Battelle
The Business of Innovation

Environmental Technology Verification Program

Advanced Monitoring Systems Center

Test/QA Plan for Verification of Ballast Water Exchange Screening Tools

Verification of
Ballast Water Exchange Screening Tools

January 30, 2007

Prepared by

Battelle
505 King Avenue
Columbus, OH 43201-2693

A2 TABLE OF CONTENTS

List of Figures Page

List of Tables

ETV Advanced Monitoring Systems Center

Test/QA Plan for Verification of Ballast Water Exchange Screening Tools

January 30, 2007
APPROVAL:

Name _____

Company _____

Date _____

A3 DISTRIBUTION LIST

Elizabeth A. Betz
U.S. Environmental Protection Agency-HEASD
National Exposure Research Laboratory
E205-01 EPA Mailroom
Research Triangle Park, NC 27711

Robert Fuerst
U.S. Environmental Protection Agency-HEASD
National Exposure Research Laboratory
D205-05 EPA Mailroom
Research Triangle Park, NC 27711

Gail Roderick
NIS Program
USCG R&D Center
1082 Shennecossett Road
Groton, CT 06340

Travis Martin
Dakota Technologies, Inc
2201 A 12 St N
Fargo, ND 58102

Jeanine Boyle
Rosanna Buhl
Mark Curran
Amy Dindal
Carlton Hunt
Clare Larson
Mary Schrock
Zachary Willenberg
Battelle
505 King Ave.
Columbus,OH 43201

SECTION A
PROJECT MANAGEMENT

A4 VERIFICATION TEST ORGANIZATION

The verification test will be conducted under the auspices of the U.S. Environmental Protection Agency (EPA) through the Environmental Technology Verification (ETV) Program. It will be performed by Battelle, which is managing the ETV Advanced Monitoring Systems (AMS) Center through a cooperative agreement with EPA. The scope of the AMS Center covers verification of monitoring technologies for contaminants and natural species in air, water, and soil.

The day to day operations of this verification test will be coordinated and supervised by Battelle, with the participation of the vendors who will be having the performance of their technologies for screening ballast water exchange verified. Testing will be conducted at Battelle in Columbus, Ohio. Each vendor will provide Battelle with their respective technology and will train the Battelle staff in their technology use. Battelle staff will operate the technologies during verification testing.

The organization chart in Figure 1 identifies the responsibilities of the organizations and individuals associated with the verification test. Roles and responsibilities are defined further below. Quality Assurance (QA) oversight will be provided by the Battelle Quality Manager and also by the EPA AMS Center Quality Manager, at her discretion.

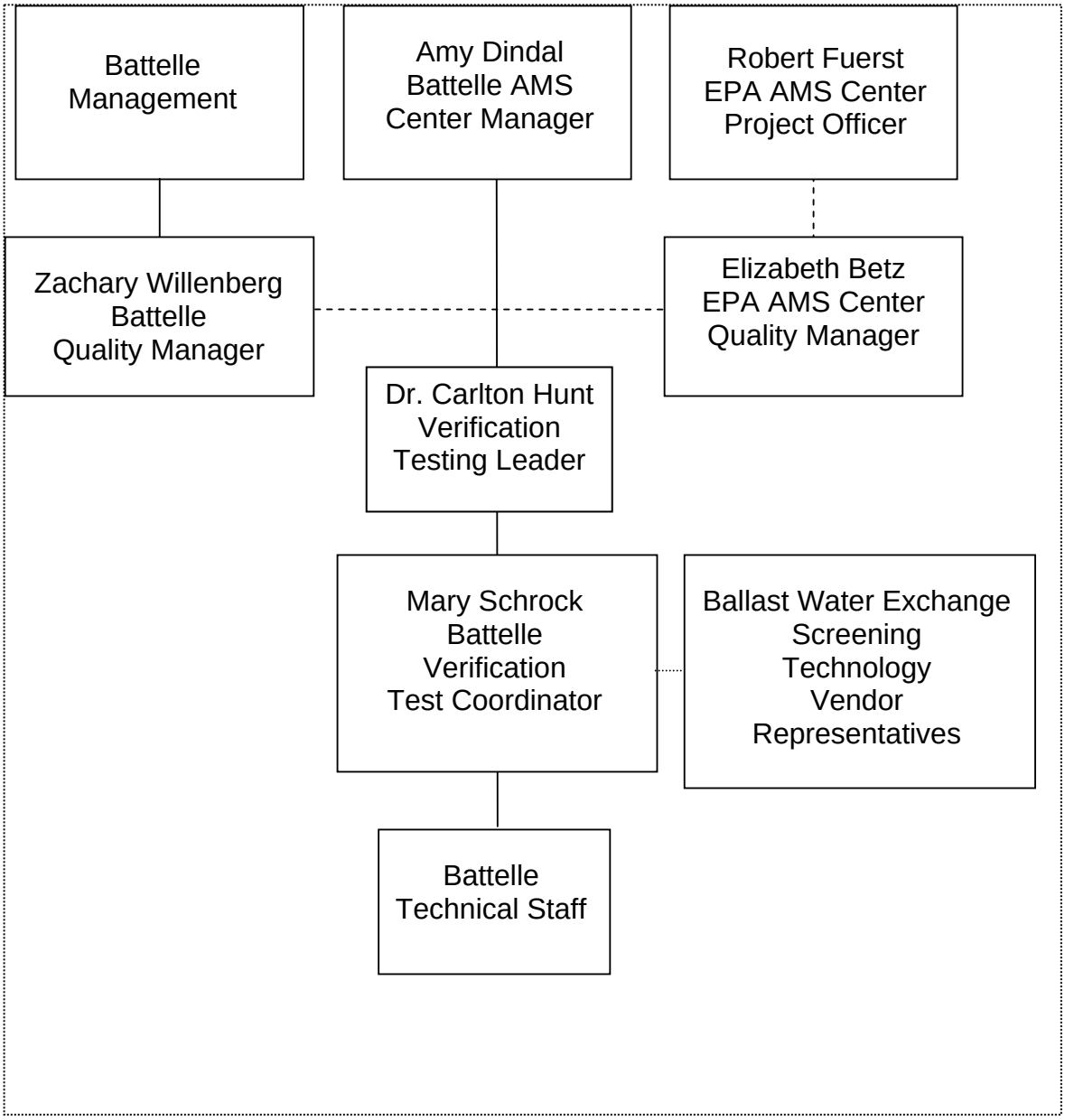

Figure 1. Organization Chart for the Verification Test

A4.1 Battelle

Ms. Mary Schrock is the AMS Center's Verification Test Coordinator for this test. In this role, Ms. Schrock will have overall responsibility for ensuring that the technical, schedule, and cost goals established for the verification test are met. Specifically, Ms. Schrock will:

