

**LONG ISLAND SOUND
AMBIENT WATER QUALITY MONITORING
PROGRAM**

**WATER QUALITY AND HYDROGRAPHIC SURVEYS
STANDARD OPERATING PROCEDURES MANUAL**

Revision
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STATE OF CONNECTICUT
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REVISIONS PAGE

Date	Review Number	Summary of Changes	Applicable Section
4/28/2011	2	Addition of pH calibration procedures	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 photosynthetically-active radiation (PAR) are collected. 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. One such ongoing project, funded through the Long Island Sound Study research grant program (2004), seeks to “quantify the impact of anthropogenic nitrogen loading to Long Island Sound with respect to natural resources” (LISS undated). The principle investigators are Dr. Mark Altabet from the University of Massachusetts- Dartmouth, School of Marine Science and Technology and Dr. Johan Varekamp from Wesleyan University. LISWQMP collects water samples for the project, fills pre-cleaned and acidified sample bottles provided by the project, and ships the bottles back to UMass- Dartmouth for analysis. Data collection for these special projects is performed following standard operating procedures specified by the principal investigator and are not included in this manual.

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.

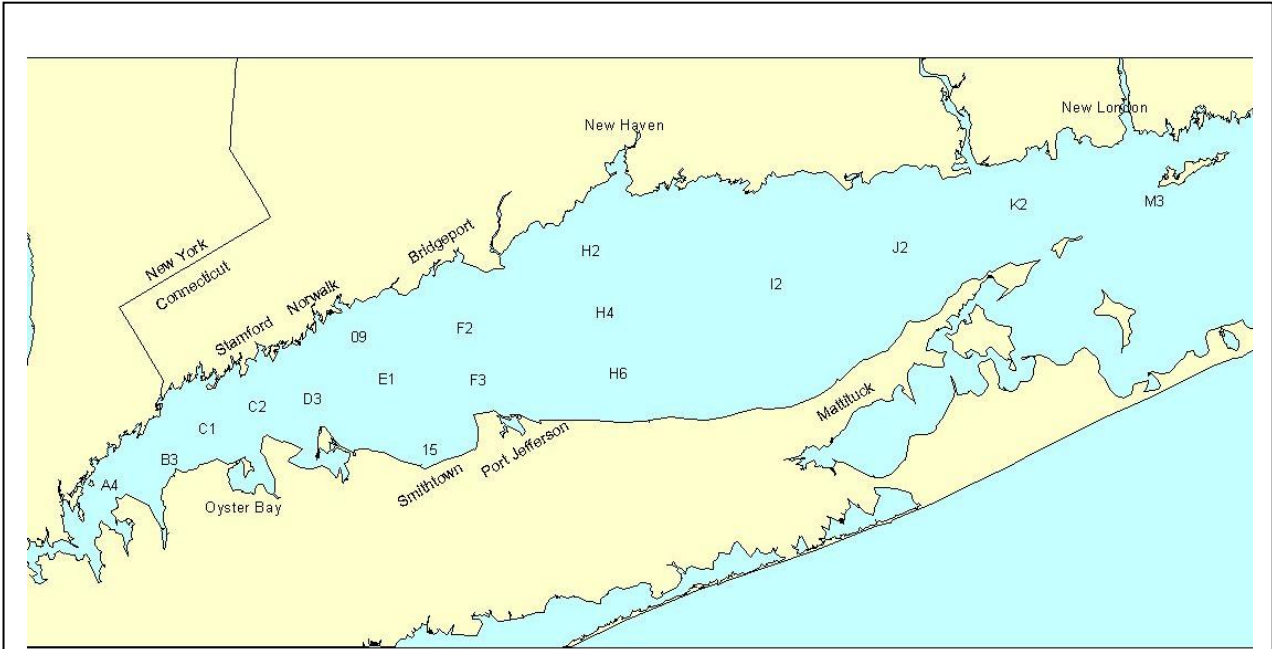


Figure 1. CT DEEP monthly water quality monitoring stations in Long Island Sound

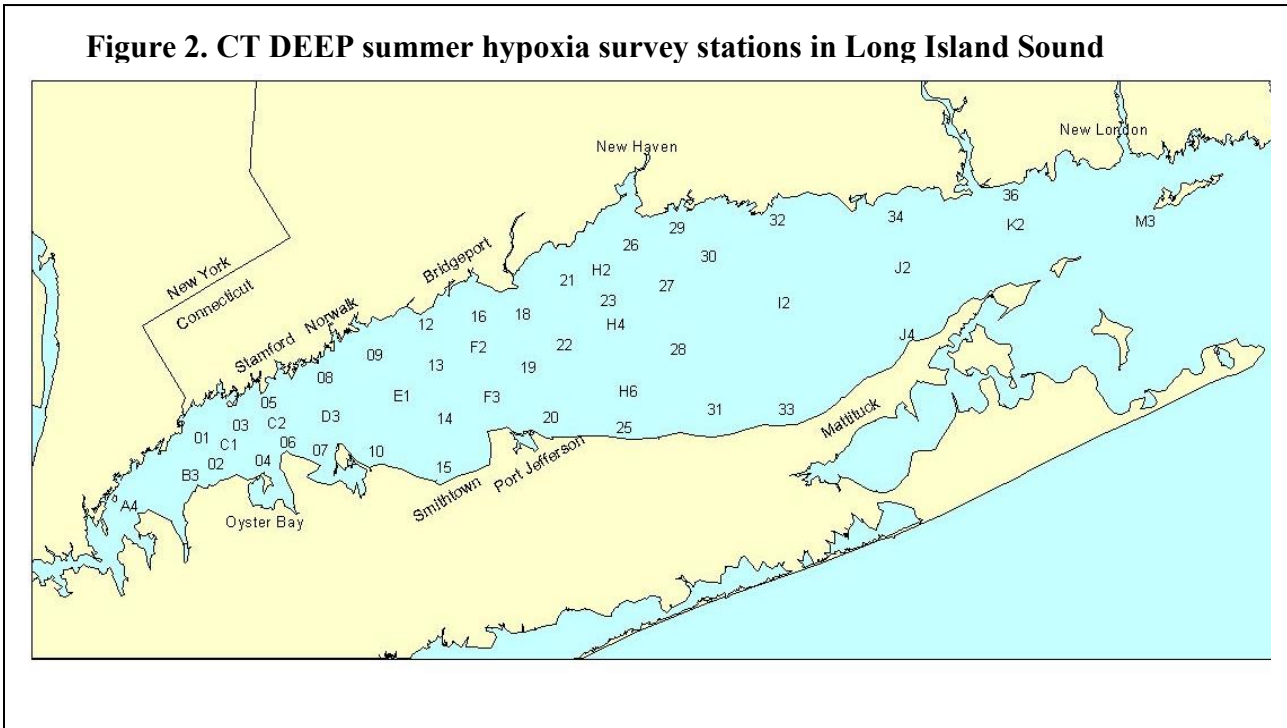


Figure 2. CT DEEP summer hypoxia survey stations in Long Island Sound

General Overview of Methods

State of Connecticut bond authorizations earmarked funds for Long Island Sound water quality monitoring to equip the 50 foot CT DEEP research vessel, the R/V John Dempsey (Figure 3), with a state-of-the-art water sampling and monitoring system. This system includes a conductivity-temperature-depth (CTD) water column profiling unit (Sea-Bird model SBE-19 SeaCat Profiler) equipped with dissolved oxygen, pH and PAR sensors as well as an in line fluorometer. This unit has an internal memory, and is capable of creating and storing data files on depth, temperature, salinity, dissolved oxygen, pH, chlorophyll and PAR at a rate of twice per second as the unit is lowered through the water column. These data can be reviewed in real-time (i.e., as the cast is taking place) via the onboard computer or can be uploaded onto the computer after cast completion.



Figure 3. R/V John Dempsey

As technology advances and equipment becomes obsolete and in order 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. In 2016, CT DEEP acquired two YSI EXO2 series multi-parameter water quality sondes. Side-by-side testing will occur in 2017 to determine data comparability with the existing platform and develop Standard Operating Procedures for the calibration and use of the EXOs.

Generally the CTD 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 CTD 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.

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, titrations, 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. CTD 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. 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.

Raw CTD data shall be reviewed by experienced Program staff. The downcast will be reviewed for significant outliers (spikes) and functional problems such as system clogging. Acceptable casts will be averaged into 0.2 meter bins and corrected with the use of a regression. Processed CTD files will be uploaded into the Program database.

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.

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 phosphorus

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	Hartford			
Field data sheets (17 for monthly surveys, 40 for hypoxia surveys)	Hartford			
Chain-of-custody forms (3 BOD COCs per survey, 3 route specific COCs per survey)	Hartford			
Map/site visit plan	Hartford			
Field writing implements- Permanent marker for labeling, Rite-in-rain pen, pencil, etc.	Hartford			
CTD (Sea-Bird model SBE-19 SeaCat Profiler with auxiliary DO, PAR, pH, and Fluorometer sensors) or YSI EXO sonde	Windsor			
Laptop computer for CTD 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			
Kimwipes	Windsor			
CRC Marine Formula 6-66	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 (Hoeffler 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 µM)	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 CTD profiler, Niskin water sampling bottles, and a rosette multi-bottle array which allows for the deployment of up to ten sampling bottles and the CTD at the same time. A laptop computer allows the CTD data to be viewed in real-time. The rosette, CTD, and Niskin water sampling bottle array is shown in Figure 4 to the right.

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.



Figure 4. Rosette, CTD, and Niskin sampling array

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

<u>Standard Operating Procedure For The Calibration Of The Seabird Seacat Profiler (SBE-19) Dissolved Oxygen Sensor</u>	16
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STANDARD OPERATING PROCEDURE FOR THE CALIBRATION OF THE SEABIRD SEACAT PROFILER (SBE-19) DISSOLVED OXYGEN SENSOR

In August 2010, CT DEP upgraded the dissolved oxygen sensor on its CTDs to the SeaBird SBE 43 Dissolved Oxygen Sensor, a polarographic membrane sensor. This sensor requires calibration less frequently, annually by the manufacturer. SeaBird states that electrochemical drift exists below the calibration certainty of 1 $\mu\text{mol/kg}$ and has not been observed in years of factory calibration data nor in long deployments. Therefore any drift is attributed to fouling of the membrane. SeaBird recommends post survey validation whereby the DO and drift are checked against replicate water collections and Winkler Titrations. See *Standard Operating Procedure for Determining Dissolved Oxygen Content of Seawater Using the Azide-Winkler Method* for instructions on performing Winkler titrations. The calibration coefficients SOC and BOC can be adjusted to the Winkler results following procedures outlined later in this document. If the adjustment is greater than 20% the unit should be returned to SeaBird for recalibration.

Selected text adapted from:

Sea Bird Electronics Inc. Application Note 64-2 Revised June 19, 2012 SBE 43 Dissolved Oxygen Sensor Calibration and Data Corrections

Summary

The CTD dissolved oxygen sensor is calibrated at least 24 hours prior to each survey at the CT DEEP laboratory in Windsor. Additionally, the CTD and DO sensor are returned to Sea-Bird Electronics for manufacturer calibration annually. This SOP outlines the steps necessary for successful calibration.

Equipment/Apparatus

- ❖ SeaBird Seacat Profiler equipped with SBE 43 Dissolved Oxygen Sensor
- ❖ Deionized Water
- ❖ Calibration Tank (100 gallons) Filled with fresh water
- ❖ Aquarium Pump, airline tubing, air stone
- ❖ Siphon hose
- ❖ Laptop computer equipped with SeaSave and Term19 (MS Dos) software
- ❖ Electro-mechanical cable
- ❖ Calibration notebook
- ❖ Writing implement
- ❖ Sodium sulfite solution
- ❖ 2 Winkler bottles
- ❖ Winkler reagents (see Winkler SOP)

Procedure

1. During the winter when water temperatures are colder, drain and refill the calibration tank with fresh water from the tap. Otherwise, place air stone into tank approximately six inches from the surface (greater depth will tend to supersaturate the water because the air will be injected at a pressure higher than atmospheric pressure). Plug in aquarium pump.
2. Connect CTD to computer with the cable. Be careful not to bend the pins.
3. Remove plastic tubing protecting the conductivity cell. Remove short section of tubing on top of the “Y” connector at the top of the Tygon tubing.
4. Put CTD cage and all into the calibration tank and let soak for at least one hour with the power **OFF**.
5. Gather additional items needed to calibrate CTD.
 - CTD calibration Notebook,
 - Sodium Sulfite,
 - Sodium Thiosulfate,
 - Manganous Sulfate,
 - Alkali-iodide-azide Reagent,
 - Starch Solution,
 - Sulfuric Acid,
 - pH buffers- 7, 10, 4
 - Winkler titration bottles,
 - rubber hose,
 - digital buret,
 - Erlenmeyer flasks
 - Automatic Pipetter
 - Pipette tips
 - Shower test bucket
6. Following the one hour soak time, turn on the laptop and log in following the prescribed security procedures.
7. Double click on the Term19 icon.
 - a. Press <F6> to wake up the CTD and establish communication.
 - b. Type DS (Display Status) at the S> prompt to check to make sure the battery is sufficiently charged (Vmain should be >10 for calibration). Also note the conductivity value and pump delay.
 - c. Next, at the S> prompt, type SP (Set Pump) to change the minimum conductivity value.
 - i. Set the Minimum Conductivity for Pump Turn On to zero (this insures that the pump will come on in the fresh water) but do not change the pump delay (should be 45 seconds). You must type 45 seconds and hit enter (Do not just hit enter because a value of 0 will be saved). To double check if the pump settings are correct, hit DS at the S> prompt and be sure that minimum conductivity is 0 and pump delay is 45.

