



## Standard Operating Procedure

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Revision History		
Version No.	Effective Date	Description
1.0	01/01/2024	Original composition by M. Kachmar

**Procedure Owners:**

**Date:**

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## **1 Purpose**

The purpose of this SOP is to provide concise guidance and methodology for deploying and storing the Aqua Troll 600 data sonde(s) on a monthly basis to accurately collect temperature, salinity, pH, dissolved oxygen (DO), and chlorophyll-a data to characterize site water quality.

## **2 Scope**

This SOP is pertaining to the EPA Long Island Sound Study funded Oyster Health project that is incorporating water quality metrics to understand host-pathogen-environment relationships.

## **3 Definitions/Acronyms**

## **4 Safety Precautions**

All team members will wear appropriate clothing dependent on weather conditions including but not limited to waders, rubber boots or protective footwear, gloves, hats, sunglasses, long sleeve shirts, and pants. Thick protective gloves (e.g garden gloves) should be worn when handling fouling organisms. Team members will wash hands thoroughly after field trips end. A first aid kit will be present for any injury. Extra water will be provided to avoid dehydration or heat stroke. Team members will take regular breaks when needed.

Exercise weather-appropriate field safety measures by monitoring conditions before and during the trip. Do not perform fieldwork during dangerous conditions (e.g. lightning, extreme winds, extreme floods). Do not visit field sites alone (use buddy system). Inform PIs of dates and times of fieldwork. Confirm safe return to the lab. At intertidal sites, perform procedures during low tide. At subtidal sites, divers are to follow NOAA diving regulations according to the instructions of the lab diving coordinator (Barry Smith).

## **5 Reagents/Media**

1. HCl
2. NaOH
3. Isopropyl alcohol

## **6 Supplies/Materials**

1. D batteries (Energizer or Durcell)

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2.  $\frac{15}{16}$  inch socket wrench
3.  $\frac{15}{16}$  inch open-end wrench
4. Adjustable wrench
5. O-rings
6. Sonde supply box
  - a. High vacuum grease
  - b. RDO sensor Calibration sponges
  - c. Extra pins and bolts for housing
  - d. Desiccant tube
  - e. Screw drivers
  - f. Hex driver
  - g. Allen key
  - h. Sonde wipers (Aqua Troll part #0078940)
  - i. Alcohol pads
  - j. D batteries (minimum of 2, Energizer or Duracell only).
  - k. Paper towels (for collecting discarded pH filling solution).
  - l. Q-tips or cotton buds
7. Towel / cloth for wiping things down.
8. Dish scrubbing pad
9. Scrub brush
10. Shucking knife
11. 500 mL empty container with sealable top
12. First aid kit
13. Hand sanitizer
14. Wire cutters
15. Gray floating tray
16. Robust zip-ties
17. PVC housing with screw cap
18. Mushroom anchors
19. Bolts (2 per site)
20. Hose clamps
21. Rope
22. NOAA buoy

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- 23. Mild soap (Dawn dish soap)
- 24. Distilled water
- 25. Deionized water
- 26. Fresh running water (sink)
- 27. Factory supplied boxes

## 7 Equipment

- 1. Field tablet (containing VuSitu app)

## 8 Quality Control

All team members will be trained to complete all field tasks, including training on data entry requirements for each specific task. Environmental monitoring data will be regularly checked for drift and sensors will be calibrated when necessary.

