



PROTOCOLS AND PROCEDURES FOR OOI DATA PRODUCTS: QA, QC, CALIBRATION, PHYSICAL SAMPLES

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Protocols and Procedures for OOI Data Products: QA, QC, Calibration, Physical Samples

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1 Introduction

1.1 Purpose of this Document

The purpose of this document is to describe the Protocols and Procedures for Quality Assurance (QA) and Quality Control (QC) for the Ocean Observatories Initiative (OOI) data and data products and physical samples. This includes calibration and field verification procedures. This is a “living document.” Updates are anticipated, as procedures are refined during the construction and operations and maintenance phases.

1.2 OOI Definitions

The definitions below in *italics* are reprinted from the official OOI DOORS reference module; other definitions are unofficial, but descriptive and pertain only to this document.

As-Found Condition: Condition e.g. of an instrument in an unaltered state, usually after recovery of the instrument after a mission. “Unaltered” implies that additional measures have been taken to preserve that state, if the instrument is stored or handled for any significant amount of time (e.g. by storage in dark, moist, cold conditions, no exposure to (UV) sunlight). For the purposes of this document, the intent of determining sensor offsets after recovery in the as-found condition is to provide a known reference point for calibration/validation at the end of the mission.

Calibrate: Test or verify the accuracy of a measuring instrument or process by comparing its output with a known reference standard. Calibration also refers to adjustments to an instrument, process, or data to attain the required accuracy.

COTS: Commercial, Off the Shelf instrumentation. Current instrumentation available for purchase from vendors.

CNC: Core Non-Commercial. Custom or one-of-a-kind instrumentation that may utilize COTS components in a novel way. Some CNC may be considered prototype for development of a future COTS instrument.

Data: Term that is not used in its unqualified form. In the oceanography domain, refers to the measurements that come from sensors and not to any data products computed based on these measurements.

Data Products: An information product that is derived from observational data through any kind of computation or processing. This includes aggregation, analysis, modeling, or visualization processes.

Data Quality Assurance: Quality Assurance (QA) is a set of review and audit procedures implemented by personnel or an organization (ideally) not involved with normal project activities to monitor and evaluate the project to maximize the probability that minimum standards of quality are being attained. With regard to data, QA is a system to assure that the data generated is of known quality and data production procedures are being followed. This assurance relies heavily on documentation of processes, procedures, capabilities, and monitoring. Reviews verify that data quality objectives were met within given constraints. QA is inherently a human-in-the-loop effort. QA procedures may result in corrections to data. Such corrections shall occur only upon authorized human intervention (e.g., marine operator, product scientist, quality analyst, principal

investigator) and the corrections may either be applied in bulk (i.e., all data from an instrument's deployment period) or to selective data points. The application of QA corrections automatically result in the reflagging of the data as 'corrected', and substantial documentation must accompany any QA action.

Data Quality Control: Quality Control (QC) is a process of routine technical operations, to measure, annotate (i.e., flag) and control the quality of the data being produced. These operations may include spike checks, out-of-range checks, missing data checks as well as others. QC is designed to:

- (i) Provide routine and consistent checks to ensure data integrity, correctness, and completeness;
- (ii) Identify and address possible errors and omissions;
- (iii) Document all QC activities.

QC operations include automated checks on data acquisition and calculations by the use of approved standardized procedures. Higher-tier QC activities can include additional technical review and correction of the data by human inspection.

Field Verification: Instrument- and deployment location-specific field test; analysis of representative physical sample; or comparison, (i.e., ground-truthing), with data from other OOI instruments (inter- or intra-array and platform) or external sources for data quality assurance. The resulting data from *in situ* measurements, physical samples or on-board standards may be used to perform secondary calibrations.

HITL QC: (Human in the Loop) Also known as "Visual QC", inspection of data performed by a subject matter expert (SME) based upon unique expertise or particular knowledge of an instrument or a data product. This may involve evaluating an entire data set or just those portions of a data set flagged as poor quality data and carrying out the step necessary to correct and reflag that data as good.

In situ Calibration: Calibration process that is performed for an instrument while it is deployed *in situ*. This process can refer to ancillary measurements (of standards) made onboard the instrument *in situ*, or just baselining the instrument for drift. The results can be used for near real-time or delayed adjustment of instrument measurements for drift relative to the primary calibration.

Marine operator: The individual responsible for operating an array or asset within the OOI while deployed. This individual is likely to be located at one of the marine-IOs and an OOI employee or contractor.

Physical Sampling: Collection of a sample(s) by four specific instrument classes on the Regional Scale Nodes (OSMOI, RASFL, PPSDN, FLOBN). This also applies to collection of a sample in the field at deployment, recovery, or anytime during deployment, to use for field verification of an instrument's measurements.

Primary Calibration: Generally performed by the vendor, unless the instrument allows the operator to change or update coefficients while deployed (e.g., *in situ* calibration). It typically uses a formula to convert instrument native measurements (e.g., counts/voltage) to physical units. Each instrument purchased will be shipped with a certificate of calibration and product data sheets by the vendor at the time of initial purchase and with factory maintenance/calibration. These documents may contain information that reflects the hardware settings/adjustments made at the factory. This formula is either programmed into an instrument or applied via software for data conversion. The OOI does not change these hardware settings or software coefficient, except in rare instances where *in situ* calibration is possible and deemed appropriate. All subsequent (secondary) calibrations only determine an adjustment needed to refine the data produced. We may informally distinguish them by speaking of hardware (instrument) vs. data calibration.

Quality Assurance: A planned and systematic means for assuring management that defined standards, practices, procedures, and methods of the process have been followed in the performance of a job.

Quality Assessment: An evaluation of the performance of something – in this case: an instrument, automated QC procedure, calibration procedure, etc.

Quality Control: The operational steps performed to enforce quality of a particular product.

Secondary Calibration: The process by which data measurements are adjusted to correct for errors arising from deployment, operations, recovery, inherent instrument drift or fouling. The data necessary for these adjustments come from tests performed by the OOI on board a ship, at a laboratory or maintenance bay, or refurbishment facility. Tests are needed at deployment and at maintenance visits or recovery of the instruments, in order to: check primary calibration, assure nothing changed during shipment and integration, and determine drift of instrument between deployment and maintenance visit or recovery.

Validation: Confirms that the product, as defined, fulfills its intended use.

Verification: Confirms that work products properly reflect their requirements. In other words, verification ensures that “you built it right.”

1.3 Parent Documents

The following documents are the parents from which this document’s scope and content derive:

- Data Management Plan (DMP) (1102-00000)
- Quality Assurance/Quality Control Plan (1003-00000)

1.4 Motivation for Data QA/QC efforts

OOI’s six major science themes form the top-level requirements of the system and flow down to requirements at subsystem and instrument levels (including accuracies, precision, and drift) for at least 49 specific data products (see the L-2 Science Requirements in DOORS modules). It is well known that sensor response and behavior changes with time due to inherent drift, aging, and biofouling. Plus, several points of potential failure and error exist between the instrument itself and final delivery of the data to a user. Therefore, a data QA/QC effort is needed to ensure that the OOI data products meet the OOI requirements in order to satisfy the stated science themes.

The key questions to be answered in evaluation of a data stream are:

1. Does the data meet the requirements (esp. accuracy)?
2. If not, can the data be adjusted sufficiently to meet the requirements?

Consequently, data QA/QC consists of two activities:

1. Assessing data quality versus the requirements, resulting in a pass/fail assessment
2. Adjusting data via calibrations, both routinely (based on both the field verification samples and “as-found” measurements done at the vendor calibration facility) and in cases of “fail” assessments

The overarching goal of the data QA/QC process is to have as few “bad” data points as possible and to accurately characterize that data as “good” or “bad” with a flag upon completing all OOI’s procedures. The OOI also has the option to correct or adjust “bad” data and reclassify it as “good,” depending upon the level of data product and the information available, subject to scope

and budget constraints. As the historical data record is augmented and the OOI program matures so will the ability to correct/adjust the OOI data products.

There are many examples of QA/QC procedures available to the OOI as reference from community white papers (e.g., OceanObs09) and workshops. Maintaining and improving the Data QA/QC procedures will be an ongoing Operations and Maintenance effort. Development of the OOI Data QA/QC procedures used the most up-to-date established community guidelines for quality assurance of real-time oceanographic data.

One source of such guiding principles is the Quality Assurance of Real-time Oceanographic Data (QARTOD) Workshops. A key outcome of QARTOD is the development of the Seven Laws of Data Management, initially defined in the 2003 workshop and later refined:

1. Every real-time observation distributed to the ocean community must be accompanied by a quality descriptor.
2. All observations should be subject to some level of automated real-time quality test.
3. Quality flags and quality test descriptions must be sufficiently described in the accompanying metadata.
4. Observers should independently verify or calibrate a sensor before deployment.
5. Observers should describe their method/calibration in the metadata.
6. Observers should quantify the level of calibration accuracy and the associated expected error bounds.
7. Human-in-the-loop (HITL) checks on the automated procedures, the real-time data collected and the status of the observing system must be provided by the observer on a time-scale appropriate to ensure the integrity of the observing system.

Future whitepapers and workshops will result in additional data QA/QC guidance and development of instrument-specific processes acceptable to the scientific community. This may result in modifications to the automated or HITL procedures applied to OOI data and data products.

1.5 Role of the Integrated Observatory Network

The Cyberinfrastructure (CI) component of OOI is developing the Integrated Observatory Network (ION). ION includes the entire software and hardware necessary to transmit, process, store, and deliver OOI data products to the user community. The capabilities of ION are detailed in documents on the CI Product Management web page¹. With respect to the capabilities needed to carry out calibration, field verification, and quality control procedures, ION will have the ability to:

- Apply an automated procedure (transform) the input data set(s) to produce data products, e.g., apply a calibration routine to produce a calibrated data product;
- Apply automated QC routines to data and data products and associate quality flags;
- Allow a user to submit data or metadata, and associate it with a data product. This would include additional QC flags and updated data versions created from manual QC or QA processes. This will be fully supported in ION Release 3.

1.6 Data Quality Control Levels

There are three levels of quality control that are applied to data. If available, the user may request data be delivered according to these three levels. The quality control levels for all products are contained in the metadata. The QC levels are:

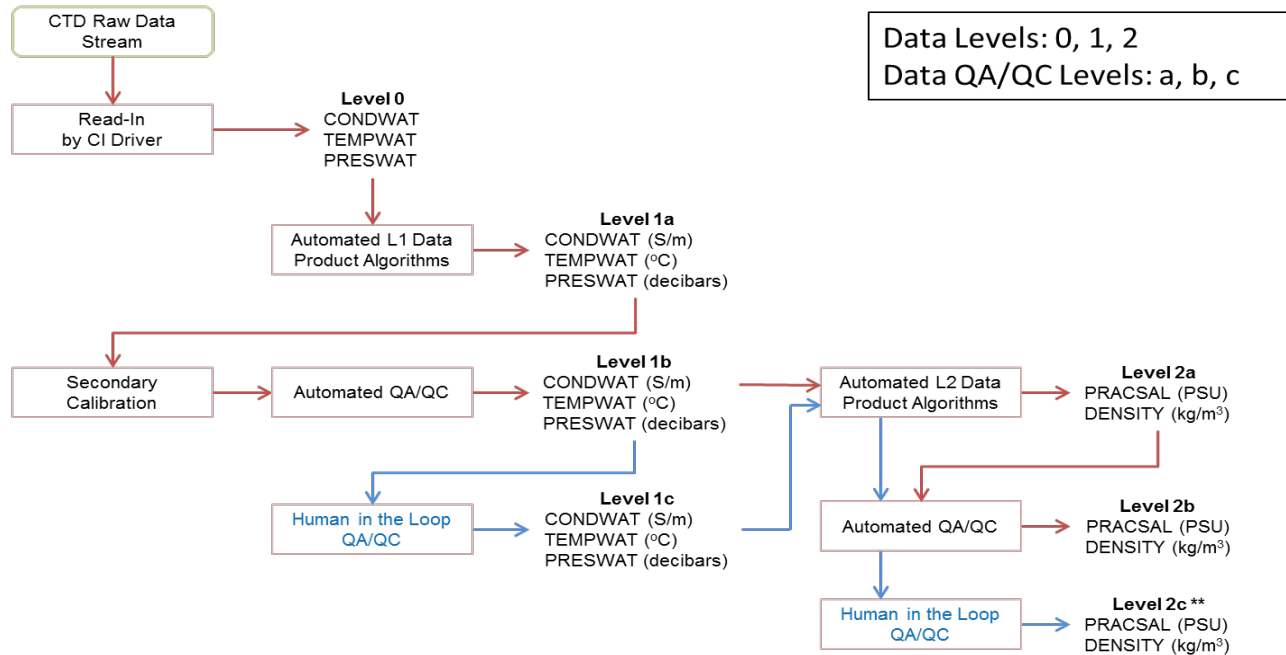
- a) No quality control has been applied to the data.

¹<https://confluence.oceanobservatories.org/display/CIPROD/CI+Product+Management>

- b) Automated QC has been applied. Adjustment/corrections from one or more of automated quality control algorithms have been applied to a data set with each data point potentially receiving one or more quality flag(s).
- c) A marine operator has performed HITL QC or validated the outcome of previous automated QC.

The following flow diagram depicts the various QC levels and functions as sensor data are converted into products. For an explanation of Data Level, please refer to the Data Management Plan (1102-00000).

CTD Example Data Products and QA/QC Flow



** Note that Level 2c data products are only produced from Level 1c data products (see blue data path). Human in the Loop QA/QC is required for both the inputs to and the outputs from the Level 2 data products algorithm.

2 Metadata Requirements

A suite of metadata will support and document the data QAQC procedures. Metadata explain the background information about the deployment and instrument characteristics that enable users to properly assess the data product. The Metadata Implementation document (1102-00001) describes the overall OOI metadata approach and lists in detailed planned metadata elements – it is the appropriate point of reference for complete information on the metadata model and the full list of attributes. In the event two instruments are involved in deriving an L2 data product the metadata for each is available in the L1 data product record. In general, the QAQC-related metadata for data products will include elements that describe:

- The contents of the data product, e.g., descriptive name, abbreviation, included parameters.
- The geospatial and temporal coverage
- The provenance of the data product, including descriptions of processing steps (e.g. algorithm name and description, inputs used), and the instrument (e.g. make/model, settings, calibration history)
- Data quality, e.g., quality flags from QC procedures; estimates of accuracy, precision, and drift from either manufacturer information or OOI evaluations; quality assessment level.

The following is the minimum list of metadata required for all data products:

- Data product descriptor (7 characters)
Example: TEMPWAT, CONDWAT, DENSITY
- Data product descriptive name
Example: Seawater temperature, seawater conductivity, seawater density
- Deployment location on platform (Reference Designator)
Example: GA01SUMO-FI003-03-CTDMO0999, GA01SUMO-FI003-04-CTDMO0999
- Instrument Make/Model
Example: Seabird SBE37IM, Seabird SBE16+V2, Seabird SBE52MP
- Instrument Serial Number (by manufacturer)
- Time of measurement
- Location of measurement (latitude, longitude, vertical axis)
- Calibration information and history of data processing steps
- Instrument settings used in collection of these data.

3 Primary Calibration

As defined above, primary calibration uses a formula to convert instrument raw measurements (e.g., counts/voltage) into science units. The primary calibration (vendor-supplied coefficients and/or settings) of a sensor may be “hard-wired” or programmed into an instrument, or applied externally (via an equation) as an adjustment/correction to a data point. These coefficients are subject to change, nominally during each service cycle. Documentation of primary calibration is included with new instruments as well as those refurbished units returned to OOI. For some instruments, a command can be sent by the marine operator to change the primary calibration prior to or during deployment. For all COTS instruments, the marine operators will be responsible for ensuring instruments are routinely maintained according to, at minimum, vendor specifications and schedule; serviced by the appropriate qualified group (which may not be the vendor) on the recommended schedule; and that the updated primary calibration documentation received following service is properly recorded in the instrument-specific metadata. Original hardcopies are

retained for the life of the program. For the following instruments, the marine operators may perform all of the calibration, service, and maintenance functions: TBD. A single instrument inter-calibration/service center per instrument is recommended by the external community to minimize handling errors and lend O&M efficiency by concentrating specific laboratory instruments or testing facilities needed for a type of instruments at one facility instead of duplicated among all IOs. Due to OOI IO structure and general practicality, each IO will instead handle servicing of their own instrumentation (which will include primary calibration by the vendor). However, all IOs will follow the same protocols set forth by an SME for each instrument. Regular cross-training exercises will minimize the variability in procedures among laboratories, and possibly minimize variability among technicians too.

The OOI approach to calibration is to maintain the vendor-supplied primary calibration (coefficients and/or settings) throughout an instrument deployment but to allow alteration of primary calibrations by the marine operator (if warranted and appropriately documented) between deployments. The only exception to this approach is when the potential exists for the instrument to reliably perform *in situ* or self-calibration (either automated or by operator command) or interim calibration is available between deployments, such as when the results of Field Verification support adjustments, or on-board standard analysis is available from *in situ* calibration, or calibration-affecting vendor software upgrades are provided. In such cases, the policy will require that the marine operator provides documentation including: 1) written justification for making changes, and 2) signed approval authority (may include science team lead or marine operator lead). Upon completion of the change the appropriate metadata fields will be populated or augmented to reflect and adequately capture the new primary calibration. This entire process shall be documented and validated through the QA procedure. Execution of the SME tasks described below is subject to budget allocation by the IOs, although they are clearly recognized as important tasks for the OOI to accomplish that dramatically influence data quality.

3.1 Development of SOP Manual(s) for Instruments and/or Data Products

Gathering high quality data starts with pre-deployment efforts. As OOI technicians dealing with sensors are often not experts in what the instrument is measuring and manufacturer user manuals are often incomplete in describing the latest best management practices, highly detailed manuals must be developed by SME's which spell out the specifics of every step associated with sensors, their secondary calibration and field verification testing, biofouling prevention/minimization, best practices and standardization across the program, and the rationale for such actions. These OOI SOP manuals should be approved by SMEs and include laboratory and field protocols – and should be written in the vein of *Manual for Dummies*.

3.2 Biofouling Prevention or Minimization

SME's should research test and employ various techniques that exist for mitigation of fouling. These always require specific actions prior to deployment – and should be spelled out in the SOP Manuals; Delauney et al. (2010) provide a recent review of conventional methods and Manov et al. (2004) discuss specific techniques for bio-optical sensors. For example, they suggest that open-face sensors be positioned to minimize particles settling on the face (i.e., not act as particle collectors) and that upward-facing sensors (e.g., radiometers sensing downward light) be positioned such that the CTD pump outflow washes off the optics windows (via water jetting on their surfaces, being careful that the radiometers are not shaded by other instruments or parts of the platform) or have a wiper.

4 Field Verification of OOI Data and Data Products

The OOI Data QA/QC approach is designed to ensure that OOI data and data products are of both measurable and the highest possible quality as defined in the L2 science requirements. An integral component of the data QA/QC procedures is the verification that the data and data products produced meet the requirements of the OOI. The Field Verification of OOI data will be a coordinated effort between shipboard personnel (Chief Scientist and scientific and engineering staff) and shore-based OOI personnel (Marine Operators, OOI Operations Managers, QA/QC Analysts). The initial verification step may consist of simple functionality test(s) of an instrument during a pre-installation temporary deployment and/or after a final deployment. The OOI Operations Manager in consultation with the appropriate scientific and engineering staff will determine if the instrument is functioning as intended. Field Verification of the OOI data and data products is the final step in this multi-stage verification of the successful integration and deployment of the OOI system infrastructure. Field Verification consists of comparison of data from different instruments or sources as well as the collection and analyses of representative physical samples or measurements *in situ* and comparison of this “ground-truth” data with that generated by the deployed OOI instruments to provide substantiation of the data and confidence in the quality of the OOI products. Execution of the tasks described in this section by qualified OOI personnel is subject to budget allocation at the IO-level, although they are clearly recognized as important tasks for the OOI to accomplish that dramatically influence data quality. A minimum level of Field Verification effort will be determined for the program, but IOs may conduct additional efforts as required by IO-specific instrumentation and subject to budget, scope and schedule.

The data collected during Field Verification are *multi-purpose*. In addition to substantiating the functioning of the instruments, the comparison of post-deployment, *in situ*, and post- or pre-recovery Field Verification data may be measurements used to adjust for inherent instrument drift and/or biofouling-associated signal degradation. For example, data generated by analyses of physical samples collected *in situ* forms the basis of Secondary Calibration procedures and/or the Human-in-the-Loop data quality assessment process described in the following sections (see Sections 5.0 and 6.0).

The purposes of Field Verification of OOI data are to:

1. provide substantiation that the deployed instrumentation is functioning as intended;
2. provide assurance that the data and data products generated are of the highest quality, within the limits of existing resources;
3. supply users of the OOI and OOI data products with associated metadata, to facilitate their own QA/QC assessments; and
4. form a basis for Secondary Calibration (Section 5.0) and/or adjustment/correction (Section 6.0) of the OOI data.

As part of the overall data QA/QC approach, OOI utilizes a variety of methods for Field Verification of OOI data and data products. Combinations of the following methods may be employed at deployment, *in situ*, or during/after recovery of the OOI core instruments:

1. simple functional tests of system-integrated instruments (e.g., bench test, field communications check) – with evaluation by Scientist or QC Analyst and/or Operations Manager to ensure that instrument is functioning as intended;
2. up/down casts with recently calibrated CTD and/or analogous instrument-equipped rosettes;
3. physical samples collected *in situ* with ship-based and/or shore-based analyses;
4. comparison of co-located sensors/instruments;
5. intra-array and inter-array comparisons of similar instruments;
6. associated shipboard underway systems;
7. analogous measurements made by ships of opportunity;
8. satellite or other remote sensor-based measurements;

9. comparison with re-evaluated OOI and other historical data sets.

An instrument-based summary of the Field Verification procedures for OOI data and data products is provided in Appendix C, and Physical Sampling procedures are found in Appendix G.

5 Physical Samples and Analytical Protocols

5.1 Introduction

The Ocean Observatories Initiative (OOI) collects a variety of physical samples. There are two distinct categories of physical samples: samples that support the field verification (aka calibration) of OOI-supported “core instrumentation,” and samples obtained by OOI core instruments that, when analyzed, constitute an OOI-supported core data product. Field verification is defined as the process by which the operation of deployed sensors and instruments and the quality of their data are substantiated. Physical samples for field verification of data and calibration purposes are collected using standard oceanographic shipboard hydrocasts and by remotely operated vehicles (ROVs). Physical samples for OOI data products are collected *in situ* by OOI-supported core instrumentation. The physical samples include seawater, high-temperature and low-temperature diffuse hydrothermal fluids, fluids from methane seeps, and DNA from diffuse flow fluids. The types of samples recovered will evolve as individual PI and community sensors are added to or removed from the OOI infrastructure.

