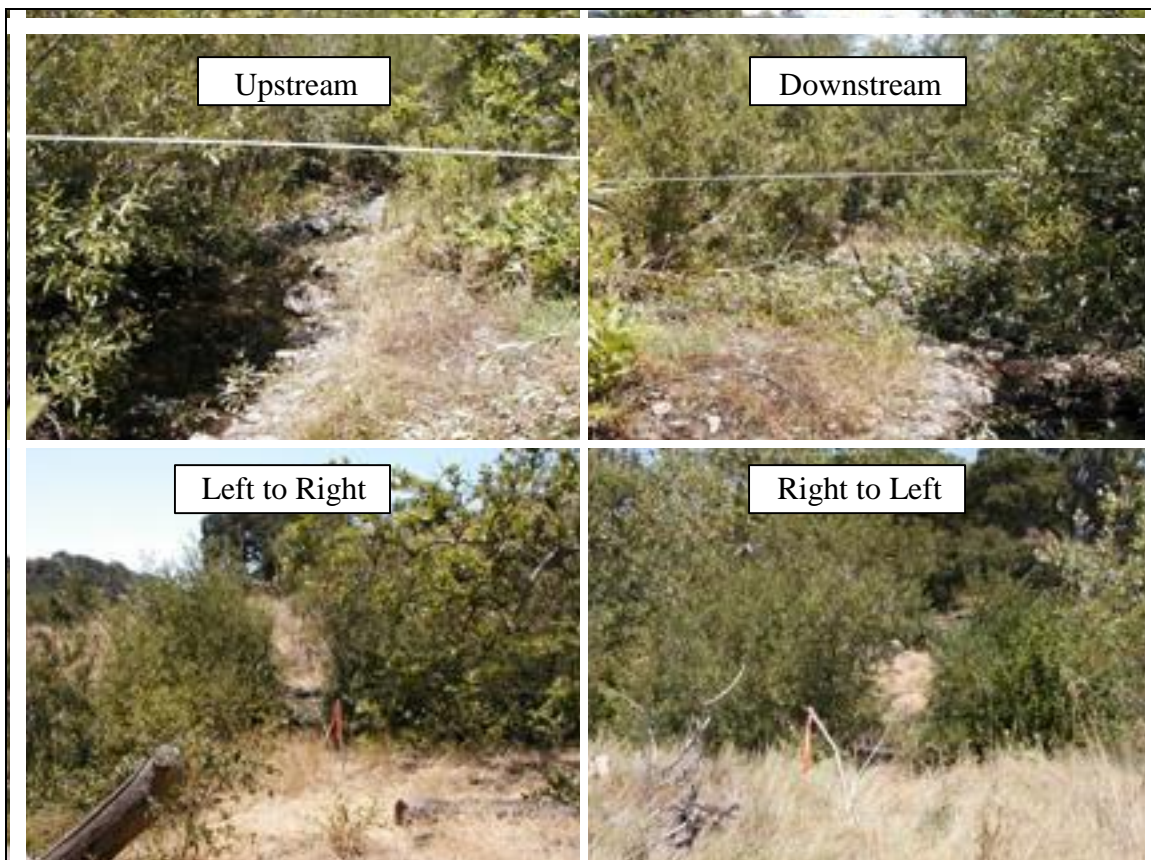


Figure 15 – Example photos

11. Clean-Up

- Remove measuring tape from rebar benchmarks, replace PVC pipes over rebar, and apply fresh flagging if needed to assist in relocating site for subsequent surveys.
- Use alcohol wipes to clean off all equipment that might have come into contact with poison oak. Then, clean any poison oak off your own skin with the alcohol wipes in the field, and wash with Tecnu when you return to the office. Also, thoroughly check yourself and your partner(s) for ticks.
- Finally, report your successful completion of the survey or any problems you encountered to the VMP staff.

Measuring Stream Gradients Using a Hand Level

Equipment:

- Stadia rod
- Hand level
- Tape measure

1. Person one (P1) stands in the upstream pool with the hand level. Stand so you have an unobstructed view of the downstream pool.
2. Person two (P2) holds the stadia rod at the water surface and takes a stadia reading of the eye level of P1.
3. P2 stands in the downstream pool and holds the stadia rod at the water surface.
4. P1 uses the hand level to take a stadia rod reading of the downstream pool.
5. Calculate the difference between the two elevation readings. This value is the rise.
6. Use the tape measure to measure the distance between P1 and P2. This value is the run.
7. Calculate the % Slope = $(\text{Rise} / \text{Run}) \times 100$

For example:

Rise: Downstream reading = 6.81; Upstream reading = 5.19 - therefore $6.81 - 5.19 = 1.62$

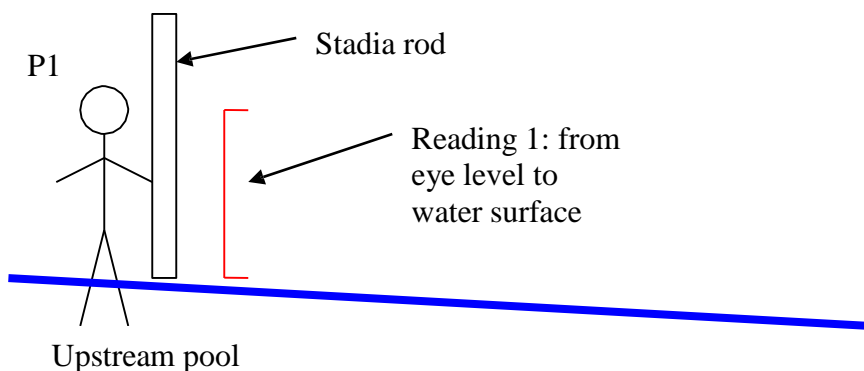
Run: using tape measure = 73

% Slope = $(\text{Rise} / \text{Run}) \times 100 = (1.62 / 73) \times 100 = 2.2 \%$

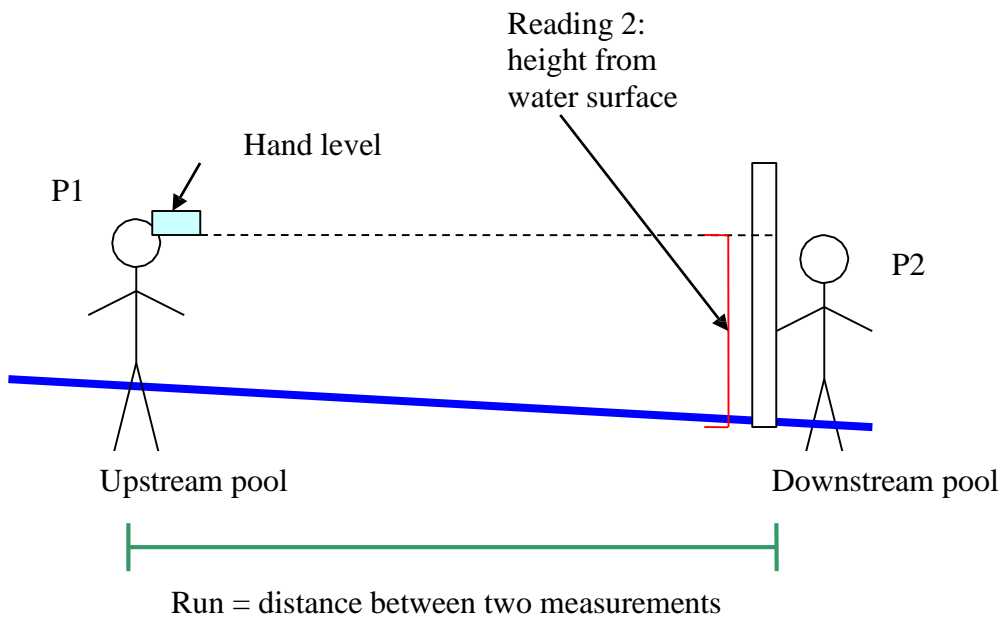
Note:

- The hand level does not magnify as much as the bigger level. To make a more accurate stadia rod reading, it helps to have the person holding the rod place their finger at different points on the rod. The person with the level can call out 'higher' or 'lower' until the rod holder's finger is at the correct reading.

Step 2.



Step 3.



Stream Profiling Datasheet

Creek:		Site ID:		Date:		VMP Staff:
Volunteers:						
Foot #	Stadia Reading	Foot #	Stadia Reading	Foot #	Stadia Reading	Bankfull and Floodplain Dimensions
0		35		70		Bankfull Width:
1		36		71		Bankfull Depth:
2		37		72		Flood-Prone Width:
3		38		73		Slope Measurements
4		39		74		Upstream Distance:
5		40		75		Upstream Reading:
6		41		76		Downstream Distance:
7		42		77		Downstream Reading:
8		43		78		Notes
9		44		79		
10		45		80		
11		46		81		
12		47		82		
13		48		83		
14		49		84		
15		50		85		
16		51		86		
17		52		87		
18		53		88		
19		54		89		
20		55		90		
21		56		91		
22		57		92		
23		58		93		
24		59		94		
25		60		95		
26		61		96		
27		62		97		
28		63		98		
29		64		99		
30		65		100		
31		66		101		
32		67		102		
33		68		103		
34		69		104		

APPENDIX F. MORRO BAY NATIONAL ESTUARY PROGRAM SURFACE
ELEVATION TABLE MONITORING

Morro Bay National Estuary Program Surface Elevation Table Monitoring for Measuring Sediment Accretion in Morro Bay Estuary

Purpose and Methods

The surface elevation table (SET) –also referred to as a sedimentation erosion table - is used to measure the sediment accumulation on the surface of a marsh or mudflat over time. In conjunction, a thin layer of feldspar can be placed around the SET as a marker horizon to measure shallow subsidence of the surface caused by compaction, desiccation, or other processes (Figure 1).

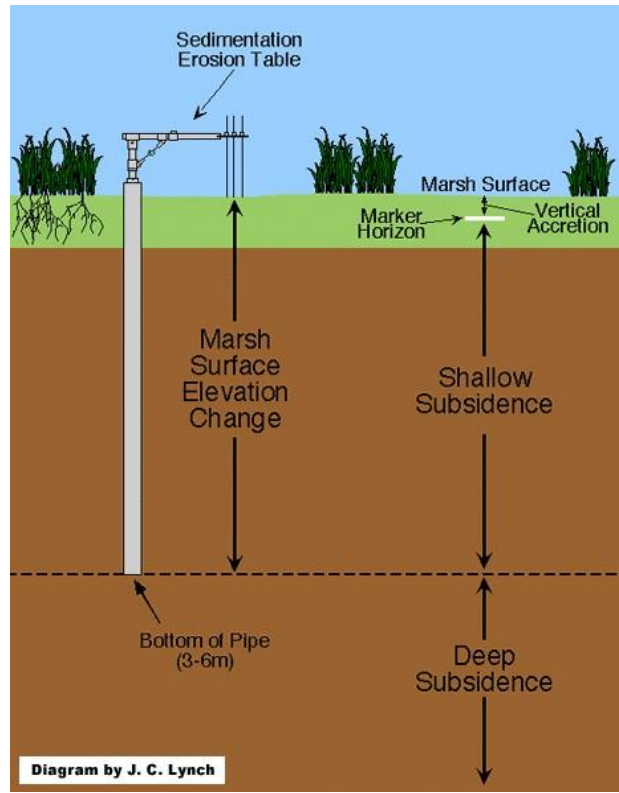
The SET is attached to a stable benchmark on the marsh surface (Figure 1). The benchmark is a 3” aluminum pipe driven into the sediment as far as possible (until refusal). The benchmarks are driven by hand, and extend 6 to 8 inches above the sediment surface. In order to take measurements, the SET is placed over the benchmark and the surface elevation is measured relative to the benchmark at radial points around the station. Since a footprint in the measurement area can contaminate the readings for years, a platform or walkway, which serves as a working surface can also be constructed (Figure 2). We anticipate using these platforms in the salt marsh as needed, but not in the mud flats.

The plan for Morro Bay is to use the SET devices mainly in and adjacent to the salt marsh, and to use marker horizons and some PVC benchmarks elsewhere in the estuary. The final site locations, will be determined based on field conditions, site access, and the expertise of the Battelle consultants. A final map showing exact locations will be made available to you upon installation. Both salt marsh and intertidal mudflats will be represented.

Recording measurements after the initial installation consists of walking to the benchmark, attaching the SET device, and measuring the elevation. The precision of measurements at the 95% confidence interval for the SET technique ranges from ± 0.4 mm to ± 1.5 mm, depending on substrate characteristics. For the horizon markers, a small core is taken and the amount of sediment that has accumulated above the horizon marker is measured from the core. The direct measurements of elevation change can be used to measure overall sediment deposition or erosion, to compare rates of sediment deposition in different regions of the estuary, and to provide data to ‘ground-truth’ the numerous sediment models of the estuary. Elevation change can be compared to the local rate of sea-level rise to determine the potential for submergence or elevation of the wetland.

We plan to take measurements pre and post wet season in the first year, and annually thereafter. We will adjust the sampling frequency depending on results, and in response to extreme events such as major storms. The time required to collect data, once on site, is relatively short (less than an hour for an experienced operator) and so man-hours depend on ease of access to the site. Our data collection program should extend for at least five years; probably longer assuming the method proves effective. Additional information on the SET methodology can be found at: <http://www.pwrc.usgs.gov/resshow/cahoon/>

Layout of Surface Elevation Tables



Surface Elevation Tables Data Collection Form

SET DATASHEET

Date _____
 Location Morro Bay
 Transect A B C
 Station low high

direction	pin	reading (mm)	notes
_____	1	_____	_____
	2	_____	_____
	3	_____	_____
	4	_____	_____
	5	_____	_____
	6	_____	_____
	7	_____	_____
	8	_____	_____
	9	_____	_____
_____	1	_____	_____
	2	_____	_____
	3	_____	_____
	4	_____	_____
	5	_____	_____
	6	_____	_____
	7	_____	_____
	8	_____	_____
	9	_____	_____
_____	1	_____	_____
	2	_____	_____
	3	_____	_____
	4	_____	_____
	5	_____	_____
	6	_____	_____
	7	_____	_____
	8	_____	_____
	9	_____	_____
_____	1	_____	_____
	2	_____	_____
	3	_____	_____
	4	_____	_____
	5	_____	_____
	6	_____	_____
	7	_____	_____
	8	_____	_____
	9	_____	_____

Surface Elevation Table Data Collection Form

MARKER HORIZON DATASHEET: SALT MARSH SAMPLING LOCATIONS

Date: _____
 Observers: _____
 Location: Morro Bay
 Transect: A B C
 Station: low high

<u>plot location</u>	<u>repl.</u>	<u>random</u> <u>x</u>	<u>random</u> <u>y</u>	<u>readings</u> 3 per core	<u>notes</u>
_____	1	29	29	_____ _____ _____	_____
	2	16	19	_____ _____ _____	_____
	_____	18	14	_____ _____ _____	_____
	_____	38	18	_____ _____ _____	_____
_____	1	20	12	_____ _____ _____	_____
	2	36	18	_____ _____ _____	_____
	_____	14	38	_____ _____ _____	_____
	_____	8	11	_____ _____ _____	_____
_____	1	44	10	_____ _____ _____	_____
	2	27	11	_____ _____ _____	_____
	_____	18	36	_____ _____ _____	_____
	_____	40	28	_____ _____ _____	_____
_____	1	15	17	_____ _____ _____	_____
	2	45	29	_____ _____ _____	_____
	_____	43	12	_____ _____ _____	_____
	_____	19	14	_____ _____ _____	_____

APPENDIX G. MORRO BAY NATIONAL ESTUARY PROGRAM
BIOASSESSMENT AND ALGAE MONITORING PROTOCOL

Morro Bay Volunteer Monitoring Program Bioassessment Monitoring Protocol

Refer to the “*Standard Operating Procedures for the Collection of Field Data for Bioassessments of California Wadeable Streams: Benthic Macroinvertebrates, Algae, and Physical Habitat*” May 2016. Prepared by the California State Water Resources Control Board Surface Water Ambient Monitoring Program.

NOTE: Sections 4.4 to 4.6 of the protocol, involving collecting and preparing algal samples, are not conducted by our organization due to lack of lab facilities and inadequate budget.

While conducting bioassessment monitoring, staff and volunteers must follow the California Department of Fish and Wildlife Aquatic Invasive Species (AIS) Decontamination Protocols, as follows.



California Department of Fish and Wildlife Aquatic Invasive Species Decontamination Protocol

The California Department of Fish and Wildlife (CDFW) is committed to protecting the state's diverse fish, wildlife, and plant resources, and the habitats upon which they depend. Preventing the spread of aquatic invasive species (AIS) in both CDFW's activities, as well as those activities CDFW permits others to conduct is important to achieving this goal. The protocols outlined below are a mandatory condition of your CDFW authorization to work in aquatic habitats. They are intended to prevent the spread of AIS, including New Zealand mudsnail (*Potamopyrgus antipodarum*), quagga mussel (*Dreissena rostriformis bugensis*) and zebra mussel (*Dreissena polymorpha*). Information about New Zealand mudsnails and quagga and zebra mussels is summarized in Attachments A and B. For complete information on the threats of AIS and aids to their identification, please visit the Department's Invasive Species Program webpage at www.dfg.ca.gov/invasives or call (866) 440-9530.

Many AIS are difficult, if not impossible to see in the environment and can be unknowingly transported to new locations on equipment. Therefore, decontamination is necessary to prevent the spread of AIS between collection locations. Equipment shall be decontaminated between each use in different waterbodies. All equipment, including but not limited to, wading equipment, dive equipment, sampling equipment (e.g., water quality probes, nets, substrate samples, etc.), and watercraft, must be decontaminated using one or more of the protocols listed below. As an alternative to decontaminating on-site, you may wish to have separate equipment for each site and to decontaminate it all at the end of the day. Listed below are three options for equipment decontamination. Use your judgment and field sampling needs to select the method(s) that are appropriate for your equipment and schedule. **Because there are currently no molluscicides registered with the California Department of Pesticide Regulation that have been demonstrated to be effective for these three species, CDFW cannot recommend chemical decontamination.** If you would like training on implementing these protocols please contact the Invasive Species Hotline at (866) 440-9530 or e-mail invasives@wildlife.ca.gov

General field procedures to prevent the spread of AIS:

- If decontamination is not done on site, transport contaminated equipment in sealed plastic bags and keep separate from clean gear.
- When practical, in flowing water begin work upstream and work downstream. This avoids transporting AIS to non-infested upstream areas.
- For locations know to be infested with AIS, use dedicated equipment that is only used in infested waters. Store this equipment separately.

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Equipment Decontamination Methods

Option 1: Dry

- Scrub gear with a stiff-bristled brush to remove all organisms. Thoroughly brush small crevices such as boot laces, seams, net corners, etc.
- Allow equipment to thoroughly dry (i.e., until there is complete absence of moisture), preferably in the sun. Keep dry for a minimum of 48 hours to ensure any organisms are desiccated.

Option 2: Hot water soak

- Scrub gear with a stiff-bristled brush to remove all organisms. Thoroughly brush small crevices such as boot laces, seams, net corners, etc.
- Immerse equipment in 140° F or hotter water. If necessary, weigh it down to ensure it remains immersed.
- Soak in 140° F or hotter water for a minimum of five minutes.

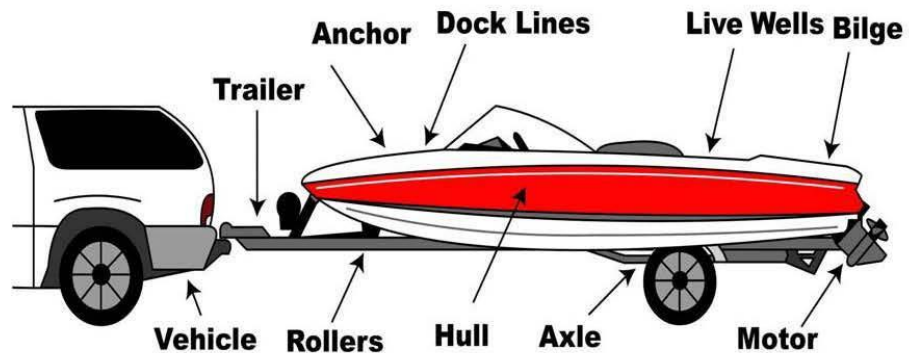
Option 3: Freeze

- Scrub gear with a stiff-bristled brush to remove all organisms. Thoroughly brush small crevices such as boot laces, seams, net corners, etc.
- Place in a freezer 32°F or colder for a minimum of eight hours.

Watercraft Decontamination

- Prior to leaving the launch area, remove all plants and mud from your watercraft, trailer, and equipment. Dispose of all material in the trash.
- Prior to leaving the launch area drain all water from your watercraft and dry all areas, including motor, motor cooling system, live wells, bilges, and lower end unit.
- Upon return to base facilities, pressure wash the watercraft and trailer with 140° F water*, including all of the boat equipment (i.e. ropes, anchors, etc.) that came into contact with the water.
- Flush the engine with 140° F water for at least 10 minutes and run 140° F water through the live wells, bilges, and all other areas that could contain water.

*To ensure 100% mortality the water needs to be 140° F at the point of contact or 155° F at the nozzle.



Reporting Aquatic Invasive Species

If you suspect you have found New Zealand mudsnail, quagga and zebra mussels, or other AIS, please immediately notify the CDFW Invasive Species Program at (866) 440-9530 or e-mail invasives@wildlife.ca.gov. Please provide your contact information, specific location of discovery, and digital photographs of the organisms (if possible).

Attachment A

New Zealand Mudsnaill

The threat posed by New Zealand mudsnails (NZMS):

- NZMS reproduce asexually therefore it only takes a single NZMS to colonize a new location.
- NZMS are prolific, and a single NZMS can give rise to 40 million snails in one year.
- Densities of over 750,000 NZMS per square meter have been documented.
- NZMS out-compete and replace native invertebrates that are the preferred foods of many fish species and alter the food web of streams and lakes.

Identifying NZMS:

- NZMS average 1/8 inch in length, but young snails may be as small as a grain of sand. Adults bear live young.
- See the photos, below, for assistance identifying NZMS. Expert identification will be necessary to confirm identification.

IDENTIFYING THE NEW ZEALAND MUDSNAIL

Size: A mature snail is usually less than 5 mm (.2 in) long. (Photo by Jane and Michael Liu.)

Shape: Shell is elongated and dextral (its whorls or spirals lean toward the right). Snail typically has between 5 to 6 whorls on its shell. (Photo by D. L. Gustafson, <http://www.esg.montana.edu/aim/mollusca/nams>.)

Color: Most snails have a light- to dark-brown shell that may appear to be black when wet. (Photo by Jane and Michael Liu.)

Embryos: Upon dissection, mature snails will have brooded embryos. (Photo by D. L. Gustafson, <http://www.esg.montana.edu/aim/mollusca/nams>.)

Operculum: The mudsnail operculum (a rounded plate that seals the mouth of the shell when the animal's body is inside) can be seen on live snails but is not easily visible on dead or preserved snails. (Photo by D. L. Gustafson, <http://www.esg.montana.edu/aim/mollusca/nams>.)

NZMS Habitat:

- NZMS can live in most aquatic habitats, including silted river bottoms, clear mountain streams, reservoirs, lakes and estuaries.
- NZMS have a temperature tolerance of 32-77° F.
- NZMS can survive out of water for more than 25 days in cool, moist environments, and have been found over 40 feet from water.

Current known locations of NZMS in California can be found at <http://nas.er.usgs.gov/taxgroup/mollusks/newzealandmudsnaildistribution.aspx>

Attachment B

Quagga and Zebra Mussels

The threat posed by quagga and zebra mussels (Dreissenid mussels):

- Dreissenid mussels multiply quickly and out-compete other species for food and space.
- Their presence can alter food webs and alter environments, negatively affecting native and game fish species.
- Dreissenid mussels attach to hard and soft surfaces, and negatively impact water delivery systems, hydroelectric facilities, agriculture, recreational boating and fishing.
- Adults can survive up to 30 days out of water in cool, humid conditions.
- Produce microscopic larvae that can be unknowingly transported in water, including live-wells, bilges, and motors.

Identifying Dreissenid mussels:

- Typically the same size as a fingernail but can grow up to about 2 inches long.
- Variable, usually dark and light alternating stripes. May also be solid cream, brown, or black.

Dreissenid mussel habitat:

- Variable, including both hard and soft surfaces in freshwater.
- From surface depth to more than 400 feet in depth.



Current known locations of Dreissenid mussels in California can be found <http://nas.er.usgs.gov/taxgroup/mollusks/zebramussel/maps/CaliforniaDreissenaMap.jpg>

APPENDIX H. MORRO BAY NATIONAL ESTUARY PROGRAM EELGRASS PERMANENT
TRANSECT MONITORING PROTOCOL

**Morro Bay National Estuary Program
Eelgrass & Algae Field Transect Protocol**

Monitoring takes place at five permanent stations located throughout Morro Bay. Each station has one to three permanent transect locations for monitoring. All transects are 50 meters long. At each site there is one ‘Long’ transect and two ‘Depth’ transects.

Long transects are 50 meter transects that are monitored at 20 distinct quadrat locations.

Depth transects are 50 meter transects that are parallel to the Long transect and are monitored at only 12 distinct quadrat locations.

Table 1. Monitoring Locations

SiteName	Transect_Type	Lat	Long
Coleman	Long	35.372120	-120.860617
Reference	Long	35.357709	-120.853375
Reference	Depth	35.357604	-120.853544
Marina	Depth	35.346079	-120.846312
Marina	Long	35.345975	-120.846031
Chorro	Long	35.337993	-120.846888
Pasadena	Long	35.333396	-120.849309
Tidelands	Long	35.359488	-120.851817

Coleman and **Marina** stations are located on the East side of the navigation channel.

Tidelands, **Pasadena**, **Reference** and **Chorro** stations are located on the West side of the navigation channel.

The procedure differs depending on the density of eelgrass present at the transect. In areas with dense eelgrass coverage, 30 shoots can be collected from each site for biomass analysis. For areas with sparse eelgrass coverage, adequate shoots may not be present. In this case, five shoots are collected from the monitoring site. The protocol is divided into steps for Dense Sites and Sparse Sites.

Figure 1. Eelgrass permanent transect monitoring locations with 2017 eelgrass extent.



Supplies Checklist

- waterproof datasheets (up to 3 per site)
- (2) 300ft standard and metric Keson Transect Tapes
- (1) 15 meter metric Keson Transect Tape
- (1) photo whiteboard
- Dry erase markers
- (2) Plastic rulers (metric)
- (4) temporary transect end stakes
- GPS unit
- Plastic clipboards & pencils
- Ziploc Bags (1 gallon)
- Waterproof biomass sample labels
- Waterproof camera
- PVC 0.25 m² meter quadrat
- PVC 0.0625m² quadrat

Setting up Transects and Quadrats

1. Review site maps for the site and determine which transect is being monitored (A, B or C).
2. Select the appropriate datasheet for which quadrats must be counted.
3. Fasten the Keson tape with the 0 end at the northern stake. (Reference site maps if needed.)
4. Take photos of the transect from each end of the transect looking in towards the area to be assessed.
5. All quadrat measurements are completed on the shallow side of the transect tape.
6. Place a sampling quadrat (using the distances provided on the datasheet) on the shallow side of the transect. Position the quadrat so that the center mark of the quadrat is on the specified Keson tape distance mark.

Monitoring Protocol for Dense Sites

Photo Documenting Quadrats

Take photos before disturbing any of the vegetation in the quadrat.

