LONG ISLAND SOUND
AMBIENT WATER QUALITY MONITORING PROGRAM

WATER QUALITY AND HYDROGRAPHIC SURVEYS

STANDARD OPERATING PROCEDURES MANUAL

Revision 4
June 2022

STATE OF CONNECTICUT
DEPARTMENT OF ENERGY AND ENVIRONMENTAL PROTECTION
BUREAU OF WATER PROTECTION AND LAND REUSE
PLANNING AND STANDARDS DIVISION
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APPROVAL PAGE

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REVISIONS PAGE

Date	Review Number	Summary of Changes	Applicable Section
4/28/2011	2	Addition of pH calibration procedures	All
4/7/2017	3	Update of DO calibration procedures, TSS procedures	All
6/16/2022	4	Update profiler use and calibration procedures, remove data processing procedures (covered in separate SOP)	All

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List of Acronyms

BIOSI	Biogenic Silica
BOD	Biochemical Oxygen Demand
CESE	Center for Environmental Science and Engineering
Chl a	Chlorophyll a
COC	Chain of Custody
CT DEP	Connecticut Department of Environmental Protection
CTD	Conductivity, Temperature, Depth recorder
DI	De-ionized
DO	Dissolved Oxygen
EMAP	Environmental Monitoring and Assessment Program
EPA	Environmental Protection Agency
HPLC	High-performance Liquid Chromatography
LISS	Long Island Sound Study
LISWQMP	Long Island Sound Water Quality Monitoring Program
MSDS	Material Safety Data Sheet
NCA	National Coastal Assessment
PAR	Photosynthetically Active Radiation
PC/PN	Particulate Carbon /Particulate Nitrogen
PFD	Personal Flotation Device
PP	Particulate Phosphorus
QA/QC	Quality Assurance/Quality Control
SOP(s)	Standard Operating Procedure(s)
UConn	University of Connecticut
UMass	University of Massachusetts

Background

The Connecticut Department of Energy and Environmental Protection (CT DEEP), with support from the Environmental Protection Agency (EPA), initiated the Long Island Sound Ambient Water Quality Monitoring Program (LISWQMP) in January 1991, following a series of comprehensive field surveys conducted during 1988, 1989 and 1990 as part of the initialization, calibration, and verification of the National Estuary Program's Long Island Sound Study (LISS) coupled hydrodynamic-water quality model. The monitoring program, performed by the CT DEEP's Bureau of Water Protection and Land Reuse, continues today. A total of seventeen (17) stations are sampled monthly throughout Long Island Sound (Figure 1). Additionally, summer monitoring (referred to as the hypoxia surveys) to determine the areal and temporal extent of low dissolved oxygen conditions in the Sound is performed bi-weekly from late June through early September along a grid of fixed stations, concentrated in the western and central Sound (Figure 2).

During the monthly water quality survey, water samples are collected for water quality analyses (including nutrients, suspended solids and chlorophyll *a*) and water column profiles of temperature, salinity, dissolved oxygen, pH, and total algae (chlorophyll a and phycoerythrin) are collected. Fluorescent Dissolved Organic Matter (fDOM) will also be monitored beginning in the late summer/fall of 2022. The data collected are considered essential to ongoing data set development, to continued evaluation of model predictions, to help in an ongoing evaluation of monitoring and research needs, and, over the long-term, to monitor the effectiveness of management actions taken in response to findings of the LISS.

The LISWQMP receives requests for assistance with data collections or special projects. Data collection for these special projects is performed following standard operating procedures specified by the principal investigator and are not included in this manual. Special projects also have their own separate Quality Assurance Project Plans.

The LISWQMP also began a project in April 2002 to examine the phytoplankton community structure of Long Island Sound through High Performance Liquid Chromatography (HPLC) phytopigment analysis. Water collected at selected sites during the monthly surveys is vacuum filtered and filters are sent to the University of Maryland’s Horn Point Laboratory for analysis. The SOP for filtering is included with this manual.

Objectives

The objectives of this survey are many. One objective is to develop and initiate a long-term monitoring program that will assist in evaluating the success of management actions in the future. This objective has been met in part through equipment acquisition, staff training, and the successful implementation of a monthly water quality monitoring survey, and CT DEEP plans to continue this monthly survey indefinitely. Cooperation with other research and monitoring efforts on Long Island Sound, providing data, arranging for shared boat time, and adding locations and parameters to the sampling scheme, for example, also help to further the effort of evaluating the Long Island Sound system and identifying research needs.

The second objective is to supplement the data set developed by the 1988 through 1990 surveys of water quality and hydrographic parameters from the East River to Block Island Sound. Although the current data set being collected and compiled is more limited in the number of stations than the earlier surveys, the continuity is valuable. In addition, an intensive hypoxia monitoring survey, performed during the summer months at a large number of stations concentrated in the western Sound provides information on areal and temporal extent of hypoxia each summer.

A third objective is to provide quality assured data to meet Clean Water Act obligations.

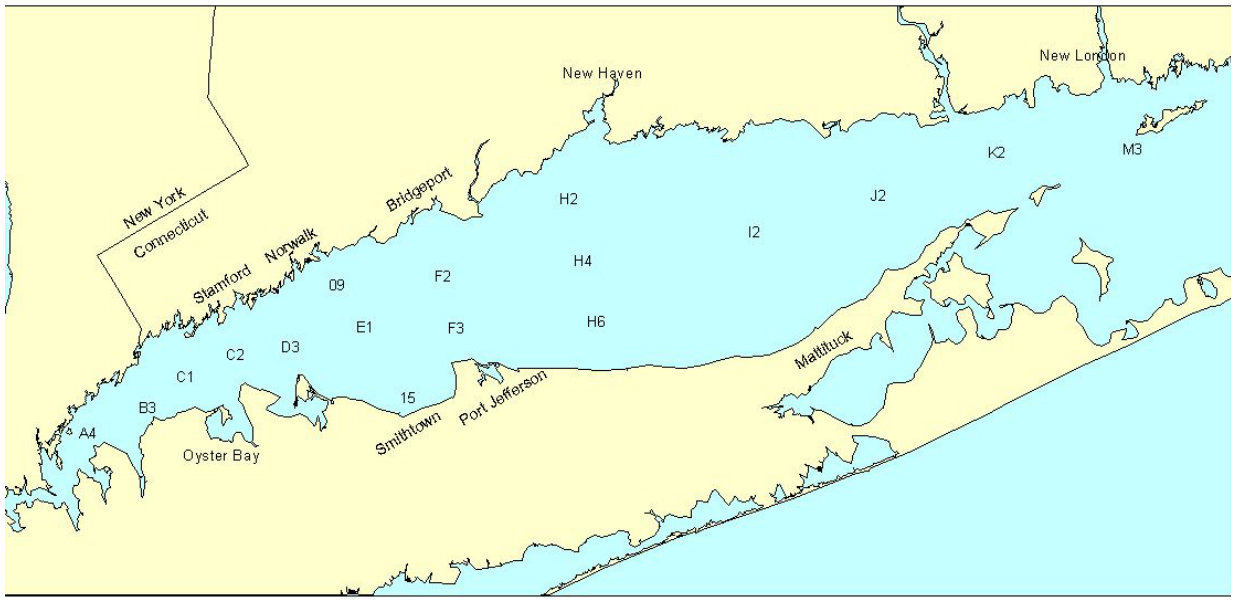
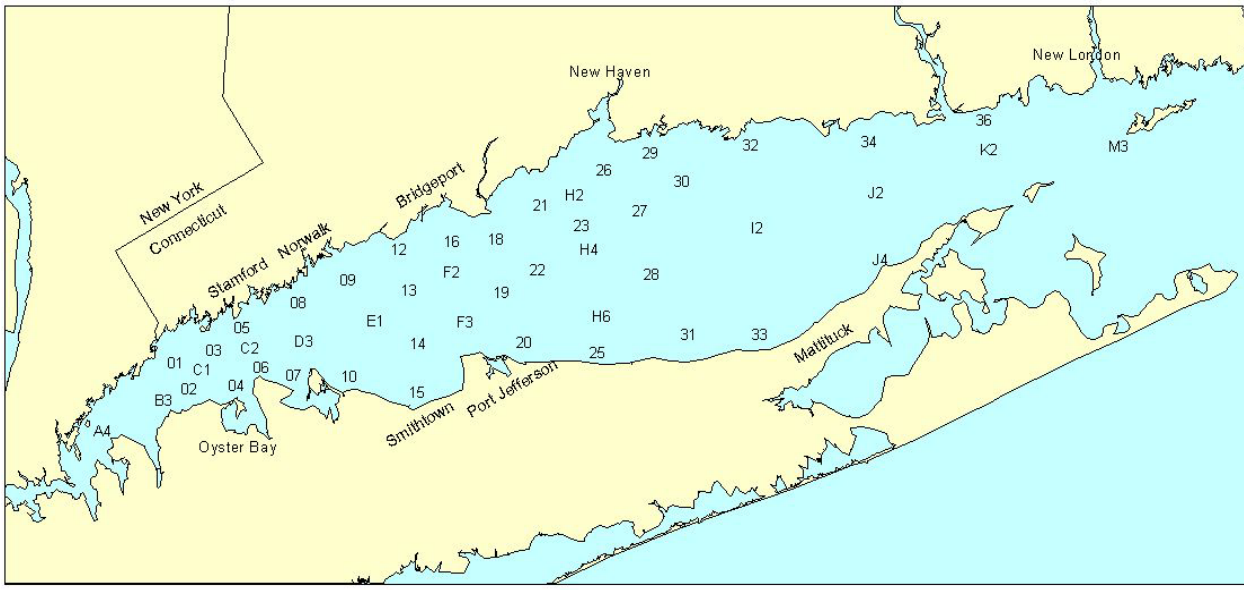


Figure 1. CT DEEP monthly water quality monitoring stations in Long Island Sound

Figure 2. CT DEEP summer hypoxia survey stations in Long Island Sound



General Overview of Methods

Long Island Sound water quality monitoring is conducted aboard the 50 foot CT DEEP Research Vessel John Dempsey (Figure 3). State-of-the-art water sampling and monitoring systems allow continuous water column profiles to be collected. As technology advances and equipment becomes obsolete and to provide more uniform data reporting and make our data consistent across all LIS and freshwater sampling, new water column profiling instruments may be utilized. The current profiling system utilizes a YSI EXO2 series multi-parameter water quality sonde. This unit is equipped with depth, temperature, salinity, dissolved oxygen, pH, chlorophyll and phycoerythrin, and fDOM sensors (fDOM installed late 2022). Data are logged at a rate of 0.5 per second as the unit is lowered through the water column. These data are reviewed in real-time (i.e., as the cast is taking place) via the onboard computer, stored on the sonde's internal memory, and uploaded onto the computer after survey completion.

Figure 3. R/V John Dempsey

Generally the profiling unit is mounted on a rosette water sampling device (General Oceanics model 1015 Rosette Multi-Bottle Array) which also holds up to ten five-liter water sampling bottles (Niskin model 1010 Water Sampling Bottles). These bottles are open as the rosette is deployed and can be closed (i.e., a water sample collected) when the real-time readout from the profiler indicates that the appropriate water sampling depths have been reached. The rosette triggering device is powered through an electromechanical cable on which the unit is lowered. This cable is attached to a deck command unit in the onboard laboratory and together this system allows remote actuation of a sequence of water sampling bottles.

Water samples are collected at a minimum of two depths for full nutrient analyses. The bottom water is sampled at approximately five meters off the bottom and surface water is sampled at a depth of two meters. These depths were chosen by the LISS consultant who developed the water quality model as the most appropriate depths for evaluating nutrient concentrations within the water column. Additional water samples may be collected for dissolved oxygen and chlorophyll determinations at mid-depths (between the surface and bottom nutrient samples), and near-bottom (within a meter of the actual bottom).

Water collected is filtered in the onboard laboratory, and filters and filtrate are delivered to an analytical laboratory for analyses for nutrients, including particulate carbon, nitrogen, phosphorus and silica (biogenic), and dissolved forms including nitrite, nitrate, ammonium, orthophosphate, and silicate, as well as chlorophyll *a*, total suspended solids, and 30-day biological oxygen demand.

Health & Safety Warnings

Sampling from a research vessel in various weather conditions, the use of reagents and acid preservatives, and handling of unknown sample constituents provide occasions for possible hazardous situations to the field monitor. The following should be taken into consideration to ensure the safety of personnel in the field.

General boating safety practices must be observed. The ship's captain and engineer are knowledgeable in safety and emergency procedures and equipment. All crewmembers are expected to follow their instructions AT ALL TIMES. This includes during the performance of regular activities aboard the research vessel, as well as when any emergency situation arises. All crew members should be familiar with where on the vessel safety and emergency equipment is located, such as life jackets, throw-ring, immersion suits, life raft, fire extinguishers, radio, and the first aid kit.

Personal flotation devices (i.e., life jackets, float coats, Mustang work suits) are to be worn when outside of the vessel cabin in rough seas; when deploying gear off the rear or side-deck; when assisting with securing the vessel to, or releasing the vessel from a dock; and when outside forward of the pilot house (bow). Hard hats are required in certain instances when there exists any potential danger from overhead gear, such as deploying the rosette.

There are always hazards when working aboard a vessel, and these hazards are increased whenever there is gear in the water. Be familiar with the operations of the boat, any cables and equipment that are on the deck, the winches and net reel, and any equipment being deployed. Most importantly, be aware of what is going on around you.

Some vessel safety factsheets, CT DEEP directive and vessel rules, and an emergency radio communication guide are included as Attachment A to this manual. The Fishing Vessel Safety Factsheets contain some information that does not necessarily apply to our work aboard the R/V John Dempsey, but is useful for a general understanding of safety equipment, and should prompt questions (most appropriately directed to the ship captain) concerning the specifics of safety gear available on the Dempsey.

The chemicals used on-board for the purpose of preserving samples can be very dangerous. They should be handled with care and should never be left open when unattended. Gloves and safety glasses should be worn when handling (dispensing) these chemicals. Report any spills IMMEDIATELY. Copies of official MSDS sheets with emergency contact information are stored in the senior project scientist’s cubicle and in the Right to Know workstation at the laboratory in Windsor.

Eyewash bottles are available and are kept above the sink in the laboratory area.

If you suffer ANY injury while working on the research vessel, let someone know immediately (captain, supervisor). First aid kits are available on the boat.

When deploying the rosette, keep hands and fingers off the circular base to avoid the possibility of having them caught between the rosette and the boat. Instead, hold the frame along the support bars radiating from the weights.

Personnel Qualifications and Training

At least one permanent staff person of the LIS Water Quality Monitoring Program will be present on the Research Vessel for each survey. Such person will have proven their ability with all aspects of survey preparation and implementation to a senior project scientist. Lower level staff, such as new permanent staff or temporary/ seasonal staff, who participate in field operations will be trained in each field function they will be required to perform (equipment handling, filtering tasks, etc.) prior to participation in a survey. All staff that will participate in field activities on a regular basis will be required to review applicable SOPs and receive safety training annually. Performance of new staff or temporary/seasonal staff will be closely observed. Staff will not be allowed to proceed unsupervised unless and until they have shown proficiency in each particular survey preparation and field activity as determined by the senior project scientist. Staff performance can continue to be evaluated by observations by the senior project scientist or field lead scientist. The project scientist or lead field scientist will correct any errors as they occur and demonstrate proper technique if necessary. If staff continues to make errors, retraining will occur and he/she will be allowed to continue with supervision until they demonstrate consistent proper technique.

Data and Records Management

All field data shall be recorded on the appropriate field data sheet and laboratory chain-of-custody form (Attachment ?). Profile data not available in real-time shall be uploaded to the field computer as soon as possible and reviewed to ensure that a full cast was recorded. Raw profile data are transferred to the Program's network drive following the survey. The field team leader shall be responsible for the accuracy and completeness of all data recorded in the field and the subsequent completion of field data entry into the Program database. All original field data sheets shall be archived, making them available for future reference if necessary. Archives will be maintained according to CT DEEP Records Retention Policies. Field data sheets are also scanned and stored electronically as PDFs in a dedicated directory on the Program's network drive. Drives are backed up nightly.

Raw profile data shall be reviewed by experienced Program staff. Processed profile data will be uploaded into the Program database following procedures outlined in the Data Entry Standard Operating Procedure.

Water samples and filters shall generally be delivered to the analytical laboratory the day they are collected. The analytical laboratory will assign a unique laboratory sample code to each sample, and such code will be recorded directly on to the chain-of-custody form delivered with the samples. The laboratory will keep the original chain-of-custody form and will return a copy to Program staff. Analytical results will be provided in both electronic (via e-mail) and hardcopy forms. Upon receipt, Program staff will review results and associated Quality Assurance/Quality Control (QA/QC) data and upload the results into the Program database.

All data in the Program database (1991 through current) are available upon request, and Program staff frequently fill requests for data. Summary reports of Summer Hypoxia Survey results are produced and distributed to interested parties immediately following each survey. These reports include surface and bottom temperature, salinity and dissolved oxygen (DO) data and a map of minimum DO levels throughout the Sound. Program data are also available for download from the University of Connecticut (UCONN) ERDDAP website. A hypoxic area calculator and

volume estimate web app is available from UCONN as well. Program data will also be uploaded to WQX and available through the Water Quality Portal.

Survey Preparation

Sample Bottles and Labels

The University of Connecticut Center for Environmental Science and Engineering (CESE) in Storrs, CT generally performs the laboratory analyses of water samples for the LISWQMP. Prior to each monthly survey, sample bottles, centrifuge tubes, and pre-weighed foil cups with filters (Table 1) must be obtained from the lab (directions are provided as Attachment B). Bottle labels are pre-printed prior to each survey with the following information.

STA ID- Station Identification (e.g., M3)
S or B= surface or bottom sample
LISS- Long Island Sound Study
Date- MM/DD/YY (current year- to be filled out in the field by project scientists)
Type of container
CENT= centrifuge tube
125 mL NALG= 125 mL wide mouth Nalgene bottle
250 mL NALG= 250 mL wide mouth Nalgene bottle
BOD= 2 L poly bottles for BOD analyses

M3 S	**LISS**
05/30/06	**CENT**

Labels are also pre-printed for foil packets that will contain filters for nutrient analysis.

PC/PN= particulate carbon/particulate nitrogen
CHLA= chlorophyll *a*
HPLC= high performance liquid chromatography
PP= Particulate phopshorus

M3 S	**LISS**
05/30/06	**PC/PN**

Table 1. List of supplies to be obtained from UConn CESE prior to monthly surveys.

Nalgene poly bottles for filtrate storage and delivery (125 ml)
Nalgene poly bottles (250 ml)
Centrifuge tubes for BioSi filter storage and delivery
BioSi filters (47mm polycarbonate membrane filter with a pore size of 0.4um)
PC/PN filters (precombusted 25mm GF/F (glass fiber) filter with a pore size of 0.7um)
Chl a filters (25mm GF/F filter with a pore size of 0.7um)
TSS/PP filters (preweighed, precombusted 47mm GF/F filter with a pore size of 0.7um)
Two-liter poly bottles for BOD samples

The University of Connecticut Marine Science Department at the Avery Point Campus in Groton, CT generally performs the laboratory analyses of plankton samples for the LISWQMP. Prior to each monthly survey, sample bottles must be obtained from the lab (directions are provided as Attachment B). Bottle labels are pre-printed prior to each survey with the following information.

STA ID- Station Identification (e.g., M3)
S or B= surface or bottom sample
(Researchers Name i.e., Dr. Hans Dam, Dr. McManus, Dr.Lin)
Date- MM/DD/YY (current year- to be filled out in the field by project scientists)
Type of Preservative (i.e., 5% Lugol's, 2.5% Formaldehyde, 10% Formalin, Lugol's)

Equipment and Supplies

The Long Island Sound Water Quality Monitoring Program shares the R/V John Dempsey with the CT DEEP's Bureau of Natural Resources, Marine Fisheries Division. The Marine Fisheries Division uses the vessel to conduct surveys of the fish populations of Long Island Sound from April to June and September to October. Therefore, sampling equipment and gear used by the LISWQMP must be loaded on and off the boat prior to and following surveys during these months. Equipment is stored in two locations, the Marine Fisheries Division Headquarters at 33 Ferry Road in Old Lyme and at the CT DEEP's Field Station, 9 Windsor Ave, Windsor. Directions to these locations are provided as Attachment B. To facilitate loading and offloading, the following checklists are provided (Table 2). These should be used to ensure that all needed equipment, reagents, and supplies are accounted for.

Table 2. Survey Preparation Checklist

	Storage Location	Loaded on Boat	Off Loaded	Comments
Field notebook with coins	Windsor			
Field data sheets (17 for monthly surveys, 40 for hypoxia surveys)	Windsor			
Chain-of-custody forms (3 BOD COCs per survey, 3 route specific COCs per survey)	Windsor			
Map/site visit plan	Windsor			
Field writing implements- Permanent marker for labeling, Rite-in-rain pen, pencil, etc.	Windsor			
Profiler	Windsor			
Laptop computer for profiler communication and real-time operation	Windsor or Hartford			
Electromechanical deployment cable and backup shielded electrical cable for real time CTD operation	Windsor			
Deionized water in carboys, (2)	Windsor			
Foil packets labeled with date and station, for PC/PN, PP and Chl-a filter storage and delivery	Windsor			
Nitrile gloves	Windsor			
Safety goggles	Windsor			
Wash bottles	Windsor			
zip-seal plastic bags	Windsor			
Slotted screwdriver	Windsor			
sample bottle loading rod	Windsor			
volt meter	Windsor			
Coolers	Windsor			

Table 2 (continued). Survey Preparation Checklist

	Storage Location	Loaded on Boat	Off Loaded	Comments
Rosette multi-bottle array (General Oceanics model 1015)	Old Lyme			
Niskin model 1010 water sampling bottles (10)	Old Lyme			
25 mm filtering apparatus (Hoeffer filtering manifold with filtrate collection tank)	Old Lyme			
47 mm filtering apparatus for PP and BioSI (home-made filtering manifold with 4 1000-ml filtering flasks and 500 ml overflow flask)	Old Lyme			
47 mm filtering apparatus for TSS (home-made filtering manifold with 2 1000-ml filtering flasks)	Old Lyme			
Filtering funnels, bases, and holders (frit glass and stainless steel)	Old Lyme			
Vacuum filtration pump with hoses (Remains on the boat)	Old Lyme			
Graduated cylinders (250 ml)	Old Lyme			
Filter forceps	Old Lyme			
Clamp for filtration tank outflow tubing	Old Lyme			
Tygon tubing, several lengths, to use as sample bottle outflow hose	Old Lyme			
Bongo Net	Old Lyme			
Small (~8 in diameter) sieve (<64 uM)	Old Lyme			
Large (~12 in diameter) sieve <64	Old Lyme			
Large (~12 in diameter) sieve >64	Old Lyme			
Saltwater wash bottle	Old Lyme			
Funnel, white plastic	Old Lyme			
Plankton Sample Bottles-				
Six 125 mL Nalgene containers	Windsor			
26 250 mL Nalgene containers (amber)	Windsor			
twelve 500 mL wide mouth containers	Windsor			
Twelve 250 mL Nalgene containers (amber)	Windsor			
Life Jackets	Windsor			
Float Coats	Windsor			

Equipment Handling

The primary pieces of equipment used for this monitoring program are the profiler, Niskin water sampling bottles, and a rosette multi-bottle array which allows for the deployment of up to ten sampling bottles and the profiler at the same time (Figure 4). A laptop computer allows the profile data to be viewed in real-time.

Figure 4. Rosette, profiler, and Niskin sampling array

This equipment is state-of-the-art oceanographic equipment and it is costly to repair. **The utmost care should be taken at all times when handling this equipment.** When gear is being deployed crewmembers should be very careful to watch for potential hazards, such as lobster pot lines or the wakes from passing vessels that could cause the instrument to be swept beneath the research vessel or entangled. Should any such hazards present themselves, let the captain or ship engineer know immediately. At any indication of such a problem, the deployed gear should be hauled back immediately. In general, the ship's captain and engineer are very aware of the hazards in the water about them, but if you should see something that you think they are not aware of do not hesitate to bring it to their attention. This equipment is too valuable to take chances.

Whenever the vessel is moving, all equipment must be secured in such a way to avoid tipping and sliding and to avoid any possible damage from other equipment nearby. The same applies whenever the equipment is transported by vehicle.

References

Long Island Sound Study. Undated. [Online] *Research Project Summaries*. Accessed 27 January 2017 from http://longislandsoundstudy.net/research-monitoring/lis-research-grant-program/2004-research-project-descriptions/

Standard Operating Procedures

STANDARD OPERATING PROCEDURE FOR THE CALIBRATION OF THE YSI EXO 2 PROFILER

In 2017, CT DEEP purchased two YSI EXO 2 multiparameter sondes. Following side-by-side testing with the previous profiling equipment (SeaBird SBE 19 Seacat), the Program transitioned to using YSI EXOs exclusively. The LISWQMP follows the manufacturer's instructions for calibration of the depth, dissolved oxygen, conductivity, pH, turbidity, Total Algae, and fDOM sensors as specified in the YSI EXO Manual. The entire manual is available online from YSI: https://www.ysi.com/file%20library/documents/manuals/exo-user-manual-web.pdf
Calibration records are stored electronically on the calibration laptop. Additionally, staff complete a paper copy calibration log which is kept in a binder in the survey prep lab in Windsor.

The sensors are calibrated in the following order:
Dissolved Oxygen
Specific Conductance
pH
Turbidity
Chl a- RFU
Chl a- ug/L
Phycoerythrin – RFU
Phycoerythrin- ug/L
fDOM
The temperature sensor cannot be calibrated but is checked biannually against a NIST certified thermometer.