Water column samples are analyzed for temperature, salinity, density, oxygen, alkalinity, pH, nutrients, chlorophyll, CDOM, and zooplankton abundance and composition (if within budget) to support field verification of data and secondary calibration of OOI core instrument-based measurements. This document describes the samples to be acquired and the sampling and analytical protocols to be used during installation and operations and maintenance (O&M) of the OOI. This document also describes the location, number and volume of samples to be collected. Sampling and analytical protocols for seawater are based on those established during the World Ocean Circulation Experiment (WOCE) and the U.S. Joint Global Ocean Flux Study (JGOFS) with updates. The document provides guidance for the types of metadata and formats to be associated with each step from sample collection through analyses.

This section also describes the chain-of-custody procedures and handling of physical samples acquired by OOI-supported core instrumentation (RASFL, PPSDN, OSMOI, and FLOBN). These physical samples will be analyzed on shore with the results constituting an OOI-supported data product that will be made available through ION. Although there are only four OOI core instrument types that acquire physical samples, it is important that guidelines be in place for future expansion of the OOI because there are significant tracts of oceanographic science that cannot be supported by *in situ* sensing alone.

This section describes the physical samples to be collected by the Ocean Observatories Initiative (OOI) from: 1) seafloor secondary infrastructure (core instruments) and 2) from ship-based instruments and ROVs as required for field verification of data and secondary calibration of OOI core assets (Figure 1). Field verification is the process by which the operation of deployed sensors and instruments and the quality of their data are substantiated. Field verification is considered a subset of the general integrated system “Verification” process and may consist of a variety of techniques, for example: 1) up and down hydrocasts in which conductivity (C) and temperature (T) are used to confirm the CTD data on profilers and 2) physical sample collections for shipboard and/or onshore analyses.

The section also describes the location, number, and volume of samples to be collected; procedures for collection and storage of samples; and analytical procedures. It also provides guidance for the types of metadata and formats to be associated with each step from sample

collection to analyses. Finally, it describes the data products and procedures and timing of data transfer to Cyberinfrastructure (CI).

5.2 Storage and Archive

OOI provides temporary storage for physical samples (collected from *in situ* samplers RASFL, PPSDN, OSMOI, and FLOBN instruments and for calibration of core instruments) in order to enable shore-based analyses and production of data products from core physical samples and perform necessary field verification of data and secondary calibration of OOI core assets. OOI will not keep physical sample archives but will, if possible, turn over samples to interested parties if requested.

5.3 Types of Samples

The arrays on the OOI system of systems are deployed in diverse environments and, therefore, calibration of the sensors for each array may have different calibration requirements and procedures resulting from both the different individual platforms and the environments (e.g., Pioneer, Global, RSN, and Endurance). Physical samples are acquired by both the Regional Scale Nodes (RSN) and the Coastal and Global Scale Nodes (CGSN).

Physical samples acquired by the RSN core infrastructure, RSN shipboard assets (hydrocasts), and by ROVs during RSN installation and maintenance cruises include: fluid samples (seawater, high-temperature vent fluids, low- temperature diffuse hydrothermal fluids and fluids from methane seeps), biological samples (DNA collected on filters), and solids associated from black smoker chimneys acquired during water sampling (Table 1). The water column samples collected by the hydrocasts are analyzed for salinity, oxygen, alkalinity, pH, nutrients, chlorophyll, and zooplankton abundance and composition (if within budget) to support field verification of data and secondary calibration of OOI core instrument-based measurements.

Table 1. OOI-RSN Physical Sample Types

Sample Type	Acquired by Core Sensor	Acquired by Ship-based CTD	Acquired by ROV	Location
Seawater	-	10 L Niskin Bottles	-	RSN PN1A and PN3A moorings; 0-3000 m [†]
High-temperature black smoker fluids	-	-	Gas-tight titanium bottles	Inferno Vent ASHES Vent field
Diffuse fluids	OSMOI Sampler	-	Gas-tight titanium bottles	Inferno Vent ASHES vent field
Methane seep	OSMOI Sampler; Benthic Fluid Flow(FLOBN)	-	Gas-tight titanium bottles	Southern Hydrate Ridge
Microbial DNA	Remote Access Sampler (RASFL) & Particulate Sampler (PPSDN)	-	-	Diffuse site, International District
Black smoker chimney	-	-	Manipulator/bio-box*	-

[†]see section 3.0 for details of water column sampling; *prior to taking gas-tight fluid samples in black smokers it is common to remove the top portion of a smoker to enable access to the orifice to insure high quality temperature measurements and fluid samples.

Seawater hydrocast samples also are collected as part of the OOI Coastal and Global Scale Nodes O&M (Table 2)

Table 2. OOI – Global, Pioneer, and Endurance Physical Sample Types

Sample Type	Acquired by Ship-based CTD	Location
Seawater	10 L Niskin Bottles (or equivalent)	Uncabled global scale arrays: Station Papa, Irminger Sea, Argentine Basin and Southern Ocean
Seawater	10 L Niskin Bottles (or equivalent)	Pioneer array uncabled moorings, autonomous vehicles
Seawater	10 L Niskin Bottles (or equivalent)	Endurance array cabled and uncabled moorings, gliders

5.4 Sample Collection

The following section describes the collection of physical samples on OOI cruises, which may include field verification samples. The metadata for the samples is included in the cruise files and includes information associated with any OOI instruments that are used on these cruises. Where non-OOI assets are utilized in the validation of OOI measurements at any point during an

instrument's deployment or immediately pre- or post-recovery, the metadata for the non-OOI asset must be submitted by the responsible IO in substantiation of the data validation.

5.4.1 Hydrocasts-RSN

Seawater samples are collected at RSN full water column mooring sites (~3000 m water depth) PN1A (Base-Slope) and PN3A (Axial Seamount) at ≤ 1 km off the moorings (distance TBD and depending on operational and safety considerations) for field verification of sensors on the deep profiler, 200 m platform and the shallow winched profiler (Figures 1 - 3). These samples are for field verification of data and possible secondary calibration of RSN core sensor data and data products (Table 3). Two vertical hydrocasts, one at each location, are completed using a 24-place rosette system with 10 L Niskin bottles (Figures 2 & 3, Table 3). One Niskin bottle is tripped at each of the following depths (meters): 5, 10, 25, 50, 75, 100, 125, 150, 175, 200, 300, 400, 500, 600, 700, 800, 900, 1000, 1250, 1500, 1750, 2000, 2500, and ~ 3000 (near seabed).



Figure 1. Site locations for the Regional Scale Nodes (RSN) and cable routes, as well as the Coastal Scale Nodes (CSN) Endurance Arrays.

The Seabird rosette is equipped with the following sensors/instruments:

- Seabird 911 plus CTD
- Pressure
- Temperature
- Conductivity:
- Pump
- Seabird SBE 43 Dissolved Oxygen
- WETLabs transmissometer (25 cm)
- Seatech Fluorometer
- pH: Seabird SBE18



Figure 2. Rosette being recovered aboard the R/V Thompson

The vertical casts are conducted immediately after deployment and successful powering up of each mooring and once it is established that the sensors are working properly.

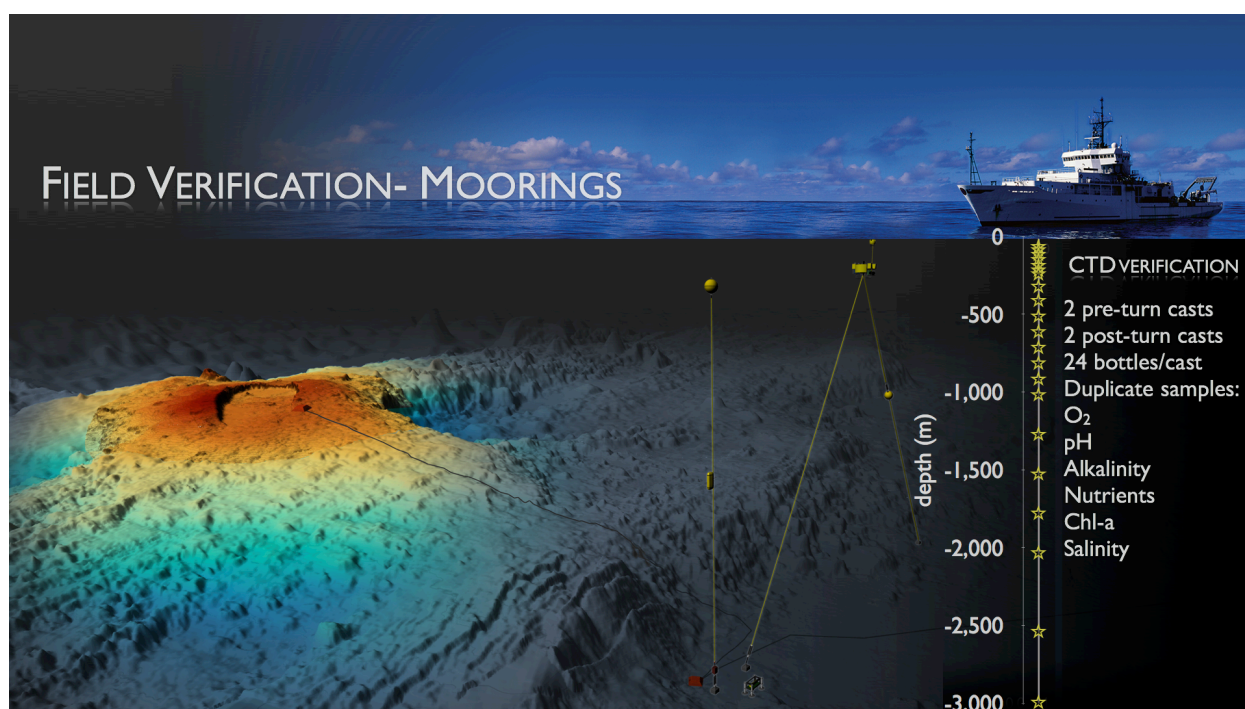


Figure 3. Schematic showing the full water column mooring at RSN site PN3A at the base of Axial Seamount on the Juan de Fuca Ridge. Also shown is the deep profiler, 200 m platform and shallow winch. The stars indicate the water sampling depths using a CTD rosette and 24 Niskin bottles.

Water sampling begins immediately after the rosette is recovered and secured on deck. Following procedures established by programs such as the U.S. Joint Global Flux Study (JGOFS), drawing order of sample types is: 1) oxygen, 2) pH, 3) alkalinity, 4) salinity, 5) nutrient, and 6) chlorophyll. Two samples are drawn for each sample type from each Niskin bottle to provide duplicates.

Table 3: Hydrocast Samples

Sensor Verification/Calibration	Measurement	Sample Volume (ea.)	Onshore/ Shipboard
CTD Seabird	Salinity	125 mL	onshore-UW [†]
Dissolved Oxygen	Dissolved Oxygen	125 mL	shipboard-RSN
Fluorometer 3 wavelength	Chlorophyll CDOM	500 mL or greater	shipboard-RSN onshore-TBD
Satlantic	Nutrients: silicate, phosphate, nitrate	45 mL	onshore-UW [†]
pCO ₂	alkalinity	500 mL	onshore-RSN
pH	pH	50 mL	shipboard-RSN

[†]Onshore analyses of salinity and nutrients are conducted at the UW School of Oceanography Marine Chemistry laboratory (<http://www.ocean.washington.edu/services/techservices.html>). TBD = to be determined. UW = University of Washington.

The same sampling protocol is used for collection of samples just prior to recovery of the deep profiler, sensor packages on the 200 m platform, and the winched profiler. Real-time data from the rosette package (e.g. CTD) are continuously collected during the casts. This post-deployment and pre-recovery physical sampling scheme is repeated during reinstallation of assets onto the

moorings during Operations and Maintenance (O&M) cruises. Storage of samples is described in 5.1. A description of analytical procedures for each measurement is included in Appendix G.

5.4.2 Pioneer Array

Physical sampling is necessary during Pioneer Array mooring turnaround cruises (2 times per year, nominally April and September) and glider service cruises (2 times per year in conjunction with mooring turnarounds and possibly 4 times per year glider/AUV cruises). The purpose of the physical sampling is to verify pre- and post-deployment laboratory calibration of core instruments to better account for drifts, offsets, and non-linear sensor changes that may occur during the operations period. The sampling scheme for the Pioneer array utilizes a CTD rosette package to obtain water column profiles and discrete water samples from which a variety of analyses can be made using laboratory techniques. The rosette is configured with multiple Niskin bottles and an instrument package to measure the following variables:

- Conductivity
- Temperature
- Pressure
- Dissolved oxygen

The CTD package should include instrumentation to measure the following variables:

- Beam transmission
- Photosynthetically Available Radiation (PAR)
- Chlorophyll fluorescence

The Pioneer Array physical sampling will occur at the three surface mooring sites (Inshore, Central, and Offshore) where the broadest range of interdisciplinary measurements are made (e.g., nutrients, dissolved oxygen, pH, pCO₂, bioacoustics). In addition, samples will be taken at each deployment/recovery site for the six active gliders (which may total fewer than six sites, depending on whether multiple gliders are deployed at a single site). Glider physical sampling locations vary with glider tracks and time of recovery. Physical sampling to verify AUV instrument performance will be combined with the mooring and/or glider sampling, as appropriate; no additional sampling will be done specifically for AUVs. A high-level summary of the sampling plan is provided below.

Physical samples are obtained at Inshore, Central, and Offshore sites and at the glider deployment site(s). One cast at each mooring site is completed pre-recovery (immediately after the ship arrives onsite and before any recovery activity begins) and one cast post-deployment (after all activity at the site is complete), similar to the RSN strategy. Since glider activities at a site are expected to be of short duration, a single cast will be sufficient. Niskin bottles on the CTD rosette are tripped during the upcast. A maximum of four depths are sampled for each cast (surface mixed layer, mid-depth layer, chlorophyll maximum layer, bottom boundary layer) and two bottles per depth are tripped for replicate samples (e.g., JGOFS protocol). Thus, a minimum of eight bottles is required on the rosette. Extra bottles will also be deployed in case of tripping problems. Either a 12 or 24 bottle configuration is used to fulfill the sampling requirements.

Water samples are analyzed for (order in list follows JGOFS protocol for order of samples taken from bottles):

1. Dissolved oxygen
2. Total dissolved inorganic carbon
3. Total alkalinity
4. Nitrate (from total dissolved nitrogen analysis)
5. Chlorophyll (laboratory fluorometry or pigments via High Performance Liquid Chromatography (HPLC))

In comparing to the JGOFS protocol it is notable that laboratory salinity analysis from physical samples is not specified in the list above. This is due to the consensus of domain experts that space/time variability in the coastal ocean is typically larger than the uncertainty in a well-calibrated CTD instrument package. Thus, additional accuracy obtained from laboratory analysis is of marginal benefit. However, in situations where the calibration accuracy of the CTD package is unknown or suspect, laboratory analyses of physical samples for salinity should be done.

Glider samples are only analyzed for dissolved oxygen and chlorophyll, due to the restricted suite of sensors carried by the gliders

A computation of the number of samples and the number of laboratory analyses for each variable listed above is as follows:

Mooring sites

- 1 cast pre-recovery, 4 depths per cast, 2 samples per depth = 8 samples per site
- 1 cast post-deployment, 4 depths per cast, 2 samples per depth = 8 samples per site
- 3 sites x 16 sample per site = 48 samples (bottles) per cruise
- 5 analyses per sample x 48 samples = 240 analyses per cruise

Glider/AUV sites (maximum number of analyses)

- 1 cast, 4 depths per cast, 2 samples per depth = 8 samples per site
- 6 sites x 8 samples per site = 48 samples (bottles) per cruise
- 2 analyses per sample x 48 samples = 96 analyses per cruise

Analyses per year

- 240 x 2 mooring cruises + 96 x 6 glider cruises = 1056 analyses per year

In principle, the analysis of water samples could be done either on the ship or on shore. Given the short duration of the Pioneer Array cruises, it is planned that the samples be returned to shore and the analyses performed at an onshore laboratory. The water samples are discarded after analysis (i.e., long-term storage of the physical samples is not required.)

5.4.3 Endurance Array

Physical sampling is necessary during Endurance Array mooring turnaround cruises (2x per year, nominally in April and September), and glider deployments (possibly up to 4x per year, various locations). The purpose of the physical sampling is to provide context for pre- and post-deployment laboratory calibration of core instruments in order to better account for drifts, offsets, and non-linear sensor changes that may occur during operational deployment.

The sampling scheme utilizes a package containing a CTD-equipped rosette to obtain water column profiles and discrete water samples from which a variety of analyses are performed using laboratory techniques. The rosette package will be configured with multiple Niskin bottles and an instrument package to measure the following variables:

- Conductivity
- Temperature
- Pressure
- Dissolved oxygen

The CTD package should include instrumentation to measure the following variables:

- Beam transmission
- Photosynthetically Available Radiation (PAR)
- Chlorophyll fluorescence

The Endurance Array physical sampling will occur at the six surface mooring sites (at 25m, 80m, and 600m depths for the Washington and Oregon lines). The Endurance sites are serviced twice a year. In addition, samples are taken at the deployment/recovery sites for the six active gliders (which may total fewer than six sites, depending on whether multiple gliders are deployed at a single site). Glider physical sampling locations vary with glider tracks and time of recovery. The high-level summary of the sampling plan is:

- Physical samples obtained at 25m, 80m, and 600m mooring sites, and at glider recovery/deployment sites
- One cast at each site is completed pre-recovery (immediately after the ship arrives onsite and before any recovery activity begins) and one cast post-deployment (after all activity at the site is complete), similar to the RSN strategy. Since glider/AUV activities at a site are expected to be of short duration, a single cast will be sufficient.
- Niskin bottles on the CTD rosette to be tripped during the upcast
- Maximum of four depths for each cast: surface mixed layer, mid-depth layer (samples will be recovered from the mid-depth layer as applicable based on the site depth, at the discretion of the project scientists aboard the vessel), Chlorophyll maximum layer, bottom boundary layer
- Two bottles per depth for replicate samples (e.g., JGOFS protocol)
- Minimum of 8 bottles needed on the rosette, so sampling can be done with a 12 bottle, rather than 24 bottle frame
- Water samples from the Endurance mooring sites are analyzed for (order in list follows JGOFS protocol for order of samples taken from bottles):
 1. Dissolved oxygen
 2. Total dissolved inorganic carbon
 3. Total alkalinity
 4. Nitrate (from total dissolved nitrogen analysis)
 5. Chlorophyll (laboratory fluorometry or pigments via HPLC)
- Glider samples are only be analyzed for dissolved oxygen and chlorophyll, due to the restricted suite of sensors carried by the gliders
- In a notable departure from the JGOFS protocols, salinity is not verified through Endurance physical sampling. This is due to the consensus of domain experts that space/time variability in the coastal ocean is typically larger than the uncertainty in a well-calibrated CTD instrument package. Thus, additional accuracy obtained from laboratory analysis is of marginal benefit. However, in situations where the calibration accuracy of the CTD package is unknown or suspect, laboratory analyses of physical samples for salinity should be done.

A computation of the number of samples and the number of laboratory analyses for each variable listed above is as follows:

Mooring sites

- 1 cast pre-recovery, 4 depths per cast, 2 samples per depth = 8 samples per site
- 1 cast post-deployment, 4 depths per cast, 2 samples per depth = 8 samples per site
- 6 mooring sites x 16 samples/site = 96 samples (bottles) per cruise
- 5 analyses per sample = 480 analyses per cruise

Glider sites

- 1 cast during glider recovery/deployment, 4 depths per cast, 2 samples per depth = 8 samples per site
- 6 sites (maximum) = 48 samples (bottles) per cruise (maximum)
- 2 analyses per sample = 96 analyses per cruise (maximum)

Given the short duration of the Endurance Array cruises, it is planned that the water samples be returned to shore and the analyses be done in shore-side laboratories at Oregon State University. The water samples are discarded after analysis, i.e., long-term archiving of the physical samples is not required.

Endurance Physical Sampling Plan			
Parameter	Collection	Analysis	Analyzed by:
ChlA	Niskin bottle	Filtering, extraction & fluorometric analysis	Ricardo Letelier
DO	Niskin bottle	Winkler, etc.	CEOAS Chemical Oceanography Group
NO3	Niskin bottle	Colorimetric analysis	CEOAS Chemical Oceanography Group
Carbon (DIC, Total carbon)	Niskin bottle	Acidification, total carbon	Burke Hales
Alkalinity	Niskin bottle	Potentiometric titration	Burke Hales
Endurance Sites: (Gray's Harbor, Newport Hydrographic, and glider lines)	25 m Mooring (GH & NH, two casts, twice a year)	80 m Mooring (GH & NH, two casts, twice a year)	600 m Mooring (GH & NH, two casts, twice a year)
Sampling Depths (meters)	2	2	2
	Mid-depth*	Mid-depth*	Mid-depth
	Chlorophyll max	Chlorophyll max	Chlorophyll max
	23	78	598
480 mooring analyses per cruise x 2 cruises/year = 960 analyses			
96 glider analyses per cruise x 4 cruises/year = 384 analyses			
Maximum number of analyses per year = 960 mooring + 480 glider = 1344 analyses			
*Mid-depth samples will be recovered as applicable based on the site depth, at the discretion of the project scientists aboard the vessel			

5.4.4 Field Verification Sample Descriptions:

5.4.4.1 Oxygen (Shipboard, following modified JGOFS Protocol):

Samples for oxygen analysis for field verification of the data from and secondary calibration of the dissolved oxygen sensor are drawn from the Niskin bottles into 125 mL iodine flask bottles. The

samples are stored in a cool dark place and analyzed after waiting 6-8 hours to ensure complete reaction and that they come to room temperature but before 24 hours. Samples are analyzed shipboard. Once analysis is complete, samples are discarded.

- 5.4.4.2 pH (Shipboard, following JGOFS/GEOTRACES protocols described by Dickson, A.G., Sabine, C.L. and Christian, J.R. (Eds.) 2007. Guide to best practices for ocean CO₂ measurements. PICES Special Publication 3, 191 pp, http://cdiac.ornl.gov/oceans/Handbook_2007.html - CH4 SOP6b):

Samples for verification of the pH sensor are drawn directly into 10 cm path-length optical cells (cuvettes) sealed with polytetrafluoroethylene (Teflon®) caps and stored in a refrigerator for shipboard analyses using a spectrophotometer. Once analysis is complete, samples are discarded.

- 5.4.4.3 Alkalinity (OOI, RSN laboratory-or other, following JGOFS/GEOTRACES protocols described by [Dickson, A.G., Sabine, C.L. and Christian, J.R. \(Eds.\) 2007. Guide to best practices for ocean CO₂ measurements. PICES Special Publication 3, 191 pp; \[http://cdiac.ornl.gov/oceans/Handbook_2007.html\]\(http://cdiac.ornl.gov/oceans/Handbook_2007.html\) - CH₄ SOP1- sampling and SOP3a- Alkalinity by open cell titration\):](#)

Samples for verification of the CO₂ sensor and analyses of alkalinity are drawn from Niskin bottles into 500 mL glass bottles with greased stoppers and stored in a refrigerator until analyzed. Once analysis is complete, samples are discarded; but because unused fluids have mercuric chloride in them, they are stored safely onboard for appropriate disposal onshore according to OOI hazardous material protocols.