To ensure data are not lost from the previous survey, Press <F9> to upload data stored on the CTD to a temporary file. You will be prompted to enter a file name. Then hit Return. When the screen appears prompting you to enter Header information hit escape. Once completed, open the temporary file and confirm all have been uploaded.

Once all the data have been uploaded, Press <F8> to Initialize the Log. At the prompt asking “Do You Really Want to Initialize Logging?” select Yes. The message “Profiler is ready for deployment” should appear and TERM19 should close out, if it doesn’t Push F10 to exit. Select Yes.

8. Double click on the Seasave7 icon.
 - a. Click on RealtimeData on the Menubar.
 - b. Select Start Acquisition.
 - c. Click on Select[.CON]File.
 - d. It should default to the previous survey’s CON file; however, be sure it is the one you desire. If not, navigate to the correct file. Select it and Click Open.
 - e. Click on Enter Output Data File Name.
 - f. Navigate to the Desktop. Create a new folder and enter the survey name (e.g., WQJAN17). Click Open.
 - g. Type first for file name. Click OK.
 - h. Click Start Acquisition.
 - i. In the Header Information Window, enter first as the station name and the time under Latitude (or Time).
 - j. Click OK.
 - k. A pop up window will appear that says “Turn on the SBE19 using the magnetic switch.” And it will count down for 60 seconds. After about 15 seconds have elapsed off the clock, turn the CTD on using the tool fashioned for this purpose (metal wire attachment on the pipe).
9. Let CTD run for at least 500 scans (readings should be stable). After the readings have stabilized record real-time temperature and DO data displayed on screen into calibration notebook.
10. In the meantime, place two BOD bottles in the shower collection bucket. Place the stopper end of rubber hose into the tank (the stopper adds weight). Pinch the tubing about 6 inches from the end not in the water. Place tubing in water up to where you pinched it. Draw out quickly to start a siphon. With water flowing, place the end of the tube into the bottom of the BOD bottle. Allow the water to overflow until the bottom of the bucket is just covered with water (~20 seconds). Continue with second bottle. Begin to perform Winkler Titrations following the Standard Operating Procedure for Azide-Winkler Titrations attached herewith this manual.
11. If the CTD value is within +/- 0.2 mg/L of the average Winkler value, the calibration is complete. If the value is greater than +/- 0.2 mg/L proceed with the steps below to adjust the SOC coefficient. If the unit drifts greater than 20% it needs to be returned to Sea-Bird for recalibration.

SOC coefficient adjustment

SeaBird states that “any sensor drift with time is primarily attributed to fouling of the membrane, either biological or waterborne contaminants (i.e., oil). Adverse fouling might take several weeks to months to occur. Fouling of the membrane eventually causes the sensor to read low of correct. When the SBE 43 sensor starts to show drift from fouling, the effect on the sensor calibration is linear, and therefore can be corrected by adjusting the linear slope term of the calibration equation (Soc term...)”.

Follow these simple steps to derive a correction factor to adjust Soc

1. Compute the correction ratio between the Reference value (Winkler) and corresponding SBE 43 sensor value.

$$\text{Ratio} = \text{Reference} / \text{SBE 43}$$

*Be sure the units for each method are consistent (i.e., ml/L; mg/L; $\mu\text{mol/kg}$).

2. Multiply the correction ratio by the previous Soc in the configuration (.con or .xmlcon) file or on the calibration sheet.

$$\text{NewSoc} = \text{PreviousSoc} * (\text{Reference} / \text{SBE 43})$$

3. Modify the .xmlcon file
 - a. Click on Configure Inputs on the Menu Bar.
 - b. Click the Modify Button.
 - c. Double click Oxygen, SBE 43
 - d. Update the calibration date with today's date in MM/DD/YY format
 - e. Click in the SOC box. Update with the newly calculated SOC.
 - f. Click in BOC box to ensure your changes are accepted in the SOC box.
 - g. Click OK.
 - h. Click Save As.
Navigate to the Desktop and your new survey folder (e.g., WQJAN17). Enter survey name as the new xmlcon file name. Click Save. Click Exit, Click OK.
4. Perform a check of the new SOC.
 - a. Click Real-time data. Click Select Output File name.
 - b. Enter POST as the file name. Click Save. Click Start. Enter post as the station name and input the time.
 - c. Turn on the CTD. Allow the CTD to run for 1500 scans.
 - d. Record the DO value in the field calibration notebook. The new DO value should be within +/- 0.2 mg/L of the average Winkler value.
5. Perform a QA oxygen check after calibration.
 - a. Click on RealTimeData, Select Start Acquisition.
 - b. Click on Select [.CON] File. Use the new .CON file you just saved. Click OK.

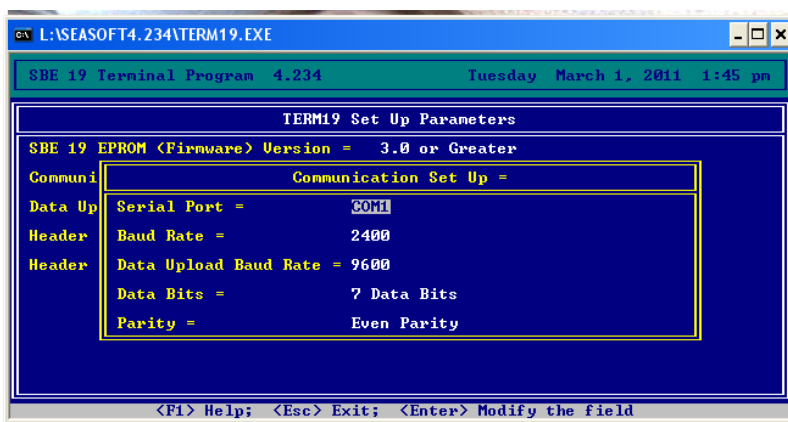
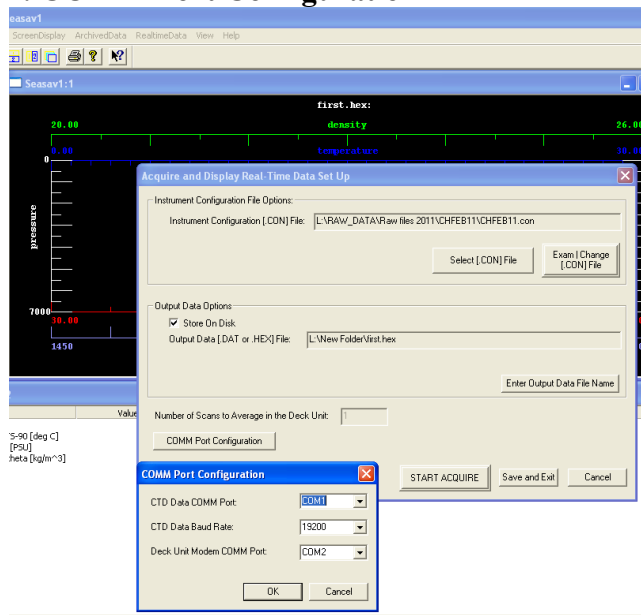
- c. Enter the output data file name as “post”.
 - d. Click Start Acquire, Turn the CTD on.
 - e. Let CTD run for at least 1500 scans; readings should be stable.
 - f. Record CTD oxygen measurement in the calibration notebook under the post heading.
6. Turn CTD off, remove from the tank, and set on the counter to dry. Attach the syringe filled with DI water over the conductivity cell and carefully dispense until water comes up through the plenum into the tubing. Draw back the water until the water level is not covering the membrane (want 100% humid environment, but do not want membrane to be submersed).
 7. Return to the laptop and double click on Term19. Press <F6> to wake up the CTD. Type “SP” at the DOS prompt. Change the pump turn on setting for conductivity back to 3270 Hz and be sure that the delay is set to 45 seconds. Press <F10> to exit.
 8. Disconnect the cable and reinstall the pin protectors and outlet tubing.
 9. Load CTD into vehicle to be brought to boat for survey or return to storage cabinet. The CTD must not be stored in the vehicle overnight- if you bring the vehicle home, bring the CTD inside. The CTD also must not be left in vehicles when temperatures exceed 75°F.

Problems and possible causes:

Oxygen value just keeps decreasing - Pump not operating (make sure you have changed the 'conductivity for pump turn-on' to zero in TERM19!) or plumbing not connected properly. Oxygen sensor consumes oxygen, so if a constant flow of water is not maintained across the membrane, the oxygen value will steadily decline.

No values appear in the SeaSav1:2 window – Check the COMM Port Configuration in SeaSave. Select RealTimeData from the menu bar, Select Start Acquisition. Click on COMM Port Configuration. The settings should be as shown below. Also check the Communication Set Up in TERM19. Double Click on Term19, Press <F2> for Setup. Arrow down until Communication Set Up = is highlighted. Hit Enter. Set the parameters to match those in Figure X below.

Figure 7. COMM Port Configuration



Routine (post-cruise) Cleaning (no visible deposits or marine growths on sensor) — Follow this two-step procedure:

1. **Flush** the sensor for **1 minute** with a **1%** solution of **Triton X-100** warmed to 30 °C (86 °F) using syringe and tubing. **Drain and flush** with warm (not hot) fresh water for **5 minutes**.
2. **Soak** the sensor for **1 minute** in a **500 - 1000 ppm** solution of **Bleach**. After the soak, **drain and flush** with warm (not hot) fresh water for **5 minutes**. **Dilute** concentrated household bleach to 50 to 1 (50 parts water to 1 part bleach) to produce the proper concentration to clean the oxygen sensor.

STANDARD OPERATING PROCEDURE FOR DETERMINING DISSOLVED OXYGEN CONTENT OF SEAWATER USING THE AZIDE-WINKLER METHOD

Summary

The Azide-Winkler Method is used to determine the dissolved oxygen content of the seawater and act as a quality control check of the CTD Dissolved Oxygen sensor. Winkler titrations are performed in the laboratory prior to each survey to determine if the DO sensor has drifted.

Explanation of the Winkler titration (Azide modification)

The Azide modification is used to eliminate the interference from Nitrite in the water. The reaction begins with the addition of two reagents Manganous Sulfate and Alkaline Potassium Iodide Azide. These reagents react to form a Floc of Manganous Hydroxide. The Oxygen in the water reacts with the Manganous Hydroxide oxidizing it to Manganic Hydroxide. This is a one to one relationship; for every molecule of Oxygen one molecule of Manganic Hydroxide is formed. Sulfuric acid is then added to convert the Manganic Hydroxide to Manganic Sulfate. At the same time Sulfate from the Manganic Sulfate reacts with the Alkaline Potassium Iodide Azide to produce Potassium Sulfate and free Iodine. Since the Sulfate for the reaction comes from the Manganic Sulfate the amount of Iodine released is directly proportional to the amount of Oxygen in the water. The Sodium Thiosulfate titrant reacts with the free Iodine to form Sodium Iodine, the solution turns clear when all the Iodine is converted to Sodium Iodine, each ml of titrant is equivalent to one mg of Dissolved Oxygen. The Starch indicator is added to enhance the end point.

Refer to *Standard Methods for the Examination of Water and Wastewater* Method # 4500-O Oxygen (Dissolved) for additional information.

Equipment/Apparatus

- ❖ Water Sample
 - ❖ 2 Glass BOD bottles with stoppers
 - ❖ Rubber tubing with stopper attached
 - ❖ Safety glasses
 - ❖ N-Dex gloves
 - ❖ Dispensettes or Automatic Pipettes
 - ❖ Manganous Sulfate Reagent
 - ❖ Alkaline Potassium Iodide Azide Reagent
 - ❖ Sulfuric acid
 - ❖ Sodium Thiosulfate
 - ❖ Starch Indicator Solution
 - ❖ Digital Buret
 - ❖ 250 mL graduated cylinder
 - ❖ 2 Erlenmeyer flask(s)
- Shower Testing Bucket

Safety Precautions

The chemicals used for dissolved oxygen determination can be dangerous. Safety glasses and gloves must be worn whenever handling these chemicals. See [Attachment C](#), which contains Material Safety Data Sheets, for additional health and safety information.

Procedure

- 1) Two BOD bottles are placed into the shower testing bucket with stoppers removed. Place the stopper end of the rubber tubing and the majority of the tubing into the calibration tank. Pinch the tube closed underwater. Start a siphon by quickly pulling part of the tubing out of the tank, release the tube, and placing it into the shower testing bucket. Once the water is flowing freely, quickly place the tubing into a BOD bottle, getting the outlet of the tubing all the way to the bottom of the bottle. Do this while the water is running. DO NOT pinch off tubing.

If the water splashes into the bottom of the bottle as the bottle fills air is being continuously added to the sample and the DO determination is useless. There will be a slight addition of air just as the bottle begins to fill, but as long as the tubing is kept at the bottom of the bottle it will not be too much, and should be forced out as the bottle is overflowed.