Equipment/Apparatus/Chemicals

- YSI EXO2 Sonde
- YSI Handheld unit for barometric pressure
- magnet
- Laptop computer equipped with KOR software
- Calibration notebook
- Writing implement
- pH Buffer- 4, 7, 10
- conductivity standard- 50,000 uS/cm
- turbidity standard- 124 NTU
- Rhodamine dye
- 2 1-L volumetric flasks
- 1-L beaker
- Wash bottle
- Pipettes
- DI Water
- quinine sulfate dihydrate
- 0.1 N (0.05 M) sulfuric acid
- Energizer D-cell batteries

DISSOLVED OXYGEN

The optical dissolved oxygen sensor is calibrated in water-saturated air prior to each survey at the CT DEEP laboratory in Windsor, as well as on each day of the survey prior to arrival at the first station. Additionally, the DO sensor cap should be replaced every 12 months.

1. Remove the calibration cup from the YSI EXO 2 and discard the liquid (tap water or pH buffer 4).
2. Reattach the calibration cup to the EXO. Tighten completely, then loosen two turns.
3. Using the magnet, turn on the EXO.
4. Power up the laptop and launch the KOR software by double clicking on the icon.
5. Connect the EXO to the laptop by clicking on the Scan for Bluetooth Devices button. Once a unit is discovered, click on Connect.
6. Allow the unit to equilibrate for approximately 10 minutes.
7. Turn on the EXO Handheld unit. Record the barometric pressure on the calibration worksheet. Turn off the Handheld.
8. Return to KOR. From the Ribbon, select Calibration. Then select the drop down arrow next to DO. The click the Calibrate Button next to %Sat. Click in the box next to Barometer. Type in the barometric pressure reading that you recorded on the calibration worksheet. Watch the screen and wait for the Data Stability row to turn green and read Stable. Click Apply. Click Complete Calibration. Click Exit.

Specific Conductance

1. Click the drop-down arrow next to Conductivity.
2. Click the Calibrate button next to Sp Cond (uS/cm).
3. Remove the Calibration Cup from the sonde.
4. Pour about half of the solution from the RINSE conductivity standard bottle into the cal cup.
5. Reattach the cup to the sonde. Gently swirl around to rinse the probes.
6. Remove the cal cup. Discard the rinse solution down the drain. Flush the drain with tap water.
7. Repeat the rinse.
8. Open the new bottle of conductivity standard. It should be 50,000 uS/cm. Pour the entire bottle into the calibration cup and reattach.
9. In KOR ensure that the standard value is correctly set to 50,000.0 uS/cm.
10. Watch the screen and wait for the Data Stability row to turn green and read Stable. Ensure that the readings are in fact stable and not increasing or decreasing. Click Apply. Click Complete Calibration. Click Exit.
11. Remove the cal cup. Pour the used standard into the RINSE bottle. Recap the bottle.
12. Using tap water, fill the cal cup about 1/8 full. Swirl around to rinse the cal cup. Discard the rinse water. Repeat. Run the EXO sonde under cool running tap water to rinse.
13. Using DI water, fill the cal cup about 1/8 full. Swirl around to rinse the cal cup. Discard. Repeat.

pH

1. Click the drop down arrow next to pH/ORP.
2. Beginning with the pH buffer 7.0 in the RINSE bottle, pour half into the calibration cup. Reattach. Swirl and agitate gently to rinse the probes. Discard. Repeat.
3. Fill the calibration cup with new pH 7.0 buffer. Reattach the calibration cup.
4. Ensure the standard value shown in KOR is 7.00. Wait for the readings to stabilize and the data stability to read stable.
5. Click Apply.
6. Click Add Another Calibration Point.
7. Pour the used 7.0 buffer into the RINSE 7.0 buffer bottle.
8. Tap water rinse the calibration cup.
9. Tap water rinse the probes.
10. DI Rinse the calibration cup.
11. DI Rinse the probes.
12. Pour half of the RINSE pH 10.00 buffer into the calibration cup. Reattach. Agitate and swirl to rinse the probes. Discard.
13. Repeat Step 12.
14. Pour new pH 10.00 buffer into the calibration cup ensuring a sufficient amount to cover the probes, up to the second line on the cal cup.
15. Reattach the calibration cup.
16. Ensure the standard value in KOR is now 10.00. Allow the probe to stabilize/equilibrate. Click Apply. Click Add Another Calibration Point.
17. Pour the used 10.0 buffer into the RINSE 10.0 buffer bottle.
18. Tap water rinse the calibration cup.
19. Tap water rinse the probes.
20. DI Rinse the calibration cup.
21. DI Rinse the probes.
22. Fill the cal cup 1/8 full with new pH 4.00 buffer. Reattach. Agitate and swirl to rinse the probes. Discard.
23. Repeat Step 12.
24. Pour new pH 4.00 buffer into the calibration cup ensuring a sufficient amount to cover the probes, up to the second line on the cal cup.
25. Reattach the calibration cup.
26. Ensure the standard value in KOR is now 4.00. Allow the probe to stabilize/equilibrate. Click Apply. Click Complete Calibration. Click Exit.
27. Pour the pH 4 buffer into the pH 4 RINSE bottle.
28. Tap water rinse the cal cup and the probes.
29. DI rinse the cal cup and the probes.

Turbidity

1. Click the drop down arrow next to turbidity.
2. Click the calibrate button.
3. Fill the cal cup with new DI water. Reattach.
4. Wrap a paper towel around the outside of the cal up to block ambient light.

5. Ensure the standard value in KOR reads 0. Wait for the readings to stabilize.
6. Click Apply. Click Add Another Calibration Point.
7. Pour half of the turbidity solution from the RINSE bottle into the calibration cup. Reattach. Swirl and agitate to rinse the cup and the probes. Discard.
8. Repeat.
9. Carefully and slowly fill the calibration cup with new turbidity standard, being careful to pour down the side of the cal cup and not add air bubbles.
10. Ensure the standard value in KOR reads 124 NTU.
11. Once the readings have stabilized, click Apply. Click Complete Calibration. Click Exit.
12. Pour the used standard into the RINSE bottle.
13. Tap water rinse the cal cup and probes.
14. DI rinse the cal cup and the probes.

Chlorophyll a RFU (Total Algae Sensor)

Chlorophyll a sensor calibration requires the preparation of a stock standard that is then diluted to prepare the calibration standards. Rhodamine dye (Kingscote Chemicals, Miamisburg, OH; item #106023) is diluted with DI water.

1. Prepare the stock 125 mg/L solution of Rhodamine WT.
    a. Fit the automatic pipette with a new, clean plastic tip.
    b. Using the automatic pipette, transfer 5.0 mL of the 2.5% Rhodamine WT dye solution into a 1000 mL volumetric flask. Fill the flask to the volumetric mark with deionized water and mix well to produce a solution that is approximately 125 mg/L of Rhodamine WT. Transfer to a storage container (1000 mL glass flask).
    c. Wrap in foil to block light and store in the refrigerator in the prep lab for up to 1 year.
2. If the stock solution has been in the refrigerator, remove and allow to come to room temperature on the counter before calibration (keep the foil on the flask at all times).
3. Fit the automatic pipette with a new, clean plastic tip. Use the automatic pipette set to 5 mL to transfer 5.0 mL of the 125 mg/L solution prepared in step one into a 1000 mL volumetric flask. Fill the flask to the volumetric mark with deionized water. Put the cap on the flask. Mix well to obtain a solution that is 0.625 mg/L of Rhodamine WT. Use this solution within 24 hours of preparation and discard it after use. Rinse the tip and discard in the trash.
4. The Zero point must always be calibrated first. Fill the cal cup with DI water to the second line. Reattach. Wrap a paper towel around the cal cup to block ambient light.
5. In KOR, select the drop-down arrow next to Chl a RFU. Click Calibrate.
6. Ensure 0 is entered for the standard value.
7. Allow the readings to stabilize and the line in the chart to turn green. This may take some time. Once the readings remain stable for 30 seconds, click Apply.
8. Click Add Another Calibration Point. Record the numbers in the calibration log worksheet.
9. Place a small amount of calibration standard (15 ml) into the EXO cal cup. Reattach the cup. Swirl to rinse the sensor. Discard.
10. Repeat.
11. Fill the cal cup to the second line with calibration standard. Reattach the cal cup. Wrap paper towel around the cal cup.

12. Note the temperature reading in the calibration standard box.
13. Using Table A (copied from the YSI EXO manual) on back of the calibration log worksheet, circle the row on the worksheet that corresponds to the observed temperature reading. Enter the corresponding value for the standard into KOR. At 22ºC the default value is 16.4.
14. Once the readings have stabilized, click Apply. Click Complete Calibration. Record the numbers in the calibration log worksheet. Click Exit.
15. Pour the standard into a large beaker to be used as rinse water for the ug/L calibration.
16. Rinse the cal cup and sonde with tap water.
17. Rinse the cal cup and sonde with DI water.

Chl a (ug/L)

1. The Zero point must always be calibrated first. Fill the cal cup with DI water to the second line. Reattach. Wrap a paper towel around the cal cup to block ambient light.
2. In KOR, select the drop-down arrow next to Chl a ug/L. Click “Calibrate”.
3. Ensure 0 is entered for the standard value.
4. Allow the readings to stabilize and the line in the chart to turn green. This may take some time. Once the readings remain stable for 30 seconds, click “Apply”.
5. Click “Add Another Calibration Point”. Record the numbers in the calibration log worksheet.
6. Discard the DI water.
7. Place a small amount of calibration standard (15 ml) from the RINSE beaker into the EXO cal cup. Reattach the cup. Swirl to rinse the sensor. Discard.
8. Repeat.
9. Fill the cal cup to the second line with fresh, new calibration standard (the same as used for Chl a RFU). Reattach the cal cup.
10. Note the temperature reading in the calibration standard box.
11. Using Table A (copied from the YSI EXO manual) on back of the calibration log worksheet, circle the row on the worksheet that corresponds to the observed temperature reading. Enter the corresponding value for the standard into KOR. At 22ºC the default value is 66.
12. Once the readings have stabilized, click Apply. Click Complete Calibration. Record the numbers in the calibration log worksheet. Click Exit.
13. Pour the standard into a clean beaker for use as a rinse in the ug/L calibration.
14. Tap water rinse cal cup and sonde.
15. DI water rinse the cal cup and sonde.

Pyhcoerythrin (RFU)

1. Use the glass pipette to transfer 0.2 mL of the 125 mg/L stock solution into a 1000 mL volumetric flask. Fill the flask to the volumetric mark with deionized water. Put the cap on the flask. Mix well to obtain a solution that is 0.625 mg/L of Rhodamine WT. Use this solution within 24 hours of preparation and discard it after use. Rinse the pipette with DI water.
2. The Zero point must always be calibrated first. Fill the cal cup with DI water to the second line. Reattach. Wrap a paper towel around the cal cup to block ambient light.
3. In KOR, select the drop-down arrow next to Phycoerythrin (RFU). Click Calibrate.
4. Ensure 0 is entered for the standard value.

5. Allow the readings to stabilize and the line in the chart to turn green. This may take some time. Once the readings remain stable for 30 seconds, click Apply.
6. Click Add Another Calibration Point. Record the numbers in the calibration log worksheet.
7. Place a small amount of calibration standard (15 ml) from the RINSE beaker into the EXO cal cup. Reattach the cup. Swirl to rinse the sensor. Discard.
8. Repeat.
9. Fill the cal cup to the second line with fresh, new calibration standard. Reattach the cal cup.
10. Note the temperature reading in the calibration standard box.
11. Using Table A (copied from the YSI EXO manual) on back of the calibration log worksheet, circle the row on the worksheet that corresponds to the observed temperature reading. Enter the corresponding value for the standard into KOR. At 22ºC the default value is 45.
12. Once the readings have stabilized, click Apply. Click Complete Calibration. Record the numbers in the calibration log worksheet. Click Exit.
13. Tap water rinse the cal cup and sonde.
14. DI rinse the cal cup and the sonde.

Phycoerythrin (ug/L)

1. Fill the cal cup with DI water to the second line. Reattach. Wrap a paper towel around the cal cup to block ambient light.
2. In KOR, select the drop-down arrow next to Phycoerythrin ug/L. Click Calibrate.
3. Ensure 0 is entered for the standard value.
4. Allow the readings to stabilize and the line in the chart to turn green. This may take some time. Once the readings remain stable for 30 seconds, click Apply.
5. Click Add Another Calibration Point. Record the numbers in the calibration log worksheet.
6. Place a small amount of calibration standard (15 ml) from the RINSE beaker into the EXO cal cup. Reattach the cup. Swirl to rinse the sensor. Discard.
7. Repeat.
8. Fill the cal cup to the second line with fresh, new calibration standard that you made in Step 1 of the Phycoerythrin (RFU) calibration. Reattach the cal cup.
9. Note the temperature reading in the calibration standard box.
10. Using Table A (copied from the YSI EXO manual) on back of the calibration log worksheet, circle the row on the worksheet that corresponds to the observed temperature reading. Enter the corresponding value for the standard into KOR. At 22ºC the default value is 126.
11. Once the readings have stabilized, click Apply. Click Complete Calibration. Record the numbers in the calibration log worksheet. Click Exit.
12. Tap water rinse and DI rinse the cal cup and sonde. Discard calibration solutions down the drain; flush with plenty of water.
13. Return the stock solution to the refrigerator and ensure flask is covered with foil to block light.
14. Rinse the glass ware thoroughly with tap water and DI water. Return to the drying rack.

SmartQC for TAL Sensors

The SmartQC Score for any TAL sensor is based on an offset from 0 RFU, and a gain factor. Each individual channel (Chlorophyll, Phycocyanin, Phycoerythrin) has a unique offset and gain factor. It is possible to have a green SmartQC Score for calibration of one channel, but a yellow or red SmartQC Score for the second channel. In this case the TAL sensor SoftQC that is shown in Kor Software will appear as the worst QC Score (yellow or red), and one must look at the individual channels to investigate where the issue is. Thus the steps outlined here are for each channel, and for each unit calibrated within that channel.

Guidance on interpretation of the SmartQC Score for this sensor is as follows:

Green: Gain and offset are within acceptable limits. Calibration was performed successfully and results are within factory specified limits.

Yellow: The sensor gain or offset is near the threshold of calibration limits. If a user calibration results in a yellow QC Score, perform the following actions:

1. Perform a Factory Reset Calibration and complete a recalibration.
    a. If performing a 1-point calibration, use fresh, clear water.
    b. If performing a 2-point calibration, use fresh, clear water and freshly made Rhodamine WT solution.
2. Ensure that the standard value was entered correctly. Calibration of TAL channels is temperature-dependent; make sure the appropriate value from the table in Section 4.24 was entered during calibration for either RFU or µg/L.
3. Ensure that the sensor is free of debris. Refer to Section 5.6 for additional information on how to properly clean the sensor in order to avoid damage.

If the QC Score returns to yellow, the sensor is still able to be used, but the user should monitor this sensor during calibrations for any further drift.

Red: The sensor gain or offset are outside of factory specified limits. If a user calibration results in a red QC Score, follow the same steps described above for a yellow QC Score.

If the QC Score remains red, please contact YSI Technical Support for further assistance.

fDOM

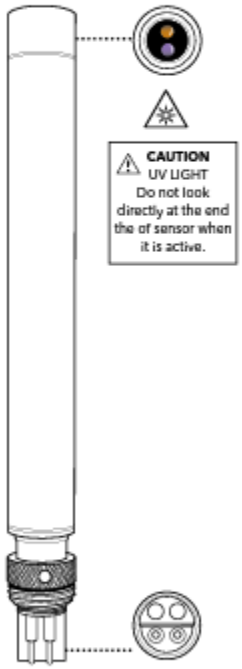
LIS WQMP staff will follow the YSI manual directions for calibration of the fDOM sensor as excerpted below. Personal protective equipment (i.e., safety glasses and nitrile gloves) must be worn when working with the calibration standards.

4.13 fDOM
Sensor Overview

The EXO fDOM (Fluorescent Dissolved Organic Matter) sensor detects the fluorescent component of DOM (Dissolved Organic Matter) when exposed to near-ultraviolet (UV) light.

Colored Dissolved Organic Matter

Users might wish to quantify colored dissolved organic matter (CDOM) in order to determine the amount of light which is absorbed by stained water and thus is not available for photosynthesis. In most cases, fDOM can be used as a surrogate for CDOM.



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Quinine Sulfate

A surrogate for fDOM is quinine sulfate, which, in acid solution, fluoresces similarly to dissolved organic matter. The units of fDOM are quinine sulfate units (QSUs) where 1 QSU = 1 ppb quinine sulfate and thus quinine sulfate is really an indirect surrogate for the desired CDOM parameter.

The EXO fDOM sensor shows virtually perfect linearity (R^2=1.0000) on serial dilution of a colorless solution of quinine sulfate. However, on serial dilution of stained water field samples, the sensor shows some underlinearity. The point of underlinearity in field samples varies and is affected by the UV absorbance of the DOM in the water. Testing shows that underlinearity can occur at fDOM concentrations as low as 50 QSU. This factor means that a field sample with an fDOM reading of 140 QSU will contain significantly more than double the fDOM of a sample that reads 70 QSU. This effect--good linearity in colorless quinine sulfate solution, but underlinearity in stained field samples--is also exhibited by other commercially available fDOM sensors and thus the performance of the EXO sensor is likely to be equivalent or better than the competition while providing the advantages of easy integration into a multiparameter package and automatic mechanical cleaning when used in monitoring studies with an EXO2 sonde.

Specifications

Units	Quinine Sulfate Units (QSU), ppb
Temperature *Operating* *Storage*	 -5 to +50°C -20 to +80°C
Range	0 to 300 ppb QSU
Response	T63<2 sec
Resolution	0.01 ppb QSU
Sensor Type	Optical, fluorescence
Linearity	R^2>0.999 for serial dilution of 300 ppb Quinine Sulfate solution
Detection Limit	0.07 ppb QSU
Optics: Excitation	 365±5 nm
Emission	480±40 nm

100

4.14 fDOM Calibration Standards

Quinine Sulfate Solution for fDOM Sensor

⚠ **WARNING:** Before using a quinine sulfate reagent (solid or solution) or sulfuric acid reagent, read the safety instructions provided by the supplier. Take extra precautions when making dilutions of concentrated sulfuric acid, as this reagent is particularly dangerous. Remember that only trained personnel should handle chemicals.

Preparation

Use the following procedure to prepare a 300 µg/L solution of quinine sulfate (300 QSU) that can be used to calibrate the EXO fDOM sensor for field use:

1. Purchase solid quinine sulfate dihydrate (CAS# 6119-70-6) with a high purity (>99%).
2. Purchase 0.1 N (0.05 M) sulfuric acid (CAS# 7664-93-3), to avoid the hazards of diluting concentrated sulfuric acid to make this reagent.
3. Weigh 0.100 g of solid quinine sulfate dihydrate and quantitatively transfer the solid to a 100-mL volumetric flask. Dissolve the solid in about 50 mL of 0.05 M (0.1 N) sulfuric acid (H_2SO_4), dilute the solution to the mark of the volumetric flask with additional 0.05 M sulfuric acid, and mix well by repeated inversion. This solution is 1000 ppm in quinine sulfate (0.1%).
4. Transfer 0.3 mL of the 1000 ppm solution to a 1000 mL volumetric flask and then fill the flask to the top graduation with 0.05 M sulfuric acid. Mix well to obtain a solution of 300 µg/L (300 QSU or 100 RFU).
5. Store the concentrated standard solution in a darkened glass bottle in a refrigerator to retard decomposition. The dilute standard prepared in the previous step should be used within 5 days of preparation and should be discarded immediately after exposure to EXO's metal components.

Degradation of quinine fluorescence by copper and chloride

NOTICE: Exposure of the quinine sulfate solution to any copper-based component of the EXO sonde and sensors (primarily the wiper assembly) will begin to degrade the solution significantly within minutes. Quinine fluorescence is also degraded by the presence of chloride or halide ions, found in estuarine or seawater, conductivity standards, and Zobell solution. Thus, clean your sensors thoroughly and perform your calibration as quickly as possible on immersion of the sensors into the quinine sulfate solution. Discard the used standard. When quinine sulfate standards are required in the future, perform another dilution of the concentrated solution.

Effect of temperature on fluorescence

The intensity of the fluorescence of many dyes shows an inverse relationship with temperature. This effect must be accounted for when calibrating the EXO fDOM sensor with quinine sulfate solution. Enter the QSU or RFU value from the table below that corresponds to the temperature of the standard.

Temp (°C)	RFU	QSU	Temp (°C)	RFU	QSU
30	96.4	289.2	18	101.8	305.4
28	97.3	291.9	16	102.7	308.1
26	98.2	294.6	14	103.6	310.8
24	99.1	297.3	12	104.6	313.8
22	100	300	10	105.5	316.5
20	100.9	302.7	8	106.4	319.2

4.15 fDOM Calibration

Review the basic calibration description in Section 4.2.
Before calibrating, be certain that the sensing window is clean (cleaning instructions, Section 5.6).

This procedure calibrates fDOM RFU or fDOM QSU/ppb. If the user has both units selected, then this procedure must be performed twice, once for each unit, to completely calibrate the parameter.

For 2-point calibrations, the first standard must be clear water (0 µg/L). The second standard should be a 300 µg/L quinine sulfate solution. For detailed instructions for mixing this solution, see Section 4.14.

> **NOTICE:** Do not leave sensors in quinine sulfate solution for a long time. A chemical reaction occurs with the copper on the sonde (wiper assembly, sonde bulkhead, copper tape) that degrades the solution and causes it to drift. Also, start with very clean sensors, as the presence of chloride and halide ions (from estuarine or seawater, conductivity standards, and Zobell solution) can compromise QS fluorescence.

QSU - 1- or 2-point

Pour the correct amount of clear deionized or distilled water into the calibration cup. Immerse the probe end of the sonde in the water.

In the Calibrate menu, select fDOM, then select QSU/ppb. Select either a 1- or 2-point calibration. Enter 0 for first standard value and 300 µg/L for second standard value.

Observe the Pre Calibration Value readings and the Data Stability, and when they are Stable, click Apply to accept this calibration point.

Remove the central wiper from the EXO2 sonde before proceeding to the next step.

Next place the sensors in the correct amount of 300 µg/L quinine sulfate standard in the calibration cup. Click Add Another Cal Point in the software. Observe the Pre Calibration Value readings and the Data Stability. While stabilizing, verify that no air bubbles reside on the sensing face of the sensor. If there are bubbles, gently shake or move the sensor to dislodge. When data are Stable, click Apply to accept this calibration point.

Click Complete. View the Calibration Summary screen and QC Score. Click Exit to return to the sensor calibration menu.

RFU - 1- or 2-point

Pour the correct amount of clear deionized or distilled water into the calibration cup. Immerse the probe end of the sonde in the water.

In the Calibrate menu, select fDOM, then select RFU. Select either a 1- or 2-point calibration. Enter 0 for first standard value and 100 RFU for second standard value.

Observe the Pre Calibration Value readings and the Data Stability, and when they are Stable, and when they are Stable, click Apply to accept this calibration point.

Remove the central wiper from the EXO2 sonde before proceeding to the next step.

Next place the sensors in the 300 µg/L quinine sulfate standard in the calibration cup. Click Add Another Cal Point in the software. Observe the Pre Calibration Value readings and the Data Stability. While stabilizing, verify that no air bubbles reside on the sensing face of the sensor. If there are bubbles, gently shake or move the sensor to dislodge. When data are Stable (or data shows no significant change for approximately 40 seconds), click Apply to accept this calibration point.

Click Complete. View the Calibration Summary screen and QC Score. Click Exit to return to the sensor calibration menu. Rinse the sonde in tap or purified water and dry the sonde. Discard the used standard.

SmartQC for fDOM Sensors (RFU or QSU)

The SmartQC Score for fDOM is based on a gain factor and an offset factor. Both of these values may change as the sensor and the optics age.

Guidance on interpretation of the SmartQC Score for this sensor is as follows:

Green: Gain and offset are within acceptable limits. Calibration was performed successfully and results are within factory specified limits.

Yellow: The sensor gain or offset is near the threshold of calibration limits. If a user calibration results in a yellow QC Score, perform the following actions:

1. Perform a Factory Reset Calibration and complete a recalibration.
    a. If performing a 1-point calibration, use fresh, clear water.
    b. If performing a 2-point calibration, use fresh, clear water and freshly made quinine sulfate solution.
2. Ensure that the standard value was entered correctly. Calibration of fDOM is temperature-dependent; make sure the appropriate value from the table in Section 4.14 was entered during calibration for either RFU or QSU.
3. Ensure that the sensor is free of contamination. Refer to Section 5.6 for additional information on how to properly clean the sensor in order to avoid damage.
4. Ensure the copper tape and the central wiper brush are removed from the sonde. Copper quenches fluorescence of quanine sulfate, which will interfere with the calibration.

If the QC Score returns to yellow, the sensor is still able to be used, but the user should monitor this sensor during calibrations for any further drift.

Red: The sensor gain or offset are outside of factory specified limits. If a user calibration results in a red QC Score, follow the same steps described above for a yellow QC Score.

If the QC Score remains red, please contact YSI Technical Support for further assistance.

STANDARD OPERATING PROCEDURE FOR FIELD SAMPLING USING A PROFILER/ROSETTE SAMPLER ABOARD THE R/V JOHN DEMPSEY

Summary

A YSI EXO2 profiler is used to obtain *in situ* water quality data from Long Island Sound. The profiling unit is mounted to a Rosette multi-bottle array Water Sampling System. The Rosette accommodates up to 10 General Oceanics Niskin bottles for the collection of water samples using "grab" techniques. The bottles are actuated remotely from a deck command unit.