- Prepare the draft test/QA plan, verification reports, and verification statements.
- Establish a budget for the verification test and manage staff to ensure the budget is not exceeded.
- Revise the draft test/QA plan, verification reports, and verification statements in response to reviewers' comments.
- Assemble a team of qualified technical staff to conduct the verification test.
- Direct the team in performing the verification test in accordance with this test/QA plan.
- Hold a kick-off meeting approximately one week prior to the start of the verification test to review the critical logistical, technical, and administrative aspects of the verification test. Responsibility for each aspect of the verification test will be confirmed.
- Ensure that all quality procedures specified in this test/QA plan and in the AMS Center Quality Management Plan[1] (QMP) are followed.
- Serve as the primary point of contact for vendor representatives.
- Ensure that confidentiality of sensitive vendor information is maintained.
- Assist vendors as needed during verification testing.
- Become familiar with the operation and maintenance of the technologies through instruction by the vendors, if needed.
- Respond to any issues raised in assessment reports, audits, or from test staff observations, and institute corrective action as necessary.
- Coordinate distribution of the final test/QA plan, verification reports, and verification statements.

Dr. Carlton Hunt will serve as Verification Testing Leader. Dr. Hunt will:

- Support Ms. Schrock in preparing the test/QA plan and designing the testing.
- Review the draft and final test/QA plan.

- Attend the verification test kick-off meeting.
- Review the draft and final verification reports and verification statements.

Ms. Amy Dindal is Battelle's Manager for the AMS Center. As such, Ms. Dindal will oversee the various stages of verification testing. Ms. Dindal will:

- Review the draft and final test/QA plan.
- Attend the verification test kick-off meeting.
- Review the draft and final verification reports and verification statements.
- Ensure that necessary Battelle resources, including staff and facilities, are committed to the verification test.
- Ensure that confidentiality of sensitive vendor information is maintained.
- Support Ms. Schrock in responding to any issues raised in assessment reports and audits.
- Maintain communication with EPA's technical and quality managers.
- Issue a stop work order if Battelle or EPA QA staff discovers adverse findings that will compromise test results.

Technical staff from Battelle will support Ms. Schrock in planning and conducting the verification test. The responsibilities of the technical staff will be to:

- Assist in planning for the test, and making arrangements for the receipt of and training on the technologies.
- Attend the verification test kick-off meeting.
- Assist vendor staff as needed during technology receipt and training.
- Conduct verification testing using the vendor's technology, if necessary.
- Conduct reference testing.
- Perform statistical calculations specified in this test/QA plan on the technology data as needed.
- Provide results of statistical calculations and associated discussion for the verification reports as needed.
- Support Ms. Schrock in responding to any issues raised in assessment reports and audits related to statistics and data reduction as needed.

Mr. Zachary Willenberg is Battelle's Quality Manager for the AMS Center. Mr. Willenberg will:

- Review the draft and final test/QA plan.
- Attend the verification test kick-off meeting.
- Conduct a technical systems audit at least once during the verification test, or designate other QA staff to conduct the audit.
- Audit at least 10% of the verification data or designate other QA staff to conduct the data audit.
- Prepare and distribute an assessment report for each audit.
- Verify implementation of any necessary corrective action.
- Request that Battelle's AMS Center Manager issue a stop work order if audits indicate that data quality is being compromised.
- Provide a summary of the QA/QC activities and results for the verification reports.
- Review the draft and final verification reports and verification statements.

A4.2 Technology Vendors

The responsibilities of the technology vendors are as follows:

- Review and provide comments on the draft test/QA plan.
- Accept (by signature of a company representative) the final test/QA plan prior to test initiation.
- Provide two units of their technology for evaluation during the verification test.
- Provide all other equipment/supplies/reagents/consumables needed to operate their technology for the duration of the verification test.
- Supply training on the use of the technology, and provide written consent and instructions for test staff to carry out verification testing, including written instructions for routine operation of their technology.
- Provide maintenance and repair support for their technology, on-site if necessary, throughout the duration of the verification test.
- Review and provide comments on the draft verification report and statement for their respective technology.

A4.3 EPA

EPA's responsibilities in the AMS Center are based on the requirements stated in the "Environmental Technology Verification Program Quality Management Plan" (EPA QMP).[2] The roles of specific EPA staff are as follows:

Ms. Elizabeth Betz is EPA's AMS Center Quality Manager. For the verification test, Ms. Betz will:

- Review the draft test/QA plan.
- Perform at her option one external technical systems audit during the verification test.
- Notify the EPA AMS Center Project Officer of the need for a stop work order if the external audit indicates that data quality is being compromised.
- Prepare and distribute an assessment report summarizing results of the external audit.
- Review draft verification reports and verification statements.

Mr. Robert Fuerst is EPA's Project Officer for the AMS Center. Mr. Fuerst will:

- Review the draft test/QA plan.
- Approve the final test/QA plan.
- Review the draft verification reports and verification statements.
- Oversee the EPA review process for the test/QA plan, verification reports, and verification statements.
- Coordinate the submission of verification reports and verification statements for final EPA approval.