- 2) Let the bottle overflow two or three times its volume. Allowing the bottle to fill and overflow for thirty seconds will accomplish this. Alternatively, count while the bottle is filling - if it takes to a count of 20 for the bottle to fill, then count to twenty two to three more times and you will have allowed the bottle to overflow its volume by that many times.
- 3) Slowly pull the tube out of the bottle with the WATER RUNNING. Stopper the bottle carefully to exclude any air bubbles.
- 4) Leave the overflow that remains around the stopper - DO NOT tip bottle and pour this out. This overflow is important for keeping air out when the reagents are added.
- 5) Repeat for the second bottle.
- 6) Standard stock reagents are then added to the samples using either bottle top dispensettes or hand held automatic pipettes.

Add one milliliter (ml) of $MnSO_4$ (manganous sulfate -reagent #1).

Procedure for using hand held pipette:

- a) Check to see that the pipette is set to dispense 1 ml - the 1-ml line should be just visible at the top of the notch.
- b) Wipe outside of pipette tip off with a Kimwipe before filling with reagent.
- c) Push the plunger down to the first stop point, submerge pipette tip in reagent, and let plunger come up slowly. Releasing the plunger too rapidly will allow reagent to go up into the pipette, and this can cause clogging or other problems that reduce the accuracy of the pipette.
- d) Lift the sample BOD bottle stopper only when you are ready to dispense the reagent.
- e) Hold pipette tip just above the surface of the liquid when dispensing the reagents. DO NOT submerge the pipette tip into the sample. This avoids contamination of the pipette tip and so avoids contamination of the reagents.
- f) Push down on the plunger to dispense reagent into the bottles, and when plunger reaches the first stop, pull gently towards you so it will go all the way down, forcing the last drop out of the tip.

Procedure for using Dispensette bottle top dispenser:

- a. Remove the cap covering the tip of the dispenser.
 - b. Turn recycler valve to the dispensing position.
 - c. Unstopper the BOD sample bottle (one at a time) and place it beneath the dispensing tip.
 - d. To dispense, gently lift the plunger up until it stops, and then gently push all the way down. The dispensing tip should be within the bottle opening but not touching the liquid.
 - e. Recap tip.
- 7) Add 1 mL of alkali-iodide-azide reagent (reagent #2) to all bottles in the same manner as described in Step 6 above using either the dedicated dispensette or place a new clean tip on the hand held pipette.
- 8) Turn the tap water on in the sink. Carefully remove the sample bottle(s) from the counter. Place your forefinger on the stopper of the bottle to prevent leakage and entry of air into the sample. Rinse the top of each bottle under running water so that reagents in the puddle on top of the bottle do not go onto the counters or floor.
- 9) Keeping one finger on the stopper invert the bottles a few times to thoroughly mix the water and the reagents. A flocculent will form and the bottles should be mixed enough so that this flocculent is homogenous throughout the bottle. Return the bottle to the counter to allow the floc to settle.
- 10) When the manganese hydroxide floc has settled to half the bottle volume, leaving clear supernatant above, the third and final reagent, sulfuric acid, is added. Be very careful with the dispensing of the acid. Wear gloves and safety glasses.
- a) Remove the cap covering the tip of the dispenser.
 - b) Turn recycler valve to the dispensing position.
 - c) Unstopper the BOD sample bottle (one at a time) and place it beneath the dispensing tip.
 - d) To dispense, gently lift the plunger up until it stops, and then gently push all the way down. The dispensing tip should be within the bottle opening but not touching the liquid.
- Continue steps b, c, and d until all of the sample bottles have been acidified.
- e) Turn recycler valve back to recycle position and replace cap on the tip of the dispensing arm.
- 11) Thoroughly mix the bottles, being sure to keep a finger tightly on the stopper. Start the mixing by inverting the bottle over the sink, rinsing the top of the bottle under running water faucet to remove any acid from the bottle lip. Mix well by inverting bottle end to end. Invert until all signs of flocculent are gone. Visible brownish specks in the bottle indicate that the floc is not completely dissolved.

Titration

- 1) Unscrew the cap of the sodium thiosulfate solution. Assemble the digital buret by inserting the filler tube into the buret. Place the buret onto the sodium thiosulfate. To avoid air bubbles, it is helpful to have the tube reach close to the bottom of the reagent bottle. Carefully screw the digital buret onto the bottle. Set the buret to recycle mode. Turn on the buret. Turn the dispensing wheel towards you until you have “dispensed” around 100mL. Next carefully remove the display panel to reveal the mechanism. Turn the wheel a few more times watching as the liquid is drawn up into the tubes. Be certain that no air bubbles are visible in the exposed tubing or at the dispensing tip. Replace the panel. Turn the buret to dispense mode. Turn the wheel, allowing the chemical to fill the dispensing tube and discharge about 5 mL into the sink to be sure all air bubbles are removed from the dispensing tube. Clear the readout.
- 2) Pour approximately 15 mL into a 250 ml graduated cylinder (one with a hole drilled at about 200 ml). Swirl and swish the cylinder to rinse thoroughly. Dispose of the rinse water down the drain. Leave the water running for a few minutes to completely flush the sink pipes and dilute the waste. Repeat.
- 3) Measure out 201 ml into the graduated cylinder. Set the cylinder aside on the counter.
- 4) Pour the remaining sample from the BOD Bottle into the Erlenmeyer flask, swirl and rinse. Drain the rinse water into sink, flush. Place the BOD bottle and stopper into the sink for cleaning later.
- 5) Pour the 201 ml from the graduated cylinder into the flask. Place the flask beneath the buret. Repeat for the second BOD bottle. Place the flasks on the right side of the buret in the order to be titrated.
- 6) Double check that the buret is reading 0 mL.
- 7) Hold the flask in one hand. Slowly add titrant by turning the wheel while continuously swirling the flask until the solution turns straw colored (light yellow).
- 8) Add a small amount of starch indicator solution (~2 mL by squirting the bottle twice around the sides of the flask). This will turn the solution a blue-purple color.
- 9) Slowly continue to add titrant, swirling constantly. The solution will turn blue. As the solution gets light blue titrant must be added ONE DROP AT A TIME. Give each drop a chance to mix in fully before adding another. The endpoint is reached when the solution turns clear. The solution will change from a very pale blue-grey to clear. The drop that makes the difference should be obvious as the solution turns clear, check the color against a white surface such as a sheet of paper.
- 10) Record the digital readout of the buret in the Winkler section of the CTD calibration notebook. If you overshoot the endpoint write O.S. on the field sheet for that bottle. Repeat for the second BOD bottle.

[(If due to a spill, less than 201 ml of the sample are available, the titration can still be performed. Be sure to record on the data sheet the volume of fixed sample available to titrate. The dissolved oxygen (DO) is calculated using the following equation:

$$\text{mg O}_2/\text{liter} = \frac{\text{ml of titrant} \times 0.025 \times 8\text{mg/meq O}_2 \times 1000 \text{ ml/liter}}{\text{ml of sample}}$$

(0.025 = normality of sodium thiosulfate solution, the titrant)

NOTE: ml of sample in the above equation refers to original sample volume. The original sample volume is modified by the addition of reagents. Therefore, if 201 ml can not be used due to a spill, calculate ml of sample (x) using the following equation:

$$x = 298y/300$$

where y = the volume of fixed sample available to titrate; and
x = original sample volume after correction for sample loss by displacement with reagents (to be used in above equation)

Example: Only 150 ml of the fixed sample are available to titrate due to a spill.
 $x = (298)(150)/300 = 149$; ml of sample = 149.]

- 11) Rinse the BOD bottles and stoppers thoroughly with water and then with Deionized water. Place the bottles upside down in the wooden rack to dry.

STANDARD OPERATING PROCEDURE FOR CALIBRATION OF THE SEABIRD 18 pH SENSOR

Summary

In August 2010, CT DEEP upgraded its CTDs to include pH sensors. The SBE 18 pH sensor is an add-on auxiliary sensor for profiling CTDs. The sensor uses a pressure-balanced glass electrode/Ag/Ag-Cl reference pH probe to provide *in situ* measurements at depths up to 1200 meters. The pH sensor is returned to the manufacturer for annual calibration along with the CTD.

SeaBird software calculates pH as:

$$V_{out} = \text{offset} + [\text{slope} * (R * T / F) * \ln(10) * (\text{pH} - 7)]$$

Where

R = gas constant = 8.31434

F = Faraday constant = 9.64867×10^{-4}

T = temperature (°K)

V_{out} = output voltage from pH sensor (0 - 5 volts)

Substituting for R, F, and ln(10):

$$V_{out} = \text{offset} + [\text{slope} * 1.98416 \times 10^{-4} * T * (\text{pH} - 7)]$$

Therefore,

$$\text{pH} = 7 + (V_{out} - \text{offset}) / (1.98416 \times 10^{-4} \times T * \text{slope})$$

The following information was excerpted from SeaBird Application Notes No 18-1 SBE 18, 27, and 30, and AMT pH Sensor Calibration (PHFIT Version 2.0) and 18-2 pH Sensor Storage, Maintenance, and Calibration. Both notes were revised February 2010.

Equipment/Apparatus

- ❖ pH buffer solution 4, 7, 10
- ❖ Small plastic beaker (~50 mL)
- ❖ Phillips screwdriver
- ❖ Small gage calibration Wire
- ❖ Wash bottle filled with deionized water

When the pH Sensor is Not in Use

1. Replace the *soaker* bottle over the plastic pH electrode by removing the soaker bottle cap, sliding it along the plastic pH electrode as far as it will go, and threading the bottle up into the cap. There should be enough fluid in the bottle to cover at least the glass electrode and Teflon reference junction.
2. Remove the bottle by reversing the sequence.

When removing or installing the soaker bottle, do not force the pH electrode sideways. The electrode's outer shell is plastic, but the inner stem is glass and can break if the electrode is handled roughly.



Figure 8. pH probe

The *soaker* fluid is pH 4 buffer solution saturated with KCL. The pH 4 solution is acidic, and will eat away most fouling of the pH electrode. The sensor will tolerate the periodic absence of the soaker bottle and can be returned to initial performance by soaking for a few hours. However, **exposure of the bare sensor to temperature extremes (e.g., strong direct sunlight on a hot day) can cause a loss of internal electrolyte.** Subsequent cooling will draw air into the sensor, which will lead to pressure-related problems.

Note: The sensor contains a non-organic electrolyte and antibacterial inhibitors designed to optimize its use in marine environments.

Prior to each survey, recalibrate the pH Sensor

Sea-Bird pH sensors are calibrated with commercial buffer solutions (± 0.02 pH). Make periodic corrections by comparison to buffers near the anticipated in situ pH, typically in the 7 - 8 pH range. Best calibration of the sensor is obtained by soaking the sensor in deionized water for 30 minutes prior to standardization with buffers.

To calibrate:

Review the SeaBird pH calibration YouTube video <http://www.seabird.com/ph-sensor-calibration>. This procedure is slightly different from our SOP, but it is helpful and informative, just the same.

1. Run Seasave V7, set it up to display the pH voltage (the voltage channel for the pH data is volt 1), and start real-time data acquisition.
2. Connect a small-gauge wire to one of the screws at the connector end of the sensor housing and the other end will be put into the small beaker containing the buffer solutions during the calibration.
3. Rinse the beaker and pH probe with DI water. Discard. Next rinse both the probe and beaker with pH 4 buffer- Fill the beaker to about 10 mL with buffer solution. Place the beaker over the probe and gently swirl around. Discard the solution. Repeat. Again fill the beaker with pH buffer 4 solution, this time to about 25 mL. Put the pH probe in the buffer solution and wait 1 minute for complete stabilization. Record the resulting voltage on the computer display in the calibration notebook.
4. Repeat this process in order with pH 7 buffer and pH 10 buffer. **Rinse the beaker and pH electrode in deionized water between measurements in the different pH buffer solutions.**

Note: In our SEASOFT V2 suite of programs, edit the CTD configuration (.con) file using the Configure Inputs menu in Seasave V7 (real-time data acquisition software) or the Configure menu in SBE Data Processing (data processing software). Select pH as a voltage sensor when editing the configuration file; the software prompts for slope and offset.

Sea-Bird provides PHFIT software to use when calibrating their pH sensors. PHFIT is part of the SEASOFT-DOS software package; the latest version of the software is available for download from the SeaBird website.

5. If the probe reading varies from the standard by more than +/- 0.15 SU, Run PHFIT:
 - a) Double click on the PHFIT shortcut on the laptop desktop.
 - b) At the prompt, enter the sensor serial number and the temperature (in °C) of the buffer solutions (should be room temperature).
 - c) At the prompt, enter the pH of 4. Hit return. Enter the output voltage (Vout). Hit return. Repeat for 7 and 10. . Once you entered the last value hit enter again. When you have finished, the program outputs the offset and slope, along with the residuals.

6. Enter the new offset and slope in the CTD's configuration (.con or .xmlcon) file.
 - a. Click on SBEDataProc.exe.
 - b. In the Configure menu, select the applicable CTD.
 - c. In the dialog box, click Open and select the applicable .con file for the CTD.
 - d. In the sensor list, double click on the pH sensor.
 - e. Enter the new offset and slope in the dialog box and click OK.
 - f. Click Save As and enter the survey date to save the changed configuration (.con) file.

