



Standard Operating Procedure

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

Procedure Owners:

Date:

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Date:

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1 Purpose

The purpose of this SOP is to provide concise guidance and methodology for quality monthly oyster sample collections from intertidal and subtidal oyster beds for disease analysis and reproductive evaluation.

2 Scope

This SOP is pertaining to the EPA Long Island Sound Study funded Oyster Health project that is incorporating disease analysis and reproductive evaluations to understand oyster population health within the Long Island Sound.

3 Definitions/Acronyms

4 Safety Precautions

All collection 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 oysters. 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, barry.smith@noaa.gov).

5 Reagents/Media

None

6 Supplies/Equipment

1. Collection Permit(s)
2. Field Permission letter(s)
3. Field backpack
 - a. Field tablet

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- b. First aid kit
- c. Povidone Iodine scrub
- d. Hand lotion
- e. Bug spray
- f. Sunscreen
- 4. Cooler with ice
- 5. Digital calipers
- 6. Labeled onion/string bags
- 7. Spare onion/string bags
- 8. 5-gal buckets (4-6 total)
- 9. Spare rebar stakes
- 10. Towel/Cloth
- 11. Shucking knives
- 12. Scrubbing brushes
- 13. Waders/ boots
- 14. Working gloves
- 15. Waterproof jacket / pants
- 16. Waterproof top
- 17. Large bottle with water
- 18. Diving:
 - a. Towable dive flag
 - b. Diver collection bags (x2)
 - c. Personal Flotation Device (PFD) for row boat
 - d. Wire cutters on lanyard
 - e. Large rubber bands
 - f. Dive tanks
 - g. AED
 - h. Oxygen supply

7 Quality Control

All team members will be trained to complete all field tasks, including training on data entry requirements for each specific task. Oysters are targeted using standard size requirements of 80-120mm with the exception of Gold Star beach ($\geq 40\text{mm}$) due to the planted population being a single cohort. To ensure completeness, [field notebooks](#) will include a checklist of all data that needs to be recorded during each visit. All datasheets will be screenshotted as back up in the event data is lost before connecting to the network.

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8 Preparation (at lab)

1. Follow the procedures for booking field work in advance using the "[Booking Field Work](#)" SOP.
 - a. Field work for the season should be pre-scheduled and booked at least one month in advance. All intertidal field trips are dependent on the best low tide (mean low water) of the month. Ideally the entire season should be pre-booked (March - November).
 - b. All field dates should be coordinated with the Oyster Health team and, for subtidal sites, NOAA divers.
 - c. If the field visit is on Long Island, NY - be sure that all ferry tickets have been purchased and printed for easy travel.
2. The week prior to the field visit make sure to contact all site hosts and collaborators that sampling is happening and team members will be at the site during specific time periods. Include the list of days for proceeding months as well.
3. Coordinate with the Oyster Health team to have a minimum of 2 individuals visiting a site. A 3 person team is ideal for safety.
4. Prepare a monthly field plan for the team describing clear goals of the trip.
5. The day before the field visit obtain the appropriate and pre-reserved government vehicle to load with necessary equipment listed in the supplies and equipment list.
6. Additionally, make sure the government vehicle has adequate gas for the specific trip.
7. Fully charge the field tablet and make sure all appropriate SOPs and [field notebooks](#) are available offline.

9 Procedures

1. **Before entering the water, inform PI's that the team has arrived at the site and are about to enter the water. This is a required safety precaution in the event of an accident.**
2. A total of 30 oysters will be randomly collected from each site in addition to 3-4 extras in the event of a causality (eg. stabbing during opening or mud oyster).
Note: Collection techniques will be slightly different between and among intertidal and subtidal that will be specified below.
3. The targeted height (Figure 1) of the oysters being collected is 80-120 mm with the exception of Gold Star beach ($\geq 40\text{mm}$) as the diseases of interest target

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oysters within and above this size/age class. Use the standardization guide or digital caliper to reference the appropriate size of oysters.

4. Ideally, try to collect individual oysters or individuals that can easily be broken off the clump rather than oysters that are heavily clustered together from settlement. This will reduce mortality from attempting to remove oysters from one another. If this becomes difficult, use a shucking knife to gently remove oysters from one another.
5. At the end of each collection, [field notebooks](#) should be updated describing the tasks that were completed and any anomalies that occurred while in the field.