- 5.4.4.4 Salinity (UW Marine Chemistry Lab, following JGOFS/GEOTRACES protocols, using a Guildline Instruments Laboratory Salinometer Model 8400):

Salinity samples for verification of conductivity are collected in 125 mL polyethylene bottles, with the bottle filled to between the shoulder and the neck of the bottle but do not overfill or fill to below the shoulder. The samples are stored at ambient temperature in the dark in the R/V biolab until analysis onshore. Samples are analyzed within a week of sampling or ASAP. Unused fluids are left in the bottle to prevent salt buildup from evaporation until they are cleaned or reused. Once analysis is complete, samples are discarded.

- 5.4.4.5 Nutrients (UW Marine Chemistry Lab, following JGOFS/GEOTRACES protocols):

Samples for verification of the nitrate instrument are drawn from the Niskin bottles for onshore analyses. Storage requirements for nutrient samples (NO₃>35 µM) are those requested by the provider of the onshore analyses, UW Marine Chemistry Lab (MCL). Shipboard, nutrient samples are stored in HDPE Nalgene 60 mL bottles with ~ 45 mL fluids enclosed in each and frozen upright, except for subsamples for silica which should only be refrigerated. Once onshore, samples are delivered to the MCL, thawed, and analyzed. Thawing prevents reuse of samples for follow-on analyses and remaining samples are discarded by MCL.

- 5.4.4.6 Chlorophyll (Shipboard, following JGOFS/GEOTRACES protocols):

Chlorophyll samples for verification of fluorometers are drawn from Niskin bottles into clean brown 1000 mL polyethylene bottles and refrigerated. Samples are never stored in a refrigerator or chilled and must be filtered in a low light environment as soon as possible (See Appendix G5), with resulting GFF filters preserved by placement in a glass centrifuge tube containing 10 mL 90% acetone, tightly capped to prevent evaporation, and stored in the dark in a -20°C freezer

until shipboard analyses. Fluids and filters are discarded following analyses using approved chemical waste disposal protocols.

5.4.4.7 Robotic Vehicle:

5.4.4.7.1 Fluid Samples:

High-temperature vent and low-temperature diffuse flow fluids are collected using an ROV at the ASHES Hydrothermal Field at Axial Seamount and at methane seeps at Southern Hydrate Ridge (Figures 1 and 4). High-temperature vent fluids are defined as particulate-laden fluids issuing directly out of chimney orifices. At Axial Seamount, the high-temperature fluids issuing from black smoker chimneys are generally $> 250^{\circ}\text{C}$. Low-temperature diffuse flow fluids are sulfide particle-free, low-temperature (generally < 80 to 150°C) fluids issuing directly from cracks in the basaltic seafloor. These sites are typically marked by various assemblages of white, filamentous bacteria, with lesser amounts of tubeworms and rare clams (Figure 4).

Samples are taken with the ROV manipulator using gas-tight titanium bottles (Figure 5). This is required for discrete volatile analyses for field verification of the mass spectrometric data of dissolved gas concentrations, and for verification of some high-temperature major and trace element chemistry associated with the *in situ* remote access sampler (RASFL) (Figure 6). Two gas-tight bottles are used for sampling each site with vent temperatures measured prior to and/or during sampling. The sampling sites are imaged prior to and after sampling using high definition (HD) and still cameras mounted on the ROV. The temperatures attained with the temperature probe are logged.

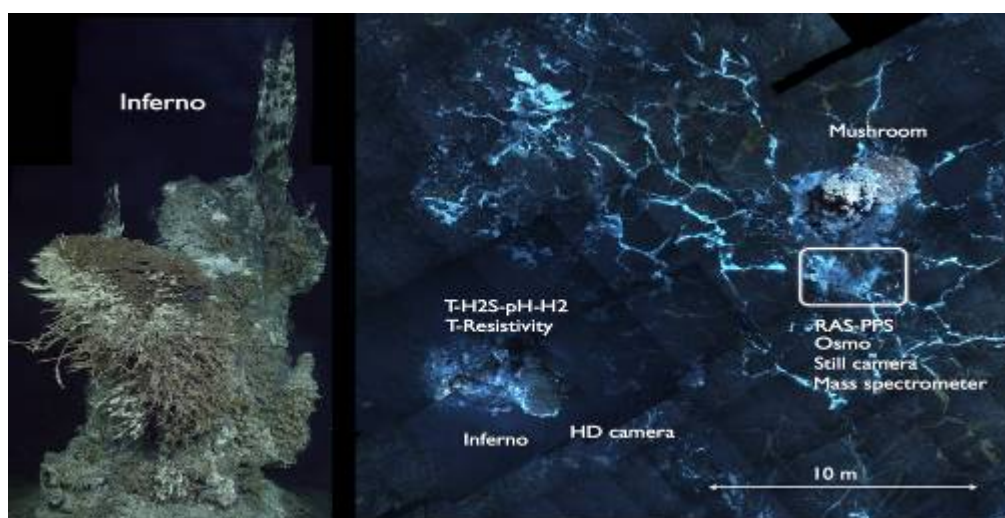


Figure 4. Photomosaic of the high-temperature smoker Inferno (left) in the ASHES hydrothermal field. The structure rises 4 m above the seafloor and hosts numerous active orifices. The image to the right is a down-looking photomosaic of Inferno and an adjacent smoker called Mushroom. Also shown is the location of sensors to be placed in orifices on Inferno, and at an active diffuse flow site located ~ 8 m to the northeast.

Vent, diffuse, and seep fluids are sampled from the titanium gas-tight bottles immediately following recovery of the ROV on deck. A split of the sample is used for volatile analyses via shipboard gas chromatography (methane, hydrogen, carbon dioxide), while major and trace element chemistry are done in an onshore laboratory using HPLC, ion-chromatography (IC), and inductively-coupled plasma mass spectrometry (ICP-MS). Depending on the analysis, fluids are stored in glass evacuated “vacutainers” or HDPE Nalgene bottles, either at room temperature, +2°C or -20°C. A description of analytical procedures for each measurement type is included in Appendix G.



Figure 5. Example of gas-tight fluid sampler acquiring diffuse flow fluids from the Lost City Hydrothermal Field with the robotic vehicle Hercules.

Table 4: Gas-Tight Fluid Samples

1.1.1 Sensor	Measurement	Volume	Onshore/ Shipboard
Verification/Calibration			
Osmo Sampler (OSMOI)	Majors, volatiles	150 mL	onshore [†]
High Temperature, H ₂ S, pH, H ₂ sensor	Majors, volatiles	150 mL	onshore [†]
Black smoker temperature-resistivity (chlorinity)	Majors, volatiles	150 mL	onshore [†]
Remote Access Sampler (RASFL)	Majors	150 mL	onshore
Mass Spectrometer	Volatiles	150 mL	shipboard [†]

[†]Some volatile species are analyzed shipboard using a GC – e.g. methane, hydrogen, carbon dioxide.

5.4.4.8 Samples Recovered by CORE RSN Sensors:

There are three core RSN seafloor instrument types that recover fluids for follow-on onshore analyses (Table 5). These three instruments include a Remote Access Fluid sampler (RASFL, see Figure 6), located in the ASHES Hydrothermal Field; two Osmo samplers (OSMOI), one each located in ASHES and at the Southern Hydrate Ridge site; and a Benthic Fluid Flow instrument (FLOBN), located at Southern Hydrate Ridge.

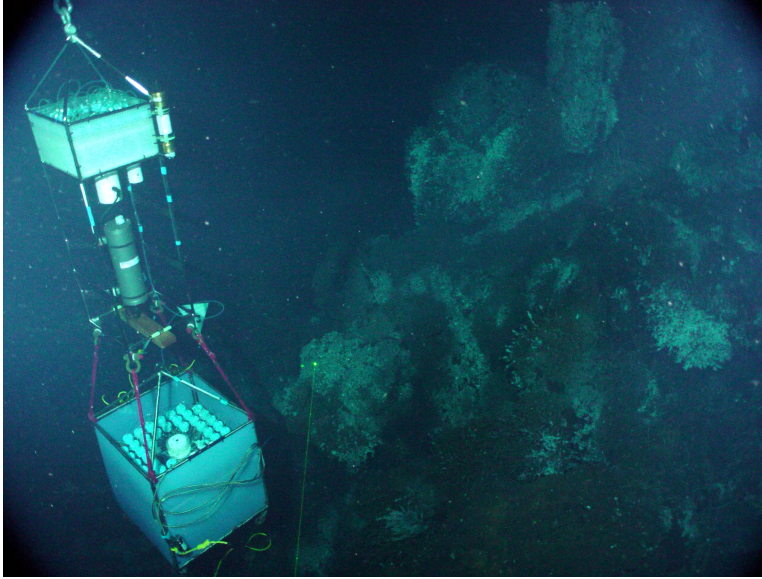


Figure 6. Example of a Remote Access Sampler and DNA Sampler at the black smoker called Hulk in the Endeavour Segment of the Juan de Fuca Ridge.

In addition to these fluid samplers, a Phytoplankton Particulate Sampler (PPSDN) filters and preserves particles 0.2µm to 500µm in size from diffuse fluids in the ASHES Hydrothermal Field (Figure 6) for laboratory-based genetic analysis.

Table 5: RSN CORE Instrument Samples

Sensor Verification/Calibration	Measurement	Volume	Onshore/ Shipboard
Osmo Sampler (OSMOI)	Major-Trace Fluid Chemistry	0-1 mL	onshore
Remote Access Sampler (RASFL)	Major-Trace Fluid Chemistry	800 mL	Onshore & shipboard
Phytoplankton Particulate DNA Sampler (PPSDN)	16S rRNA	0-100 µg	onshore
Benthic Fluid Flow (FLOBN)	Tracer and Major Ion Fluid Chemistry	0-0.35 mL	onshore

5.4.4.8.1 Remote Access Fluid Sampler (RASFL) (Butterfield 1997; 2004):

This instrument collects 48, 800 mL fluid samples from a diffuse flow site in the ASHES Hydrothermal Field (Figures 4 and 6). The RASFL consists of a nozzle, placed within a site of diffuse flow, and a pump, which draws fluids into sterile collapsible plastic bags housed inside rigid plastic containers. The instrument also monitors temperature at the intake nozzle continuously. Sampling ports and schedules are controllable through an RS-232 port allowing remote control from shore through the cabled network. The instrument package is recovered annually and samples processed shipboard. Aliquots of fluids are subsampled for shipboard analyses of pH, alkalinity, hydrogen sulfide, dissolved silica, and ammonia. Extracted water (acidified with sulfamic acid) is analyzed onshore for major, minor and trace elements at the Pacific Marine Environmental Laboratory (PMEL) and UW. Once analysis is complete, samples are discarded.

5.4.4.8.2 Osmo Sampler (OSMOI):

This instrument collects low-temperature diffuse flow fluids and seep fluids at the ASHES Hydrothermal Field and Southern Hydrate Ridge, respectively (Figure 7). The sampler collects osmotically-pumped fluids into small bore (0.8 mm I.D.) Teflon® tubing initially filled with distilled water. The tubing is coiled within small canisters and fluids are drawn in through a small nozzle placed at the point of sampling.



Figure 7. Two loaded Osmo (OSMOI) Sampler canisters are attached to a data logger and microbial incubator awaiting deployment in a black smoker chimney on the Endeavour Segment of the Juan de Fuca Ridge. In this example, the inlets are housed within the barrel of the incubator.

These instruments are recovered annually during O&M cruises. Upon recovery, the tubing is removed from the canisters and the Teflon tubing is cut into 1-2 m long sections with the fluid expelled from each section into microcentrifuge tubes. The tubes are stored in a refrigerator for follow-on onshore analyses with analytical protocols similar to those used for processing of diffuse flow fluids. Once analysis is complete, samples are discarded.

5.4.4.8.3 Benthic Fluid Flow Instrument (FLOBN):

This instrument injects a chemical tracer into seawater and/or pore fluids near the seabed and continuously collects the same fluids using osmotic pumps and narrow-bore Teflon tubes. The tubes containing the fluids are appropriately partitioned and the collected fluids are preserved until shore-based analysis for tracer and major ion concentrations. The tracer distributions and depth profiles of the major ions in the sediments will enable the determination of benthic fluid flow rates for diffuse fluids emanating from the seabed of Southern Hydrate Ridge.

5.4.4.8.4 *Phytoplankton Particulate Sample (PPSDN):*

This instrument collects 24 filtered and fixed (preserved) samples for onshore analyses of 16S rRNA. The instrument operates in a similar manner as the RASFL, but fluids are drawn through stacked 500 μm and 0.2 μm filters for particulate DNA collection. A reservoir of Ambion RNAlater® allows injection of preservative for sample fixation. This instrument is coupled to the RAS in a manner similar to that shown in Figure 6, allowing co-registered temperatures, fluids, and DNA to be collected. The PPSDN is recovered annually at the same time as the RAS is recovered. The filters are removed and stored in sterile Falcon Tube-type containers and placed immediately within a -20°C freezer for onshore analyses. Samples are transferred to UW and stored in a -20°C freezer in MSB. Once analysis is complete, samples are discarded.

5.4.5 Global Arrays

The following water samples are taken on the maintenance cruises of the global arrays (Station Papa, Irminger Sea, Southern Ocean, Argentine Basin) for the purpose of calibration and verification of the OOI instruments on the moorings and gliders. Each sample type is understood as being collected with a rosette system attached to a lowered CTD.

- **Salinity.** Purpose: verification of moored and glider CTD instruments.
- **Dissolved oxygen.** Purpose: verification of moored and glider oxygen instruments.
- **Nitrite/nitrate.** Purpose: verification of moored nutrient instruments.
- **Chlorophyll.** Purpose: verification of moored and glider fluorescence/turbidity instruments.
- **Total dissolved inorganic carbon & alkalinity (TDIC&Alk).** Purpose: verification of moored pCO_2 and pH instruments.

The protocols for collecting and analyzing these water samples are standard industry practice, and are outlined in [reference to be provided].

5.5 Cruise Documentation, Metadata, and Data Distribution

Rigorous and thorough documentation of all aspects of cruise operations are critical to insure that:

- installation requirements are met daily, as well as for a given field season;
- problems with infrastructure or deployments can be dealt with in a timely manner;
- the IO and non-IO communities are well-informed of at-sea operations.

This document only provides suggested guidance for metadata associated with physical samples collected as part of the OOI. Table 6 summarizes metadata that should be collected routinely during cruise operations and onshore analyses.

5.6 Cruise Documentation

The OOI maintains a digital daily log of all shipboard operations in a Daily Operations Science and Engineering Report. These reports form the foundation of a Cruise Report which will include additional operational details such as comprehensive final site layouts with appropriate maps, dive track lines with post-cruise corrected navigation etc.

During each OOI cruise, the cruise report includes a section summarizing the physical samples taken on the cruise in support of OOI, associated metadata (see Section 5.7.1), and results of shipboard and onshore analyses in support of OOI. Hard-copy sample sheets are transferred to digital format, made available to CI to allow OOI data user access and the hard copies delivered to the cruise chief scientist for disposal or retention at their discretion. Hard copies are not retained by the OOI. A more thorough development of what should be included in OOI cruise reports will be presented in follow-on operational documents. Metadata are consistent and complete for each of the OOI cruises and include Event Logs, Station Logs, and Sampling Logs. This information will initially be provided as Excel spreadsheets that are updated daily, and included in daily operations reports (engineering and science). These documents are checked and verified by the responsible logger as well as by the Data Manager shipboard. Using the NSF-funded Marine Geoscience Data System portal as an example, the following types of metadata forms are completed as part of the cruise reports:

- Metadata: Cruise metadata are analogous to those in the Marine Geoscience Data System (<http://www.marine-geo.org/>)(MGDS) and to GLOBEC. The MGDS forms include:
 - MGDS_M01_1CruiseInfo – Summary of cruise including Funded Projects, PI's, start/end dates and ports
 - MGDS_M01_2CruisePersonnel – List of cruise participants: Name, Role, Institution, e-mail
 - MGDS_M01_3DataSummary – Summary of instruments used and data types produced during the cruise: Field Data Type (photographs, navigation, sample-biology, rock, fluid), temperature, bathymetry, navigation, CTD etc.; Device (camera, CTD, magnetometer), Device make, Platform (ROV, Ship)
 - MGDS_M01_4_TetheredInstrumentSummary - Basic information about deployment and recovery of tethered instruments including CTDs, Towed Nets, Towed Camera Systems (excluding ROVs)
 - MGDS_M01_5_LaunchSummary - Basic information about dives that includes platform name, start date, start-end location
 - MGDS_M01_6_DeployedInstrumentSummary -For reporting information about instrument deployed and recovered during a single

- expedition: Device make, model, station type, deployment-recovery location, depth, vehicle heading, altitude, etc.
- MGDS_M01_7_Long-Term DeployedInstrumentSummary -For reporting information about instrument deployed and recovered during multiple expeditions.
 - MGDS_M01_8_Ship-BasedSamples - For all samples acquired from the ship, includes samples acquired with tethered instruments such as CTDs etc. sample location, wire out etc.
 - MGDS_M01_9_DiveSamples - For all samples acquired with a dive platform (HOV, ROV, and AUV): Dive platform type, dive number, sample type, device type, date, time, location etc.
 - MGDS_M01_10_DataFiles - For any data files including grids, images, EXCEL spreadsheets, shapefiles; cruiseID, file name, format, data type, device type, start-stop dates, location, coordinate type, X resolution, Y resolution etc.
 - MGDS_M01_11_Configurations - Catalog sensor details with positions, serial numbers etc.
 - MGDS_M01_12_Vocabularies

5.6.1 Underway Ship Data (e.g. CTD):

Routine underway ship data from US research vessels are provided directly to a data center, e.g. the Rolling Deck to Repository (R2R) (<http://www.rvdata.us/>). These data can provide information that is important for the interpretation of OOI core data (e.g. for validation purposes). The OOI will ingest, redistribute, or link to such data if they are relevant to the OOI core data. To the extent that additional ship and science activities are deemed part of the OOI project, their generated data/metadata will also be transferred to OOI systems, as described in the following section.

5.6.2 Event Logs:

Cruise Event Logs provide an overall chronological summary of activities that occur on the cruise. For OOI cruises, these activities include OOI specific ROV dives, deployment and recovery of OOI infrastructure from the ship (e.g., moorings), and OOI hydrocasts supporting product calibration and verification. Events also include suspension and resumption of operations. A representative set of fields is presented in Table 6, and builds from the Data Management Guidelines Manual (V1.0 2008) from the Biological and Chemical Oceanography Data Management Office (BCODMO: <http://bcodmo.org/>) funded by NSF. Fields that are important to include in the Event Log are shown below:

Table 6. Event Log Metadata

Field Name	Description
eventEvent	The unique event number identifier for each sampling event (start and end). Example: shipCRYMMDD.hh.mm where ship = ship abbreviation CR = cruise number YY = last 2 digits of year MM = the 2 digit month DD = the two digit day hh = the two digit hour mm = the two digit minute
instrument	Instrument name (CTD)
station	Station number
cast	Cast number
dive	Dive number
ev_type	Event type descriptor (e.g. a combination of instrument and cast number)
date and time (YYYYMMDD in GMT expressed as hhmm)	Date as YearMonthDay the measurement was taken (8 digits as YYYYMMDD) Year as YYYY, Month as MM (01-12) and Day as DD (01-31); January 1, 2008 would be 20080101. Time (24 hour clock, as hhmm with hh being the two digit hour and mm being the two digit minutes)
time_local	Local time as hhmm
time_zone	UTC -08, or PST
s_e	Start/end flag of the activity.
latitude	Positive values = northern hemisphere, (required format is decimal degrees) Format is ddd.dddd for decimal degrees
longitude	Positive values = est. of the prime meridian, (required format is decimal degrees)
water_depth	Water depth in meters
cast_depth	Cast depth in meters
Investigator	Name of person responsible for sampling the event
custom program-specific fields	Standard station of sampling grid identifier
comments	Optional comments

Example Event Log*

Cruise # and Vessel

Event	Date	GMT	Time-L	TZ	station	longitude	latitude	instrument	event type
TN_268_1108172012	20110817	2000	1300	-7	1	49.93619	130.01417	CTD	CTD002
TN_268_1108172012	20110820	1900	2000	-7	2	49.93619	130.01417	ROPOS	R217

*investigator is not included in this example to conserve space; CTD = hydrocast, ROPOS = ROV, R217 = ROPOS Dive 217

5.6.3 Station Logs:

Station logs are kept for each station. They include information relating to the position and time of the sampling station, ROV operation (or bathymetric mapping where appropriate). Station logs

also include the weather conditions and other details important to sampling. The following information is included:

Station Latitude and Longitude (units as above)
Station Date (yyyymmdd)
Station Time (designated as "GMT/UTC" or local)
Station Identifier
Meteorological Observations
Station Sounding Bottom Depth

5.6.4 Sample Logs:

Sample logs are kept as running chronological logs to include all samples taken during a given day, but also as individual logs that may contain additional information (e.g., images of rock samples; images of chimney prior and after sampling; or water temperature, pressure, salinity, nutrients required for calculation of inorganic carbon chemistry). This Sample Log example closely follows that developed for the NSF-funded Marine Geoscience Data System (MGDS - <http://www.marine-geo.org/index.php>). The following information is included in running chronological logs:

	Example	Comments
Sampling Platform	ROV	ROV = Remotely operated vehicle
Platform Name	ROPOS	ROV = ROPOS
Dive or Station #	R217	Dive 17
Device Type	Niskin, Manipulator	Collected with ROPOS manipulator.
Sample Name	R217_1108_2017	Dive number, month, day and time
Latitude	-130.01417	
Longitude	49.93619	
Platform depth during sampling	1507 m	
Seafloor Depth	1507 m	
Navigation Type	USBL	Ultra short baseline
Vehicle heading	020	
Local X(m)	4023	Vehicles may set up their own origin
Local Y (m)	5045	
Local Origin Latitude	-130.01417	
Local Origin Longitude	49.93619	
Sample Repository	UW	University of Washington
IGSN #	OOIUW1245	System for Earth Sample Registration #
Describer	Jane Doe	Person describing and archiving sample
Details	Sulfide sample from ASHES vent field containing coarse-grained chalcopyrite, fine-grained sphalerite and anhyrite	
Rock Sample Classification	Hydrothermal	
Rock Description	Sulfide	
Expected data type	chemistry-rock	
Preservation type	frozen	

For hydrocasts, this additional information should be logged:

Start time
Start Latitude
Start Longitude
Target Sample Depth
Actual Sample Depth
Stop Time
Stop Latitude
Stop Longitude
Wire out (m)

Sample Bottle
Sample type (e.g. salinity, chlorophyll-a, nutrients)

In addition to the cruise sampling scheme adopted by the OOI, archived samples from the OSMOI, RASFL and PPSDN will also be registered with the System For Earth Sample Registration (SESAR) (<http://www.geosamples.org/>) providing a unique international nine-digit sample number.