## 9 Procedures

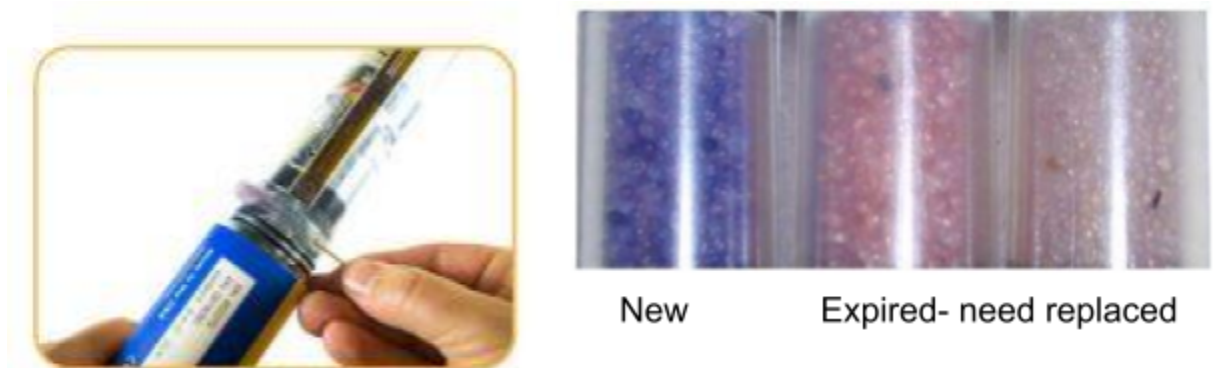
### 1. Sonde Assembly

- a. Once you receive your equipment and unbox, the first step is to install the batteries. The AquaTroll 600 takes alkaline D batteries (preferably Energizer or Duracell - factory recommendation).
  - i. To install the batteries, open the battery compartment (Figure 1) by twisting. Install the batteries into the sleeve.
  - ii. Use an Allen wrench to remove and check the desiccant compartment/color (Figure 2). If there is no desiccant installed, insert a new desiccant pill. If the present desiccant is pink, that is the indicator that it needs to be replaced.
  - iii. Once the batteries are installed and desiccant is inserted, add vacuum lubricant to create a seal when closing the battery compartment. If the batteries are installed properly, the LCD screen should activate and illuminate blue.

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**Figure 1:** Anatomy of the Aqua Troll 600.



**Figure 2:** Demonstration of removing desiccant pill (left). Color indicators of desiccant pill replacement status. Pink color indicates that the desiccant pill should be replaced (right).

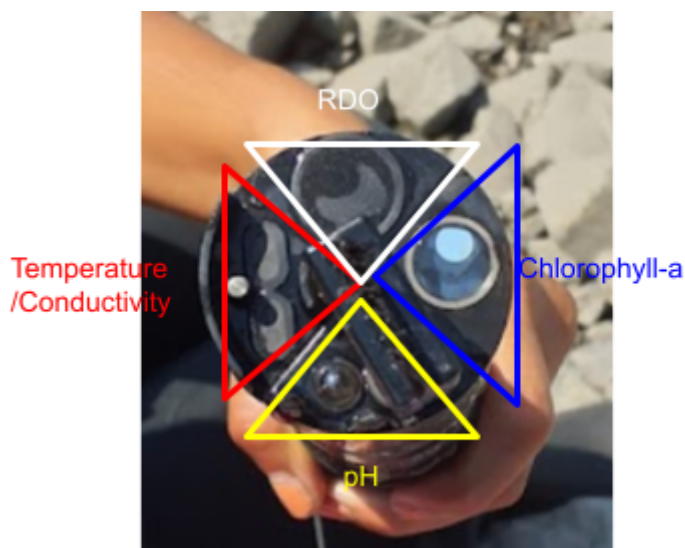
- b. Next, Install the wiper motor and sensors to the appropriate ports.
  - i. Remove the metal guard (often called the restrictor) exposing the ports.
  - ii. Add vacuum lubricant to the O-rings of the wiper motor and install the motor into the very center port.

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**Figure 3:** Wiper parts and where to install them into the sonde port.

- iii. Once the wiper motor is secured into the port, obtain the sensors. Add lubricant to the O-rings of the sensors.
- iv. Align the sensors with the interlocking grooves in the wiper motor and slide the sensors, one at a time, into the ports. Configure the sensors as shown in Figure 4.
- v. Tighten the small screws at the bottom of the sensors.



**Figure 4:** Sensor identification and setup configuration.

*c. Note about sensors: the RDO sensor requires a 'fast cap' (product #0038520). This will come with the sensor and should be installed prior to*

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*sonde installation. The wiper can reduce the life of the cap so be sure to monitor any damage to replace the cap when necessary.*