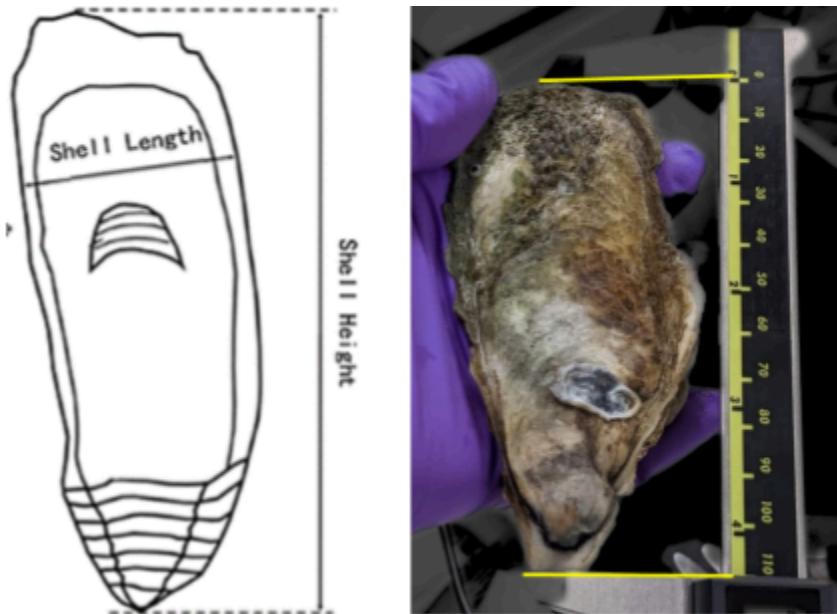


Figure 1: Shell measurements to be collected using calipers. Schematic showing height and length measurement landmarks (left). Shell height measurement on live oyster (middle).

6. Intertidal Sites:

Note: Only one person should be walking across the oyster bed at a time to reduce damage and disturbance of the bed.

a. **Ash Creek (Fairfield, CT):**

- i. To access Ash Creek oyster bed, there is a path that leads directly from the parking lot to the shoreline where the bed begins.

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- ii. In the parking lot, make sure all equipment is accounted for and everyone has put on the appropriate gear (waders, boots, gloves, etc).
- iii. Carry all equipment and supplies down the path and to the shoreline.
- iv. Ash Creek is broken up into two sections of the oyster bed (Figure 2 & 3).
- v. Collections will take place on both portions of the oyster bed. (Figure 3)

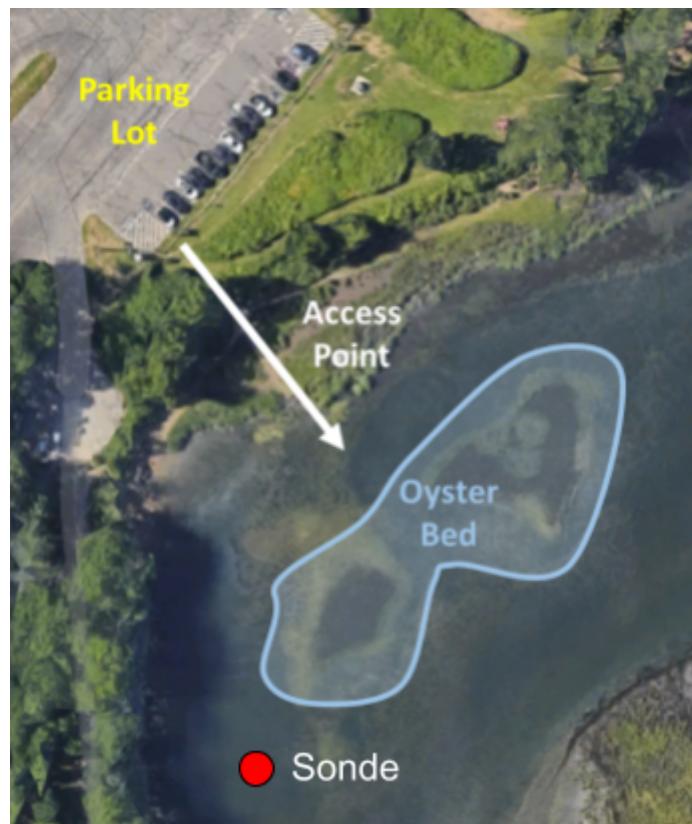


Figure 2: Access point and visualization of the Ash Creek oyster bed.