Equipment/Apparatus

- Electromechanical cable(s)
- profiling unit (YSI EXO2)
- Niskin Bottles
- Rosette Sampler
- ½ inch socket driver
- ½ inch double end Hex Box wrench
- Phillips head screw driver
- Regular head screw driver (~1/8 inch wide)/loading rod
- Quarter
- Laptop computer equipped with KOR software

Procedure

A. Attach the electromechanical cable to the rosette and net reel

Communication between the laptop, rosette, deck command unit, and the YSI profiler occurs through an electomechanical cable that is wound onto the net reel. A Signal Output Adapter (SOA) Modbus Communication Adapter is installed inside the cabin on the starboard side wall of the lab and the net reel cable is connected. The net reel cable must also be spliced to a pigtail that connects to the YSI and rosette. Information on installation and connection of the SOA is available in the YSI manual and not covered in this SOP. Details on splicing the pigtail are also not covered in this field manual/SOP. The LISWQMP shares the R/V John Dempsey with the LIS Trawl Survey. During the months of April, May, June, September, and October, when

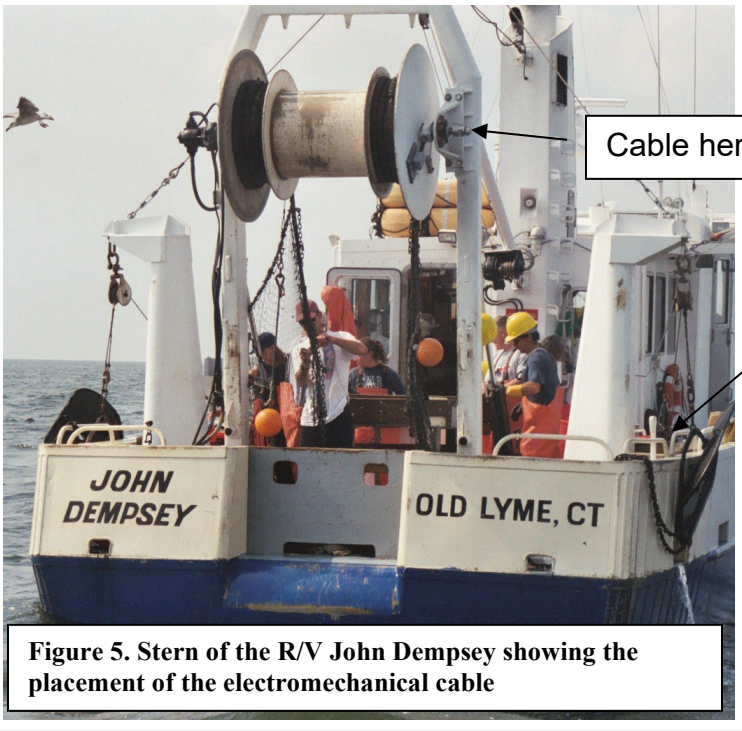


Figure 5. Stern of the R/V John Dempsey showing the placement of the electromechanical cable

the trawl survey operates, LIS WQMP equipment and gear must be loaded on and off the boat prior to and following each survey, respectively. To ensure communication between the rosette, deck command unit, and the laptop, and before disembarking from the dock, the electromechanical cable must be connected to both the net reel and the laptop computer. Follow the procedure below to connect the cable. This is not done every survey; only during months when changeover occurs.

Enter the onboard laboratory. Remove the foam plug from the hole to the left of the desk on the starboard side. Feed the cable through the hole out onto the deck. Uncoil the cable and run it along the starboard side of the deck towards the net reel. Carefully, climb up the support to attach the cable to the slip-ring mounted on the net reel. You can brace yourself on the rosette table and one foot on the net reel support. Remove the protective tape wrap. Spray connections with CRC 6-66 silicon if needed. Connect the cable to the slip-ring. Be sure to also connect the safety line. Return inside. Replace the foam plug from the inside to prevent water from entering the cabin. Plug the USB adapter into the SOA and then into the laptop.

B. Setting the Niskin Bottles in the Array

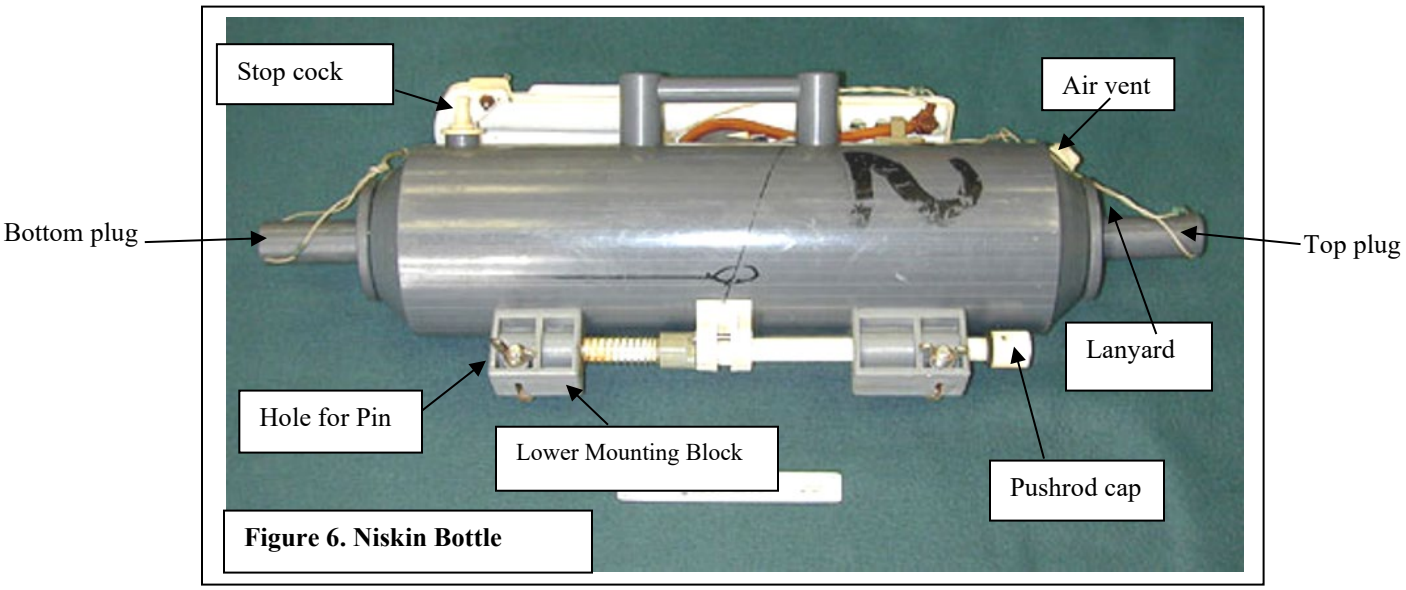


Figure 6. Niskin Bottle

1. Insert the Niskin Bottle in to the array (Figures 6 and 7).
   Grasp the bottle by the handle.
   Hold the bottle in the upright position with the air vent at the top and the stop cock on the bottom. Gently tilt the bottle so that the air vent is pointing towards you.
   Line up the hole on the bottom of the lower mounting block with the pin on the array.
   Slid the pushrod down. Tilt the bottle back to the vertical position. Release the pushrod so that the round white cap fits into the hole on top of the array.

Continue placing bottles on the rosette working counterclockwise. Avoid empty positions between bottles.

Figure 7. Niskin bottles in the sampling array.

2.Using a slotted screwdriver or a coin, turn the ramp/shaft at top center of the rosette until the beveled edge points toward the location one position to the left (counterclockwise) of the first bottle to be actuated (see Figure 12). If the first bottle is in position #1, then the beveled edge should point towards #12; if the first bottle is in position #2, the beveled edge should point towards #1, etc. The shaft will turn clockwise or counterclockwise, **but remember: a counterclockwise turn towards an untriggered bottle will trigger that bottle (it will scare you and can hurt you if you are not expecting it!);** and the shaft will not turn clockwise if a bottle at the clockwise position is set.

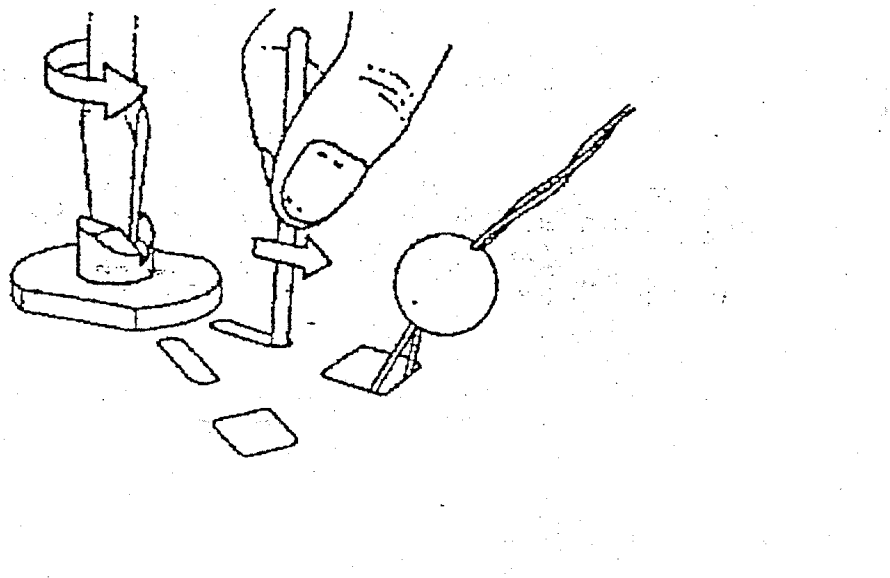


Figure 8. Close-up of top of rosette multibottle array showing ramp/shaft alignment (note beveled edge) and technique for attaching bottle lanyard by pushing release pin into forward locking position with loading rod. Note that beveled edge of the shaft end should be aligned with a position where there is NO bottle (position prior to first bottle) and should be aligned before the lanyards are secured in place.

3. Unclip the lanyard from the bottom plug.

4. Pull open the top plug.

5. Holding the lanyard against the closing force, insert the lanyard loop (at the end of the short extension) into the lanyard opening at the top of the rosette.

6. Using the loading rod (small flat head screwdriver with flat side pointed away from you), gently push the release pin down and forward toward the loop (see Figure 12). A faint click should be heard when the release pin has locked, and the little white ball will move forward and lock the lanyard loop in place. If you miss the loop and the ball locks, gently push the pin up and back. Repeat for all bottles on the rosette working counterclockwise.

7. Pull the bottom plug open and snap the clip on the long extension of the top lanyard through the loop attached to the bottom plug. Do this connection on the right side of the bottle. Be

sure that the white ball in the center of the long lanyard extension is not caught behind or between neighboring bottles. It is important that this ball be on the outside of the bottles so that the bottles are free to close. It is also necessary that the bottles are all set in the same direction, in other words, that the lanyards connecting the top and bottom all be on the same side of the bottles - to the right when you are looking at the bottles. This ensures that no bottle interferes with the operation of another. Repeat for all bottles on the rosette working counterclockwise.

8. Close all air vents by turning clockwise until tightened. These are the screw closures on the top of each bottle. It is imperative that these air vents be closed because no air should be introduced into the water before it is drawn from the bottle for dissolved oxygen determination.

9. Close all stopcock assemblies. These are at the bottom of each bottle and are used to draw water from the bottle. Pull out the outer ring (away from the bottle) until a click is heard (or felt), and then rotated slightly so that the small hole in it no longer lines up with the pin beneath.

10. Check that the lead weights attached to the bottom of the rosette frame are secured. These are held on with hose clamps, with some tie wraps providing additional support. A daily check that these are secure will avoid the loss of a weight.

C. Field calibrate the YSI DO sensor.

1. Power on the laptop and log in.
2. Launch the KOR software.
3. Remove the YSI from the hard travel case.
4. Unscrew the calibration cup. If the storage solution is tap water, this can be poured down the drain. If the YSI is stored in pH 4 buffer, pour the buffer into the waste buffer container. DI rinse the cal cup. Rinsate may be discarded down the sink.
5. Replace the calibration cup. Loosely tighten the threads, but do not tighten the cal cup on completely.
6. Allow the YSI to equilibrate with the water saturated air for approximately 10-20 minutes as directed in the YSI manual.
7. Using the magnet, turn the YSI on and enable Bluetooth.
8. Doubleclick on the Scan for Bluetooth devices tab in the KOR software.
9. Click on the calibration tab.
10. Click on the drop down arrow for DO.
11. Click the button next to % sat.
12. Turn on the YSI Handheld. Obtain the barometric pressure.
13. Enter the barometric pressure into the KOR DO calibration software.
14. Allow the readings to stabilize.
15. Click Accept.
16. Click Complete Calibration.
17. Click Exit.
18. Next calibrate the Depth sensor.
19. Close out of the calibration menu.
20. Disconnect the YSI from the laptop.
21. Turn off the Bluetooth with the magnet.
22. Approximately 5 minutes prior to arrival at the first station, attach the YSI to the rosette, using the same method as you did to attach the Niskins.
23. Unscrew the thumbscrew on the carabineer. Pass it through the carrying handle of the YSI. Retighten the thumbscrew so it is hand-tight.
24. Attach the pigtail from the electromechanical cable to the YSI. Line up the pins and gently push the connector down onto the YSI. Tighten the white protective cap hand-tight. (This will connect the YSI to the KOR software.)
25. The YSI sensors need to remain moist. Place a zip top bag around the YSI. Carefully close the bag as far as you can.
26. The YSI should not be left out on deck in winter months when the temperature is below freezing. If the air temperature is less than 32ºF, bring the YSI back into the cab upon departing the station.

E. Launching the YSI to record profile data

The LISWQMP senior scientist has previously configured and saved all station templates to the laptop(s) that are utilized by the Program. Templates are set to log data in 0.5 second intervals using the Rapid System-wide Averaging Mode. Adaptive logging is disabled.

1. Approximately three (3) minutes prior to arrival on station, ensure that the YSI is connected to KOR. Click the Deployment Tab.
2. Click the Open Template button.
3. Click the drop down arrow under Site Name. In the box, type in the name of the station at

which you will be collecting profile data. Hit the Enter button on the keyboard.
4. Click the Save and Apply Template to Sonde Button.
5. Answer Yes to the popup box that says "Would you like to begin internal logging to the sonde?"
6. Select the NOW radio button. Click Start to begin logging data.

F. Rosette Deployment

1) Don hardhat and life jacket. Proceed to the rosette table at the stern of the boat.

2) Remove the plastic zip top bag that is keeping the YSI sensors moist. Place in the bungie cord around the hydraulic control station.

3) There should always be two crewmembers handling the rosette at deployment time. When assisting with this deployment, be careful to keep track of where the arms of the net reel are. Hold onto the rosette by the frame along the radiating support bars not where the weights are. DO NOT grab onto a sampling bottle or to the profiler to maneuver the rosette. Help to guide the rosette up off of its stand, keeping it from banging into the arms of the net reel. **If the rosette swings be careful to keep hands on radiating support bars of the rosette and not on the outer rim of the circular base where there is the danger of hands and fingers getting caught between the rosette and the boat. The profiler unit should always be facing out/away from the transom to avoid damage to the profiler in the event the rosette should swing into the boat.**

5) Double check that the profiler is submerged with the tops of the Niskins completely covered. Let the hydraulics operator know if the rosette needs to go deeper. If the water is choppy the rosette will need to be set slightly deeper.

6) The rosette must be allowed to soak beneath the surface of the water for at least three minutes before the downcast is started.

This serves a number of purposes: it allows the profiler to come to equilibrium with the surrounding water - this is especially important for temperature. If the unit has been on the deck for a couple of hours and been warmed by the sun, it needs time to cool down to the temperature of the water. The equilibration period gives the plumbing system a chance to fill with water, pushing out any air, and gives the pump a chance to turn on - there is a 45-second delay between the time the CTD enters the water and the time the pump turns on (this allows the plumbing to fill with water, the pump will not pump water effectively if any air is trapped in the plumbing, the pump can be damaged by prolonged operation in air). Finally, the three-minute equilibration period allows the dissolved oxygen sensor time to polarize, which is essential for adequate response time and performance.

While the rosette is soaking record station observations on the field data sheet- % cloud cover, current weather, sea state, latitude/longitude from the boat's Global Positioning System, the station depth from the boat's depth finder, time on station, etc. See Attachment E for an example field sheet and explanations for cloud cover, sea state, etc.

G. Profile Cast and Water Sample Collection

1) Following the equilibration period, notify the hydraulics operator to lower the rosette to the bottom. When the rosette reaches the bottom, tell the hydraulics operator that the bottom had been reached and record the depth on the field sheet under the Profiler depth field.

   The downcast is performed without stopping the rosette until it reaches the bottom. The descent rate should be approximately 0.2 meters per second. The hydraulics operator must be told if the descent rate is too fast. The boat's depth-finder provides an approximate depth at the station, and the real-time readout on the computer screen provides the actual depth of the rosette. The real-time readout will indicate when the bottom has been reached - by depth readings that do not change - there is a delay in this readout.

   During regular monthly water quality surveys the rosette should immediately be raised 5 meter off the bottom.

   During hypoxia surveys when a near-bottom sample is to be collected instruct the hydraulics operator to raise the rosette to 1 meter off the bottom.

   Record the profile data on the field sheet.

   If a separate real-time data cable is used, this must be hauled in by hand as the upcast is underway. Wind this cable neatly back into the grey tote as the rosette is raised through the water column. This will ensure that the cable will unwind freely for the next deployment.

2) Collect bottom water samples by pushing the red sample (trigger) button on the rosette deck command unit; a yellow light comes on, there is a delay before the trigger actually closes the bottle - a click can be heard and the dial advances to the bottle location number that was just actuated; the green light then comes on. [During this operation, a stepping motor in the rosette rotates the shaft and ramp one position at a time. At each step the ramp frees a release pin which in turn releases the nylon lanyard and trips the corresponding sampling bottle.] Repeat the procedure to collect successive samples. The number of bottles collected at each station varies with the type of survey being performed (e.g., monthly or hypoxia) and if samples are collected for plankton community analyses. Refer to Attachment G as a guide to the number of bottles collected at each station and depth. This is a guide only. If guest researchers are aboard, and we are collecting water for them, this number will change.

Figure 9. Deck Command Unit

   To ensure samples are actually being collected, connect the volt meter to the deck command unit. When the readings on the volt meter are 52± 2 the bottles have been triggered.

3) Mid-depth samples are taken on the upcast at stations where plankton samples are collected. The rosette will be stopped at appropriate depths with the depth of these samples approximately evenly spaced between surface and bottom samples. The depth of sample, from the real-time computer readout, must be recorded on the appropriate data sheet. The up-cast rate of retrieval should also be 0.2 meters/second.

4) Lastly, the rosette is brought up to a depth of 2 meters (surface sample), and the procedure for recording profile data on the field sheet and filling bottles is repeated.

H. Retrieving the Rosette

1) The same care taken when the rosette was deployed should be taken upon retrieval. BE READY TO KEEP THE ROSETTE FROM SWINGING AND BANGING AGAINST THE STERN OF THE BOAT. GRAB THE CROSSBARS OF THE BASE WITH TWO HANDS AND HELP GUIDE THE ROSETTE ONTO ITS STAND. TAKE CARE TO KEEP HANDS/FINGERS OFF THE OUTER EDGE/RING OF THE ROSETTE BASE TO AVOID THE POSSIBILITY OF HAVING THEM CAUGHT OR CRUSHED BETWEEN THE ROSETTE AND THE BOAT.

1) Place the plastic bag back over the YSI to ensure the sensors remain moist.

2) Stop the Deployment. Go to the laptop. Click the Deployment Tab. Click Stop Deployment.

I. Water sample handling

1). One crew person will remove the Niskin bottle containing the bottom sample and one will remove the bottle containing the surface water sample. These bottles are brought into the laboratory in the wheelhouse. The bottom sample is placed in the right slot in the rack and the surface sample is placed in the left slot.

Bottles for special projects such as the Altabet study may be filled on deck.

Water for biochemical oxygen demand analysis is also drawn on the deck (See BOD SOP below).

At select stations (see phytoplankton section) water is composited from the Niskin bottles into a large carboy for plankton analysis. This may require a second cast with the rosette (See Plankton SOP below).

J. Reset Niskin Bottles for the next station.

K. Repeat above steps F-H for remaining stations.

L. End-of-Day Equipment Care

Bring the Profiler into the wheelhouse. Wash with fresh tap water. Fill the calibration cup to the fill line with TAP water (NOT DI Water). Attach the calibration cup. Return the YSI to the storage rack in the wheelhouse if sampling the following day or to the hard, plastic traveling case at the end of the survey. Be sure the Bluetooth is off and the unit has been disconnected from the KOR software.

Remove all Niskin bottles from the rosette, empty of water if needed, and store in plastic crate on board.

Rinse the plankton sieves with fresh water. Store in the laboratory overnight.

Store all coolers and sample collection bottles that are not being shipped to CESE in the lab.

STANDARD OPERATING PROCEDURE FOR THE COLLECTION OF SECCHI DISK DEPTH MEASUREMENTS

Summary

One of the major diagnostic tools in the analysis of eutrophication is the measurement of water transparency. Algal blooms decrease light penetration by light absorption, and scattering water transparency and light penetration are proportional to the density of the algal bloom.
A simple method of estimating light penetration in the vertical direction is with a Secchi disk, where the disappearance depth is defined as the Secchi depth. The Secchi disk is submerged into the water from the shady side of an anchored boat or from the end of a pier. The Secchi disk is lowered to a point where it is no longer visible and then raised to a level where it again becomes visible. The Secchi Depth at this point is measured (meters) and recorded.

Safety Warnings

Take care when leaning over the gunwales of a boat not to fall in. Be sure to wear a lifejacket.

Equipment/Apparatus

- Weighted 8 inch (20cm) Secchi disk with alternating black and white quadrants
- Calibrated, Non-stretch Line (tenths of meters)

Procedure

1. Position yourself on the **shady side** of the boat.
2. Remove sunglasses and hats.
3. Slowly lower the Secchi disk into the water until it is no longer visible. Note the depth at the waterline.
4. Slowly raise the Secchi until it becomes just visible again. Note this depth.
5. Average the two depths and record on the Field Data Sheet.

Interferences

There are many types of interferences or sources of variation for Secchi disk readings. These include but are not limited to surface glare, ripples and waves, angle of the sun, cloudy vs. clear sky, variations in eyesight of the observers, shadows of the boat, weeds, and resuspension of sediments from the bottom. Additional variations are caused by variations in size of disks used (20 cm is used here), variations in color (white is commonly used also, but a black and white disk is used here) and use of a viewscope to reduce glare.

Quality Assurance/Quality Control

Quality control involves participation in a yearly field training session for new staff. During field training, new staff must replicate readings taken by experienced staff following procedure above, and must agree within 20 percent.

Annual checks of the calibrated line against a meter stick are performed to ensure the line has not stretched and to remark the calibrations in the event of fading.

STANDARD OPERATING PROCEDURE FOR THE COLLECTION OF WATER FOR BIOCHEMICAL OXYGEN DEMAND

Summary

The Biochemical Oxygen Demand (BOD) is an empirical test in which standardized laboratory procedure is used to determine the relative oxygen requirements of wastewaters, effluents, and polluted waters. The test measures the oxygen required for the biochemical degradation of organic material (carbonaceous demand) and the oxygen used to oxidize inorganic materials, such as sulfides and ferrous iron.

The method consists of placing a sample in a full, airtight bottle and incubating the bottle under specified conditions for a specific time. Dissolved Oxygen (DO) is measured initially and after incubation. The BOD is computed from the difference between the initial and final DO. For specific analytical procedures, refer to the CESE SOP.

References

1979 US EPA Manual entitled, "Methods for Chemical Analysis of Water and Wastes," EPA 600/4-79-020, Revised 3/83, Method 405.1, (p. 405.1).

"Standard Methods for the Examination of Water and Wastewater," 18th Edition 1992, Method 5210 B, (p. 5 - 2-6).

Equipment/Apparatus

- Niskin Bottle filled with sample water
- Tygon tubing
- Pre-labeled 2 L plastic sample containers

Procedure

1. Sample rinse the collection bottle twice before collecting the sample.

   Attach small tygon tube to the stopcock of the Niskin. Push in the stopcock, open the air vent and allow water to flow out of tube. Pinch tube gently at the stopcock to remove air bubbles. Pinch the tube to stop the flow. Remove the cap from the BOD sample bottle, fill with a small volume of the appropriate sample, remove tube (pinch to stop flow), recap the bottle and mix to rinse. Pour out. Repeat.

2. Collect the sample.

   Remove the cap again. Allow the water to flow out of the tube. Insert the tygon tubing all the way to the bottom of the sample container and allow the bottle to fill until overflowing. Remove the tube with the water still flowing. Pull out the stopcock or wrap tygon tubing up and around lanyard to stop water flow. Squeeze the bottle slightly and recap so that a minimum amount of air is trapped in the bottle.

3. Samples must be kept cool (4ºC) and in the dark. Place the sample bottles in the appropriate cooler.

Sample Delivery

Deliver to the laboratory within the 24 hour hold time, usually immediately after the day's sampling. Be sure to include the completed chain of custody form. See Attachment F for an example COC.

If samples cannot be delivered to CESE and the analysis started within 36 hours of collection time (e.g., collected on a Friday), consult with senior project scientist and CESE Chief Chemist or Lab Supervisor for direction on whether or not to even collect BOD samples.

Quality Control

A duplicate surface water sample is collected at Station B3.