7. Run a QC Check using one of the pH standards to confirm the calibration was successful.

8. Rinse the pH probe with DI water. Replace the sensor soaker bottle. Clean up.

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

Summary

A SeaBird SeaCat SBE 19- CTD (Conductivity, Temperature, and Depth) Recorder equipped with a LyCor PAR sensor, SBE 18 pH sensor, Wet Labs fluorometer and SBE 43 dissolved oxygen sensor is used to obtain *in situ* water quality data from Long Island Sound. The CTD 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.

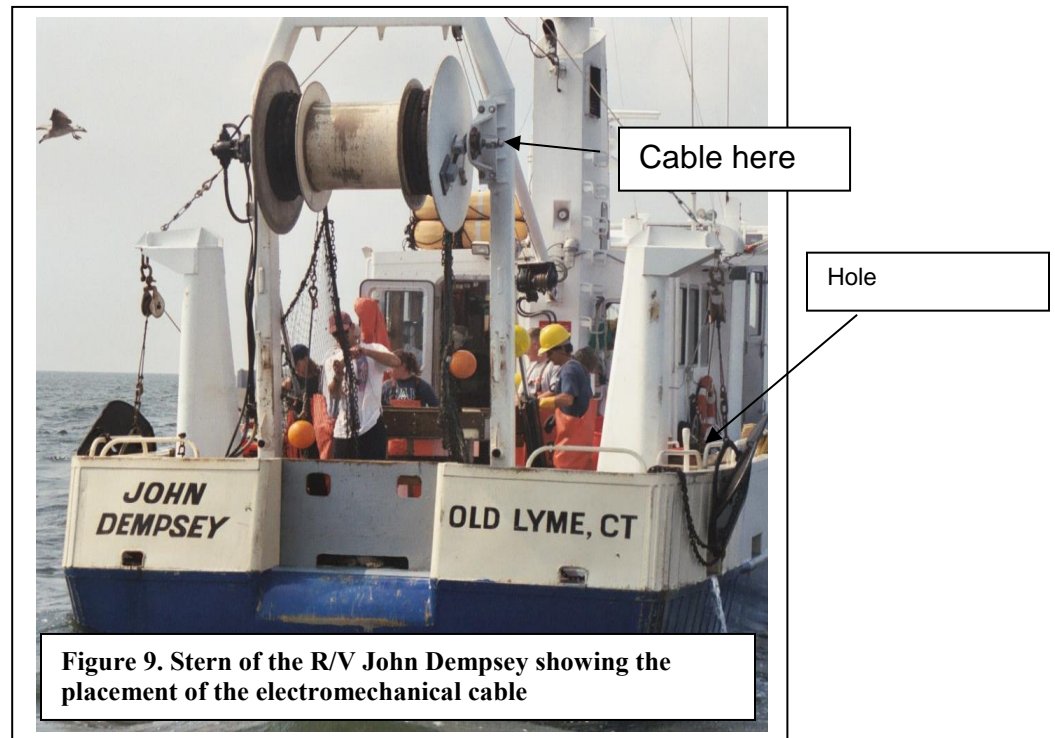
Equipment/Apparatus

- ❖ Electromechanical cable(s)
- ❖ CTD unit
- ❖ 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
- ❖ Coins
- ❖ Laptop computer equipped with TERM19 and SEASAVE software

Procedure

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

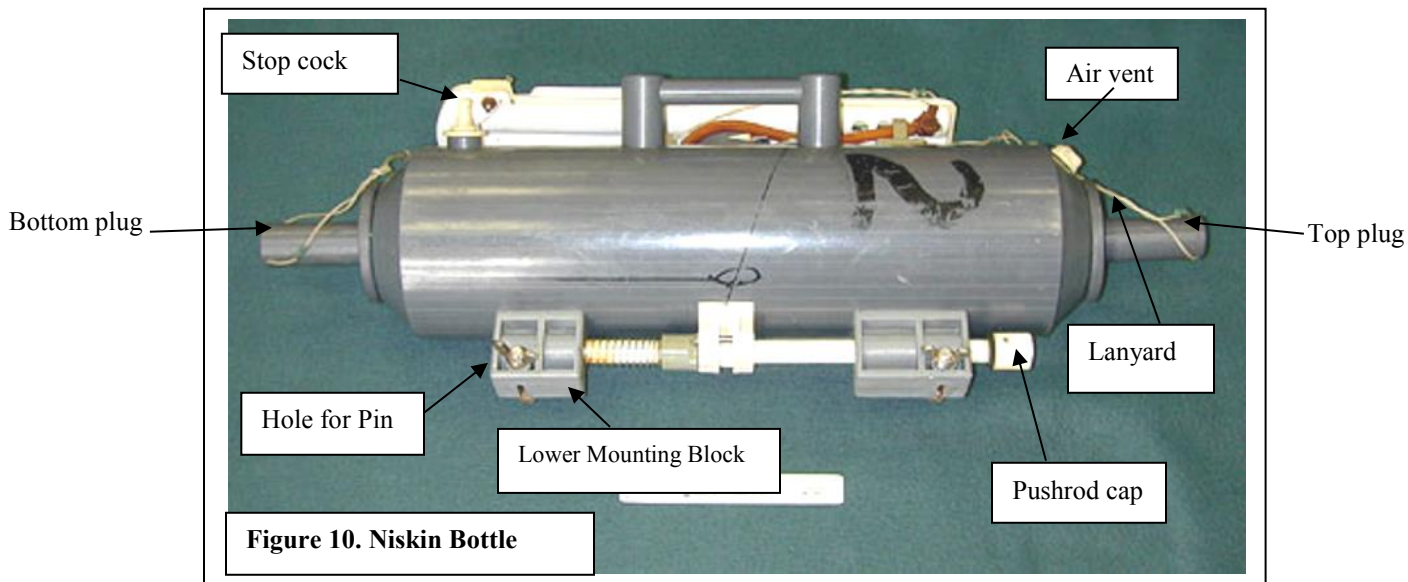
Before disembarking from the dock, the electromechanical cable must be connected to both the net reel and the laptop computer. Enter the onboard laboratory. Remove the foam plug from the hole to the left of the desk on the starboard side. Bring the cable out on deck. Place the end with the green metal connector carefully into the hole through the wall. Return inside. Replace the foam plug from the inside to prevent water from entering the cabin.



Connect the cable to the gray electrical box mounted to the wall above the hole. Go back out on 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.

B. Setting the Niskin Bottles in the Array



1. Insert the Niskin Bottle in to the array (Figure 10 and 11).

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 pushbar so that the round white cap fits into the hole on top of the array.

Continue placing bottles on the rosette working counterclockwise starting in position #3. There should be no empty positions between bottles.

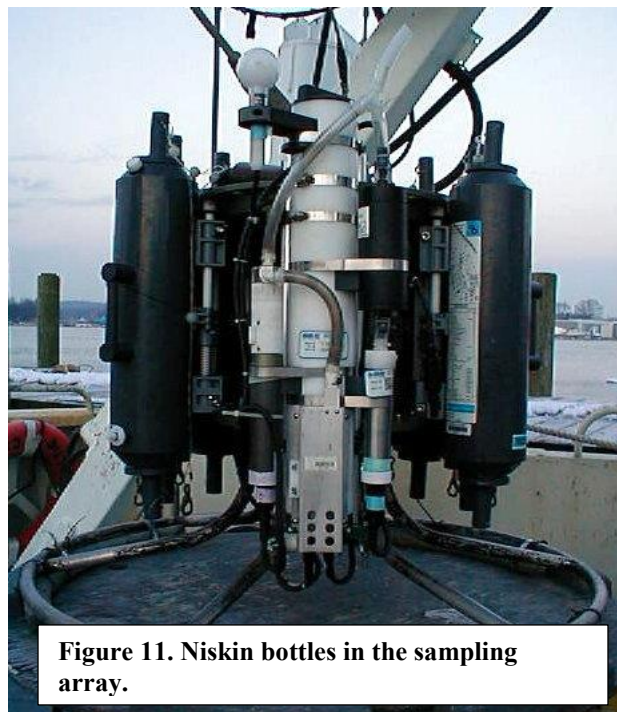
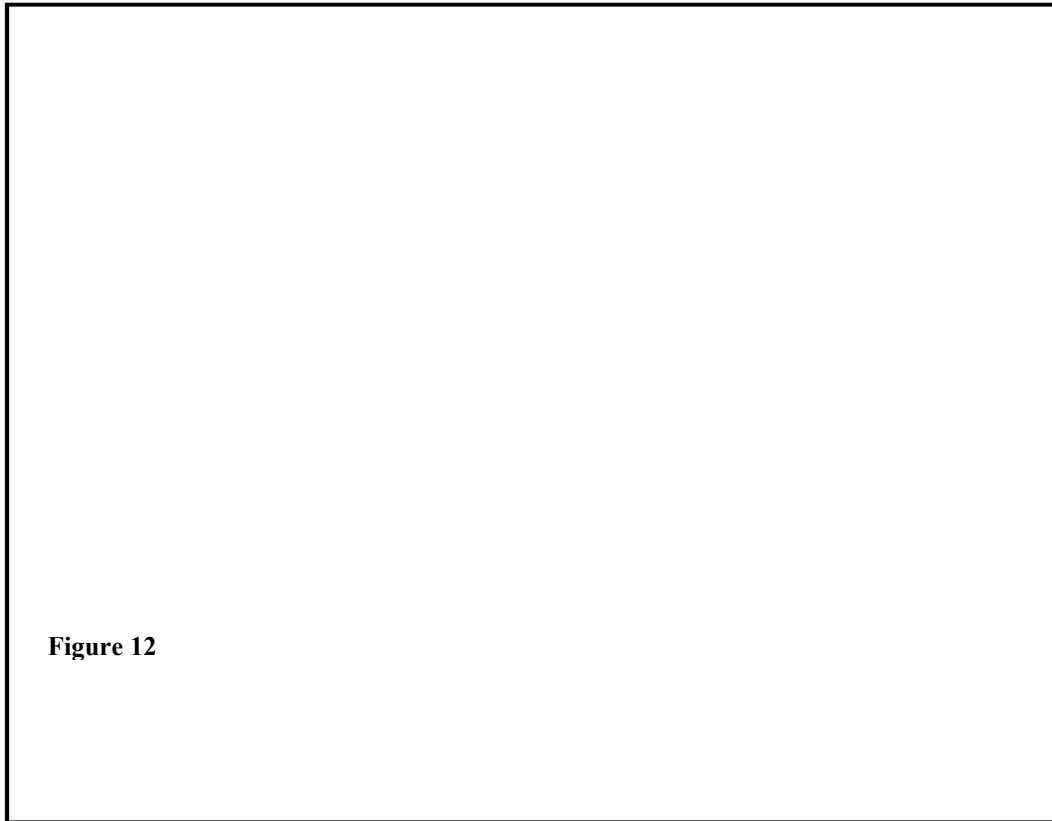


Figure 11. Niskin bottles in the sampling array.

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.



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. Moving CTD between the Cage and the Rosette



Figure 13. CTD in cage

The CTD is always transported and stored in its cage and so must be removed from the cage (Figures 11 and 13) and secured in place on the rosette mount (Figure 11). **NOTE: When you are ready to remove the CTD to move it between the rosette and the cage stay with it. Do not leave it half secured in either place and do not put it down on any surface.** Avoid transferring the CTD from the cage to the rosette when conditions are rough and the rear deck is being constantly sprayed with saltwater. Under such conditions, request that the captain slow the vessel in order to make the transfer with a minimum risk of bare electrical connectors coming in contact with saltwater.

The CTD must first be moved from the cage to the rosette. The rosette mount attaches to the rosette. Following the day's activities the CTD can be stored in the on-board lab in the mount. The mount is secured to the wall in the Niskin holder assembly on the port side of the vessel.

- 1) Unscrew and remove the bolts and associated plastic washers attaching the base of the CTD to the lower crossbar of the cage. Unscrew the stainless steel thumbscrew from the top of the CTD mount.
- 2) Loosen bolts securing the upper support, but do not remove these bolts yet.
- 3) It may be necessary to unplug the two cables on the bottom of the CTD if they are around the lower crossbar (usually happens upon return from factory calibration). Loosen the collar by turning it to the left (counter clockwise). Carefully, pull the connectors off STRAIGHT. It is very important that these cables be released and re-secured very carefully. The connectors are pins and if they are removed or plugged in without extreme care the pins can bend, weakening them and making them more likely to break. Do not unplug them unless you are ready to move the instrument. If possible reconnect the cables so that they are free from the crossbar.
- 4) Finish loosening the bolts securing the upper ring and carefully remove CTD from cage. Immediately take the CTD to the rosette or place it in the Niskin assembly. The CTD mount is fashioned out of a repurposed Niskin bottle equipped with a stainless steel pushrod and a locking washer and thumbscrew. Mount the CTD onto the rosette or into the in lab assembly exactly as if mounting a Niskin. Be sure to install and tighten the thumbscrew to prevent the pushrod from
- 5) Carefully unplug the cap from the connector. Unplug the cap from the cable attached to the rosette. Carefully push the connector from the cable onto the connector from the CTD. Always pull connectors off and push them on STRAIGHT to avoid bending the pins. When it is cold, the rubber of these connectors is not pliable and it is more difficult than usual to plug the connectors in. Do not force them. Most of the connectors have a raised bump on the outside that corresponds to the position of the largest pinhole; this

bump should be lined up with the largest pin. Do not leave the instrument unattended when any of the connectors are unplugged. Salt water is very damaging to these connectors so it is imperative that they not get wet. A silicone o-ring compound is used to keep moisture out. If it appears that the grease is gone, add a very small amount around the rubber ring beneath the pins. If the pins get some salt water spray on them, carefully wipe off with a damp (fresh water) towel and then dry thoroughly before plugging in.