1. Fill out the whiteboard with the transect and quadrat information for the quadrat picture:
 - a. Site Name
 - b. Transect Name (A,B or C)
 - c. Quadrat Number (location on tape)
 - d. Date
 - e. Time
2. Place the whiteboard outside the quadrat frame, within the area captured in the photo.
3. Take a photograph of the quadrat. Take the photograph as vertical as possible, including the entire quadrat frame, the whiteboard photo label, and the Keson tape measure. Try to avoid shadows and reflections in the photograph.

Example photos:



Measuring Quadrat Percent Coverage and Canopy Height

1. Identify the seagrass and algae species present in the quadrat.
2. Estimate % cover of each species (seagrass and algae) in the quadrat. Minimum % cover increments are 5%. Record the percent coverage for each vegetation type in the **Coverage %** columns on the datasheet.
3. Randomly select a clump of eelgrass blades from within the quadrat. Extend the leaves sideways to their maximum length without uprooting.
4. Use the 15-meter Keson tape to measure the length *in centimeters* from the substrate to the leaf tips. Ignore the longest 25% of the eelgrass leaves in the clump.
5. If grass density is very low and a clump of blades cannot be grouped, make measurements on 5 individual shoots within the quadrat. Record the 5 separate measurements in the **Canopy Height (cm)** column.
6. If the quadrat contains algae but no eelgrass, use a ruler to measure the depth of the algae mat (in centimeters) from the surface of the mud.
7. Record the measurements in the **Canopy Height (cm)** column on the datasheet.
8. Note whether the canopy height measurements were taken for algae or eelgrass.

Counting Eelgrass Shoots. Measuring Water Depth

1. Carefully separate eelgrass shoots around the outside of the transect frame to determine which shoots are rooted within the frame. Shoots that are lying down into the frame should be folded back out of the frame to avoid confusion.



2. If the frame is very dense with evenly distributed shoots, count 25% or 50% of the quadrat. Use the small white quarter squares to mark off which portions of the quadrat will be counted.
3. Fill in the % area counted (25%, 50%, 100%) in the **Quad Area** column on the datasheet.
4. Count shoots individually, feeling down to the base of the shoot at the sediment. Where possible, count all shoots in the quadrat (100% of the quadrat).
5. Record the total number of shoots for the area counted in the **EG Shoot Count** column.
6. If the majority of the quadrat is submerged in water, use the plastic ruler to measure water depth in centimeters. Record the water depth in the **Water Depth (cm)** column on the datasheet.
7. Do not record water depth if less than 50% of the quadrat is submerged. Record zero if water depth is less than 1cm.

Photo Documenting Quadrats

1. Once the quadrat is placed, fill out the whiteboard with the following information:
 - a. Site Name
 - b. Transect Name (A,B or C)
 - c. Quadrat Number (location on tape)
 - d. Date
 - e. Time
2. Take a photo of the quadrat.

Eelgrass Monitoring Datasheets:

- Long Transect Datasheet
- Depth (Shallow) Transect Datasheet
- Depth (Mid) Transect Datasheet
- Depth (Deep) Transect Datasheet

A (Shallow Transect)

Date: _____ Time Arrived: _____ Monitor Names: _____

Station Name: _____ Transect Letter: _____

Coleman =COL Tidelands =TID Manning= MAR
 Chorro =CHO Pasadena = PAS

Algae Biomass Collected: _____ Y or N # Samples _____

Eelgrass Biomass Collected: _____ Y or N # Shoots _____

Transect Site Photos: _____ North facing: _____ South facing: _____

Trans Name	Trans Letter	Quad Position	Coverage %				Canopy Height (cm)	Canopy Notes	EG Shoot Count	Water Depth (cm)	Additional Notes
			Eelgrass	Ulva	Gracilaria	Other					
		2									
		7									
		8									
		16									
		18									
		25									
		26									
		33									
		38									
		40									
		44									
		46									

MBNEP Long Transect Form

Date: _____ Time Arrived: _____ Monitor Names: _____

Station Name: _____ Transect Letter: _____

Coleman =COL Tidelands = TID Marina= MAR
Chorro =CHO Pasadena = PAS

A= Shallow B= Middle C=deep

Algae Biomass Collected: _____ Y OF N _____ # Samples _____

Transect Site Photos: _____ North facing: _____ South facing: _____ Eelgrass Biomass Collected: _____ Y OF N _____ # Shoots _____

Station Name	Trans Letter	Quad Position	Coverage %				Canopy Height (cm)	Canopy Notes	EG Shoot Count	Water Depth (cm)	Additional Notes
			Eelgrass	Ulva	Gracilaria	Other					
		1									
		3.5									
		6									
		8.5									
		11									
		13.5									
		16									
		18.5									
		21									
		23.5									
		26									
		28.5									
		31									
		33.5									
		36									
		38.5									
		41									
		43.5									
		46									
		48.5									

B (Middle Transect)

Date: _____ Time Arrived: _____ Monitor Names: _____

Station Name: _____ Transect Letter: _____

Coleman =COL Tidelands = TID Merrimac = MAR
 Chorro =CHO Passadena = PAS

A= Shallow B= Middle C=deep

Algae Biomass Collected: _____ Y or N # Samples _____

Eelgrass Biomass Collected: _____ Y or N # Shoots _____

Transect Site Photos: North facing: _____

South facing: _____

Trans Name	Trans Letter	Quad Position	Coverage %				explain OTHER	Canopy Height (cm)	Canopy Notes	EG Shoot Count	Water Depth (cm)	Additional Notes
			Eelgrass	Ulva	Gracilaria	Other						
		9										
		10										
		15										
		17										
		22										
		25										
		28										
		31										
		35										
		37										
		39										
		45										

C DEEP

Station Name: _____ Staff Names: _____

Date & Time Arrived: _____ Time Departed: _____ Tide Height & Time: _____

Trans Name	Trans Letter	Quad Position	Photo	Coverage %				Canopy Height (cm)	Quad Area	EG Shoot Count	Water Depth (cm)	Flowering/ Grazing
				Eelgrass	Ulva	Gracilaria	Other					
		5										
		7										
		10										
		18										
		19										
		22										
		26										
		34										
		35										
		38										
		43										
		44										

Transect Codes:
 A = Shallow
 B = Middle
 C = Deep

Station Names:
 Coleman
 Tidelands
 Pasadena

COL **Chorro** **CHO**
TID **Marina** **MAR**
PAS

Substrate Type:
 Sand
 Fines
 Mixed

WQ Measures:
 Temp:
 Salinity:

APPENDIX I. MORRO BAY NATIONAL ESTUARY PROGRAM EELGRASS INTERTIDAL BED
CONDITION MONITORING PROTOCOLS

Morro Bay Intertidal Eelgrass Bed Condition Monitoring Protocol

Intertidal eelgrass monitoring will take place in 4 of the most significant beds throughout Morro Bay. The goal of monitoring is to determine eelgrass bed conditions through density, patchiness, and observational data. Typically, a 150 meter, 7 quadrat survey, can be completed in one tide window with 2-3 people (3 ideal). Ideal tides for intertidal monitoring are below -0.5 feet. Beds are considered separate if they are 50 meters or more apart.

Eelgrass monitoring should be conducted two times a year: in the fall (August – November with preference for September and October) and again in the spring (April – May). Intertidal monitoring will be conducted at eelgrass beds at Coleman Beach, North Sandspit, Reference Bed and Windy Cove. Refer to the attached map for eelgrass bed locations.

Supplies

- 0.50m x 0.50m quadrat
- 50m or longer Keson Transect Tape
- (1) photo whiteboard
- Dry erase board markers
- Meter stick or metric ruler
- GPS unit
- Waterproof camera
- Plastic clipboard & pencils
- Rite-in-the-rain datasheets (one per quadrat)
- Squirt bottle for cleaning eelgrass
- Field notebook
- Personal Gear: wading boots or wetsuit, depending on how bed is accessed.
- Kayak, paddleboard or surfboard depending on bed locations

Protocol:

Complete a quadrat every 25 meters along the transect. Begin at transect start point, which is defined by landmarks and lat/long coordinates. Record transect orientation on datasheet (e.g. Survey direction from North to South)

1. Fill out the site name (not site code), date, time, surveyors, GPS point and transect orientation on datasheet.
2. Record the quadrat meter mark on the datasheet. (the first quadrat is quadrat 0).
3. Anchor the meter tape at the start of the transect.
4. Place the quadrat at the starting point along the landward edge of the bed, but slightly water ward so that the quadrat is not at the very edge, if possible. Placement should be in eelgrass. Place along the landward edge of the bed to prevent double sampling during subtidal surveying.
 - Note: If eelgrass is not present at the zero mark, move forward up to 12 meters to place the quadrat. If there is no eelgrass within 12 meters, then circle “No Eelgrass” next to stipe counts and move on to the 25 m mark on the transect. For example, if there is no eelgrass present at meter 0, but there is eelgrass at meter 10, the quadrat would be done at meter 10. Quadrat meter mark on datasheet would be recorded as ‘10’, and GPS location

would be recorded for the 10 m mark. Regardless of where the quadrat ends up, the next one should be attempted at the 25 m mark (and if no eelgrass there, that point can be shifted +/- 12 meters to find eelgrass).

5. Take a GPS point for the quadrat and record lat/long on datasheet.
6. Record site name, date and quadrat number on the white board. Take a photo of the white board and then a photo of the quadrat.
7. **Quadrat:** Using the point intercept method, count the total number of corners or intersection points (25 total) that have the following directly under the intersection or at the inside corners or edges of quadrat. Within each category below there can be a maximum of 25 counts, but there can be multiple categories present at each point. Use finger to check if material is directly under the point (only include in count if directly under) and check for layering (e.g. gracilaria may be under or over eelgrass, but either way it should be counted if under that point).
 - a. Eelgrass (**live** blade or stipe)
 - b. Gracilaria: red and stringy
 - c. Ulva: green and leafy
 - d. Fauna (indicate type: e.g. sand dollar, small brown snail)
8. **Stipe Count:** Count the number of stipes in each of the 4 center squares (see Figure 1) of the quadrat to measure density. Keep counts separate for each square, though order of counts doesn't matter.

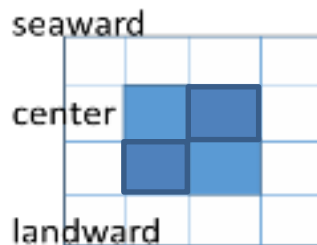


Figure 2: Count the number of stipes in each of the 4 center squares

9. **Blade Condition:** At the nine points on the quadrat (as shown in Picture 2), repeat the following steps.
 - a. At the first of the nine points, select the nearest stipe that is within a half square distance of the point in all directions (including outside of the quadrat). For this stipe, measure the length of the longest blade from the sediment to the tip of the blade. Record to nearest millimeter. Record if the longest blade is broken or not by putting an astrick next to blade length.
 - b. Wash the whiteboard off and then label the whiteboard with date, quadrat number and time. Spread the same stipe on the whiteboard so that all blades are visible if possible. Use a bottle of water/spray bottle to wash off the blades and board before taking the photo.
 - c. Take one photo that includes the label and one photo close up to the eelgrass with the blade spread out. These photos will be used later for image classification.

- d. Count the total number of blades connected to the stipe (E.g.: 5 blades in Figure 3)
 Out of the total blade count, record how many of the blades have dark spots, breakage at the tip, epiphytes/bryozoans, or epifuana, respectively. If epifuana or epiphytes are present, be sure to label what type of species they are (E.g.: 4/5 have dark spots, 3/5 have turf algae and bryzoan, 5/5 have small brown snails)
 If no stipe within $\frac{1}{2}$ a squares distance in all directions, skip (leave cells on datasheet blank).
- e. Repeat with eight remaining intersection points.

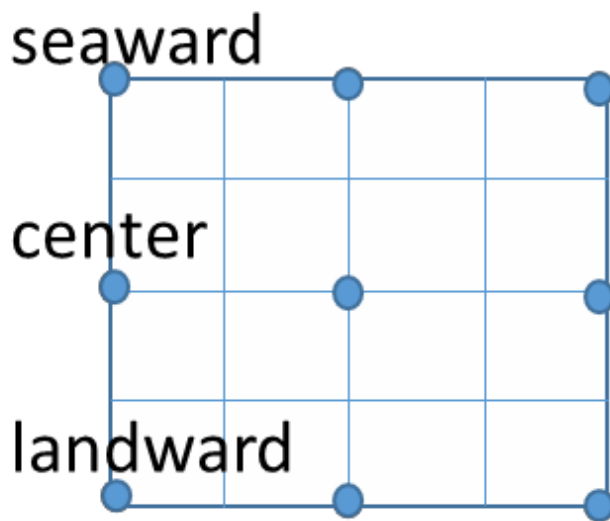


Figure 3 Blade details



Figure 4 Photo for blade condition

Intervals between quadrats (mapping)

10. Anchor the meter tape or have one person hold the end. Continue from the quadrat for 25 meters using the meter tape. Lay the meter tape out following the margin of the water's edge. At each meter between quadrats, score each meter as yes (1) or no (0) for the presence of eelgrass above (exposed) and below (submerged water).

- E.g. Eelgrass present above and below at meter 1 would be recorded as 1 1. No eelgrass above or below would be recorded as 0 0.
 - Note: that its actually hard to keep at the margin's edge, so we usually just go in a straight line and look toward the water edge.
11. Every 25 meters, complete steps 2-9 until 7 quadrats (150m) have been completed. If there is no eelgrass at the next measurement point on the transect, move to the nearest eelgrass within 12 meters (forward or back), which will not necessarily be located in the most dense eelgrass. If there is no eelgrass within that distance in either direction, move to the next 25m point and circle “no eelgrass” next to “**Stipe Counts**” on the datasheet.

Upon returning from the field

Datasheets and record keeping:

1. Check that all fields of the datasheets were completed, site info was filled out correctly and writing is legible.
2. Scan data sheets. Email datasheets out if necessary. Save a scanned copy of each datasheet here:
 - a. N:\Projects\Restoration\Eelgrass\Monitoring\2016 Monitoring\Scanned Datasheets
 - b. Save it as Site Name Date Quadrat (example: Reference Bed 02052016 Q64)
 - c. File original datasheets in Eelgrass datasheet folder. Make a photocopy to bring into the field when necessary.
3. Record field notes in the field notes Word document:
 - a. Include: date, site, survey type (intertidal/subtidal), crew, tide and a brief description of what was done. Include comments such as if that was a good tide for that site or additional gear to bring for future surveys.
4. Download photos from camera. File photos in appropriate MBNEP file, upload photos to dropbox for CalPoly.
5. Rinse and put away gear (including GPS and field camera)

Site Descriptions

Coleman Beach: (MBNEP Site code: ECO)

- Direction of survey: S → N
- Lat/Long of start:
- Lat/Long of end:
- Survey type: intertidal and subtidal
- Launch point: Coleman Beach Parking lot
- Best tide window for subtidal surveys:

This bed is the longest, most continuous bed and consistently has the best visibility. It seems to have an even amount of subtidal and intertidal eelgrass.



Figure 5 Start of Coleman surveys. Start point is at the edge of the building and extends north 150 m, ending at the elbow in Coleman Beach.

North Sand Spit: (MBNEP Site Code: ENS)

This bed has also been referred to as Tidelands on past datasheets.

- Direction of surveys: N → S
- Lat/long start: 35.368839, -120.858842
- Lat/long end: 35.36817, -120.85741
- Survey type: intertidal and subtidal
- Launch Point: Target Rock for north end. This site can also be accessed by launching from the NEP office and walking down the sandspit.

This bed starts across from the T-Pier and continues south. This is a patchy bed. There are side channels throughout the bed that punch holes in the bed. There is a steep drop off in depth, so the bed is narrow, with more intertidal than subtidal eelgrass. The 150m mark is across from the South T-pier.

Reference Bed: (MBNEP Site Code: ERE)

- Direction of surveys: N → S (subtidal), S → N (*intertidal*) unfortunately the subtidal and intertidal were completed in opposite directions)
- Lat/long start:
- Lat/long end:
- Survey type: intertidal and subtidal
- Launch Point: easiest from Tidelands Park steps, could also be done from MBNEP office.

This site has mid-range visibility. Eelgrass becomes pretty patchy in this section of the bay. This is the shortest bed. Intertidal and subtidal both extend for 65 meters. There are patches that continue south, but they are farther than 50m apart and subtidal portions are difficult to find under most conditions. On very calm days with good visibility or on very low tides they are easier to find.

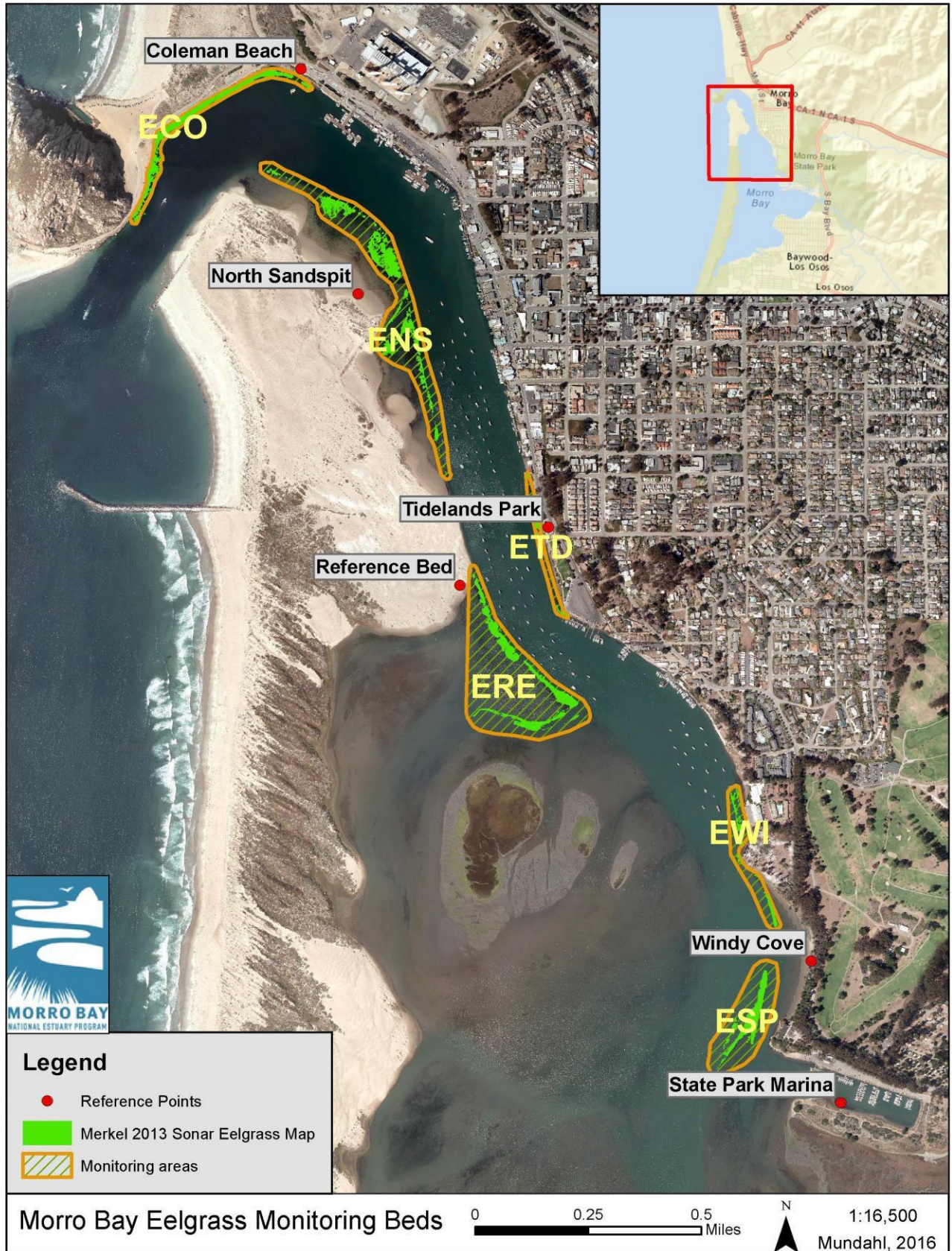
Windy Point (MBNEP Site Code: EWI)

- Direction of surveys: N → S
- Lat/long start: 35.351251, -120.845271
- Lat/long end:
- Survey type: intertidal and subtidal
- Launch Point: easiest from Tidelands Park steps, could also be done from MBNEP office.
- Description of start point: Start point is just north of the concrete stairs, but the nesting point.

State Park Marina (MBNEP Site Code: ESP)

- Direction of surveys: N → South
- Lat/long start: 35.34782, -120.84490
- Lat/long end:
- Survey type: subtidal only
- Launch Point: State Park Marina or Windy Cove.
- Description of start point: 50m north of the north red buoy.

This bed is subtidal only. It is best surveyed on a low tide (around -0.5ft). Most of the bed lines the navigation channel and visibility is very poor, so diving is not recommended. We surveyed this at a -0.5ft tide and weren't able to reach the bottom of the eelgrass when just floating on the surface. This is not a good bed to survey on windy days because it lines the navigation channel. It is hard to keep kayaks out of the navigation channel. The 0 quadrat is 50m north of the red buoy.



APPENDIX J: MORRO BAY NATIONAL ESTUARY PROGRAM
MULTI-SPECTRAL EELGRASS MAPPING GROUNDTRUTHING METHOD

Multi-spectral Eelgrass Mapping Effort: Ground Truth Methodology

Goals: To support the multi-spectral eelgrass mapping effort to create a bay-wide map of intertidal submerged aquatic vegetation, the Estuary Program works with a contractor to collect the imagery and create the vegetation classification. To support this effort, Estuary Program staff collect data on the ground to verify the classification scheme. Data is collected prior to the flight to provide groundtruthing data to support create of the classification. Data is also collected following the flight to provide confirmation on areas that the contractor is having trouble classifying.

Equipment: Data was collected on a EOS Arrow Gold using ESRI Collector software.

Locations: These vary by year and are dictated by the contractor's needs.

Methodology:

- All data points are collected on foot, though a paddleboard can be to access different locations within the bay. All points are collect on foot to ensure adequate time for the GPS unit to stabilize and collect as accurate of location points as possible. Because of this, low tide windows and areas shallow enough to stand in are targeted.
- Areas that needed groundtruthing for the previous aerial flight are targeted, as well as additional locations requested by the contractor. Areas with the following characteristics are targeted: Areas with mixed vegetation; areas with discrete substrate or vegetation; and areas with different densities of eelgrass (dense, medium, patchy).
- At each location, the GPS unit is held in place until it stabilized to submeter accuracy (typically around 20 cm). Dominant vegetation and secondary vegetation types are recorded. If the point is not mixed, the dominant vegetation is also entered as the second dominant vegetation. Georeferenced photos are also taken in some locations.
- Additionally, ground truthing at some locations consist of landscape photos with written descriptions of the area rather than specific coordinates, as requested by the contractor.

APPENDIX K. MORRO BAY NATIONAL ESTUARY PROGRAM EELGRASS RESTORATION
MONITORING PROTOCOL

Gear:

- 1m x 1m plot with 16 intersections
- Meter stick
- Meter tape and metal garden stake
- Camera
- Datasheet on Rite-in-the-Rain paper
- GPS Unit
- Site maps and diagrams

Safety Protocol:

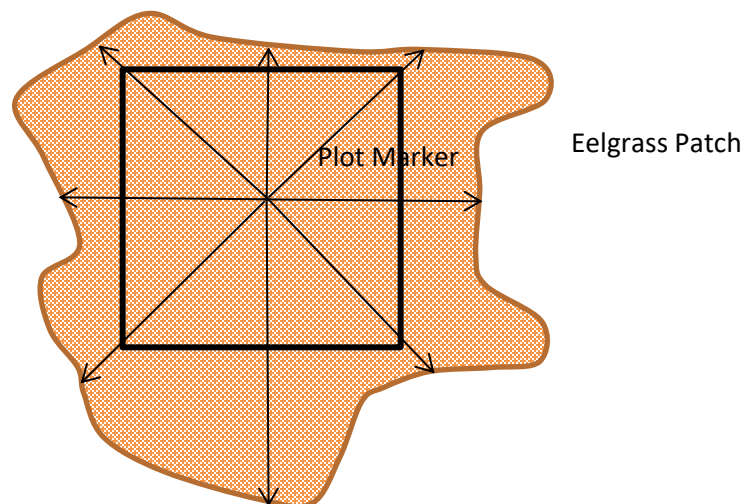
As some sites require paddleboard/kayak/boat, check the weather before leaving the office and verify conditions are safe for paddling. Notify designated person of the float plan for the day, including launch location, number of people going out and estimated return to the office. A USCG approved personal floatation device (PFD) must be worn at all times when using a SUP/Kayak. General guidelines are winds must be less than 10kts for paddling. Refer to Morro Bay National Estuary Program Kayaking/SUP safety protocol for additional information

Field Protocol:

Photos: take an overview photo of each transplant site (all plots in one photo) facing north and south.

At each plot:

- Photo: Lay down the quadrat with the PVC in the south west corner and take an overview photo of the plot. Do this before moving anything out of the way. Take the photograph as vertical as possible, including the entire quadrat frame. Try to avoid shadows and reflections in the photograph. Record photo ID's on the datasheet.
- Plot Expansion Measurement: We will estimate the growth of the plot. Anchor a measuring tape to the center of the plot with a metal garden stake. Using the below guide as a reference, measure the distance from the center of the plot to the edge of eelgrass growth at 8 locations. You can walk around the outside of the plot to quickly get the 8 measurements.



- Point Intersect: At each string intersection and quadrat corner (25 points total), record the presence/absence of eelgrass, macro algae and invertebrates. Also give a qualitative percent cover estimate for each. Do this before moving things out of the way.
- Shoot Count: Count the total number of shoots in each of the sixteen sub squares. This can be scaled down to just four sub squares for plots with lots of new growth.

At each rebar piece:

- Photo: Take the photo of the rebar piece and plot marker as vertically as possible. Try to avoid shadows and reflections in the photo. Record photo IDs on the datasheet.
- Shoot Count: Count the total number of shoots and immediate adjacent to the piece. This may not be practical if the rebar pieces are doing well.

Observations: Note if any shoots are flowering, and if so, record the stage.

Measure the length of the pvc pole (this will show sediment deposition or aggradation over time).

Datasheets and record keeping:

1. Rinse and put away gear (including GPS and camera)
2. Check that all datasheets were completed and all necessary info was filled out correctly and writing is legible.
3. Scan datasheet.
4. Download photos from the camera and file accordingly with the site name, plot number, and date.



MORRO BAY
NATIONAL ESTUARY PROGRAM

0 0.5 1 Miles

May 2020



Restoration Monitoring Datasheet- 12 Months- Plots & Rebar at Site

Site: _____ Date: _____ Surveyors: _____ Start Time: _____ End Tide: _____
 Photo ID of Site (Looking North): _____ (Looking South): _____ Tide: _____

Plot Name	Photo ID	Length of PVC pole	Additional Notes:
Plot 1			
Plot 2			
Plot 3			
Rebar 1			
Rebar 2			
Rebar 3			

Plot Expansion: Measure plots (in meters) from center of plot frame; measure rebar pieces from center of "patch".

Plot Name	1	2	3	4	5	6	7	8	Average
Plot 1									
Plot 2									
Plot 3									
Rebar 1									
Rebar 2									
Rebar 3									

Percent Cover: Estimate percent cover of each of these parameters for each plot, not rebar pieces.