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Figure 3: Photo taken from the access point at Ash Creek oyster bed, showing the two portions of the oyster bed (right and left).

- vi. Obtain buckets and shucking knives and wade/walk to the right or left portion of the oyster bed to collect samples. *Note: The channel between the right and left portion can become deeper (hip to chest height) depending on the mean low water.*
- vii. 33 oysters will be collected over both portions of the oyster bed (30 for processing and 3 extra). A total of 15 oysters will be randomly selected across the oyster bed on the right side. A total of 18 will be randomly selected across the oyster bed on the left side.

b. Fence Creek (Madison, CT):

- i. To access Fence Creek oyster bed, there are two points of access from the parking pull-off (Figure 4). A note for safety, you will need to go over a wood guard fence to get to the paths.
- ii. In the parking area, make sure all equipment is accounted for and everyone has put on the appropriate gear (waders, boots, gloves,

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etc). Carry all equipment and supplies down the path and to the shoreline.

- iii. The oyster bed covers three sections - an old aquaculture lease, under the bridge on Seaview Ave., and out towards the Long Island Sound/ocean. (Figure 5) Collection will only occur beneath the bridge and towards the Sound. Care should be taken to avoid walking on the aquaculture lease area.

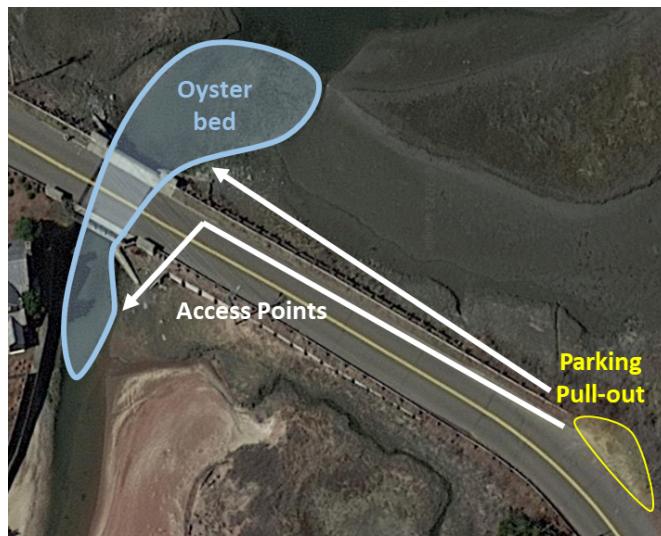


Figure 4: Visualization of the access points and parking at the Fence Creek oyster bed.

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Figure 5: Aerial view of Fence creek oyster bed areas.

- iv. Obtain buckets and shucking knives and wade/walk to the desired area of the oyster reef. Samples will be randomly collected throughout all sections of the oyster bed.
- v. *Notes: The middle of the channel under the bridge is deep. Walk along the walls of the bridge to avoid flooding waders and to safely get to the ocean side. (Figure 7)*