- 6) Be sure that all locking rings around the electrical connectors are secure; but DO NOT over-tighten.

D. Preparing CTD for Deployment

- 1) An ice pack is secured against the dissolved oxygen sensor to keep the sensor from heating up while on the deck between casts. It is important to secure the ice pack at the start of each day so that the CTD will be ready for the first cast. (During some times of year this may not be necessary because air and water temperatures are very similar. An ice pack can also work to conserve heat though, so if there is risk of freezing while unit is on deck, an ice pack can help avoid this.)
- 2) Remove the syringe with tygon tubing that attaches to the bottom of the conductivity cell. Put this syringe in a safe place in the lab. **The conductivity cell is glass and should be handled especially carefully - DO NOT force any tubing on or off.** This tubing can remain off during the course of a day. Remove the plastic cap over the tubing at the top of the CTD.
- 3) Check to be sure that the plumbing is hooked up correctly (see Figure) and that all of the connections are secure. The correct hook-ups allow water pumped in through the bottom of the conductivity cell to move across the DO sensor, through the fluorometer and then through the pump and out the outlet at the back of the pump.
- 4) Check the screws on the magnetic ON/OFF switch. These screws have a tendency to loosen and should be checked daily. Do not over tighten. Switch should move freely but not slide on its own.
- 5) Check to be sure that all cables are connected and that all locking sleeves are tightened. The I/O port of the CTD needs to be connected to the Y-cable that connects the end of the electromechanical cable to both the rosette and the CTD, or to a separate real-time cable.
- 6) Leave ice pack in place until just before cast is to begin.

E. Start/set up Laptop

In general, the CTD memory is cleared at the beginning of each day. **Be sure that all stored files have been uploaded to the computer before initializing the log.** If the unit is operating in real-time mode the files are automatically uploaded to the computer. It doesn't hurt to double check the C:/windows/survey name directory before initializing the log.

Initialize the log

- 1) Plug the communication cable from the CTD (from the gray box in the lab) into the com port on the computer. Turn on the computer.
- 2) Double click the TERM19 icon. At the prompt hit the F6 key to wake up the CTD.
- 3) At the S:> prompt type DS to check instrument status.
Check main battery voltage, which should be at least 10.3. If less than 10.3, a freshly charged battery should be installed (batteries are rechargeable nickel-cadmium battery pack or 9 alkaline D-cells). See end of this section for directions.
Check Minimum Conductivity for Pump Turn-on: **3270** for the open sound (or 0 for near shore waters (NCA)). Check date and time; sample rate: 1 scan per 0.5 seconds; etc. If the pump turn-on is set to zero, type sp at the prompt to set pump. Type desired setting and then make sure pump delay is set to 45 seconds.
- 4) Press F8 to clear the CTD memory (initialize the log). Select Yes at the prompt. The computer will say please wait, then it will say logger prepared for deployment and it will exit TERM19.

Enable real-time acquisition

1. Double click on the SEASAVE icon.
2. Select the proper .CON file when prompted at the start. Be sure that the scroll display (SEASAV1:3) has the display file survey.dsf selected under setup. Also change the headers to match the survey. On the menu bar click configure and then header.
3. Next click "Real Time Data" from the menu bar.
4. Then select "Start Acquisition".
5. Go to "Enter output data file name" make sure that the path is correct and enter the file name e.g. A4080206 (station name/month/day/year).
6. Click "Start Acquire" with the mouse and enter the station name and the time on station.
7. Press enter to start acquiring data.
8. The computer will begin to count down and you have 60 seconds to turn the CTD unit on at the rosette.

NOTE: CTD downcast should be continuous; at a rate of approximately 0.2 meters/second (slower is OK).

F. Rosette Deployment

- 1) Don hardhat and life jacket. Proceed to the rosette table at the stern of the boat.
- 2) TURN ON THE CTD. Be sure to push the magnetic switch up firmly and fully.
- 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 CTD 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.

- 4) Guide the rosette off the stern, pushing it out slightly and keeping it stable so it does not swing and/or bang up against the stern of the boat. Continue to push it out and guide it as it is lowered into the water. Look for air bubbles coming from the CTD as the rosette is lowered. This ensures that the plumbing is not clogged. If working with a real-time data cable separate from the electromechanical cable, this cable must be fed out as the rosette is lowered. DO NOT allow this cable to slack as it could be drawn beneath the vessel and become tangled with the propeller (very bad).
- 5) Let the hydraulics operator know when the CTD is submersed just enough to cover the small extension of tubing that extends from the tubing connector - where the air is forced out of the plumbing system through a small release hole. Be sure that the CTD is submersed enough so that the tubing extension remains submersed. If the water is choppy the rosette will need to be set slightly deeper, because if the small tubing extension comes out of the water, air can get into the system and the pump can lose its prime.
- 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 CTD 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. CTD 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 CTD 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 m off the bottom.

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

Record the CTD 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



Figure 14. Deck Command Unit

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). Refer to [Attachment G](#) as a guide to the number of bottles collected at each station and depth. Generally, two bottles are filled at this depth.

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 CTD data on the field sheet and filling bottles is repeated; generally two bottles are filled at this depth.

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, and grab the base and help guide the rosette onto its stand. **TAKE CARE TO KEEP HANDS/FINGERS OFF THE OUTER EDGE OF THE ROSETTE BASE TO AVOID THE**

POSSIBILITY OF HAVING THEM CAUGHT OR CRUSHED BETWEEN THE ROSETTE AND THE BOAT.

- 2) TURN THE CTD OFF.
- 3) Secure ice pack around dissolved oxygen sensor.

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.

During hypoxia surveys, where no filtering is to take place or where near bottom and mid samples are collected, sample bottles for DO determination are usually filled on deck. Additionally, bottles for special projects such as the Altabet study may also 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.

J. Reset Niskin Bottles for the next station

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

L. End-of-Day Equipment Care

Make sure the CTD is turned OFF.

Return the CTD to the storage rack in the wheelhouse if sampling the following day or to the cage at the end of the survey. Reattach the syringe and flush DI water through the conductivity cell into the tygon tubing above the DO sensor by pushing and pulling the plunger a few times. Store the CTD with DI water in the chamber but not covering the membrane to keep 100% humidity environment. Remove all Niskin bottles from the rosette and store in plastic crates on board.

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.
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. Squeeze the bottle slightly and recap so that a minimum amount of air is trapped in the bottle.
3. Samples must be kept cool (4°C) and in the dark. Place the sample bottles in the appropriate cooler.
4. 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 for direction on whether or not to even collect BOD samples.

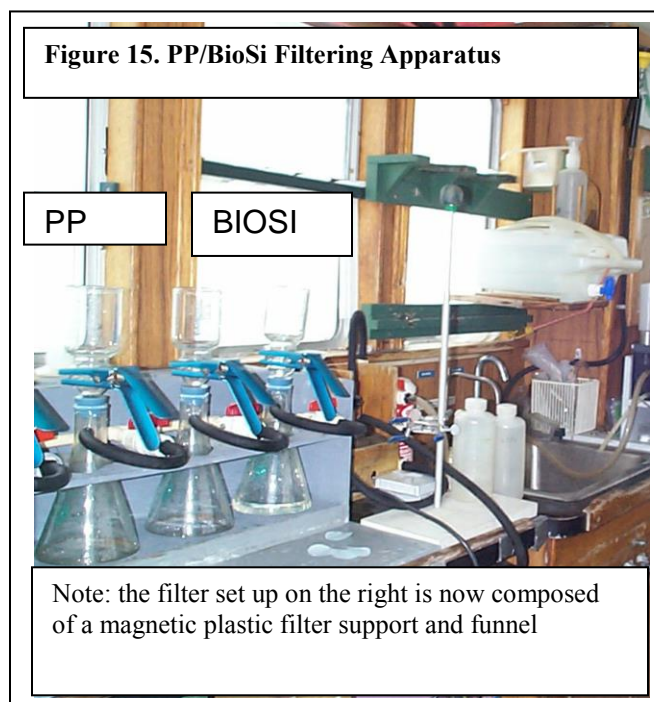
STANDARD OPERATING PROCEDURE FOR FILTERING WATER SAMPLES FOR PARTICULATE PHOSPHOURS (PP), DISSOLVED NUTRIENTS, BIOGENIC SILICA (BIOSI), AND DISSOLVED SILICA ANALYSIS

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
- ❖ 2 metal clamps
- ❖ 2 plastic filter supports
- ❖ 2 magnetic 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 μ M pore-size) Whatman 1825-47
- ❖ 47mm polycarbonate membrane filter (0.4 μ 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



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 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
 - a) 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.
 - b) 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.
 - c) 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.
 - d) 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.
 - e) 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 vacuum pump 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. Remove the filter support and funnel. Place into the wooden hole at the back of the apparatus. Swirl filtrate around the flask and then discard. Reconnect tubing and the filter support and funnel. Repeat.
 - d) Pour remaining 440 mL through the funnel. Turn off the vacuum once all the sample has passed through the filter.
 - e) 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) Repeat with second 250 mL. Turn off the vacuum once all the sample has passed through the filter.
- 9) In the meantime, 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 (right side of the refrigerator/freezer) at CESE. All other samples are placed into the freezer (left side). 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 Hoefler 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.



Equipment/Apparatus

- ❖ 25mm filtering apparatus (Hoefler filtering manifold with filtrate collection tank, associated vacuum tubing)
- ❖ 250 mL graduated cylinder with hole drilled at 200 mL
- ❖ plastic funnels
- ❖ precombusted 25mm GF/F (glass fiber) filters (0.7 μ m pore size) for PC/PN analysis
- ❖ 25mm GF/F (glass fiber) filter (0.7 μ 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.

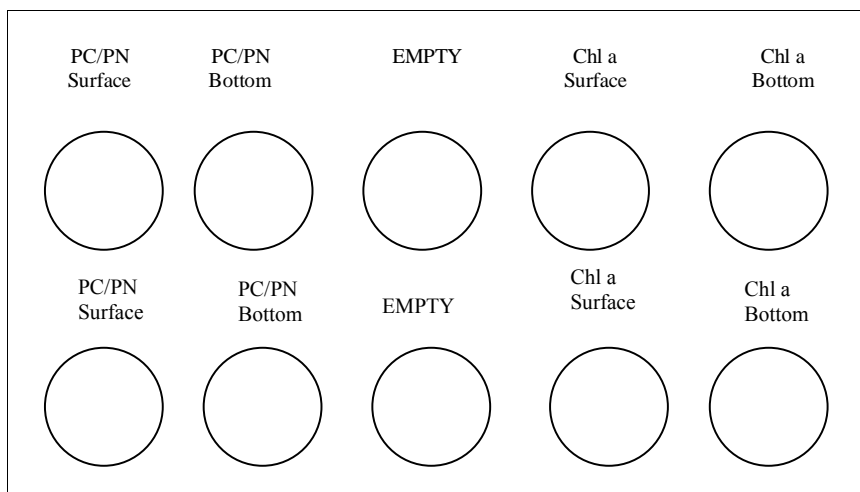


Figure 17. Diagram of filter set up for PC/PN, Chl a analysis. Surface water is filtered for HPLC analysis. Filters are set up following PC/PN and chl a filtering in the spaces marked above as chl a surface.

- 1) Rinse the flat tipped forceps with DI Water.
- 2) 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. Place the PC/PN filter on the filter holder rough side up beginning with the space designated PC/PN surface (Figure 17). 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 from back to front then left to right, Rinse the forceps with DI water Repeat the process for the chl a filters.
- 3) Carefully grasp the funnel by the metal funnel holder. 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.
- 4) Turn the vacuum on at the red-valved 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.

FILTRATION

- 1) Begin filtering with the bottom sample. Carefully remove the Niskin bottle from the right side of the wall rack. 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)
- 4) Sample rinse the 200 mL graduated cylinder by twice then fill to 200 mL mark. Re-wrap the tygon tubing
- 5) If it isn't already on, turn on the pump. Open the red valve to the filters.
- 6) Begin by pouring the sample into the funnel for bottom water PC/PN. 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.
- 7) Refill graduated cylinder with bottom water. Pour into the second funnel.
- 8) Continue on to the chl a filters with the bottom water, working back to front.
- 9) 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 back to front, left to right.
- 10) Allow the vacuum to remove excess water from the filter. Turn off the vacuum. Open the valves to the unoccupied filters to release the pressure.
- 11) Carefully remove stainless steel holders/funnels and set on a paper towel on the counter to the left of the Hoeffler unit.
- 12) Rinse both sets of forceps with DI water. Carefully open the foil packet 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.
- 13) Begin with the surface PC/PN filter and work from back to front, left to right. Slide the pointed forceps down the slot in the manifold. Gently lift the filter. Using the flat forceps, grasp the filter on the edge. Fold filter in half. Using the flat forceps, place the filter in the foil packet in the bottom of one corner. Repeat for second replicate filter. It's important that they NOT TOUCH one another.
- 14) Fold foil over twice to ensure a complete seal. Rinse the forceps in between with DI water. Proceed to the next set of filters. Once all filters have been removed, place foil packets in freezer.
- 15) 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. Surface water is now filtered for HPLC analysis.
Place 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. 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.