	Eelgrass (LIVE blade or stipe)	Gracilaria (Red and stringy)	Ulva (green and leafy)	Fauna (indicate type: e.g. sand dollar)
Plot 1				
Plot 2				
Plot 3				

Shoot Counts: Count total number of shoots present in four sub squares of the quadrant.

Plot 1	Plot 2	Plot 3
1 st Square-	1 st Square-	1 st Square-
2 nd Square-	2 nd Square-	2 nd Square-
3 rd Square-	3 rd Square-	3 rd Square-
4 th Square-	4 th Square-	4 th Square-
Average		

Only If Extra Time...

Rebar 1	Rebar 2	Rebar 3

Observations (black spots on blades, seeding of blades, drying out of blades, grazing of blades, etc.):

APPENDIX L. MORRO BAY NATIONAL ESTUARY PROGRAM
SHOREBIRD SURVEY PROTOCOL

Morro Bay Volunteer Monitoring Program

Shorebird Survey Protocol

(from Point Reyes Bird Observatory – Pacific Flyway Project)

Shorebirds can be counted more easily than many birds due to their use of tidal flats and other open habitats. Further, shorebirds generally move shoreward as the tide rises, affording land-based observers an improving view until the flood tide forces the birds to alternative high tide habitat. However, due to their habits of aggregating, sometimes in very large, mixed-species flocks, and flying (often unexpectedly during a count!), skill and experience are necessary to obtain accurate counts.

COUNTING METHODS

Counting methods vary, depending on the number of birds, their edginess, and census conditions. The number of birds in a flock can be obtained by:

1. Counting birds individually.
2. Counting birds in groups of some number
3. Estimating the number of birds in a flock without actually counting them.

The first two methods are always more accurate and preferred over the third, which should be used only for preliminary observations of flocks or to salvage an interrupted count.

Counting birds individually: If there are only a few birds in an area, (less than 100 total), they should be counted individually.

Counting birds in groups of some number: If you are counting larger flocks, say 200 birds, begin at one end of the flock - count the first 10 birds, then the second 10 birds, continuing until you can easily see the birds in groups of 10. At that point, start counting by 10-bird groups. This method can be generalized to counting by 20-bird, 25-bird, 50-bird, or even larger groups, for larger flock sizes. This method can also be used for counting birds in flight.

Estimating the number of birds: It is wise to initially estimate numbers of all the birds quickly, to determine the relative proportions of the most common species, to roughly determine the numbers of the less common species, and to determine whether the species are segregated or homogeneously mixed throughout the flock. Then, if the actual count is interrupted, some reasonable estimates of numbers will be possible. Making a rapid estimate of the number of birds may be the only way to salvage a count in which birds fly away before they are counted.

SPECIES IDENTIFICATION

1. Try to identify all the birds (except dowitchers) to species.
2. Whenever possible, allocate unidentified shorebirds to species groups (such as Least Sandpiper or Western Sandpiper) rather than just listing them as unidentified large or small shorebirds.

On the census form we have provided species categories that might commonly be used. Four groupings: yellowlegs spp., dowitcher spp., phalarope spp., and Least or Western sandpiper are self-explanatory. The plover species would include Semipalmated Plover, Snowy Plover, and Killdeer. The large sandpiper grouping could include any combination of Willet, Marbled Godwit, Greater Yellowlegs, Long-billed Curlew, or Whimbrel. If you use any of these categories please indicate which species you believe could have been involved. Write in any species not on the census sheet in one of the empty columns at the bottom of the census form and provide details on the sightings.

APPORTIONING THE COUNT OVER SEVERAL SPECIES

1. Count out each species separately.
2. Count all species together.
3. Count several species together.

Counting each species separately: Counting each species separately is time consuming if there are many species to count but can be the least confusing method if there are few abundant species per counter.

Counting all species together: If the group of shorebirds comprises primarily one species, with a few individuals of other species, it is often easiest to count the total number of birds, then to count out the rarer species and subtract their numbers from the total, to get the number of the common species. An assistant, recording for the counter, can aid in this process by either counting the rarer species in the flock or by making the subtractions on the recording form. In cases where there are tens of thousands of shorebirds and several species are very abundant, it may be necessary to count all species together and to sample species composition at many places in the flock in order to determine the proportion of the total made up by each species.

Counting several species simultaneously: In a mixed flock, it is often fastest to make a combined count of similarly sized species. For example, say there are six species present: Dunlin, Western Sandpiper, Least Sandpiper, Willet, Marbled Godwit, and Long-billed Curlew. It might be easiest to count the first three species together, by writing down the number of the less common species as they are encountered and keeping a running tally of the most common species in your head until the flock has been scanned completely. Similarly, the Willet, Marbled Godwit, and Long-billed Curlew could be counted together. Counting several species simultaneously can often be done most effectively if there is both a counter and a recorder. The counter can give the recorder the counts of the less common species as they are seen but accumulate the count for the most common species until the flock is completely counted.

WHAT TO DO WHEN BIRDS FLY DURING A CENSUS

Movement of birds during a census creates some particularly troublesome problems. Our basic strategy is to count the birds before they move from tidal flat areas. Counts at shoreline segments should not take longer than about 1 hour so that the overall amount of bird movement is minimized. It is also important that counters in the same general area of the Bay are counting at the same time. Using the guidelines below, we hope that you will be able to best judge whether or not you should count certain birds.

1. **If you are covering a shoreline segment:** Count birds that fly into and stay in your area during the first hour of your count. You should count birds that fly out of your area only if they are leaving for non-tidal, high tide roosts (such as salt ponds). If a bird (or flock) moves to another tidal flat area and it is less than an hour since your starting time for that area and there is still a counter visible to you in the area into which the bird(s) moved, you should not count the bird(s) for your area. Alternatively, if a bird (or flock) moves into your area in the first hour of your count, you should count them, unless it is apparent to you that they have been counted by an observer in an adjacent area and the observer did not see them move to your area.
2. **If you are covering a non-tidal area at the same time that shoreline segments are being covered:** Only count flying birds in your area that leave for other tidal areas. If birds leave your area for the tidal flats (this shouldn't happen too often) do not count them if it is less than an hour since tidal flat counts have begun. Do not count birds that fly into your area once the shoreline segment counts have begun.
3. **For both tidal and non-tidal sites:** If you do not know where departing birds go, note them separately on your form with the time they leave and general direction that they go. Make similar notes on arriving birds. If birds fly by your area without stopping, note them separately with the time and direction of flight, if possible. Do not include any of the birds noted separately in the totals for your area; we will decide after the census if they have been counted by any other team. However, the best decisions on whether or not a flock should be counted are often most easily made in the field.

FOR FIRST TIME CENSUSERS: WHAT YOU 'LL WANT TO REMEMBER TO BRING

- Binoculars
- Telescope (if you have one)
- Wrist watch
- Letter or card indicating permission to restricted land (if applicable)
- Recording pad (use census form for final tally only)
- Pencils (mechanical pencils don't need to be sharpened if they break)
- Rubber boots (your letter will indicate if needed for your area)
- Warm layers of clothes, water, sunscreen
- These forms and instructions
- A field guide (although there usually isn't time to refer to it during the census)

Morro Bay Volunteer Monitoring Program Shorebird Census Location Information

See Map for reference. All boat sections concentrate the on the main body of the bay excluding the salt marsh and Chorro/Los Osos creek outlets. Those areas will be covered from the shore.

1. Section 1: Land Section. Use scope near Morro Rock, T-Piers, and several street-end viewing docks along the Embarcadero.
2. Section 2 & 3: Boat Section. Depart from embarcadero. Sections 2 & 3 are usually completed together. Concentrate on Grassy Island & the shore of sand spit. The boundary of Section 4 starts at the southern tip of Grassy Island and runs from the sandspit east to Fairbanks Point.
3. Section 4: Boat Section. Depart from State Park Marina. South of Grassy Island. Concentrate on shore of sandspit and any exposed mudflats. The southern boundary of Section 4 runs east to west from just north of the Marina to the sandspit.
4. Section 5: Boat Section. Depart from State Park Marina. Concentrate on sand spit and any exposed mudflats south of the Marina to approximately the northern tip of Pasadena Point. The southern boundary of Section 10 runs east to west from Cuesta Inlet to the sandspit.
5. Section 6: Land Section. Use scope around State Park Marina (to the south side) and east along the Salicornia (pickle weed) as far as necessary.
6. Section 7. Land Section. Can be done with Section 6 from the same vantage points. Boundary between Sections 6 & 7 is approximately along the Chorro Creek channel.
7. Section 8. Land Section. Use scope from Bush Lupine Point in Elfin Forest. Park at north end of 13th Street in Los Osos. If vantage point further east is necessary, go along boardwalk to Siena's View, the next point at which the boardwalk affords a bay view.
8. Section 9. Land Section. Use scope (if needed) from pullout on Pasadena Drive, from Baywood Pier, Sweet Springs Nature Preserve (enter at Broderson St.), & further west at public access near intersections of Mitchell Dr. & Doris St.
9. Section 10: Boat Section. Depart from Cuesta Inlet. Concentrate on sand spit and any exposed mudflats south to approximately Cuesta Inlet. The southern boundary of Section 10 runs east to west from Cuesta Inlet to the sandspit.
10. Section 11. Land Section. Starting from west end of Butte Dr. checking near shore, then when closer to Shark's Inlet (back corner of bay), look across to shore of sand spit. Boundary with Section 10 is north end of a large bare dune.
Note: If sand spit side of Section 10 is done on foot, the Section 11 team can do the portion of Section of 10 that is Cuesta by the sea shoreline.
11. Section 12. Land Section. Use scope from bay side of South Bay Blvd. across from Turri Rd. Further north access is available just south of Chorro Cr. Bridge.

12. Section 13. Land Section. Ocean side of sandspit. Use Montana de Oro access. Census the beach while walking north, then walk back.

Note: Can arrange pick up with boat on north sandspit rather than walking back.

13. Sections 14/15. Land Section. Walk from Morro Rock to North Point (or vice versa). To avoid wading Morro Creek, walk north from Morro Rock to the creek, walk back and drive to north side of creek (Highway 41 west to the end). Park and walk from north side of Morro Creek to North Point. With two people & a desire to walk less, a car can be dropped off at North Point, return to Morro Rock to begin survey, census and finish at North Point and awaiting car.

We need at least 11 bird survey leaders.

Morro Bay Volunteer Monitoring Program Shorebird Monitoring Datasheet

Location _____ Date _____

Team Members (first, last names) _____

Area _____

Start Time _____ End Time _____

Black Bellied Plover		Western Sandpiper		Great Blue Heron	
Snowy Plover		Least Sandpiper		Great Egret	
Semipalmated Plover		Least/Western sandpip.		Snowy Egret	
Killdeer		(*) Ratio >>		Cattle Egret	
Am. Black Oystercatcher		Dunlin		Green Heron	
American Avocet		Least/West/ Dunlin		Blck-cr. Night Heron	
Greater Yellowlegs		(*) Ratio >>		Virginia Rail	
Lesser Yellowlegs		Baird's Sandpiper		White Pelican	
yellowlegs spp.		Short-bill. Dowitcher		Brown Pelican	
Willet		Long-bill. Dowitcher		Turkey Vulture	
Wandering Tattler		dowitcher spp.		Osprey	
Spotted Sandpiper		Wilson's (common) Snipe		White-tailed Kite	
Whimbrel		Wilson's Phalarope (u)		Northern Harrier	
Long-billed Curlew		Red-necked Phalarope		Red-tailed Hawk	
Marbled Godwit		Red Phalarope (u)		American Kestrel	
Ruddy Turnstone		phalarope spp.		Merlin	
Black Turnstone		Brant		Peregrine Falcon	
Surfbird		Other species (d)		Red-shouldered Hawk	
Red Knot					
Sanderling					

(u) indicates species we suspect will be very uncommon or local

Dowitchers--We suggest the censusers count all dowitchers as **dowitcher spp.** If dowitchers are identified to species we would like some information on how the determination was made.

Mixed Species Groups -- Indicate the possible species and number. For example, if you saw a mixed flock of 155 Willet and Marbled Godwit, you would write Willet/Marbled Godwit and 155. **Whenever possible, indicate proportions of mixed species groups.**

(*) Use these spaces only to record numbers of sandpipers that you are able to separate into Least or Western sandpipers. Count the species together and record the total number. **If possible, list the ratio of the unseparated species.**

APPENDIX M. MORRO BAY NATIONAL ESTUARY PROGRAM'S SUSPENDED SEDIMENT
MONITORING PROTOCOL

Morro Bay National Estuary Program's Suspended Sediment Monitoring Protocol

Background

As the main entity conducting monitoring for the Morro Bay Sediment TMDL, the MBNEP recognizes the importance of tracking sediment loads as best management practices and restoration projects are installed throughout the watershed. This monitoring effort is intended to supplement existing ambient turbidity monitoring (captured via the Water Quality Monitoring effort) by collecting near continuous monitoring data during large storm events. The goals are to monitor suspended sediment concentration, nephelometric turbidity and stream discharge to allow for better calculations of suspended sediment load. This effort employs automated samplers for sample collection at a higher frequency than could be attained by field visits.

Overview

Automated samplers are deployed in the field to collect water samples during storm flows. Permanent equipment shelters have been installed at creek sites to house automated samplers. If a storm is forecasted, the samplers are stocked with empty bottles and programmed to begin sampling to collect samples of a specified volume on a determined frequency. Once all 24 sampler bottles have been filled VMP staff return to the site to swap out the bottles and re-program the sampler if further sampling is desired. Samples are processed in our Water Science Lab located at Cuesta College. Samples are weighed and then poured onto pre-weighed filter. The dried filters provide the mass of sediment contained in the sample and based on the volume of water originally collected, a suspended sediment concentration can be obtained corresponding to various flow levels in the creek.

A. Field Sampling Protocol

1. Bottle Preparation for Sampling

Each site has four assigned bottle sets (A, B C, or D). Bottles are individually labeled with the site code (three letters), set code (A, B, C or D) and bottle number (1 through 24). For example, each bottle has a unique three part ID: CAN-A-13. Each bottle has a unique tare weight, written on both sides of the bottle. There are two extra sets of bottles (site code EXT) that may be used at any of the stations if needed. To prepare bottles for field sampling, follow the method below.

1. Rinse bottles thoroughly with DI water.
2. Fill halfway with DI water, cap, shake vigorously, and pour out DI water.
3. Bottles from especially dirty or contaminated bottle sets should be washed with dishwashing soap and the bottle brush, and rinsed thoroughly with DI water.
4. Place bottles on drying rack over sink.
5. After bottles have rested for 20 minutes, tip forward to pour out water settled in the bottle neck.
6. Place bottles back on rack until completely dried.
7. Place caps into 500 mL beaker, and then fill with DI water and pour out several times.

Once bottles have dried completely:

1. Wipe out sampler base to remove any debris, dirt or water.
2. Gather bottles into matching sets of 24. (Sets are A, B or C for each site).
3. Inspect bottles to ensure that the bottle ID number, tare weight, etc. are legible. If needed, use a Sharpie to refresh these labels.
4. Remove any remaining stickers with prior sampling information.
5. Install bottles in numeric order, with bottle numbers matching numbers stamped on the sampler base.
6. Double check that bottles 3, 15 and 21 are notched into the molds of the base.
7. Fasten the round bottle plate to the base with the three elastic bands.
8. Lightly screw caps onto each bottle.

2. Battery Charging and Installation

There are numerous 12 volt, 32amp hour batteries available for use in the field to power the ISCO samplers. The small black batteries are used primarily for field testing; the large batteries are deployed for continuous sampling.

Charging the 32 amp Battery:

- NEVER charge a frozen battery
 - NEVER set battery on top of charger
 - Never place charger directly above battery being charged. The gases from the battery will corrode and damage the charger
1. Be sure to set the charger to the 12V setting
 2. Place the battery on top of the Styrofoam or wooden block.
 3. Note the polarity of the battery posts by checking the identification marks.
 4. Connect the POSITIVE (red) output clamp from the charger to the POSITIVE (red) post on the battery.
 5. Position yourself as far away from the battery as possible, and then connect the NEGATIVE (black) output clamp from the charger to the free end of the cable (black).
 6. Plug in charger power cord.
 7. Select the appropriate charge rate and battery voltage. The yellow LED light labeled “CHARGING” should light up and the charging process should start.
 8. Check to make sure the charging is on the lowest 2V setting for trickle charging.
 9. Battery charging takes approximately 6 hours.
 10. After charging is done, disconnect the charger using the following steps. First, unplug the charger power chord. Second, remove the output clamp from the NEGATIVE (black) post. Third remove the POSITIVE (red) output clamp from the positive (red) post.
 11. Wash hands each time you finish handling the battery and charger.

3. Calibrate Volume

Check and calibrate volume before each new run

1. Disconnect the pump tube from the bulkhead fitting.

2. Place end of tube over a collection container.
3. From the main screen, go to Other Functions → Manual Functions → Calibrate Volume → enter the desired volume (400 mL) → press the enter button.
4. It will then purge the line and then pump the desired volume. After it has finished pumping measure the volumes delivered and enter the amount when prompted.

4. Filling out the Field Logbook

Upon arrival at the site, start a new record in the field logbook. Each field record should include the following:

1. Current date and time.
2. Staff gauge height (recorded off the staff gauge on the bridge wall)
3. Code for the bottle set being removed
4. Description of samples collected in bottle set being removed. Note if any bottles failed to fill, overtopped, or drew an incorrect volume.
5. Name of battery, how much used
6. If battery needs switching out
7. Code for new bottle set being installed – check that bottles are in order and caps are off
8. Code for new battery being installed
9. Program start date and start time
10. Program sampling frequency (typically 30 or 60 minutes.) Program sampling volume (400ml.)
11. Note if tubing was changed

5. Programming the Sampler

Standard Programming

With standard programming, the sampler purges the sampling line, draws a sample, and then purges the line again after sampling. Only in the extended programming mode does the sampler rinse the line before sampling or conduct retries for sampling.

Each sampler has been pre-programmed (initialized) with a site description, which does not need to be changed. Suction line length has also been entered and should not be changed unless changes are been made to the sampler intake.

When setting a program it is important to keep in mind the duration of the sampling period that can be accommodated by the 24 available bottles and the timing of the anticipated storm flow event. A 30-minute sampling interval will capture a period of about 11.5 hours. A 1-hour sampling interval will capture a period of 24 hours.

Sampling Intervals with a Manual Start

1. Press the power button to turn the sampler on.
2. Arrow to **Program**, press **Enter**

- a. (Selected options will begin blinking when they are selected.)
3. At “Site description change?” arrow to **No**, and press **Enter**.
4. At “Number of Bottles,” arrow to **24**, and press **Enter**.
5. At “Bottle Volume,” type in **1000** mL, and press **Enter**.
6. At “Suction Line Length,” check that lengths are the same as shown below, press **Enter**. (Should not need to be changed except in special circumstances)
7. Line lengths by site are:
CANET: 76
SAN LUISITO: 35
WALTERS: 55
8. At the screen for “Time Paced or Flow Paced” sampling, select **Time Paced**, and press **Enter**.
9. At “Time Between Sample Events,” type in the **Number of Hours and Minutes**, and press **Enter**.
10. At “Sequential, Bottles/Sample, Samples/Bottle” select **Sequential**, and press **Enter**.
11. At “Run Continuously?” select **No**, and press **Enter**.
12. At “Sample Volume” type in **400** ml, and press **Enter**.
13. At the next menu “No Delay to Start, Delayed Start, or Clock Time” scroll to **Clock Time**, and press **Enter**.
14. At “First Sample at --:--“, type in the start time, and press **Enter**. Time should be entered as time on a 24-hour clock. (Take note of the clock time displayed on the sampler, the sampler cannot begin sampling in the past!)
15. At “Select Days” scroll through days, press **Enter** to select or un-select. Scroll to **Done**, press **Enter**.
16. The final screen should show the time the program is scheduled to start (top line) and the countdown to the first sample (bottom line). Double check that the start time is consistent with when you want to begin sampling, and that the countdown is accurate.

Setting the Clock

1. Synchronize sampler clock to time and date on a cell phone. Remember to change clocks with daylight savings time.
2. Go to **Other Functions**, and press **Enter**.
3. Go to **Maintenance**, and press **Enter**
4. Press **Enter** again to select **Set Clock**.
5. Enter **Hour**, and press **Enter**. Enter **Minutes**, and press **Enter**. Enter **Day**, and press **Enter**.
Type in Months, and press **Enter**. Type in the **Year**, and press **Enter**.
 - a. Hours must be set using the 24 hour clock. If it is after 12pm, add 12 to the standard time to get the 24 hour time. (4:00PM + 12 = 16:00)
6. Press **Stop** to return to the main menu.

6. Sample Preparation and Handling

Resetting the Pump Counter:

After changing the pump tube, you must reset the pump counter. Go to **Other Functions** → **Maintenance** → **Pump Tube Alarm**. The sampler displays a screen to give you the current pump-count information. The first line will show the pump counts tallied since the counter was last reset. The second

line shows the current alarm setting. To reset the counter to zero, press YES. Always reset the counter after replacing a tube. Select **NO** if just checking the current count. Press **STOP** to return to main menu.

In the field:

1. Immediately upon removing sampler base, cap bottles tightly.
2. Confirm bottles are in the correct order. Note any that are out of order in the log book
3. Fill out the log book with all necessary information (provided in the log book)
4. Note in the field log any bottles where the volume collected appears to be inaccurate.
5. Bottles should be transported to the lab the same day they are removed from the field.

At the lab:

1. Remove capped bottles from the sample base.
2. Check the water level of each bottle, note bottles with suspect volumes.
3. **Clean and dry outside of each bottle**, and look for floating debris (leaves, sticks or algae).
4. Remove the bottle cap.
5. Set bottle on high capacity scale.
6. Record the weight of the sample on the appropriate RITR label.
7. Re-cap bottle tightly and remove from scale.
8. Apply the RITR label(s) to the bottle.
9. Bottle is ready for storage. Bottles may be stored in the refrigerator, or in one of the grey storage bins in the lab.

Sample Storage at the Lab

If samples will not be processed immediately:

1. Make sure sample is completely and clearly labeled and weighed in. Do NOT enter the information for the bottle into the Sediment Data Management System until you begin filtering the samples.
2. Re-cap tightly and place in the refrigerator or storage container.
 - a. Samples with a high sediment/solids concentration should preferably be stored in the refrigerator.
3. Fill out all necessary information onto the white board

Sample Handling QA

To ensure sterility and accuracy, one blank sample should be run for each bottle set that comes in from the field. The blank bottle is filled with DI water, stored at the lab, and filtered alongside samples in the field bottle set.

To prepare a QA Blank:

1. Locate one-liter Nalgene bottles from the BLK bottle set at the lab or fill an empty and clean bottle from that bottle set.
2. Fill the bottle approximately halfway with DI water.
3. Weigh the un-capped bottle on the high capacity scale.
4. Create a RITR label noting the QA Blank sample, the date it was prepared, and the corresponding field bottle set.
5. Record the weight of the sample on a RITR label.
6. Re-cap bottle tightly and remove from scale.
7. Apply the RITR label(s) to the bottle.
8. Place bottle into storage with the appropriate field bottle set, or continue with login for sample processing.

7. Turbidity Measurements

Selected samples may be measured for turbidity. Measurements are typically conducted on 12 bottles in a 24 bottle set. **Turbidity measurements can only be conducted on samples that have already been weighed in, and have been brought to room temperature to avoid error due to condensation.**

Gloves should be worn at all times when measurements are being conducted. It is not advised to process samples the same day that turbidity is measured. The sample will filter slowly and use more filters than normal.

1. Check to make sure the instrument is in Automatic range by pressing the RANGE button.
2. Check to make sure that Signal averaging is turned on by pressing the SIGNAL AVG button.
3. Make sure sample bottle is tightly capped.
4. Vigorously shake sample bottle to re-suspend sediment. Allow sample to rest for 30 seconds after mixing.
5. Over a clean glass beaker, pour 30ml of sample into the sample cell to the line indicated on the glass sample cell. Take care to handle the sample cell by the top. Pour any spilled water back into the bottle. Use DI water to thoroughly rinse the beaker into the bottle.
6. Cap the sample cell, wipe off any water spots or smudges with KimWipes.
7. Apply a thin bead of silicone oil, and use the velvet oiling cloth to wipe the oil over the cell.
8. Place the sample cell in the instrument, aligning the arrow on the cell to the notch in the instrument compartment.
9. Close the lid of the compartment.
- 10. The unit will begin measurements immediately, record the result indicated within the first 10 seconds of measurement. Turbidity readings will decline as the sediment settles out of the sample over time.**
11. Record the result on a new RITR label, and place on the bottle next to the sample information label.
12. Pour the water from the sample cell back into the sample bottle. Rinse the sample cell with DI water, and pour the rinseate into the sample bottle. *It is important to recapture all of the suspended sediment from the original sample for processing. It is also important not to add any weight (DI water rinseate) to the bottle unless it has already been weighed in! Water weight is important- but once the sample has been weighed in, extra weight from DI rinseate water won't*

be factored into the lab record. (So don't re-weigh after putting the rinseate DI water into the bottle.)
13. Recap the bottle and place into storage container.

B. Laboratory Protocol

1. Equipment Accuracy and Calibration Checks

All equipment should be checked and re-calibrated on a weekly basis. Deionized

water:

1. The deionizing filter for the building needs to be replaced periodically. Check the conductivity of the deionized water weekly to ensure that the filter is working correctly.
2. Rinse a 500 mL beaker with DI water and pour out.
3. Fill a 500 mL beaker with DI water.
4. Insert the conductivity probe, and allow reading to settle.
5. If DI water conductivity is over 100 uS/cm, run the tap an additional 1-2 minutes, collect and test a new sample.
6. Once DI water is reading below 100 uS/cm, fill all squeeze bottles that will be used during sample processing.
7. Record conductivity on log sheet.
8. If conductivity is below 100 μ S/cm, it cannot be used. Cuesta needs to be notified to change their filters.

Oven Temperature

1. Flip the middle switch to 'Power' to turn on the oven.
2. Flip the lower switch upwards to 'Heat'.
3. Set the dial thermostat to 225° F.
4. The red (Power) and orange (Heating) indicator lights on the front of the oven should illuminate.
5. When the orange indicator light has gone off, open the oven door and record the temperature on the internal thermometer.
6. Oven temperature should be between 103° and 110° Celsius.
7. Make adjustments to thermostat dial as needed.

Hach 2100AN Turbidity Meter

This instrument is designed for long-term stability and minimal need for recalibration. It is recommended that recalibration be performed at least once every three months, or more often if experience indicates a need. **The meter should be left on 24 hours a day if the instrument is to be used regularly.** To guarantee accurate calibration, use only the formazin standards issued by Hach.

The formazin calibration standards have a long shelf life, but must be stored properly. Standards should be stored in their respective kit with cover in place, out of direct sunlight. For long term storage, refrigeration at 5° C is recommended, do not store above 25° C.

1. Make sure the meter has been on for at least 1 hour.

2. Shake the standard vigorously for 2-3 minutes to re-suspend particles. Do NOT shake the < 0.1 NTU standard, only the 20 NTU, 200 NTU, 1000 NTU, 4000NTU, and the 7500 NTU standards.
3. Allow standard to stand undisturbed for 5 minutes.
4. Gently invert the cells 5 to 7 times.
5. Prepare the sample cell for measurement by applying a thin film of silicon oil and wiping with the velvet cloth.
6. Let the cell stand for one minute after oiling.
7. Press the **CAL/Zero** button. The small green LED digits in the mode display will flash “00”.
8. Insert the <0.1 ntu standard cell, align the orientation marks on the cell and chamber. Close the lid and press the **ENTER** button to begin calibration.
9. The meter will count down from 60 to 0, read the sample and calculate the correction factor.
10. The display will show the 20.00 NTU value on the screen. Insert the 20.00 NTU standard with correct alignment and press the **ENTER** button.
11. The meter will count down from 60 to 0, and display the will show the value of the next standard required.
12. Repeat these procedures sequentially for the remaining standards as the meter prompts.
13. After reading the final standard (7500 NTU), press the **CAL/ZERO** to accept the calibration.