5.6.5 Integrated Observatory Network (ION)

As indicated in the OOI Data Management Plan (Document Control Number 1102-00000), OOI data/products are managed by the OOI Program Office in cooperation with the IOs. The first step in this management is achieved by transmitting the cruise data, and any other appropriate ship and science data, to the Integrated Observatory Network, either by leveraging planned R2R communications and databases or through independent transmission. Wherever possible, the transferred data are delivered to the ION as quickly as possible, including in near real time when technologies allow.

All cruise data and metadata (including from scientific instruments), once ingested into the CI, are made immediately available to users of the OOI (with the exception of data for which a specific exception has been granted, per the OOI Data Policy). For RSN core instruments that collect physical samples, the data will not be transferred through ION but instead be transferred to CI after final analysis at the shore site.

The processes and policies for transforming the cruise-derived data into formal OOI data products are determined by the OOI Program. The ION network facilitates the implementation of these processes and policies. In most cases, the network operates workflows to carry out the processes, according to the policies, as soon as the data are available.

6 Secondary Calibration and Data Adjustments

6.1 Purpose and Objectives

The primary calibrations initially applied in the process of generating a Level 1 data product may not be optimal for the entire deployment period and/or may contain other errors. In addition, information may become available to improve data accuracy after the primary calibrations have occurred. Field verification is the mechanism by which OOI determines whether data values need to be corrected via secondary calibrations and data adjustments. These efforts are driven by the nominal accuracies of sensors, instruments, and data products as specified in the OOI requirements and by the COTS instrument vendors.

Secondary calibration and other data adjustments may change with time and are the key mechanisms to account for sensor drift. The application of any of these adjustments happens entirely within the shore-side cyberinfrastructure, as opposed to onboard the marine platforms, and the adjustments will result in updated versions of data products that are fully traceable back to the originating raw data and all intermediate processing steps. The secondary calibration equation shall be added to the metadata. For most data products, complex sensor response functions are addressed by the primary calibration equations; and, therefore, the secondary calibration equations are typically conceptually and computationally simpler (e.g., changes in gain and offset).

6.2 Source of Calibration Information

The source of the secondary calibration data or adjustment can be any of the following:

- test of instrument calibration, e.g., post-recovery, at manufacturer's facility;
- test of instrument intercalibration of all instruments deployed, e.g., post-recovery, at a facility of an OOI implementing organization (or subcontracted facility);
- shipboard, or otherwise *in-situ*, measurements made during an OOI cruise, e.g., physical samples;
- *in situ* calibration process on deployed instruments;
- other sources of information gathered through Field Verification of instrument measurements such as intercomparison of instruments;

The rationale and general approach of the Field Verification process is described in Section 4 with additional instrument-specific details provided in Appendix C.

Table C-1 in Appendix C lists when and how secondary calibration information is obtained for each core instrument on the system, by reference designator.

7 Data Quality Assessment and Flagging

The quality of OOI data is assessed by a combination of automated algorithms and visual inspection of the automated output (flags) by a SME. In both cases, the data values themselves are not modified. Instead, the assessment produces annotations to the data that may be either granular for individual data points, or aggregated for multiple data points such as subsets of data or even the entire dataset from one deployment.

The ultimate goal of data quality assessment is to determine whether data achieve the specified nominal accuracy or, if not, the degree of degradation. However, many tests only detect certain known and gross failure modes, the absence of which may not be sufficient to confirm high accuracies. Therefore, data quality assessment should be considered an informed estimate.

7.1 Automated Algorithms

The OOI uses a library of automated algorithm tests that assess data quality. This list is likely to expand in the future, but initially they are:

1. Global and Local range (DCNs 1341-10004 and 1342-10005, respectively:)
2. Spike (DCN: 1341-10006)
3. Stuck Value (DCN: 1341-10008)
4. Trend (DCN: 1341-10007)
5. Temporal and Spatial Gradient (DCN: 1341-10010)

Other QC-related algorithms include:

1. Polynomial Value (DCN: 1341-10003)
2. Interpolation Value (DCN: 1341-10002)
3. Modulus Value (DCN: 1341-10001)
4. Solar Elevation. (DCN: 1341-10011)

These are described in individual QC specification documents, which should be consulted for reference on the detailed descriptions of each algorithm and how they work. QC tests usually produce pass/fail flags for the tests that they perform on the data. Data product flow charts for

each data product identify which automated algorithms are applied against the data product at Levels 1 and 2. By definition Level 0 data products have no QA/QC applied and referred to as quality control level “a” in Section 1.6 above. Combination of the flags from multiple automated QC algorithms into one merged assessment allows for a best-guess estimate of data quality based on automated algorithms alone, and constitutes quality control level “b” of an OOI data product.

7.2 Data Quality Flags

OOI will assign flags to data that indicate data quality. Metadata documents which data points are being described, and what the flags mean. It is possible for a data point to have multiple flags, e.g. resulting from multiple QC algorithms being performed on it.

7.3 Visual Inspection by SME

If data products require human-in-the-loop (HITL) QA/QC, they must be reviewed by SMEs, subject to budget and operational constraints. The data product flow diagrams described above denote when this inspection occurs during the data QA/QC adjustment. The inspection results in any of the following:

- approval of data and flagging
- additional annotation to data (e.g., specify different accuracy, descriptions of quality of subsets of data)
- identification of a need to adjust data and documentation of why
- identify person who authorized data adjustment
- data adjustment (subject to scope and budget)

Review and either approval, or proper annotation and correction (subject to scope and budget), of data constitutes quality control level “c” of an OOI data product.

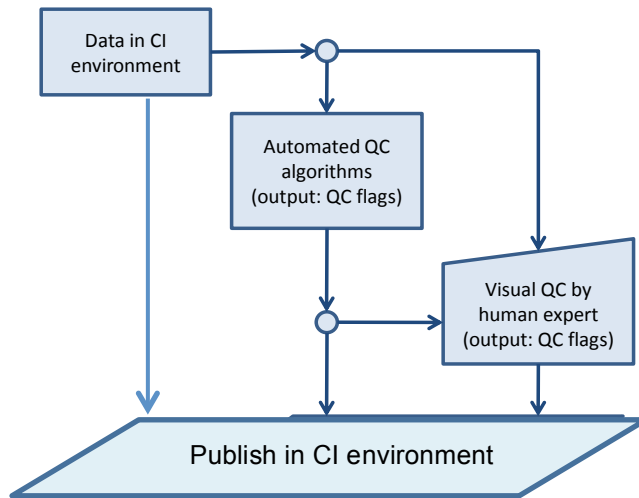
Prior to OOI Release 3, the OOI may develop statistics on the performance of the automated QC tests vs. human-in-the-loop (HITL) visual inspection, subject to scope, budget and time. The need for specialized, or more sophisticated, QC algorithm development and the amount of HITL visual inspection needed to deliver high quality level 1 and 2 data products will be evaluated, subject to scope and budget.

Prior to the first operational buoy deployment (but after glider deployment), the OOI O&M group (with collaboration from construction) identifies initial SME's for the appropriate data streams and the data products requiring HITL QA/QC assessment. The OOI tasks these SMEs to develop data product-specific procedures for HITL assessment that are delivered to the O&M team prior to the first deployment of each array. O&M implements these procedures in their Standard Operating Procedures along with a process for their evolution as the OOI program matures. The amount of HITL implemented in O&M will be constrained by balancing scientific community needs with available budget.

Appendix F contains checklists for the SME review, both generic and instrument-specific. These serve as a guideline for the visual inspection process and development of the data product-specific HITL assessment procedures mentioned above.

7.4 Data flow during QC process

Data QC occurs within ION (i.e., after the data have been transferred from the initial shore station into the cyberinfrastructure.) Automated algorithms run in ION, and visual inspection is performed through an ION interface. The following drawing visualizes the concept:



ION has the capability of associating quality flags and metadata that are created through quality control processes with data products.

8 Refining Global and Local Ranges

Acceptable data ranges for OOI deployments of instruments are dependent on the measured variable, depth, and instrument location. The initial acceptable ranges have been determined based upon historical data as recorded in data repositories and primary scientific literature and/or scientific judgment. Acceptable ranges are refined as more data are collected during the planned lifetime of OOI. These ranges as well as all automated tests, as described in Section 7.1, are contained in ION look-up tables. These tables are maintained under configuration management.

9 References

- Butterfield D.A., I.R. Jonasson, G.J. Massoth, R.A. Feely, K.K. Roe, R.E Embley, J.F. Holden, R.E. McDuff, M.D. Lilley, and J.R. Delaney (1997) Seafloor eruptions and evolution of hydrothermal fluid chemistry. *Phil. Trans. Royal Soc. London A.*, 355, 369-386.
- Butterfield., D.A., K.K. Roe., M.D. Lilley, J.A Huber, J.A Baross, R.W. Embley, and G.J. Massoth (2004) Mixing, reaction and microbial activity in the sub-seafloor revealed by temporal and

spatial variations in diffuse flow vents at Axial Volcano. In *The Subseafloor Biosphere at Mid-Ocean Ridges*. (eds) W.S.D. Wilcock, E.F. Delong, D.S. Kelley, J.A. Baross, and S.C. Cary. Geophysical Monograph 144, Amer. Geophys. Union, Washington, DC, 269-303.

COL-NASA Data QC Workshop Final Report. E. Boss, M.B. Neely, J. Werdell. 2012. 26 pp.

Dickson, A.G., Sabine, C.L. and Christian, J.R. (Eds.) (2007) *Guide to best practices for ocean CO₂ measurements*. PICES Special Publication 3, 191 pp.

Appendix A: Examples of Existing Community Standards and Practices

There are many existing instrument-specific and general methods for data QA/QC and supporting data formats in oceanography, many of which have been established for decades. This broad spectrum of existing standards and formats provides ample opportunity for incorporation and/or adaptation of accepted protocols into OOI QA/QC procedures and reduces the need to develop new ones. Nevertheless, the selection of the most appropriate procedures acceptable to the scientific community OOI will be an ongoing effort during the early stages of the OOI data QA/QC development. The following sections describe some of the existing community standards and data formats and comment on their relevance to the OOI. OOI metadata standards are described in the Metadata Approach Document.

Argo

Overview: Argo is a global array of profiling floats. Floats are autonomous instruments that drift passively under water. Argo floats periodically come to the surface, measuring CTD profiles (and possibly additional data) during the ascent and/or descent. Argo floats are expendable; hence the sensors receive no post-mission calibration. In addition to simple QC algorithms, Argo has developed sophisticated QC algorithms to detect sensor drift through comparison with an evolving climatology.

Relevance to OOI: Some of Argo's simple data QC algorithms can be adapted easily to mooring and glider applications as used by the OOI and form the basis of many algorithms described in this document. The OOI data categories are likely to follow a model similar to Argo, which distinguishes between "real-time" and "delayed-mode" data. Argo's data format (netCDF with standardized naming and metadata conventions) is an example of data formats relevant to OOI.

References:

- Internet, general:
 - <http://www.argo.ucsd.edu>
 - <http://wo.jcommops.org/cgi-bin/WebObjects/Argo>
 - <http://www.argodatamgt.org/>
- Argo data management, User's Manual:
 - <http://www.usgoda.gov/argo/argo-dm-user-manual.pdf>
- Argo data management, Argo quality control manual:
 - <http://www.argodatamgt.org/content/download/341/2650/file/argo-quality-control-manual.pdf>

NDBC

Overview: NOAA's National Data Buoy Center (NDBC) operates the network of weather buoys and coastal weather stations around the country, as well as buoys and bottom platforms in the Tropical Atmosphere Ocean (TAO) and Deep-ocean Assessment and Reporting of Tsunamis (DART) arrays.

Relevance to OOI: NDBC operates many platforms similar to OOI platforms. NDBC's operational routines are likely relevant role models to OOI data operations. NDBC has developed QC procedures that are applicable to OOI platforms as well, particularly for meteorological, wave, and acoustic Doppler current profiler (ADCP) data.

References:

- Internet: <http://www.ndbc.noaa.gov>
- *Handbook of Automated Data Quality Control Checks and Procedures*. NDBC Technical Document 09-02, National Data Buoy Center, Stennis Space Center, Mississippi 39529-6000, August 2009.

NODC

Overview: NOAA's National Oceanographic Data Center (NODC) is the national permanent archive for oceanographic data.

Relevance to OOI: It is likely that NODC will play a role in the archiving of OOI data. Data formats used by NODC are relevant to OOI.

References: Internet: <http://www.nodc.noaa.gov>

OceanSITES

Overview: OceanSITES is an international collaboration to bring together data from open-ocean, long-term reference stations. Investigators contribute their data into an open-access system, following well documented procedures and formats.

Relevance to OOI: The global-scale nodes (GSN) data may become part of OceanSITES. Data format and naming conventions, including metadata, are relevant to OOI. The OceanSITES QC flag definitions are a candidate for the OOI. OceanSITES is developing documents describing data QA, including calibration, Field Verification procedures, and data flow.

References:

- Internet:
 - <http://www.oceansites.org>
- OceanSITES User's Manual

QARTOD

Overview: QARTOD (Quality Assurance of Real-Time Oceanographic Data) is an effort to address the Quality Assurance and Quality Control issues of the Integrated Ocean Observing System (IOOS) and broader international community. QARTOD has convened a number of meetings, each discussing specific instruments and procedures.

Relevance to OOI: QA/QC of oceanographic data is the topic of this document (QARTOD meeting minutes contain relevant information). While OOI's and IOOS missions are distinctly different, the complementary nature of the programs and similar requirements for high quality data highlight the relevance of QARTOD developments.

References: Internet: <http://www.qartod.org>

GO-SHIP

Overview: The Global Ocean Ship-based Hydrographic Investigations Program (GO-SHIP) is an international effort to coordinate ship-based oceanographic expeditions with a multi-disciplinary focus. GO-SHIP continues hydrographic observations following the examples set forth e.g. by WOCE (<http://woce.nodc.noaa.gov/wdiu/wocedocs/>) and JGOFS.

Relevance to OOI: OOI will conduct multidisciplinary data-collecting cruises on research ships. GO-SHIP has a comprehensive collection of documents about ship-based measurements, including data QA, calibration, and best-practice handling of equipment and data.

References: Internet: <http://www.go-ship.org>

Appendix B: SME assignments, instrument characteristics, and required references

This section includes SME assignments and HITL models, calibration center assignments, descriptions of the basic instrument characteristics, including typical cases of malfunction and recommended calibration equations, and required references for Operations and Maintenance.

This section is a “work in progress” as instrument procurements have not been completed as of this version. The Confluence ‘ICD’ pages for each instrument and eventually the “Instrument Application” will replace most aspects of this appendix and as operational details are finalized this can be updated in the next version to show the SME and Calibration locations and POCs for the instruments.

Instrument Class ADCPS (Velocity profile, 500-700 m range)

Model 1 Subject Matter Expert:

Calibration Center:

Manufacturer, make & model: Teledyne RDI 75 kHz Workhorse Long Ranger

General description: Workhorse Long Ranger ADCP measures current structure.

Calibration Information: Only adjustments of the following are recommended:

- Magnetic variation: rotate data by an additional angle α :
 - If data are given as current speed s and direction d :
$$d_{\text{new}} = d_{\text{old}} + \alpha$$
 - If data are given in eastward and northward components of velocity, u and v :
$$u_{\text{new}} = u_{\text{old}} * \cos(\alpha) - v_{\text{old}} * \sin(\alpha)$$
$$v_{\text{new}} = u_{\text{old}} * \sin(\alpha) + v_{\text{old}} * \cos(\alpha)$$
- Sound speed correction for vertical distance from transducer:
 - The vertical distance z of each bin from the transducer head depends on the actual sound speed c . The instrument usually uses a default value, c_0 . The following equation corrects z accordingly:
$$z_{\text{new}} = z_{\text{old}} * (c / c_0)$$

Typical Signal Shapes:

Typical Failure Modes:

External Sources of Documentation:

- <http://www.rdinstruments.com/longranger.aspx>

Instrument Class CTDMO (CTD used in surface and subsurface moorings)

Model 1 Subject Matter Expert:

Calibration Center:

Manufacturer, make & model: Sea-Bird Electronics SBE-37

General description: These instruments measure temperature, conductivity, and pressure.

Calibration Information:

Internal equations are used to convert data in raw counts to physical units. Once in physical units, the following equations describe how to correct the data if such corrections are needed (cf. manufacturer's “application note” 31 at link below):

- Temperature data should be corrected using an additive constant a :
$$T_{\text{new}} = T_{\text{old}} + a$$
- Conductivity data should be corrected using a multiplicative factor m :
$$C_{\text{new}} = m * C_{\text{old}}$$
- Pressure data should be corrected using both a multiplicative factor m and an additive constant a :
$$P_{\text{new}} = m * P_{\text{old}} + a$$

Conductivity data are computed using pressure data (to compensate mechanical deformation of the sensor under stress). For highest accuracy, it may be necessary to re-apply this computation to conductivity if pressure corrections are applied (cf. manufacturer's "application note" 10 at link below).

Typical Signal Shapes:

Typical Failure Modes:

External Sources of Documentation:

- http://seabird.com/products/spec_sheets/37imdata.htm
- http://seabird.com/application_notes/AN31.htm
- http://seabird.com/application_notes/AN10.htm

Instrument Class FLOR (3-wavelength fluorometer for measurement of fluorescence, Chl-a, CDOM, and backscatter)

Model 2 Subject Matter Expert:

Calibration Center:

Manufacturer, make & model: WETLabs ECO triplet-w

General description: ECO triplet-w measures optical backscattering at 117 degrees at both 470 nm and 700 nm in addition to chlorophyll fluorescence.

Calibration Information:

Typical Signal Shapes:

Typical Failure Modes:

External Sources of Documentation:

- <http://www.wetlabs.com/products/eflcombo/triplet.htm>

Instrument Class PHSEN (pH)

Model 2 Subject Matter Expert:

Calibration Center:

Manufacturer, make & model: Sunburst SAMI²-pH

General description: Sunburst SAMI²-pH (Submersible Autonomous Moored Instrument–pH) measures pH, and total alkalinity. Calibration Information:

Typical Signal Shapes:

Typical Failure Modes:

External Sources of Documentation:

- <http://www.sunburstsensors.com/>

Instrument Class FDCHP (Direct Covariance Fluxes, High Power)

Model 2 Subject Matter Expert:

Calibration Center:

Manufacturer, make & model: WHOI HP-DCFS

General description: WHOI DCFS (Direct Covariance Flux System) consists of a Solent three-axis sonic anemometer-thermometer and a Systron Donner MotionPAK. The sonic anemometer records wind velocity to derive direct estimates of the wind vector stress using the eddy correlation technique. The MotionPAK corrects the effects of platform motion.

Calibration Information:

Typical Signal Shapes:

Typical Failure Modes:

External Sources of Documentation:

- <http://uop.whoi.edu/projects/stratus/docs/Stratus3Cruise.pdf>

Instrument Class METBK (Bulk Meteorology)

Model 1 Subject Matter Expert:

Calibration Center:

Manufacturer, make & model: Star Engineering ASIMET

General description: The ASIMET suite consists of seven models; barometric pressure, relative humidity and air temperature, precipitation, shortwave radiation, RM Young wind speed and direction, Gill sonic wind speed and direction, long wave radiation.

Calibration Information:

Typical Signal Shapes:

Typical Failure Modes:

External Sources of Documentation:

- <http://frodo.who.edu/asimet/>

Instrument Class PCO2A (Delta pCO₂ air/sea)

Model 2 Subject Matter Expert:

Calibration Center:

Manufacturer, make & model: Pro-Oceanus CO₂ Pro

General description: Sunburst PCO₂ system monitors atmospheric and surface water pCO₂.

Calibration Information:

Typical Signal Shapes:

Typical Failure Modes:

External Sources of Documentation:

- <http://www.pmel.noaa.gov/co2/>
- <http://www.pmel.noaa.gov/co2/story/Buoys+and+Autonomous+Systems>
- <http://www.battelle.org/ASSETS/C1D16CFE72F349359228B117921D3B8D/pCO2%20Monitoring%20Brochure.pdf>

Instrument Class WAVSS (Surface Wave Spectra)

Model 2 Subject Matter Expert:

Calibration Center:

Manufacturer, make & model: Axys Technologies Triaxys

General description: TRIAXYS Directional Wave Sensor collects continuous wave sampling.

Calibration Information:

Typical Signal Shapes:

Typical Failure Modes:

External Sources of Documentation:

- <http://www.axystechnologies.com/BusinessUnit/AXYSMarineEnvironmentalMonitoring/WaveCurrentWaterLevelMonitoringSolutions/TRIAXYSWaveandMotionSensors/tabid/109/Default.aspx>

Instrument Class ZPLSG (Zooplankton Sonar)

Model 2 Subject Matter Expert:

Calibration Center:

Manufacturer, make & model: Simrad ES60 fish finding echo sounder

General description: ES60 echo sounders range from relatively low-cost single beam to large multi-frequency systems containing several split-beam channels.

Calibration Information:

Typical Signal Shapes:

Typical Failure Modes:

External Sources of Documentation:

- <http://www.simrad.com/www/01/nokbg0397.nsf/AllWeb/2CEFD0293AED3771C125718F003A950A?OpenDocument>

Instrument Class CTDPF (CTD used on profilers)

Model 1 Subject Matter Expert:

Calibration Center:

Manufacturer, make & model: Sea-Bird Electronics SBE-52MP

General description: SBE 52-MP measures conductivity, temperature, and pressure by travelling vertically beneath a buoy or from a buoyant sensor package that is winched up and down from a bottom-mounted platform. A Dissolved Oxygen Sensor Module (SBE 43F) is optional.

Calibration Information: EEPROM-stored calibration coefficients permit data upload in ASCII engineering units (mmho/cm, °C, decibars, ml/l). Alternatively, the user can select to upload data in hexadecimal or binary.

Typical Signal Shapes:

Typical Failure Modes:

External Sources of Documentation:

- http://www.seabird.com/products/spec_sheets/52data.htm

Instrument Class DOSTA and DOFST (Dissolved Oxygen)

Model 2 Subject Matter Expert:

Calibration Center:

Manufacturer, make & model: Sea-Bird SBE43

General description: measures absolute oxygen concentration and % saturation.

Calibration Information:

Typical Signal Shapes:

Typical Failure Modes:

External Sources of Documentation:

Instrument Class NUTNR (Nitrate)

Model 2 Subject Matter Expert:

Calibration Center:

Manufacturer, make & model: Satlantic ISUS and SUNA models

General description: ISUS and SUNA measures nitrate concentration using advanced UV absorption technology.

Calibration Information: Calibration parameters are obtained at Satlantic by measuring absorption spectra of samples in the range of 0-40 μM nitrate, 0-35 psu and 0-20 $^{\circ}\text{C}$ temperature. The parameters for the nitrate calculations are loaded into the instrument.