The filtrate from this filtering apparatus is not used for any chemical analyses. However, this filtered water is used in the plankton processing.

Watch the water level in the filtrate collection carboy. It must be emptied periodically so that water is not sucked into vacuum line.

Quality Control/Quality Assurance

Duplicates

For quality assurance purposes, surface duplicate samples are filtered at stations M3, A4, and E1 following the same general procedures as outlined above. However, it is preferred that the apparatus is set up to filter all surface samples at once (i.e., surface PC/PN, surface DUP PC/PN, surface chl a, surface DUP chl a) followed by bottom samples.

Blanks

Blanks are prepared following filtering at stations M3, A4, and E1 for PC/PN, chl a, and HPLC analysis. Blanks can be done at any time after the first station is filtered, however they are generally collected at the same time as the bottom samples from stations M3, A4, and E1. Set up the apparatus as usual. Pour the bottom water through the funnels as usual. Do not pour any water through the surface spaces. The blanks will be collected from the surface spaces on the filtering unit. Place the filters into the foil packs as outlined above.

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.

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 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.

Chain of custody forms ([Attachment F](#)) are completed and 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
- ❖ 2-1000 ml filtering flasks
- ❖ 2 metal clamps
- ❖ 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 μ M 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.

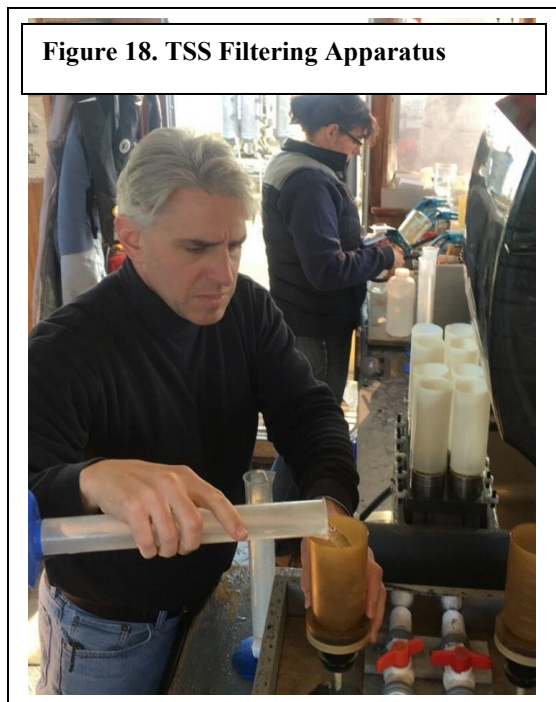


Figure 18. TSS Filtering Apparatus

Procedure

- 1) Rinse all graduated cylinders, filtering flasks, filtering funnels, and filter holders 2 times with DI water before setting up new filter pads. Connect the vacuum tubing. Place the filter supports on the filtering flasks.
- 2) Set up the TSS filters.
 - a) The TSS filters come from the lab in a box with four stacks of foil tins. Select one stack from the box. Rinse the dedicated TSS 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 on the support of the filtering apparatus closest to the wall (labeled surface) rough side up. Be sure the filter is centered so that the sample cannot get around the filter.
 - b) Remove the filter from the second tin and place it on the filter support in the front (labelled bottom), rough side up.
 - c) Place the plastic magnetic filter funnel on top of the filter. Be careful not to move the filter.
 - d) 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 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 front (bottom) flask in the TSS station.
- 6) Pour ~200 mL of sample carefully 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) Repeat with second 250 mL.

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.
- 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 back (surface) flask for TSS analysis. Filter 500mL of bottom water sample as above. DI Rinse the filter as above.
- 15) Turn off the vacuum pump.
- 16) Disconnect the tubing.
- 17) Lift the funnels off the supports (one at a time) and place on the shelf behind the apparatus. Make sure that the filter stays on the filter support.

- 18) 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. Carefully open the stack to the tin that corresponds to the filter. It is imperative that each filter be returned to its original tin.
- 19) 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. Use the flat side of the forceps to flatten the filter out at the fold. Remove filter and place in 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. Repeat for the second flask.

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

Duplicates

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

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) Rinse graduated cylinders 2 times with DI water and rinse the funnel as above.
- c) Handle filters only by the edges and only with forceps.
- d) Place filters in appropriate foil tin 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

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. 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 ([Attachment C](#)) 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. Store in the refrigerator at 4 °C in darkness until delivery to Dr. Senjie Lin at UConn, Avery Point. Samples will be delivered with appropriate chain of custody forms ([Attachment F](#)).

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.

Equipment/Apparatus

- ❖ Rosette with Niskin bottles
- ❖ 50L Nalgene carboy
- ❖ 8 lengths of Tygon tubing to drain Niskin bottles
- ❖ Bongo plankton net fit with 200 μm 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 ([Attachment C](#)) for additional health and safety information.

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

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% Lugol's 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 19. 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 μm 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 $\sim 1/8$ sample volume of 37% formaldehyde.
- 13) Samples are placed into the refrigerator.
- 14) Samples are delivered to Dr. McManus at UConn, Avery Point Campus with appropriate chain of custody forms.



Collecting net samples

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.

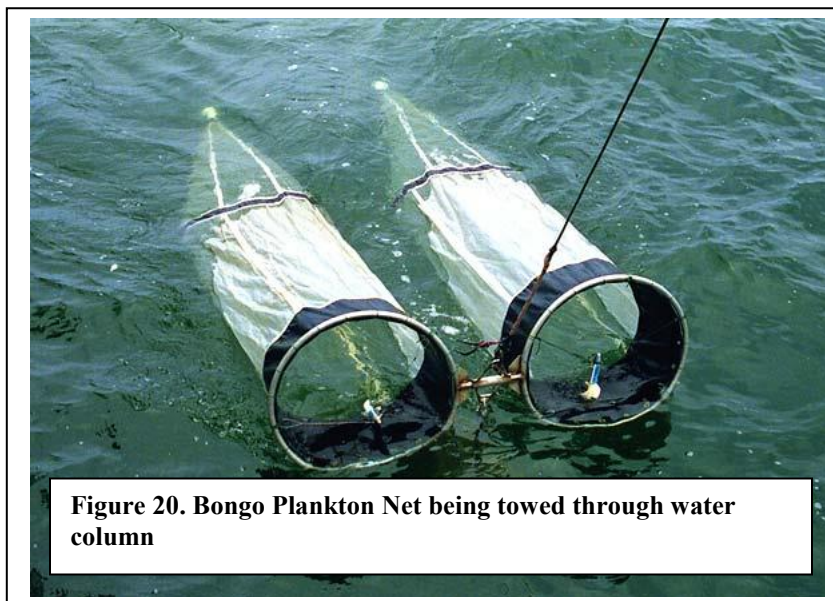


Figure 20. Bongo Plankton Net being towed through water column

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 operators 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.

Processing Net Samples

- 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 $>64 \mu\text{m}$ 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. Place sample in refrigerator.
- 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.

18) Samples are delivered to Dr. Hans Dam at UConn, Avery Point in Groton with appropriate chain of custody forms ([Attachment F](#)).

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

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.

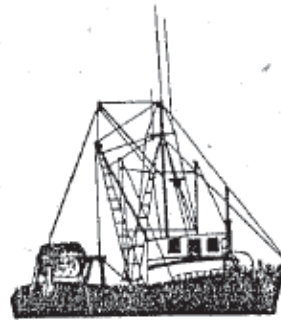
Issued by: /S/ Commissioner Amey Marrella

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.

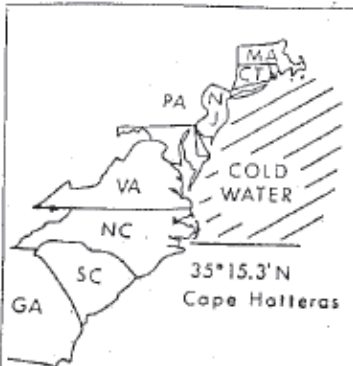


Figure 1. December through April

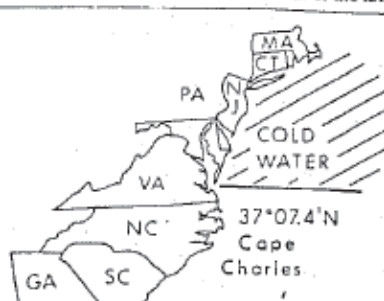


Figure 2. May and November

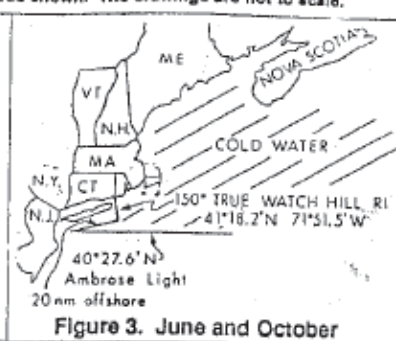


Figure 3. June and October

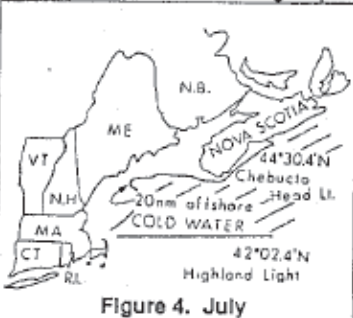


Figure 4. July

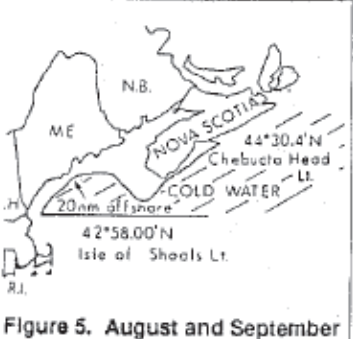


Figure 5. August and September

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.

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FISHING VESSEL SAFETY

FACT SHEET

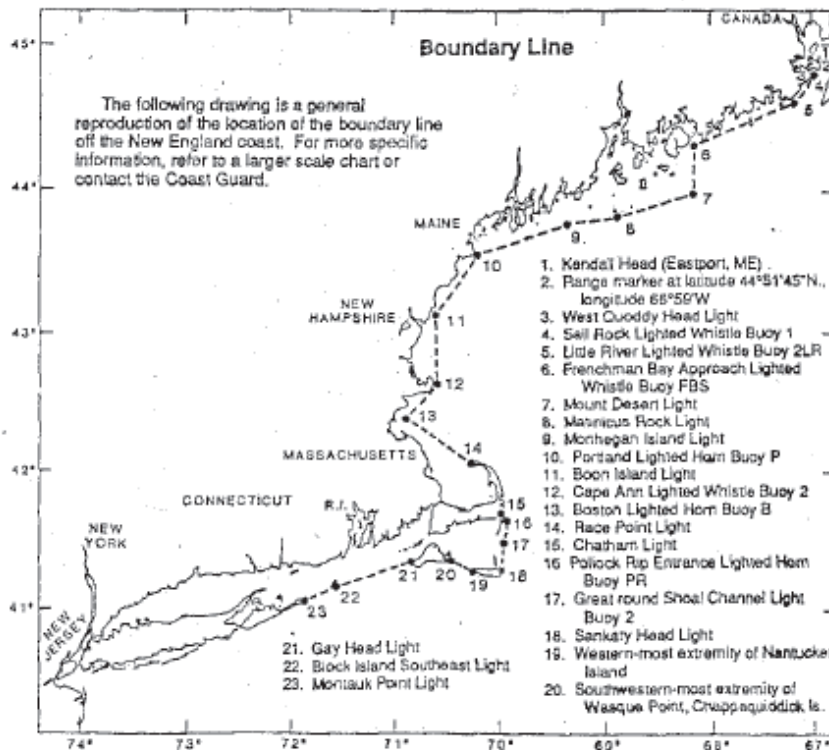


#2 Boundary Line and Coastal Waters

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.

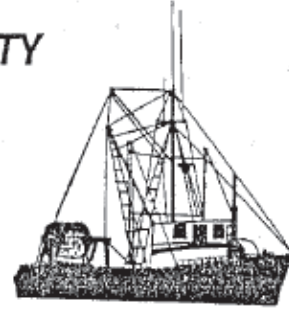


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FISHING VESSEL SAFETY

FACT SHEET

3 Immersion Suits



Who Needs One?

Immersion suits (or gummy 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 i.e. 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 26.110.—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	26.135; 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 ¹	26.135; 25.25-5(a); 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 ¹	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.

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STEARNS®



HYPOTHERMIA AND COLD WATER SURVIVAL FACTS



IFS-589 Industrial Flotation Suit



FS-7580 Recreational Flotation Suit

Time runs out fast¹ 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, Vision® 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 R/V 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

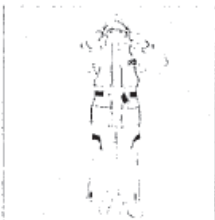
It is recommended that all closures be secured as shown before entering the water, where possible, or immediately after immersion to ensure maximum Hypothermia protection.



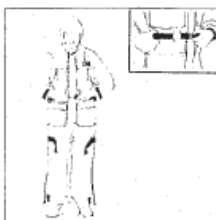
1. Pull on as you would a pair of coveralls.



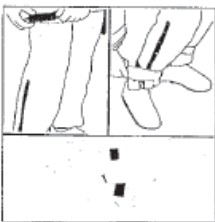
2. Close chest and leg zippers with a slow even pull. Secure velcro storm flap.