Troubleshooting

Error Code	Description	Solution
ERR01	The turbidity of the dilution water is greater than 0.5 NTU.	Start the calibration again with lower turbidity dilution water. <i>Note: Ignore ERR01 when the sample cell diameter is less than 25 mm. Push UNITS/Exit to go back to measurement mode</i>
ERR02	<ul style="list-style-type: none"> • Two calibration standards have the same value. • The difference between two calibration standards is less than 60.0 NTU. • The turbidity of Standard 1 is too low (less than 10 NTU). 	<ol style="list-style-type: none"> 1. Inspect the preparation of standards. 2. Do the calibration again. <i>Note: Ignore ERR02 when the sample cell diameter is less than 25 mm. Push UNITS/Exit to go back to measurement mode.</i>
ERR03	Low light error	<ol style="list-style-type: none"> 1. Put the sample in the instrument again. 2. Make sure that the lamp light is on. 3. Make sure that an object is not in the light path. 4. Do sample dilution if necessary.
ERR04	Memory malfunction	<ol style="list-style-type: none"> 1. Turn the instrument off and then back on. 2. Contact Technical Support if the error occurs again.
ERR05	A/D is over the range	<ol style="list-style-type: none"> 1. Make sure that the light shield is closed. 2. Contact Customer Service if necessary.
ERR06	A/D is under the range	<ol style="list-style-type: none"> 1. Make sure that no object is in the light path.

		2. Contact Customer Service if necessary.
ERR07	Light leak	1. Make sure that the cover for the sample cell compartment is closed. 2. Turn the instrument off and then back on.
ERR09	Printer time out error	1. Make sure that the external printer is connected correctly. 2. Make sure that the external printer is selected (online).
ERR10	System voltage out of range	1. Turn the instrument off and then back on. 2. Contact Customer Service if the error occurs again.
ERR11	System loop test error	1. Turn the instrument off and then back on. 2. Contact Customer Service if the error occurs again.

Turn Ratio on: Ratio on mode decreases the effects of light absorbing particles, color, absorbance and high turbidity interferences.

Ratio on provides very good linearity, calibration stability and a wide measurement range. Ratio on helps correct for interference when color is present in the sample that absorbs at the wavelength of incident light. The manufacturer recommends that Ratio on be used for most measurements. Ratio must be on to measure samples greater than 40 NTUs (268 nephelos or 9.8 EBCs)

Turn the instrument on 30 minutes (Ratio on) or 60 minutes (Ratio off) before measurement. Keep the instrument on 24 hours a day if the instrument is used regularly.

When automatic ranging is selected, the display flashes 9s when the sample is greater than the maximum range of the instrument. The display flashes 9s when Ratio is off and the measurement is greater than 40 NTUs (268 nephelos or 9.8 EBCs). Turn Ratio on to increase the range.

When automatic ranging is selected, the display flashes all 0s when the measurement is less than the range of the instrument or a negative value. Calibrate the instrument.

Calibrate the turbidimeter at least every 3 months or as specified by the regulating authority when data is used for USEPA reporting. The instrument is ready for calibration 60 minutes after start-up. Keep the instrument on 24 hours a day if the instrument is used regularly.

Scales:

The scales need to be warmed up before it is used, the longer the better. Once hour is the minimum and two hours is ideal. If the scales are being used frequently (weekly) it is better to leave it on all the time. If it is used less frequently (monthly) then it is better to turn them off in between uses. During times of intensive use, the should be left on.

Temperature fluctuations: fluctuations in temperature affect drift on the scale (even 2-3C) can affect it.

To use the scale, put an item on the scale and wait for it to stabilize. Count to five, then record that value. If the item sits on the scale for too long, the weight will continue to change due to the mechanics of the scale. Don't leave anything on the pan for too long as it will throw off the calibration of the unit. Place items on the scale gently.

Fine Scale Balance

The fine scale balance should be checked daily for accuracy before weighing in any filters (new or used). The scale should be calibrated when the temperature in the room has stabilized. If the oven will be used concurrent with the scale, allow the oven to warm the room for 15 minutes before calibrating the scale. It is also important to watch the scale for signs of drift when weighing for an extended period of time. If the weight of the scale drifts more than 0.0002 grams when closed and stabilized, recalibration may be necessary. Calibrate weekly during intensive times of use, or every time during low times of use. Make sure the scale has been on for 1-2 hours first.

1. Position the scale so that the glass doors will open fully to enable weighing the samples.
2. Make sure the bubble level in the back left corner of the scale is centered. If it isn't, use the dials located on the front sides of the scale to level the scale.
3. Turn the scale on. The scale display should fill in completely to indicate a self-check.
4. Make sure the surface of the scale is clean and all of the glass sides are closed.
5. Tare the balance by pressing the '**ZERO**' button. The scale will beep when zeroing is complete.
6. Remove the 'Fine Balance Weights' (in the grey box) and scale calibration log book from the drawer underneath the scales.
7. Scale calibration weights should only be handled with gloves. It is preferable to use the plastic tongs to transfer the fine calibration weights from the box to the scale.
8. When the scale has zeroed and stabilized, press the **CAL** button to begin calibration.
9. Place the indicated calibration weight in the center of the scale and gently close the glass doors (50.0000 grams).
10. When the scale has stabilized it will beep and display the calibration weight with weight units.
11. Remove the calibration weight and replace it in the grey box. Perform several weight checks with the smaller weights in the calibration weight set. Perform weight checks with the 30.0000, 20.0000, 5.0000, 2.0000 gram weights in the set.
12. While testing with calibration weights, record the time and date of the test, weight of the calibration weight, and scale reading in the scale logbook.

The scale capacity is 70.0000 grams, do not exceed this capacity when performing weight checks.

High Capacity Balance

The high capacity balance should be checked for accuracy before weighing in each round of samples. The calibration weights are kept in three separate black plastic boxes in the drawer underneath the scales.

1. Make sure the bubble level in the back left corner of the scale is centered. Adjust the front feet of the scale to relevel if needed.
2. Turn the scale on. The scale display should fill in completely to indicate a self-check.
3. Make sure the surface of the scale is clean and that there is no water on the metal plate.
4. Tare the balance by pressing the 'ZERO' button. The scale will beep when zeroing is complete.
5. Remove the Gross Scale Weights from the drawer. There are separate black boxes for calibration weights of 200.00g, 300.00g and 500.00 g.
6. Wear gloves for moving the calibration weights from the storage boxes to the scale.
7. When the scale is stable, press the CAL button to begin calibration.
8. Place the indicated calibration weight on the center of the scale (500.00 grams)
9. After calibration, the calibration weight is displayed with weight units.
10. Remove the calibration weight.
11. Perform weight checks with the smaller weights in the calibration weight set. Perform weight checks with the 200.00 and 300.00 gram weights. Do not exceed the capacity of the scale: 810grams.
12. When testing with calibration weights, record the time and date of the test, weight of the calibration weight, and scale reading in the scale logbook.

If the scale is left on for an extended period or bumped off balance, it may drift. If the scale drifts by 1.2 or more, clean and recalibrate the scale.

Annual Scale Maintenance and Calibration

The High Capacity Balance and Fine Scale balance are checked and calibrated annually by an independent calibration firm.

Desiccator Cabinet

At the beginning of each lab session, check the desiccant in each desiccator. Desiccant may need to be re-charged in the oven if the ambient humidity exceeds 15%. (Alternatively, more fresh desiccant can be added to the cabinet for a short-term fix.)

Check the humidity indicator card in both desiccators at the beginning of each lab session. The indicator circles will change from blue to pink if the humidity of the cabinet increases, To recharge Silica Gel Dessicant, bake at 105-110°C until beads turn deep orange. Beads are green when they are "wet". To bak Drie-Rite desiccant crystals, spread the desiccant into baking pans and heat to 230°C (450° F) for one to two hours and crystals are blue.

Upon removal from the oven, desiccant should be placed immediately into the cabinets or storage containers. Loosely cap storage containers until desiccant has cooled (15-minutes) then screw the cap onto the container.

The desiccator cabinet is very sensitive to being opened on humid days. Even opening it for a few seconds might impact the color of the indicator circles. When possible, minimize how long the door is open and keep a close eye on the indicator cards. Add more desiccant if necessary.

There are two cabinets – one for prepped, clean filters and one for used filters (filters with sediment on them). Do not cross contaminate the cabinets and never put “wet” items into the cabinets. Only put items that have properly dried in the oven in first.

Vacuum Carboy

1. Empty the vacuum carboy frequently when processing samples.
2. The carboy should be emptied when the water depth in the carboy reaches the label marked on the side.
3. Unscrew the white cap on the top of the carboy, being careful not to disconnect tubing from the cap.
4. Pour carboy contents into the sink.
5. Replace white cap on carboy and screw on tightly.

Inspect the tubing that runs from the vacuum to the carboy, there should not be any moisture in the tubing. This is important for maintaining the life of the vacuum pump and should be checked on a regular basis when the lab is in consistent use. To remove moisture from the tubing:

1. Loosen the white plastic cap on the carboy so that air can circulate out of the carboy.
2. Disconnect the clear tubing line at the ‘Vacuum’ side of the pump.
3. Attach the tubing to the nozzle on the ‘Pressure’ side of the pump.
4. Open the valves on the filtration manifold to allow pressure to escape.
5. Turn on the pump and let it run for one to two minutes. This will push accumulated moisture away from the pump, and back towards the storage carboy.
6. When you cannot see any more water droplets in the clear suction line, or the pump has run for about 2 minutes, turn off the pump.
7. Disconnect the tubing from the ‘Pressure’ side, and re-attach to the ‘Vacuum’ side.
8. Replace white cap on carboy and screw on tightly.

2. Lab Safety Equipment and Procedures

Water samples should be considered and handled as samples containing pathogenic material. Historic water quality data from sediment monitoring sites has shown high levels of pathogenic bacteria, thus safety precautions are necessary for sample handling.

Sample Handling Safety Procedures

1. Powderless gloves should be worn at all times when samples are being weighed or filtered.
2. In isolated instances, face shields should be worn when filtering to avoid potentially hazardous splashing.
3. Lab personnel should wash hands with anti-bacterial soap before and after all sample and battery handling.

Filter Contamination Prevention

Filter weights and subsequent sediment concentration data can be influenced by contamination. The following steps should be taken to prevent filter contamination.

1. Lab personnel should wash hands prior to commencing any work in the lab.
2. Lab personnel should put on powderless gloves before handling any filters.
3. Lab personnel should not conduct lab work while wearing wet clothing, or with wet hair.
4. Filters should be transported to the manifold and into foil trays using clean forceps. Following rinsing on the manifold, filters should not be touched with bare hands.
5. Filters that have been rinsed and dried for one hour should be moved into the 'clean desiccator' using clean tongs immediately upon removal from the oven.
6. 'Dirty' filters should never be put in the 'clean desiccator' cabinet and vice versa.

General Lab Safety

A First Aid kit is mounted on the wall near the sink of the lab. An updated red folder of MSDS forms is available in the file holder near the desiccator cabinets.

Potentially hazardous materials in the lab include:

NiCd batteries (4)
AGM Batteries (1)
Turbidimeter calibration standards
Contaminated water samples
Compressed air

3. Filter Preparation and Storage

Important: Filters must always be handled with forceps, and gloves must be worn during filter preparation and use.

1. Using forceps, remove filters from the box and separate carefully.
2. Visually inspect filters for holes or tears. Discard defective filters.
3. Rinse manifold platforms and filter funnels with DI water.
4. Remove the magnetic filtration cups from the six manifold stages.
5. Place new filters on all six manifold stages, rinse with DI water to create adherence to the manifold stages.
6. Open the pressure valves to pull DI rinse water down through the filters.
7. Check filters for holes by listening for a whistling sound, emitted as air passes through a hole in the filter.
8. Watch for filter centers to change from transparent to white, indicating that most of the moisture has been removed. (Pressure on the vacuum valve will be between 14- 17 in. Hg)
9. Carefully remove filters with forceps.

- When removing filters, grasp with forceps at the most outer edges of the filters, never grasp from the center of the filter.
10. Place each filter into a clean, numbered foil tray.
 11. Place foil trays on clean baking sheet and allow to air dry until the full sheet of foils is filled.
 12. Place baking sheet into oven and heat at 103° to 105°C for a minimum of 2 hours, more if there are sand trays in the oven*.
 13. Remove baking sheet from oven.
 14. Using tongs, place foil trays into Clean Filter Desiccator for cooling and storage.
 15. Foil trays must cool in desiccator for at least 30 minutes (to reach room temperature) before being used for filtration.

4. Crucible Preparation and Storage

Samples with a visibly elevated sediment content may be evaporated in glass crucibles or larger foil trays to reduce processing time and error.

1. Glass crucibles and foil trays are permanently numbered beginning with crucible number 901.
2. Clean crucibles and trays may be stored in the Clean Filter Desiccator between uses.
3. Crucibles and trays are washed with soap, rinsed with DI water after each use.
4. Place crucibles on a baking sheet and dried in the at 103° to 105°C for 30 minutes.
5. Remove baking sheet from oven.
6. Visually inspect crucibles for cracks, chips, or dirt.
7. Using tongs, place crucibles into Clean Filter Desiccator for cooling and storage.
8. Crucibles must cool in desiccator for at least 60 minutes before being used for sample processing.

5. Sample Log in and Filter Assignment.

Samples are not logged in to the computer until they are ready to be filtered and processed. All samples are logged in using the ‘Samples’ form in the Suspended Sediment Database, seen below.

Before the sample login information is entered, it is important to determine whether the sample should be analyzed separately for sand and fine particles, or just fine particles. Samples with high levels of turbidity (100NTU) are good candidates for separate sands/fines analysis. This can vary based on the bottle set. If in doubt, do a sand-fines split.

If the sample will only be processed for ‘fines’, only one sample login is recorded. If the sample will be processed separately for sands and fines, two different records will need to be created for the same sample.

Sediment Sample Login Page

Record Created	<input type="text"/>	<i>These fields are locked and cannot be edited. They are updated automatically when records are created or changed.</i>
Record Edited	<input type="text"/>	
Data Entry Tech	<input type="text"/>	
Date Sample Collected	<input type="text"/>	<i>Example 01/01/2011 01:00 AM</i>
Site Code	<input type="text"/>	
Bottle ID	<input type="text"/>	
Incoming Weight (grams)	<input type="text"/>	<i>Enter whole number, no units.</i>
Sample Analysis Tech	<input type="text"/>	
Date Sample Processed	<input type="text"/>	<i>Enter date of laboratory analysis.</i>
Sample Type	Fines <input type="text"/>	
Turbidity (NTU)	<input type="text" value="0"/>	
Remarks	<input style="width: 100%;" type="text"/>	
Stage Height (ft)	<input type="text" value="0"/>	
Filter Records		
Filter Tare Weight	<input type="text"/>	Remark <input style="width: 100%;" type="text"/>
Dish Number	<input type="text"/>	
Filter with Sample	<input type="text"/>	Invalid <input type="checkbox"/>
Record: 14 of 1 of 1 No Filter Search		
True Wet Weight (grams)	<input type="text"/>	<i>These fields are calculated automatically, do not enter values in these fields.</i>
Sediment Weight (grams)	<input type="text"/>	
Coarse Fraction mg/L	<input type="text"/>	
Fines Fraction mg/L	<input type="text"/>	

NOTE: You must start entering with Date Sample Collected, site code, bottle ID. Do not start with weighing in filters, they will not get recorded properly.

Field Title	Purpose	Entry Instructions
Record Created	Tracks when records are created.	Locked field, no entry permitted.
Record Edited	Tracks when records are changed or updated.	Locked field, no entry permitted.
Data Entry Tech	Records the name of staff person logging sample data into the database	Enter the name of the staff person logging sample information and assigning filters to samples.
Date Sample Collected	Records date and time when samples are collected in the field	Enter date and time on bottle label, using 24-hour clock. For midnight enter 00:00:01.
Site Code	Records which site samples are from.	Enter 3 letter site code, SLU, WAL CAN. For blanks enter BLK.
Bottle ID	Records which bottle is being used, subtracts tare weight	Enter 3 letter site code, bottle set letter, bottle number. Only bottles already logged into the

		system will be permitted for entry in this field.
Incoming Weight	Records weight of bottle with sample. Should not include weight of cap.	Enter incoming wet weight, in grams, as written on bottle label.
Sample Analysis Tech	Records name of staff person filtering the sample through the manifold.	Enter the name of the staff person filtering the sample on the manifold.
Date Sample Processed	Tracks when samples and filters are processed.	Enter date that sample processing starts.
Sample Type	Denotes whether the processing is for sand sized particles or fine sized particles.	Choose either sands or fines from the drop-down menu.
Remarks	Notes anything unusual about the sample.	Note low sample volume, if sample has spilled, if there is algal growing in the bottle.
Turbidity (NTU)	Record turbidity reading for the sample.	Enter this value if a turbidity measurement has been conducted on the sample.
Stage Height	Identifies creek stage when sample was collected.	Enter the corresponding stage height if available.
True Wet Weight (grams)	Calculates the weight of the sample. Automatically subtracts the tare weight of the bottle containing the sample.	This field is locked and calculates results automatically. It is shown only to reference the weight of the sample being analyzed.
Sediment Weight (grams)	Calculates the net weight of sediment collected on all assigned filters or crucibles by subtracting the tare weight of all assigned crucibles and filters.	This field is locked, and is shown to indicate the raw net sediment weight of the sample.
Coarse Fraction (mg/L)	Uses the 'True Wet Weight' and 'Sediment Weight' fields to calculate the fine sediment concentration of the sample in parts per million.	This field is locked, and is only shown to display the sediment concentration of the sample.
Fine Fraction (mg/L)	Uses the 'True Wet Weight' and 'Sediment Weight' fields to calculate the coarse/sands concentration of the sample in parts per million.	This field is locked, and is only shown to display the sediment concentration of the sample.

The filters assigned to each sample are recorded in a sub-form of the sample login form. The Filter Records form functions independently from the sample login form, but is electronically linked to the information in the sample login form. This form allows the data entry tech to assign unlimited filters to a single sample.

Filter weights are transferred directly to the sub-form electronically. To enable the transfer of weights to the database the CPS Plus software must be opened and running concurrent with the Access database. This will allow for automatic transmission of weights to the database.

To transfer weights to the database- point the cursor to the desired cell block and click to activate the cursor. Press 'PRINT' on the scale to transmit the weight to the cell block. For samples that weigh more than 9.99999 grams, the first digit of the weight will not transfer automatically from the scale to the database. Manually enter this value so that the correct weight value is entered.

Field Title	Purpose	Entry Instructions
Filter Tare Weight (grams)	Records the weight of the clean filter and dish, or crucible, assigned to the sample.	When the filter has been placed on the fine balance and the weight has stabilized, press 'Print' on the balance to transmit data to this field directly.
Filter With Sample (grams)	Records the weight of the used filter and dish, or crucible, assigned to the sample.	When the fine balance has stabilized, press 'Print' on the balance to transmit data to this field directly.
Dish Number	Tracks the dishes or crucibles assigned to the sample.	Type in the three digit number of the filter, written on the tab of the foil dish.
Remarks	Notes any problems with the filter or crucible during processing.	Enter notes as needed.
Invalid	Checkbox used to invalidate filters or crucibles that have been incorrectly assigned or damaged. Checking this box halts all automatic data calculations for the sample.	Check this box when filters have been damaged or contaminated. Checking this box will cancel the calculation of the fines or coarse fraction for the whole sample.

Basic (Fines) Sample Login

1. Remove samples from long term storage and inspect visually.
2. Bottles from each set should be dry, with roughly equal volume samples.
3. In the 'Suspended Sediment' master database, open the form called 'Samples'.
4. Enter the following fields in order:
 - a. Data entry tech (the staff person logging samples into the database)
 - b. Date sample collected. (Time entered as 24 hour clock.)
 - c. Site code
 - d. Bottle ID (An error message will pop up if your bottle is not identified in the database.)
 - e. Incoming Wet Weight
 - f. Sample analysis tech (the staff person performing the filtration on the sample)
 - g. Date Sample Processed
 - h. Sample Type (Fines)

5. Turbidity: Enter turbidity measurement indicated on bottle if available.
6. Stage Height: Enter stage height measurement indicated on bottle if available.
7. Remarks: Use this field to note unusual sample volumes (bottles that may have underfilled), floating debris, or macroinvertebrates in the sample.
8. Activate the 'Filters' sub-form of the Samples form to assign filters to the sample in accordance with protocol below.

Assigning Filters or Crucibles

Depending on the sediment content of samples, they may require from one to six or more filters to process the entire sample. Samples with substantial sediment content may be processed in crucibles as well as filters. Reviewing the stage data and processing samples in the order they were taken in the field will help determine the number of filters needed for each sample.

1. Use tongs to remove clean and dry filters from 'Clean Filter Desiccator'.
 - a. Only remove as many filters as you can weigh-in in a ten-minute period. Filters will pull moisture from the air which will result in an artificially high tare weight. A good rule of thumb is to start with two filters.
2. Place one filter or crucible on the fine scale balance and close all of the glass doors.
 - a. Close glass doors gently! Pulling doors closed with force can upset the balance of the scale, and produces an immediate effect on the weight of the filter. The weight of the filter will take longer to stabilize if the door is not closed gently.
3. Place the mouse pointer into the 'Filter Tare Weight' field and activate the field.
4. When the weight has stabilized on the balance, press 'Print' on the balance to transmit data to the field.
5. Enter the three digit number written on the foil tab or crucible into the 'Dish Number' field.
6. To assign an additional filter, move down to the next line within the filter subform. Do not create a new record for additional fine filters.

Sands/Fines Sample Login

1. Create a full record for 'Fines' processing for the sample. Assign filters as needed to collect fine particles.
2. Create a second Sample record for the 'Sands' information. Enter the identical sample information from the bottle as noted in Step 4 of the Basic Sample Login. Make sure that the new record specifies 'Sands' as the sample type.
3. Assign a separate series of crucibles or filters to the sample record for 'Sands' using the filters subform in the database.
 - a. Typically, this will involve a glass crucible, or large foil dish for collecting the sands.
4. Proceed with sample filtration only when BOTH the sands and fines login records have been created for the sample.

6. Filtering Samples

It is important to observe the vacuum pressure exerted on the manifold when filtering samples. Pressure applied to the filters should not exceed 20 in. Hg. If filtering on several stations

concurrently, leave one station clear so that vacuum pressure can be relieved. It is also important to NOT SHAKE THE BOTTLES. Shaking the bottles will make it more difficult to process the sample.

For samples that require a single filter, no sand split

1. Turn on the vacuum pump and open all of the valves on the manifold.
2. Rinse down all stations on the manifold to clear any debris or dust that might have settled on the surface.
3. Remove one of the magnetic cups and center the filter on top of the black filtering surface.
 - a. Filters should be handled with forceps and should not be 'flipped'. The side of the filter 'face-up' in the foil dish should be the side 'face-up' on the manifold.
 - b. If filters are not carefully centered on the manifold they may prevent the magnetic cup from sealing properly, and the sample may leak out of the cup.
4. Place the empty foil dish directly beneath the filter station. Filters and dishes are not transferable! It is important to keep the filter with the assigned dish throughout the process.
5. Wet down the filter with DI water and inspect the filter for contamination or tears.
6. Replace the magnetic cup on top of the manifold station over the wet filter.
7. Slowly pour the sample into the magnetic cup, being careful not to splash water out of the cup.
8. Hold the sample bottle cap over the magnetic cup and rinse down the cap with DI water.
9. Hold the sample bottle over the magnetic cup and rinse down the inside of the bottle with DI water. Tilt the bottle at an angle to flush out any particles trapped in the neck of the bottle.
10. When all of the sample and rinse water has filtered through the filter, rinse down the inside of the magnetic cup with DI water.
11. Watch the filter carefully to see that all water drains through the filter.
12. Switch off the suction to the station and remove the magnetic cup.
13. Using forceps, carefully slide the filter back into the assigned foil tray.
 - a. Handle the filter along the edges only to avoid removing any particles that are accumulated on the center of the filter.
 - b. It is important not to tilt or flip the wet filter as sands or other large particles may fall off of the filter.
14. Place the foil tray with filter onto a large baking sheet.
15. Allow wet filters to air dry on baking sheet until the baking sheet is full.

*Process the blank sample using the protocols for filtering with a single filter. One blank sample should be run for each bottle set (24 bottles) that comes in from the field.

For Fines samples that require multiple filters, no sand split:

1. Turn on the vacuum pump and open all of the valves on the manifold.
2. Rinse down all stations on the manifold to clear any debris or dust that might have settled on the surface.
3. Remove the magnetic cups and center the filters on top of the black filtering surface.
 - a. Filters should be handled with forceps and should not be 'flipped'. The side of the filter 'face-up' in the dish should be the side 'face-up' on the manifold.

- b. Do not place filters on more than 5 stations at the same time. One station must remain open to relieve pressure to the vacuum.
4. Place the empty foil dish directly beneath the filter station. Filters and dishes are not transferable! It is important to keep the filter with the assigned dish throughout the process.
5. Wet down the filters with DI water and check the filter for contamination or tears.
6. Replace the magnetic cup on top of the manifold station.
7. Place filters on up to 5 stations. Leave the sixth station clear so that vacuum pressure can be relieved if needed.
8. Slowly pour between 50 to 100ml of the sample onto each of the filters in turn.
 - a. While pouring, a white cross will appear across the center of the filter. The disappearance of this cross (filter turning uniformly brown) is an indication that the filter may be nearing capacity for filtering water.
9. Watch the vacuum pressure as each of the manifolds begin to pull water through the filters.
 - a. If the draw down of water is initially quick, but then slows, purge the vacuum line of water by opening the valve on the empty station. Allow the empty station to draw air into the suction line and push the filtered water towards the carboy.
 - b. If vacuum pressure rises to 19 in. Hg, open the valve on the empty station to relieve the pressure.
10. Close the valve at the empty station to increase the pressure on the filtering stations.
11. Open and close the valve at the empty station as needed to continue filtering without exceeding 19in Hg pressure.
12. When the majority of the sample volume has been poured out, carefully use DI water to dislodge remaining particles in the bottom of the bottle.
13. Distribute the remaining sample among the filters.
14. Hold the sample bottle over one of the magnetic cup and rinse down the inside of the bottle with DI water. Tilt the bottle at an angle to flush out any particles trapped in the neck of the bottle.
15. Hold the sample bottle cap over one of the magnetic cups and rinse down the cap with DI water.
16. When all of the sample and rinse water has filtered through the filter, rinse down the inside of the magnetic cups with DI water.
17. Watch the filters carefully to see that all water drains through the filters.
18. Switch off the suction to the station and remove the magnetic cup.
 - a. Particles may stick to the inner rim of the magnetic cup if a sample with particularly heavy sediment content is filtered on a single sample.
 - b. Once the magnetic cup has been removed from the completed sample, place the magnetic cup inside the cup of another filter that has been assigned to the same sample.
 - c. Rinse the inner edges of the nested magnetic cup into the cup that still contains a filter. This will capture any sediment in the sample that may have been lodged to the inside of the sample cup.
 - d. Rinse the last magnetic cup into the last filter tray. Do not rinse the cups in the sink until the bottle is completely processed.
19. Using forceps, carefully slide the filters back into their assigned foil trays. It is important not to tilt or flip the wet filter as sands or other large particles may fall off.
20. Place the foil trays with filters onto a large baking sheet.
21. Allow wet filters to air dry on baking sheet until the baking sheet is full.