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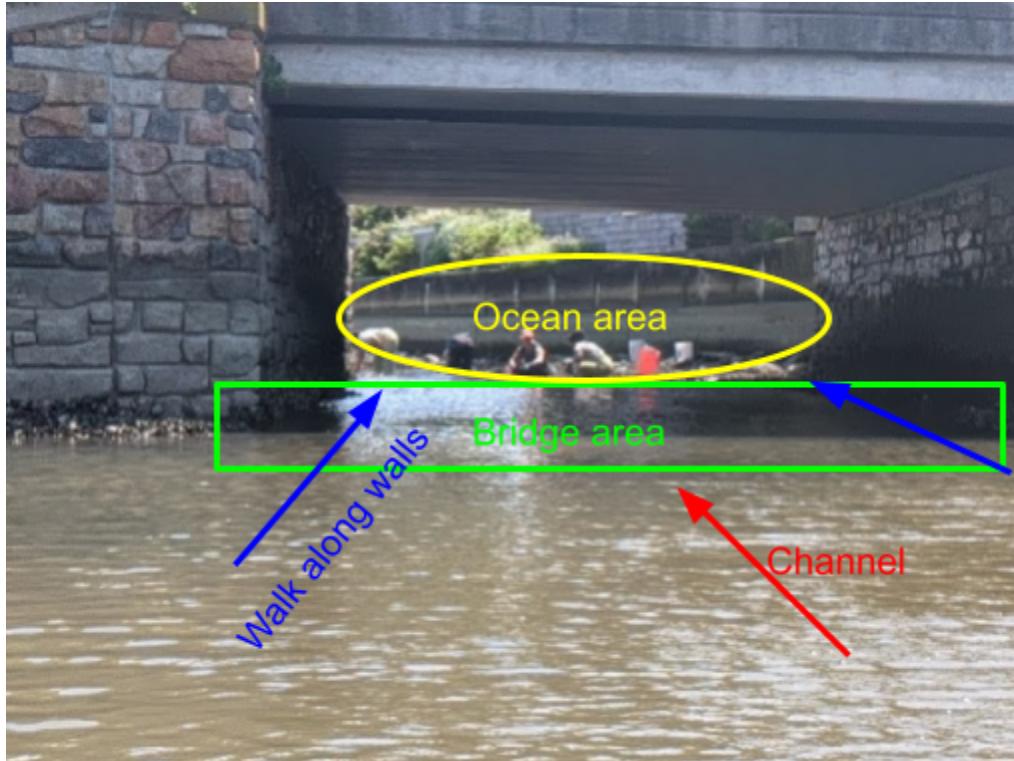


Figure 7: Visualization of where to safely walk under the bridge (blue arrows) and a reference of the designated collection areas at Fence Creek.

- vi. 34 oysters total will be collected from the oyster bed: 17 from the bridge section, 17 from the ocean/sound region.
- c. Once oysters are collected, place them in an onion bag with the site labeled. The bag of oyster samples will then be put into the cooler on ice for transport back to the lab.
- d. Once oysters are collected and all other monthly requirements are fulfilled (see '[Monthly Sonde Maintenance](#)', '[Monthly HOBO maintenance](#)', and '[Water sampling and Chemistry](#)'), collect all gear and return back to the government vehicle to remove waders, and reorganize gear into the vehicle.
- e. Wash hands or use hand sanitizer after oyster collections. Ensure that no one has any cuts or injuries.
 - i. *At Fence Creek, due to high bacterial load, use the Povidone Iodine scrub to cover your hands/arms and allow it to sit for 1 minute on intact skin or a minimum of 5 minutes (maximum of 10 minutes) on*

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any cuts or wounds. If you are prone to skin irritation with povidone iodine scrub, discontinue/avoid use. Be sure to remove any clothing or jewelry that you do not want to become stained. Rinse thoroughly with water.

- ii. Immediately follow the above procedure for povidone iodine application if a cut is acquired while at any site.*

7. Subtidal sites:

- a. For collecting samples, each diver should have a large dive collection bag (metal handles and mouth to open and close the bag with ease).
- b. Assist the divers with getting into their gear and record their PSI (amount of air) before entering the water and record in the NOAA dive log. Collection bags will be hooked to them (Figure 8).



Figure 8: Depiction of oyster collection bags and how to hook them onto the divers.
(Diver in photo is LTJG TJ Houck)

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- c. Divers will collect oysters randomly across the oyster bed areas in the collection bags targeting an amount that will yield 33 oysters.
- d. The land team will watch the divers while under water for safety. Divers will have a flag tailing them to know where they approximately are in the event of an emergency. It is also important to watch for their surfacing bubbles (Figure 9).



Figure 9: Image of the divers underwater with the dive flag surfaced and tailing their movements.

e. Gold Star Beach (Huntington, NY):

- i. To access Gold Star Beach oyster bed, there is a dock landing adjacent to the Cornell Cooperative Extension Building in the parking lot (pass required). If the dock is closed use the access code to open the gate (press 2 & 4 simultaneously, then press 3). The reef is accessible from the dock (estimated coordinates of the reef: 40.897870, -73.435303) (Figure 10).