Typical Signal Shapes:

Typical Failure Modes:

External Sources of Documentation:

- <http://www.satlantic.com/details.asp?ID=11&CategoryID=2&SubCategoryID=0>
- http://www.satlantic.com/documents/624333_SAT-DN-425_Operation_Manual_ISUS_V3_RevA5.pdf

Instrument Class OPTAA (Spectrophotometer, optical attenuation and absorption)

Model 2 Subject Matter Expert:

Calibration Center:

Manufacturer, make & model: WETLabs ac-s

General description: ac-s measures absorption and beam attenuation. It has 9 wavelengths of absorption (a) and attenuation (c)

Calibration Information: Factory calibration for absorption and attenuation consists of temperature characterization, optically clean water calibration, and air calibration. Field calibration needs to be considered. Details of calibration information can be found in the manufacturer website (see links in External Sources and Documentation)

Typical Signal Shapes:

Typical Failure Modes:

External Sources of Documentation:

- <http://www.wetlabs.com/products/ac/acall.htm>
- <http://www.wetlabs.com/technicalnotes/China%202.pdf>
- www.wetlabs.com/appnotes/docs/v4v4ch2.doc
- www.wetlabs.com/appnotes/docs/v4v4ch3.doc

Instrument Class PCO2W (Delta pCO₂ Waterside)

Model 2 Subject Matter Expert:

Calibration Center:

Manufacturer, make & model: Sunburst SAMI-CO₂

General description: SAMI-CO₂ measures partial pressure of CO₂ in water

Calibration Information:

Typical Signal Shapes:

Typical Failure Modes:

External Sources of Documentation:

- http://www.sunburstsensors.com/Product_SAMI.html

Instrument Class SPKIR (Spectral Irradiance)

Model 2 Subject Matter Expert:

Calibration Center:

Manufacturer, make & model: Satlantic OCR-507

General description: OCR-500 series radiometers are available in four or seven channel discrete wavelengths from 400-865 nm.

Calibration Information:

Typical Signal Shapes:

Typical Failure Modes:

External Sources of Documentation:

- http://www.satlantic.com/Multispectral_Radiometers/ocr-500_series_radiometers.asp

Instrument Class VEL3D (Velocity (point), turbulent U, V, W)

Model 1 Subject Matter Expert:

Calibration Center:

Manufacturer, make & model: Nobska MAVS-4, Nortek Vector

General description: MAVS3 (Modular Acoustic Velocity Sensor 3rd Generation) is a 3-D single point acoustic current meter that uses time travel technology. Nortek Vector's instrument is a Doppler current meter with three capabilities beams focused on one small sample volume.

Calibration Information: In Menu mode and MAVSoft mode, velocity offsets, compass offsets, and tilt offsets can be calibrated. Within the Nortek software, the compass is calibrated from the On-line menu.

Typical Signal Shapes:

Typical Failure Modes:

External Sources of Documentation:

- <http://www.nobska.net/>
- <http://www.nobska.net/Kobe%20Tutorial.ppt>
- <http://www.nortekusa.com/usa/products/acoustic-doppler-velocimeters/vector-1>

Instrument Class CTDGV (CTD for gliders)

Model 1 Subject Matter Expert:

Calibration Center:

Manufacturer, make & model: Sea-Bird Electronics SBE-41CP

General description: SBE41 module uses the proven MicroCAT temperature, conductivity, and pressure sensors. SBE41CP is designed to perform a Continuous Profile.

Calibration Information:

Typical Signal Shapes:

Typical Failure Modes:

External Sources of Documentation:

- <http://www.seabird.com/alace.htm>

Appendix B-1 Tests Specific to Sensor Make/Model (subject to scope and budget)

Sea Water Electrical Conductivity: Problems with a conductivity sensor are most easily spotted in salinity. Spikes and temporary shifts in calibration are fairly common and can be explained by dirt particles entering the sensor. The operator should monitor salinity closely for these symptoms.

Sea Water Pressure: Sensor drift is a common occurrence in pressure sensors. The operator should monitor pressure time series closely for this symptom. In cases where a tethered pressure sensor changes depth naturally, such as a mooring wire that tilts in ambient currents, it may be possible to consider only those periods without the motion (i.e. visually interpolate / ignore tilt events). Likewise, for mobile platforms (gliders, AUVs, profiled sensors) it is possible to only consider periods at known / constant depths (surface, AUV docking stations, profiler park positions). The goal is to have a minimum baseline to confirm absence of drift behavior for every pressure sensor. Because seawater pressure is often used to infer the vertical position of the instrument, quality control is particularly important for this quantity.

Appendix B-2 Parameter-Specific Tests (subject to scope and budget)

Automated QC algorithms are a significant component of the data QC process of the OOI network, and shared algorithms will be used for common and/or similar instrument classes deployed across IOs. The following are QC tests for measurements of specified quantities; some are to be implemented on specific sensors only. Future work includes evaluating other QC tests for inclusion into automated algorithms. Additional parameter-

specific tests are developed as Instrument Make/Model information becomes available and the QC procedures are better defined through incorporation of community-accepted QA/QC standards. CI will only implement algorithms described in approved DPSs.

Sea Water Pressure at Surface (sensor specific). For pressure sensors in sea water that are on platforms at or near the sea surface, a test shall be performed that validates that the pressure reading is close to zero. If the reported value falls outside the limits defined by a multiple of the nominal sensor accuracy A away from the expected value (zero at surface), the test shall fail, and all subsequent pressure measurements be flagged accordingly. That is, to pass the test, the measurement X taken at the sea surface must fulfill:

$$|X| < 3A$$

Examples of platforms to which this test shall be applied are gliders while at the surface, moored profilers while at the surface, and surface buoys. If the measurement is taken at a known but short and constant distance D below the surface (e.g. on the bottom of a surface buoy), the above equation becomes:

$$|X - D| < 3A \quad (D \text{ in m, } X \text{ and } A \text{ in dbar})$$

Sea Water Pressure in Subsurface Moorings (sensor specific). A test shall be applied to underwater mooring systems with multiple pressure sensors at different depths to validate that the pressure readings are not further apart vertically than the wire length between them allows. If the test fails, all pressure data involved shall be flagged accordingly, and a note to the mooring operators shall be sent out to alert them of the situation.

Given two pressure measurements $P_{1,2}$ (in dbar) of accuracies A and a wire length W (in m) between them, the test passes if:

$$|P_2 - P_1| < \max(\{1.1W, W + 5A\})$$

Sea Water Conductivity, Temperature, and Pressure: Vertical Density Gradient Test (implemented). A test shall be performed for all platforms that measure sea water electrical conductivity, temperature, and pressure simultaneously at different depths, in order to identify unrealistic vertical density gradients. Failure of the test results in all conductivity, temperature, and pressure measurements involved being flagged accordingly. The test works as follows:

Compute densities $\rho_{1,2}$ from the measurements at two depths, where ρ_2 is from the deeper location.

Derive the measurement accuracy A from the accuracies in conductivity, temperature, and pressure.

The test passes if:

$$\rho_2 - \rho_1 > -5A$$

Appendix C Field Verification of OOI Data and Data Products

Appendix C contains a summary of the methods that can be used for Field Verification of OOI instruments and their data/data products. For instruments that produce more than one type of data, multiple methods may be listed. The methods may also be instrument model, platform, and/or location specific.

The Field Verification methods summarized in Table C-1 assume that basic functionality of the instrument upon deployment has already been verified, and that the generated data are reasonable, based on the operational requirements and locations of the instruments. In the enclosed table, the “Primary Field Verification” column lists the preferred methods of Field Verification of data for each of the instruments. The “Secondary Field Verification” column lists alternative methods that may be used in addition to the primary procedures or in lieu of them if weather conditions during Operations & Maintenance (O&M) preclude the preferred method.

This list of Field Verification methods is not all inclusive, and it is anticipated that this selection of applicable methods will be modified and/or augmented as the historical record of OOI data develops, the operational characteristics of the OOI instruments are better delineated, the OOI O&M program matures, and additional sources of data (e.g. ships of opportunity, satellite radiances, additional deployed instruments) become available.

Any data collected during Field Verification may also be used for Secondary Calibration of OOI data and data products and/or data adjustment during the Data Quality Assessment process of the QA/QC program, as described in Sections 5 and 6, respectively.

KEY TO TABLE C-1

LOCATION

CE	Coastal Global Scale Nodes (CGSN)– Endurance Array
CP	CGSN – Pioneer Array
GA	CGSN – Argentine Basin
GI	CGSN – Irminger Basin
GP	CGSN – Ocean Station Papa
GS	CGSN – Southern Ocean
RA	Regional Scale Nodes (RSN) – Axial Seamount
RH	RSN – Hydrate Ridge
ALL	All of Above

FIELD VERIFICATION METHOD

NA	Not applicable
TBD	To be determined
ColorCard	Visual evaluation of HD video and digital still images using white balance and color correction/calibration cards post deployments and/or during maintenance visits.

CTD-Nearby	Comparison with CTD data from a nearby profiler/mooring/glider/profiler. Temperature-Salinity (T-S) plots also utilized during inter-instrument comparisons.
CTDCast-PD/PR	Comparison with data from the up/down casts of a recently calibrated CTD and/or target instrument (attached to rosette) either post-deployment (PD) and/or pre-recovery (PR) of the instrument of interest. Temperature-Salinity (T-S) plots also utilized during inter-instrument comparisons. The locations of the Field Verification CTD casts are as described for the hydrocasts in Section 5 Physical Samples and Analytical Protocols).
Inst-Nearby	Comparison of data/data products with those from a nearby instrument on profiler/mooring/glider/seafloor with analogous measurement capabilities. Specific approaches are highly location-specific.
PhysSamp-PD/PR	Physical Samples collected post-deployment (PD) and/or pre-recovery (PR) of instrument and analyzed by ship-based and/or shore-based procedures. See –Section 5 Physical Samples and Analytical Protocols . Analytical results from physical samples used for Field Verification are provided as metadata to the core OOI data products.
MPR_ROV	Measurement of relative water column pressure (relative depth) between seafloor benchmarks near deployed instruments and a benchmark at a reference location instrument using a recently calibrated Mobile Pressure Recorder (MPR) system mounted on a ROV. While included as a Field Verification method, its intent is primarily to correct for drift in pressure data from the BOTPT instrument which is used to determine seafloor inflation/deflation.
Temp_ROV	Measurement of temperature near instrument post deployment and/or during maintenance visits using a recently calibrated thermistor/thermocouple mounted on the ROV.
T-S_Plot	Temperature-Salinity (T-S) plot used for sensor evaluations on CTDs, data curves should be consistent.
Vel-Nearby	Comparison with an instrument (VELPT/ADCP) co-located on the platform or on a nearby OOI profiler/mooring/glider. Vessel-mounted reference instrument may also be utilized.

Table C-1 Field Verification Methods for OOI Data and Data Products

Instrument Class	Instrument Name	Inst Model	Location	Data Type Data Product	Primary Field Verification Method	Secondary Field Verification Method
ADCPA	velocity_profile_mobile_a sset		CE,CP	Velocity Profiles Acoustic BS	Vel-Nearby	TBD
ADCPS	velocity_profile_600m		ALL (exc RA)	Velocity Profiles Acoustic BS	Vel-Nearby	TBD
ADCPT	velocity_profile_300m		CE,CP RA,RH	velocity profiles Acoustic BS	Vel-Nearby	TBD
BOTPT	pressure_bottom_tilt		RA	Bottom Pressure	MPR_ROV	NA
				Seafloor Tilt	NA	NA
CAMDS	camera_digital_still_strob e		CE RA,RH	Still image	ColorCard	NA
CAMHD	camera_digital_video_HD		RA	HD Video image	ColorCard	NA
CTDAV	CTD_AUV		CP	CTD data Practical Salinity Density	T-S_Plot; CTD-Nearby	TBD
CTDBP	CTD_bottom_pumped		CE,CP	CTD data Practical Salinity Density	T-S_Plot; CTDCast-PD/PR; PhysSamp-PD/PR	CTD-Nearby
CTDGV	CTD_glider		CE,CP GA,GI GP,GS	CTD data Practical Salinity Density	T-S_Plot; CTD-Nearby	CTD-Nearby
CTDMO	CTD_mooring		GA,GI GP,GS	CTD data Practical Salinity Density	T-S_Plot; CTDCast-PD/PR; PhysSamp-PD/PR	CTD-Nearby
CTDPF	CTD_profiler		ALL	CTD data Practical Salinity Density	T-S_Plot; CTDCast-PD/PR; PhysSamp-PD/PR	CTD-Nearby
DOFST	oxygen_dissolved_fastres p		CE,CP RA,RH	Dissolved Oxygen	CTDCast-PD/PR; PhysSamp-PD/PR	Inst-Nearby
DOSTA	oxygen_dissolved_stable		ALL	Dissolved Oxygen	CTDCast-PD/PR; PhysSamp-PD/PR	Inst-Nearby
FDCHP	flux_direct_cov_HP		CE,CP GA,GI,GS	Covariance Flux of Heat, Moisture, Momentum	Inst-Nearby	TBD
				Humidity, Air Temp	Inst-Nearby	TBD
				Wind Velocity	Inst-Nearby	TBD
FDCLP	flux_direct_cov_LP		CP	Covariance Flux of Heat, Moisture, Momentum	TBD	TBD
				Humidity, Air Temp	Inst-Nearby (METBK)	TBD
				Wind Velocity	TBD	TBD
FLOBN	flow_benthic		RH	Benthic Flow	TBD	TBD
FLORD	fluorometer_two_wavelen gth		GA,GI GS,GP RA,RH	Chlorophyll-a Optical BS	CTDCast-PD/PR; PhysSamp-PD/PR	Inst-Nearby
FLORT	fluorometer_three_wavel ength		CE,CP RA,RH	Chlorophyll-a CDOM Optical BS	CTDCast-PD/PR; PhysSamp-PD/PR; CDOM - TBD	Inst-Nearby
HPIES	IES_pressure_velocity		RA,RH	RATT, Bottom Pressure, Horiz Electric Fields, Horiz Avg Velocities, Water Prop Prof Time Series Water Column Heat	TBD	TBD
HYDBB	hydrophone_BB_passive		CE,RA,R H	Frequency, Acoustic Pressure	TBD	TBD

Protocols and Procedures for OOI Data Products: QA, QC, Calibration, Physical Samples

				Waves		
HYDLF	hydrophone_LF_passive		RA,RH	Acoustic Pressure Waves	TBD	TBD
MASSP	mass_spectrometer		RA,RH	Dissolved Gas Concentrations	PhysSamp-PD/PR	Inst-Nearby
METBK	meteorology_bulk		CE,CP GA,GI, GP,GS	Assortment of Meteorological Data	Dual Sensors	Inst-Nearby (FDCHP)
NUTNR	nutrient_Nitrate		ALL	Nitrate Concentration	PhysSamp-PD/PR	Inst-Nearby
NUTR4	nutrient_four_channel		CP	NO2,NO3,PO4, SIO4 Concentrations	PhysSamp-PD/PR	Inst-Nearby
OBSBB	seismometer_BB_tri axial _accel		RA,RH	Broadband Ground Acceleration and Velocity	Inst-Nearby	TBD
OBSBK	seismometer_BB_tri axial _keck		RA,RH	Broadband Ground Acceleration and Velocity	Inst-Nearby	TBD
OBSSP	seismometer_shortperiod		RA,RH	Short Period Ground Velocity	Inst-Nearby	TBD
OPTAA	attenuation_absorption_o ptical		All	Optical Attenuation and Absorption	Inst-Nearby	TBD
OSMOI	watersample_chem_trace		RA,RH	Water Trace Chemistry	PhysSamp-PD/PR	Inst-Nearby
PARAD	PAR		CE,CP RA,RH	PAR	CTDCast-PD/PR; Inst-Nearby	TBD
PCO2A	pCO2_air-sea		CE,CP GA,GI GP,GS	pCO2 in air	Inst-Nearby (PCO2W)	TBD
PCO2W	pCO2_water		ALL	pCO2 in water	PhysSamp-PD/PR	Inst-Nearby
PHSEN	pH_stable		ALL	pH	PhysSamp-PD/PR	Inst-Nearby
PPSDN	DNA_particulate		RA	16s rRNA of Physical Sample	PhysSamp-PD/PR	TBD
PRESF	pressure_SF		CE,CP	Seafloor Pressure	Inst-Nearby	TBD
PREST	pressure_SF_tidal		RA,RH	Seafloor Pressure – Tidal	MPR_ROV	TBD
RASFL	watersample_chem_trace _H2S_pH		RA	Water Chemistry of Physical Sample	PhysSamp-PD/PR	TBD
SPKIR	spectral_irradiance		ALL	Downwelling Spectral Irradiance, Diffuse Attenuation	CTDCast-PD/PR	Inst-Nearby
THSPH	temp_H2_H2S_pH		RA	Vent Fluid Temp, H2, H2S, pH	PhysSamp-PD/PR	Inst-Nearby
TMPSF	temperature_seafloor		RA	Spatial Distribution of Temperature	Temp_ROV	Inst-Nearby
TRHPH	temp_resist		RA	Vent Fluid Temp, Redox State, Chlorinity	PhysSamp-PD/PR	Inst-Nearby
VADCP	velocity_profile_50m_turb		RA,RH	Turbulent Velocity Profiles, Acoustic BS	Vel-Nearby	TBD
VEL3D	velocity_point_3D_turb		ALL	Turbulent Point Water Velocity	Vel-Nearby	TBD
VELPT	velocity_point		CE,CP GA,GI GS	Mean Point Water Velocity	Vel-Nearby	TBD
WAVSS	wave_spectra_surface		CE,CP GA,GI GS	Platform Motion	TBD	TBD
ZPLSC	plankton_ZP_sonar_coas tal		CE,CP	MF Acoustic BS	TBD	TBD
ZPLSG	plankton_ZP_sonar_glob al		GA,GI GP,GS	MF Acoustic BS	TBD	TBD

Primary and Secondary Calibration Methodology by Instrument

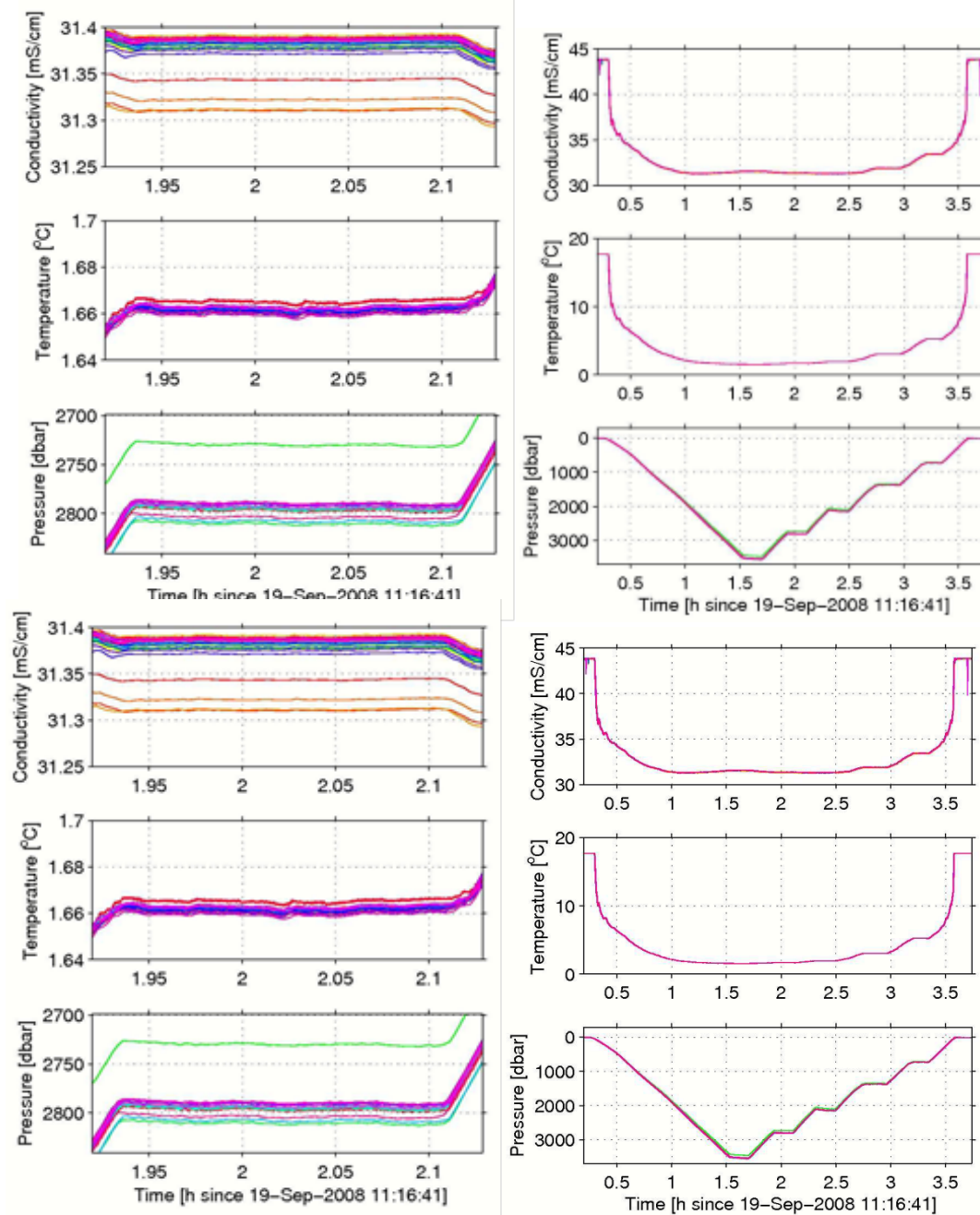
Reference Designator	Calibration Methodology
GA01SUMO-FI***-**-CTDMO****	Primary: at manufacturer Secondary: on ship-borne CTD/rosette system: Use electronic data from ship-borne system. Temperature: no water samples needed Pressure: no water samples needed Conductivity: take water samples for salinity and adjust ship-borne system as needed
GA01SUMO-FI***-**-ADCPS****	Primary: at manufacturer, plus compass calibration in port prior to ship departure Secondary: none
GA01SUMO-FI***-**-FLORD****	Primary: at manufacturer Secondary: on ship-borne CTD/rosette system, using electronic data if available, else water samples
GA01SUMO-FI***-**-VELPT****	Primary: at manufacturer, plus compass calibration in port prior to ship departure Secondary: none
GA01SUMO-FI***-**-PHSEN****	Primary: at manufacturer Secondary: on ship-borne CTD/rosette system, using water samples of total dissolved organic carbon and alkalinity
GA01SUMO-SB***-**-METBK****, GA01SUMO-SB***-**-FDCHP****	Primary: at manufacturer Secondary: in-situ with ship-borne reference system, using ship meteorology suite, standing by buoy short distance downwind for 4h
GA01SUMO-SB***-**-PCO2A****	Primary: at manufacturer Secondary: on ship-borne CTD/rosette system, using water samples of total dissolved organic carbon and alkalinity
GA01SUMO-SB***-**-WAVSS****	Primary: at manufacturer Secondary: none

Appendix D: Secondary Calibration Procedures on Global Deployment Research Vessel during an OOI Cruise Procedure “On Ship-Borne CTD/Rosette System”

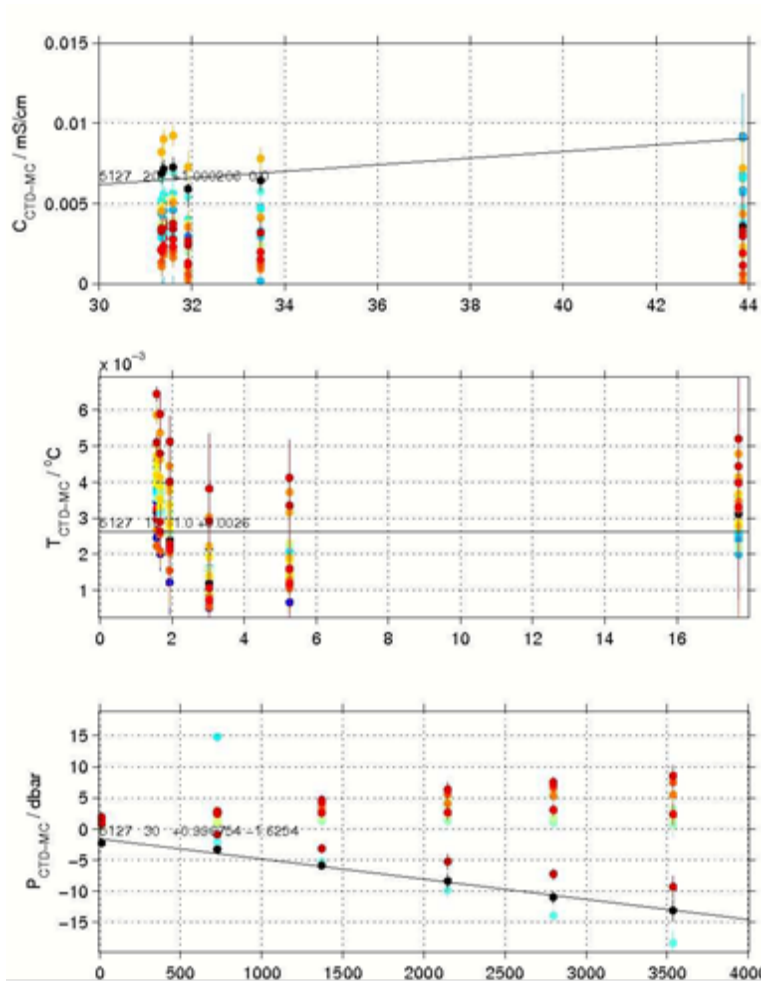
Moored conductivity, temperature, and pressure sensors shall be cross-calibrated with a shipborne CTD system immediately prior to deployment and after recovery. For that purpose, the shipborne CTD system must be calibrated accordingly. WOCE standards should suffice for the shipborne CTD; it will typically be necessary to correct the CTD conductivity using salinometry. Best results are obtained if the CTD is stopped for several minutes at selected depths during the upcast. The procedure for the cross-calibration is as follows (illustrated in figures below):

1. Attach mooring sensors to shipborne CTD system, as close to sensor inlets as possible (esp. in the vertical). Mooring sensors should sample at sufficiently high temporal rate.
2. Perform CTD cast: lower to target depth and identify five depths with low vertical gradients in temperature and salinity
3. During upcast, pause at these five depths for ten minutes each. If desired, collect water samples for salinometry here as well.
4. After cast, extract data from CTD system at these depths as well as from mooring sensors
5. Apply all calibrations to CTD system and derive field calibrations for mooring sensors from differences between their values and the CTD values at the stop positions.
6. In lab, analyze water samples and utilize for calibration as applicable

The corrections to be applied to the mooring data must be interpolated in time to account for the time passed between pre-deployment field calibration and post-recovery field calibration. The quantity to be interpolated is the to-be-applied correction, using linear interpolation in time.