STANDARD OPERATING PROCEDURE FOR FILTERING WATER SAMPLES FOR PARTICULATE PHOSPHOURS (PP), DISSOLVED NUTRIENTS, BIOGENIC SILICA (BIOSI), AND DISSOLVED SILICA ANALYSIS

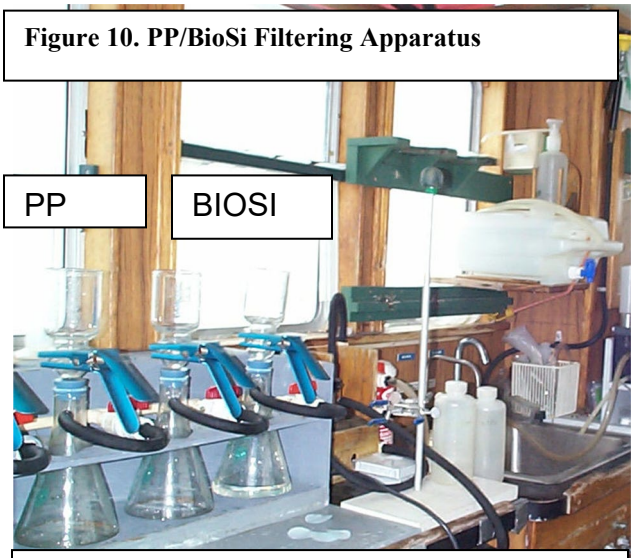
Summary

Water samples are filtered to separate dissolved nutrient components from particulate nutrient components and suspended solids. The filtrate from this apparatus is used for dissolved nutrient analyses-the filtrate from the PP filters is used for dissolved nitrogen, phosphorus, and carbon; the filtrate from the BioSi filters is used for dissolved silica (silicate) analysis.

Equipment/Apparatus

- Oil-free Vacuum pump
- Filtering manifold
- 4-1000 ml filtering flasks
- 2 glass fitted filter supports
- 2 glass funnels
- 4 metal clamps
- 2 plastic filter supports
- 2 plastic funnels
- 4-250 mL plastic graduated cylinders with holes drilled slightly above the 250 mL mark.
- 2-250mL plastic graduated cylinders with holes drilled at the 202 mL mark
- 47 mm GF/F filters (0.7 µM pore-size) Whatman 1825-47
- 47mm polycarbonate membrane filter (0.4*u*m pore size)
- 2 Stainless steel forceps
- Nalgene wash bottle
- 125 mL Nalgene sample bottles
- 250 mL Nalgene sample bottles
- Centrifuge tubes
- Foil packets
- Sample labels
- Deionized Water

Figure 10. PP/BioSi Filtering Apparatus



Note: the filter set up on the right is now composed of a magnetic plastic filter support and funnel

Note: The oil free vacuum pump is stored on the R/V John Dempsey and is located under the sink. The pump is turned on and off via a switch located on the underside of the counter to the left of the sink.

The two flasks on the right of the apparatus (See Figure 15) are designated for Biogenic silica (and associated dissolved nutrients). The two on the left are designated for PP (and associated dissolved nutrients).

Procedure

1) Rinse all graduated cylinders, filtering funnels, and filter holders (frit glass or stainless steel) 2 times with DI water before setting up new filter pads. Connect the vacuum tubing. Place the filter supports on the filtering flasks.

2) Set up the PP filters.
    a) The PP filters come from the lab in a cardboard box but are stored in a plastic box. Rinse the dedicated PP forceps with DI water. Shake off excess water. Using the forceps, remove the filter from the box. The filters have two sides, a smooth side and a rough side. The smooth side will have a waffle pattern, the rough side, will not. Place the filter onto the fritted glass support of the filtering apparatus rough side up. Be sure the filter is centered so that the sample cannot get around the filter.
    b) Repeat for the second filter.
    c) Place the glass filter funnel on top of the filter and secure with the clamp. Be careful not to move the filter. Repeat for the second filter.

3) Set up the BIOSI filters
    1) The BIOSI filters are stored in a plastic box with circles of paper separating each filter. Rinse the dedicated BioSi forceps with DI water. Shake off excess water. Use the forefinger of one hand to gently push back the paper and expose the filter. Using the forceps in the other hand, pick up the filter by the edge.
    2) The filter is placed on the support shiny side down. Generally, the filters come packaged shiny side up. Double check that this is the case. Maneuver the filter so the shiny side is down.
    3) Bring the filter to the edge of the support. Carefully slide the filter up the edge and onto the support. This is somewhat tricky. It is highly likely that you will end up with folds/creases/wrinkles in the filter. The support has two gaps that allow you to place the forceps under the filter and reposition it. You can also carefully smooth the forceps over the filter to push the fold to the edges. Repeat for the second filter.
    4) Once the filter is centered on the support and free from folds, turn the vacuum pump on for ~10 seconds. This will firmly seat the filter and draw out any tiny wrinkles.
    5) Place the magnetic funnel over the filter. Attach the clamp.

4) Mix the surface water sample in the Niskin bottle on the left side of the rack in the lab. Remove the Niskin bottle from the rack and invert (shake) end to end several times. The water sample in the bottle must be kept well mixed as suspended material in the water settles to the bottom of the bottle making some samples very concentrated and leaving other samples very dilute.

    If not previously done, attach small tygon tube to the stopcock of the Niskin. Push in the stopcock, open the air vent and allow water to flow out of tube. Pinch tube gently at the stopcock to remove air bubbles. Pinch the tube to stop the flow. Wrap the tube through the lanyard and secure against the rack until ready to dispense the sample.

5) Sample rinse the 4-250 mL-graduated cylinders with the holes drilled just above the 250 mL mark. To do this, fill one of the cylinders with ~10 mL of sample. Swirl the water around the cylinder to rinse, shake, and then dump into the sink. Repeat. Begin

to fill the cylinder to the 250 mL mark. Continue until all four have been rinsed and filled. If you overfill, pour out the hole. Place the cylinders in the holding rack above the filtering apparatus.

6) Sample rinse and fill one 250 mL cylinder to the 200mL mark as in Step 5 above. Place the filled cylinder on the shelf of the filtering apparatus behind the left BioSi flask.

7) Mix the Niskin bottle containing bottom water. Rinse one 250 mL cylinder and then fill to the 200 mL mark as above. Place this cylinder on the shelf behind the right BioSi flask or on the counter to the right of the apparatus.

This filtering process is time consuming and you will need to multitask to complete the filtering between stations. It is best to filter for PP and BIOSI at the same time.

8) Begin the process of filtering for PP (500mL)
   a) Turn the valve on to the left flask for PP analysis.
   b) This flask needs to be sample rinsed 2 times before collecting filtrate for dissolved nutrient analysis.
   c) Pour ~20 mL of sample into the funnel and allow to pass through the filter. Turn off the vacuum. Disconnect the tubing. Carefully, remove the filter support and funnel, being sure to keep the two together and not to disturb the filter. Place into the wooden hole at the back of the apparatus. Swirl filtrate around the flask and then discard either down the sink or out the back door. Return the flask to the apparatus. Reconnect tubing and the filter support and funnel. Repeat.
   d) Pour remaining 210 mL through the funnel. Turn off the vacuum once all the sample has passed through the filter.
   e) Now move to the PP flask on the right. Turn on the vacuum pump to the flask on the right in the PP station.
   f) Pour ~200 mL of sample carefully into the glass funnel again being careful to pour down the side of the funnel rather than directly onto the filter.
   g) Swirl remaining ~50 mL to re-suspend settled materials and pour into funnel.
   h) Return to the PP flask on the left. Pour the second 250 mL carefully into the funnel. Allow to drain.
   i) Return to the left flask. Pour the second 250 mL into the funnel. Allow to drain. Turn off the vacuum once all the sample has passed through the filter.
9) Next proceed to filter for BIOSI (200 mL)
   a) Turn on the vacuum pump to the BioSi filters.
   b) **Surface and bottom samples will be collected at the same time for BIOSI analysis**. The filter flasks are labeled Surface on the left and Bottom on the right. The flasks will collect the filtrate for dissolved silica analysis and must be sample rinsed a minimum of two times.
   c) Pour ~10 mL of sample from the surface cylinder into the funnel on the left and allow to pass through the filter. Turn off vacuum pump. Disconnect vacuum hose. Remove the filter support and funnel as one piece and carefully set into wooden hole on the back of the apparatus. Swirl the 10mL around the flask then discard. Reconnect the vacuum tubing and the filter support and funnel. Repeat.
   d) Slowly pour remaining sample into the plastic funnel. Use caution to pour

down the side of the funnel and not directly onto the filter.

e) Swirl the last 30 mL of sample to re-suspend settled materials and pour into the funnel. Allow the entire sample to filter through.
f) Repeat for the bottom water sample.

10) Sample rinse the dissolved nutrient sample bottle.
Select the correct 250mL Nalgene sample bottle (surface) to sample rinse before filling with sample. Disconnect the vacuum tubing from the filter flask on the left. Remove the entire filter assembly as you did for sample rinsing the flask and place it into the hole. Remove the cap from the sample bottle but do not set it down on anything (i.e., keep it in your hand while you hold the bottle). Pour a small amount ~10 mL from the flask into the sample bottle. Replace cap. Swirl and shake to rinse. Discard rinse water. Repeat.

11) Collect the filtrate for the dissolved nutrient analyses using the bottle you sample rinsed in Step 10. Fill the sample bottle with sample water to the shoulder. As these samples are frozen, it is important to leave headspace (~1/4 inch) to allow for expansion. Recap the bottle. Replace the filter assembly on the flask and connect the vacuum tubing. Place the sample bottle in the freezer.

12) **VERY IMPORTANT: THE FILTERS MUST BE RINSED WITH DI WATER before removing them from the filter holder/funnel assembly. This step is for comparison purposes with the TSS filters.** This rinsing removes the salt from the filter, which would otherwise invalidate the analysis for TSS.
    a) Using the wash bottle, squirt DI around the sides of the graduated cylinders that were used to measure the samples for the filter to the 10 mL mark. Carefully, pour both of these together into the funnel, being sure not to pour directly onto the filter. Turn on the vacuum and draw through. Repeat.
    b) Using the wash bottle, squirt DI water around the inside of the funnel, about three turns (5-10 ml). Do not squirt or otherwise force water directly onto the filter - this can damage the filter or force some of the particulate material on it through.
    c) Repeat steps a and b for the second filter.
    d) Allow the vacuum to remove excess water from the filter. Turn off the vacuum. Disconnect the tubing.
    e) Disconnect the clamp holding the funnel onto the support.
    f) Lift the funnel off the support and place in the hole on the shelf behind the apparatus. Make sure that the filter stays on the filter support.
    g) Rinse the designated PP forceps with DI water, shake to remove excess. Carefully open the correctly labeled foil pack by sliding the forceps into the foil and opening the forceps. Then, carefully slide one side of the forceps under the filter, grab the filter by the edge only, where there is no sample, and gently fold over. Use the flat side of the forceps to flatten the filter out at the fold. Remove filter and place in the foil packet.
    h) Repeat for the second flask. Place the filter into the same foil packet.
    i) Fold the top of the foil packet over three times to secure the filters inside. Place the packet into the freezer.

10) Return to the BIOSI filters

By now the sample should have filtered and the filter should be dry.

a) Turn off the vacuum and remove the tubing from the filtrate flasks.
b) Remove the magnetic funnels and place on the shelf behind the apparatus.
c) Open the correctly labeled surface centrifuge tube. Hold the cap in your hand with the tube.
d) Rinse the designated BioSi forceps with DI Water. Using the forceps, gently lift a corner of the filter. Grasp the filter on the edge and fold it in half.
e) Place the filter into the centrifuge tube. Try to get the filter to the bottom of the tube. Recap the tube.
f) Repeat for second filter.
g) Place the tubes in the freezer.
h) Sample rinse the 125mL Nalgene bottle labeled for the surface sample twice as for the filtrate from the PP apparatus.
i) Fill the sample bottle to the neck, leaving headspace. Place this sample bottle in the freezer.
j) Repeat for the bottom water sample.

11) Once the filtrate has been collected empty the flasks into the sink or out the door.

12) Rinse all supports, funnels, and flask that hold filtrate with DI water.

13) Set up the apparatus again.

14) Repeat the procedure for the PP filters for the bottom sample.

15) Once finished with the bottom sample, set up the apparatus for the next station.

16) At the end of the trip, all "glassware" is rinsed in DI water. The flasks are turned upside down in the holders to dry. The supports and funnels are placed in the holes in the shelves to dry.

17) Record volumes filtered on the filtering information data sheet. The results of the chemical analyses are on a per volume basis, so an accurate record of the volume filtered is necessary.

Changes to the standard volume filtered can (and should) be made if suspended material concentrations are high and it is taking 5 minutes or more to filter a sample. In such a case, the pores of the filter clog, changing the relative pore size of the filter and thus changing the size of the material that is being caught by the filter. In addition, if suspended material concentrations are very low and little or no color is visible on the filter, the volume filtered should be increased. Always record the volume filtered on the data sheet, and be sure that all replicate filters have identical amounts of sample filtered through them.

Quality Control/Quality Assurance

Duplicates

For quality assurance purposes, surface duplicate samples are filtered at stations M3, A4, and E1 following the same procedures as outlined above. Duplicate surface samples are collected before bottom samples. On the BioSi apparatus, the surface and surface duplicates are run at the same time, followed by the blank (See below) and bottom samples.

Field Filter Blanks

Filter blanks are prepared and included with the other samples for analysis. These Blanks provide a way to measure any background contamination on the filters caused by field handling procedures. The Blank filters should be treated in the very same manner as a sample filter, except that no sample is filtered. Blanks are generally prepared following filtering at stations M3, A4, and E1 but can be run at any time during the day after the first station has been filtered.

a) Set up filters and assemblies as you would to prepare for sample filtering.

b) For PP filter blank only: Rinse graduated cylinders 2 times with DI water and rinse the funnel as above.

c) For BioSi: turn on vacuum pump briefly to draw any DI water off that the filter picked up from rinse water on the holder. DO NOT run any DI water through these filters.

d) Handle filters only by the edges and only with forceps.

e) Place filters in appropriate foil packet or centrifuge tube and freeze.

Check to be sure that the sample code on the foil pack or container corresponds to the sample that was prepared.

Sample Delivery

Foil packets, centrifuge tubes, and 250 mL Nalgene sample bottles (dissolved nutrients) are placed into the freezer after all filtering is completed until the vessel arrives at the dock. Dissolved silica samples (125 mL Nalgene bottles) are also stored in the freezer until arrival at the dock. At the dock, foil packets, centrifuge tubes, and TSS tins are placed together into a large Ziploc bag and then into a cooler along with the dissolved nutrients and BOD samples to be transported to the analytical lab (CESE at UConn in Storrs, CT). Chain of custody (COC) sheets (Attachment F) are filled out and accompany the samples to the lab. Samples are relinquished to a freezer/refrigerator designated by the laboratory. The 125mL dissolved silica samples are placed into the refrigerator at CESE. All other samples are placed into the freezer. Copies of the completed COCs are returned (faxed) to LISWQMP staff with the data package.

STANDARD OPERATING PROCEDURE FOR FILTERING WATER SAMPLES FOR PARTICULATE CARBON (PC), PARTICULATE NITROGEN (PN), CHLOROPHYLL A (CHL A), AND HIGH PERFORMANCE LIQUID CHROMATOGRAPHY (HPLC) ANALYSIS

Summary
Surface and bottom water samples collected from Long Island Sound are filtered through a Hoefer filtering manifold. The filters are analyzed for particulate carbon/particulate nitrogen, chlorophyll a content and undergo high performance liquid chromatography analysis, which separates and quantifies pigments to determine phytoplankton composition. This SOP describes the process for filtration.

Figure 11. Chl a, PC/PN, and HPLC Filtration Apparatus

Equipment/Apparatus

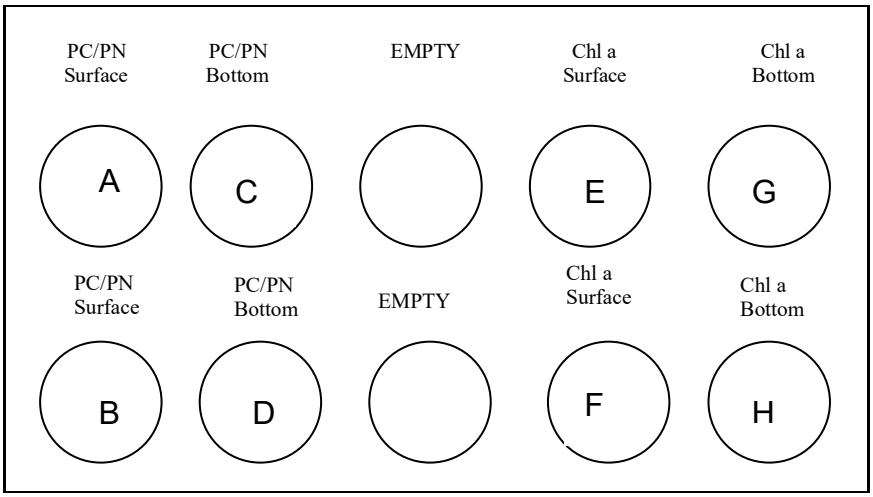
- 25mm filtering apparatus (Hoefer filtering manifold with filtrate collection tank, associated vacuum tubing)
- TSS sampling apparatus
- 250 mL graduated cylinder with hole drilled at 200 mL
- precombusted 25mm GF/F (glass fiber) filters (0.7*u*m pore size) for PC/PN analysis
- 25mm GF/F (glass fiber) filter (0.7*u*m pore size) for chl a and HPLC analysis
- foil packets
- 2 pairs of forceps
- *oil free vacuum pump

* Vacuum pump is the same as that used for TSS/PP/BIOSI filtering. The pump remains on the vessel under the sink. The power switch is located under the counter to the left of the sink.

Procedure

SET UP

PC/PN filtration will take place on the left side of the apparatus and chl a will take place on the right. The apparatus is labeled as such. Additionally, surface and bottom samples are filtered at the same time. From left to right the apparatus should be set up to filter surface sample PC/PN, bottom sample PC/PN, surface sample chl a, and bottom sample chl a. See Figure 17. PC/PN and chl a filters are kept in separate labeled plastic boxes under the filtrate collection tank.



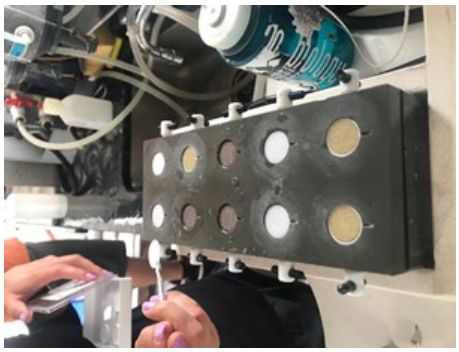


Figure 12. Diagram of filter set up for PC/PN, Chl a analysis. Surface water is filtered for HPLC analysis. Filters are set up following PC/PN and chl a filtering in the spaces marked above as chl a surface.

1) Rinse the filter base with Deionized (DI) water. Gently shake off excess. Place the base onto the square filtrate collection box. There are slots next to the filter pad holes that are utilized for removing the filters after the sample has been collected. Typically, the slots are set up on the right side (for right-handed analysts). The base can be flipped 180º for left-handed analysts.

2) Rinse the flat tipped forceps with DI Water.

3) Using the rinsed forceps, select a PC/PN filter from the box. The filters have two sides, a smooth side and a rough side. The smooth side will have a waffle pattern, the rough side, will not and may appear stringy. Place the PC/PN filter on the filter holder rough side up beginning with the space designated PC/PN surface A in Figure 17 above. This allows larger particles to be caught first by the filter, while smaller particles get through the first couple of layers and are caught later. The filter pad will not clog as fast if set-up properly and so the pore size is not as likely to be affected. Continue to place PC/PN filters on the apparatus beginning with space B, then moving to C and D. Rinse the forceps with DI water.

4) Repeat the process for the chlorophyll a filters.

5) Carefully grasp the funnel by the metal funnel holder. DI rinse the funnel: begin by squirting DI water around the inside of plastic funnel then move to the outside of the metal base.

6) Firmly seat the metal funnel holder onto the filter. Be aware that there is a metal ring on the bottom of the holder that can cut the filter. Repeat with the remaining 7 funnels. Depending upon your comfort level, you may rinse two funnels at once. Do not set the newly DI rinsed funnel onto the counter.

7) Turn the vacuum on (top red-valve) at the manifold coming from the pump. Next turn the

vacuum on to each filter to confirm that the funnel and filter are properly positioned. If a hissing is heard, adjust the funnel by turning it gently. If the hissing persists, lift off the funnel and check to make sure the filter is not damaged, replace any damaged filters. Be sure the valves to the center two slots without filters are off. Turn off the vacuum at the red valve until ready to filter. Only turn the valves on the filter base 90 degrees in either direction to turn off and on. They will back completely out of the unit if turned too much.

FILTRATION

1) Begin filtering with the bottom sample. Carefully remove the bottom water Niskin bottle from the right side of the wall rack. Push down on the plunger. Gently tilt the bottle forward. Then lift up. Holding the bottle by the handle with one hand, place the other on the backside to support the bottle. Pinch the tygon tubing between the handle and your hand to prevent water from spraying everywhere. Invert the bottle a few times to fully mix the sample. Return the bottle to the rack.

2) If not previously done, attach small tygon tube to the stopcock of the Niskin. Push in the stopcock, open the air vent and allow water to flow out of tube. Pinch tube gently at the stopcock to remove air bubbles. Pinch the tube to stop the flow. Wrap the tube through the lanyard and secure against the rack until ready to dispense the sample.

3) If it isn't already on, turn on the pump. Open the red valve to the filters.

4) Sample rinse the 200 mL graduated cylinder twice by swirling about 15 mL of sample around the inside of the cylinder, shake and discard. Fill to 200 mL mark with sample. The bottom of the meniscus should be at the 200 mL mark. Re-wrap the tygon tubing around the lanyard of the Niskin bottle.

5) Begin by pouring the sample into the rear G funnel (Figure 17) for bottom water chlorophyll a. Brace yourself by putting one foot under the cabinet. Extend thumb and index finger to act as brace. Place cylinder against fingers while pouring. DO NOT brace against "funnels". If seas are extremely rough, rinse milk jug cap with DI and place inside the top of the cylinder to prevent over pouring.

6) Refill graduated cylinder with bottom water. Pour into funnel H.

7) Continue on to the bottom PC/PN filters (C &D), working back to front. Re-wrap the tygon tubing once completed.

8) Remove the surface water Niskin from the left side of the wall rack. Mix surface water sample. Rinse the graduated cylinder two times with sample water. Fill to the 200 mL mark. Pour into the funnels labeled surface samples, proceeding from E to F (back to front), then A and finally B.

9) Allow the sample to completely filter through. Visually check through the square filtrate collection box and through the top of the funnel base. Carefully lift off the filter bases by tilting forward towards you and lifting up. Proceed from left to right. Place on the paper towel on the counter, next to the Hoeffer unit. Turn off the vacuum. Open the valves to the unoccupied filters to release the pressure.

10) Rinse both sets of forceps with DI water. Tap together gently to remove excess water. Carefully open the foil packet labeled surface PC/PN by sliding the closed flat forceps into the packet and then allowing them to open gently. Be sure that the label on the foil packet corresponds to the sample that was filtered. Do not set the forceps down on the counter.

11) Beginning with the surface PC/PN filter "A", you will work from back to front, left to right to place the filters into pre-labeled foil packs.

    If you are right handed, grasp the pointed forceps in your right hand and the flat forceps in your left. Using both hands and sets of forceps, proceed to fold the filter in half. Slide the pointed forceps down the slot in the manifold. Gently lift the filter up off the base about a quarter of an inch. Gently grasp the outer white ring where there is no sample with the flat forceps. Using the pointed forceps, fold the filter. Using the flat forceps, place the filter in the foil packet in the bottom of one corner. Gently close the foil pack around the filter. Repeat for second replicate filter (e.g. "B"), placing it in the opposite corner of the foil pack. It's important that they NOT TOUCH one another inside the foil pack or they will freeze together.

12) Fold foil over twice to close and ensure a complete seal. Rinse the forceps in between with DI water. Proceed to the next set of filters (e.g., bottom PC/PN). Repeat twice move for surface chl a and bottom chl a. Once all filters have been removed, place foil packets in designated zip top bag in the freezer.

13) Once the procedure is complete for the PC/PN and chl a filters, remove the filter support. Rinse with DI water. Rinse the holder/funnel with DI water. If HPLC analysis is to be performed at the stations, set up the apparatus by:

    Placing two of the chl a filters on the apparatus in the surface chl a spots. Put the filter holders/funnels over the filters. Turn on the vacuum. Utilizing surface water, sample rinse the graduated cylinder and fill to the 200mL mark. Pour 200 mL of surface sample into each of the funnels. Allow vacuum to remove excess water. Remove holder/funnel. Using the forceps place the filters into the foil packet as above.

IMPORTANT NOTES-

Rinse all holders, funnels, and the filter support with DI water in between stations. Sample rinse the graduated cylinder between surface and bottom samples and between stations.

Watch the water level in the filtrate collection carboy. It must be emptied periodically (e.g., every other station) so that water is not sucked into the vacuum line. The filtrate from this filtering apparatus is not used for any chemical analyses. However, this filtered water is used in the plankton processing. Check with the senior scientist to see if the wash bottles need to be filled before dumping. Excess may be poured down the sink in the lab or over the gunwales back into LIS.

Quality Control/Quality Assurance

Blanks and Duplicates

For quality assurance purposes, surface duplicate and blank samples are collected at stations M3, A4, and E1.