A5 BACKGROUND

The ETV Program's AMS Center conducts third-party performance testing of commercially available technologies that detect or monitor natural species or contaminants in air, water, and soil. The purpose of ETV is to provide objective and quality assured performance data on environmental technologies, so that users, developers, regulators, and consultants can make informed decisions about purchasing and applying these technologies. Stakeholder committees of buyers and users of such technologies recommend technology categories, and technologies within those categories, as priorities for testing. Technologies for ballast water exchange screening tools were identified as a priority technology category through the AMS Center

stakeholder process since they have the potential to make the evaluation of ballast water exchange monitoring procedures efficient and timely.

Mid-ocean ballast water exchange (BWE) will become mandatory for all vessels entering U.S. waters from outside the 200-mile exclusive economic zone. To support such regulation, accurate and portable verification tools are needed for determining that ballast water exchange has taken place. One parameter which has been proposed as a means of distinguishing between coastal and open ocean water content in ballast water is colored dissolved organic matter (CDOM). [3,4,5] CDOM refers to the fraction of dissolved organic matter that absorbs light and fluoresces in the ultra-violet (UV) and visible regions of the spectrum. This test/quality assurance (QA) plan provides procedures for a verification test of BWE screening tools which use CDOM fluorescence to evaluate ballast water exchange. The objective of this verification test is to evaluate the performance of BWE screening tools in measuring CDOM relative to a standard CDOM measurement approach using laboratory bench scale excitation-emission spectrometry under controlled laboratory conditions. This test will not verify that the technologies successfully quantify CDOM concentrations or detect ballast water exchange, but rather how well the technologies measure fluorescence from CDOM compared to a standard technique for measuring fluorescence. This test will also not represent all types of waters which may be encountered in ballast water screening, but will attempt to represent a range of water (and subsequently the range of fluorescence measurements generated by various types of water) that may be expected in practical application. Critical characteristics of the BWE screening tool technologies that will be assessed during this testing include the following:

- Accuracy
- Linearity
- Precision
- Method Detection Limit
- Inter-unit reproducibility
- Temperature effects
- Matrix effects
- Data completeness
- Operational factors such as ease of use and maintenance.

A6 VERIFICATION TEST DESCRIPTION AND SCHEDULE

A6.1 Summary of Technology Category

Technologies to be evaluated in the verification of BWE screening tools include those which provide a portable, quick measurement of CDOM. The technologies function by measuring the amount of fluorescent CDOM in ballast water. CDOM has been proposed as a means of distinguishing between coastal and open ocean water content in ballast water.[3,4,5] This verification test will not evaluate whether the CDOM measurement is capable of detecting ballast water exchange, or even if the CDOM measurement accurately measures CDOM in water, rather it will evaluate whether the screening tool CDOM measurements are comparable to CDOM measurements obtained by a standard technique. The screening tool CDOM measurements will be reported via a digital display or electronic output signal.

A6.2 Verification Test Schedule

Table 1 shows the planned schedule of testing and data analysis/reporting activities to be conducted in this verification test. The planned dates for conducting verification tests of BWE screening tools are January – March 2007 at Battelle's laboratories in Columbus, Ohio. It will be necessary for participating vendors to provide their technologies to Battelle by a specified date so testing staff may become familiar with operating the units before testing begins. Vendor staff will provide training in operating the technologies either in person or by teleconference. The period of operation for verification testing will be approximately two to four weeks. The test procedures are described in Section B of this test/QA plan.

Subsequent to the verification test, a separate verification report will be drafted for each participating technology. These reports will be reviewed by the respective vendor and by peer reviewers, and submitted to EPA for final signature. Technologies and associated equipment (but not consumables) will be returned to the vendors at the completion of report writing, unless other arrangements have been made with Battelle.

Table 1. Planned Verification Test Schedule

Dates	Testing Activities	Data Analysis and Reporting
January-February 2007	Set up of ballast water exchange screening tools and training of verification testing staff on technology use.	
January –March 2007	Conduct verification testing.	Review and compile test data and records as they become available. Review and summarize verification testing staff observations.
February-March 2007		Prepare report templates and complete common sections of reports. Evaluate and analyze data generated during testing.
March- April 2007		Complete draft reports and submit for vendor and peer reviews.
April-May 2007	Return equipment to vendors.	Revise draft reports and submit final reports for EPA approval.

A6.3 Test Site

Laboratory analyses will be conducted in Battelle laboratories in Columbus, Ohio. There will be no field portability testing during this technology verification.

A6.4 Health and Safety

Battelle will conduct all verification testing and reference CDOM measurements following the safety and health protocols in place for the laboratory and facilities. This includes maintaining a safe work environment and a current awareness of handling potentially toxic chemicals. Exposure to potentially toxic chemicals will be minimized, personal protective equipment will be worn, and safe laboratory practices will be followed.

A7 QUALITY OBJECTIVES AND CRITERIA FOR MEASUREMENT DATA

In performing the verification test, Battelle will follow the technical and QA procedures specified in this test/QA plan and will comply with the data quality requirements in the AMS Center QMP. [1] The objective of this verification test is to evaluate the performance of BWE screening tools in their ability to measure CDOM versus a standard CDOM measurement technique under controlled laboratory conditions. This evaluation will assess the capabilities of the BWE screening technologies in both laboratory prepared and real-world open ocean and

coastal environmental samples (no actual ballast water samples are anticipated), and will include a comparison between the BWE screening tool results and those of a standard technique as described in Section B4. Additionally, this verification test will rely upon verification testing staff observations to assess other performance characteristics of the BWE screening tools. Below is a discussion of the quality objectives and the criteria for measurement data that have been established to assure that the test objectives are met.