For samples that are processed for Sands and Fines separately:

1. Separate the filters and crucibles that have been assigned to the Fines record from those that have been assigned to the Sands record.
2. Filtration for **Fines** takes place first, followed by **Sands**. Group and set aside the filters and dishes that have been assigned to the **Sands** record.
3. Rinse down all stations on the manifold to clear any debris or dust that might have settled on the surface.
4. Remove the magnetic cups and center filters on top of the black filtering surface.
 - a. Filters should be handled with forceps and should not be 'flipped'. The side of the filter 'face-up' in the dish should be the side 'face-up' on the manifold.
 - b. Do not place filters on more than 5 stations at the same time. One station must remain open to relieve pressure to the vacuum.
5. Place the empty foil dishes directly beneath the filter stations.
 - a. Filters and dishes are not transferable! It is important to keep the filter with the assigned dish throughout the process.
6. Wet down the filters with DI water and check the filter for contamination or tears.
7. Replace the magnetic cup on top of each manifold station.
8. Place the metal sands filter in the top of one of the magnetic cups that overlies a filter.
9. Slowly pour sample water through the sands filter into the magnetic cup.
10. Pour slowly and evenly, avoid suspending or mixing particles that have settled to the bottom of the sample. While pouring, a white cross will appear across the center of the filter. The disappearance of this cross (filter turning uniformly brown) is an indication that the filter may be nearing capacity for filtering water.
11. When the first filter has reached capacity, carefully move the sands filter to the magnetic cup of a second filter. Proceed with pouring the sample through the sands filter and into the cup until the second filter has reached capacity.
12. Once the majority of the sample has been poured through the sands filter and onto the fines filters, swirl the remaining liquid (50-100ml) to re-suspend the settled sediment within the sample.
13. While suspended, pour the remaining sample into the sands filter while overlying a clean fines filter.
14. Thoroughly rinse the sand particles in the sands filter to flush any fine sediments down into the magnetic cup.
 - a. The rinse water from the sands filter should be running clear to ensure that all fine particles have been flushed onto the fines filters.
15. When all of the sample and rinse water has been poured from the ISCO bottle and filtered through the sands filter and onto the fines filters, rinse down the inside of the magnetic cups with DI water.
16. Watch the filters carefully to see that all water drains through the filters.
17. Switch off the suction to the station and remove the magnetic cup.
 - a. Particles may stick to the inner rim of the magnetic cups if a sample with particularly heavy sediment content is filtered on a single sample.
 - b. Once the magnetic cup has been removed from the completed sample, place the magnetic cup inside the cup of another filter that has been assigned to the same sample.

- c. Rinse the inner edges of the nested magnetic cup into the cup that still contains a filter. This will capture any sediment in the sample that may have been lodged to the inside of the sample cup.
18. Using forceps, carefully slide the filters back into their assigned foil trays. It is important not to tilt or flip the wet filter as sands or other large particles may fall off.
 19. Place the foil trays with filters onto a large baking sheet.
 20. Once all of the fines have been flushed onto assigned filters the sands can be transferred to the assigned crucible or foil dish.
 21. Use DI water to rinse all of the sand into one corner of the sands filter
 - a. It is permissible to rinse the sands in the filter over the sink, using as much DI water as needed to accumulate all the particles into one mass. All of the fines should already have been flushed onto filters, so capturing rinsewater is not necessary at this stage.
 - b. Take care not to rinse any sands OUT of the filter.
 22. Hold the sands filter over the assigned crucible or foil tray and use DI water to flush all of the sands into the crucible.
 - a. All of the sand particles and a small volume of DI water (~100ml) should be captured in the dish.

*Process the blank sample using the protocols for filtering with a single filter. One blank sample should be run for each bottle set (24 bottles) that comes in from the field.

Drying Samples

Used filters and crucibles should be placed on a baking sheet in preparation for drying. Once the baking sheet is full, place baking sheet into the oven to dry samples. Crucibles can also be placed into one of the square or rectangular baking pans.

1. If not already on, turn on the oven. and set the thermostat to 225° F
 1. Place baking sheets with filters, or pans with crucibles into oven.
 - a. Never mix 'clean filters' and 'used filters' on the same baking sheet.
 2. Bake from three to 24 hours. Minimum drying time for processed filters is 3 hours if there are not sands trays in the oven.
 3. Crucibles must be baked for 1 hour after all visible water has evaporated. Minimum drying time for crucibles is around 8 hours.
 4. Remove baking sheets from oven.
 5. Using tongs, carefully transfer foil trays with filters and crucibles into the 'Sediment Sample Desiccator'.
 6. Filtered samples should cool in the desiccator for 30 minutes before they are weighed. Glass crucibles should cool in the desiccators for 60 minutes before they are weighed.

Final Filter and Crucible Weights

The fine scale balance should be checked for accuracy before weighing in any filters or crucibles. It is also important to watch the scale for signs of drift when weighing filters for an extended period of time.

1. Make sure the bubble level in the back left corner of the scale is centered.

2. Turn the scale on. The scale display should fill in completely to indicate a self-check.
3. Make sure the surface of the scale is clean and all of the glass doors are closed.
4. Complete the scale calibration and weight checks described in the Equipment Accuracy and Calibration section of this protocol.
5. Open the form called 'Final Weights'.
6. Locate the dish number that corresponds to the dish being weighed. Dishes are listed in ascending order.
7. Using the tongs, place the dish with filter or crucible onto the fine scale balance and close the glass doors.
8. Place the mouse pointer into the 'Filter With Sample' field and activate the field.
9. When the weight has stabilized on the balance, press 'Print' on the balance to transmit data to the field.
10. After weighing several dishes, save changes and close the form. Re-opening the form will limit the display to only the filters that are missing a final weight.

7. Data Management

The database should be copied and saved to a USB data transfer device twice each month. The database is then downloaded from the USB device to a staff computer at the MBNEP office and saved to a specified folder on the server. The back-up of the server created weekly and stored in a fireproof box.

Database copies can be made and transferred more frequently when a high volume of samples is being processed at the lab.

A query has been developed so that a hard copy of all sample results can be created. The query is run twice a month during times of sample processing. These paper records are stored at the lab.

C. SLQA Sample Processing

The Estuary Program participates in a twice a year quality assurance study with USGS. The study consists of 9 bottles of either increasing sediment, or three triplicate samples. This is a blind study, where we do not know the concentrations of sediment in the bottles. Samples are processed according to the above protocol, with a few differences.

16. All samples are processed with a Sands/Fines split
17. A sample blank is run for each DAY samples are processed
18. All samples are reweighed.
19. Samples are processed in pairs as much as possible.

When the bottles arrive:

- Inspect them for leaks
- Weight them in at sed lab, WITH LIDS ON
- Record weights in an excel spreadsheet (filed in the appropriate SLQA folder at the sed lab).
Make sure to remove the tape before weighting the bottles in.

Processing samples

- Check DI water conductivity
- Check scale, calibrate if necessary (Calibrated weekly)

- Clean manifold, cups, sands filter etc.
- Re-dry clean filters if necessary. Filters need to be cooled at room temperature, in a desiccant cabinet for one hour before use.
- Get a new sand tray. Inspect thoroughly (using forceps and kimwipes).
- Each bottle needs a sands/fines split.
 - o Weight in sand trays and then reweigh to confirm accurate weight. After it is weighed in, check for leaks by filling with deionized water. Then, set aside
 - o Process bottles as normal for conducting a sands/fines split
 - o Thoroughly inspect the bottle for sands stuck in there. Even if you think the bottle is clean, re-rinse it. This helps rinse the sands in the tray, which needs to be done anyways.
 - o Sands need to be thoroughly cleaned. Expect to spend up to 5 minutes doing this.
 - o Once you are sure the bottle is fully rinsed and the sands are as clean as they can get, clean the sands into the sands tray using deionized water.
 - Note – first, use a squirt bottle to get all the sands into the lower part of the sands filter while over the fines filter first. Then, carefully flip the filter over the sands tray. Use DI water to get all the sands into the sands tray.
 - o Put all filters in oven as normal
- Each day, process a blank following protocols in the sediment protocol

Drying and weighing in samples:

- Dry samples for 12 hours at 225 F
- Carefully transfer samples to the “dirty” desiccant cabinet to cool for one hour.
- Starting with the sample blank, weight in the samples. Only remove as many samples from that cabinet that can be weighed in within 10 minutes.
 - o If the blank does not have the same weight,
- All samples need to be rebaked, cooled and reweighed.

APPENDIX N. MORRO BAY NATIONAL ESTUARY PROGRAM
EQUIPMENT CALIBRATION PROTOCOLS

Morro Bay Volunteer Monitoring Program Equipment Calibration Protocols

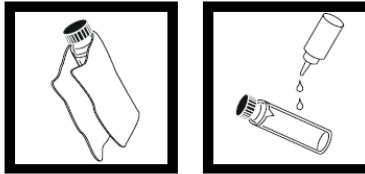
Hach 2100 Turbidimeter

This instrument is designed for long-term stability and minimal need for recalibration. It is recommended that recalibration be performed at least once every three months, or more often if experience indicates a need. To guarantee accurate calibration, use only the formazin standards issued by Hach.

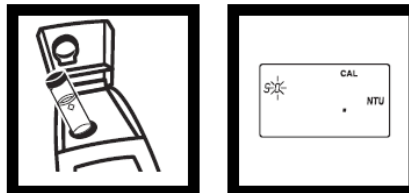
The formazin calibration standards have a long shelf life, but must be stored properly. Standards should be stored in their respective kit with cover in place, out of direct sunlight. For long term storage, refrigeration at 5° C is recommended, do not store above 25° C.

Calibration:

- Shake the standard vigorously for 2-3 minutes to resuspend particles. Do NOT shake the < 0.1 NTU standard, only the 20 NTU, the 100 NTU and the 800 NTU standards.
- Allow standard to stand undisturbed for 5 minutes.
- Gently invert the bottle 5 to 7 times.
- Prepare the sample cell for measurement using traditional preparation techniques. (Oil the sample cell and wipe with velvet cloth).



- Let the cell stand for one minute after oiling.
- Insert the <0.1 ntu standard cell, align the orientation marks on the cell and chamber. Close the lid and press the **I/O** button to turn on the meter.
- Press the **CAL** button, both the **SO** and **CAL** buttons will be displayed and the '0' will flash.



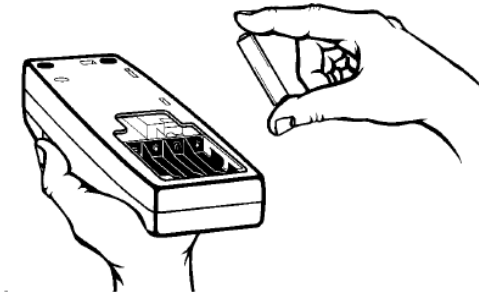
- Press **READ** and the meter will count down from 60 to 0, read the sample and calculate the correction factor.



- The display will show the 20.00 ntu value on the screen. Insert the 20.00 ntu standard with correct alignment and press the **READ** button.
- Repeat these procedures sequentially for the remaining standards as the meter prompts.
- After reading the final standard (800 ntu), the meter will re-display the **S0** screen from the beginning of the calibration.
- Remove the 800 ntu standard and press **CAL** to accept the calibration.

Battery Replacement:

- This meter works on 4 AA-size alkaline batteries. Brand name ‘digital’ batteries are recommended for best performance.
- Remove the battery compartment cover on the bottom of the meter to install new batteries.
- Check the polarity diagram on the inside of the panel to ensure correct installation.



YSI 2030 Conductivity Meter Calibration Protocol

Calibration:

The YSI 2030 meter requires only a single point calibration to adjust the meter for all parameters. As the VMP uses these meters in brackish and saline waters, a 1413 us or 12880 us standard is suitable for most testing.

- Turn the instrument on and allow it to complete its self test procedure.
- Select a calibration solution most similar to the sample the meter will be measuring.
- Pour at least 3 inches of solution into a clean glass calibration jar.
- Insert the probe into the beaker deep enough to completely submerge the oval shaped hole on the side of the probe. Do not rest the probe on the bottom of the container, suspend it above the bottom by at least ¼ inch.
 - Allow at least 60 seconds for the temperature reading to become stable.
 - Move the probe vigorously from side to side to dislodge any air bubbles from the electrodes.
- Press and hold the CAL button until a new window pops up, then select specific conductance, and uS.
 - Use the UP ARROW or DOWN ARROW key to adjust the reading on the display until it matches the value of the calibration solution you are using.
 - Once the display reads the exact value of the calibration solution being used (the meter will make the compensation for temperature variation) press the ENTER key.

Battery Installation

The instrument requires 2 alkaline C-cell batteries. Under normal conditions, the average battery life is 425 hours at room temperature without using the back light. A battery symbol will blink in the lower, left corner of the display to indicate low batteries when approximately 1 hour of battery life remains.

To install or replace the batteries:

1. Turn the instrument off and flip over to view the battery cover on the back.
2. Unscrew the four captive battery cover screws.
3. Remove the battery cover and remove the old batteries if necessary.
4. Install the new batteries, ensuring correct polarity alignment (figure 1).
5. Place the battery cover on the back of the instrument and tighten the four screws. Do not over-tighten.

The waterproof instrument case is sealed at the factory and is not to be opened, except by authorized service technicians. Do not attempt to separate the two halves of the instrument case as this may damage the instrument, break the waterproof seal, and will void the warranty.

YSI 2030 Dissolved Oxygen Meter Calibration Protocol

Before calibrating, determine if meter is being calibrated for fresh water (creeks) or salt water (bay) monitoring. Determine the approximate altitude of the area where the meter will be used.

Calibration:

- Ensure that the sponge inside the instrument's calibration chamber is saturated with de-ionized water.
- Rinse the probe with de-ionized water.
- Insert the probe into the calibration chamber.
- Turn the instrument on by pressing the ON/OFF button.
- Wait for the dissolved oxygen and temperature readings to stabilize, approximately 15 minutes.
- Press and hold the Calibrate key for 3 seconds. Using the up or down arrow key, highlight Dissolved Oxygen and press enter. The Pro2030 will indicate 'Calibrating %DO' on the display. The instrument will automatically calibrate the sensor to the current barometric pressure. If DO Local% is enabled, the sensor will calibrate to 100%. This may take up to 2 minutes depending on the age of the sensor and membrane. You can press the Cal key at this time to cancel the calibration.
- 'Calibration Successful' will display for a few seconds to indicate a successful calibration and then the instrument will return to the Run screen.
- If the calibration is unsuccessful, an error message will display on the screen. Press the Cal key to exit the error message and return to the Run screen. See the Troubleshooting guide for possible solutions.

SENSOR MAINTENANCE: CONDUCTIVITY

The openings that allow sample access to the conductivity electrodes should be cleaned regularly. The small cleaning brush included in the Maintenance Kit is intended for this purpose. Dip the brush in clean water and insert it into each hole 10 to 12 times. In the event that deposits have formed on the electrodes, it may be necessary to use a mild detergent (laboratory grade soap or bathroom foaming tile cleaner) with the brush. Rinse thoroughly with clean water, then check the response and accuracy of the conductivity cell with a calibration solution.

SENSOR MAINTENANCE: DO

Polarographic Sensors – Model # 605203

The cap membrane and KCl (potassium chloride) electrolyte solution should be changed every 2-4 weeks during regular use. In addition, the electrolyte solution and membrane should be changed if (a) bubbles are visible under the membrane; (b) significant deposits of dried electrolyte are visible on the membrane; and (c) if the sensor shows unstable readings or other sensor-related symptoms.

During membrane changes, examine the gold cathode at the tip of the sensor and the silver anode along the shaft of the sensor (figure 12). If either the silver anode is black in color or the gold cathode is dull, the sensor may need resurfaced using the 400 grit wet/dry sanding discs included in the membrane kit. Do not sand the electrode every membrane change as this is not routine maintenance. In fact, visually, the anode may appear tarnished and operate properly. YSI recommends using the sanding disc if the sensor has difficulty stabilizing or calibrating after a regular membrane change.

Gold Cathode

For correct sensor operation, the gold cathode (figure 12) must be textured properly. It can become tarnished or plated with silver after extended use. Never use chemicals or abrasives that have not been recommended or supplied by YSI.

First dry the sensor tip completely with lens cleaning tissue. Wet a sanding disc and place it face up in the palm of your hand. Next, with your free hand, hold the sensor in a vertical position, tip down. Place the sensor tip directly down on the sanding disc and twist it in a circular motion to sand the gold cathode. The goal is to sand off any build-up and to lightly scratch the cathode to provide a larger surface area for the electrolyte solution under the membrane. Usually, 3 to 4 twists of the sensor are sufficient to remove deposits and for the gold to appear to have a matte finish. Rinse the sensor thoroughly with distilled or deionized water and wipe the gold cathode with a wet paper towel before putting on a new cap membrane. If the cathode remains tarnished, contact YSI Technical Support or the YSI authorized dealer where you purchased the instrument. Figure 12, DO sensor with membrane removed.

Silver Anode

After extended use, a thick layer of Silver Chloride (AgCl) builds up on the silver anode (figure 12) reducing the sensitivity of the sensor. The anode must be cleaned to remove this layer and restore proper performance. The cleaning can be chemical and/or mechanical: Mechanical cleaning: In order to sand

the silver anode along the shaft of the sensor, simply hold the sensor in a vertical position. Wet the sanding disc and gently wrap it around the sensor and twist it a few times to lightly sand the anode (the goal is to simply sand off any build-up without scratching or removing layers of the anode itself). Usually, 3 to 4 twists of the sanding disc are sufficient to remove deposits. After completing the sanding procedure, repeatedly rinse the electrode with distilled or deionized water and wipe with lens cleaning tissue to remove any grit left by the sanding disc. Thoroughly rinse the entire tip of the sensor with distilled or deionized water and install a new membrane.

IMPORTANT: Be sure to: (1) Use only the fine sanding discs provided and (2) Sand as mentioned in the above procedures. Not adhering to either of these instructions can damage the electrodes. i 34 35
Chemical cleaning:

Remove the cap membrane and rinse the sensor with deionized or distilled water. Soak the sensing section of the sensor in a 14% ammonium hydroxide solution for 2 to 3 minutes or in a 3% ammonia solution overnight for 8-12 hours (most household ammonia cleaners are typically around 3%). Rinse heavily in cool tap water followed by a thorough rinsing with distilled or deionized water. The anode should then be thoroughly wiped with a wet paper towel to remove the residual layer from the anode. Trapping residual ammonia under the new membrane cap can quickly tarnish the electrode and/ or give false readings and should therefore be avoided. After performing a chemical cleaning on the polarographic sensor, lightly sand the cathode and anode following the mechanical cleaning procedures described previously. If this procedure is unsuccessful, as indicated by improper sensor performance, contact YSI Technical Support or the YSI authorized dealer where you purchased the instrument.

Oakton pH Test 30 pH meter

Calibration:

This meter must be rinsed with tap water and not with deionized water.

- Ensure that the sponge in the cap of the meter is damp.
- Remove the cap and rinse the meter with tap water.
- Turn the meter on and insert into the first calibration solution. We will be using 7.0 and 10.0 solution.
- Wait two minutes for the reading to stabilize, and record value as the pre-calibration reading on the calibration sheet.
- Remove the meter from the solution. Rinse the probe and dry with a Kim Wipe. Place in the second calibration solution.
- Wait two minutes. Record the value as the pre-calibration reading on the calibration sheet.
- Push the CAL button to enter the calibration mode. The CAL indicator will be displayed. The upper display will show the measured reading based on the last calibration while the lower value is the pH standard solution.

- Place the meter in the first calibration solution. Allow two minutes for the meter to stabilize.
- Press the HOLD/ENT button to accept the first calibration point. The upper display will be calibrated to the pH calibration standard and the lower will be toggling between the readings of the next pH standards.
- Rinse the probe with tap water, dry with a Kim Wipe and insert in the next calibration solution.
- Wait two minutes and then push the HOLD/ENT button to accept the second calibration point.
- The meter is now flashing to be calibrated in a third calibration standard, but we do not use a third standard. Push CAL to skip the remaining calibration point and exit to the measurement mode.
- Place the now calibrated meter in one of the calibration solutions. Wait two minutes for the meter to stabilize, and then record a post-calibration reading.
- Rinse the probe, dry and then take a post-calibration reading in the other calibration standard and record on the datasheet.
- Rinse meter and replace cap. Store in plastic box.

The pH meters must be calibrated on a weekly basis.

**Protocol for Winkler Titration to Verify DO Meter Accuracy
To be Conducted by VMP Staff Only**

Equipment:

- 1 L beaker
- Magnetic stirrer
- Stirring plate
- DI water
- Dissolved Oxygen Test Kit (HACH, Cat. No. 1469-33)

Procedure:


1. Rinse all testing materials with DI water.
2. Conduct the High Range Test (1-20 mg/L)
3. Fill the beaker with DI water.
4. Place a stirrer in the water and place the beaker on the stirring plate.
5. Set the stirrer to the maximum rate and allow the water to mix for 2 minutes.
6. Turn off the stirrer and use the beaker to fill the DO bottle (round bottle with glass stopper). Allow sample water to overflow the bottle and avoid turbulence and bubbles in the sample while filling.
7. Replace the beaker on the stirring plate and turn the stirrer on to the maximum.
8. Place the DO meter probes into the beaker, making sure that they are fully submerged.
9. Conduct the Winkler DO test on the sample. Incline the bottle slightly and insert the stopper with a quick thrust to avoid trapping air bubbles. If bubbles become trapped, discard the sample and repeat the test.
10. Remove the stopper and add the contents of one Dissolved Oxygen 1 Reagent Powder Pillow and one Dissolved Oxygen 2 Reagent Powder Pillow. Stopper the bottle carefully to avoid trapping air bubbles. If bubbles become trapped, discard the sample and repeat the test.
11. Shake the bottle vigorously to mix. Flocculant (floc) precipitate will form. Brownish-orange precipitate indicates oxygen is present.
12. Wait for floc to settle to approximately half the bottle volume.
13. Shake the bottle vigorously.
14. Wait for floc to settle halfway.
15. Remove the stopper and add the contents of one Dissolved Oxygen 3 Reagent Powder Pillow. Stopper the bottle carefully to avoid trapping air bubbles. If bubbles become trapped, discard the sample and repeat the test.
16. Shake the bottle vigorously to mix. Floc will dissolve and the sample will turn yellow if oxygen is present.
17. Fill plastic tube full (to the top) with prepared sample.
18. Pour the contents of the tube into a square mixing bottle.
19. Add Sodium Thiosulfate Standard Solution one drop at a time to the mixing bottle. Count each drop. Swirl to mix after each drop. Add drops until the sample becomes colorless.
20. The total number of drops of titrant equals the total mg/L Dissolved Oxygen. Compare to the read-outs on the DO meters.
21. Record all values on the Equipment Calibration Log Sheet.
22. Discard waste in bottle labeled "Winkler Titration Waste Container."
23. Triple rinse all DO testing bottles with DI water.

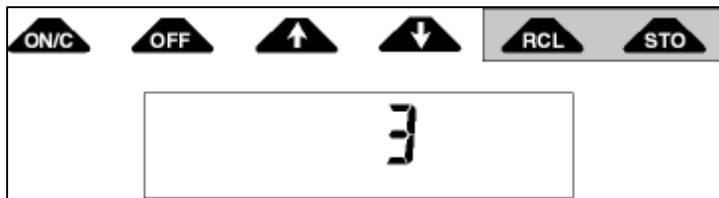
Protocol for Flo-Mate Zero Check and Zero Adjust

Zero Check

- First wipe down the sensor because a thin film of oil on the electrodes can cause noisy readings.
- Then place the sensor in a five gallon plastic bucket of water. Keep it at least three inches away from the sides and bottom of the bucket.
- To make sure the water is not moving, wait 10 or 15 minutes after you have positioned the sensor before taking any zero readings.
- Use a filter value of 5 seconds. Zero stability is ± 0.05 ft/sec.

Zero Adjust

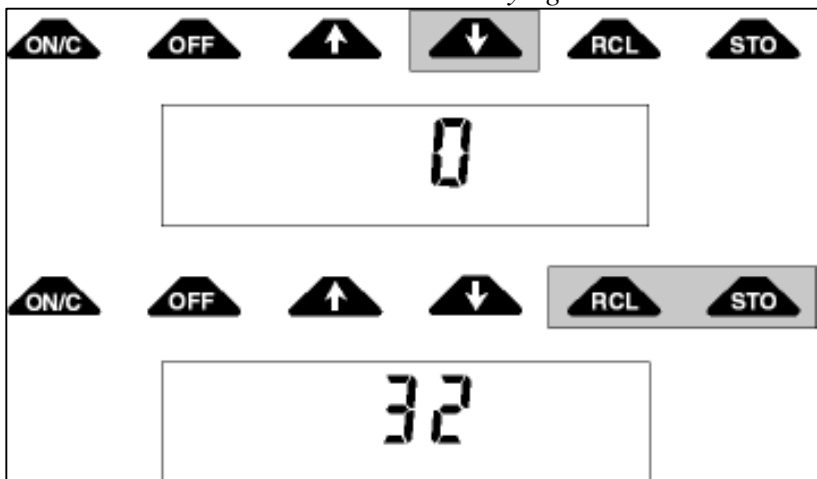
- Position the sensor as described in the zero check procedure.
- To initiate the zero start sequence, press the STO and RCL keys at the same time. You will see the number 3 on the display.
- Decrement to zero with the  key.



- The number 32 will be displayed.
- The unit will decrement itself to zero and turn off. The unit is now zeroed.

Comment:

Each key in the zero adjust sequence must be pressed within 5 seconds of the previous key. If the time between key entries is longer than 5 seconds or if a wrong key is pressed, the unit will display an ERR 3. Turn the unit OFF then back ON and try again.



Equipment Calibration Log

**Morro Bay Volunteer Monitoring Program
Equipment Calibration Log: pH**

Meter ID: PHEL-MBV-_____

Meter Model #: Oakton pHTestr 30

Date	Time	pH Standard 7		pH Standard 10		Initials
		Pre-cal	Post-cal	Pre-cal	Post-cal	

Bacteria Laboratory Equipment

SHEL LAB SMI2 Incubators and Thermometers

This instrument is to provide a controlled dry heat environment from 8°C to 70°C \pm 0.35 °C for culture tubes or Petri dish incubations.

Each time a volunteer or staff member places samples in the incubator or removes samples, the temperature on the certified thermometer inside the incubator is recorded on a log sheet. The temperature on the certified thermometer should be at 34°C \pm 0.5°C for IDEXX Colilert-18 incubation and 41°C \pm 0.5°C for IDEXX Enterolert incubation. If the incubator is not at the desired temperature, the temperature control will be adjusted until the temperature is correct.

If the incubator thermometer is not within \pm 0.5°C of the certified thermometer, the temperature difference will be recorded and the incubator temperature will be adjusted accordingly.