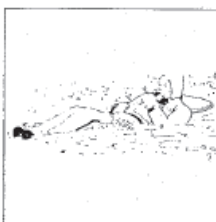
3. Pull hood over head. Adjust draw cord to a snug position.



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.

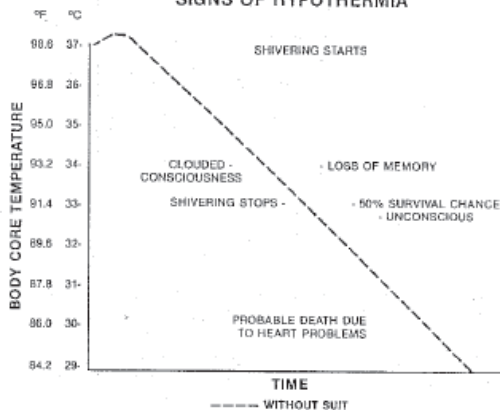
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HYPOTHERMIA

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



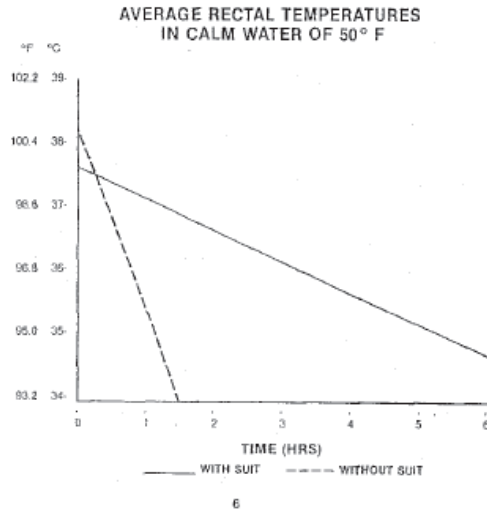
The absence of shivering at a core temperature of approximately 33°C indicates that the body has given up its defenses against the cooling. A state of unconsciousness follows shortly thereafter. Death, as a result of body cooling, may occur when the heart temperature falls below 30°C.

5

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.



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 capsizing, 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.

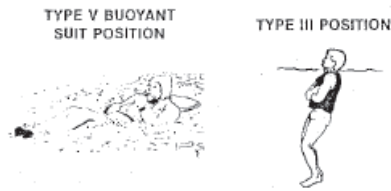
5. Will swimming increase my survival time?

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.



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, re clothe 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 artificial 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.

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



An Anthony Industries company

12

FOR MAXIMUM
HYPOTHERMIA PROTECTION...

Available for use
on R/V John
Dempsey

COLD WATER IMMERSION SUIT



"COLD WATER IMMERSION SUIT"

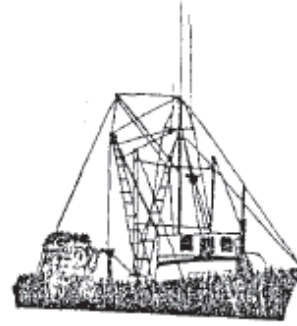
ISS-590i

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 material 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.

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 (46CFR 160.072) 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.

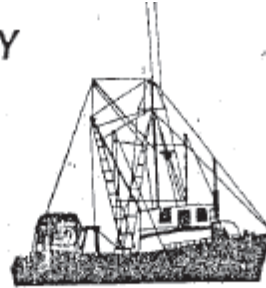
Fill in blanks with vessel name, call sign and description using a permanent marker or a ballpoint.

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 it's 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 or 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 manufacturer's 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

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 readily 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.

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

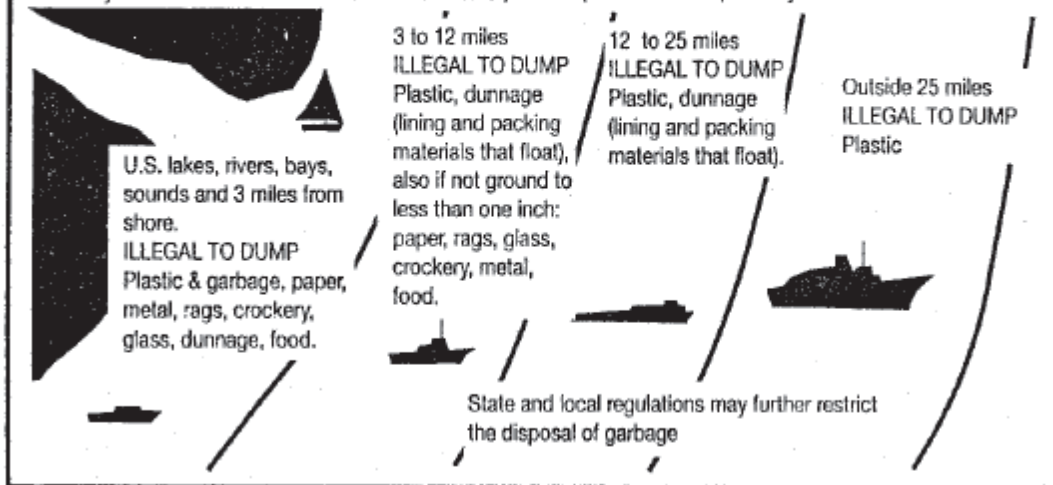
Area	Vessel type	Survival craft required
Beyond 20 miles of coastline.....	All.....	Inflatable buoyant apparatus.
Beyond Boundary Line, within 20 miles of coastline, cold waters.....	All.....	Inflatable buoyant apparatus.
Beyond Boundary Line, within 20 miles of coastline, warm waters.....	All.....	Life 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 foot (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.



ATTACHMENT B

Directions

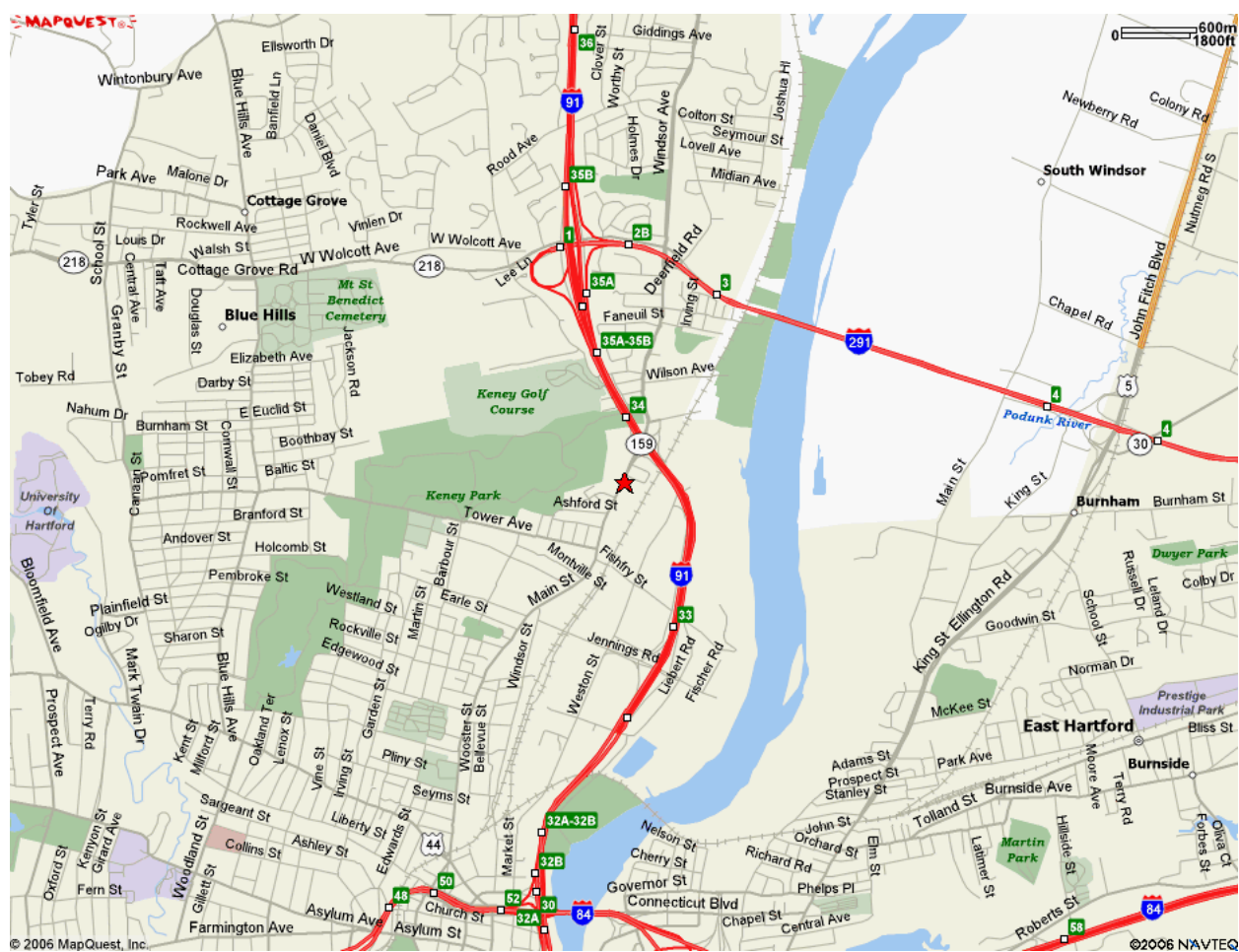
Directions to Windsor Water Survey Prep Lab 9 Windsor Ave, Windsor, CT

From 79 Elm St., Hartford

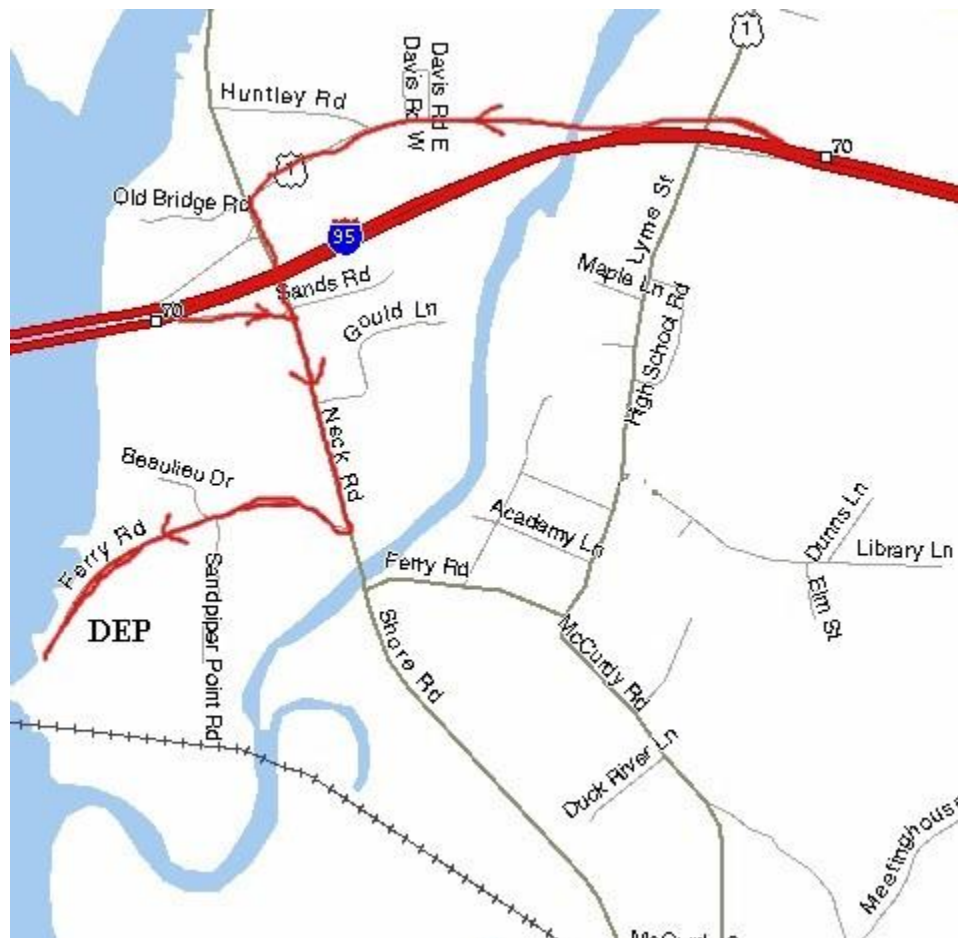
Take Exit for Route 91 North.

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.