A7.1 Quality Objectives

Data quality objectives indicate the minimum data quality required to meet the BWE screening tool verification objectives. Data quality objectives for this verification test include those related to the reference method performance, those related to the BWE screening tool technology performance, as well as those related to documenting verification testing staff observations. Data quality objectives for the reference method (see Section B4) are presented in terms of data quality indicator (DQI) criteria for the critical measurements associated with the reference method and are listed in Table 2 and discussed in Section A7.2. The reference method data quality relies, in part, on proper sample preparation, proper application of the reference method, and proper maintenance of reference method instrumentation. Battelle will rely on the vendor's data quality objectives for each BWE screening tool in order to insure that the screening technology is performing properly during testing. This will include adhering to each vendor's criteria for calibration and performance of positive and negative control samples. The screening technology data quality relies on proper operation and maintenance of the BWE screening tools and proper sample preparation. Quantitative data quality objectives for the operator observations have not been defined but are incorporated into documentation requirements and data review, verification, and validation requirements for this verification test.

A7.2 Criteria for Measurement Data

Table 2 presents the DQIs and criteria for the reference method critical measurements. The reference method measurement quality will be assured by adherence to these DQI criteria. The reference method measurement quality will be monitored by comparing the Varian Cary Eclipse spectrometer output at the wavelengths of interest with calibration standards (naphthalene-anthracene) traceable to National Institute of Standards and Technology (NIST) standards, observation of quinine sulfate at reference levels, and by inclusion of negative control

samples and positive control samples for stability monitoring. Criteria for the BWE screening tool technologies for critical measurements related to calibration standards and recommendations for appropriate positive and negative controls and their critical measurements will be provided by each vendor. The Battelle Quality Manager or his designee will perform a technical systems audit (TSA) at least once during this verification test to review these QA/QC requirements. The EPA AMS Center Quality Manager also may conduct an independent TSA, at her discretion.

A8 SPECIAL TRAINING/CERTIFICATION

Documentation of training related to technology testing, data analysis, and reporting is maintained for all Battelle technical staff in training files at their respective Battelle location. The Battelle Quality Manager may verify the presence of appropriate training records prior to the start of testing. If Battelle staff operate and/or maintain a technology during the verification test, the technology vendor will be required to train those staff prior to the start of testing. Battelle will document this training with a consent form, signed by the vendor, that states which specific Battelle staff have been trained and determined by the vendor to be competent in operation of the vendor's technology. Battelle technical staff will have a minimum of a bachelor's degree in science/engineering or have equivalent work experience.

Table 2. DQIs and Criteria for Critical Measurements for Reference Method

DQI	Method of Assessment	Frequency	Minimum Acceptance Criteria	Corrective Action
Precision	Triplicate analyses of test samples	All test samples	3% relative standard deviation (RSD)	Data considered suspect and reanalyzed or reported with qualifiers if reanalysis is not possible
Bias and Accuracy of Instrument --Zero	Zero Check-observe signal with emission beam blocked	Every 4 hours	< 2 counts with Time Constant = 5 seconds.	Re-zero the instrument with the emission beam blocked.
Bias and Accuracy of Overall Instrument Sensitivity	Using conditions simulating tests, observe NIST calibration standard (naphthalene-anthracene)	Beginning of sample set analyzed each day, at least every 4 hours during analysis of the set, and at conclusion of the set.	Target: constant intensity throughout the experiment. If >1% variation of signal, then adjust voltage.	Recalibrate to target fluorescence value with NIST calibration standard. Constant intensity is re-established by varying the photomultiplier tube (PMT) operating voltage. If variation is >3%, re-analyze previous sample set or report with qualifiers.
Bias and Accuracy of Sample Measurements	Initial Calibration-various levels quinine sulfate prepared in water (Burdick and Jackson HPLC grade, or equivalent). Start with unspiked water, then observe 4 or more additional concentrations of quinine sulfate prepared as per ASTM E 579-04[6]	Perform initial calibration at the outset of verification testing and after any parameter changes or overall instrument sensitivity changes of >4%.		Investigate sources of standard contamination or changes in instrument parameters; clean the cuvette; reanalyze fresh control or standard; run Varian validation tests for operation.
	Calibration Check Sample-one concentration of quinine sulfate (i.e, 10 ppb quinine sulfate).	Minimum 5% of all samples tested.	Check standard is within ± 2% change in response from initial calibration after adjustment of Overall Instrument Sensitivity Reanalyze affected samples since last successful check if calibration check change is >4%.	Adjust Overall Instrument Sensitivity; check new calibration check sample (i.e., different concentration of quinine sulfate or freshly prepared standard); repeat initial calibration; run Varian validation tests for operation.
	Positive Control Sample- 50 ppb SR fulvic acid	Minimum 5% of all samples tested.	No more than ± 3% change in response during the sample set analyzed on the same day.	
	Negative Control Sample- (Burdick and Jackson HPLC grade, or equivalent)	Minimum 10% of all samples tested.	Within 10% of counts of original zero in initial calibration standard, which is near zero counts.	Clean cell (multiple times if necessary); check instrument zero.

DQI	Method of Assessment	Frequency	Minimum Acceptance Criteria	Corrective Action
Selectivity	Negative Control Sample- - (Burdick and Jackson HPLC grade, or equivalent)	Minimum 10% of all samples tested.	Within 10% of counts of original zero in initial calibration standard.	Adjust Overall Instrument Sensitivity; check new control sample; repeat initial calibration; run Varian validation tests for operation.
	Positive Control Sample- 50 ppb SR fulvic acid	Minimum 5% of all samples tested.	No more than ± 3% change in response during the sample set analyzed on the same day.	
	Environmental Samples-coastal and open ocean	All Environmental Samples used in verification testing.	Environmental Samples will be monitored for meeting vendor criteria for distinguishing between coast and open ocean samples.	If an Environmental Sample has a CDOM measurement which does not correspond to the vendor criteria for the type of sample being tested (i.e., a coastal sample does not meet vendor's criteria for identification as a coastal sample) and the above corrective actions have been taken, flag the results in the report. Full excitation-emission matrix (EEM) measurements may also be obtained on the sample in question in order to further evaluate sample properties.
Completeness	Amount of valid data obtained	Overall number of data points collected for test	90% of overall data points collected should be valid.	If feasible, analyze additional samples to meet the acceptance criterion.
Representativeness	Performance Test and Environmental Samples (see Table 5)	Overall for test	Target: samples with fluorescence measurements which span the responses obtained in the initial calibration curve.	Additional samples or samples fortified with quinine sulfate or fulvic acid may be considered to replace some Environmental Samples if those initially tested do not represent a range of fluorescence measurements that span the responses obtained in the initial calibration curve. Such samples will only be used to provide a range of fluorescence measurements for side-by-side comparison of screening tool measurements to reference method measurements.