APPENDIX O. MORRO BAY NATIONAL ESTUARY PROGRAM
DATA MANAGEMENT PROTOCOLS

Morro Bay Volunteer Monitoring Program Data Management Protocols

Revised March 2019

Overview

All of the data that is collected for the volunteer monitoring program is managed in either an Access database or in Excel spreadsheets. Data managed in the Morro Bay Monitoring Program (MBMP) Access database, which includes water quality, bacteria and bay water quality data, are submitted to CEDEN. Bioassessment data is managed in a separate Access system designed by SWAMP. Suspended sediment data is managed in a Sediment Access database. The remaining projects have separate datasheets that were created by the VMP specifically for each project. Following is a list of the data that is collected and the file where it is entered.

Project	Data Entry File
Bacteria	v:/database/MBMP.accdb
Water Quality and Flow	v:/database/MBMP.accdb
Water Quality and Bacteria QA	v:/database/MBMP.accdb
Dawn Patrol	v:/database/MBMP.accdb
Phytoplankton Bioassessment and algae	...monitoring\data management\MBVMP database\MBVMP_Vers_*.xlsx ... monitoring\data management\Macro Database\yyyy\BioassessmentShortform
Suspended Sediment Data	...VMPDATA\SEDIMENT\SUSPENDED SEDIMENT.ACCDB
Eelgrass	...VMP DATA\EELGRASS\DATA-SUMMARIES\ VMP FIELD DATA or VMP BIOMASS DATA
Bird Survey	...VMP DATA\BIRD SURVEYS\DATA- SUMMARIES\BIRD SURVEY DATA.xlsx
Stream Profiles	...VMP DATA\STREAM PROFILES\DATA- SUMMARIES\PROFILE DATA or SUBSTRATE DATA
Volunteer Hours	...VMP ADMIN\RECOGNITION\VOLUNTEER HOURS\VOLUNTEER HOURS.xlsx

Project	Data Entry File
Continuous Water Temperature	...VMP Data\Temperature (TIDBIT)*site code*.xlsx
Stormwater Monitoring	... VMP Data\Stormwater\SPM Parking Lot Project Monitoring\SPM Stormwater Monitoring Data Results yyyy.xlsx
New Volunteers	Outlook

CEDEN

The California Environmental Data Exchange Network (CEDEN) is a cooperative effort by many partners, including the Surface Water Ambient Monitoring Program and the Water Boards to exchange environmental data and make it accessible to the public. In order to load data into CEDEN, it must be entered into a series of upload templates.

The VMP created an Access-based data management system to manage data, import lab data, and export data into the CEDEN template formats. These templates are then submitted to the appropriate Regional Data Center for checking and loading into CEDEN. We call this database the Morro Bay Monitoring Program (MBMP) Database.

The following types of data are managed in the MBMP Access system:

- Creek water quality – conventional analytes and nutrients
- Creek instantaneous flow data
- Bacteria data for creeks and bay
- Dawn Patrol – bay water quality data
- Seeps water quality data
- QA data for water quality and bacteria monitoring
- Monitoring sites
- Monitoring equipment

Suspended sediment data is managed in a Sediment Access database.

Bioassessment data for macroinvertebrates, physical habitat and water quality are managed in the SWAMP format templates which are fed into an Access data management system by the Regional Data Center.

The following types of data are managed in Excel:

Monitoring Effort	Description of Data
Minisondes	Continuous water quality meter data
Pressure transducers	Water depth data

Stream profiling	Cross-sectional profile data
Continuous water temperature	Continuous temperature data

Bacteria / Bacteria QA

1. Use the printed IDEXX table, or the IDEXX MPN Generator (in the Start Menu), to verify that the volunteer correctly identified the MPN.
2. If the sample was diluted, verify that the MPN was multiplied by the dilution factor.
3. For *E. coli*, if multiple dilutions were run, enter data from the results that are closest to 80% positive wells (77 of 97 wells). This may be from separate dilutions for the separate types of bacteria. Indicate on the datasheet and in the Notes column of the database what dilution, for each type of bacteria, was used on the data entered.
4. For total coliform, always enter the result for the dilution that is less than 80% positive wells. This may not be the result that is closest to 80% positive wells.
5. If the results for a site are above this range then increase the dilution factor the following month, and likewise, if the results for a site are below this range, decrease the dilution the following month.
6. The data is entered in the Bacteria Monitoring form in the MBMP.acddb
7. For entering the QA data:
 - a. For precision data: In the detail record, choose Hydrologic Unit = QA. For QA Purpose, choose 'Volunteer Precision'. Fill in the QA Result, QA Qualifier and QA Dilution. If the results are within the R_{log} range, then the precision test is a 'pass'.
 - b. For accuracy data: For QA Purpose, choose 'CRM'. Fill in the QA Result, QA Qualifier and QA dilution. If the volunteer result is within the target range provided by the manufacturer, then the accuracy test is a 'pass'.
 - c. For sterility data: For QA Purpose, choose 'Sterility'. Enter the QA Result, QA Qualifier and QA dilution. If the QA Result is less than the MDL, then the result is a 'pass'.
 - d. When finished entering data, initial and date the datasheet in the bottom left hand corner then file in the folder 'Data Entered – Needs QA'. The program manager will then double check the data, initial and date it, then either file it into the 'Need to File' folder or the appropriate project folder.

Dawn Patrol

1. The data is entered in the MBMP database in the Dawn Patrol form.
2. When finished entering data, initial the datasheet in the upper right hand corner then file in the folder 'Data Entered – Needs QA'. The program manager will then double check the data, initial it, then either file it into the 'Need to File' folder or the appropriate project folder.

Water Quality

1. The data is entered in the Water Quality section of the MBMP database.
2. In the Field Observations section, enter observations about the site regarding algae, flow conditions, etc. If no field observations were collected, check the 'No Field Observations Made' box. Also enter any notes about equipment problems, site conditions, etc.

3. If data was discarded due to failure of QA criteria, flag the data as 'Exclude' so that it will not be included in analysis.
4. If duplicate readings are taken for any of the analytes (e.g. dissolved oxygen), enter the second reading in the Volunteer Precision field. The % RPD will be calculated. For RPD values < 25%, choose 'Pass'. For RPD values > 25%, choose 'Fail'.
5. For conductivity at SYB the meter will read in mS/cm. All of the other site's data is collected in μ S/cm. Therefore, the value that the volunteer records needs to first be converted (multiply by 1000) before being entered.
6. For the nutrients, if there were not any levels detected then the Result entered would be the PQL of the equipment and the Result Qualifier would be '<'.
7. For QA accuracy data entry:
 - i) This lab-generated data is imported in an automated process. From the main menu, choose Lab Import and then Water Quality Lab Import.
 - ii) Hit 'Choose File' to select the file to import. Then choose the 'Import' button.
8. For QA precision data entry:
 - i. Complete the Result and Result Qualifier information for the precision result.
 - ii. The %RPD will automatically calculate.
 - iii. If the RPD is > 25%, choose 'Fail'. If the RPD is < 25%, then choose 'Pass'.
9. For Sterility data entry:
 - i. Enter the Result and Result Qualifier information for the blank.
 - ii. If the Result is less than the PQL, then enter 'Pass'.
10. Flow data is entered in the flow detail record. The Q value is automatically calculated by the system. .
11. When finished entering data, initial the datasheet in the upper right hand corner then file in the folder 'Data Entered – Needs QA'. The program manager will then double check the data, initial it, then either file it into the 'Need to File' folder or the appropriate project folder.

Suspended Sediment

1. All data is entered directly into the suspended sediment database. No worksheets or paper copies are generated from the sample collection or analysis.

Bioassessment

1. Bioassessment data is entered into CEDEN, per the protocol provided by SWAMP. The protocols are available at:
 - a. <http://swamp.mpsl.mlml.calstate.edu/resources-and-downloads/database-management-systems/swamp-25-database/documentation-25/swamp-data-management-plan>
 - b. <http://swamp.mpsl.mlml.calstate.edu/resources-and-downloads/database-management-systems/swamp-22-database/documentation-22>

Eelgrass

1. Eelgrass field data is entered in files separate from biomass data from each site. For each year of monitoring, create a new file in each folder
2. ..\..\Monitoring\vmp\VMP Data\Eelgrass\Data - Summaries\VMP FIELD DATA

3. Create separate worksheets within the spreadsheet for each site and label with site code and year.
4. Enter all data as shown on the datasheet, calculate average coverage for each site.
5. When finished entering data, initial the datasheet in the upper right hand corner then file in the folder 'Data Entered – Needs QA'. The program manager will then double check the data, initial it, then either file it into the 'Need to File' folder or the appropriate project folder.

Stream Profiles

1. The stream profile data is entered in a separate excel database for each site. These files are located in the folder U:\Monitoring\vmp\VMP Data\Stream Profiles\Data - Summaries\Profile Data
2. The file All Sites – Channel Data.xls is where all of the descriptive channel data is entered. This includes bankfull measurements and calculations of slope and entrenchment for all of the sites.
3. For each of the profile sites, open the file corresponding to that site's name. Under the 'Stadia Reading' heading, create a column that corresponds to the current year and enter the stadia reading data.
4. Under the 'Elevation' heading, also create a column that corresponds to the current year then enter the formula to compute the elevation data from the stadia data. Drag the formula down through all of the cells.
5. Open the profile plot worksheet. Update the source data by adding a new series in order to display the new data. Then remember to update the heading on the plot to reflect the correct time frame.
6. There is also a file, U:\Monitoring\vmp\VMP Data\Stream Profiles\Data – Summaries\Dates Sites Surveyed.xls, which shows a table that indicates which of the sites were measured in a given year. Update this file as well as sites are revisited.
7. When finished entering data, initial the datasheet in the upper right hand corner then file in the folder 'Data Entered – Needs QA'. The program manager will then double check the data, initial it, then either file it into the 'Need to File' folder or the appropriate project folder.
8. FYI – in the past, veg and substrate data was collected in conjunction with the stream profiling data. This data is saved in CCAMP ## in the TransData2 worksheet.

Pressure Transducers

See Appendix N for description of data protocols.

Minisondes

See Appendix C for description of data protocols.

Volunteer Hours

1. There is a separate worksheet for each year, which has listed only the volunteers that participated during that year, and shows both monthly and annual participation hours.
2. The 'All Volunteer Hours' worksheet lists the names of all the volunteers that have ever participated, their annual participation hours and overall total for all years. This worksheet should be manually updated each December.
3. The volunteer list is sorted by last name so search for the appropriate location to add the volunteer.
4. Insert a row then in the first cell, type in the volunteer's last name. In the second cell, type in the volunteer's first name.

5. Now monthly hours can be added for that volunteer.
6. The previous worksheets have been protected to prevent accidental data loss from those years.
7. Should these sheets need to be edited, the protection can be removed by clicking on the pull down menu 'Tools', selecting 'Protection', and 'Unprotect Sheet'.
8. Similarly, to protect a sheet once data has been entered click on the pull down menu 'Tools', select 'Protection', and 'Protect Sheet'.
9. You will be prompted for a password which, in this case, should be left blank.

New Volunteers

1. All participants in program activities need to fill out a contact information/liability release form.
2. If the volunteer is interested in receiving monthly emails with program updates, enter their contact information into the Access database. Open the database and choose Data Input/Lookup.
3. Enter contact info into appropriate cells.
4. Give the release form to Karissa so she can add their email address to the active mailing list. She will return the sheet to you to file in the volunteer info binder.
5. If someone only participates in a one-time event and is not interested in getting email updates, do not enter their contact info into the database. Simply file their sheet in the brown Pendaflex folder.
6. Enter completed volunteer training in the Volunteer Training Log Sheet located at u:\monitoring\vmp\vmp admin\recognition\volunteer hours.xls. Enter the volunteer name, hours of training, type of training, location of training, and staff member conducting training.

APPENDIX P. MORRO BAY NATIONAL ESTUARY PROGRAM PRESSURE TRANSDUCER
PROTOCOL

Morro Bay National Estuary Program Pressure Transducer Protocol

The MBNEP is responsible for the maintenance and monthly data download of four pressure transducers located throughout the Chorro Creek Watershed.

Site Code	Creek Name	Site Description	Latitude	Longitude
310CCC	Chorro Creek	Chorro Creek upstream from Chorro Flats	35.3575	-120.812
310 CPN	Pennington Creek	Pennington Creek upstream of Rancho El Chorro Outdoor School	35.345265	-120.728912
310 PFL	Pennington Creek	Pennington Creek at fish ladder	35.344597	-120.729984
310WFL	Walters Creek	Walters Creek at flume	35.348090	-120.750501

At each site flow is measured using the MBNEP protocol, stage height is recorded using a staff plate or stadia rod, and the pressure transducer data is downloaded.

Flow

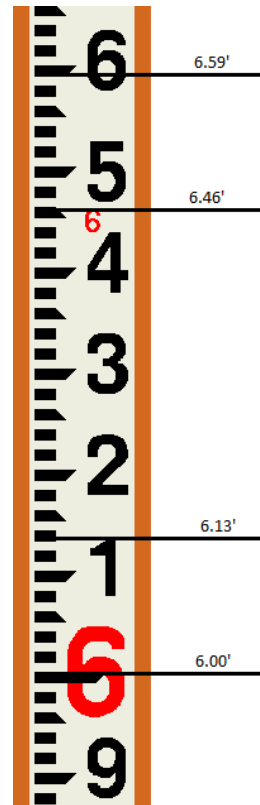
Flow is measured at each site using the MBNEP protocol. Generally, the measurements should be taken at the same location each month, however, as flow regimes change, taking measurements at the same location may not be feasible. The location can be moved to another spot in the general location, just as long as there are no tributaries, seeps, springs etc. contributing to the flow of the new monitoring location.

During low flow conditions it is especially difficult to take flow measurements. It is important to move any rocks, organic material, or anything that may cause an eddying effect in the water column. If any “grooming” of the channel is conducted be sure to do it before and not during measurements. Please review the MBNEP flow monitoring protocol prior to heading out into the field.

Stage Height

Depending on the monitoring location, stage height is measured using a semi permanently installed staff plate or stadia rod. At 310CCC, 310CPN, 310PFL, and 310 WFL, a staff plate is installed directly adjacent to the pressure transducers. The stage height will be read directly off these plates.

All stage measurements are in tenths of feet.



Pressure Transducer Data Download

The pressure transducer data is downloaded every month using a tablet computer.

Connecting the Pressure Transducer to the Computer



1. To access the end of the pressure transducer cable, remove the cap from the PVC pipe that is located on the stream bank. To remove the desiccant housing, hold the textured section of the cable connector in one hand and the desiccant in the other. Twist in opposite direction to unlock the desiccant housing from the cable.
2. Position the TROLL Com and the cable connector so they will connect properly. Insert the Troll Com connector firmly into the cable connector.



3. Hold the textured section of the sleeve in one hand and the TROLL Com in the other. Push and twist until you hear a click. The click ensures the cable and TROLL Com are securely attached.

4. Attach the TROLL Com to the USB 2.0 to Micro USB adapter. Insert the Micro USB adapter into the tablet computer.

Connecting the Pressure Transducer to the Win-Situ 5 Software

1. Open the Win-Situ 5 Software by double clicking the Win-Site 5 icon.
2. When you open the Win Situ 5 Software, you are asked if you want to connect to your device. Click **Yes**. Synchronize the instrument clock to the tablet clock. 
3. Parameters and measurement are displayed in light gray on the home screen if a connection is established. Click the **Play** button  to view live readings. The software displays an error message if a connection cannot be established.

Set Up a Data Site

1. Click the **Site** button.



2. Select either **Default Site** or click the **New** button to set up a custom site.
3. Enter Site Name. Entering coordinates and uploading a picture are optional.
4. Click the **Check Mark** to save the data site.

Set up a Data Log

1. Click the **Logging** tab



2. Click the **New** button.



3. Select your site, and name the new log.
4. Click the **right arrow** to continue after each step.
5. Select **Pressure (PSI), Temperature (C), and Depth (ft)**.
6. Select **Linear** logging method.
7. Configure the logging method interval to **15 Minutes**.
8. Select **Manual Start** condition, **No Stop Time**, and **Disable** overwrite when full.
9. Select **Depth to Water** as level reference.
10. Enter a specific gravity value of **0.999**.
11. The software displays a summary. Review your selections carefully before clicking the check mark.
12. To start a manual log, go to the logging tab, highlight the log name, and tap the **Start** button.

Downloading Data to a PC

1. With the pressure transducer connected to the tablet computer, select the **Logging** tab.
2. Select the log you intend to download.
3. Choose a Running, Suspended, Stopped, or Deleted log.
4. Click the **Download** button.
5. In the next screen, select **New Data** (data logged since the last download).
6. The log is copied to the tablet into your Win-Situ working directory folder. View or change the working directory using **File>Settings**.
7. At the end of the download, Win-Situ gives you the option of viewing the data.
8. Select **Yes** and the log will be displayed in the data screen.
9. Click **Disconnect** to disconnect the pressure transducer from the tablet computer.

Maintenance

In addition to the flow measurement, stage readings, and data download, it is important to perform a few maintenance measures to insure that the equipment is functioning properly.

Desiccant

Each cable end is equipped with a chamber filled with desiccant. Desiccant protects the electronics from condensation, which can cause irreparable damage and loss of data. The indicating desiccant changes from blue to pink as it becomes saturated with moisture. Check the condition of the desiccant each time you visit the site.



Desiccant stages (from left to right): New, nearly expired (replace now), expired
When the desiccant is expired, bake at 225 °C for approximately 30 minutes or until the desiccant has turned blue.

Clearing of Debris

It is important, especially after a storm event, to clear any debris or sediment that has accumulated around the pressure transducer. Also, check to see if the pressure transducer is secure to the creek bank and channel bottom.

APPENDIX Q. MORRO BAY NATIONAL ESTUARY PROGRAM HOBO CALIBRATION
AND DEPLOYMENT PROTOCOL

MBNEP HOBO Calibration and Deployment Protocol

The Hobo U26 logger measures temperature and dissolved oxygen. Dissolved oxygen requires a calibration before each deployment.

Storage:

The Hobo loggers need to be stored in the calibration boot with a damp sponge. To do so, wet the sponge with freshwater, place the sponge in the end of the calibration boot and then insert the logger in the boot.

Maintenance:

To clean the hobo sensor cap:

1. Remove the protective guard but leave the sensor cap on the sensor.
2. Rinse the logger with clean water.
3. Gently clean the cap with a soft bristled brush when needed. Use a cloth to remove dirt from the logger.
4. If extensive debris or mineral build up is present, soak the cap end in vinegar for 15 minutes, then soak in deionized water for another 15 minutes.

To clean the logger body:

1. Make sure sensor cap is installed on the logger
2. Gently scrub logger body with plastic brush or wipe down with a towel. Use Alconox to remove grease, vinegar to remove mineral deposits.
3. Rinse logger body with deionized water.

To launch the hobos:

The hobos need to be calibrated before each deployment by putting the logger in 100% saturation by placing it in water saturated air. In addition, we take field measurements with a YSI 2030 when launching and retrieving the hobo.

To connect the logger to a computer:

1. Plug Optic USB Base Station into the office laptop (Black Toshiba)
2. Unscrew the pointed cap on the communication end of the logger
3. Attach the coupler to the base station (Blue coupler).
4. Insert the logger into the coupler, aligning the bump/arrow on the coupler with the notches on the logger. When plugged into the computer, the Status light should be green.

Calibrating the Logger with Lab Calibration Tool:

Necessary materials: fresh water, calibration boot and sponge, barometric pressure and office laptop

1. Make sure the logger sensor is clean and dry.
2. Connect the logger to the computer as described in the previous section. Stop the logger if it is currently logging.
3. Open the Lab Calibration by going to **Device → Lab Calibration**
4. In “Step 1: 100% Saturation” enter the barometric pressure for your current location. If barometric pressure reading has been adjusted for sea level, select the box for “if

using sea level barometric pressure, enter elevation” checkbox and enter in the elevation.

5. Make sure logger has protective guard installed so that sensor is covered
6. Wet the small sponge with fresh water, squeeze out excess water
7. Place sponge in the end of the calibration boot
8. Insert logger in the calibration boot so that there is approximately 1cm (0.5 inch) overlap between the end of the boot and the body of the logger. This will ensure there is enough space between the end of the logger and the sponge. The logger should not be pressed up tightly against the sponge.
9. Wait approximately 15 minutes until the logger reaches temperature equilibration, but less than 30 minutes so the logger doesn't go to sleep
10. Click “Get DO values from logger” button to display the 100% saturation results. You can click this button as many times as needed. The results are updated each time you click the button. To check for equilibrium, click the “Get DO value from the logger” button several times in a row to check the current DO Concentration from logger at 100% Saturation value. If the value remains the same or varies very little with each button click, then temperature equilibrium has likely been reached. *Note-it takes a few seconds for the value to update after clicking the button*
11. Once the results have stabilized, click the Next button.

Lab Calibration

HOB0 Dissolved Oxygen U26-001, S/N: 90000004

Logger's Gain Adjustment: 0.00
Logger's Offset Adjustment: 0.00
Lab Calibration last completed on: Never

If you would like to directly edit your logger's gain and offset, then you may skip the calibration steps.

I know my values, skip to Finish

Step 1: 100% Saturation | Step 2: 0% Saturation (optional) | Step 3: Finish

Enter a value for barometric pressure: 760 mm Hg

If using sea level barometric pressure, enter elevation: 0 m

Place logger in 100% saturated environment for 15 minutes, then click the Get DO from Logger button:

Get DO value from Logger

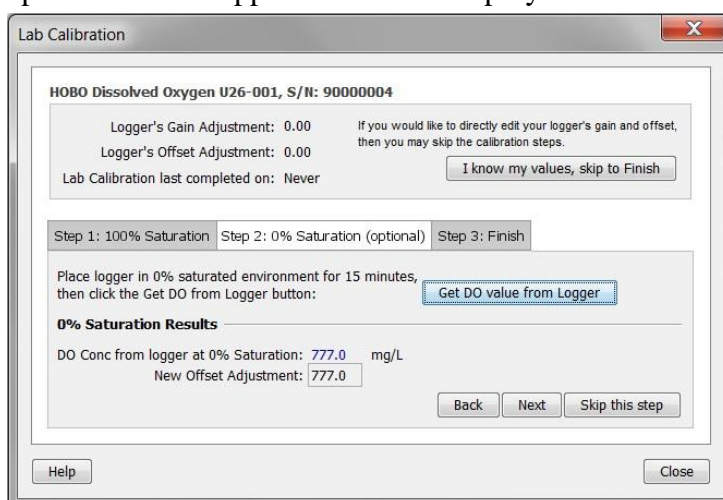
100% Saturation Results

DO Conc from logger at 100% Saturation: 9.24 mg/L
DO Conc calculated at 100% Saturation: 8.8 mg/L
New Gain Adjustment: 0.953

Next

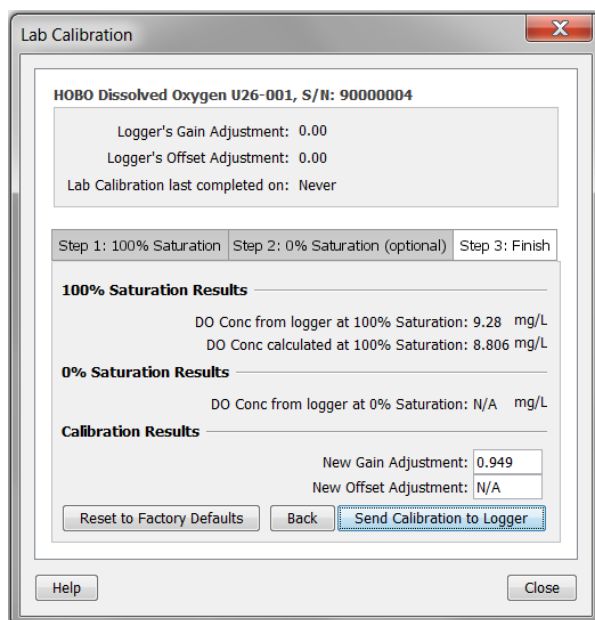
Help Close

12. 0% Saturation is optional and not applicable for our deployment. Click the button for



“Skip this Step”.

13. The results for the steps are displayed with the overall calibration results and the new gain and offset adjustment values. If you are satisfied with the results, click the “Send Calibration to Logger” button. The logger will then be calibrated based on the new values. The values will not take effect until the logger is launched.



Launching the Logger:

After calibrating, the logger needs to be launched to configure it before taking into the field for a deployment. Once launched, the logger will record samples and events. Samples are sensor measurements recorded at each logging interval. Events are independent occurrences triggered by logger activity, such as Bad Battery or Host Connected. Events help you determine what was happening while the logger was logging. To launch the logger:

1. Connect the logger to the computer. From the Device menu, select Launch (short cut CTRL + L)
- 1) Change the Name to Site Code Date deployed (ex UCR 04-12-2016). The serial number of the device will not change (10871201 = SO-MBV-3)

- 2) Under Sensors, select both DO and Temperature channels to log.
- 3) Under Deployment, select the desired logging interval (30 minutes unless told otherwise). We will typically pre-program the logger to start. To do that, under “Start Logging” select on Date/Time. Select the desired date and the time you want it to start logging. Select the time on half hour intervals (e.x. have it start at 2:30:00, not 2:29:15).
- 4) Select a logging interval (typically 30 minutes)
- 5) Choose when to start logging and when finished, hit “**Delayed Start**”
 - a. Choose a start time right at or before you anticipate the logger to be in the water. This allows us to use the YSI field measurements for field calibration when necessary. Be sure to record the time the logger is placed IN the water in the field book, and we can delete any bad data when the logger is downloaded.
- 6) Record Equipment ID, site deployed, date deployed, time started and deployment number in Rite-In-Rain notebook.
- 7) Note: the loggers will continue logging until we connect it to the computer and click “Stop Device”.

Deploying the Logger:

1. The hobo should be stored with the black boot on at all times. Remove the black calibration boot before placing hobo in water. Place the logger inside the PVC housing.
2. We typically place the loggers in the same location at each site, but check that the logger will be located in a spot where it will receive good flow and can be safely secured to an object (around a tree etc). Loop one cable loop through the U lock and thread the U lock through the housing. Lock to something on the bank.
3. Make sure the logger is fully submerged and not in direct sunlight to minimize temperature changes that are unrelated to water temperature. Gently shake the housing underwater to release any air bubbles that may be trapped in the housing and near the sensor.
4. Record the time the logger is deployed in the field log book. Take four YSI measurements at least a minute apart and record them in the notebook. Wait until the temperature of the logger has equilibrated with the water temperature, typically 30 minutes after being submerged. Try to take YSI measurements near an interval of 15 minutes (e.g. 10:00, 10:15, 10:30).

Picking up and Redeploying:

1. We typically leave sensors out for 1 week at a time, pick them up and then drop them off at another location. When picking up the sensors, check out the office laptop. Bring the laptop, shuttle, logbook and YSI.
2. When you get to the site, take another set of YSI readings in the manner above (excepting the temperature equilibration) and record in the notebook. Record the time and date that you picked up the logger. Plug the logger into the computer and press “Stop Device”
3. Offload the data files onto the computer. Open the data file in HOBOWare. In the plot setup window, select the Dissolved Oxygen Assistant and click Process. Compare the temperature reading from the YSI measurement to the begin- and end-points for the DO assistant and make sure they are similar.

4. Plot the data and save it as a project file. Check over the data to make sure the numbers make sense.
5. If re-deploying, wipe the sensor cap with a KimWipe and go through the DO calibration processes again. Note, you can plug the hobo in at the first site and allow it to equilibrate as you travel to the next field site, but be careful about the temperature in the car. The hobo needs to be calibrated at room temperature. Once at the next site you can finish the process to save time. Go through the same steps as before to calibrate and launch the logger
6. When you get to the next site, take YSI measurements and record all the deployment information in the notebook.