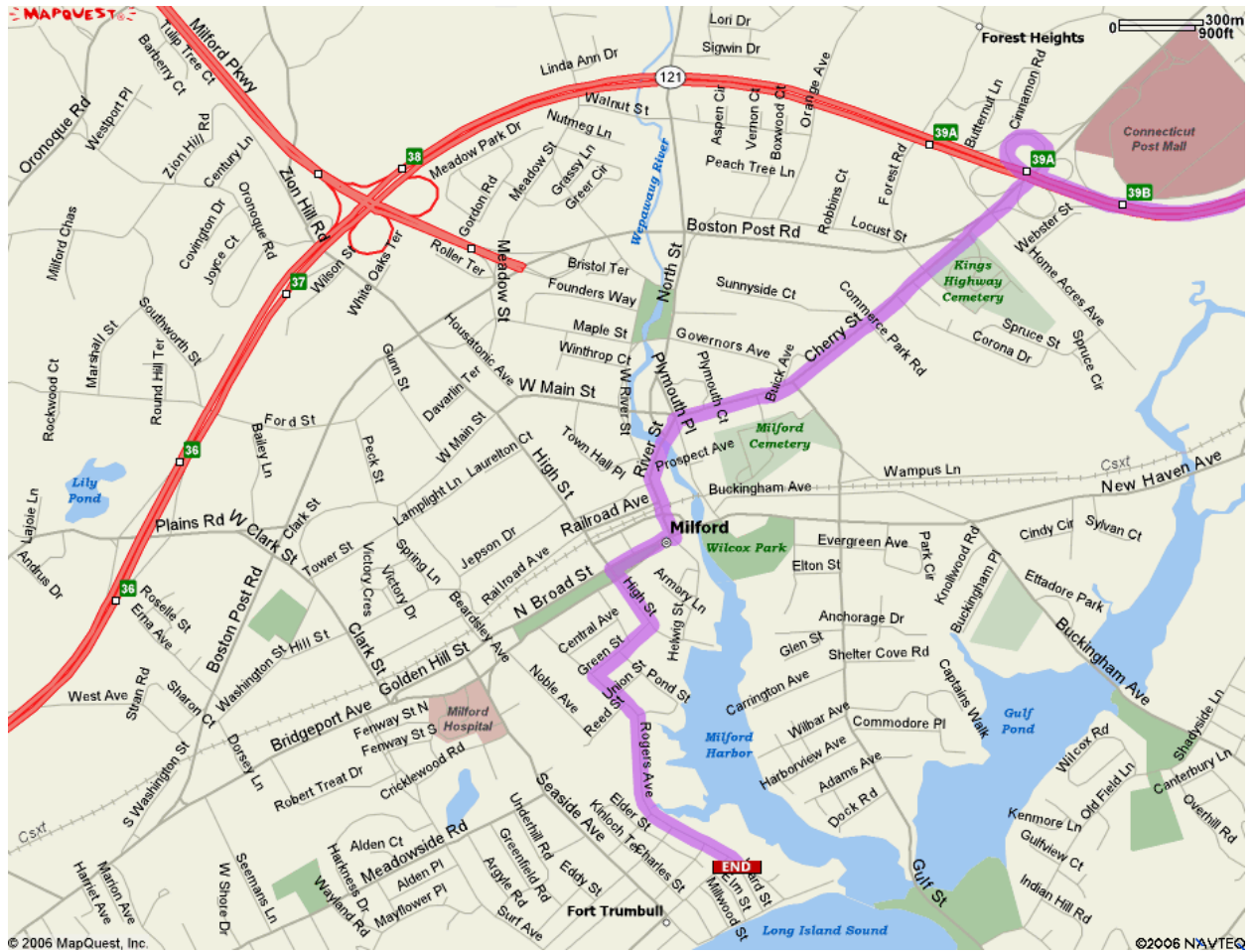
From I-95 North take exit 70, take right at bottom of ramp, take first right onto Ferry Road, Marine Headquarters is on Left at end of road.

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.



Directions to CESE in Storrs

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

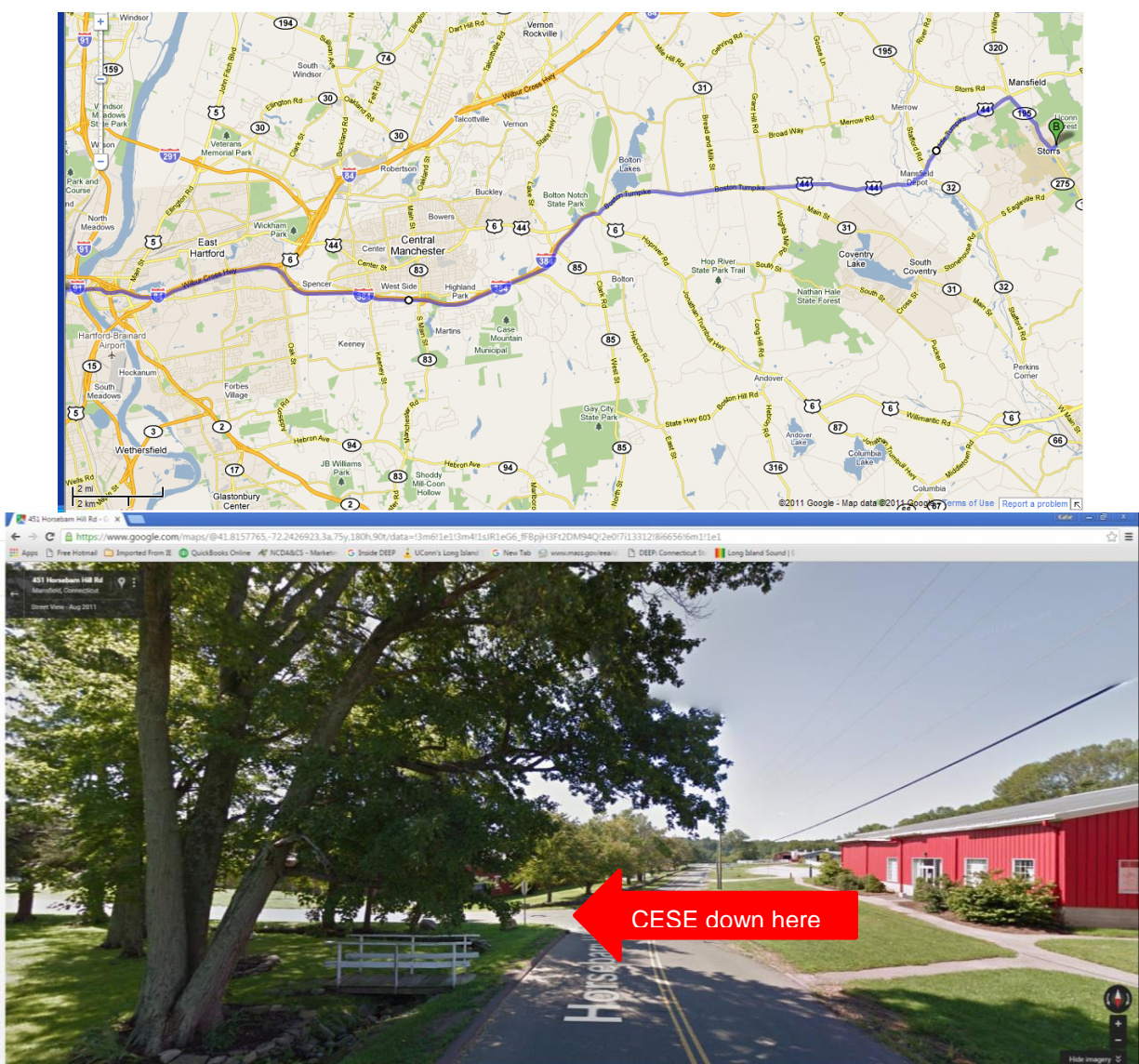
Take I-84 East.

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

MSDS Sheets

Chemicals used by the Long Island Sound Monitoring Program are not necessarily obtained from the manufacturers identified on these MSDS sheets. 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.

[KCl](#)

[Iodine](#)

[Manganous Sulfate solution](#)

[Alkali-iodide-azide Reagent](#)

[Sulfuric acid](#)

[Starch Indicator Solution](#)

[Sodium Thiosulfate 1.0 Normal](#)

[Formaldehyde 37% Solution](#)

[pH buffer 4.0](#)

[pH buffer 7.0](#)

[pH buffer 10.0](#)

[Conductivity Standard](#)

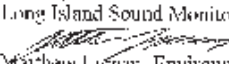
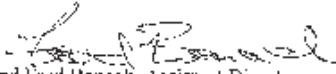
ATTACHMENT D

Memorandum- Winkler Waste Disposal Procedures

Memorandum

February 26, 2003

To: Long Island Sound Monitoring File

From:  Matthew Lysnar, Environmental Analyst 2 and  Fred Banach, Assistant Director
Bureau of Water Management

Subject: Winkler Waste Disposal Procedures

A question was raised during a recent meeting of the Water Bureau's Health and Safety Committee concerning the practice of disposing Winkler dissolved oxygen test wastewaters generated as a result of our Long Island Sound monitoring via discharge to a Publicly Owned Treatment Works (POTW). After discussing this practice with Jim Grier and Dick Mason from the Permitting and Enforcement Division it was determined that the discharge of Winkler wastewaters to a POTW is allowed under the *General Permit for Miscellaneous Discharges of Sewer Compatible (MISC) Wastewater* (issued June 12, 2002). The General Permit allows the discharge of the Winkler wastewaters to a POTW because the discharge qualifies as an "Undesignated MISC Wastewater", it does not contain any prohibited substances listed in Appendix B, Tables II, III, V or Appendix D of Section 22a-430-4 RCSA (Appendix F of the General Permit), and the volume is less than 2% of the POTW design flow. There is no need to formally register the discharge because the volume is less than 500 gallons per day.

In accordance with the conditions of the General Permit, the Long Island Sound monitoring staff will maintain an on-site log for each discharge. The log will include date and time of discharge, volume, description of wastewater discharged and the process that generated the wastewater. This log will be maintained at the CT DEP Lab building at 9 Windsor Avenue in the water survey preparation room.

cc. Tom Morrissey
Tessa Gutowski
Sheila Jenkins
Dick Mason
Paul Sucey
Christine Olsen

ATTACHMENT E

Field Data Sheet

Explanations/Codes

Station Observations The following information on the field data sheet is completed while the CTD is equilibrating (soaking) for three minutes.

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 °C; obtain from the digital thermometer located on the Dempsey to the left of the desk in the onboard lab

CTD #- indicate which CTD is in use

CTD#1= SN #0765

CTD#2= SN #1724

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 180° 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 state 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 CTD 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 (the I68 temperature not the oxygen temperature)

Salinity

Dissolved Oxygen

Winkler Data- Record the bottle numbers and corresponding depth codes for Winkler samples. When the titrations are performed, the person titrating fills in his/her initials and enters the values from the buret. The average of all the Winkler values from each depth is recorded in the Winkler dissolved oxygen column.

Coastal 2000 Station Number- Fill in station number if collecting NCCA samples

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)

+							
---	--	--	--	--	--	--	--

CTD

--	--	--	--	--	--	--	--

Profiling Method

--	--	--	--	--	--	--	--

Surface Bottle Method

--	--	--	--	--	--	--	--

Tide Stage

--	--	--	--	--	--	--	--

% Cloud Cover

--	--	--	--	--	--	--	--

Current Weather

--	--	--	--	--	--	--	--

Sea State

--	--	--	--	--	--	--	--

Station Depth (m)

--	--	--	--	--	--	--	--

CTD READING

Station Depth (m)

--	--	--	--	--	--	--	--

DEPTH FINDER

Raw PAR @ surface / Constant = Incident PAR

						1.62			
--	--	--	--	--	--	------	--	--	--

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

Coastal 2000 Station Number

--	--	--	--	--	--	--	--	--	--

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

Cast #

--	--	--	--	--	--	--	--

ATTACHMENT F

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: <div style="border: 1px solid black; padding: 2px; width: 100px; margin: 0 auto;">7/7/2005</div>			Date of Delivery: <div style="border: 1px solid black; padding: 2px; width: 100px; margin: 0 auto;">7/8/2005</div>					
LIM Number	Sample Code	Volume Filtered (mL)				Filter #s TSS/PP	Comments	
		PC/PN	Chl-a	BioSi	TSS/PP			
		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: (SIGNATURE)			Date & Time		RECEIVED BY: (SIGNATURE)			Date & Time

CT DEP FIELD SAMPLING SHEET
 Long Island Sound Ambient Water Quality Monitoring Program
 HPLC Phyt pigment 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: Laboratory of Dr. George McManus
UCONN Dept of Marine Sciences
1080 Shennecossett Road
Groton, CT 06340
(860) 405-9164

FROM: 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	10	
	K2 -64	10	
	I2 -W	10	
	I2 -64	10	
	F2 -W	10	
	F2 -64	10	
	H4 -W	10	
	H4 -64	10	
	D3 -W	10	
	D3 -64	10	
	B3 -W	10	
	B3 -64	10	
	-W	10	
	-64		
	-W	10	
	-64		
	-W	10	
	-64		

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

-64 = sample from concentrating x-liters of whole water composite with 64 μ 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: Laboratory of Dr. Hans Dam
 UCONN Dept of Marine Sciences
 1080 Shennecossett Road
 Groton, CT 06340
 (860) 405-9164

FROM: 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:

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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		Surface Yes/No	Bottom Yes/No
		Yes	No		
A4					
B3					
C1					
D3					
E1					
F2					
H4					
I2					
J2					
K2					

Notes:

Relinquished by: _____ Date/Time: _____

ATTACHMENT G

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

	Station	Approximate Maximum Depth (m)	Latitude	Longitude	CTD	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	
	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	-72.45766667	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	
	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				
	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	*will require 2nd cast due to station depth; 2 nd cast bottles are collected on down cast, stopping at 6 m intervals starting at the 2 m depth; 1 bottle at each depth	
	D3	3 bottles	Up 6 m – 1 bottle*	Up 6 – 1 bottle	Up 6 – 1 bottle	Up 6 – 1 bottle	3 bottles*		
	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		