A9 DOCUMENTATION AND RECORDS

The records for this verification test will include the test/QA plan, sample chain-of-custody forms, laboratory record books (LRB), data collection forms, electronic files (both raw data and spreadsheets of sample results or statistical calculations), and the final verification reports and verification statements. All of these records will be maintained in the Verification Test Coordinator's office (or at the test location during the test) and will be transferred to permanent storage at Battelle's Records Management Office at the conclusion of the verification test. The location (e.g., specific personal computer, server, or media type and storage location) of final versions of the electronic files will be noted in the test records. All Battelle LRBs are stored indefinitely, either by the Verification Test Coordinator or Battelle's Records Management Office. EPA will be notified before disposal of any files. Table 3 has further details regarding the data recording practices and responsibilities.

All written records must be in ink. Any corrections to notebook entries, or changes in recorded data, must be made with a single line through the original entry. The correction is then to be entered, initialed, and dated by the person making the correction. In all cases, strict confidentiality of data from each vendor's technology, and strict separation of data from different vendors' technology, will be maintained. Separate files (including manual records, printouts, and/or electronic data files) will be kept for each technology.

Table 3. Summary of Data Recording Process

Data to Be Recorded	Where Recorded	How Often Recorded	By Whom	Disposition of Data
Dates, times, and details of test events, technology maintenance, down time, etc.	ETV LRBs or data recording forms	Start/end of test procedure, and at each change of a test parameter or change of technology status	Battelle	Used to organize and check test results; manually incorporated in data spreadsheets as necessary
Technology calibration information	ETV LRBs, data recording forms, or electronically	At technology calibration or recalibration	Battelle	Incorporated in verification report as necessary
Technology readings	Either recorded electronically by the technology and downloaded to an independent computer or hard copy data printed by the technology and taped into the ETV LRB or hand entered into ETV LRBs or data recording forms.	Recorded continuously for electronic data and printed after each measurement for hard copy print-outs or recorded manually with each reading.	Battelle	Converted to or manually entered into spreadsheet for statistical analysis and comparisons
Sample collection and reference method analysis procedures, calibrations, QA, etc.	LRBs, chain-of-custody, or other data recording forms	Throughout sampling and analysis processes	Battelle and others assisting in sample collection	Retained as documentation of sample collection or reference method performance
Reference method results	Electronically or manually into ETV LRBs or data recording forms. Where possible at least the same number or a maximum of one number more significant figures as the BWE screening technology result will be reported for the reference method.	Every sample or QC analysis	Battelle	Transferred to spreadsheets for calculation of results, and statistical analysis and comparisons

SECTION B
MEASUREMENT AND DATA ACQUISITION

B1 EXPERIMENTAL DESIGN

BWE screening tool technologies will be tested only in a laboratory under controlled laboratory conditions; no field testing will take place during this verification test. This will allow comparison of the technology results to a reference method under a specified set of conditions. Performance test and environmental samples will be analyzed in triplicate and evaluated for accuracy compared to expected measurements based on the reference CDOM analyses, instrument linearity across the range of concentrations tested, and precision among the replicate measurements obtained. Two units of each technology will be used to measure the test samples. Measurements with the two units will be concurrent with each other and with the reference method measurement, so that inter-unit precision can be evaluated. All measurements made for direct comparison with the reference method will be conducted at ambient room temperature. However, because these technologies will be used in a wide range of temperatures in practical application and because temperature can affect CDOM fluorescence, a subset of test samples will be analyzed with the BWE screening tool units only at two additional temperatures, one below ambient and one above ambient (i.e., approximately 4 °C and 34 °C, respectively) in order to evaluate the screening tool's variability due to temperature effects. Note that while temperature is one of several variables that might affect practical application (other possibilities include humidity, ambient light, exposure to the elements, etc.), this verification test will only test the affect of varying temperature on the screening technology performance.

The analyses will be performed according to the vendor's recommended procedures as described in the user's instructions or manual, or during training provided to Battelle staff. Similarly, calibration and maintenance of the technologies will be performed as specified by the vendor. Results from the technologies being verified will be recorded manually by the operator on appropriate data sheets or captured in an electronic data system and then transferred manually or electronically for further data workup. Qualitative characteristics of each technology such as ease of use will be assessed through observations made by the Test Coordinator and operators throughout the verification test. The results from each technology will be reported individually. No direct comparison will be made between technologies, but each technology will undergo similar testing so it is convenient for end users to evaluate the ETV testing results.

B1.1 Test Procedures

The BWE screening tool verification test will focus on analyzing a broad range of samples which will provide a variety of CDOM concentrations. The first sample type will be performance test samples where analytes or compounds known to cause fluorescence (i.e., quinine sulfate or Suwannee River fulvic acid) will be added at multiple concentration levels to Burdick and Jackson HPLC grade (or equivalent) water and analyzed by the participating technologies to assess their ability to detect the CDOM fluorescence in water. Analytes will be added to Burdick and Jackson HPLC grade water (or equivalent) at a range of concentrations that span the technologies' linear range or at a minimum cover a range of fluorescence values expected to be encountered between open ocean and coastal water samples. The second sample type will be environmental samples and will consist of water collected from various coastal and open ocean areas. The environmental samples, which are expected to represent a range of CDOM concentrations that may be encountered in real-world water samples, will be collected from a maximum of 15 locations and will be measured by the reference method in order to screen their CDOM response before use in testing. Table 4 lists proposed locations for collecting environmental samples. A minimum of 10 samples will be selected from those collected to challenge the technologies with real-world samples of varying CDOM concentrations. If necessary to achieve a good range of CDOM concentrations, some open ocean or coastal samples may be fortified in the laboratory (i.e., with quinine sulfate or Suwanee River fulvic acid) to adjust their CDOM concentration, and used in place of coastal samples. Note that any fortified samples will be used for side-by-side comparison of the screening tool results to those obtained with the reference method only and are only for the purposes of providing a range of fluorescence measurements. Full excitation-emission matrix (EEM) measurements using the reference method instrumentation will be obtained on these 10 environmental samples used in verification testing in order to provide additional spectroscopic information on these samples. For full EEM measurements, the Varian Cary Eclipse spectrometer automatically corrects