1) Set up the apparatus following the same general procedures as outlined above.
2) Blank and bottom samples are collected first at these stations followed by all surface samples at once (i.e., surface PC/PN, surface DUP PC/PN, surface chl a, surface DUP chl a).
3) Mix the Bottom Sample Niskin. Sample rinse the graduated cylinder with bottom water.
4) Fill to the 200 mL mark with bottom water. Pour into bottom chlorophyll G. Repeat for funnels H, C and D.
5) Do not pour anything through filters A, B, E and F. These will be your blank filters.
6) Remove the funnels and place the filters into the correct pre-labeled foil packs as above.
7) Reset the apparatus.
8) Mix the Surface Water Sample Niskin. Sample rinse the graduated cylinder. Fill to the 200 mL mark with surface water. Pour into funnel G.
9) Continue pouring and filtering surface water through the remaining 7 funnels.
10) Remove the funnels. Following the procedures above, remove filters A&B and place into the PC/PN Surface foil pack.
11) DI rinse the forceps.
12) Place filters C&D into the PC/N surface duplicate foil pack.
13) Continue on to the chlorophyll samples, placing filters into the correct foil packs, DI rinsing the forceps as you move right to left, in between surface and surface duplicate.
14) HPLC duplicates are collected only at Station A4. Following PC/PN and chlorophyll blank & bottom and surface & surface duplicate sample processing, set up the apparatus yet again with four (4) chlorophyll filters in the E, F, G, and H slots.
15) Mix the surface water Niskin. Sample rinse the graduated cylinder. Begin filtering 200 mL of surface water, as above, through funnel G. Repeat until you have run water through all 4 funnels. Remove funnels, place filters into the correct foil packs: filters E & F will be surface while G & H will be duplicates.
16) DI Rinse all funnels and the base. Set up for the next station. At the end of the day, rinse all equipment with tap water, then DI water, and allow to air dry.

Sample Delivery

Chl a and PC/PN samples are placed into a Ziploc bag and delivered to CESE (UConn, Storrs) on ice along with the nutrient samples usually at the end of each sampling day.

HPLC samples are stored in the boat's freezer until the end of the survey then brought back to the Department's Windsor Ave laboratory and stored in the -80ºC freezer. The HPLC samples are then sent overnight delivery in batches on dry ice to Horn Point at the University of Maryland via a commercial carrier such as UPS. Samples are sent Monday-Wednesday to ensure staff at Horn Point are available to receive the samples.

Completed Chain of Custody forms (Attachment F) accompany the samples to the labs.

STANDARD OPERATING PROCEDURE FOR FILTERING WATER SAMPLES FOR TOTAL SUSPENDED SOLIDS (TSS) ANALYSIS

Summary

Water samples are filtered to separate dissolved nutrient components from suspended solids. Two filters from each sample are provided to the laboratory.

Equipment/Apparatus

- Oil-free Vacuum pump
- Filtering manifold
- 2 plastic filter supports
- 2 magnetic plastic funnels
- 1-250 mL plastic graduated cylinder with hole drilled slightly above the 250 mL mark.
- Pre-weighed 47 mm GF/F filters (0.7 µM pore-size) Whatman 1825-47 in foil tins (with bar codes from lab)
- Stainless steel forceps
- Nalgene wash bottle
- Deionized Water

Note: The oil free vacuum pump is stored on the R/V John Dempsey and is located under the sink. The pump is turned on and off via a switch located on the underside of the counter to the left of the sink.

Figure 13. TSS Filtering Apparatus

Procedure

1) Rinse all graduated cylinders, filtering flasks, filtering funnels, and filter holders 2 times with DI water before setting up new filter pads prior to arrival at the first station. Connect the vacuum tubing. Place the filter supports on the PVC filtering bases.

2) Set up the TSS filters.

a) The TSS filters come in pre-weighed foil tins. It is important that the filters are returned to their respective tins. There are four stacks of tins per box received from the lab. Select one stack from the box (it doesn't matter which one).

b) Rinse the flat forceps with DI water. Shake off excess water. Using the forceps, remove the filter from the first foil tin. Note that the filter has a waffle pattern on one side and a more random, stringy, rough pattern on the other. Place it rough side up on the support of the filtering apparatus on the left (labeled surface). Be sure the filter is centered so that the sample cannot get around the filter.

c) Remove the filter from the second tin and place it on the filter support on the right (labelled bottom), rough side up.

d) DI Rinse one plastic magnetic filter funnel and place it on top of the filter. Be careful not to move the filter. Repeat with the second magnetic funnel.

e) Turn the two empty foil tins 180 degrees from the others (so you know which have been used).

3) Mix the bottom water sample in the Niskin bottle on the right side (bottom water) of the rack in the lab. Remove the Niskin bottle from the rack and invert (shake) end to end several times. The water sample in the bottle must be kept well mixed as suspended material in the water settles to the bottom of the bottle making some samples very concentrated and leaving other samples very dilute.

4) Sample rinse the 250 mL-graduated cylinder with the hole drilled just above the 250 mL mark. To do this, fill one of the cylinders with ~10 mL of sample. Swirl the water around the cylinder to rinse and then dump into the sink. Repeat. Fill the cylinder to the 250 mL mark. Replace the tygon fill tube on the Niskin bottle.

5) Turn on the vacuum to the right (bottom) filter base in the TSS station.

6) Pour ~200 mL of sample into the funnel being careful to pour down the side of the funnel rather than directly onto the filter.

7) Swirl remaining ~50 mL to re-suspend settled materials and pour into funnel.

8) Re-fill the cylinder with another 250 mL bottom water. Pour the sample into the funnel as in steps 6 & 7.

VERY IMPORTANT: THE TSS FILTERS MUST BE RINSED WITH DI WATER BEFORE REMOVING THEM FROM THE FILTER HOLDER/FUNNEL ASSEMBLY. This rinsing removes the salt from the filter, which would otherwise invalidate the analysis for TSS.

9) Using the wash bottle, wash down the sides of the graduated cylinder that was used to measure the samples for the filter, using approximately 15 mL. Pour into the funnel. Turn on the vacuum and draw through. Repeat.

10) Using the wash bottle, squirt DI water around the inside of the funnel, about three turns (5-10 ml). Do not squirt or otherwise force water directly onto the filter - this can damage the filter or force some of the particulate material on it through.

11) Allow the vacuum to remove excess water from the filter.

12) Turn off the vacuum pump to this filter.

13) Mix the surface water sample bottle in the Niskin bottle on the left side of the rack. Sample rinse the graduated cylinder twice, fill to the 250 mL mark.

14) Turn the vacuum pump on to the left (surface) filter base for TSS analysis. Filter 500mL of bottom water sample as above. DI Rinse the filter as above.

15) Turn on the vacuum pump to the bottom filter.

16) Lift the funnels off the supports (one at a time) and place on the counter in front of the apparatus. Make sure that the filter stays on the filter support. Turn off the vacuum to

both filters.

17) Rinse the forceps with DI water, shake to remove excess. Pick up the stack of foil tins in one hand and the forceps in the other. Using one hand, carefully open the stack to the tin that corresponds to the filter. It is imperative that each filter be returned to its original tin. The filter on the left (surface) will go into the first tin; the one on the right will go into the second (bottom). Do not remove a tin from the stack and place the tin down on the counter; this will contaminate the tin underneath. Instead, hold the stack of tins in your hand.

18) Carefully slide one side of the forceps under the filter, then grab filter by the edge only, where there is no sample, and gently fold over in half. Use the flat side of the forceps to flatten the filter out at the fold. Do not press so hard as to cause sample to seep out. Remove the filter and place in the appropriate foil tin. If the filter should tear, be sure to place all pieces into the tin as TSS is determined by weight. If a portion of the filter is lost (e.g., torn with missing piece, dropped on the floor, etc.) discard the filter and the foil tin it was in and re-run the sample

19) Repeat for the bottom filter.

As conditions allow and especially if it's a hot day, tins should be placed into the freezer in between stations.

Quality Control/Quality Assurance

Blanks and Duplicates

For quality assurance purposes, surface duplicate samples are filtered at stations M3, A4, and E1 following the same procedures as outlined above.

Filter blanks are prepared and included with the other samples for analysis. These Blanks provide a way to measure any background contamination on the filters caused by field handling procedures. The Blank filters should be treated in the very same manner as a sample filter, except that no sample is filtered. The filters are rinsed with DI water. Blanks are generally prepared following filtering at stations M3, A4, and E1 but can be run at any time during the day after the first station has been filtered.

a) Set up the filters as you would to prepare for sample filtering.
b) Begin as you would for regular sample filtering, starting by mixing the bottom water Niskin. Sample rinse the graduated cylinder. Fill to the 250 mL mark with bottom water sample. Pour into the assembly on the right. Repeat with additional 250 mL sample for a total of 500mL filtered. Swirl DI Water around the inside of the cylinder to rinse the graduated cylinder (~15 ml). Gently pour down the side of the funnel, not directly onto the filter. Repeat. Swirl DI water around the funnel 2-3 times to rinse down the sides.
c) Collect the blank sample. Rinse graduated cylinders with ~15 mL DI water. Pour into the funnel on the right (usually used for surface). Repeat. Swirl DI water around the funnel 2-3 times to rinse down the sides.
d) Place filters in appropriate foil tin following procedure above. The filter on the left will be the blank. The one on the right is the bottom.

e) Reset the filter assembly.
f) Mix the surface water sample in the Niskin.
g) Sample rinse the graduated cylinder twice. Fill to the 250 mL mark. Pour into the assembly on the right. Repeat with an additional 250 mL for a total of 500mL of sample filtered.
h) DI Rinse the cylinder and the funnel as above.
i) Filter an additional 500 mL of surface water through the assembly on the left. DI rinse the graduated cylinder and funnel.
j) Place filters into the appropriate tins as above. The filter on the left is the surface sample. The one on the right is the surface duplicate. Check to be sure that the sample code on the foil pack or container corresponds to the sample that was prepared.

Sample Delivery

Tins for TSS are placed into the freezer after all filtering is completed until the vessel arrives at the dock. For delivery to the analytical lab (CESE at UConn in Storrs, CT) tins are placed into a Ziploc bag and then placed on ice and transported in coolers.

Chain of custody (COC) sheets (Attachment F) are filled out and accompany the samples to the lab. It's important to enter the TSS tin numbers onto the COC in the appropriate spot, following the order of collection. Samples are relinquished to a freezer/refrigerator designated by the laboratory. Copies of the completed COCs are returned (faxed) to LISWQMP staff with the data package.

STANDARD OPERATING PROCEDURE FOR THE COLLECTION OF PHYTOPLANKTON SAMPLES

Summary
Water samples for phytoplankton identification will be collected from ten stations (B3, D3, F2, H4, I2, K2, A4, C1, E1, and J2) using 5-L Niskin bottles mounted on the General Oceanics Rosette Multi-bottle sampling array. Samples will be collected on the upcast from the bottom and at 2 m below the surface of the water.

Equipment/Apparatus
- Rosette with Niskin bottles
- 250 mL graduated cylinder
- 250 mL Amber Nalgene sample bottles
- Lugol's solution (40 g resublimed Iodine, 60 g granular Potassium Iodine, 1000 mL Deionized water)

Safety Precautions
Be sure to wear eye protection and gloves when preserving samples with Lugol's solution (eye and skin irritant). See MSDSs available in the Windsor Lab for additional health and safety information.

Procedure
Samples may be obtained either following collection of BOD samples on the deck, or following the collection of nutrient samples in the laboratory.

1) Remove the surface Niskin bottle from the rack and mix the sample thoroughly by inverting the bottle a few times. Return the bottle to the rack.

2) Sample rinse the graduated cylinder two times with ~10 mL of sample water.

3) Fill the graduated cylinder to the 200 mL mark.

4) Remove the top from the pre-labeled sample bottle. Pour the sample into the bottle. Recap. If the bottle is being re-used from previous surveys, be sure to rinse with DI water prior to filling.

5) Repeat for the bottom water.

6) Using the automatic pipette, preserve with 4 mL of Lugol's solution. Gently swirl the bottle to ensure ample mixing of the preservative throughout the sample.

Sample Delivery
Preserved samples are held onboard in coolers in the dark until delivery to Dr. Senjie Lin's lab at UConn, Avery Point. Samples will be delivered with appropriate chain of custody forms (Attachment F) at the end of the survey.

STANDARD OPERATING PROCEDURE FOR THE COLLECTION OF ZOOPLANKTON SAMPLES

Summary
Mesozooplankton are defined as those animal species within the plankton that are collected with a 200-micron mesh net, whereas, microzooplankton are those plankton species that will pass through a 200 micron mesh net. The primary goals of the Long Island Sound mesozooplankton analysis are to (1) evaluate the spatial and temporal variation in mesozooplankton species composition and abundance, (2) evaluate relationships between mesozooplankton or particular species abundance and nutrient or hydrographic conditions, and (3) provide direct mesozooplankton biomass data for model applications. Mesozooplankton samples will be collected with paired 200 µm mesh, 0.5 m diameter, 2.5 m long plankton nets (SeaGear Corp, Melbourne, FL) each fitted with a calibrated flowmeter attached within the opening to provide an estimation of sampling effort. Microzooplankters are fragile and hence easily damaged or destroyed by nets or pumps. Whole water samples will be collected with the use of 5-liter Niskin bottles from discreet depths within the water column and composited.

Zooplankton samples are only collected from stations B3, D3, I2, F2, H4, and K2.

Equipment/Apparatus

- Rosette with Niskin bottles
- 50L Nalgene carboy
- 8 lengths of Tygon tubing to drain Niskin bottles
- Bongo plankton net fit with 200µm codends and in line flow meters
- (side winch, shackle, 30 lb weight with line)
- >64 µm stainless steel sieve
- 180 µm stainless steel sieve
- 2 mm stainless steel sieve
- 250 mL graduated cylinder
- filtered seawater (obtained from PC/PN filtering) in wash bottles
- Nalgene sample bottles
- Lugol's solution
- 37% formaldehyde

Safety Precautions
Be sure to wear eye protection and gloves when preserving samples with Lugol's solution (eye and skin irritant) and 37% formaldehyde (irritant, corrosive). See attached MSDS available in the Windsor lab for additional health and safety information.

Procedure

Collection of composite sample for whole water plankton sample and >64 µm sample

Water for this plankton analysis is collected using the Niskin bottles on the rosette. Once on station, the rosette is deployed for the CTD cast and to collect water for nutrient analysis. In addition, as bottles are available, samples are collected for the composite on the upcast at 4-6 meter intervals. Depending on station depth and the number of bottles necessary for nutrient analysis, a second cast might be necessary, in which case, water is collected on the downcast.

1) Once the rosette is retrieved, rinse a large carboy (50L) with a small amount of sample water and then drain.
2) Attach Tygon tubes to each of the Niskin bottle stopcocks.
3) Place the other end of the tubes into the top of the large carboy. Make sure the spigot on the bottom of the carboy is turned to OFF!
4) Drain the water from the Niskin bottles into the carboy.
5) Once drained, remove the tubes from the carboy.
6) Replace the screw cap on the carboy and place the carboy in the shade until it can be processed. Reset the Niskin bottles for the next station.

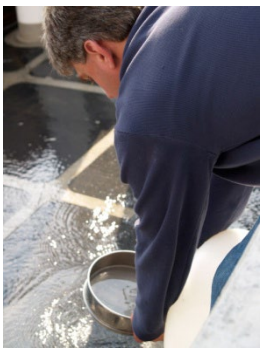
Processing Composite sample

Whole water sample

1) Thoroughly mix the composite sample by gently laying the carboy on its side and lifting back up a minimum of three times.
2) After the sample is mixed place the carboy on a cooler.
3) Loosen the screw top a few turns.
4) The pre-labeled 250mL Nalgene sample container needs to be rinsed prior to collecting the sample.
5) Remove the top from the sample bottle, but don't put the top down.
6) Open the spigot and collect a small amount of sample (~10mL) in the bottle.
7) Close the spigot.
8) Place the top on the bottle and shake/swirl to rinse.
9) Pour out the rinse water.
10) Repeat.
11) To collect the sample, open spigot on the carboy, letting water run for a second.
12) Move the uncapped sample bottle under the spigot and collect sample.
13) Pull the sample bottle out of the flow of water, then shut the spigot on the carboy.
14) Pour off a small amount of sample to leave headspace and room for the preservative. Recap the sample bottle.
15) Whole water samples are preserved with 5% Lugols' solution. Using the dispensette, carefully squirt 8 mL of Lugol's into the sample. (The dispensette is set to dispense 4 mL of solution, so you will need to repeat the procedure twice.) Samples are placed into the refrigerator.
16) Samples are delivered to Dr. McManus at UConn, Avery Point Campus.

Figure 14. Whole Water Zooplankton Sample Processing

>64-µm sample



1) After collecting the whole water sample, tighten the top on the carboy and place it back on the deck.
2) Mix the sample thoroughly by laying the carboy on its side as above.
3) Note the level of water in carboy. This method requires that 10L be filtered through a 64-micron mesh steel sieve.
4) Place the carboy on top of one of the other small stainless steel sieves. Place the 64 uM sieve under the spigot. Remove the cap from the carboy.
5) Open the valve and allow 10 L to filter through the sieve. Close the valve.
6) (if not already full) Fill the designated filtered seawater wash bottle with water collected from the chl-a/PCPN filtering station.
7) Using the wash bottle, rinse the contents of the sieve to one side.
8) Rinse the small white funnel with filtered seawater. Place the funnel into the pre-labeled 125 mL Nalgene sample bottle.
9) Place the sieve on top of the funnel and with the least amount of rinse water possible, carefully rinse the contents of sieve into the bottle.
10) Rinse around the inside of the funnel.
11) Cap the bottle.
12) >64 samples are preserved by adding ~1/8 sample volume of 37% formaldehyde.

<u>Collecting net samples</u>

1. Take the plankton net out of the carrying/storage bag and unfurl.

2. Using a threaded shackle attach the winch cable from the side winch to the top tow ring in the yoke of the net. Be sure that the flow meter outside of the nets is on the upper side of the net frame.

3. Attach the weight to the bottom tow ring.

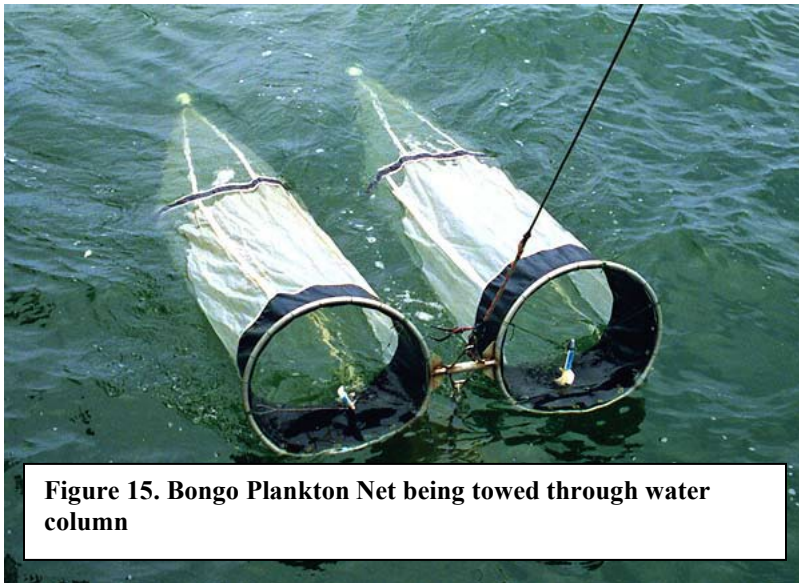


Figure 15. Bongo Plankton Net being towed through water column

4. Attach the codend buckets to the net. Slip the net into codend bucket. Line up the metal retaining clips and close the clamps.

5. Record on the field data sheet the starting numbers for each of the three flow meters. Flow Meter A corresponds to net A and meter B corresponds to Net B. The flow meter

that is attached to the yoke is labeled as meter D.

6. Once on station, don hardhat and life jacket.

7. The hydraulics operator will raise the net off the deck. IMPORTANT- DO NOT lift the net off the deck; let the winch do the work. If you lift the net you could cause the line to go slack and come off the block, damaging it. The operator will also usually lift the weight over the gunwales as well. Assist the operator by putting the codends over the gunwales. Keep the net from swinging into the operator's head or the side of the boat.

8. Swing the net at the yoke so that the net is fully extended in the water with the codends parallel to the boat. Be sure flow meter D is on top.

9. The net is lowered to the bottom at a moderate rate. The scientist will have contact with the line as the net is lowered to determine when the bottom is contacted.

10. Once the weight contacts the bottom, pull up on the cable to keep the net from digging into the bottom sediments and notify the hydraulics operator.

11. The net is then retrieved at a constant slow speed (without stopping).

12. Once the net has returned to the side of the boat, the operator will raise it up and bring in the weight.

13. Using the saltwater deck hose, carefully rinse down the net over the side of the boat (Watch out not to soak the hydraulics operator) from the opening to the codend. Be sure to rinse from outside the net.

15) The net is then lowered to the deck. Leave the codends attached to the net and stand them in an upright position until ready to process. Cover the entire net assembly with the storage bag to block the sun and prevent degradation of the sample and net.

16) Record the flow meter readings on the field sheet.

If the net contacts bottom sediments, rinse out net and codends and recast net. Make sure you record flow meter reading or reset to 0.

If flow meters in nets are reading significantly lower, more than 20%, than flow meter outside of net, recast net after rinsing out net, ctenophores and brown micro-algae often clog net during summer months.

<u>Processing Net Samples</u>

1) With the deck hose rinse down the last few feet of the net into the codend again by holding the net over the gunwale and rinsing from the outside. This is to wash any plankton that got stuck on the net into the cod end. Allow the water to drain to a level below the codend bucket.

2) Stack the 2 mm mesh sieve onto the 180 micron mesh sieve. Tilt the 2 mm sieve at a 45-degree angle and rest it over the 180. The 2 mm sieve is to retain larger zooplankton,

ichthyoplankton, and “gelatinous forms”.

3) Remove the codend bucket from the net. Rinse any material from the codend attachment into the codend with the filtered seawater wash bottle.

4) Pour the contents of the codend through the sieve.

5) Rinse down the sides of the codend bucket with filtered seawater at least two times.

6) Carefully rinse the plankton through the 2 mm sieve into the 180 µm sieve.

7) If gelatinous forms are present measure their volume by pouring them from the 180 mm sieve into the designated graduated cylinder. Record the total volume and species composition on datasheet. After recoding volume and composition, dispose of gel forms overboard.

8) Rinse the contents of the 180-micron sieve into the >64 µm sieve. This is done for ease of getting the sample into the container with the smallest volume possible.

9) Rinse the contents of the >64 sieve to one side.

10) Set the sieve aside at an angle.

11) Select the smallest volume Nalgene sample bottle possible based on the contents of the sieve allowing sufficient space for the sample, wash water, and preservative. Rinse the funnel with filtered seawater and rinse the sample bottle.

12) Put the funnel into the sample bottle. Carefully rest the sieve on the funnel and using the wash bottle, carefully rinse the plankton into the funnel and then into sample bottle. Be sure to leave room for the fixative.

13) Repeat the entire procedure with net B.

14) Samples are preserved with 37% formaldehyde. Mark a line on the sample bottle to indicate the volume of sample. Estimate one quarter this volume. Mark a line that is one quarter above the sample volume line. Add formaldehyde to this line.

15) Rinse the sieves, graduated cylinder and codend bucket with saltwater and freshwater in between tows.

16) Reattach the cod ends and reset the flow meters for the next station.

17) At the end of the day, disconnect net from the weight and side winch cable. Return the net to the storage bag. If possible, rinse down the net with the freshwater hose and dry slightly before storage.

Sample Delivery
Preserved samples are held onboard in coolers in the dark until delivery to Dr. Hans Dam or Dr. George McManus at UConn, Avery Point in Groton with appropriate chain of custody forms (Attachment F) following completion of the survey.

ATTACHMENTS

ATTACHMENT A

Research vessel policies, rules, and safety information

PFD Directive

Updated Dempsey Safety Manual

Fishing Vessel Sheet 1 - Cold Water

Fishing Vessel Sheet 2 - Boundary Line and Coastal Waters

Fishing Vessel Sheet 3 - Immersion Suits

Stearns Survival Suit

Fishing Vessel Sheet 4- Visual Distress Signals

Fishing Vessel Sheet 5 – Survival Craft

MARPOL treaty card

MANUAL CODE **5560**

D **11**

Department of
Environmental Protection

DIRECTIVE

SUBJECT: USE OF PERSONAL FLOTATION DEVICES BY DEP STAFF

PURPOSE: To establish department policy regarding the use of Personal Flotation Devices (Life Jackets) for department staff.

POLICY: It is the policy of the Department of Environmental Protection that all employees in performance of their duties shall wear an appropriate Personal Flotation Device (PFD) when aboard and underway on any vessel. This policy shall be extended to vessels at anchor when conditions (i.e. weather, sea, work) and the safety of the employee dictate, or the supervisor deems it appropriate, that an approved PFD be worn. Such PFD's shall be U.S. Coast Guard approved and in serviceable condition. For definitions of the different types of PFD's and when they should be worn, see the Connecticut Boaters Guide.

The only exceptions to this policy are:

1. when on a vessel required to have a certificate of inspection issued by the Coast Guard unless directed by the First Mate or the Captain (i.e. *RV John Dempsey* or any passenger ferry), or

2. when below deck or in an enclosed cabin or in the cabin of a large patrol vessel (i.e. 35 feet or longer), or

3. for Lifeguards, when engaged in patrol surveillance from a vessel where the use of a PFD would endanger the lifeguard or hinder the rescue or ability to rescue. Lifeguards on patrol surveillance shall possess rescue equipment required by DEP Lifeguard Policies and Procedures.

Supervisors are responsible for ensuring that their employees are aware of this policy and that they have access to and training regarding the use of the appropriate equipment.

Employees in violation of this directive will be subject to disciplinary action.

Issued by: /S/ Commissioner Amey Marrella

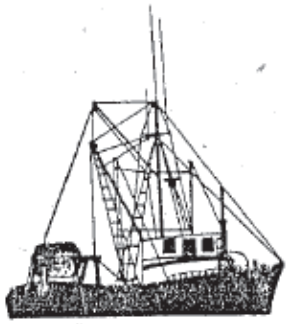
Date: April 19, 2010

Special Instructions: Replaces Manual Code 5560 D11 dated September 29, 2009

Distribution: Electronic

FISHING VESSEL SAFETY

FACT SHEET



#1 Cold Water

Figures 1-5 illustrate Cold Water locations. Cold Water continues north of the latitude shown. The drawings are not to scale.