- d. For newly received sondes that have not been previously deployed or factory calibrated, Calibrate the sensors following the [“Sonde Calibration” SOP](#).
- e. To connect to the tablet using the VuSitu app. Hold the sonde vertically with the sensors facing upward. the screen will activate indicating the bluetooth connection is available.
- f. Set up the sonde:
  - (i) When connected to the VuSitu app. Click connect and then the serial number associated with the device.
  - (ii) In the VuSitu app, click *Instrument Settings*.
  - (iii) Check the Instrument Clock settings (apply mobile device time to the sonde), and check the Wiper Settings (activate wiper every reading). Other settings can be set when creating a logging session.
  - (iv) Go back to the home screen via arrow prompts at top left of screen.
- g. Initiate a new logging session:
  - (i) On the home screen of the connected VuSitu app, click *Logging*.
  - (ii) On the *Logging* screen, click *New Log*.
  - (iii) Follow the prompts to create a new logging session.
    - Choose to record all parameters,
    - Give the session a name (Log\_MMYYSiteCode; eg. Log\_0423ASHC)
    - Choose the location (add new location name, if necessary; It’s ok to ignore lat/long info), (eg. site name = FENC, ASHC, GOLD, or STAR)
    - Set salinity to automatic (method used for calculating DO),
    - Choose logging method = linear,
    - Record new measurement every 30 min,
    - Start logging = manual start (or scheduled if needed),
    - Stop logging = manual stop,
    - Log wrapping = overwrite old data in this log when memory is full,



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- **Save** and follow any further prompts

## 2. Deploy the data sonde and housing

*a. Note: If transporting the sonde between the lab and the field site, keep the sensors wet/moist during transport following the steps in [Section 9.3.a](#).*

### b. Housing Deployment:

- i. For details describing how to make sonde housing see [Appendix A](#). See Figure 5 for example of intertidal site sonde housing. See Figure 6 for example of subtidal site sonde housing.



**Figure 5:** Setup of the sonde stations deployed at intertidal sites (Ash Creek and Fence Creek) showing the the PVC screw cap (top arrow), which the sonde is tethered to; the top security bolt passing through the PVC housing (middle arrow); and the bottom bolt, which the sonde sits



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upon.



**Figure 6:** Depiction of subtidal sonde housing and deployment method.

**c. Sonde deployment:**

- i. *Before deploying the sonde in the field, make sure that the copper guard (covering the probes) is attached in the data logging position and that the sonde has been set to record data as described above in Section 9.1.f.*
- ii. The copper guard vent holes should be at the tip of the sonde, allowing external water to flow across the sensors (Figure 7).

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Copper guard holes near the sensors to allow water flow.

**Figure 7:** Proper configuration of the copper guard for deployment.

- iii. If it is in calibration/storage position with the copper guard vent holes near the middle of the sonde, preventing external seawater flowing over the sensors, unscrew the copper guard from the sonde, reverse it, then screw it back on in the data logging position. The black screw cap of the copper guard will also need to be unscrewed and repositioned at the free end.
- iv. **Intertidal sites:**
  1. **At Ash Creek and Fence Creek:**
    - a. Wade to the sonde station. Insert the sonde into the PVC housing and gently lower it with the probes facing downward. It will sit on a cross bolt threaded near the bottom of the PVC housing.

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- b. Screw the PVC cap (tethered to the data sonde) into the top of the PVC housing. Don't over-tighten it.
- c. Insert the stainless steel security bolt through the top of the PVC housing through the two holes drilled near the top of the housing, then screw on its corresponding nut until it's snug. *Note: At Fence Creek, the locking nut does not need to be tight against the PVC housing.* (Figure 8)