Above: Example of a CTD cast with mooring sensors attached for calibration. Right: Conductivity, temperature, and pressure data as a function of time. Note the stops in the upcast, from which calibrations will be derived. Left: Close-up of one stop, showing the multiple sensors delivering slightly different values.



Above: Differences between the CTD and mooring sensor values from previous figure at the stop positions. Black lines highlight to-be-applied field calibrations for one exemplary sensor.

Procedure “*In situ* with Ship-Borne Reference Sensor”

1. approach instrument *in situ* with ship-borne reference system
2. if possible, re-task instrument to high sampling rate
3. Inter-comparison time period: either...
 - a) maintain reference system in vicinity of in-situ system for defined period of time, collecting data with reference system, or
 - b) collect water samples in vicinity of in-situ system
4. afterwards, extract data from all systems from the inter-comparison time period
5. analyze water samples, as applicable
6. Obtain calibration coefficients from inter-comparison between extracted instrument data versus...
 - a) extracted ship-borne reference data, or
 - b) water samples

Appendix E: Guidelines and Instructions for Visual Inspection QC

Visual inspection by a human expert is an important part of the overall QC procedure. While automation of the QC process dramatically reduces the need for manual intervention, periodic QC inspection by human experts ensures that automated algorithms are effective and that resulting data flags are appropriate. This “visual data QC” also provides the most up-to-date qualification of the data streams via human judgment, i.e. before automated algorithms may be appropriately updated due to changes in the instruments and/or environment being monitored. Furthermore, external calibration of data streams is an important component of data quality assurance.

At the time of visual inspection the data must already have been adjusted for primary and secondary calibrations and have automated quality control test result flags attached. The visual inspection then investigates all instances where the data have not passed the automated tests, as indicated by QC summaries generated during the automated procedures. In addition, the procedures outlined in this section are performed. To summarize:

- Confirm or reject all issues arising out of the automated tests
- Perform all metadata and variable tests described below in this section
- Ensure propagation of bad flags to derived variables and data products, including possibly revised results

Metadata

Platform configuration: Compare the computer database of platform configuration (sensor locations on the platform and sensor serial numbers) with the manual versions at instrument deployment and recovery. Resolve any discrepancies.

Clock drift: Verify that the instrument clock drift has been accounted for in the dataset.

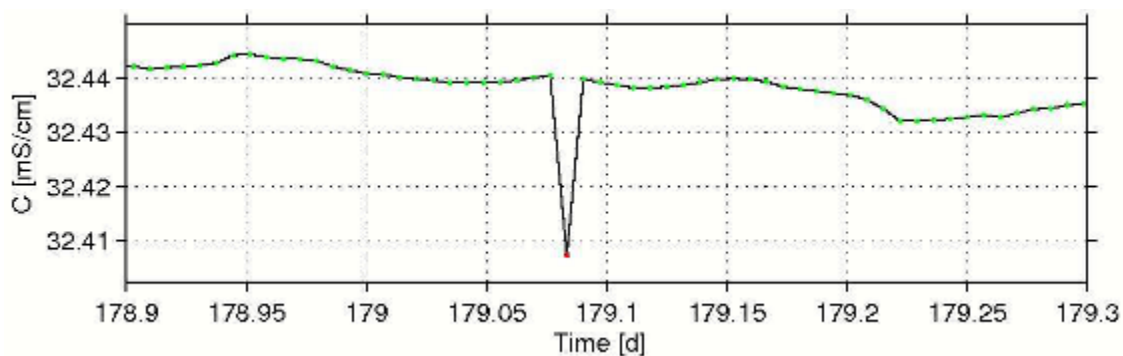
Location: Compare the deployment / recovery locations (latitude and longitude) from the manual logs to the computer database and resolve any conflicts.

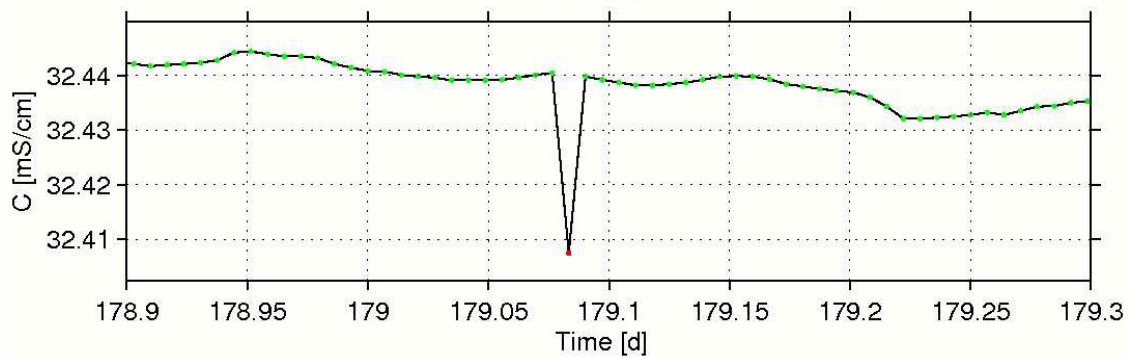
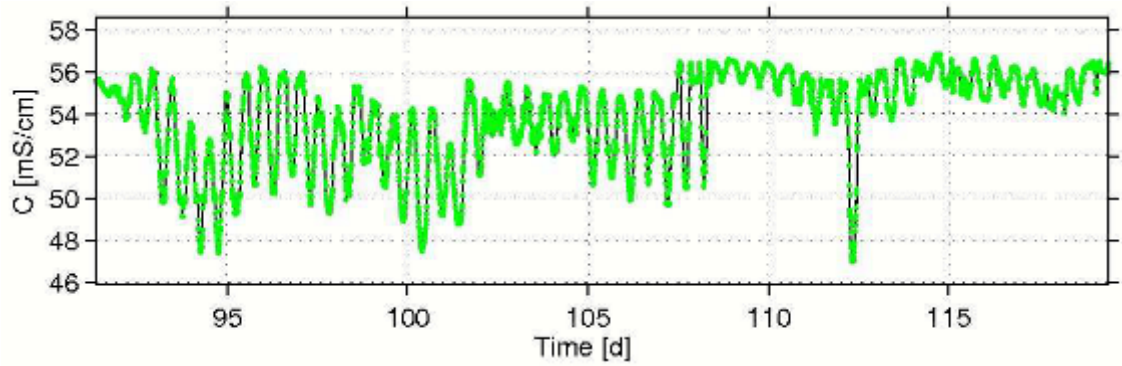
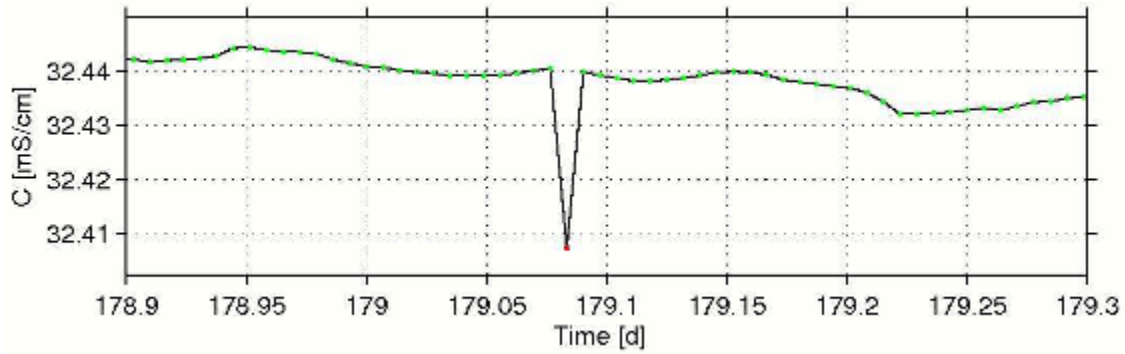
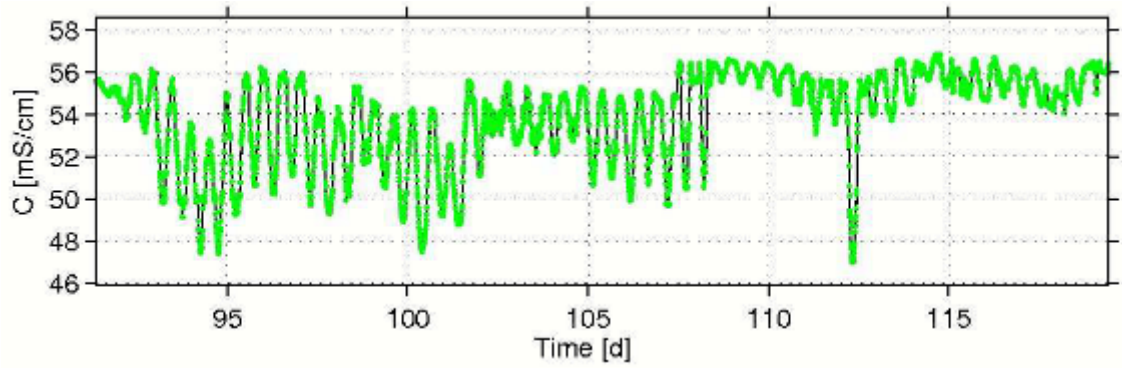
Tests for all Variables

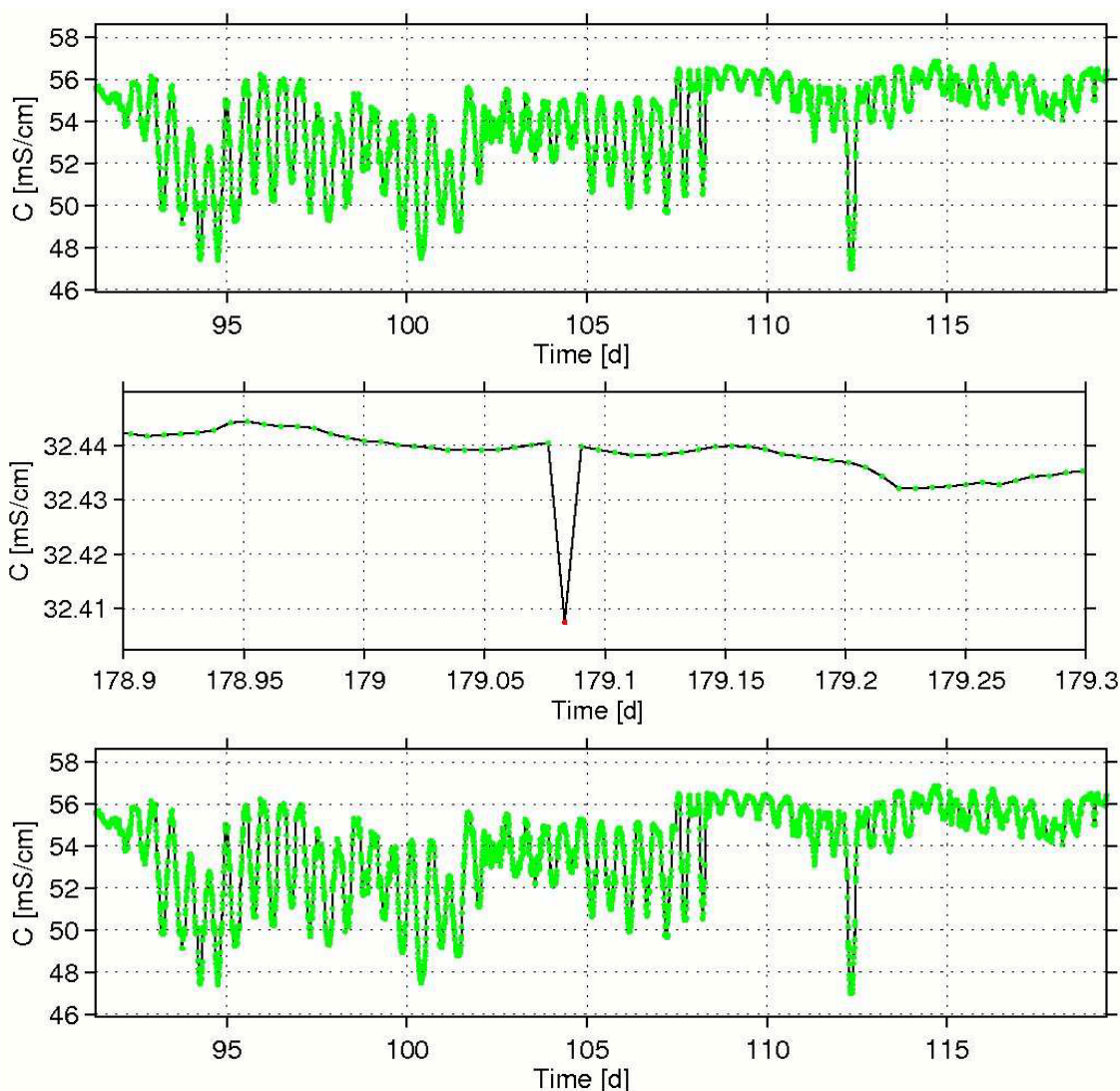
Visual inspection of the data shall explicitly look for occurrences of the following, presumably bad, behavior in the data:

- Spikes, single-point outliers
- Sudden jumps or episodes of other-than-usual values, multi-point outliers
- Slow drift
- Sensor-specific behavior

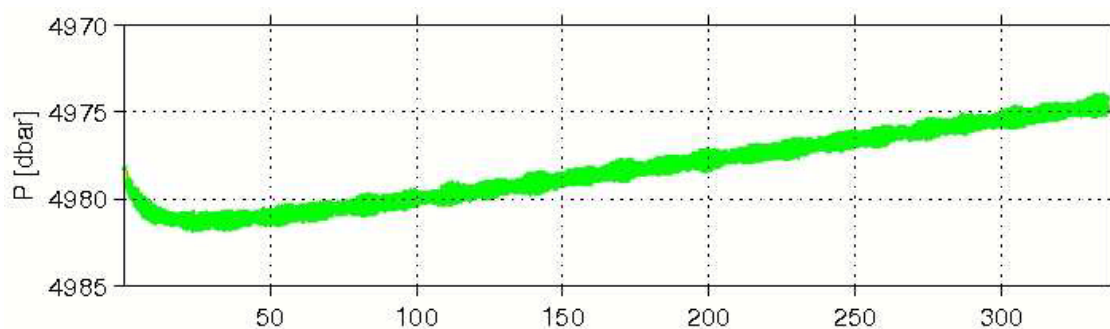
Examples are shown in the following figures:

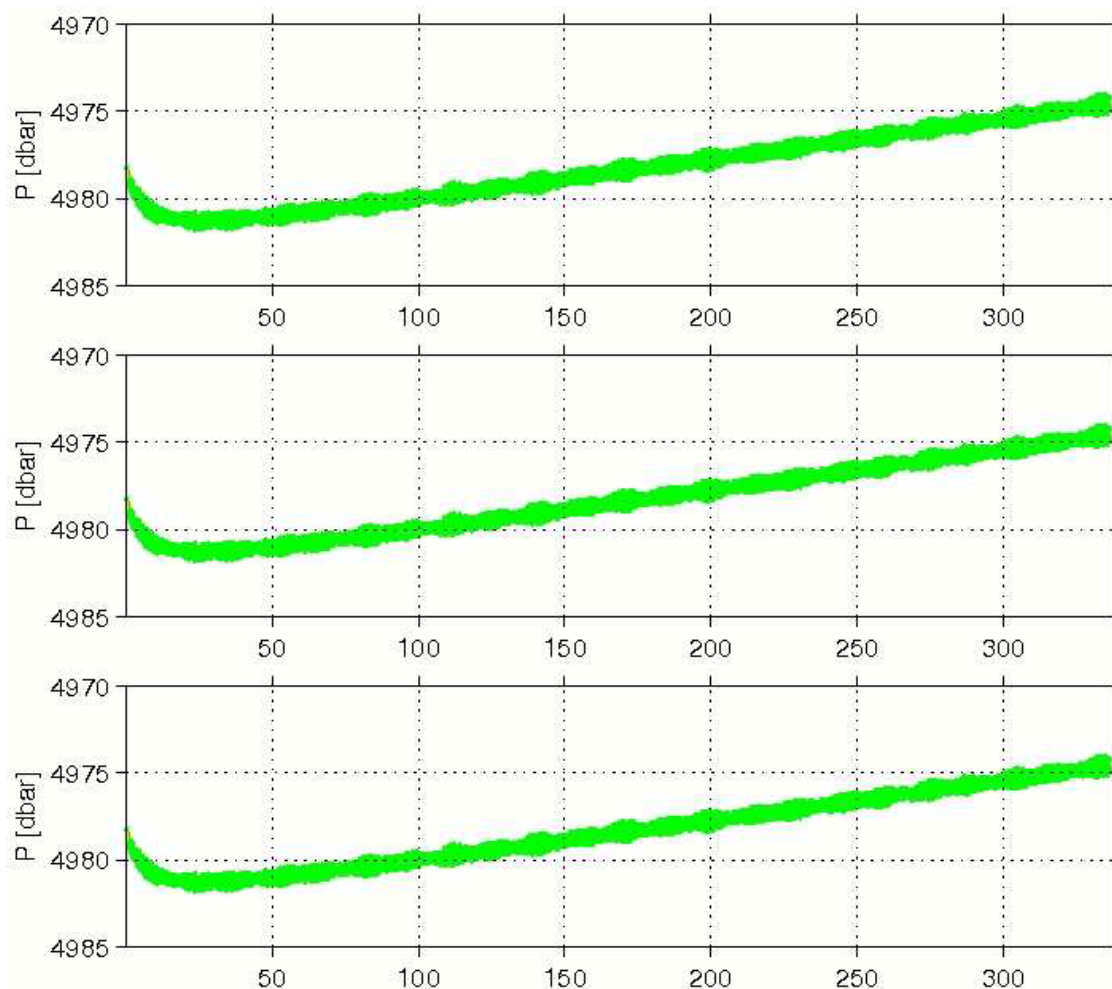




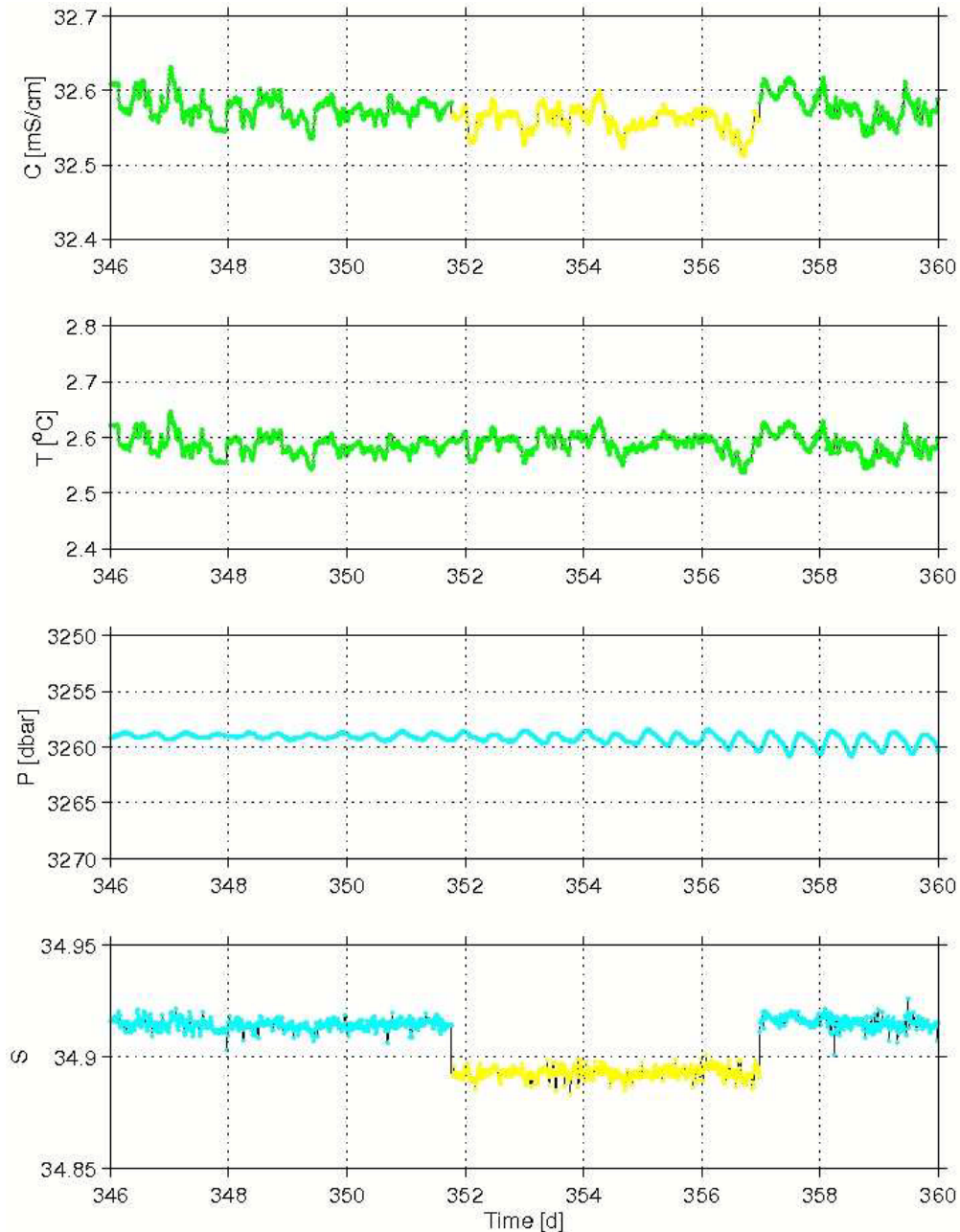


Above; Examples of spike detection in time series of seawater electrical conductivity. While a spike of very small amplitude is easily detected in the upper panel, the lower panel presents a situation where natural oscillations by much larger amplitude defeat both automatic detection algorithms and human eye.