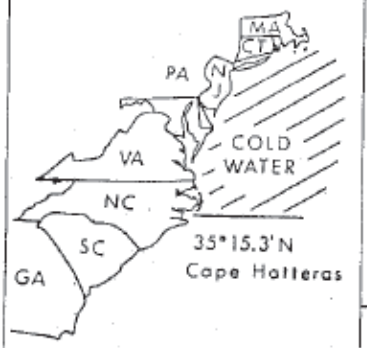


Figure 1. December through April

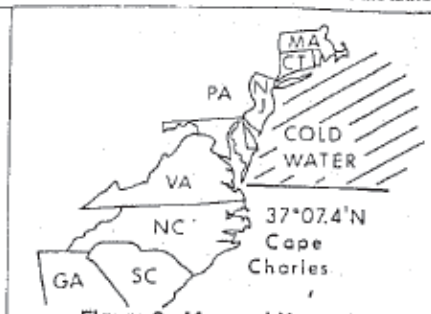


Figure 2. May and November

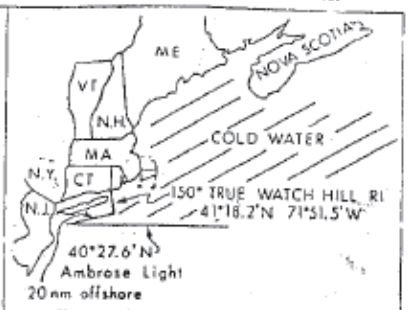


Figure 3. June and October

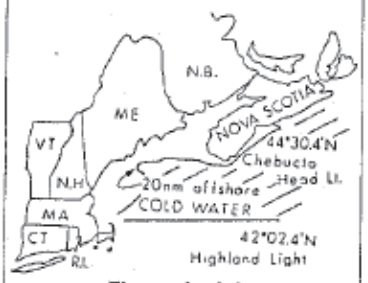


Figure 4. July

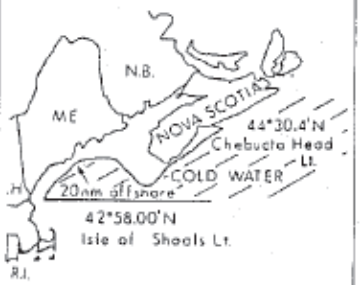


Figure 5. August and September

The Fishing Vessel Safety Regulations define *COLD WATER* as water where the monthly mean water temperature is normally 59 degrees Fahrenheit or colder. The *COLD WATER* southerly line changes depending on the time of year (Refer to Figures 1-5).

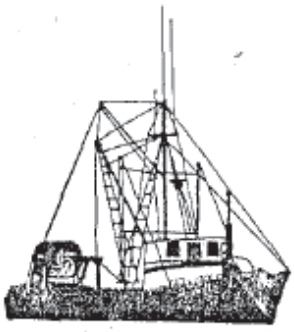
Much of the equipment required by the Fishing Vessel Safety Regulations depends on if you operate a vessel, regardless of size, in *COLD WATER*. The reason behind this regulation is the possibility of hypothermia. Examining many of the accident reports, it was discovered that many deaths were not a result of the accident itself, but of unprotected people being exposed to cold water causing hypothermia. Hypothermia basically means that your body is unable to control heat loss, because the temperature surrounding it is too cold. Your body core temperature drops, causing a variety of symptoms, including unconsciousness and death. The colder your environment, the faster your body loses heat. Water robs your body of heat much faster than the same air temperature. A temperature of 59° F seems to be a critical temperature for a person with no protection. This temperature is painful and hypothermia seems to progress much faster than in warmer temperatures. In waters warmer than this, most people are able to survive at least a couple of hours.

For more information on *COLD WATER*, please refer to United States Coast Guard Navigation and Vessel Inspection Circular (NVIC) No# 7-91.

The Fishing Vessel Safety Fact Sheet series is being jointly sponsored by the Rhode Island Sea Grant Program, the University of Rhode Island Cooperative Extension Service and the US Coast Guard, First District.

FISHING VESSEL SAFETY

FACT SHEET

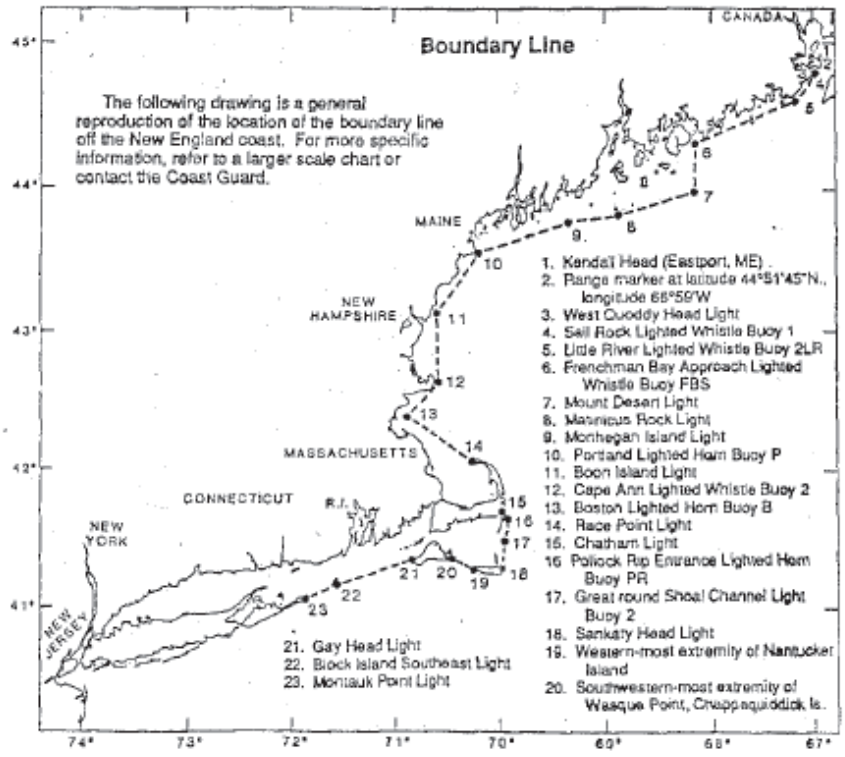


#2 Boundary Line and Coastal Waters

The *boundary line* is defined in 46 CFR, part 7 as a line that generally follows the trend of the seaward high water shoreline and cross entrances to small bays, inlets and rivers. In some areas, the *boundary line* is found as much as 12 miles from shore, and in other areas, the line comes ashore.

This line should not be confused with the other lines used in reference to other regulations. These are the *Territorial Sea* line (found 3 nautical miles offshore), the *Contiguous Zone* line (found 12 nautical miles offshore) and the *Fishery Conservation Zone* line or *Exclusive Economic Zone* (found 200 miles offshore).

The *Territorial Sea* line divides the **coastal waters** from the **high seas** (in reference to the EPIRB requirements only). Waters out to the *Territorial Sea* line are termed **coastal waters. Coastal waters** also include those waters directly connected to the territorial seas , such as bays, sounds, harbors, rivers, inlets, where any entrance exceeds 2 nautical miles between opposite shorelines.



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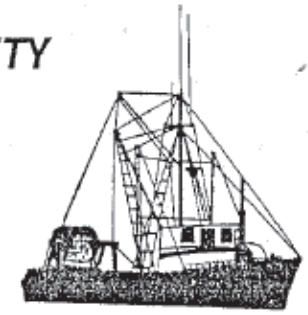
FISHING VESSEL SAFETY

FACT SHEET

3 Immersion Suits



Who Needs One?

Immersion suits (or gumby suits as many of us affectionately call them) are one of the most important safety items we can have with us at sea. They stuff an awful lot of benefit into a relatively small package. In the event that we end up in the water, a well-cared for suit will provide us with protection against the cold water, keep us floating and hopefully from drowning, will provide a brighter, bigger object for the rescuers to locate, and we can attach all sorts of accessories ie. strobes, mirrors, dye packs, and EPIRBs to help us attract attention. And they are REQUIRED as of November 15, 1991 on the following vessels:

FOR ALL VESSELS

Those vessels that operate in COASTAL WATERS or beyond in COLD WATER (refer to Fact Sheet 1), an immersion suit or exposure suit of the proper size is required for each and every person on board with an approved PFD light. For those vessels operating in warm waters, a PFD of the appropriate type may be substituted for an immersion suit.

FOR DOCUMENTED VESSELS

Those vessels that operate seaward of the BOUNDARY LINE (Refer to Fact Sheet 2) as defined by 46 CFR part 7, an immersion suit or exposure suit of the proper size is required for each and every person on board with an approved PFD light.

TABLE 28.110.—PERSONAL FLOTATION DEVICES AND IMMERSION SUITS

Applicable waters	Vessel type	Devices required	Other regulations
Seaward of the Boundary Line and North of 32° N; or South of 32° S; or Great Lakes.	Documented vessels	Immersion suit or exposure suit [1]	28.135; 25.25–9(a); 25.25–13; 25.25–15.
Coastal waters or beyond *cold* waters (includes Great Lakes).	All vessels	....do [1]	Do.
All other waters	40 feet (12.2 meters) or more in length	Type I, Type V commercial hybrid, immersion suit, or exposure suit [2].	28.135; 25.25–5(e); 25.25–5(f); 25.25–9(a); 25.25–13; 25.25–15.
Do	Less than 40 feet (12.2 meters) in length.	Type I, Type II, Type III, Type V commercial hybrid immersion suit, or exposure suit [2].	Do.

[1] Until September 1, 1995, individuals weighing less than 44 pounds (196 Newtons) may substitute an approved personal flotation device of the appropriate size for a required immersion suit or exposure suit.
[2] Certain Type V personal flotation devices are approved for substitution for Type I, II, or III personal flotation devices when used in accordance with the conditions stated in the Coast Guard approval label.

Although seemingly straight-forward, there are some situations where confusion can arise. For example, a fisherman from Rhode Island fishing the open coastline of Rhode Island in any vessel, would need an immersion suit only in the months of October, November, December, January, February, March, April, May and June. But that same fisherman would not need an immersion suit during the months of July, August and September (because it is not defined as cold water) unless he had a documented vessel operating seaward of the boundary line.

The Fishing Vessel Safety Fact Sheet series is being jointly sponsored by the Rhode Island Sea Grant Program, the University of Rhode Island Cooperative Extension Service and the US Coast Guard, First District.

STEARNS®

HYPOTHERMIA AND COLD WATER SURVIVAL FACTS

IFS-580 Industrial Flotation Suit

IFS-7580 Recreational Flotation Suit

Time runs out fast in a freezing sea.

5 minutes to realize you're missing.
5 minutes to organize the search.
5 minutes to pull you to safety.

Every second counts when the water is shocking cold. Flotation gear alone will keep you afloat, but offers minimal protection against hypothermia. *Stearns Rule of 50* illustrates the danger of cold water: "In water of 50°F, you have a 50-50 chance of surviving beyond 50 minutes."

That's why Stearns developed "The Work Suit" (IFS-580). This industrial flotation suit looks and wears like an ordinary work coverall. But it's much, much more.

Core-Guard® design features high-buoyancy PVC to meet U.S. Coast Guard Type V requirements. This closed-cell foam also acts as insulation to help retain the body's core temperature.

"The Work Suit" features a foam lined hood, inflatable head support, Vislon® zippers, plus take-up straps around wrists, thighs and ankles.

FOREWARD

In water-related activities there is always an element of risk. Falling into even relatively warm water can soon lead to cooling of the body (Immersion Hypothermia), resulting in disorientation, unconsciousness, and ultimately heart failure. Other threats in a survival situation include despair and trauma . . . any of which is harmful and may lead to death depending upon the specific situation.

The scientific studies at the Hypothermia-Cold Water Institute at the University of Minnesota - Duluth School of Medicine, and human subjects immersion tests conducted in the frigid waters of Lake Superior have contributed significantly to our efforts to increase the survival time and raise the level of safety and protection of the mariner against unreasonable risks associated with cold-water immersion.

In a survival situation, your personal water safety relies significantly on your knowledge and ability to meet life-threatening conditions. **There is no substitute for preparedness.** The following information requires your careful consideration, and should improve your chances for survival in cold water.

Refers to Work Suit coverall — available for use on RJV John Dempsey

Meaning of

SPECIAL USE APPROVAL

This pamphlet explains the "Special Use Approval" of this PFD by the United States Coast Guard . . . and provides additional information about the performance, protection and safety features afforded by this product which may not be provided by the more conventional PFDs discussed in the accompanying PFD information pamphlet.

1. **Special approvals** are granted by the United States Coast Guard for PFDs which do not meet all the requirements for approval under Types I, II, III and IV . . . but which offer other safety features.

2. **What is meant by "restricted approval" of this Type V PFD?**

   This device cannot be donned as quickly as a conventional PFD and, therefore, it must be worn at all times to be accepted as a U.S. Coast Guard approved device.

   This PFD provides significant Hypothermia protection as explained in this pamphlet. **For recreational use,** this device may be used to meet the requirements for carrying a Type III PFD. **For use on commercial inspected vessels,** it may be carried only as additional equipment, such as a work vest.

3. **What is the purpose and use of the head support on this Type V Buoyant Suit?**

   The head support is designed to increase the amount of freeboard, and to improve the field of vision by placing the head at an angle which enhances the wearer's ability to sight search and rescue craft and floating debris.

   It also keeps more of the head (which is a high heat loss area) out of the water.

4. **How do I care for the head support?**

   Before each use the head support should be inspected to ensure satisfactory operation in a possible emergency:

- **CHECK THAT IT IS FREE FROM RIPS, TEARS OR PUNCTURES. THE INFLATABLE SHOULD BE CHECKED FOR LEAKAGE.**

  To check for leaks, inflate the device until firm and leave overnight. If the device has not lost its shape overnight, it is fine. If it has lost its shape, a leak may be looked for by holding the device under water.

  A leaking valve can be easily detected and may be washed or blown clear to work again. A leak in the inflation chamber may be recognized by an increase in bubbles with an increase in pressure on the chamber. Anything more than a mild squeeze is unnecessary.

- **STORAGE**

  When not in use, the suit should be stored on a coat hanger in an area where it will not be damaged. A cool, dry area is ideal.

  Prevent sharp or heavy objects from coming in contact with the suit and head support.

  A wet suit should be allowed to dry naturally but not allowed to remain damp for long periods. Do not dry in a dryer or in front of a direct source of heat such as a radiator.

5. **Can I wear an additional conventional PFD with this Type V Buoyant Suit?**

   Yes. When this Buoyant Suit is worn with a PFD it is recommended that the conventional PFD be placed over this Type V Buoyant Suit.

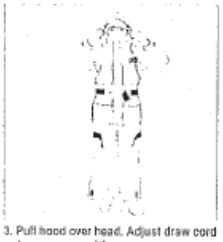
NOTE: ANY "TURNING MOMENT" WHICH MAY BE PROVIDED BY THE CONVENTIONAL PFD WILL BE DECREASED WHEN WORN WITH THIS TYPE V BUOYANT SUIT.

INSTRUCTIONS FOR USE
DONNING

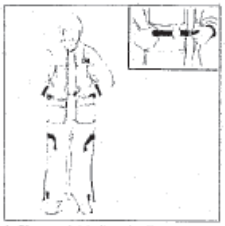
It is recommended that all closures be secured as shown before entering the water, where possible, or immediately after immersion to ensure maximum Hypothermia protection.

1. Pull on as you would a pair of coveralls.

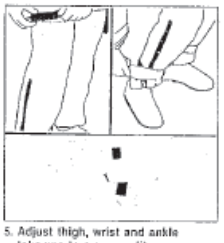
2. Close chest and leg zippers with a slow even pull. Secure velcro storm flap.



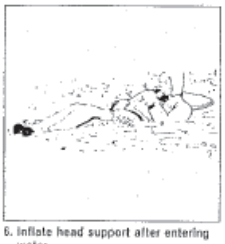
3. Pull hood over head. Adjust draw cord to a snug position.



4. Close waist belt and adjust to snug fit.



5. Adjust thigh, wrist and ankle take-ups to a secure fit.



6. Inflate head support after entering water.

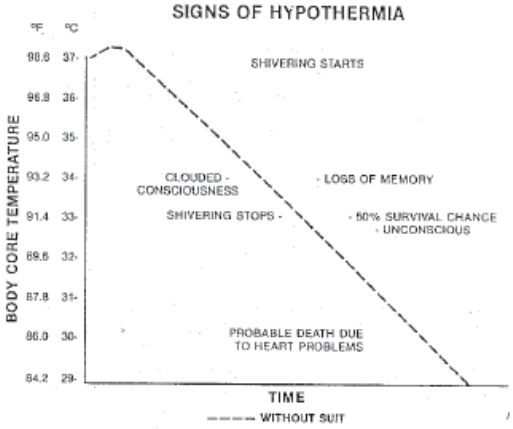
NOTE: ACTUAL DONNING AND IN-WATER TESTING IS RECOMMENDED.

HYPOTHERMIA

1. What is Hypothermia?

It is the lowering of the body-core temperature (heart, brain and other vital internal organs) of approximately 2°C or more (from the normal 37°C). The skin and muscles cool rapidly in cold water, while the temperature of the heart, brain and other vital internal organs generally begin to fall after 15 to 20 minutes. The body attempts to increase heat production by shivering, but the effort yields only a small amount in comparison to the heat loss from the body when exposed to cold water.

SIGNS OF HYPOTHERMIA



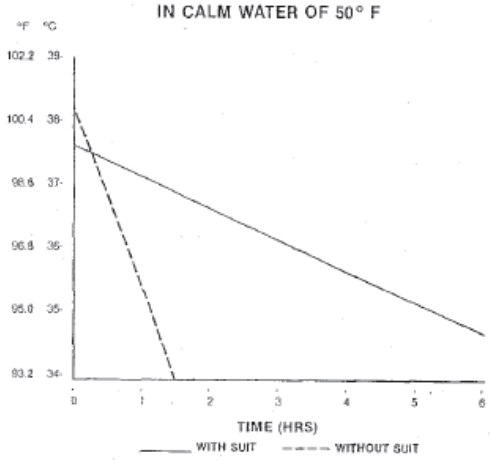
The absence of shivering at a core temperature of approximately 33°C indicates that the body has given up its defenses against the cooling. A state of unconsciousness follows shortly thereafter. Death, as a result of body cooling, may occur when the heart temperature falls below 30°C.

2. **How long can I survive in cold water while wearing this Buoyant Suit?**

Wearing a PFD of any kind will not ensure survival in water, although all types can help. Several other factors will influence the length of time a person can survive in water, including body-type, body-attitude, physical condition, amount of subcutaneous (beneath the skin) body-fat, clothing, temperature of the water and the will to survive. There is no universal rule as to temperature and survival time, as resistance to cold and instinct for self-preservation differ greatly.

However, the predicted survival time for average adult humans immersed in calm water of 50°F (10°C), wearing this Type V Buoyant Suit over light clothing, is about 14 hours; whereas, without the suit the predicted survival time is approximately 3 hours. **Times derived from human subjects testing of this type are only approximate.**

AVERAGE RECTAL TEMPERATURES
IN CALM WATER OF 50° F



An individual unexpectedly immersed in cold water without a flotation device or PFD has virtually no chance of conserving or minimizing heat loss. In fact, many individuals, upon capsize, seem to lose the ability to clamber back on board or to hang onto some craft or object, as there is a progressive decline in muscle strength following immersion in cold water.

An individual with a vest-style PFD can reduce the rapid heat loss by assuming a heat conservation position in the water, depending on the type of PFD being worn (see page 8).

3. **While awaiting rescue, should I attempt to get out of the water?**

Yes. Heat loss to cold air is much less than that to cold water . . . even when the air temperature is much lower than the water. **Always try to get out of the water.**

4. **Will I have difficulty climbing out of the water while wearing this Buoyant Suit?**

You may. Even an immediate effort to climb aboard a capsized boat, a floating piece of wreckage, or a life raft is difficult because of increased bulk and temporary entrapment of water in the suit. Your extremities are quickly numbed by cold since the body reduces its blood supply to the skin, arms and legs.

You can reduce the difficulty of climbing out of the water by opening the arm and leg closures . . . which will allow much of the entrapped water to escape from the suit.

REMEMBER: THE MORE OF YOUR BODY YOU CAN GET OUT OF THE WATER THE SLOWER YOUR HEAT LOSS.

5. **Will swimming increase my survival time?**

No. An average adult in light clothing cannot swim more than approximately 350 feet in water temperatures of 10°C (50°F) before losing consciousness as a result of body cooling. A person wearing a vest-style PFD can survive about 30% longer when completely still in cold water than when moving vigorously or swimming.

6. **What should I do in the event of accidental immersion?**

Try to climb back into the boat, on top of an overturned craft or onto any other floating wreckage, since water draws heat from the body as much as 30 times faster than air of the same temperature.

If you are wearing a PFD, stay in place quietly. For maximum heat conservation, press the PFD to your chest, hold both arms against your body, and keep your legs tightly together.

TYPE V BUOYANT SUIT POSITION

TYPE III POSITION

7. **What is the proper treatment of Hypothermia?**

Since there is no simple, universal method of treatment, it is not possible to state which method is best. When possible, it is advisable to call a doctor as quickly as possible.

The following section is designed to offer general guidelines for use by the medically inexperienced person who must attempt to rewarm a victim without the benefit of monitoring equipment. **THE TREATMENT OF HYPOTHERMIA IS REWARMING.**

Every victim of Immersion Hypothermia is a candidate for passive rewarming. The following points should be considered in all situations of Immersion Hypothermia.

- If at all possible, lift the victim out of the water horizontally (a person lifted vertically out of the water may suffer sudden heart failure).
- Avoid rough handling (this may open the blood vessels in the skin, sending warmer blood from the body's core to the cooler regions, leading to sudden further drop in temperature).

  A cold heart is very sensitive to mechanical disturbances. Rough handling may contribute to heart failure.

If conscious:

- Gently remove the victim's wet clothing; and,
- If possible, reclothe in dry clothing . . . cover the head and wrap a scarf around the neck; and,
- Encourage movement . . . to stimulate shivering and subsequent generation of heat; and,
- Give warm sweet drinks . . . but under no circumstances is alcohol to be used.
- If the victim is unable to walk, consider wrapping the victim in blankets or a sleeping bag.
- Apply external warm compresses to the head, neck, trunk and groin.

If unconscious:

- Make certain the victim has an open air passage . . . is breathing . . . and has a pulse.

BREATHING AND PULSE MAY BE SLOW AND SHALLOW, AND DIFFICULT TO DETECT. TAKE UP TO A FULL MINUTE TO MEASURE THESE VITAL SIGNS.

- Seek immediate medical assistance.
- **Remember:** Never try to give an unconscious or semi-conscious person anything to swallow.

If Lifeless:

. . . don't assume the person is dead just because he is very cold.

One of the human response patterns to immersion in cold water is the oxygen-conserving dive reflex . . . an involuntary suspension of respiration. This is triggered by the sudden contact of very cold water with the face. It serves to help protect the brain from severe oxygen deprivation . . . and even though the person may not be breathing, the reflex directs oxygenated blood to the trunk of the body, thereby prolonging critical function of the life support organs.

- CLEAR THE AIR PASSAGE WAYS.
- APPLY CPR.
  Perform artifical ventilation (moving air into and out of the lungs).
  Perform external chest compression.
- TRANSPORT TO A MEDICAL FACILITY.

DON'T GIVE UP!

ALCOHOL

1. **Does the use of alcohol contribute to the effects of Hypothermia?**

   Yes. It can lead to Hypothermia because it reduces the shiver response . . . another source for heat production; and, it alters the thermal regulatory process, reducing the effectiveness of the body's cold stress response mechanisms . . . to a point so drastic as to trigger the onset of severe Hypothermia with a thermal stress.

Also, research studies at the Hypothermia-Cold Water Institute indicate that the use of alcohol intensifies disorientation which may cause death.

SPECIAL TIPS:

1. All PFDs increase survival time because they allow you to float without using energy. Some PFDs help because of the insulation they provide.
2. Life-support equipment must always be in serviceable condition.
3. A PFD with a well-insulating hood and gloves is recommended, as heat loss from the head and hands is substantial.
4. Before abandoning the ship, wear a PFD (properly donned) . . . and put on as much warm clothing as possible.
5. If abandoning ship by direct entry into the water:
   a. Be sure your PFD is secured correctly.
   b. Use one hand to protect your nose, and the other to hold on to the PFD.
   c. Keep your feet together, check below for obstructions, and jump feet first.
   d. Survivors should remain together for distress relief.
6. Always try to get out of the water onto floating wreckage or an overturned boat.
7. Control your breath . . . cold water in the face provokes choking and eventual panic.
8. Do not swim unless it improves your situation.
9. When in water: Keep your legs together, and your arms close to the body in order to reduce heat loss.
10. The will to survive is your best weapon. Concentrate always on how to improve the situation.

NEVER GIVE UP!

Questions to which this pamphlet may not have responded may be referred to:

Stearns Manufacturing Company
St. Cloud, Minnesota, USA 56302
Telephone: (612) 252-1642
Telex: 291105
Fax: (612) 252-4425

This informative booklet was prepared and printed by Stearns Manufacturing Company, as a service to the boating public in the interest of greater safety in and about the water through a better understanding of the proper use and function of life-support equipment.

Printed in U.S.A.
SMC 1189-15M

an Anthony Industries company

Available for use on R/V John Dempsey

FOR MAXIMUM HYPOTHERMIA PROTECTION...