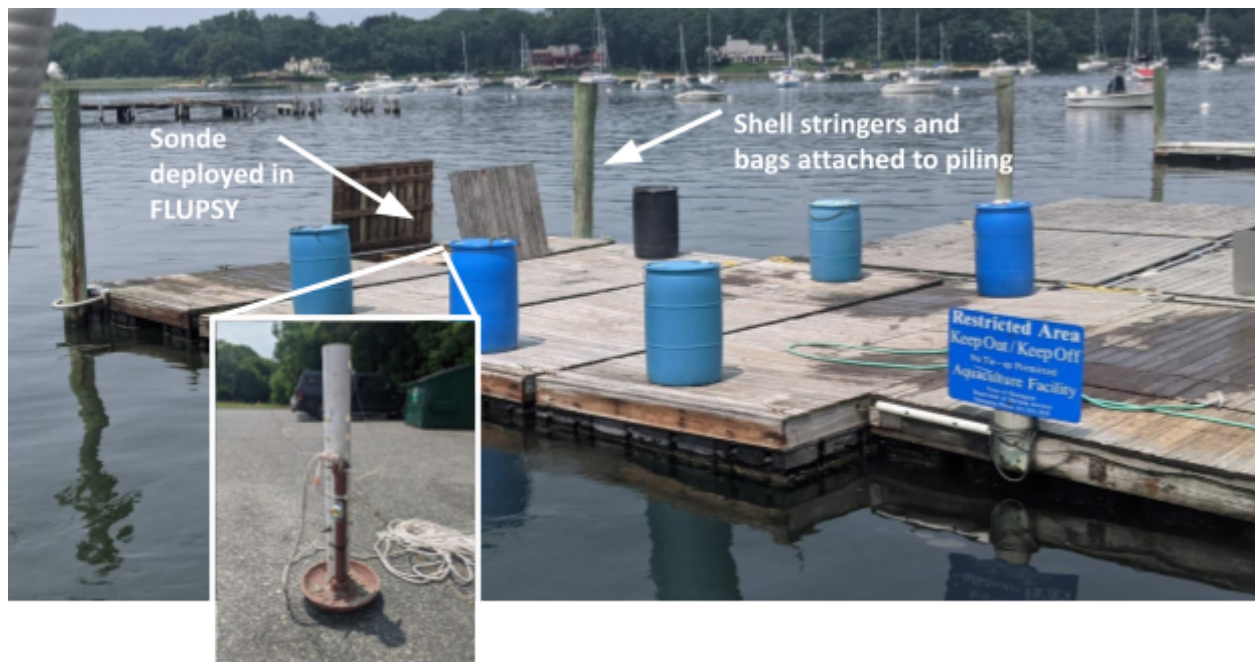
**Figure 8:** Depiction of inserting the sonde into the housing, closing the cap, and screwing the bolt into place at intertidal oyster bed sites.

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## v. Subtidal sites:

### 1. At Gold Star Beach (Figure 9)

- Place the sonde inside the PVC housing with the probes facing downwards.
- Clip the tether from the anchor onto the top of the data sonde.
- Pass a zip-tie through one of PVC vent holes and out another vent hole (both about half way up the sonde) and use it to secure the sonde tight against the inside of the PVC housing.
- Slowly lower the mushroom anchor to the seabed and fasten it on a cleat. (want to be sure the sonde and housing remain in an upright position during deployment)



**Figure 9:** Map showing approximate location of all gear deployments at Gold Star. Inset shows an image of the sonde housing secured to the mushroom anchor.

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3. Sonde storage:

a. Short term (Figure 10)

- i. Short term storage should be used for transportation or holding less than a week.
- ii. Before transporting the sonde to or from the field, remove the copper guard from the sonde and the end cap. Put the guard on the Aqua Troll Sonde in the 'calibration mode' position. The holes on the guard should be closest to the screen.
- iii. Pour ~15mL of clean water (fresh or seawater) into the copper guard to keep the sensors moist during transportation. Cap the copper guard. (This will keep sensors from drying out)



**Figure 10:** Steps to put the sonde into short term storage.

b. Long term

i. Cleaning

1. Before the sonde can be fully stored for long term purposes, all parts of the sonde (Figure 1) and the sensors should be thoroughly cleaned.
2. Do not remove the sonde from short term storage set up (section 3.a) until the body of the sonde has been thoroughly cleaned to protect the integrity of the sensors. The body of the sonde (battery compartment, screen, copper guard, cap) can be cleaned by removing any biofouling using a scrub brush or shucking knife. Rinse the sonde thoroughly and clean with water and mild soap. Rinse the sonde again and allow it to air dry.
3. Once the body of the sonde has been cleaned and dried, the copper guard and cap can be removed. First, remove the cap and drain the water from the housing. Then, remove the copper guard. Rinse the sensor area with fresh water to remove any mud or loose dirt from the area.
4. Cleaning the sensors:



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- a. Remove the wiper and discard. Using the thin screwdriver (in the sonde supply box), unscrew the wiper from the wiper poll. The screw is very small and is in the bottom corner adjacent to the wiper poll. If fouling is covering the screw, gently remove fouling from that area using a shucking knife. *Be sure to never touch the probe sensors with the shucking knife to avoid damage.* Gently lift the wiper up off of the wiper poll and discard.
- b. Using Q-tips gently remove fouling from the sensors. Rinse periodically with water.
  - i. *The pH sensor should not be directly touched using the Q-tips. Only clean the area around the sensor.*
  - ii. Conductivity, Temperature, Chlorophyll-a, and RDO can be gently wiped using the Q-tip and water.
  - iii. If there are calcified organisms in crevices, use the shucking knife or Hex screwdriver to remove. *Do not use these tools to remove calcified animals directly on sensor screens. (see section 9.3.b.iii for removal)*
5. Once sensors are clean, remove fouling from the inside of the copper guard and cap using a scrub brush, warm water, and mild soap. Pat excess water and air dry.
- ii. Taking apart:
  1. Using a hex screwdriver, loosen the screws at the bottom of each sensor. The screws will not fall out.
  2. Once the screws have been loosened, gently push the desired sensor upward from the bottom where the ridges are. The sensor may feel stuck due to the vacuum seal around the connection probe and port.
  3. Repeat for all sensors.
  4. Once the sensors are removed they will require individual maintenance (see section 9.3.b.iii. below).
  5. Next, remove the wiper pole from its port. Gently pull the wiper poll out. Clean the wiper pole by wiping down with a damp paper towel. *Be careful not to get water into the probe area.*

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6. Gently and carefully use a damp paper towel to wipe any dirt or mud from around the port grooves. *Do not get water in the ports.*
- iii. Individual sensor maintenance and storage:
  1. pH sensor:
    - a. If the ORP platinum electrode is dull or dirty, it can be cleaned with a swab soaked in methanol or isopropyl alcohol. Rub the electrode until it is shiny.
    - b. The pH probe must be kept moist for the life of the sensors.
    - c. *Do not directly wipe the glass bulb.* To clean the pH sensor, gently rinse with cold water. If further cleaning is required, consider the nature of the debris.
    - d. To remove crystalline deposits:
      - i. Clean the sensor with warm water and mild soap. Soak the sensor in 5% HCl solution for 10 to 30 minutes. If deposits persist, alternate soaking in 5% HCl and 5% NaOH solutions (*See Appendices B & C for reagent recipes*).
    - e. To remove oily or greasy residue:
      - i. Clean the sensor with warm water and mild soap. Methanol or isopropyl alcohol may be used for short soaking periods, up to 1 hour. *Do not soak the sensor in strong solvents, such as chlorinated solvents, ethers, or ketones, such as acetone.*
    - f. To remove protein-like material, or slimy film:
      - i. Clean the sensor with warm water and mild soap. Soak the sensor in 0.1 M HCl solution for 10 minutes and then rinse with deionized water. (*See Appendix D for reagent recipe*)
    - g. After cleaning, rinse the sensor with water, and then soak overnight in a pH 4 buffer (Apera Instruments, SKU AI1102).
    - h. Storage:
      - i. Obtain the sensor storage cap, and dampen the sponge inside with the pH Storage Solution or pH 4 calibration standard. Cap the sensor and use electrical tape to seal the storage cap.



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*Do not store the pH sensor in DI water because it will deplete the reference solution and drastically reduce the life of the sensor.*

- ii. Add the blue cap to the port.

### 2. RDO:

- a. Leaving the sensor cap, rinse the sensor with clean water. gently wipe with a soft cloth or brush if biofouling is still present. If extensive fouling or mineral buildup is present, soak the sensor in vinegar for 15 minutes, then soak in deionized water for 15 minutes. *Do not use organic solvents—they will damage the sensor cap. Do not remove the sensor cap when rinsing or brushing.*
- b. After cleaning the sensor, perform a 2-point calibration ([see Sonde Calibration SOP](#)).
- c. To clean the optical window, remove the cap and gently wipe the sensing window with the lens cloths. *Do not wet the lens with any liquid.*
- d. Storage: store the sensor body and cap in the factory supplied containers.