Above: Example of a time series with an erroneous drift. The instrument was a pressure sensor moored at circa 5000 m depth.



Above: Example of data with a temporary malfunction in a conductivity sensor (marked yellow). Panels from top to bottom show time series of electrical conductivity, temperature, pressure, and derived salinity from an instrument in seawater. The episode of poor performance in conductivity stands out more clearly in the salinity data.

Appendix F Checklists for Visual Inspections of Data by Subject Matter Expert (SME)

Generic Checklist for all Data Products

Data Product ID: _____
 Operator name: _____
 Date of inspection: _____

Pass	Fail	N/A	Description
			Dataset complete (i.e. starts/ends at nominal start/end date, no gaps)?
			Any anomalies reported in deployment/recovery log sheets?
			Complete chain of custody documents and analysis results (including documentation of laboratory Standard Operating Procedures [SOPs] and QA/QC statistics) for any physical samples?
			Reference designator, instrument model and serial number correct in metadata?
			Location (latitude, longitude, depth) correct in metadata?
			Time information correct, clock drift corrected, timing uncertainty specified?
			Primary calibrations applied to data and documented in metadata?
			Secondary calibrations applied to data and documented in metadata?
			Any obvious anomalies or outliers in data?
			Data values at start consistent with end values of previous deployment?
			Data values at end consistent with start values of successive deployment?
			Data values consistent with data from neighboring related instruments? Which instrument(s):
			Data values consistent with dedicated Field Verification data... ...at beginning of deployment? ...during deployment? ...at end of deployment?
			No drift/trend/jump in data that could be due to sensor drift, malfunction, bio-fouling?
			Unusual amount of problems detected by automated checks?
			Unusual amount of false positive/false negative results by automated checks?

Checklist for all CTD Instruments**

Data Product ID: _____
Operator name: _____
Date of inspection: _____

Pass	Fail	N/A	Description
			Generic checklist completed?
			T-S plot of single sensor: all data on one consistent curve?
			T-S plot of all sensors on same platform (if applicable): all data on consistent curve? Data sets used: _____
			T-S plot of previous deployment (multiple sensors if applicable): all data on one consistent curve? Data sets used: _____
			T-S plot of simultaneous measurements on different platform: all data on consistent curve? Data sets used: _____

Checklist for all DOSTA and DOFST Instruments

Data Product ID: _____
Operator name: _____
Date of inspection: _____

Pass	Fail	N/A	Description
			Generic checklist completed?
			T-O plot of single sensor: all data on one consistent curve?
			T-O plot of all sensors on same platform (if applicable): all data on consistent curve? Data sets used: _____
			T-O plot of previous deployment (multiple sensors if applicable): all data on one consistent curve? Data sets used: _____
			T-O plot of simultaneous measurements on different platform: all data on consistent curve? Data sets used: _____

Checklist for all ADCP Instruments (Classes ADCPT, ADCPS, ADCPA, VADCP)

Data Product ID: _____
 Operator name: _____
 Date of inspection: _____

Pass	Fail	N/A	Description
			Generic checklist completed?
			Confirm that internal compass calibration has been performed prior to deployment (see log sheets or configuration files).
			Confirm that settings for magnetic variation are correct: Desired value: + / - _____ ° Value applied internally by instrument: + / - _____ ° Value applied externally but not in CI: + / - _____ ° Value applied externally in CI: + / - _____ °
			Bottom / surface echoes properly identified by automated QC tests?

Checklist for all VEL3D and VELPT Instruments

Data Product ID: _____
 Operator name: _____
 Date of inspection: _____

Pass	Fail	N/A	Description
			Generic checklist completed?
			Confirm that internal compass calibration has been performed prior to deployment (see log sheets or configuration files).
			Confirm that settings for magnetic variation are correct: Desired value: + / - _____ ° Value applied internally by instrument: + / - _____ ° Value applied externally but not in CI: + / - _____ ° Value applied externally in CI: + / - _____ °

Checklist for all METBK Instruments

Data Product ID: _____
 Operator name: _____
 Date of inspection: _____

Pass	Fail	N/A	Description
			Generic checklist completed?
			Confirm that internal compass calibration has been performed prior to deployment (see log sheets or configuration files).
			Confirm that settings for magnetic variation are correct: Desired value: + / - _____ ° Value applied internally by instrument: + / - _____ ° Value applied externally but not in CI: + / - _____ ° Value applied externally in CI: + / - _____ °

Checklist for all FDC* Instruments

Data Product ID: _____
Operator name: _____
Date of inspection: _____

Pass	Fail	N/A	Description
			Generic checklist completed?
			Confirm that internal compass calibration has been performed prior to deployment (see log sheets or configuration files).
			Confirm that settings for magnetic variation are correct: Desired value: + / - _____ ° Value applied internally by instrument: + / - _____ ° Value applied externally but not in CI: + / - _____ ° Value applied externally in CI: + / - _____ °

Appendix G – Sampling Protocols and Analyses

G1. Dissolved Oxygen Analyses

Oxygen analyses are conducted according to the procedures outlined by Steve Emerson, University of Washington

OXYGEN ANALYSIS NOTES

1. Boxes of O₂ sample bottles (24 each) have been provided. The volumes of these bottles are located in each box. Transfer the sheets to the O₂ Analysis log book.
2. Reagents for each cruise should be made fresh. Always bring back unused reagents so that reagents and standards may be checked post-cruise.
3. All the O₂ sample bottles should be washed with lab soap and water before and after each cruise. If the bottle has been used for a sample, it should be washed before reuse. Ship's water is adequate for the distilled water rinse.
4. 5 mL glass pipettes should be used to dispense the KIO₃ standards. These pipettes are calibrated and labeled with the calibration. If possible, a spare Dosimat (calibrated for 5 mL delivery) may be used to deliver the main KIO₃ standard.
5. The Dosimat should be secured to the counter. Tape or string should be used to keep the exchange unit (includes the glass thiosulfate bottle and the fill/empty piston assembly) firmly locked in place.

Note: An error message "No exch. Unit" appears if the exchange unit is not firmly seated:

6. The following procedure should be used when turning on the Dosimat:
 1. The ON-OFF button is located on the rear of the Dosimat. Hold in the FILL button on the front of the Dosimat and push the ON-OFF button. Release the FILL button.
 2. A message will appear: "RAM init." Press the GO button. Another message will appear: "RAM passed".
 3. Push the CLEAR button. The screen will then read "DOS 0.000 mL".
 4. These steps can be bypassed by simply pushing the ON-OFF button. However, if a message "error 5" appears, it is necessary to go through the above steps.
7. Use a small amount of thiosulfate to rinse out the reagent bottle on the Dosimat exchange unit. Then fill with thiosulfate. Always shake the thiosulfate reagent bottle before each session (and during if sessions are extended). 20 - 30 mL of thiosulfate should be primed through the system in the following way:
 1. If using the keyboard, touch the MODE button. Keeping hitting the MODE button until the screen reads : "DIS R". Press ENTER. "DIS R 0.000 mL" appears.
 2. Touch the VOLUME button. "V DIS 1 mL" appears on the display. Enter the volume desired and hit ENTER. Increase the rate of delivery dial to 10 for rapid delivery.

3. Hit GO and the Dosimat automatically empties and fills until that volume is delivered. The total volume delivered appears on the screen. Flick the tubing leading from the reagent bottle and to the sample to remove trapped air bubbles.

4. After the prime is completed, touch the MODE button until "DOS" appears. Hit ENTER and "DOS 0.000 LI" will appear. The Dosimat is now ready to run samples.

Note: The first prime usually requires 30 mL to clear the water and air in the tubing. If the Dosimat has sat for an extended period (>4 hours), a prime of 10 mL should be run to clear old reagent and air bubbles.

5. If using only the push-button control, hold down the button until 5 ml have been delivered. The button must be pushed again after the burette fills. Continue till the prime is completed.

WARNING: If the Dosimat is not returned to the "DOS" mode before a sample is run, the Dosimat will continue to deliver reagent until the preset volume is delivered. If you forget, turn the Dosimat off using the ON-OFF button, and restart.

8. A UW KIO₃ standard should be the main standard and should be run before every session. Any other UW standard should be run as well so that sufficient data is generated to assess the normality of the weighed standards.

Note: The main KIO₃ standard may be placed in the reagent bottle of a calibrated Dosimat, if a spare Dosimat is available. This aids in delivery of the standard and gives highly reproducible delivery of the standard. Rinse the reagent bottle with a small amount of the KIO₃ standard before filling. 5 mL glass pipettes can be used for the other standards.

9. Run the standards and blanks in distilled water. This needs to be done only at the beginning of a cruise or if discrepancies arise during a cruise. After the initial standard run in distilled water, run all standards and blanks in low O₂ seawater - usually from 600-1000 m water.

10. Standards are run according to the normal method, adding the reagents in reverse and titrating 5 mL of KIO₃ standard just to the endpoint. Another 5 mL of KIO₃ standard is then added and again titrated to the endpoint. The difference of the two (1 - 2) is the blank.

Note: The samples should be calculated using the main UW KIO₃ standard and blank run in seawater. The normality of each standard can be calculated from any other standard by the following formula:

$$\text{Normality of Std A} = \text{mL thio Std A} / \text{mL thio Std B} \times \text{Normality of Std B}$$

Check the normality of the standards using the titrations in distilled water.

11. Standards should be run every day to detect changes in the standard or the thiosulfate. Additionally, several standards should be run if the last session was more than 6 hours before.

12. Samples should be collected as soon as the CTD is safely on deck. O₂ samples are the first sampled unless prior agreement has been reached for other analyses (CFC's, pH, etc.). If the air temperature is quite warm, a tarp may be necessary to shade the CTD for sampling. Niskin water temperatures should be taken and recorded right after the O₂'s are sampled.

13. The samples should be fixed immediately. Make sure no air bubbles are present in the reagent dispensers before using. Prime several times before starting to sample the CTD.

14. Add the MnCl_2 and NaI-NaOH reagents, being sure that the dispenser tip is well below the surface of the sample bottle. The reagents are dense and sink to the bottom. Shake vigorously and allow the sediment to settle for twenty (20) minutes. Remix and store in a cool dark place. If the samples are not to be analyzed for some time, cap the sample with a layer of low O_2 seawater before storage.

15. Samples should be allowed to come to room temperature before analysis. Gently remove the overlaying water without disturbing the sediment and dry the excess.

16. If a whole box (24) of samples is to be analyzed, it is convenient to acidify all of the samples ahead of time. After removing the overlaying water, remove the stopper and add 1 mL of H_2SO_4 , placing the dispenser tip well below the surface. Do not disturb the sediment. Carefully drop the stopper back into the bottle, press down hard and check for air bubbles. If a bubble exists, repeat. Mix the sample and return to the box.

Note: Acidified samples may sit for several days. It is best to acidify just before analysis. Seawater dries on the outside of the bottle and is opaque. It is helpful to rinse the outside of the bottle with water and dry before analysis.

17. If possible, try and run a series of duplicates with other groups doing O_2 analysis. This allows for an immediate check on both systems and eliminates possible contamination errors.

18. Duplicate should be run on all samples. This is both an aid to obtaining the best possible data and a check of the system integrity (taking of samples on-deck, handling of samples, reading of endpoint, etc.).

Oxygen KIO_3 Standard Preparation

1. Always make up two standards, an Aldrich KIO_3 standard and a WAKO KIO_3 standard. The standards should be run against each other to determine the accuracy of the standards.
2. Place a small amount of bulk Aldrich and WAKO standards into glass drying dishes. Only use an amount sufficient to make one standard (~ 0.5 g). The standards are kept in a desiccator at all times.
3. Dry the standards in the drying oven at 90 - 100 °C for at least four hours. Remove the ground glass lid before drying the standards.

Note: Watch carefully as the oven heats so that the temperature does not exceed 100 °C. KIO_3 deteriorates at higher temperatures. It is best to have the oven at temperature before drying the standards.

4. At the end of the drying period replace the lid and put the drying dishes into a desiccator to cool. Wait at least 15 minutes before weighing out the standards.
5. A KIO_3 standard of 0.025N is usually made. Weigh out **~ 0.4458 grams** of KIO_3 to make up 500 mL of 0.025N standard. It is not necessary to weigh out this amount exactly.
6. Weigh out an appropriate amount of each standard. Write this amount into the notebook kept by the desiccator containing the bulk standards. Designate which standard is used.
7. Place the weighed standard into a calibrated 500 mL volumetric flask. It is best to dry the neck of the flask so that the reagent does not stick to the rim. Rinse the weighing paper and the sides of the flask to wash all of the reagent to the bottom.
8. Fill the volumetric flask exactly to the line with the bottom of the meniscus touching the line. Place a stir bar into the flask and mix on a stirrer for 20 - 30 minutes. Remove the stir bar and transfer the reagent to a clean, dry amber bottle.
9. Label the bottle with the standard name, date, weight of dried reagent and normality of the standard. Normality of the standard is calculated thus:

Aldrich KIO_3 standard:

weight of KIO_3 / .445875 g * .025 * 500 / volume of 500 mL flask * 0.997

WAKO KIO_3 standard:

weight of KIO_3 / .445875 g * .025 * 500 / volume of 500 mL flask

Note: The 0.997 term corrects for the impurity of the Aldrich KIO_3 standard which has a purity of 99.5 - 100.5%. The WAKO standard is a certified standard with a purity of 99.98%.

10. Dispose of the unused standards and replace the weighing dishes in the desiccator.

Winkler O2 Analysis Reagent Preparation

MnCl_2 - dissolve **300 grams** of reagent grade **MnCl_2** in ~ 300 mL deionized water in a 500 mL Erlenmeyer flask with stirring. Make up to 500 mL in a volumetric flask. Transfer to a 500 mL brown HDPE bottle.

NaI-NaOH - dissolve **300 grams** of **NaI** in ~ 300 mL of deionized water in a 500 mL Erlenmeyer flask with stirring. Dissolve slowly, adding only 50 - 70 grams at a time. Make sure that the reagent stays clear. Add **160 grams** of **NaOH** a little at a time. Make up to 500 mL in a volumetric flask. Transfer to a 500 mL brown HDPE bottle.

Note: This reaction is exothermic. Do not make final volumetric determination until the reagent has cooled.

H_2SO_4 - add **160 mL** of concentrated **H_2SO_4** to 250 mL of deionized water in a 500 mL volumetric flask. Place a container of ice on a stirrer. Place the flask with water in the container of ice before adding the H_2SO_4 . **Very exothermic!!** Make up to 500 mL after reagent has cooled. Transfer to a 500 mL brown HDPE bottle.

starch - dissolve **5 grams** of reagent grade **starch** in ~ 300 mL of deionized water in a 500 mL Erlenmeyer flask. Heat for 3 - 4 minutes in a microwave on high power. Stir after every minute or so. Place the flask in a container of ice on a stirrer to mix. Make up to 500 mL in a volumetric flask. Transfer to a 500 mL brown HDPE bottle.

Note: If starch precipitates out during preparation, repeat the microwave step to get it back into solution. Filter if some precipitate remains.

$\text{Na}_2\text{S}_2\text{O}_2 \cdot 5\text{H}_2\text{O}$ - dissolve **49.64 grams** of **$\text{Na}_2\text{S}_2\text{O}_2 \cdot 5\text{H}_2\text{O}$** in deionized water in a 2000 mL volumetric flask. Make up to 2000 mL. Transfer to two 1000 mL brown HDPE bottles.

Make enough reagent to run all the expected samples and half again. Each sample takes about **1.5 mL** of thiosulfate and **1.0 mL** of each of the others. The **KIO_3** standard needs to be dried at least four (4) hours at 100 °C. Make at least two different standards.

OXYGEN ANALYSIS NOTES

1. Equipment:

Dosimat(s)
keypads

- connecting cables
- exchange units
- thiosulfate bottle
- delivery tip
- SS rod
- delivery tip clamp
- stir-bars
- wash bottles
- Kimwipes
- pipettes (5 mL)
- magnet remover
- power strip(s)
- plastic waste bottles
- light(s)
- spare light bulbs

Reagents:

- MnCl₂
- NaI-NaOH
- H₂SO₄
- starch
- KIO₃ standard(s)
- pipettes (1 mL)
- Repipettes (1 mL)

Miscellaneous:

- wash basin
- bottle brushes
- tie-down rope
- eyebolts
- Tygon tubing (5/16")
- lab notebook
- O₂ analysis spreadsheet
- floppy disks
- Micronox lab soap
- office supplies
- duct tape
- blue diapers
- clipboard

Set-Up

1. Do an immediate check-list to determine that all the equipment is on board.
2. Pick an area that allows the equipment to be set up with room for reagents, pipettes, wash bottles, wiring, etc. If possible, set up the KIO₃ standard Dosimat in the center of the other two so that it is accessible to both techs. The reagents for standards can also be in the center of the counter. Use only one set of reagents at a time.
3. All equipment should be tied down before the ship leaves the dock. This includes chairs and stools, sample boxes, etc. Reagents should be placed in an area bounded by wood strips so that they cannot slide. If no wood is available, duct tape the bottles to the counter or keep in a plastic basin or box.
4. Duct tape the thiosulfate reagent bottle to the Dosimat so that it does not move.

5. Wash all of the O₂ bottles with soap and water at the start and end of the cruise: also wash each bottle after it has been used for a sample. It is not necessary to wash standard bottles with soap between uses. Rinse well and reuse.

Note: If you have used a box of bottles filled with seawater for standards, wash the bottles with soap and water before taking samples.

6. Do not leave bottles out of the box except the sample you are running. Nothing...and I mean nothing...is safe from rolling or falling.

Standards

1. Use the UW KIO₃ standard as the main standard. After designating a Dosimat for the standard, rinse the reagent bottle with a small amount of standard and then fill **half full**. This keeps the standard from sitting too long in the reagent bottle and evaporating.

Note: When refilling the standard bottle, empty the remaining into a spare brown bottle. Then refill with fresh standard as before, with a rinse first.

2. Other standards are run using glass volumetric pipettes (5 mL). These pipettes must be kept scrupulously clean; watch for spotting or drops left inside the pipette after delivery. Wash with Micro and rinse thoroughly. Rinse with DZD or ship's water after each session. Rinse twice with KIO₃ standard at start of each session.

Note: DZD may not be available at all times. After the first day at sea, the ship's water is essentially distilled water and may be used for most rinsing purposes.

3. At the start of the cruise, run all standards at least four (4) times in deionized water. Each standard should agree within +/- 3 mliters. After establishing that the system is working, switch to running standards in seawater. Use low O₂ water (600-1000 m) so that it is free of particles (plankton, algae, etc.). Again run all standards four (4) times each.

Note: It is very important to establish the reliability and integrity of the standards at the beginning of the cruise. This is the time, when the schedule permits, that allows for a thorough analysis of the system, the normality of the standards, the integrity of the reagents, and the conditions for proper reading of the endpoint. At this time only, the reagents, the standards and you are fresh! By determining the status of the system at this point, discrepancies later on can be put into context. Write down everything...date and time everything...leave nothing to memory!

4. Only use the seawater standards and blanks to calculate the samples. A spreadsheet is included that allows you to enter the depth, Niskin #, temp, salinity, bottle number, cast data, etc. At the beginning of the cruise, enter the bottle volumes as they are in the example. These are located at the top of the spreadsheet. Continue across the page, entering the bottle # in line 1 and the volume in line 2. There is a LOOKUP function in the spreadsheet that fills in the volume, given the bottle #. Check occasionally to see that the volumes match.

Note: It is easiest to copy one line from the previous calculation to a space below. Then copy this row of data down for all the samples in one cast. Change the cast data, depths, temps, etc. and enter the bottle #, mL thio for each sample. After the first cast is entered and assuming that the cast sequence stays the same, the entire cast can be copied to another place and only the pertinent data changed.

5. Check the precision of your duplicate pairs. The last two columns of the spreadsheet give the STD of the duplicate pair in microliters thiosulfate and %diff from the average. The %diff should be less than 0.2%. This may increase for low O₂ samples but high values indicate problems, either in titration, sample taking, cleanliness, etc.

Note: If a certain bottle pair (I usually start a cast with an empty box so that I am always using the same bottles for duplicates (e.g., 1 and 2, 3 and 4, etc.) gives consistently high

errors, one of the volumes may be incorrect. Check the volume in the spreadsheet. If it matches the volume sheet, do not use that duplicate pair again.

On Deck

1. Prepare a cast sheet before each cast where samples will be taken. Include the date, time, location, cast name and number, depths expected, O₂ bottle numbers, salinities (if taken), and temperatures of Niskins (if taken). Give a copy to the person in charge of the cast. Take a copy with you to sample the CTD. It is best to designate one person to be in charge of determining the sample order so that no confusion arises. Have that person read off the next Niskin to be sampled and the O₂ bottle to fill.

2. Take the O₂ fixing reagents out on deck. Make sure that the bottles are primed and that no air bubbles are being delivered. Place the reagents in a place with easy access to the CTD and tie down if necessary.

Note: Between casts, it is best to rinse the plungers with hot water to clean out the reagents. Never leave the reagents sitting in the plungers for long periods of time.