Stopping the logger

If you are picking up the hobos and not redeploying:

1. Record site, date and time in the hobo log book. Take a YSI reading and record all parameters in the book.
2. Grab the hobo and bring back to car. Wipe off the hobo and the PVC housing with a towel, rinse the membrane end with tap water and wipe with a KimWipe.
3. Put the black calibration boot back on the bottom of the hobo. Unscrew the small cap on the other end of the hobo and attach it to the Optic USB BaseStation. Plug it into the laptop, turn on laptop and open HoboWare. Press “Stop Device”.
4. Offloading the files can be done at the office, or in the field, but to preserve battery life and eliminate bad data, stop the device in the field.

Field Calibration Data:

At the start and end of each deployment we take YSI measurements. Hoboware Assistant adjusts the values accordingly.

When you download a file and the “Plot Setup” field opens click “Process Data”. Dissolved Oxygen Data Assistant should open up. Make sure the box is checked for “Perform Field Calibration”. Enter in the Dissolved Oxygen value for the start of the deployment and the end of the deployment

1. Check that the times are correct. Adjust accordingly from the dropdown menu if they are not correct.
2. Make sure the correct boxes are checked. If using end calibration only, check that the Beginning Calibration box is not checked.
3. Export file to excel. Save in format mm_dd_yyyy_site. This format keeps pair sites together. Save in this folder: **N:\Monitoring\vmp\VMP Data\Minisondes and Hobos**
4. Highlight any data that looks weird. Create a notes section in a separate tab and record any weird data or events that can explain data (i.e. conductivity went high and then dropped low-2” of rain in 24 hours).

APPENDIX R. MORRO BAY NATIONAL ESTUARY PROGRAM EELGRASS SEED
TRANSPLANT MONITORING PROTOCOL

Eelgrass Seed Transplant Monitoring Protocol

Gear:

- USCG approved Personal Floatation Device with whistle
- Stand Up Paddleboard (SUP) and paddle
- Cellphone in waterproof bag
- Datasheet and clipboard with pencils
- GPS unit
- Laminated sheet with eelgrass conditions/seed phases
- Waterproof field camera
- 1mx1m Quadrat
- Proper clothing for the conditions (waders, wetsuit, board shorts etc.)

Safety Protocol:

Before leaving the office, check weather and verify conditions are suitable for paddling. Notify designated person of the float plan for the day, including launch location, number of people going out and estimated return to the office. Complete the Float Plan (found below) prior to leaving the office. A USCG approved personal floatation device (PFD) must be worn at all times when using a SUP/Kayak. General guidelines are winds must be less than 10kts for paddling. You will encounter seals, sea lions, and sea otters both around and within eelgrass beds. Remember to give them ample space and avoid areas where they are hauled out/resting, even if this means skipping a site for the day

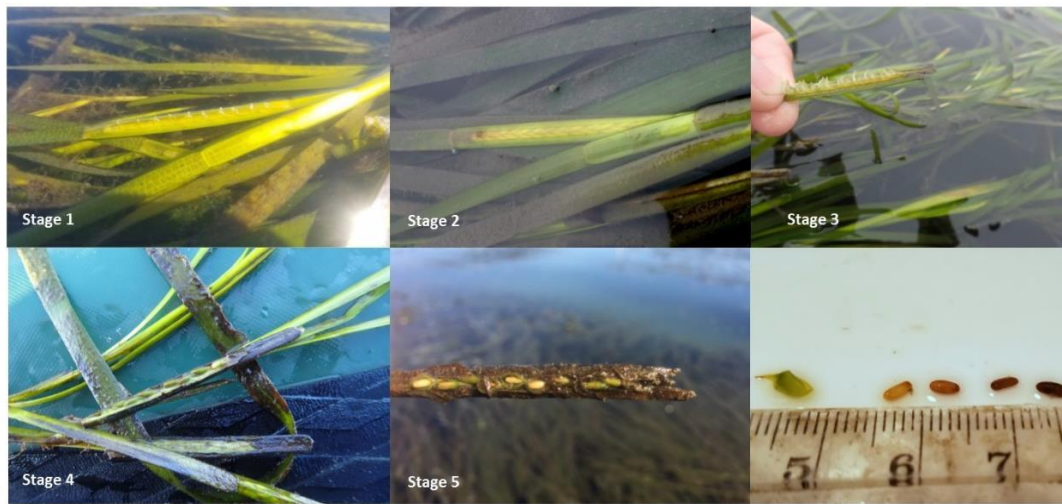
Seed Stage Monitoring Field Protocol:

The primary survey method is to paddle over each bed on a SUP at a lower tide (does not have to be a negative tide). Due to morning tides and typical wind patterns during the summer months, the best route is typically to start by paddling to the northernmost portion of the N. Sandspit bed and ride the tide back into the bay to both Reference and Windy Point beds.

Paddle: Order of beds: North Sandspit → Reference Bed → Windy Point

1. Launch SUP at the Whaler boat slip by the Estuary Program Office, or by Tidelands Park. Check the wind forecast and tides to help decide which location to launch from
2. Within each bed, choose three different locations to take seeding density measurements (Northern, middle, Southern)
3. Paddle directly into eelgrass bed and stop all momentum
4. Lay down quadrat in a representative area and photograph from above
5. Record GPS coordinates, photo ID's, and number of flowering shoots within the quadrat
6. Determine the reproductive stage at which each flowering shoot is at and record on data sheet
 - a. If unsure of stage – take a photo and record the photo ID
7. Record the location of floral shoots within the bed (outside, middle, inside).
8. Record any notes you may have
9. Move on to next location within the bed

10. Move on to next bed



Seed Transplant Field Protocol:

In the summer/fall of 2017, the National Estuary Program monitored and collected flowering eelgrass blades from healthy beds within the bay and planted them at three locations in the fore to back bay – by Channel Marker 18, State Park Marina and off Mitchel Drive. The locations are illustrated in XX. There are slightly different numbers of seeds planted in each location, but each location has seeds planted in burlap seed sacks, and then seeds planted directly into the ground. Seeds were planted in the fall of 2017.

Supplies:

- Boogie boards
- 1m x 1m quadrat with 16 intersections
- Ruler
- Camera
- Datasheets on Rite-in-the-Rain paper
- Site maps and diagrams

At each site:

1. All plots are marked with at least one PVC pole. Refer to the site maps and diagrams before approaching to make sure you do not step on the plot.
2. Place the quadrat down using the PVC markers for reference. Use the boogie boards if necessary to avoid kneeling down in the plot and disrupting the surrounding sediment. Count the number of seedlings in the plot and mark their location on the datasheet.
3. Take an overview photo of the plot.
4. For each seed bag, record the length of the longest blade and note any comments (blade condition, algae, animals etc.) Skip if touching the blade might harm the shoot/cause it to become unrooted.

Figure 1. Seed bag deployment monitoring data sheet.

Seed Bag Deployment Monitoring

Site: _____ Date: _____ Time: _____ Crew: _____

Shoot Count: record number of shoots in each seed bag. Also, note directions.

Blade Length: For each seed bag, record the length of the longest blade and note any comments (blade condition, algae, animals etc.)

Total Number of shoots: _____

	LENGTH	Comments
1		
2		
3		
4		
5		
6		
7		
8		
9		
10		
11		
12		
13		
14		
15		

General Notes and Observations:

Upon returning from the field:

5. Rinse and put away gear (including GPS and camera)
6. If using the green inflatable SUP – deflate until no longer rigid
7. Check that all datasheets were completed and all necessary info was filled out correctly and writing is legible.
8. Record field notes in the field notes Word document.
 - a. Include: date, times started, tide, crew and a brief description of what was done. Include comments such as predominate reproductive stage, general location of shoots within bed, and any other pertinent information
9. Download photos from the camera and file accordingly.
10. Scan datasheet and file in appropriate location on server. Put hard copies of datasheets in appropriate location.

Figure 2. Eelgrass seed transplant and monitoring locations.

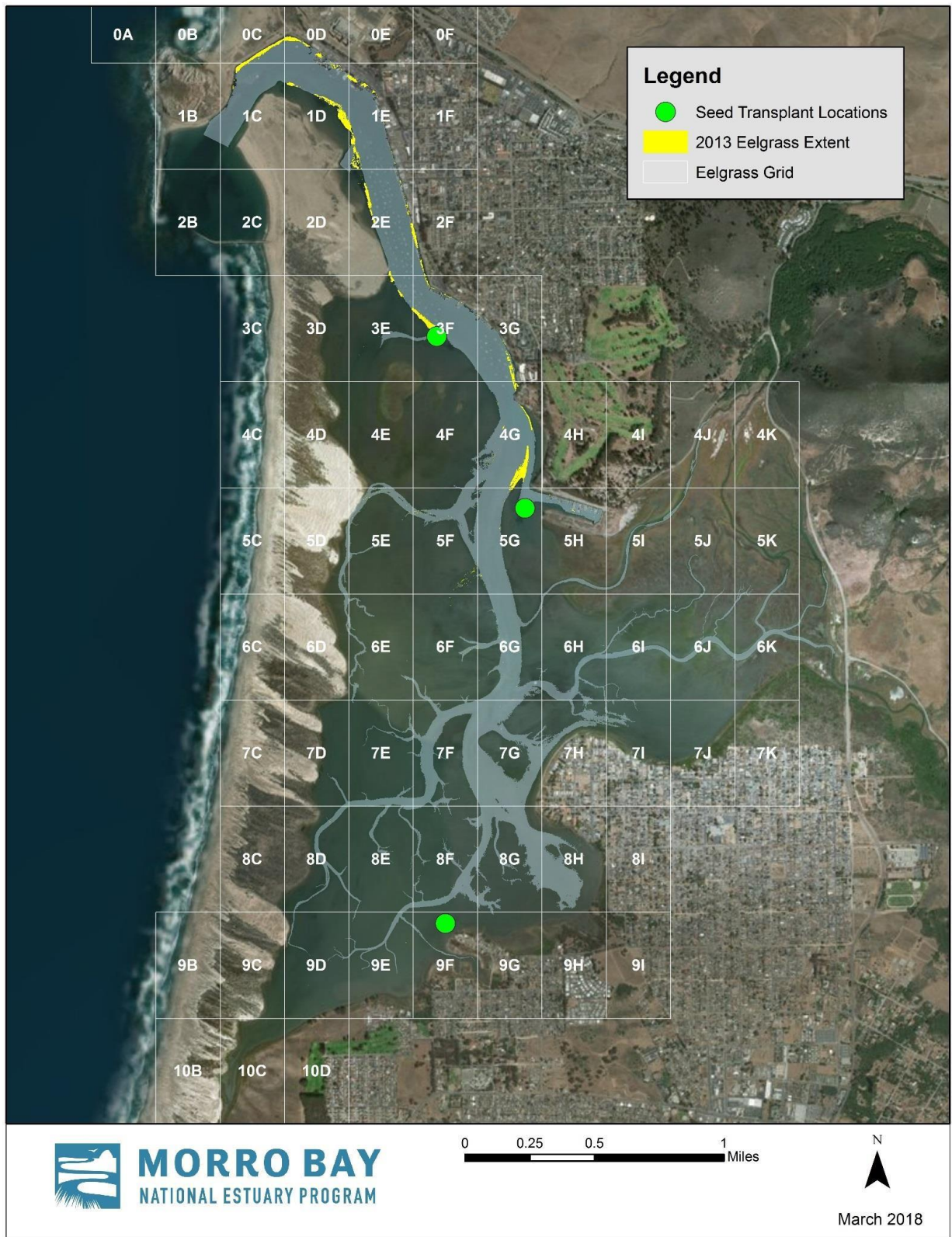


Figure 3. MBNEP Kayak/SUP Float Plan.

Morro Bay National Estuary Program Kayak/SUP Float Plan	Harbor Patrol: 805-772-6254
Date: _____	Name(s): _____
Launch location: _____	
Return Location: _____	
Survey Purpose: _____	
Surveying at/around: _____	
Departure time: _____	
Estimated time of return: _____	
Vessel: _____	
Additional Notes:	

Morro Bay National Estuary Program Kayak/SUP Float Plan	Harbor Patrol: 805-772-6254
Date: _____	Name(s): _____
Launch location: _____	
Return Location: _____	
Survey Purpose: _____	
Surveying at/around: _____	
Departure time: _____	
Estimated time of return: _____	
Vessel: _____	
Additional Notes:	

APPENDIX S. MORRO BAY NATIONAL ESTUARY PROGRAM CONTINUOUS TEMPERATURE
LOGGER PROTOCOL

Overview: The HOBOMobile MX2203 TidbiT is a continuous temperature monitor that can log up to 96,000 measurements and is accurate to $\pm 0.2^{\circ}\text{C}$ (or $\pm 0.36^{\circ}\text{F}$). These are sealed units that require a Bluetooth-enabled device and an Android or iOS device with the HOBOMobile app to interface with the TidbiT software. These devices are left in streams to monitor the daily fluctuations and seasonal trends of water temperature in Morro Bay watershed creeks. In addition to detecting small changes in temperature when submerged in water, the Tidbits can also be used to detect when the monitored section of a stream runs dry.

Protocol

Before Deploying: New loggers must be configured and loggers that have been deployed previously may need to be re-configured if they are being used at a new site.

- 1) Connect to TidbiT using HOBOMobile application. Press down on the magnetic button to activate the device's Bluetooth signal.
- 2) Configure the TidbiT settings. If this is a new logger, create a new page in the equipment log book following the naming scheme used in previous loggers. The HOBOMobile configuration options are listed below:
 - a. **Name:** Our convention for these models is "SO-MBV-# - SiteCode" e.g. *SO-MBV-6 – SLU*. Beside the site code, these names should not change, so that they are consistent with database entry. TidbiTs may be moved from one creek to another, so in the above example, the logger could be renamed to *SO-MBV-6 – DCH* if the logger was redeployed at Dairy Creek. Serial numbers always remain constant, so these are the most reliable way to check records for errors in naming.
 - b. **Group:** Not used as of 2020
 - c. **Location:** Not to be relied upon for deployment history – the logger only takes the mobile user's current location.
 - d. **Deployment Number:** Not edited by users
 - e. **Logging Interval:** Loggers should be set to 30 minute intervals.
 - f. **Start Logging:** Start the log on an interval of 30 minutes e.g. 1:00, 1:30, or 2:00 PM on the day you plan to deploy.
 - g. **Stop Logging:** Never (wrap option). Data should be downloaded with enough frequency that the memory does not fill. Complete a final download and delete on-board data at the end of each water year.
 - h. **Alarm:** Not used
 - i. **Bluetooth/Power Saving Mode:** Always Off. This will save battery. Bluetooth connection is only needed for a few moments to download. Note that the Bluetooth signal does not penetrate water.
 - j. **Logging Mode:** Normal, fixed interval. Statistics may be enabled if desired, but each of them can be obtained easily in Excel.
 - k. **Show LED:** On. This serves as a visual check in case the mobile device cannot connect.
- 3) Start the TidbiT. After completing the configuration settings above, select "Start" so that the configuration file is sent to the TidbiT. The main page should then show you that the logger is "Awaiting Delayed Start". At this point, the logger is ready to be placed in the creek.

Note: HOBOMobile also allows users to adjust the app's settings. Select SI units for downloads. US units are acceptable, but require conversion in Excel.

Deploying: These devices need to be placed in an appropriate location within the creek and properly secured.

- 1) Use four (4) zip ties to secure the TidbiT to one looped end of a steel cable. Normally, once these devices are attached to a cable, they should not be removed unless their attachment seems faulty or they need to be sent for servicing.

- 2) Using additional zip ties, attach the other looped end of the cable to a sturdy root or branch that would be unlikely to come loose in a large flow event. Attach them to live branches and roots, not dead ones.
- 3) Site selection is important – pools and deep runs are ideal. If possible, the TidbiT should be placed out of direct sunlight. Its depth should be below the level of downstream control. The cables are flexible and may move around, so imagine the TidbiT's possible locations in fast and turbulent flow and whether it could be “stranded” somewhere dry after a passing high flow.
- 4) Take photos of the spot where the cable is anchored.
- 5) Take a temperature reading with a handheld device such as a YSI meter to confirm the downloaded temperature later.

Note: Certain TidbiTs require a MBNEP secured password in order to access data. All passwords are 3834.

After Deploying: After leaving the device, it is important to make permanent notes of the deployment details. For a new year of deployment, create a folder under **N:\Monitoring\vmp\VMP Data\Temperature (TIDBIT)\Deployment Notes**. For 2020, there is a folder called “2020 Deployment” and a file called “2020 TIDBIT Deployment Log.docx,” which gives a detailed record of the deployment site, including a written description and photos of the site. For each new year, create a folder and a corresponding deployment log. Upload any photos you took of the TidbiT deployment site, as well as a detailed written description. Other deployment details, such as YSI temperature comparisons, last time checked, and previous deployment sites should be put under **N:\Monitoring\vmp\VMP Data\Temperature (TIDBIT)\Deployment Notes** in a file called “Deployment Record Database.xlsx.”

Downloading and Data Management: TidbiT data should be downloaded during regular site visits for water quality or other regular monitoring events. Additional download intervals are only necessary when retrieving a device before a storm or if the data is needed sooner for a report or research.

- 1) Take a temperature reading with a YSI 2030 or similar handheld device. Write down the temperature.
- 2) Pull the device completely out of the water and press the magnetic button to activate the Bluetooth signal while the HOBOMobile app is open. The app should automatically detect the signal and display the active TidbiT.
- 3) Note the amount of used memory and battery level.
- 4) Select “Readout” under the Actions menu. The file will begin to download and can be found in the Data Files tab afterward. Open the data file and review the data for anomalous behavior like spikes or erratic recordings.
- 5) Compare the last reading with the YSI result. Temperature values should be within 0.5°C of each other if they are taken at the same time.
- 6) Put the logger back into the water or cut it from the anchor if it needs to be removed. Note any significant changes in the site since the last visit.
 - a. If you are ending a deployment, select “Stop Logging” to end the current program. Data can still be downloaded after it is ended.
- 7) Back at the office, use the HOBOMobile app to export the downloaded graph to an .XLSX file. The file should be added to the appropriate site folder under **N:\Monitoring\vmp\VMP Data\Temperature (TIDBIT)**. Different mobile devices will have different methods to upload the data file – one reliable method is to send the Excel file in an email to your MBNEP account.
 - a. Update the “Deployment Record Database.xlsx” file with YSI and TIDBIT temperature readings and note if you have completed a deployment, specifying the TidbiT's new location in the VMP room or wherever it is stored.
- 8) Review the file again in Excel using a graph. Flag data points that may have resulted from exposing the logger to the air during a download or before/after deployment.

Morro Bay National Estuary Program

Stormwater Monitoring Protocol

Goal

The purpose of the monitoring effort is to identify the presence of typical pollutants in the stormwater runoff from the State Park Marina parking lot before and after construction of a project to reduce runoff to Morro Bay.

Sampling Preparation

1. Note all coolers and sample bottles labeled with Site and Sample ID codes.
2. Note site locations and site codes.
3. Visit sites during daylight to become familiar with monitoring locations and site needs.

Storm Event Sampling

1. Assemble all equipment:

Coolers and ice packs

Gloves

Bucket and Cup

Rain jackets and boots

Sampling Pole

Datasheets and Pencils

2. At the site, rinse the bucket or sampling cup 2-3 times in the outflow before filling it.
3. Use the bucket to fill the sample bottles. **DO NOT RINSE SAMPLE BOTTLES** as they have been sterilized and contain preservatives. **DO NOT SUBMERGE PRESERVATIVE BOTTLE TO FILL** as the preservative could mix with and contaminate the sample.
4. Record time of sample collection.
5. Complete datasheet.

Stormwater Sampling Bottles

Oil and Grease

- BC Laboratory Analysis
- 1-quart glass bottle, sulfuric acid preservative
- Fill as far as possible, a little head space is all right
- 28-day hold time

Dissolved Metals: Copper, Zinc, and Lead

- BC Laboratory Analysis
- 1-Liter plastic bottle, no preservative
- Leave some head space in the bottle
- 181-day hold time

Total Suspended Solids

- BC Laboratory Analysis
- 1-Liter plastic bottle, no preservative
- Leave some head space in the bottle
- 7-day hold time

Total Petroleum Hydrocarbons (TPH)-Gasoline

- BC Laboratory Analysis
- VOA, Glass Vial 40 mL, 200 uL HCl preservative
- Try to leave NO head space when filling
- 14-day hold time

Total Petroleum Hydrocarbons (TPH)-Diesel

- BC Laboratory Analysis
- 1 Liter glass amber bottle, no preservative
- Leave some head space in the bottle
- 14-day hold time

Bacteria (*E. coli*, Total Coliform)

- San Luis Obispo County Public Health Agency
- 1 sealed sterile IDEXX bottle with sodium thiosulfate
- Leave some head space in the bottle
- 24-hour hold time

Upon Completion of Sampling

1. Return to office.
2. Confirm that all sample bottles are labeled correctly.
3. Store samples in refrigerator.
4. Review datasheet for completeness.
5. Complete the sample chain of custody form.
6. Contact lab to notify that samples have been collected.
7. Notify lab courier or plan for GSO to deliver samples.
8. Conduct bacteria analysis for E. coli and Enterococcus. Analyze with a 1:10 dilution and a 1:100 dilution. If the SLO County lab is available, they should conduct the analysis using the IDEXX method.

BC Laboratories, Inc.

Chain of Custody

4100 Atlas Ct Bakersfield, CA 93308
 (661)327-4911 (661) 327-4911

Report & Bill to:

Analysis Requested

Name: Morro Bay National Estuary Program Phone: 805-772-3834

Address: 601 Embarcadero Suite 11	Project: State Park Marina Stormwater Monitoring
City: Morro Bay	Project #:
State: CA ZIP: 93442	Samplers Name:
Attn: Ann Kitajima	

	TPH-Gas	TPH-Diesel	Oil & Grease	Total Suspended Solids	Dissolved Lead	Dissolved Copper	Dissolved Zinc							
Lab #														
Sample Description														

Relinquished by: (Signature) Received By: Date & Time

Relinquished by: (Signature) Received by: Date & Time

(A)= Analysis (C)= Container (P)= Preservative

Relinquished by: (Signature) Received By: Date & Time

293/313

Sample Chain-of-Custody for First Flush

Sample Data Collection Form

Sample Collection

Sample ID	Time Collected	Collected by	Bottle Type

Site Conditions: (please circle one)

Precipitation: Mist Drizzle Rain Downpour

Water Clarity: Clear Cloudy Murky Muddy Dark/Black

Wind: Calm Light Medium Strong

Flow: 5 gal/min 10 gal/min 25 gal/min 50 gal/min

Observations:

Oily Sheen?	Yes	No
Odor?	Yes	No
Floating Trash?	Yes	No
Bubbles/Foam Plume at outlet?	Yes	No
Dark Plume at outlet?	Yes	No

Other Observations and Notes:

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APPENDIX U. CALPOLYSEABIRD SEAFET PH SENSOR DEPLOYMENT AND
CALIBRATION PROTOCOLS

SeaBird SeaFET pH Sensor Deployment and Calibration Protocols

Maintenance, calibration, deployment, and recovery can only be performed in direct consultation with Emily Bockmon.

*Much of the information included in this SOP is collected and synthesized from the SeaFET User Manual and Best Practices Guide.

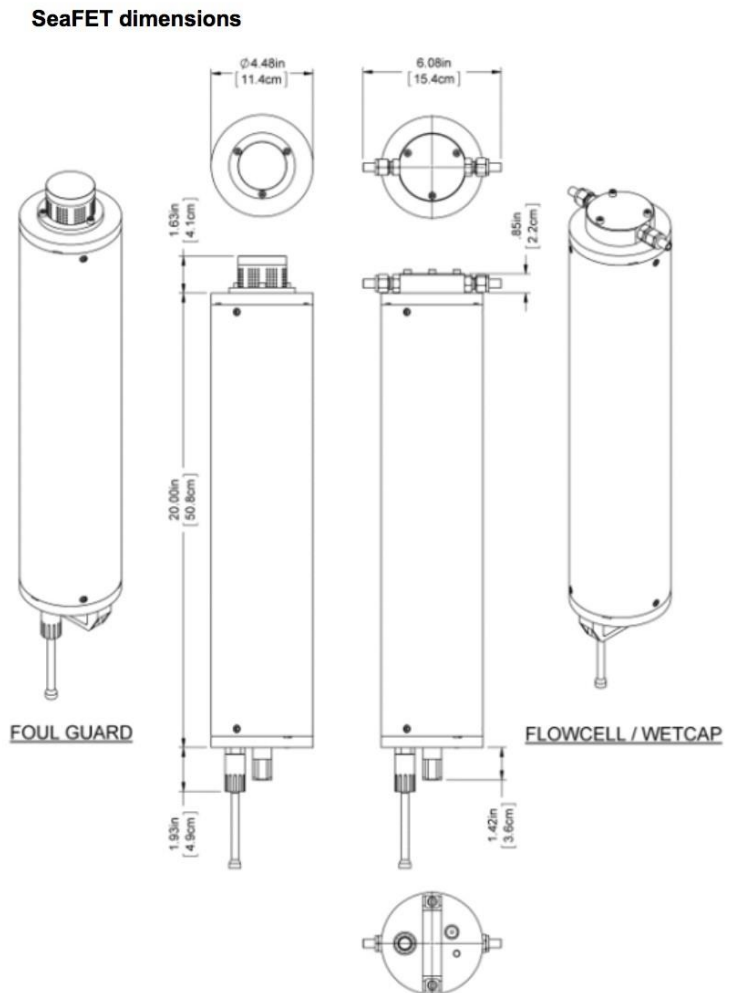
Instrument Specifications

The SeaFET V2 is a commercial oceanographic pH sensor available from Sea-Bird Scientific. It uses an ion sensitive field effect transistor to sense pH in marine environments. Two reference electrodes incorporated within the SeaFET allow for extra verification of the data validity. The internal reference is an electrode in a saturated KCl solution which is incorporated within the DuraFET® sensor. The external reference is an Ag/AgCl reference electrode that responds to the seawater pH and salinity directly. In addition to pH the DuraFET sensor also measures and records temperature.

Diagram of the instrument
(per User Manual):

According to the manufacturer
(Sea-Bird User Manual) the
SeaFET has the following
specifications:

- Rated depth: 50m
- Temperature range: 0-50 °C
- pH measurement range: 6.5 – 9.0
- pH accuracy +/- 0.05 pH units
- pH precision: <0.001 pH units
- pH Resolution: 0.0001 pH units
- pH Stability: 0.005 pH units per month

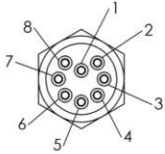


Communication and Power

The SeaFET communicates using a bulkhead connection with pinouts as listed in Table 1.1.1 of the User Manual. For independent deployment, the SeaFET is is connected to a computer using RS232 protocol through a USB convertor for communication and data transfer. For cabled deployment, real-time communication is done through a SeaBird StorX cable connected to the bulkhead.

- Examine bulkhead connectors before every deployment.
- If needed, clean with isopropyl alcohol and then blow dry with air
- Apply a small quantity of 3M Spray Silicone Lubricant to the pin end of the connector. Let it dry.

1.1.1 Bulkhead connector

Contact	Function	MCBH-8-MP
1	Voltage in	
2	Power supply return-signal ground	
3	USB V in	
4	CTD/pump V in (12 V, 650 mA, optional)	
5	TXD/D+	
6	RXD/D-	
7	CTD TXD (optional)	
8	CTD RXD (optional)	

The SeaFET operates powered by 12 D batteries. It can also be deployed with an external power source (e.g., through a StorX cable for communication relay), but the internal batteries must be installed so that the sensing element has uninterrupted power.