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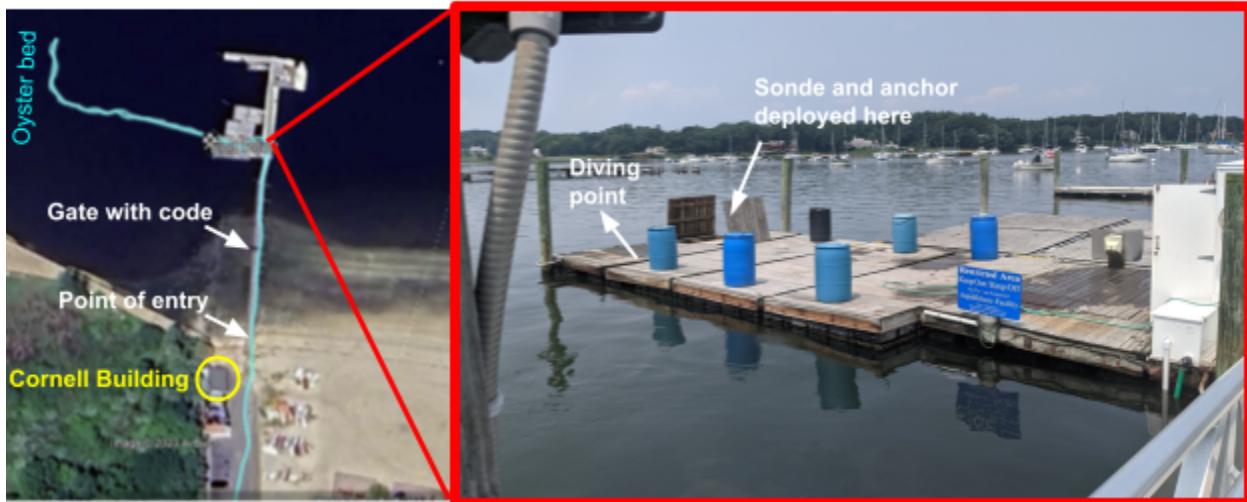


Figure 10: Aerial depiction of access points at Gold Star beach.

- ii. Once parked and access to the gate is granted, there is a wagon/wheel barrel on the dock, next to the white building, that is available to use for the diver gear. Bring that to the government vehicle for divers to fill with any gear they need.
- iii. Divers will enter the water by jumping from the dock.
- iv. Gold Star beach consists of two plantings (Fall 2022 and Fall 2023). These plantings are two separate cohorts and will be sampled as separate reef structures. The plantings were designed as mounds of oysters positioned next to each other (~ 2-3 feet apart). Year 1 and year 2 plantings are parallel to each other. Year 1 reef will be approached first with year two being planted several feet behind it (further swimming distance out from the dock).
- f. Divers will resurface with collection bags filled with oysters. Wearing appropriate protective wear (gloves, etc.) the land team will sort oysters while the dive team finishes other tasks (see [monthly sonde maintenance](#), [monthly hobo maintenance](#), and [water sampling and chemistry](#)), using a shucking knife if necessary to break apart clusters carefully. If the divers did not collect enough oysters they can return back to the oyster bed area to collect the specific amount needed.

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- g. Once 33 oysters have been collected, place them in the appropriately labeled (site specific) onion bag and on ice in the cooler to be transported back to the lab for processing.

10 Lab return and clean up

1. Oysters should be immediately taken to Building 1 Room 29 for cleaning and processing (see [oyster tissue processing](#) SOP).
2. Rinse all tools (wrenches, pliers, wire cutters) with fresh water, dry them with a towel, and spray with WD40 (kept on field gear shelves, extra in Rm 29).
3. Assist divers with cleaning and returning gear if necessary.
4. Hose down waders and any other gear that was submerged in salt water. Hang to dry in the Oyster Health gear area in the garage.
5. Take the Samsung tablet upstairs and connect it to the WiFi in order to sync the shared Google drive (contains the downloaded data files from the sonde and HOBO).
6. Make sure all notes have been updated in the appropriate [field notebooks](#).
7. Used batteries can be disposed of in a box in the 1st floor administrative office.
8. Quality control and assurance (QAQC) data following the [github repository](#).

11 References**12 Appendices**

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