COLD WATER IMMERSION SUIT

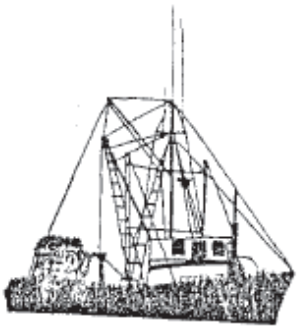
"COLD WATER IMMERSION SUIT™" ISS-590i

Designed for the most severe offshore conditions. U.S.C.G. and IMO requirements state that a temperature drop of no more than 2°C is to be experienced over a six hour period in 35°F (1.7°C) water. The Stearns ISS-590i worn over normal clothing, easily surpasses those specifications. Tests indicate the ISS-590i Cold Water Immersion Suit provides hypothermia protection for an extended period during cold water immersion.

Features include 100% neoprene construction, face shield that allows 120° unrestricted vision, waterproof zipper, attached full finger gloves, marine whistle, light pocket, retro-reflective materials front and back. Each suit comes in its own storage bag, color-coded by size. Suits are International Safety Orange.

FISHING VESSEL SAFETY

FACT SHEET



#4 Visual Distress Signals

A visual distress signal is anything that makes you bigger, brighter or more noticeable to someone trying to find you. By yourself, you are a pretty small target in a very large ocean, even under ideal conditions.

Table 28.145 lists the distress signal requirements for fishing vessels. These are in addition to those required to be carried in the service pack (SOLAS A, B or Coastal Service Pack) found in your inflatable buoyant apparatus. The type of distress signal you are required to carry depends on how far offshore you operate.

TABLE 28.145.—DISTRESS SIGNALS

Area	Devices required
Ocean, more than 50 miles from coastline.	3 parachute flares, approval series 46 CFR 160.136; plus 6 hand flares, approval series 46 CFR 160.121; plus 3 smoke signals, approval series 46 CFR 160.122.
Ocean, 3–50 miles from the coastline; or more than 3 miles from the coastline on the Great Lakes.	3 parachute flares, approval series 46 CFR 160.136, or 160.036; plus 6 hand flares, approval series 46 CFR 160.121 or 160.021; plus 3 smoke signals, approval series 46 CFR 160.122, 160.022, or 160.037.
Coastal waters, excluding the Great Lakes; or within 3 miles of the coastline on the Great Lakes.	Night visual distress signals consisting of one electric distress light, approval series 46 CFR 161.013 or 3 approved flares; plus Day visual distress signals consisting of one distress flag, approval series 46 CFR 160.072, or 3 approved flares, or 3 approved smoke signals.[1]

[1] If flares are carried, the same 3 flares may be counted toward meeting both the day and night requirement.

The approval series number to meet fishing vessel safety requirements are very important. Check in Table 28.145 to see what applies to your vessel. The approval number series that begin with 160.0XX are not SOLAS approved. Only those approval numbers in which a "1" has replaced the "0" (ie. 160.1XX) meet the requirements of SOLAS. Note that vessels traveling more than 50 miles from the coastline are required to have SOLAS approved flares and smoke signals. Carefully check the numbers before you purchase them.

Vessels that operate in coastal waters must have night signaling devices and day signaling devices. If flares are carried, the same three flares may be counted toward meeting both the day and night requirement. Otherwise, you may choose an electric light (46CFR 161.013) for the night requirement. You can choose between a distress flag (46CFR160.072) or any 3 USCG approved flares, or any 3 USCG approved smoke signals for the daytime requirement.

Vessels operating between 3 and 50 miles from shore and those greater than 50 miles from shore must carry the same number of distress signaling devices. The difference is that the vessels operating more than 50 miles offshore MUST have SOLAS approved distress signaling devices. The approval numbers must be 160.1XX.

Flares must be treated carefully. You should store them in a cool, dry place in a watertight container. They should be protected from banging into one another. Pyrotechnics have expiration dates, usually of three years. Expired signals do not count as part of the requirements. Dispose of any expired or bulging pyrotechnics immediately and properly. Contact your raft repacker for detailed instructions. Flares contain flammable metal powders which are classified as class D substances and if involved in a fire must be treated with a dry powder fire extinguisher. Never use water as some metals react violently when in contact with water.

Remember, only use flares when you know a rescuer is in the area, otherwise you have wasted it!

The Fishing Vessel Safety Fact Sheet series is being jointly sponsored by the Rhode Island Sea Grant Program, the University of Rhode Island Cooperative Extension Service and the US Coast Guard, First District.

Marine Distress Communications Form – *Speak slowly, clearly and calmly.*

1) Make sure your radio or radiotelephone is on.

2) Select 156 MHz (channel 16) or 2182 KHz.

3) Press microphone button and say "Mayday — Mayday — Mayday."

4) Say: "This is _____, _____, _____, _____, over."

5) Release the microphone button briefly and listen for acknowledgement. If no one answers, repeat steps 3 and 4. If there is no acknowledgement or if the Coast Guard or another vessel responds,

6) Say: "Mayday _____."

7) Describe your position in lat/long coordinates, in Loran-C coordinates, or by range and bearing from a known point.

8) State the nature of your distress.

9) Give the number of persons aboard and the nature of any injuries.

10) Estimate the present seaworthiness of your boat.

11) Describe your boat: _____

_____

12) Say: "I will be listening on channel 16/2182."

13) End message by saying: "This is _____, _____, over."

14) If your situation permits, stand by the radio to await further communication from the Coast Guard or another vessel.

Fill in blanks with vessel name, call sign and description using a permanent marker or a ballpoint.

Produced by **University of New Hampshire Sea Grant Extension and Cooperative Extension**

Provided by the **Rhode Island Sea Grant Program**

FISHING VESSEL SAFETY

FACT SHEET

#5 Survival Craft

There are so many different types of survival craft referred to in the safety regulations that its worthwhile defining the terms before we start. These are listed in ascending hierarchical order. In other words, you can always substitute something that exceeds the requirements for your vessel (as long as it meets Coast Guard Approval).

1. Buoyant Apparatus
These are "ready to use" rigid ring or oval platforms that require no inflation. Although they may be big enough for one of two people to climb up out of the water onto the platform, they are equipped with lifelines for people to tie-off to. They must have retroreflective material, lifelines, painter, pendant and an electronic light, and must be marked with the name of the vessel. They have no canopy or equipment pack.

2. Life Floats
These are rigid ring shaped apparatus which are ready to use. They do not provide enough space for people to climb out of the water, however, they have tie-off lifelines. These must also have retroreflective tape, painter, pendant, electronic light and be marked with the name of the vessel. They have no canopy or equipment pack.

3. Inflatable Buoyant Apparatus
These are similar to inflatable life rafts except that they carry no canopy or equipment packs. These must have a lifeline, pendant, painter and an electronic light. These are packed with vessel identification and retroreflective material.

4. Inflatable Life Rafts.
A Coast Guard approved liferaft is constructed in accordance with 46 CFR 160.018. A liferaft consists of side and end buoyancy chambers with equipment and provision compartments surrounding a watertight compartment or well deck. Liferafts are designed to allow all the people for which the raft is approved for to enter the raft and be protected from the elements. All inflatable liferafts have canopies.

Tables 28.120 (a), (b) and (c) provide the breakdown as to which survival craft you must have on board your vessel. The regulations are broken down into those for documented vessels, undocumented vessels with not more than 16 persons on board, and undocumented vessels with more than 16 people on board. Additionally, requirements depend on the area of vessel operation.

In general, documented vessels operating in the most exposed routes require an inflatable liferaft with enough capacity to accommodate all individuals on board (Don't take the manufacturers word for it-see if your crew fits inside). They must contain a SOLAS A or B pack. For example, a documented vessel operating beyond 50 miles of the coast in any water temperature, must have an inflatable liferaft with a SOLAS A pack. If the vessel operates in cold water between 20-50 miles from the coast, it must have a SOLAS B pack. If the vessel is undocumented with more than 16 people on board, it must have an inflatable liferaft with the appropriate pack, depending on the area of operation. IMPORTANT: In the tables, if no pack is specified with an inflatable liferaft, it must be

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The Fishing Vessel Safety Fact Sheet series is being jointly sponsored by the Rhode Island Sea Grant Program, the University of Rhode Island Cooperative Extension Service and the US Coast Guard, First District.

equipped with a Coastal pack. For vessels on less exposed routes (usually close to shore), a less sophisticated survival craft may be used. Inflatable buoyant apparatus, rigid buoyant apparatus or life floats can be used depending on the area of operation. In other warmer, more protected waters, survival craft may not be required at all.

At this time, vessels with less than 4 people on board operating within 12 miles of coast are not required to carry survival craft. However, this situation is being examined and future supplemental notice of proposed rulemaking will be forthcoming.

WHEN DO I NEED ONE?

Except for new vessels or those undergoing a major conversion, the following dates are phase-in times for survival craft installment.

1. *September 1, 1992*: Documented vessel that operates in the North Pacific area.

2. *September 1, 1993:* Documented vessels that operate in the Great Lakes or in the Atlantic Ocean north and east of a line drawn at a bearing of 150° true from Watch Hill, Rhode Island.

3. *September 1, 1994:* All other documented vessels.

4. *September 1, 1995:* All other vessels.

For example, a 36 ft undocumented vessel with less than 16 persons on board, operating off the coast of Maine in December (remember, the cold water areas move around) inside the boundary line has until Sept 1, 1995 to equip the vessel with a buoyant apparatus. A documented vessel operating between 20-50 miles of the coast of Rhode Island in May (refer to cold water line) must have an inflatable liferaft (SOLAS B pack) by September 1, 1993.

WHAT ABOUT OLD EQUIPMENT?

Survival craft installed on a vessel before September 15, 1991 may continue to be used IF:

1. It is of the same type required in Tables 28.120 (a), (b) or (c).

2. It is maintained in good and serviceable condition.

3. It is equipped with the proper equipment pack.

SAFETY DRILLS

As required under the Instruction section of the safety regulations, one of the monthly drills must cover abandoning the vessel and one must cover launching survival craft operations (If you are required to have one). Drills must be conducted on board the vessel as if there were an actual emergency and must include the participation of all individuals on board. This regulation is in effect *now*. There is no substitute for learning by doing. You can ask the Coast Guard to observe your drills during their complimentary dock side exam if you have questions about the drill content or procedure.

STORAGE OF SURVIVAL CRAFT

Although there is some disagreement as to where to put your survival craft, the fishing vessel safety regulations state that:

1. Each inflatable liferaft required to be equipped with a SOLAS A or B equipment pack must be stowed so as to float free and automatically inflate in the event the vessel sinks.

2. Each inflatable liferaft, inflatable buoyant apparatus, and any auxiliary craft used in their place, must be kept ready accessible for launching or be stowed so as to float free in the event the vessel sinks.

3. Each hydrostatic release unit used in a float free arrangement must be approved. (under part 160, subpart 160.062 of this chapter). These approved types are listed in the Coast Guard Equipment List publication.

4. Each float free link used with a buoyant apparatus or with a life float must be certified. (to meet Part 160, Subpart 160.073 of this chapter). These are listed in The Coast Guard Equipment List publication.

FACT SHEET #5, PAGE 3

TABLE 28.120 (a).—SURVIVAL CRAFT FOR DOCUMENTED VESSELS

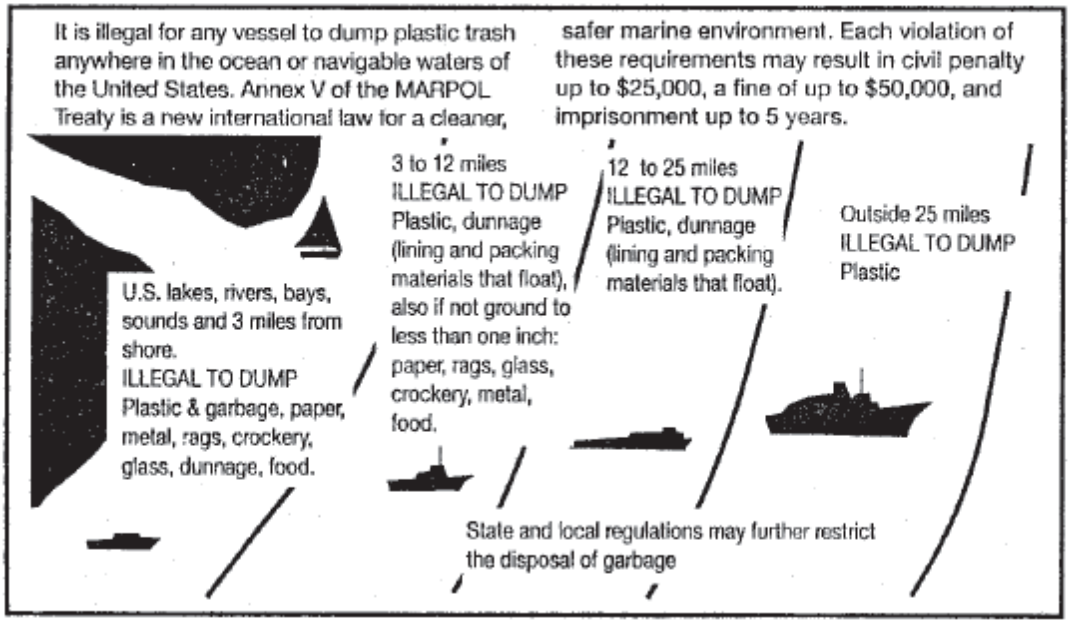
Area	Vessel type	Survival craft required
Beyond 50 miles of coastline	All	Inflatable liferaft with SOLAS A pack.
Between 20–50 miles of coastline, cold waters	All	Inflatable liferaft with SOLAS B pack.
Between 20–50 miles, of coastline, warm waters	All	Inflatable liferaft.
Beyond Boundary Line, within 20 miles of coastline, cold waters	All	Inflatable liferaft.
Beyond Boundary Line within 20 miles of coastline, warm waters.	All	Life float.
Inside Boundary Line, cold waters; or Lakes, bays, sounds, cold waters; or Rivers, cold waters.	36 feet (11 meters) or more in length	Inflatable buoyant apparatus.
Do	Less than 36 feet (11 meters) in length	None.
Inside Boundary Line, warm waters; or Lakes, bays, sounds, warm waters; or Rivers, warm waters.	All	None.
Great Lakes, cold waters	36 feet (11 meters) or more in length	Inflatable buoyant apparatus.
Do	Less than 36 feet (11 meters) in length	Buoyant apparatus.
Great Lakes, beyond 3 miles of coastline, warm waters	All	Buoyant apparatus.
Great Lakes, within 3 miles of coastline, warm waters	All	None.

TABLE 28.120(b).—SURVIVAL CRAFT FOR UNDOCUMENTED VESSELS WITH NOT MORE THAN 16 INDIVIDUALS ON BOARD

Area	Vessel type	Survival craft required
Beyond 20 miles of coastline	All	Inflatable buoyant apparatus.
Beyond Boundary Line, within 20 miles of coastline, cold waters	All	Inflatable buoyant apparatus.
Beyond Boundary Line, within 20 miles of coastline, warm waters.	All	Life float.
Inside Boundary Line, cold waters; or Lakes, bays, sounds, cold waters; or Rivers, cold waters.	36 feet (11 meters) or more in length	Buoyant apparatus.
Do	Less than 36 feet (11 meters) in length	None.
Inside Boundary Line, warm waters; or Lakes, bays, sounds, warm waters; or Rivers, warm waters.	All	None.
Great Lakes, cold waters	All	Buoyant apparatus.
Great Lakes, beyond 3 miles of coastline, warm waters	All	Buoyant apparatus.
Great Lakes, within 3 miles of coastline, warm waters	All	None.

TABLE 28.120(c).—SURVIVAL CRAFT FOR UNDOCUMENTED VESSELS WITH MORE THAN 16 INDIVIDUALS ON BOARD

Area	Vessel type	Survival craft required
Beyond 50 miles of coastline	All	Inflatable liferaft with SOLAS A pack.
Between 20-50 miles of coastline, cold waters	All	Inflatable liferaft with SOLAS B pack.
Between 20-50 miles of coastline, warm waters	All	Inflatable liferaft.
Beyond Boundary Line, within 20 miles of coastline, cold water	All	Inflatable liferaft.
Beyond Boundary Line within 20 miles of coastline, warm waters.	All	Life float.
Inside Boundary Line, cold waters; or Lakes, bays, sounds, cold waters; or Rivers, cold waters.	36 feet 11 (meters) or more in length	Inflatable buoyant apparatus.
Do	Less than 36 feet (11 meters) in length	None.
Inside Boundary Line, warm waters; or Lakes, bays, sounds, warm waters; or Rivers, warm waters.	All	None.
Great Lakes, cold waters	36 feet (11 meters) or more in length	Inflatable buoyant apparatus.
Do	Less than 36 feet (11 meters) in length	Buoyant apparatus.
Great Lakes, beyond 3 miles of coastline, warm waters	All	Buoyant apparatus.
Great Lakes, within 3 miles of coastline, warm waters	All	None.

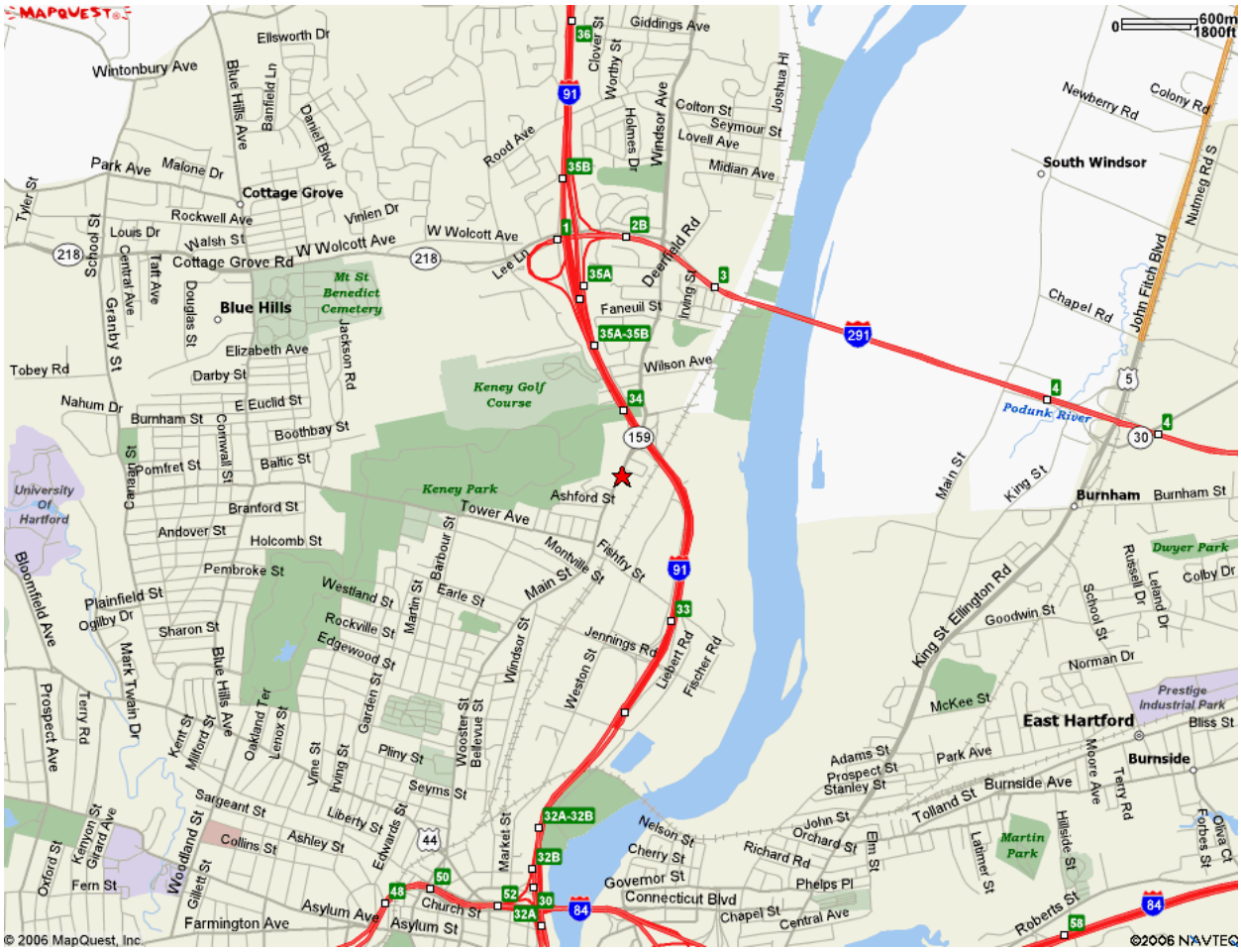

It is illegal for any vessel to dump plastic trash anywhere in the ocean or navigable waters of the United States. Annex V of the MARPOL Treaty is a new international law for a cleaner, safer marine environment. Each violation of these requirements may result in civil penalty up to $25,000, a fine of up to $50,000, and imprisonment up to 5 years.
U.S. lakes, rivers, bays, sounds and 3 miles from shore.
ILLEGAL TO DUMP
Plastic & garbage, paper, metal, rags, crockery, glass, dunnage, food.
3 to 12 miles
ILLEGAL TO DUMP
Plastic, dunnage (lining and packing materials that float), also if not ground to less than one inch: paper, rags, glass, crockery, metal, food.
12 to 25 miles
ILLEGAL TO DUMP
Plastic, dunnage (lining and packing materials that float).
Outside 25 miles
ILLEGAL TO DUMP
Plastic
State and local regulations may further restrict the disposal of garbage

ATTACHMENT B
Directions

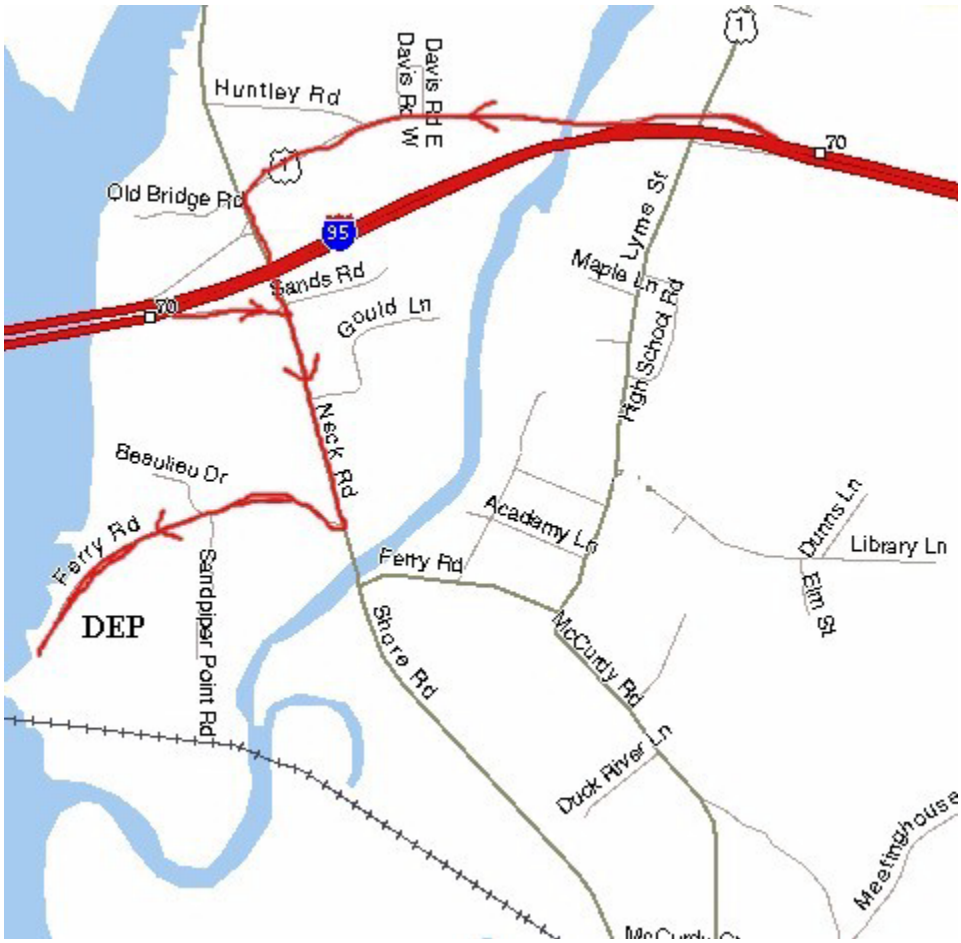
Directions to Windsor Water Survey Prep Lab
9 Windsor Ave, Windsor, CT

From 79 Elm St., Hartford

Take Exit for Route 91 North.
Take Exit 34 Route 159 Windsor. At the bottom of the Exit turn Left.
Turn Left at the stop light. Pass under the highway, pass the Citgo gas station. The Windsor Lab is the next driveway on the right.