### 3. Conductivity/Temperature:

- a. To clean the conductivity sensor face, gently rinse with clean, cold water. If further cleaning is required, consider the nature of the debris.
- b. To remove crystalline deposits:
  - i. Clean the sensor face with warm water and mild soap. Use a soft brush to gently clean the sensor pins and temperature button. Ensure removal of all debris around the base of the pins and button. If crystalline deposits persist, soak in 5% HCl for 10 to 30 minutes followed by warm soapy water and soft brushing. If deposits persist, alternate soaking in 5% HCl and 5% NaOH solutions followed by warm soapy water and soft brushing.
- c. To remove oily or greasy residue:
  - i. Clean the sensor face with warm water and mild soap. Using a soft brush, gently clean the sensor pins and temperature button. Ensure

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removal of all residue around the base of the pins and temperature button. Isopropyl alcohol may be used for short soaking periods, up to one hour. *Do not soak in strong solvents such as chlorinated solvents, ethers or ketones (such as acetone).*

- d. To remove protein-like material, or slimy film:
  - i. Clean the sensor face with warm water and mild soap. Using a soft brush, gently clean the sensor pins and temperature button. Ensure removal of all material/film around the base of the pins and temperature button. Soak the sensor in 0.10% HCl for 10 minutes and then rinse thoroughly with distilled water.

- 4. Storage: Prior to installation, store the sensor in the factory supplied container. Once installed on the sonde, the Temperature Sensor and Conductivity Sensor can be stored wet or dry depending on the sensor configuration of the sonde.

- iv. Storing the Sonde:
  - 1. The sonde and its assigned sensors should be placed into the factory provided box. Each box will have a serial number. Be sure to box the appropriate sonde based on the serial number on the box. The serial number on the sonde can be found near the screen.

## 10 Waste Disposal

1. Throw away all the dirty paper towels. Wash or rinse and cloth towels.
2. Rinse and wipe out the sink.
3. HCl and NaOH should be disposed of in their respective chemical waste containers that are appropriately labeled.

## 11 References

## 12 Appendices

- A. Sonde housing structure and deployment needs
  - a. Intertidal sites:

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- i. The sonde will need protective housing for deployment and a place of anchor. At the current intertidal sites there are no docks so a structure must be built and deployed (see Figure 4) .
    - ii. The structure is a PVC pipe with a screw cap that is attached to a metal pole adhered to a cement tire. The pole and cement tire are meant to act as the anchor structure.
    - iii. The PVC housing is attached to the metal pole using stainless steel hose clamps.
    - iv. The PVC pipe has holes drilled throughout to allow for water flow.
    - v. A red flag attached to a thin PVC pole should be tethered to the structure using heavy duty zip ties to be able to identify the location of the sonde at various tide levels.
    - vi. Additionally, there are two bolts (Figure 4) that will be installed in the top and bottom of the PVC pipe housing. The bottom bolt is used as a shelf for the sonde to sit. The top is for security and anti-theft.
  - b. Subtidal sites:
    - i. A 20lb mushroom anchor will be used to suspend the sonde subtidally near the oyster beds.
      1. This mushroom anchor will have rope attached to it to properly pull to the surface (Gold Star beach) or to attach a buoy at the surface (Laurel Hollow). See below in Section C.
    - ii. PVC housing will be attached to the mushroom anchor using heavy duty zip-ties. The housing has holes drilled into it for proper water flow. Additionally, there are two bolts that will be installed in the top and bottom of the PVC pipe housing. The bottom bolt is used as a shelf for the sonde to sit. The top is for security and anti-theft.
- B. 5% HCl Solution**
- a. Use the calculation  $C1V1 = C2V2$  to determine how much HCl concentrate should be mixed with water to create a 5% HCl solution.
    - i. ex: Starting % HCl (Volume 1) = 5% HCl (740ml)
    - ii. So, for a solution of 37% HCl, add 640 ml of water to 100ml of HCl solution.
  - b. Acid should be added to water slowly.

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### C. 5% NaOH Solution

- Use the calculation  $C_1V_1 = C_2V_2$  to determine how many grams NaOH should be mixed with water to create a 5% NaOH solution.
- A 5% solution of NaOH is equivalent to 5g of NaOH dissolved in 100ml of water. (50 g in 950 ml of water)

### D. 0.1M HCl Solution

- Use the calculation  $C_1V_1 = C_2V_2$  to determine how much HCl concentrate should be mixed with water to create a 0.1M HCl solution.
- A 37% HCl solution has a molarity of 12.08M.
- Acid should be added to water slowly.

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