- Change the 12 D cell batteries prior to each deployment.
 - Ensure the wet cap is attached to the sensors
 - Remove the vent plug
 - Put the sensor on its side on a flat surface.
 - Put the 8/32" thumb screw into the center thread of the vent plug.
 - Pull on the thumb screw until it does not come out any further.
 - Use a 5/32" hex wrench to remove the three 10-31 x 3/8" screws from the battery end flange.
 - Insert a flat head screwdriver into the slot between the pressure housing and the end flange.
 - Loosen the end flange with the screwdriver until the end flange can be pulled from the pressure housing with one hand.
A safety line attaches the end flange to the battery plate to keep the battery connector wires attached.
 - Press the locking tab on the white end flange connector to disconnect and

gently pull it out.

- Remove the desiccant packs and put them in a sealed bag.
- Remove the two 1/4" nuts that hold the battery plate.
 - Loosen evenly so that battery plate does not get stuck
- Remove and replace batteries
 - Do not mix used and new batteries

- Use only industrial 1.5V alkaline D-cell batteries
- Examine, clean, and if necessary, lubricate the O-rings on the end flange.
- Examine and clean the surface on the inside of the battery compartment where the O-rings sit.
- Replace the desiccant packs.
- Put the battery plate into position and tighten the two nuts equally until the battery plate is flush with the white connector.
- Connect the battery connector. Make sure that it "locks" in the receptacle.
- Install the end flange. Make sure that the wiring is not pinched.
- Use a hex wrench to install the three 10-31 x 3/8" screws.
- Fully insert the vent plug.
- Connect the sensor to the software and turn on the internal batteries.
 - The voltages in the sensor *Dashboard* are 12V and 6V. If they are not, open the sensor and correct the problem.
- Remove batteries if storing the sensor for an extended time

Storage between deployments

Both the SeaFET sensor and the external reference electrode should be kept wet at all times (dry for a maximum of ten minutes). Allowing the sensors to dry can cause leaking of KCl gel, rapid sensor drift, and other damage.

- Clean the sensors using high-purity isopropyl alcohol (90% or higher) and lint-free wipes and swabs.
 - First rinse thoroughly to remove any grit that may scratch the ISFET
 - Polish the external reference electrode using the supplied polishing film.
- Use a dilute hydrochloric acid solution to remove mineral deposits.
- Fill the flow cell with filtered natural seawater and seal with plastic plugs. Do not use artificial seawater.

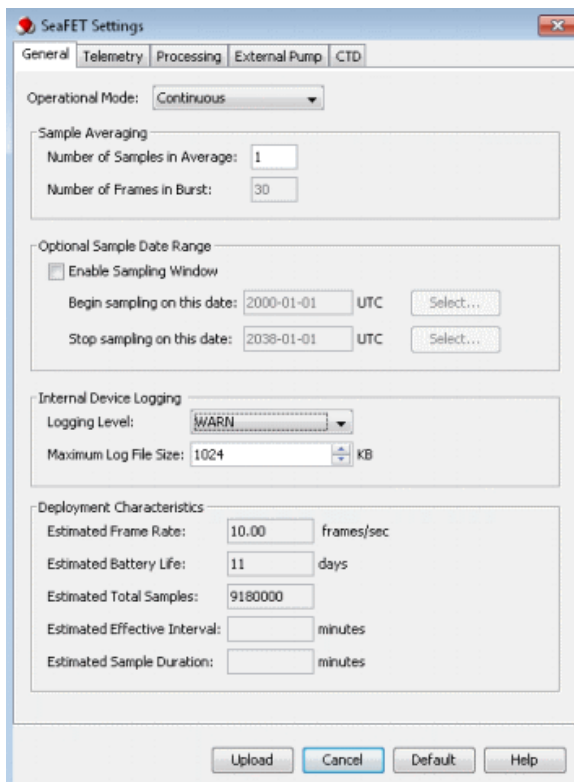
Deployment and Recovery

Preparation

To prepare the sensor for deployment, the following steps must occur:

1. Make sure that the batteries are installed and turned on.
2. Make sure that the sensor is connected to the software and PC and has power.
3. Set the sensor internal clock.
 1. Start the software.
 2. Select Sensor, *Set Clock* from the main menu.
 3. Push Synch Time to align the sensor clock with the clock of the PC.

4. Note that the sensor clock is always set to Coordinated Universal Time (UTC). To change this, go to the *Preferences* menu, then *Display*, and remove the check in the box next to "Use UTC Time."
4. Select the Sensor menu, then *Settings* and change the settings in the General tab.
 1. Set periodic mode. The sensor will collect data at a user-specified sample interval
 1. "Sample Interval": Enter the interval between data collection events: 15 minutes
 2. Sample Averaging
 1. "Number of Samples in Average:" Enter the number of pH measurements that are averaged into a data frame: 10
 2. "Number of Frames in Burst:" Enter the number of data frames in a "burst" of data frames.
 3. "Enable Sampling Window"
 1. Put a check in the box to specify when data collection starts and stops.
 2. Set Sample Date Range
 4. Internal Device Logging
 1. Choose the INFO level of recording, keeping a log (file) of all high-level operations about its performance.
 2. These files are saved as MESSAGES.LOG until they are at the maximum size specified in the "Maximum Log File Size" box. The first file is backed up as MSG_OLD.LOG and MESSAGES.LOG is overwritten.



5. Deployment Characteristics

1. "Estimated Frame Rate:" The frame rate, the data that is output from the sensor per second. Frame rate is inversely proportional to the "number of samples in average."
2. "Estimated Battery Life:" Related to the nominal battery capacity, the number of data collection events and the duration of each event.
3. "Estimated Total Samples:" Related to the frame rate, the number of data collection events, and the duration of each event.
4. "Estimated Effective Interval:" Periodic mode only. Related to the "Sample Interval" setting except when the "Estimated Sample Duration" is longer than the specified "Sample Interval."
5. "Estimated Sample Duration:" Periodic mode only. Related to the "Number of Samples in Average" and the "Number of Frames in Burst" settings.

5. Make a summary report recording status of the sensor
 1. Select Sensor at the main menu, then *Summary Report*.
 2. Enter the "Operator Name" and any "Summary Report Comments."
 3. Put a check in the "View Report" checkbox to see the report in the default .pdf software on the PC.
 4. To change the directory in which the summary is saved, push Browse.
6. Deploy the instrument and disconnect the communication cable.
7. Remove the wet cap that the sensor is stored with.
 1. Unplug the cable from the sensor.
 2. Remove the two stainless steel plugs from the wet cap. Keep them for future use.
 3. Put the sensor over a sink or bucket and empty the solution of artificial seawater from the wet cap.
 4. Put the sensor upright again.
 5. Use a 5/32" hex key to remove the three 10-32 x 5/8" socket head cap screws from the wet cap. Keep the screws to use with the anti-fouling guard.
 6. Remove the wet cap.
 7. Use a clean dry cloth to clean the sensor and the wet cap of any artificial seawater.
 8. Make sure to keep the wet cap, O-rings and stainless-steel plugs for future use.
8. Install the anti-fouling guard and connect the dummy plug.
 1. Put the sensor on a flat surface with the external reference electrode end flange face up.
 2. Put the anti-fouling guard over the ISFET and external reference electrode.
 3. Use a 5/32" hex key to install the three 10-32 x 5/8" socket head cap screws that secured the wet cap.
 1. Make sure to tighten completely.
 4. Disconnect the test cable from the sensor and attach the dummy plug and lock collar onto the connector.

Placement of SeaFET

Exact placement and positioning of the sensor is dependent on location.

Planned deployments include:

Morro Bay T Pier	35°22'10.1"N	120°51'22.2"W
Bay South Station	35°19'58.2"N	120°50'56.0"W

Alternate location: State Park Marina	35°20'45.4"N	120°50'35.9"W
Grassy Bar Oyster Co	35°20'26.6"N	120°50'56.7"W

- When submerging the SeaFET, deploy vertically so that all bubbles escape.
- Then, invert the sensor to deploy facing downwards.
 - The ISFET is sensitive to light – limit exposure when possible.
- Secure SeaFET to piling using large hose clamps

Mid-deployment checks and cleaning

Sensor will be returned to monthly for:

- Collection of discrete samples for calibration
- Biofouling check and cleaning
- Instrument status verification
 - Apply a magnet to the indicator and evaluate per the table:

No Flash: SeaFET V2 batteries/memory are not ready for deployment	Red Flash: SeaFET V2 has not received a sampling command	Green Flash: SeaFET V2 is sampling or waiting to begin sampling
<p>The LED will not flash if any one of the following conditions are true:</p> <ul style="list-style-type: none"> • The RTC battery is not sufficient (below 2.5 V) • The isolated battery is not sufficient (below 4.0 V) • The main battery or external power supply is not sufficient (below 7.0 V) • The memory is full 	<p>The SeaFET V2 batteries and memory are ready to deploy, but the instrument has not received a sampling command.</p> <p>Connect the SeaFET to a computer and click on "Start" in UCI, or send "startnow" or "startlater" before deploying.</p>	<p>The SeaFET V2 batteries and memory are ready to deploy, and the SeaFET V2 has received the "startnow" or "startlater" command.</p> <p>It is either sampling or waiting to begin sampling at the time predetermined in UCI or with the "startdatetime=mmddyyyyhhmmss" command.</p>

Recovery

Upon completion of a deployment prepare for instrument recovery:

- Divers will collect final discrete check sample next to the sensor
- Loosen hose clamps to free SeaFET from piling or post
- Return to surface
- Remove anti-biofouling cap and replace with wet cap filled with seawater
- Return sensor to lab for cleaning and data download.

Instrument Calibration

SeaFETs are exceptionally stable instruments, but need calibration to produce accurate data. This is accomplished through collection of discrete samples alongside the sensor throughout deployment. These data will allow for calibration as well as an estimate of error associated with the data.

Three separate calibrations will be performed.

1. Sensor will be returned to manufacturer for calibration (and maintenance) annually
 - a. Gel cap will be replaced
 - b. External reference electrode is replaced (as needed)
2. Verify calibration in laboratory prior to deployment
 - a. Fill cap with sample
 - b. Record data until stable
 - c. Compare to sample analyzed on spectrophotometric pH instrument
 - d. To save new coefficients to the sensor, go to Menu, Sensor, Advanced, Upload Coefficients.
3. In situ discrete sample calibration

Upon deployment the sensor can be slow to equilibrate. Thus, in situ calibration during deployment, using a discrete seawater sample analyzed in the laboratory is essential.

- Discrete sampling will be completed monthly at a minimum, or whenever the sensor is cleaned.
- An additional sample will be taken directly before instrument recovery.

Discrete sample collection

Equipment – this kit is stored in Dr. Bockmon's lab at Cal Poly. Building 52-D20.

- Niskin
- Yellow Rope
- Bottom Weight
- Trigger Weight
- Notebook & Pen
- Vinyl Tubing
- Headspace syringe
- Grease
- Kim-Wipes
- Black Bands
- White Clips
- Mercury solution
- Mercury pipet & tips
- Safety Goggles
- Gloves

- Sharpie

Procedure

Niskin Deployment from Dock or Boat:

1. Attach Bottom weight to rope and make sure rope is secured to Niskin
2. Cock open both ends of Niskin
3. Pull out nipple to its closed position & tighten top valve
4. Lower Niskin into ocean (1-2 meters) and wait 1 minute for the water to flush
5. Gently deploy trigger weight to close Niskin
6. Record exact time of sample.
7. Pull back up to dock or boat to transfer to bottle.

Alternative Niskin Deployment by Diving:

1. Remove rope and bottom weight from Niskin
2. Cock open both ends of Niskin
3. Pull out nipple to its closed position & tighten top valve
4. Diver submerges, holding Niskin as close to sensor as possible, wait 1 minute for the water to flush
5. Deploys trigger by hand to close Niskin.
6. Record exact time of sample.
7. Surface for transfer to bottle.

Niskin to Sample Bottle:

1. Label the sample bottle with sharpie: YYMMDD Location SampleID
2. Attach Vinyl Tubing to nipple and put the other end into the sample bottle
3. Push in nipple; no water should flow.
4. Slowly open (rotate) the top valve on Niskin to allow water flow
5. As the water flows through the tubing, squeeze & move the tubing to remove all bubbles
6. Rinse: Allow a small amount of water sample into bottle, tighten valve to stop flow, rinse bottle, and pour water out.
7. Start flow into sample bottle again taking care to avoid bubbles.
8. Allow water to gently fill the bottle, and then overflow at least half the sample volume.

Preparing Bottle for Storage/Transport:

1. Put Sample Bottle into secure holder
2. Use syringe to remove water to create headspace
3. Use Kimwipe to dry the inside of the bottle neck
4. Add 120 micro liters of 100% saturated Mercury II Chloride to sample via pipet.
 - a. Or, add 240 micro liters of 50% saturated Mercury II Chloride to sample via pipet.
5. Grease glass stopper, push it into bottle, then rotate until tight to securely seal bottle
6. Pull down black band and connect white clip around bottle neck
7. Invert bottle several times to fix thoroughly

Bottle may be stored in the laboratory for up to 6 months before analysis.

Discrete sample analysis

Equipment

- Custom Dissolved Inorganic Carbon instrument using a Licor 7000 NDIR analyzer
- Custom Total Alkalinity titrator built according to Dickson et al. 2003
- Custom automated spectrophotometric pH instrument using *m*-cresol purple indicator dye.

Procedure

Sample analysis for each bottle starts on pH:

1. Lamps must be on for 45 minutes before they are stable
2. Warm-up instrument and confirm reproducibility (precision).
 - a. Minimum 3-4 replicate junk samples
 - b. Do not start sample analysis until DIC instrument is warm and ready
3. Run a Standard Reference Material (SRM) to check instrument accuracy and provide value for instrument calibration.
 - a. Visually inspect these data before continuing.
 - b. SRMs should be run every ~10 samples, or when concerned about the instrument.
4. Check seal on bottle for salt and cracks in grease before opening
5. Analyze samples in duplicate at a minimum
 - a. Run a third if values are not reproducible or if you suspect instrument problems.
6. Mark date & time opened on bottle.
7. Re-insert stopper.

Pass bottle to DIC for immediate analysis:

DIC provides a backup value and a second parameter to characterize the seawater carbonate system.

1. Warm-up instrument and confirm reproducibility (precision).
 - a. Minimum 3-4 replicate junk samples
2. Run a Standard Reference Material (SRM) to check instrument accuracy and provide value for instrument calibration.
 - a. Visually inspect these data before continuing.
 - b. SRMs should be run every ~10 samples, or when concerned about the instrument.
3. Analyze samples in duplicate at a minimum
 - a. Run a third if values are not reproducible or if you suspect instrument problems.
4. Insert stopper and band & clip.

Perform Alkalinity analysis within 48 hours:

Alkalinity is a conservative parameter not impacted by gas exchange.

1. Warm-up instrument and confirm reproducibility (precision).
 - a. Minimum 3-4 replicate junk samples
2. Run a SRM to check for instrument accuracy.
 - a. Visually inspect these data before continuing.
 - b. SRMs should be run every ~10 samples, or when concerned about the instrument.
3. Begin sample analysis.

- a. If system is running well (both the VSD and fit are good) samples can be run a single time.
 - b. Otherwise run duplicates for value confirmation.
4. When finished with a sample, pouring remaining sample into Junk Seawater Carboy.

Discrete data processing and calibration

Instrument data is processed according to best practices and calibrated using reference materials. The pH data is also corrected for dye purity and dye perturbation. These finalized pH values are used for calibration.

After recovery of the instrument and downloading the data, the data will be adjusted multiplicatively or by offset according to a single calibration point from a discrete bottle pH. See Kapsenburg et al. 2017.

Additionally, data can be further temperature and salinity corrected if data is available.

Quality control and Data Management

Discrete pH samples are also used to assess error; establishing an error envelope for the sensor time-series calculated as a function of the discrete pH error. This error will be reported with the dataset.

Error analysis should be performed, per Bresnahan et al 2015, by creation and analysis of the following plots:

- a. Time-series anomaly plots to identify periods of sensor conditioning, drift, fouling, and failure.
- b. Property-property plots to examine agreement between the sensor pH and independent reference pH values from bottles or other calculated means (e.g. O₂).

For each deployment, once satisfied that the data is robust and well-calibrated, it will be finalized and a report created. The report will include:

- Deployment details including location, dates, and depth.
- Calibration procedures applied to the dataset
- Estimates of accuracy and precision
- Plots of the pH (total pH scale) and temperature vs date & time

The data will be archived as a csv file (easily accessible both by Excel and MATLAB) and stored along with the report in the Cal Poly One Drive. Additionally the raw data, plots, and report will be sent to the NEP and eventually published on the NEP website.

References

Bresnahan, P. J. J., Martz, T. R., Takeshita, Y., Johnson, K. S., and LaShomb, M. (2014). Best practices for autonomous measurement of seawater pH with the Honeywell Durafet. *Methods Oceanogr.* 9, 44–60. doi: 10.1016/j.mio.2014.08.003

Carter BR, Radich JA, Doyle HL, Dickson, AG (2013) An automated system for spectrophotometric seawater pH measurements. *Limnol. Oceanogr.: Methods* 11, 2013, 16–27. doi: 10.4319/lom.2013.11.16

Kapsenberg L, Bockmon EE, Bresnahan PJ, Kroeker KJ, Gattuso J-P and Martz TR (2017) Advancing Ocean Acidification Biology Using Durafet® pH Electrodes. *Front. Mar. Sci.* 4:321. doi: 10.3389/fmars.2017.00321

Sea-Bird Scientific (2018). User manual: SeaFET sensor, pH and optional CTD-DO. Document No. SeaFET170601

Sea-Bird Scientific (2018). Technical Note: Best Practices for the SeaFET™ V2: Optimizing pH Data Quality.

APPENDIX W. MORRO BAY NATIONAL ESTUARY PROGRAM
FRESHWATER SEEPS MONITORING

Morro Bay National Estuary Program Freshwater Seeps Monitoring for Nitrates

Overview:

To assess the impact of the Los Osos Water Reclamation Facility project, monitoring of freshwater seeps along the bay shoreline was begun in the spring of 2014. Sites are monitored on a bi-monthly basis for nitrates as N, with analysis conducted by BC Laboratories.

Sites:

All sites are located along the shoreline in Los Osos. Access is from Third Street at the ramp south of the intersection with El Morro Avenue.

- NPS: North of Pump Station
- SOD: South of Dock
- PSS: Third Street/Pismo Seep
- NPW: North of Pismo Wetland
- SSO: Sweet Springs Outlet

Scheduling:

Monitoring should be scheduled for a low tide to allow access to the shoreline sites. The tide height should be less than 2'.

BC Labs can accept nutrient samples on Monday through Friday. Couriers typically come to Morro Bay on Mondays and Wednesdays. If sampling occurs on a Tuesday or Thursday, they must be overnight shipped to the lab. Sampling cannot take place on a Friday because the lab cannot process samples on a Saturday. The samples have a 48-hour hold time.

Equipment:

- Knee boots
- Cooler and ice packs
- 8 oz. plastic BC Labs bottles
- Sharpie
- YSI 2030 meter
- DI water

Protocol:

- At each site, measure specific conductance and salinity.
- Label the bottle.
- Collect a sample, ensuring that it is free of sediment.
- Store in cooler.

At the office, complete a chain of custody form for the lab. Fill out the date and time of sample collection. Sample is picked up by a courier for delivery to the lab.

APPENDIX X. MORRO BAY NATIONAL ESTUARY PROGRAM MASTER SITE LIST

Site Code	Site Description	Latitude	Longitude	Monthly Water Quality	Bacteria	Pressure Transducer	Dawn Patrol	Bi-monthly Nutrients	Continuous Temperature	Instantaneous Flow	Suspended Sediment	Bioassessment	Stormwater	Seeps	Sediment Elevation	Stream Profiling
310CCC	Chorro Creek upstream from Chorro Flats	35.35762	-120.812	X	X	X			X	X						
310CPN	Pennington Creek @ bridge	35.3453	-120.729	X	X	X			X	X						
310PEN	Pennington Creek @ Rancho El Chorro	35.33999	-120.736	X	X					X						
310DAM	Dairy Creek near dog park	35.33558	-120.728	X	X			X		X		X				
310SLU	San Luisito Creek @ Adobe Rd	35.35595	-120.786	X	X			X	X	X						
310TWB	Chorro Creek at South Bay Boulevard	35.35433	-120.828	X	X			X		X		X				
310UCR	Chorro Creek @ upper Chorro Creek Ecological Reserve	35.33862	-120.768	X	X			X	X	X						
310BAY	Morro Bay Shoreline at Baywood Pier	35.32678	-120.841		X											
310CIN	Morro Bay Shoreline at Cuesta Inlet	35.31986	-120.849		X											
310COL	Morro Bay Shoreline at Coleman Beach	35.3722	-120.86		X											
310PAS	Morro Bay Shoreline at Pasadena Point	35.32999	-120.844		X											
310SIN	Morro Bay Shoreline at Sharks Inlet	35.31805	-120.857		X											
310SPM	Morro Bay Shoreline at State Park Marina	35.34627	-120.843		X											
310TID	Morro Bay Shoreline at Tidelands Park	35.35982	-120.852		X											
310WIN	Morro Bay Shoreline at Windy Cove	35.34934	-120.844		X											
310GS1	Turri Rd. 0.35 mi from South Bay Blvd	35.33382	-120.819	X	X			X		X						
310APN	Pennington Creek upstream from horse corral	35.35371	-120.722							X						
310ATP	Morro Bay offshore at Tidelands Park	35.35738	-120.851				X									
310CAN	Chorro Creek at Canet Road	35.35315	-120.789								X					
310CHD	Chorro Creek below Chorro Creek Dam	35.3343	-120.689									X				
310CHI	Morro Bay offshore at North of Cuesta Inlet mouth	35.32375	-120.855				X									
310CLK	Los Osos Creek Clark Valley Road	35.28187	-120.79									X				
310CSI	Morro Bay offshore at Near Cuesta Inlet	35.32042	-120.85				X									
310DAL	Dairy Creek at bottom of El Chorro Regional Park	35.33039	-120.73													
310DAU	Dairy Creek above Cattle Enclosure	35.34752	-120.715						X			X				
310LO2	Morro Bay offshore at Los Osos Creek outlet	35.336	-120.841				X									
310LSL	San Luisito Creek Lower	35.35961	-120.775									X				
310MNO	San Bernardo Creek Middle	35.37584	-120.791									X				

310MSL	San Luisito Creek Middle	35.36241	-120.757																
310OSL	San Luisito Creek above upper site	35.37027	-120.739																
310PSP	Morro Bay offshore at Pasadena Point	35.3337	-120.848																
310SHI	Morro Bay offshore at Sharks Inlet	35.3144	-120.866																
310SPO	Morro Bay offshore at State Park Marina	35.3468	-120.845																
310SYB	Los Osos Creek @ Turri Rd	35.33065	-120.817																
310USL	San Luisito Creek Upper	35.36592	-120.751																
310UWR	Warden Creek 1.5 mi upstream from Turri Rd	35.29384	-120.753																
310WLB	Walters Creek @ bridge	35.3466	-120.755																
310WLF	Walters Creek @ flume	35.34811	-120.75																
310WLM	Walters Creek downstream 0.2 mi from bridge	35.34578	-120.757																
310WRP	Warden Creek @ Wetlands Restoration Preserve	35.3206	-120.806																
310PFL	Pennington at the former Fish Ladder	35.34464	-120.73																
310SBE	San Bernardo Creek @ Adobe Rd	35.36174	-120.805																
310UPN	Pennington Creek @ corrals	35.34798	-120.726																
310CER	Chorro Creek in the Ecological Reserve	35.3474	-120.773	X															
310COO	Coon Creek at Pecho Valley Road	35.25423	-120.885	X															
310LVR	Los Osos Creek at Los Osos Valley Rd.	35.30589	-120.812	X															
310CHO	Chorro Creek at Highway 1 bridge on Camp SLO	35.32197	-120.722	X															
310CLV	Los Osos Creek 1.0 mi downstream from 310CLK	35.28809	-120.802	X															
310TUR	Warden Creek @ Turi Road	35.30231	-120.776	X															
310ACR	Chorro downstream of CMC WWTP on CDFW	35.32822	-120.758																
TBD	AboveReservoir	35.34552	-120.683																
TBD	UpperPennington	35.36246	-120.718																
310SSO	Sweet Springs Outlet freshwater seep	35.32288	-120.841																
310NPW	North Pismo Wetland freshwater seep	35.32374	-120.841																
310PSS	Pismo freshwater seep	35.32418	-120.841																
310SOD	South of Dock freshwater seep	35.32494	-120.84																
310NPS	North Pump Station freshwater seep	35.3257	-120.84																
B High	Marsh	35.34717	-120.833																
C High	Marsh	35.34787	-120.829																
A High	Marsh	35.34585	-120.833																
B Low	Marsh	35.34115	-120.837																
A Low	Marsh	35.34183	-120.842																
C Low	Marsh	35.33852	-120.83																
F Mud	Mudflat	35.3189	-120.853																
G Mud	Mudflat	35.3167	-120.862																
E Mud	Mudflat	35.32582	-120.841																

B Near	Mudflat	35.34032	-120.836																X
B Far	Mudflat	35.3398	-120.836																X
C Near	Mudflat	35.33807	-120.831																X
C Far	Mudflat	35.33787	-120.832																X
A Near	Mudflat	35.34162	-120.843																X
A Far	Mudflat	35.3415	-120.843																X
D Mud	Mudflat	35.33253	-120.839																X
CHN 01	Chorro Creek stream profiling site	35.35334	-120.829																X
CN2	Chorro Creek stream profiling site	35.35223	-120.83																X
CS2	Chorro Creek stream profiling site	35.35207	-120.829																X
CU1	Chorro Creek stream profiling site	35.33379	-120.69																X
CU2	Chorro Creek stream profiling site	35.33297	-120.691																X
CU3	Chorro Creek stream profiling site	35.33149	-120.692																X
CU4	Chorro Creek stream profiling site	35.33052	-120.694																X
CU5	Chorro Creek stream profiling site	35.32909	-120.696																X
DAM0	Dairy Creek stream profiling site	35.34027	-120.721																X
DAM1	Dairy Creek stream profiling site	35.34026	-120.721																X
DAM2	Dairy Creek stream profiling site	35.33792	-120.724																X
DAM3	Dairy Creek stream profiling site	35.3369	-120.727																X
DAM4	Dairy Creek stream profiling site	35.33662	-120.727																X
DAM5	Dairy Creek stream profiling site	35.33597	-120.727																X
PEN1	Pennington Creek stream profiling site	35.33776	-120.739																X
PEN2	Pennington Creek stream profiling site	35.33727	-120.74																X
PEN3	Pennington Creek stream profiling site	35.33637	-120.74																X
PEN4	Pennington Creek stream profiling site	35.33615	-120.741																X
PEN5	Pennington Creek stream profiling site	35.33483	-120.743																X
XS1	Chorro Flats stream profiling site	35.36041	-120.818																X
XS3	Chorro Flats stream profiling site	35.35899	-120.815																X
XS4	Chorro Flats stream profiling site	35.35836	-120.814																X
XS5	Chorro Flats stream profiling site	35.3581	-120.815																X
XS6	Chorro Flats stream profiling site	35.36026	-120.821																X
CS3	Chorro Flats stream profiling site	35.35196	-120.829																X