Table 4. Proposed Environmental Samples

Location	Description	Sample Type
Duxbury Bay, MA	Off the dock at Battelle Duxbury Operations in Duxbury, MA	Coastal seawater
Boston Harbor, MA	Inside of Neponset Estuary in Boston Harbor, MA	A mixture of freshwater and coastal seawater with expected high CDOM fluorescence
Massachusetts Bay – NF7, MA	Nine miles east of Deer Island, MA	Coastal seawater
Massachusetts Bay – NF10, MA	Nine miles east of Deer Island, MA	Coastal seawater
Sequim Bay, WA	Off the dock at Battelle Marine Sciences Lab in Sequim, WA	Coastal seawater
Puget Sound, WA	Outside of Ediz Hook in Port Angeles, WA	Coastal seawater
East Coast, FL -1	Inter-coastal water way in West Palm Beach, FL	A mixture of freshwater and coastal seawater with expected high CDOM fluorescence
East Coast, FL - 2	Atlantic Ocean beach off Palm Beach, FL	Coastal seawater
Open Ocean - 1	NASS-5 Open Ocean Seawater Reference Material for Trace Metals (available from National Research Council Canada)	Open ocean seawater with expected low CDOM fluorescence
Open Ocean - 2	MOOS-1 Seawater Certified Reference Material for Nutrients (available from National Research Council Canada)	Open ocean seawater with expected low CDOM fluorescence
Long Island Sound, NY	Dock in Port Jefferson, NY	Coastal seawater
New York Harbor, NY	East River, NY	A mixture of freshwater and coastal seawater with expected high CDOM fluorescence
New York Bight, NY	Atlantic ocean sample from a beach in South Hampton, NY	Coastal seawater
San Diego Harbor, CA	San Diego Harbor, CA	Coastal seawater
Narragansett Bay, RI	Off 2-14 Great Island Rd, Narragansett, RI	Coastal seawater

excitation, and emission will be calibrated so as to ensure the highest removal of instrumental scaling effects. The full EEM measurements are for informational purposes only. Reference method measurements used for comparing the technology results to reference method results will be obtained using only the wavelengths and technique that the BWE screening tool uses and not the full EEM measurements. The third sample type will be quality control samples. Quality control samples are discussed further in Section B5. Table 5 shows the samples that will be evaluated during this verification test.

Table 5. Verification Test Samples

	Performance Factor	Sample Description	Replicates per BWE Screening Tool Unit		
Performance Test			**~4 °C**	**~ 24° C**	**~ 34 ° C**
Quinine sulfate prepared in Burdick and Jackson HPLC grade water (or equivalent) per ASTM E579-04[6]	Accuracy, linearity, precision, temperature effects	unspiked	3	3	3
		1 ppb quinine sulfate	3	3	3
		5 ppb quinine sulfate	3	3	3
		10 ppb quinine sulfate	3	3	3
		50 ppb quinine sulfate	3	3	3
		100 ppb quinine sulfate	3	3	3
	Method detection limit	Quinine sulfate at 5 x vendor detection limit	-	7	-
Fulvic acid prepared in Burdick and Jackson HPLC grade water (or equivalent). The pH of the fulvic acid solutions will not be adjusted, but will be checked and recorded prior to their use.	Accuracy, linearity, precision, temperature effects	unspiked	3	3	3
		1 ppb Suwanee River (SR) fulvic acid	3	3	3
		5 ppb SR fulvic acid	3	3	3
		10 ppb SR fulvic acid	3	3	3
		50 ppb SR fulvic acid	3	3	3
		100 ppb SR fulvic acid	3	3	3
	Method detection limit	SR fulvic acid at 5 x vendor detection limit	-	7	-
Environmental Samples					
Location 1-open ocean	Matrix Effects	unspiked	-	3	-
Location 2-open ocean			-	3	-
Location 3-coastal			-	3	-
Location 4-coastal			-	3	-
Location 5-coastal			-	3	-
Location 6-coastal			-	3	-
Location 7-coastal			-	3	-
Location 8-coastal			-	3	-
Location 9-coastal			-	3	-
Location 10-coastal			-	3	-
Quality Control					
Negative control		Burdick and Jackson HPLC grade water (or equivalent)	8 (minimum)		
Positive control		50 ppb SR fulvic acid	8 (minimum)		
Calibration Check		10 ppb quinine sulfate	Single measurement per every 9 verification sample measurements		
		TOTAL	**185 (minimum)**		

Shading indicates samples to be tested and number of replicates using the reference method.

ETV verifications usually include a comparison of the results generated by the technologies being verified with the results of analysis of the same samples using a standard reference method that measures the same endpoint. Reference methods are discussed in Section B4.

The technologies will be evaluated for the following parameters:

B1.1.1 Accuracy

Accuracy will be determined by comparing the BWE screening tool technology CDOM measurement to the CDOM measurement generated by a Varian Cary Eclipse spectrometer at a single temperature (i.e., data from ambient temperature will be used) for both performance test samples and environmental samples. See Section B4 for complete details of how the reference method measurements will be made in comparison to how the BWE screening tool measurements are made. Percent difference (PD) between the technology result and the Varian Cary Eclipse spectrometer result will be determined.

B1.1.2 Linearity

Linearity will be determined by plotting the CDOM measurements taken while analyzing varying concentrations of analytes which are known to fluoresce against the analyte concentration and performing linear curve fitting to determine the slope and regression statistics.