Directions to Old Lyme, DEP Marine Fisheries
333 Ferry Road, Old Lyme, CT



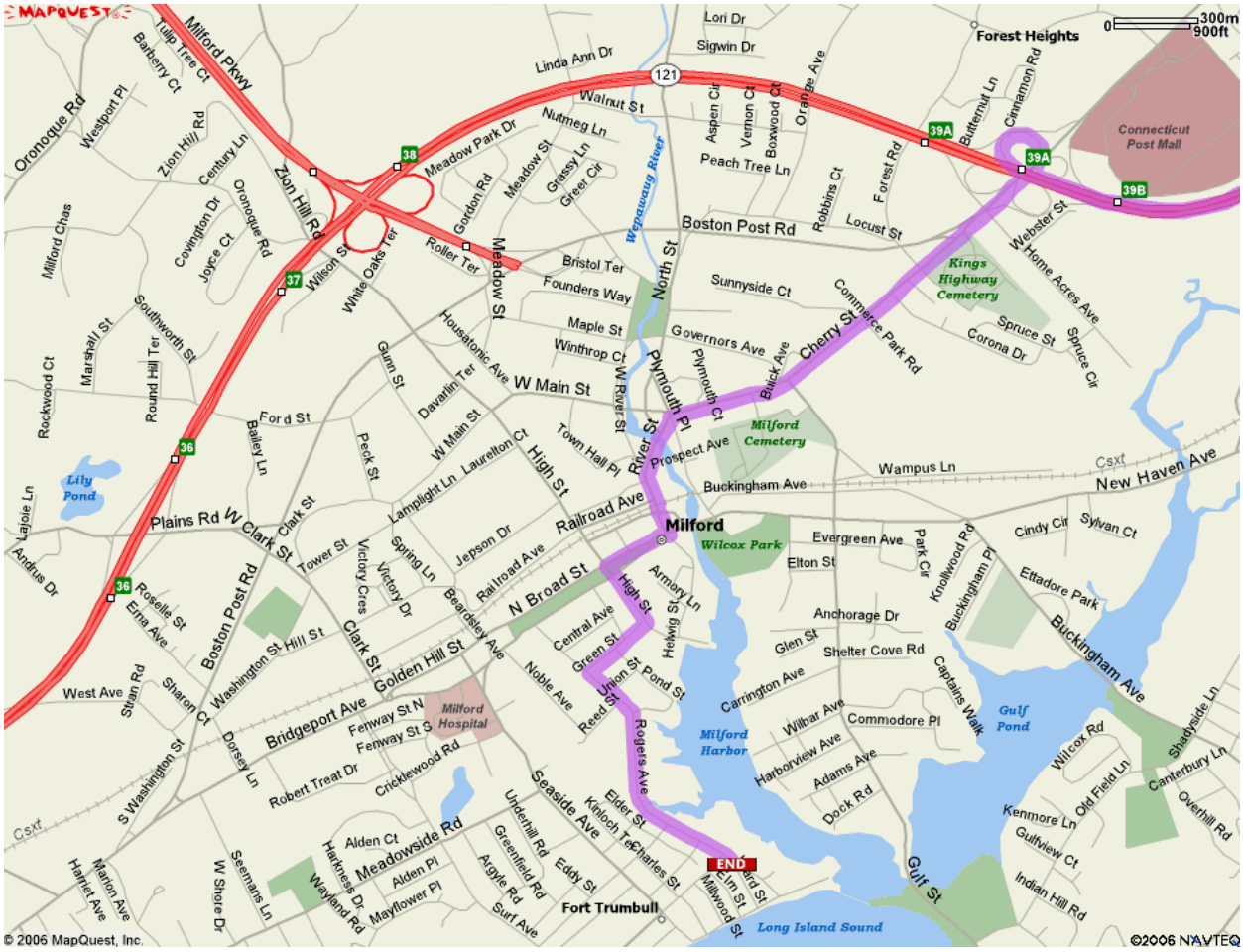
From Hartford/Windsor take Route 91 South to Route 9 South to Route 95 North.

From I-95 North take exit 70, take right at bottom of ramp, take first right onto Ferry Road, Marine Headquarters is on Left at end of road.

From I-95 South take exit 70, go straight at bottom of ramp. Go to end of road and take a Left, you will go back under I-95. After you go under I-95 take first right onto Ferry Road, Marine Headquarters is on Left at end of road.

Directions to Dept. of Agriculture, Bureau of Aquaculture
190 Rogers Ave, Milford, via Route 95

Take Exit 39A- Route 1 South off Route 95 South. At the bottom of the ramp, turn right and then merge left. Bear slightly left/stay straight to travel on Cherry Street. Bear slight Left onto River Street. In the center of town, turn Right onto North Broad Street. Turn Left in the center of the green onto High Street. Go straight through the light. Take next Right onto Green Street. At the end turn Left onto Rogers Ave.


MAPQUEST
0
300m
900ft
Milford Pkwy
Tulip Tree Ct
Barberry Ct
Oronoque Rd
Westport Pl
Zion Hill Rd
Century Ln
Covington Dr
Joyce Ct
Milford Chas
Linda Ann Dr
Lori Dr
Sigwin Dr
121
Walnut St
Nutmeg Ln
Meadow Park Dr
Meadow St
Grassy Ln
Greer Ct
Gordon Rd
Roller Ter
Wilson St
White Oaks Ter
Wepawaug River
Aspen Cir
Vernon Ct
Boxwood Ct
Orange Ave
Peach Tree Ln
Forest Heights
Butternut Ln
Cinnamon Rd
Connecticut Post Mall
39A
39B
Boston Post Rd
Robbins Ct
Forest Rd
Locust St
Webster St
Home Acres Ave
Kings Highway Cemetery
Spruce St
Corona Dr
Spruce Cir
Commerce Park Rd
Cherry St
Sunnyside Ct
Bristol Ter
Founders Way
North St
Maple St
Winthrop Ct
Governors Ave
Plymouth Pl
Plymouth Ct
Buick Ave
W River St
Housatonic Ave
Davarlin Ter
Glenn St
W Main St
Southworth St
Marshall St
Round Hill Ter
Rockwood Ct
Ford St
Bailey Ln
Peck St
Lamplight Ln
Laurelton Ct
Town Hall Pl
High St
River St
Prospect Ave
Milford Cemetery
Buckingham Ave
Wampus Ln
Csxt
New Haven Ave
Lily Pond
Lajoie Ln
Andrus Dr
Plains Rd
W Clark St
Clark St
Tower St
Victory Cres
Victory Dr
Spring Ln
Jepson Dr
Railroad Ave
Milford
Wilcox Park
Evergreen Ave
Elton St
Park Cir
Knollwood Rd
Buckingham Pl
Cindy Cir
Sylvan Ct
Ettadore Park
Buckingham Ave
Roselle St
Erna Ave
Boston Post Rd
Washington St
Hill St
N Broad St
Beardsley Ave
Central Ave
Green St
Union St
Pond St
Armory Ln
Hekwig St
Anchorage Dr
Glen St
Shelter Cove Rd
Captains Walk
Gulf Pond
West Ave
Stran Rd
Sharon Ct
Golden Hill St
Bridgeport Ave
Fenway St N
Fenway St S
Milford Hospital
Noble Ave
Reed St
Rogers Ave
Carrington Ave
Wilbar Ave
Commodore Pl
Milford Harbor
Harborview Ave
Adams Ave
Dock Rd
Wilcox Rd
Old Field Ln
Shadyside Ln
Canterbury Ln
Overhill Rd
Kenmore Ln
Gulfview Ct
Indian Hill Rd
Gulf St
Robert Treat Dr
Cricklewood Rd
Dorsey Ln
S Washington St
Seaside Ave
Elder St
Kinloch Ter
Charles St
Underhill Rd
Greenfield Rd
Eddy St
Argyle Rd
Surf Ave
Alden Ct
Alden Pl
Meadowside Rd
Mayflower Pl
Wayland Rd
Harkness Dr
Seemans Ln
W Shore Dr
Harriet Ave
Marion Ave
END
Fort Trumbull
Elm St
Millwood St
Long Island Sound
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©2006 NAVTEQ

Directions to CESE in Storrs

From Hartford to Building 4 Annex, 3107 Horsebarn Hill Road, Storrs.

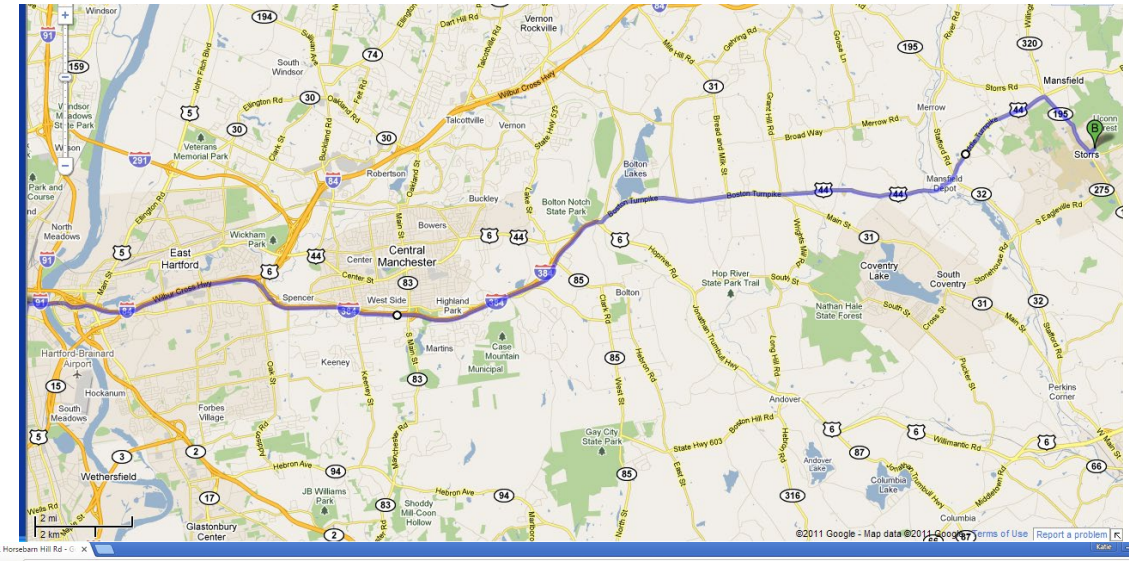
Take I-84 East.

Merge Onto I-384 East via Exit 59 Towards Providence.

I-384 East Becomes Rte 44 East. Route 44 becomes Middle Turnpike. Continue on Route 44.

Turn Right onto Route 195, Turn Left onto Horsebarn Hill Rd.

Alternately, continue on I-84 East to Exit 68, Route 195. Turn Right at the bottom of the exit onto Rte. 195, travel for 6.6 miles, and turn Left onto Horsebarn Hill Rd.



Attachment C
Field Data Sheet

Explanations/Codes
Station Observations The following information on the field data sheet is completed while the profiler is equilibrating (soaking).

Date- fill in the date in mm/dd/yy format
Time on/off station- military time- when you arrived at station and left
Field data recorder- initials of person filling out data sheet
Station Latitude-/Longitude – record lat/long from boat GPS (or computer)
Air temperature- in ºC; obtain from the digital thermometer located on the Dempsey to the left of the desk in the onboard lab

Profiler #- indicate which is in use
YSI#1= SN #1313
YSI#2= SN #1314

Profiling Method- indicate how profiles were collected
R= Rosette
S= Side Winch

Surface Bottle Method- indicate how the surface bottles were collected
R= Rosette
M= Manual

Tide Stage: *This is filled in upon return to the office following a standard procedure. See the Data Management SOP for directions.*
1= Ebb
2= Ebb Slack= Low Slack= water level below mean and velocity near zero
3= Flood
4= Flood Slack= High Slack= water level above mean and velocity near zero

% Cloud cover- % cloud cover is determined by walking out on the stern of the vessel, forward of the net reel and looking up. Stand with your arms out at the shoulders, perpendicular to your body (form a "T"). Estimate the percent of clouds covering the 180º arc from one hand to the other. Record this on the field sheet. This information is very subjective. New staff should be trained by experienced staff and perform side-by-side comparisons until estimates agree within 10%.

Current Weather: Fill in the code that corresponds to the current weather while on station

00= Clear, no clouds at any level
01= Partly cloudy, scattered or broken
02= Continuous layers of clouds
03= Sandstorm, duststorm, blowing snow
04= Fog, thick dust, haze
05= Drizzle
06= Rain
07= Snow or mixed precipitation
08= Showers
09= Thunderstorms

Sea State: Fill in the code that is closest to the sate of the sea while on station

0= Calm-Glassy (0 meters)
1= Calm- Rippled (0-0.1 meters)
2= Smooth- Wavelet (0.1-0.5 meters; 0.33 -1.65 feet)
3= Slight (0.5-1.25 meters; 1.65 - 4.125 feet)
4= Moderate (1.25-2.5 meters; 4.125-8+feet)

Secchi Depth- Fill in the Secchi depth measurement in meters

Upcast Raw Data- These data are recorded as the profiler is being brought to the surface- the upcast. Grab samples are collected at the bottom (defined as 5 meters up from the bottom) and surface (defined as 2 meters below the surface of the water). Depending upon the survey (regular monthly or hypoxia), additional samples are taken at near bottom depths (1 meter above the bottom) and mid-depths (determined by the maximum depth of the station roughly divided in half). After the on-deck command unit is triggered to collect the grab samples, write down the in situ data on the field sheet.

Sample depth
Depth code (NB= near bottom, B= bottom, M= mid, S= surface)
Time sample collected (the grab sample)
Water temperature
Salinity
Dissolved Oxygen
pH

EXO2 Profile Number- Fill in the file number associated with the profiler deployment

WATER QUALITY SURVEY

Connecticut Department of Environmental Protection
Bureau of Water Protection and Land Reuse, Planning and Standards Division
Long Island Sound Ambient Water Quality Monitoring Program

Station Name

Field Data Sheet

Date (MMDDYY) | Time ON Station | Time Off Station | Field Data Recorder

Station Latitude: 4 ____ . ____ N

Station Longitude: 7 ____ . ____ W

+/- Air Temp. (C): + ____ . ____

Profiler | Profiling Method | Surface Bottle Method | Tide Stage | % Cloud Cover | Current Weather | Sea State

Station Depth (m) ____ . ____ PROFILER READING

Station Depth (m) ____ . ____ DEPTH FINDER

Raw PAR @ surface ____ / Constant 1.62 = Incident PAR

UPCAST RAW DATA - DO NOT USE IN DATA ANALYSIS

Sample Depth (m)	Depth Code	Time Sample Collected	Water Temperature (C)	Salinity (PSU)	CTD Dissolved Oxygen (mg/l)	pH

Initials

Depth Code	Winkler Bottle #'s	Winkler Titrations				Winkler Dissolved Oxygen (mg/l)

Secchi Depth (m)

First ____ . ____ m

Second ____ . ____ m

Third ____ . ____ m

EXO2 Profile Number

Notes (any changes to normal sampling methods, changes, etc.):

Cast #

ATTACHMENT D
Chain of Custody Sheets

FIELD SAMPLES/DELIVERY RECORD

NUTRIENT ANALYSES

Sample Source:	LONG ISLAND SOUND
Sample Collector	M. Lyman
Project:	LISS
Job:	
SDG:	

FROM:

Matthew Lyman
Bureau of Water Management
CTDEP
79 Elm St.
Hartford, CT 06106-5127
(860) 424-3158
FAX 424-4055

Date of Collection:	Date of Delivery:
7/7/2005	7/8/2005

LIM Number	Sample Code	Volume Filtered (mL) PC/PN	Chl-a	BioSi	TSS/PP	Filter #s TSS/PP	Comments
		200	200	200	500		
	K2S	200	200	200	500	P0059 / P0058	
	K2B	200	200	200	500	P0057 / P0056	
	M3S	200	200	200	500	P0055 / P0054	
	M3B	200	200	200	500	P0051 / P0050	
	M3S DUP	200	200	200	500	P0053 / P0052	
	BLANK B	NA	NA	NA	NA	P0049 / P0048	
	J2S	200	200	200	500	P0047 / P0046	
	J2B	200	200	200	500	P0045 / P0044	
	I2S	200	200	200	500	P0043 / P0042	
	I2B	200	200	200	500	P0041 / P0040	
	I2M	200	200	200	500	P0039 / P0038	
	H2S	200	200	200	500	P0253 / P0251	
	H2B	200	200	200	500	P0250 / P0249	
	H4S	200	200	200	500	P0257 / P0256	
	H4B	200	200	200	500	P0255 / P0254	
	H6S	200	200	200	500	P0037 / P0036	
	H6B	200	200	200	500	P0259 / P0258	

Test Variables:

NH4, NO2+NO3, TDN, PN, DIP, TDP, PP
PC, DOC, TSS, SiO2, BioSi, Chl A

RELINQUISHED BY: (SIGNATUR	Date & Time	RECEIVED BY: (SIGNATURE)	Date & Time

FIELD SAMPLES/DELIVERY RECORD

BOD Series

Sample Source:	LONG ISLAND SOUND
Sample Collector:	M Lyman
Project:	LISS
Job:	
SDG:	

FROM:

Matthew Lyman
Bureau of Water Management
CTDEP
79 Elm St.
Hartford, CT 06106-5127
(860) 424-3158
FAX 424-4055

Date of Collection:	Date of Delivery:
7/7/2005	7/8/2005

Lab Number	Sample Code	Comments
	M3 S	
	M3 B	
	J2 S	
	J2 B	
	I2 S	
	I2 B	
	H6 S	
	H6 B	

Test Variables:
BOD30 Series

Lab Number	Sample Code	Comments

Test Variables:
BOD05 Series

RELINQUISHED BY: (SIGNATURE)	Date & Time	RECEIVED BY: (SIGNATURE)	Date & Time

CT DEP FIELD SAMPLING SHEET
Long Island Sound Ambient Water Quality Monitoring Program
HPLC Phytopigment Project

Contact: Matt Lyman, (860) 424 3158, matthew.lyman@ct.gov OR
Katie O'Brien-Clayton (860) 424-3176 katie.obrien-clayton@ct.gov

Cruise Name:____WQSEP06_____

Note: Please keep samples frozen. Transfer samples to the Health Department's deep freezer as soon as possible.

*** Please remember to record the volume filtered.

Station	Date	Volume filtered	Notes
K2	8/29/06	200 mL	
Blank B	8/29/06		
I2	8/29/06		
J2	8/29/06		
F2	8/30/06		
H4	8/30/06		
A4	8/31/06		
B3	8/31/06		
A4S-DUP	8/31/06		
C1	8/31/06		
D3	8/31/06		

Additional notes:

CT Department of Environmental Protection
Sample Summary Sheet
HPLC Phytopigment Project

Project contact:
Christine Olsen
Phone / fax: (860) 424-3727 / 4055
Christine.olsen@ct.gov
79 Elm Street, Hartford, CT 06106

Samples to be sent to:
Meg Maddox
Horn Point Laboratory
UMCES
2020 Horns Point Rd
Cambridge, MD 21613
(410)-221-8375

Date and time samples sent:
Number of samples included:
Number of sample field sheets included:
Samples were taken from cruises: WQJAN11, WQFEB11, CHFEB11, WQMAR11, CHMAR11, WQAPR11, WQMAY11

Horn Point Lab Notes:
Samples received by: _____
Date and time samples received: _____
Condition of samples upon receipt:_____
Any dry ice left in the package: _____
Other notes:

** Please return this sheet to CT DEP with results.

FIELD SAMPLES/DELIVERY RECORD
LONG ISLAND SOUND MicroZOOPLANKTON

TO:	FROM:
Laboratory of Dr. George McManus UCONN Dept of Marine Sciences 1080 Shennecossett Road Groton, CT 06340 (860) 405-9164	Matthew Lyman CTDEP Bureau of Water Management Long Island Sound Monitoring 79 Elm St. Hartford, CT 06106-5127 (860) 424-3158 (FAX 860-424-4055) e-mail: matthew.lyman@po.state.ct.us

Date of Delivery:

Date of Collection	Sample Code		Concentrated Volume (liters)	Comments
	K2	**-W**		
	K2	**-64**	**10**	
	I2	**-W**		
	I2	**-64**	**10**	
	F2	**-W**		
	F2	**-64**	**10**	
	H4	**-W**		
	H4	**-64**	**10**	
	D3	**-W**		
	D3	**-64**	**10**	
	B3	**-W**		
	B3	**-64**	**10**	
		-W		
		-64		
		-W		
		-64		
		-W		
		-64		

-W = **W**hole water (composite) sample (250 ml); w/Lugol's

-64 = sample from concentrating *x* -liters of whole water composite with 64 *u* m sieve; w/formaldehyde (target volume 10-liters)

RELINQUISHED BY: (SIGNATURE)	Date & Time	RECEIVED BY: (SIGNATURE)	Date & Time

FIELD SAMPLES/DELIVERY RECORD
LONG ISLAND SOUND MesoZOOPLANKTON

TO:	FROM:
Laboratory of Dr. Hans Dam UCONN Dept of Marine Sciences 1080 Shennecossett Road Groton, CT 06340 (860) 405-9164	Matthew Lyman CTDEP Bureau of Water Management Long Island Sound Monitoring 79 Elm St. Hartford, CT 06106-5127 (860) 424-3158 (FAX 860-424-4055) e-mail: matthew.lyman@po.state.ct.us

Date of Delivery:

ALL SAMPLES CONTAIN FORMALIN (~10%)

Date of Collection	Sample Code		Volume Sampled	Comments
	K2	**-A**		
	K2	**-B**		
	I2	**-A**		
	I2	**-B**		
	F2	**-A**		
	F2	**-B**		
	H4	**-A**		
	H4	**-B**		
	D3	**-A**		
	D3	**-B**		
	B3	**-A**		
	B3	**-B**		
		-A		
		-B		
		-A		
		-B		
		-A		
		-B		

-A = Zooplankton sample from Net A **-B** = Zooplankton sample from Net B

RELINQUISHED BY: (SIGNATURE)	Date & Time	RECEIVED BY: (SIGNATURE)	Date & Time

CT DEP FIELD SAMPLING/C-O-C SHEET
Long Island Sound Ambient Water Quality Monitoring Program
Phytoplankton Identification Project

Contact: Matt Lyman at (860) 424 3158
Or Katie O'Brien-Clayton at (860) 424 3176

Samples to:

Dr. Senjie Lin
UCONN Marine Sciences
Avery Point
(860) 405-9168

Cruise Name:_____ Date of Sample Delivery:_____

Samples included (all Surface water samples unless otherwise indicated):

Station	Date Sampled	Lugol Yes	Lugol No	Surface Yes/No	Bottom Yes/No
A4					
B3					
C1					
D3					
E1					
F2					
H4					
I2					
J2					
K2					

Notes:

Relinquished by:_____ Date/Time:_____

ATTACHMENT E

Table 1. Sampling Matrix for the Long Island Sound Water Quality Monitoring Program

	Station	Approximate Maximum Depth (m)	Latitude	Longitude	Profile	BOD	QA/QC	Parameters									
								TSS/PP	Dissolved nutrients	BioSi	Dissolved silica	PC/PN	Chl a	HPLC	Phyto-plankton	Plankton Tow	Composite Plankton
Cruise Day 1- East	K2	35-38	41.23433333	-72.26583333	X			X	X	X	X	X	X	X	X	X	X
	M3	40-42*	41.23716667	-72.05333333	X to 40 m	X	surface duplicate	X	X	X	X	X	X				
	J2	12- 30	41.182	-72.457666670	X	X		X	X	X	X	X	X	X	X		
	I2	26-28	41.1375	-72.655	X	X		X	X	X	X	X	X	X		X	X
	H6	37-42	41.026	-72.9135	X	X		X	X	X	X	X	X				
	H4	22-25	41.10166667	-72.934	X			X	X	X	X	X	X	X	X	X	X
	H2	13-15	41.178	-72.9605	X			X	X	X	X	X	X				
Cruise Day 2- West	9	10	41.07083333	-73.33616667	X			X	X	X	X	X	X				
	D3	35-44	40.99383333	-73.41133333	X	X		X	X	X	X	X	X	X	X	X	X
	C2	23	40.98433333	-73.50216667	X			X	X	X	X	X	X				
	C1	18-21	40.95583333	-73.58033333	X	X		X	X	X	X	X	X	X	X		
	B3	18-23	40.91833333	-73.64283333	X	X	surface BOD duplicate	X	X	X	X	X	X	X	X	X	X
	A4	34.2	40.8725	-73.73416667	X	X	surface duplicate	X	X	X	X	X	X	X	X		
Cruise Day-3	F2	18-21	41.08033333	-73.16533333	X			X	X	X	X	X	X	X	X	X	X
	F3	38-42	41.01783333	-73.1445	X	X		X	X	X	X	X	X				
	15	14-17	40.93133333	-73.22116667	X			X	X	X	X	X	X				
	E1	36-40	41.01933333	-73.29133333	X	X	surface duplicate	X	X	X	X	X	X	X	X		

* M3 is sampled between 40 and 42 meters; station depths range up to 80m

Table 2. Depth Interval and number of sample bottles to be collected during LISWQMP surveys

	Station	Bottom- up 5 m, unless otherwise noted, always collected	"Depth Interval" - intermediate samples taken from bottom depth to 2 meter depth at X meter intervals, dependent on tide stage, for plankton analysis. Depending upon the survey, samples may be collected from mid-water depths and processed for nutrients (e.g., NCA)				Surface- 2 meters, always collected	Notes
Cruise Day 1- East	K2	2 bottles	up 4m- 1 bottle	up 4	up 4	up 4	2 bottles	
	M3	2 bottles					3 bottles	
	J2	2 bottles					2 bottles	
	I2	3 bottles	up 4m- 1 bottle	up 4	up 4	up 4	3 bottles	
	H6	2 bottles					2 bottles	
	H4	2 bottles	up 4m- 1 bottle	up 4	up 4	up 4	2 bottles	
	H2	1 bottle					1 bottle	
Cruise Day 2- West	9	up 3 meters, 1 bottle					1 bottle	
	D3	3 bottles	Up 6 m – 1 bottle*	Up 6 – 1 bottle	Up 6 – 1 bottle	Up 6 – 1 bottle	3 bottles*	*will require 2nd cast due to station depth; 2nd cast bottles are collected on down cast, stopping at 6 m intervals starting at the 2 m depth; 1 bottle at each depth
	C2	1 bottle					1 bottle	
	C1	2 bottles					2 bottles	
	B3	2 bottles	Up 4m – 1 bottle*	up 4*	up 4*	up 4*	2 bottles	* may require 2nd cast; if so, collect on down cast, stopping at 4 m intervals, starting at the 2 m depth; 1 bottle at each depth
	A4	2 bottles					3 bottles	
Cruise Day- 3 Central	F2	2 bottles	up 4m- 1 bottle	up 4	up 4		2 bottles	
	F3	2 bottles					2 bottles	
	15	1 bottle					1 bottle	
	E1	2 bottle					3 bottles	