3. Oxygen samples are taken first from the Niskins, unless prior arrangements have been made for other tests (CFC's, pH's, etc.). Sample the Niskins from the deepest depths first as these change temperature the fastest. Sample the Niskins in order and have only one person do each duplicate pair.

Note: If other tests are performed before O₂, ask that person to close the siphon after their sampling.

4. Place a 5/16" Tygon tube onto the nipple of the Niskin. Open the bottle holding the bottle and stopper in your hand. Push the nipple into the Niskin and then open the siphon. Water will start to flow. Place the Tygon tube into the O₂ bottle and turn the bottle upside-down. Pinch the Tygon tube at the nipple to clear any air bubbles in the tube. Rinse the bottle until it is well rinsed and then turn right side-up, keeping the Tygon tube at the bottom. Allow 2 - 3 volumes of water to overflow the bottle. Slowly remove the tube while the water is still flowing. Pull out nipple after tube is out of the bottle.

5. Carry the bottle to the fixing reagents. Add 1 mL of MnCl₂, placing the delivery tip well below the surface. The reagents are dense and sink to the bottom. Do not press down too vigorously so that reagent swirls up to the top of the bottle. Immediately add 1 mL of NaI-NaOH and replace the stopper, making sure that no air is trapped inside the bottle. Mix well - at least 20 sharp hard shakes. Replace in box.

6. After all samples are taken, carry the sample box(es) back to the lab. After twenty (20) minutes or so, remix the bottles. Cap the bottles with a layer of low O₂ seawater for storage. Store in a cool, dark place if possible.

Note: Only run the samples after they get to room temperatures. This assures that all samples are analyzed under the same conditions.

Sample Analysis

1. After coming to room temperature, the samples are ready to be analyzed. If this is the start of a new session, standards and blanks need to be run. Several sets of two different standards should be run - 2 to 3 each, as long as they are within +/- 2 milliliters of each other.

Record the date and time of each standard run, as well as the time of analysis for each box of samples. This allows you to determine which standard and blank should be used to calculate which samples, since replacing reagents and standards during the cruise may occur.

Note: It is not necessary to run standards after each box of samples. Standards should be rerun only if 6 - 8 hours has passed since the last standard run.

2. It is easiest to do a box of samples at a time, and to acidify all the samples before analysis. Remove the overlaying water and dry the excess. Open the stopper and add 1 mL of H₂SO₄ to the sample, keeping the tip well below the surface and not disturbing the sediment. Carefully drop the stopper into the bottle, pushing down hard. Do not allow a bubble to be trapped; if this happens, repeat the process. Mix well until the sediment dissolves.

Note: Salt water dries to an opaque, oily sheen on the bottles. Rinse the bottle under tap water and dry. This makes seeing the endpoints easier.

3. Once the entire box is acidified, it is ready for titration. The bottles can sit acidified for up to a day or more, but should be done as soon as possible. If no sink is close by, have a basin or bucket nearby.

4. Transfer the cast information (cast #, depth, Niskin #, bottle #) to a data analysis sheet. Record the start time for each box of samples so the correct standards and blanks may be used in calculation.

5. Remove a sample from the box, take out the stopper and rinse the stopper into the bottle with DZD. Place a magnet in the bottle and place on the stirrer. Place the tip of the titrator just under the surface. If the sample appears to have a low O₂ concentration, start with the delivery dial on 2; otherwise, start on 4. Titrate down to a pale yellow color. Add 1 mL of starch - do not deliver directly into the bottle but along the sides, so that no bubbles are entrained. Complete the titration. Record the mL thio in the data analysis sheets.

6. If a sink is nearby, empty the sample bottle into the sink, making sure that the magnet is not lost (a magnet remover helps). Rinse once with DZD and replace in the box. After the box is completed, wash all bottles in soap and water. If a sink is not nearby, empty the sample into a wash basin or bucket, rinse with DZD from a wash bottle and replace in the box.

G2. Alkalinity Analyses

CO₂ Sampling and Analyses

<http://www.pmel.noaa.gov/co2/story/Laboratory+analysis+details>

<http://hahana.soest.hawaii.edu/hot/protocols/chap24.html>

G3. pH Analyses

G4. Salinity Analyses

Directly from JGOFS Protocols Manual 1994 (JGOFS Report Nr. 19)

Chapter 5. Salinity Determination

1.0 Scope and field of application

This procedure describes the method for the determination of seawater salinity. The method is suitable for the assay of oceanic levels (0.005–42). The method is suitable for the assay of oceanic salinity levels of 2–42. This method is a modification of one published by Guildline Instruments (1978).

2.0 Definition

The method determines the practical salinity (S) of seawater samples which is based on electrical conductivity measurements. The Practical Salinity Scale 1978 (PSS 78) defines the practical salinity of a sample of seawater in terms of the conductivity ratio (K15) of the conductivity of the sample at a temperature of 15°C and pressure of one standard atmosphere to that of a potassium chloride (KCl) solution containing 32.4356 g of KCl in a mass of 1 kg of solution.

3.0 Principle

A salinometer is used to measure the conductivity ratio of a sample of seawater at a controlled temperature. The sample is continuously pushed through an internal conductivity cell where electrodes initiate signals that are proportional to the conductivity of the sample. Using an internal preset electrical reference, these signals are converted to a conductivity ratio value. The number displayed by the salinometer is twice the conductivity ratio. The internal reference is standardized against the recognized IAPSO standard seawater.

4.0 Apparatus

Guildline model 8400A Autosol Salinometer. The Autosol has a 4 electrode cell which measures the conductivity ratio of a sample seawater in less than one minute. The salinity range of the instrument is about 0.005–42 and has a stated accuracy of ± 0.003 by the manufacturer. In practice, accuracies of 0.001 are possible with careful analysis.

5.0 Reagents

IAPSO Standard Seawater. Standard seawater for instrument calibration.

6.0 Sampling

Salinity samples are collected from Niskin bottles at all depths. These samples are collected after the oxygen and CO₂ samples have been drawn. The bottles used are 125 and 250 mL borosilicate glass bottles with plastic screw caps. A plastic insert is used in the cap to form a better seal. The remaining sample from the previous use is left in the bottles between uses to prevent salt crystal buildup from evaporation and to maintain an equilibrium with the glass. When taking a new sample, the old water is discarded and the bottle is rinsed three times with water from the new sample. It is then filled to the bottle shoulder with sample. The neck of the bottle and inside of the cap are dried with a Kimwipe. The cap is then replaced and firmly tightened. These samples are stored in a temperature controlled laboratory for later analysis (1–5 days after collection). Every six months the bottles are acid washed (1 M HCl), rinsed with deionized and Milli-Q water. After this cleaning they are rinsed five times with copious amounts of sample before filling.

7.0 Procedures

The samples are analyzed on a Guildline AutoSal 8400A laboratory salinometer using the manufacturer's recommended techniques.

The salinometer is calibrated with IAPSO standard seawater. Two standards are run prior to running the samples. If those two standards agree, the samples are run. At the end of the run, two new standards are run to check for instrument drift. The drifts are generally found to be zero. Using this procedure, the instrument can give a salinity precision of ± 0.001 –0.002.

8.0 Calculation and expression of results

The calculation of salinity is based on the 1978 definition of practical salinity (UNESCO, 1978). The following gives the necessary computation to calculate a salinity (S) given a conductivity ratio determined by the salinometer:

$$S = a_0 + a_1 R_T + a_2 R_T^{\frac{1}{2}} + a_3 R_T^{\frac{3}{2}} + a_4 R_T^2 + a_5 R_T^{\frac{5}{2}} + [(T-15)/(1+kT-15)] \{ b_0 + b_1 R_T + b_2 R_T^{\frac{1}{2}} + b_3 R_T^{\frac{3}{2}} + b_4 R_T^2 + b_5 R_T^{\frac{5}{2}} \}$$

where:

$$\begin{aligned} a_0 &= 0.0080 \\ a_1 &= -0.1692 \\ a_2 &= 25.3851 \\ a_3 &= 14.0941 \\ a_4 &= -7.0261 \\ a_5 &= 2.7081 \end{aligned}$$

$$k = 0.0162$$

$$\begin{aligned} b_0 &= 0.0005 \\ b_1 &= -0.0056 \\ b_2 &= -0.0066 \\ b_3 &= -0.0375 \\ b_4 &= 0.0636 \\ b_5 &= -0.0144 \end{aligned}$$

R_T = conductivity ratio of sample (=0.5 salinometer reading)

T = bath temperature of salinometer (°C)

$$\sum_{i=0}^5 a_i = 35.0000$$

$$\sum_{i=0}^5 b_i = 0.0000$$

for:

$$-2^{\circ}\text{C} \leq T \leq 35^{\circ}\text{C}$$

$$2 \leq S \leq 42$$

9.0 Quality assurance

9.1 *Quality control:* The bottle salinities are compared with the downcast CTD profiles to search for possible outliers. The bottle salinities are plotted against potential temperature and overlaid with the CTD data. Historical envelopes from the time-series station are further overlaid to check for calibration problems or anomalous behavior.

9.2 *Quality assessment:* Deep water samples (>3000 m) are duplicated. These replicate samples are found to agree in salinity of ± 0.001 .

9.3 Regular inter-calibration exercises should be performed with other laboratories.

10.0 References

Guideline Instruments. (1981). Technical Manual for 'Autosal' Laboratory Salinometer Model 8400.

UNESCO. (1978). *Technical Papers in Marine Science*, **28**, 35pp.

G6. Nutrient Analyses

Directly from JGOFS Protocols Manual 1994 (JGOFS Report Nr. 19) Chapters
9,10,11,12

G7. Chlorophyll Analyses

Chlorophyll HOTS

<http://hahana.soest.hawaii.edu/hot/protocols/chap12.html>

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Chapter 24 FLUOROMETRIC ANALYSIS OF CHLOROPHYLL A AND PHAEOPIGMENTS

SUMMARY: Seawater is collected from known depths using CTD-rosette sampling procedures. Subsamples are filtered onto glass fiber filters and placed into cold, 100% acetone to extract photosynthetic pigments. Concentrations of chlorophyll a and phaeopigments are measured by fluorometry.

24.1. Rationale and Assay Principle

A common feature of photosynthetic microorganisms in the sea, both *Bacteria* and *Eucarya*, is chlorophyll a (Chl a), which functions as a major light-harvesting pigment. This observation provides the primary justification for estimating photoautotrophic biomass from measurements of Chl a. Chl a can be easily extracted from particulate matter using acetone and measured with a high level of precision and with adequate sensitivity to obtain quantitative determinations (Holm-Hansen et al., 1965; Holm-Hansen and Riemann, 1978). Although Chl a can be measured by fluorometry, spectrophotometry or HPLC, fluorometry is widely used in oceanographic field studies. HPLC methods (see Chapter 21) can isolate and quantify the entire pigment-biomarker spectrum, including the major chlorophylls (monovinyl and divinyl Chl a, monovinyl and divinyl Chl b, Chl c), carotenoids and most other accessory pigments (Mantoura and Llewellyn, 1983; Latasa et al., 1996). Such a detailed analysis is essential when using pigment signatures for taxonomic characterization of the photoautotrophic microbial assemblage.

The half-life of Chl a after cell death has not been well studied. It is generally believed that Chl a is rapidly decomposed by a combination of bacterial degradation and, at least in near surface waters, by photochemical processes. Nelson (1993) estimated a half-life of approximately 10 d for detrital pigments, assuming that the primary removal mechanism is photo-oxidation. The presence of cell-free Chl a is a potential limitation to its use as a biomass-indicator molecule.

The primary limitation of this biomarker approach is that the Chl a : C ratio can vary significantly and systematically among algal species and within a given organism as a function of many factors, including time, cell size, temperature, nutrient conditions, growth rate, and especially, light history (reviewed by Falkowski, 1981). Variations of 26-fold have been observed for the Chl a : C ratio of a single algal species as a function of environmental conditions and growth rate (Laws et al., 1983). Consequently, reliable extrapolation of Chl a concentrations to photoautotrophic biomass is dependent upon the availability of additional information on species composition, nutrient concentration and limitation, light levels and growth rate.

In the HOT program, Chl *a* is measured both by fluorometry and by high performance liquid chromatography (see Chapter 21). In the fluorometric determination, concentrated particulate matter is immersed into absolute acetone and the fluorescence of a portion of the solvent extract, containing the photoautotrophic pigments, is read in a filter fluorometer.

24.2. Sampling, Filtration, Extraction and Storage

- 24.2.1. Seawater samples are collected at standard water depths using 12 l PVC water bottles attached to the CTD-rosette sampler and from the primary productivity cast (see Chapter 25) using Go-Flo bottles attached to a Kevlar line.
- 24.2.2. Subsamples from each bottle are transferred to clean volume-calibrated 125 mL opaque HDPE bottles. The HDPE bottles are rinsed three times with 100-200 mL of sample before the collection of the final sample.
- 24.2.3. Filtration and storage
 - The subsamples are filtered through 25 mm GF/F filters.
 - Immediately following filtration, the filter is transferred to a glass screw cap tube containing 5 mL of cold (-20°C) 100% acetone. Then the tubes are wrapped in aluminum foil and stored at -20°C to prevent photodegradation of pigments.
- 24.2.4. Sample Analysis
 - Samples are brought to room temperature 2 hr. before the analysis and the fluorometer is allowed to stabilize for 30 min before the samples are analyzed.
 - The external standard, prepared with reagent grade Chl *a* (see section 20.3), is read before other samples.
 - Samples are analyzed from deepest to shallowest water depth. Each time the door of the fluorometer is changed (i.e., instrument sensitivity), the dial is brought to the "zero" setting using 100% acetone as a blank.
 - Each sample is acidified by adding 2 drops of HCl (1 M) to the sample in the cuvette. The new reading must be stable before recording the value and is made using the same door used for the reading before the acidification.
 - The fluorometer door used, the readings before and after acidification and the dilution factor are recorded in the chlorophyll log book.

24.3. Standard Preparation and Analysis

- 24.3.1. A Chl *a* standard stock is prepared every 4 months using commercially available Chl *a* (Sigma Chemical Co.) and 100% acetone. The stock is wrapped in aluminum foil to prevent photo-degradation and stored at -20°C.
- 24.3.2. To determine the concentration of the stock, a 50 mL sample is brought to room temperature avoiding exposure to the light. An absorption spectrum from 350-750 nm is obtained using a scanning spectrophotometer and the concentration is calculated assuming an extinction coefficient of 88.15 (l g⁻¹ cm⁻¹) for the absorbance at 662 nm.

- 24.3.3. Using the same stock sample, three serial dilutions are prepared gravimetrically using 100% acetone and read in the fluorometer. During this step, Chl *a* must be kept under dim light.
- 24.3.4. A calibration of the fluorometer is performed every 6 months following the procedure described by Strickland and Parsons (1972).

24.4. Data Reduction and Calculations

Concentrations of Chl *a* and phaeopigments are calculated using the following equations:

$$\text{Chl } a = (T/(T-1)) \cdot (R_b - R_a) \cdot F_d \cdot \text{vol}_{\text{ex}} / \text{vol}_{\text{filt}}$$

$$\text{phaeo} = (T/(T-1)) \cdot ((T \cdot R_a) - R_b) \cdot F_d \cdot \text{vol}_{\text{ex}} / \text{vol}_{\text{filt}}$$

where: Chl *a* = concentration of Chl *a* (mg m⁻³)
phaeo = concentration of phaeopigments (mg m⁻³)
T = acidification coefficient (R_b/R_a average obtained during the calibration of the fluorometer)
R_b = reading before acidification
R_a = reading after acidification
F_d = door factor (μ g/(mL* reading units))
vol_{ex} = volume of extraction (mL)
vol_{filt} = volume filtered (l)

24.5. Equipment/Supplies

filtration system
glassware freezer
fluorometer (Turner model 111)
spectrophotometer (Varian model DMS-100S)

24.6. Reagents

DDW
Filtered seawater
acetone (100%)
HCl (1 M)
chlorophyll *a* standard (Sigma Chemical Co., #_____)

24.7. References

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Directly from JGOFS Protocols Manual 1994 (JGOFS Report Nr. 19)

Chapter 14. Measurement of Chlorophyll *a* and Phaeopigments by Fluorometric Analysis

1.0 Scope and field of application

Chlorophyll *a* measurements have historically provided a useful estimate of algal biomass and its spatial and temporal variability. The fluorometric method is extensively used for the quantitative analysis of chlorophyll *a* and phaeopigments. However, errors can be introduced into the results when chlorophylls *b* and/or chlorophylls *c* are present. Chlorophyll *b* is the main source of error in this method. While generally not abundant in surface waters, chlorophyll *b* can be as high as 0.5 times the Chl *a* concentration in the deep chlorophyll maximum, causing slight underestimations of the Chl *a* concentration, and drastic overestimations of the phaeopigment concentrations. Divinyl-chl *a* also interferes and is taken as Chl *a* by this method. The procedure described here is appropriate for all levels of Chl *a* concentration in the marine environment. Filtration volumes should be modified for the different environments. Scientists who employ this or other methods to measure pigments should make themselves aware of the current and historical issues that surround these techniques and make appropriate decisions about specific methodologies for their application based on the scientific requirements and constraints of their individual programs.

2.0 Definition

The concentrations of chlorophyll *a* and phaeopigments in seawater are given as $\mu\text{g kg}^{-1}$.

3.0 Principle of Analysis

Algal pigments, particularly chlorophyll *a*, fluoresce in the red wavelengths after extraction in acetone when they are excited by blue wavelengths of light. The fluorometer excites the extracted sample with a broadband blue light and the resulting fluorescence in the red is detected by a photomultiplier. The significant fluorescence by phaeopigments is corrected for by acidifying the sample which converts all of the chlorophyll *a* to phaeopigments. By applying a measured conversion for the relative strength of chlorophyll and phaeopigment fluorescence, the two values can be used to calculate both the chlorophyll *a* and phaeopigment concentrations.

4.0 Apparatus

Filtration system and Whatman GF/F filters
Liquid nitrogen and freezer for storage and extraction
Glass centrifuge tubes for extraction, 15 mL
Turner fluorometer, fitted with a red sensitive photomultiplier, a blue lamp, 5-60 blue filter and 2-64 red filter.

5.0 Reagents

100% acetone
90% acetone
1.2M HCl (100 mL HCl in 900 mL de-ionized water)

6.0 Sample Collection and Storage

Water samples are collected from Niskins into clean polyethylene bottles with Tygon® tubing.

Samples are immediately filtered through 47 mm GF/F filters using polycarbonate in-line filters (Gelman) and a vacuum of less than 100 mm Hg. Filters are folded in half twice and wrapped in aluminum foil, labeled, and stored in liquid nitrogen (to avoid formation of degradation products) until shore analysis. Alternatively, filters can be placed immediately in acetone for pigment extraction if analysis is to be carried out onboard ship.

In oligotrophic waters, for this measurement coupled with HPLC determined pigments, 4 liters are filtered. For fluorometric analysis alone, a smaller volume (0.5 -1.0 L) may be sufficient. In coastal regions, a volume of 0.1-0.5 L may be adequate. In this case, use of 25 mm GF/F filters may be appropriate.

7.0 Procedure

7.1 After removal from liquid nitrogen or freezer), the pigments are extracted by placing the filters in 5.0 mL 100% acetone. For 47 mm GF/F filters, 0.8 ml of water is retained adjusting the final extraction solution to 86% acetone and the final extraction volume to 5.8 mL. The samples are covered with Parafilm to reduce evaporation, sonicated (0°C, subdued light) and allowed to extract for 4 hours in the dark at -20°C. Following extraction, samples are vortexed, filters are pressed to the bottom of the tube with a stainless steel spatula and spun down in a centrifuge for 5 minutes to remove cellular debris. For fluorometric analysis (not HPLC), decantation can replace centrifuging.

7.1.1 The addition of 5.0 mL acetone for pigment extraction is necessary to completely submerge 47 mm GF/F filters in 15 mL centrifuge tubes. This volume may be altered depending on the size of the filter and volume of the extraction tube.

7.2 The fluorometer is allowed to warm up and stabilize for 30 minutes prior to use.

7.3 The fluorometer is zeroed with 90% acetone.

7.4 1.0 mL of pigment extract is mixed with 4.0 mL 90% acetone in a cuvette and read on the appropriate door to give a reading between 30 and 100. The sample is then acidified with 2 drops of 1.2 M HCl. Further dilutions may be necessary for higher chlorophyll *a* concentrations.

7.5 Standardization

7.5.1 For laboratory use, the fluorometer is calibrated every 6 months with a commercially available chlorophyll *a* standard (*Anacystis nidulans*, Sigma Chemical Company). If the fluorometer is taken to sea, it is recommended that the fluorometer be calibrated before and after each cruise.

7.5.2 The standard is dissolved in 90% acetone for at least 2 hours and its concentration ($\mu\text{g L}^{-1}$) is calculated spectrophotometrically as follows:

$$\text{Chl } a = \frac{(A_{\text{max}} - A_{750 \text{ nm}})}{E \times l} \times \frac{1000 \text{ mg}}{1 \text{ g}}$$

where:

A_{max} = absorption maximum (664 nm)

$A_{750 \text{ nm}}$ = absorbance at 750 nm to correct for light scattering

E = extinction coefficient for Chl *a* in 90% acetone at 664 nm ($87.67 \text{ L g}^{-1} \text{ cm}^{-1}$)

l = cuvette path length (cm)

7.5.3 From the standard, a minimum of five dilutions are prepared for each door. Fluorometer readings are taken before and after acidification with 2 drops 1.2 M HCl.

7.5.4 Linear calibration factor (K_x) are calculated for each door (x) as the slope of the unacidified fluorometric reading vs. chlorophyll *a* concentration calculated spectrophotometrically.

7.5.5 The acidification coefficient (F_m) is calculated by averaging the ratio of the unacidified and acidified readings (F_o/F_a) of pure chlorophyll *a*.

7.5.6 Samples are read using a door setting that produces a dial reading between 30 and 100. The fluorometer is zeroed with 90% acetone each time the door setting is changed.

8.0 Calculation and expression of results

The concentrations of chlorophyll *a* and phaeopigments in the sample are calculated using the following equations:

$$\text{Chl } (\mu\text{g L}^{-1}) = [F_m/(F_m-1)] \times (F_o-F_a) \times K_x \times (\text{vol}_{\text{ex}}/\text{vol}_{\text{filt}})$$

$$\text{Phaeo (Chl equiv. weights)} = (F_m/(F_m-1)) \times [(F_m \times F_a)-F_o] K_x - \text{vol}_{\text{ex}}$$

where:

F_m	=	acidification coefficient (F_o/F_a) for pure Chl <i>a</i> (usually 2.2)
F_o	=	reading before acidification
F_a	=	reading after acidification
K_x	=	door factor from calibration calculations
vol_{ex}	=	extraction volume
vol_{filt}	=	sample volume

9.0 References

- Herbland, A., A. Le Bouteiller, and P. Raimbault. (1985). Size structure of phytoplankton biomass in the equatorial Atlantic Ocean. *Deep-Sea Res.*, **32**: 819-836.
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