B1.1.3 Precision

Precision will be measured among replicate measurements of the same sample by determining relative standard deviation (RSD).

B.1.1.4 Method Detection Limit

Method detection limit will be determined by analyzing 7 replicates of known fluorescing analytes at a concentration five times the vendor expected detection limit for the analyte.

B1.1.5 Inter-unit Reproducibility

Inter-unit reproducibility will be determined by calculating the relative percent difference (RPD) between the average of replicate CDOM measurements of the same sample taken at the same temperature for all PT and environmental samples made using two different units of the BWE screening tool technology.

B1.1.6 Temperature Effects

The BWE screening tool CDOM measurements made at ~ 4 °C and ~ 34 °C will be compared to the BWE screening tool CDOM measurements made at ambient temperature (~ 24 °C) by determining RPD for each concentration of the curve analyzed at multiple temperatures. Note that reference method measurements will only be taken at ambient temperature and so there will not be any direct comparison of the BWE screening tool results to reference method measurements at ~ 4 °C and ~ 34 °C. Comparisons will only be between the BWE screening tool results at ~ 4 °C and ~ 34 °C to the BWE screening tool results at ambient temperature. During the measurements at ~ 4 °C and ~ 34 °C, the testing solutions will be equilibrated at the testing temperature for at least 30 minutes prior to the start of testing. BWE screening tool units will be equilibrated per vendor's recommendations for when there is a difference in instrument versus sample temperature, if available. If this information is not available from the vendor, then a minimum 5 minute equilibration time will be used for instrumentation.

B.1.1.7 Matrix Effects

Matrix effects will be evaluated by comparing the environmental sample PD values obtained for determining accuracy in Section B.1.1.1 with those that were obtained for the PT samples to determine if the environmental sample PD values are statistically different from the PT sample PD values. Data will be evaluated to determine if there are trends in PD with the environmental samples that could be due to matrix effects. Full EEM data may be used to help assess differences in environmental sample matrices.

B.1.1.8 Data Completeness

Data completeness will be determined as the number of valid measurements out of the total number of measurements taken.

B1.1.9 Operational Factors

Operational factors such as maintenance needs, calibration frequency, data output, consumables used, ease of use, repair requirements, waste production, and sample throughput will be evaluated based on operator and Verification Test Coordinator observations. An LRB or data sheets will be used to document observations. Examples of information to be recorded include the daily status of diagnostic indicators for the technology, use or replacement of any consumables, the effort or cost associated with maintenance or repair, vendor effort (e.g., time on

site) for repair or maintenance, the duration and causes of any technology down time or data acquisition failure, operator observations about technology ease of use, clarity of the vendor's instruction manual, user-friendliness of any needed software, overall convenience of the technologies and accessories/consumables, or the number of samples that could be processed per hour or per day. These observations will be summarized to aid in describing the technology performance in the verification report on each technology.

B1.2 Statistical Analysis

The statistical methods and calculations used for evaluating quantitative performance parameters are described in the following sections.

B.1.2.1 Accuracy

Accuracy will be determined by calculating the PD between the BWE screening tool technology CDOM measurement (M_1) and the CDOM measurement generated by a Varian Cary Eclipse spectrometer (M_2) at a single temperature (i.e., data from ambient temperature will be used) for both performance test samples and environmental samples using the following equation.

$$PD(\%) = \frac{\left| M_1 - M_2 \right|}{M_2} \times 100 \qquad (1)$$

B1.2.2 Linearity

Linearity will be determined by plotting the CDOM measurements taken while analyzing varying concentrations of analytes which are known to fluoresce (y-axis) against the analyte concentration (x-axis) and performing linear curve fitting to determine the slope (m) and intercept (b) in the equation y=mx +b. Correlation coefficients such as the Pearson's *r* values and coefficient of determination (r^2) values will be calculated.

B1.2.3 Precision

The standard deviation (S) of the results for the replicate analyses of the same sample will be calculated as follows.

$$S = \left[\frac{1}{n-1} \sum_{k=1}^{n} \left(M_k - \overline{M} \right)^2 \right]^{1/2} \quad (2)$$

where n is the number of replicate samples, M_k is the CDOM measurement for the k^{th} sample, and M is the average CDOM measurement of the replicate samples. The BWE screening tool technology precision for each sample will be reported in terms of the relative standard deviation (RSD), which will be calculated as follows.

$$RSD(\%) = \left| \frac{S}{\overline{M}} \right| \times 100 \quad (3)$$

B1.2.4 Method Detection Limit

The method detection limit (MDL) will be determined according to procedures described in 40CFR136 Appendix B[7] and will be assessed from seven replicate analyses of a fortified sample. Fortified samples will be generated by adding known fluorescing compounds such as quinine sulfate and SR fulvic acid to Burdick and Jackson HPLC grade water, or equivalent. The target analyte will be added at a concentration approximately five times the vendor-stated detection limit. The MDL will be calculated using the following equation:

$$MDL = t \times S \quad (4)$$

Where t is the Student's value of 3.143 for a 99% confidence level when the degrees of freedom (N-1) equals six, and S is the standard deviation of the replicate samples.

B.1.2.5 Inter-unit Reproducibility

Inter-unit reproducibility will be determined by evaluating the RPD between the average of replicate measurements made for each sample tested using two separate units of the technology being verified. The equation for RPD, reported in percent, is as follows:

$$RPD(\%) = \frac{|M_1 - M_2|}{M_1 + M_2} \times 200 \quad (5)$$

where M_1 is the average of replicate measurements made by the first unit of the technology and M_2 is the average of replicate measurement made by the second unit of the technology being evaluated.

B.1.2.6 Temperature Effects

Temperature effects will be determined by measuring the PD (using the equation listed in B.1.2.1) between the average of replicate measurements made for each sample at either 4 °C or 34 °C (M_1) against the average measurement taken at 24 °C (M_2). T-tests may be applied to the data to help determine whether PD measurements are significant.

B.1.2.7 Matrix Effects

Matrix effects will be determined by comparing the accuracy PD measurements for the PT samples to the accuracy PD measurements for the environmental samples. PD will be determined as described in B.1.2.1. T-tests may be applied to the data to help determine whether the differences in PD measurements between the PT and environmental samples are significant.

B.1.2.8 Data Completeness

Data Completeness will be calculated as the percentage of the total possible data by taking the number of valid data measurements generated by each technology (M_{valid}), and dividing by the total number of data measurements included in verification testing (M_{total}).

$$Completeness(\%) = \frac{M_{valid}}{M_{total}} \times 100 \quad (6)$$

The cause of any substantial loss of data will be established from operator observations or technology records, and noted in the discussion of the data completeness results.

B1.2.9 Operational Factors

There are no statistical calculations applicable to operational factors. Operational factors will be determined based on documented observations of the testing staff and the Verification Test Coordinator.

B1.3 Reporting

The data obtained in the verification test will be compiled separately for each vendor's technology, and the data evaluations will be applied to each technology's data set without reference to any other. At no time will data from different vendor's technology be intercompared or ranked. Following completion of the data evaluations, a draft verification report and verification statement will be prepared for each vendor's technology, stating the verification test procedures and documenting the performance observed. For example, descriptions of the data acquisition procedures, use of vendor supplied proprietary software, consumables used, repairs and maintenance needed, and the nature of any problems will be presented in the draft report. Each report will briefly describe the ETV Program, the AMS Center, and the procedures used in verification testing. The results of the verification test will be stated quantitatively, without comparison to any other technology tested, or comment on the acceptability of the technology's performance. Each draft verification report will be submitted for review by the respective technology vendor and by EPA and other peer reviewers. Comments on the draft report will be addressed in revisions of the report. The peer review comments and responses will be tabulated to document the peer review process. The reporting and review process will be conducted according to the requirements of the AMS Center QMP.[1]

B2 SAMPLING REQUIREMENTS

B2.1 Sample Collection, Storage and Shipment

Pre-cleaned 500 mL amber glass jars (ESS QC grade, or equivalent) with Teflon-lined caps will be used as containers for all samples collected specifically for this study. Some samples that have been collected for other programs may be used for verification testing and in those cases, the samples will be provided in the container selected by the other program and contamination control and storage conditions from that program will be documented. For

samples collected specifically for this study and not in conjunction with any other program, just prior to sample collection, the sample jars and caps will be rinsed three times with water from the collection location. Where possible these samples will be collected from a dock or other similar feature. If wading is necessary to collect the samples, care will be taken to obtain waist deep water and to avoid any sediments resuspended while wading.

Sample collection will also avoid any surface sheen, which may cause interference with CDOM fluorescence measurements. If surface sheen is present and a better location cannot be found, the jar will be submerged at least 6 inches below the water surface with the cap on. The cap will then be unscrewed and the sample collected and the jar recapped while under water. If surface sheen is not present, the jar will be uncapped and filled just below surface level. In all instances, once the sample has been collected, a small portion of the sample will be poured off and the jar recapped in order to provide an air space that will facilitate later mixing. Duplicate samples will be collected at each location to provide backup samples in case of loss during shipping and handling, and to provide a backup archive sample during testing. The samples will be refrigerated or frozen (if frozen, care must be taken to ensure that the sample can expand without breaking the sample jar) until shipment to Battelle on ice.

Shipments will be via a trackable overnight delivery service to the Battelle sample custodian. At Battelle, the samples will be stored refrigerated or frozen (depending on how the sample was sent to Battelle) until testing. While some samples may be filtered in the field upon collection, all samples (including those previously filtered in the field) will be filtered just prior to measuring the sample fluorescence in the laboratory. Samples (both those for use with the BWE screening tool and those used for the reference method) will be filtered with the size and type of filter recommended by the BWE screening tool vendor for their technology. All samples, including PT, environmental, and QC samples, will be equilibrated at the testing temperature (~ 4 °C, ~ 24 °C, or ~ 34 °C) before fluorescence measurements commence. The actual testing temperature will be recorded during testing.

B3 SAMPLE HANDLING AND CUSTODY REQUIREMENTS

Sample custody will be documented throughout collection, transport, shipping (if necessary), and analysis using standard chain-of-custody (COC) forms provided by Battelle or supplied by others providing samples for testing, as appropriate. Samples transferred within Battelle may be documented in bound sample login LRBs. Each COC form will summarize the

samples collected. The COC forms will track sample release from the sampling location to Battelle. Each COC form will be signed by the person relinquishing the samples once that person has verified that the COC form is accurate. The original sample COC forms will accompany the samples; the shipper will keep a copy. Upon receipt at Battelle, COC forms will be signed by the person receiving the samples once that person has verified that all samples identified on the COC forms are present. Any discrepancies will be noted on the form and the sample receiver will immediately contact the Verification Test Coordinator to report missing, broken, or otherwise compromised samples. Copies of all COC forms will be delivered to the Verification Test Coordinator, and maintained with the test records. Samples being transferred within Battelle may be documented in a sample login LRB which will be used to note the current location of all samples housed at Battelle.

B4 LABORATORY REFERENCE METHOD

The BWE screening tools provide a measure of CDOM by measuring fluorescence at various wavelengths using a hand-held instrument. Reference measurements for CDOM will be performed by Battelle using a laboratory bench scale Varian Cary Eclipse spectrometer. Settings for the Varian Cary Eclipse spectrometer (e.g., wavelengths, slit widths, angle for fluorescence measurement, path lengths, etc.) will be set to resemble those used by the BWE screening tool. For example, slits of 10 nm will be used on the Varian Cary Eclipse spectrometer if filters of 10 nm bandwidth are used on the BWE screening tool.

Results from the Varian Cary Eclipse spectrometer will be calculated and reported in the same manner that the BWE screening tool calculates and reports results. For example, if the screening tool measures CDOM as a ratio of intensities at two wavelengths, then the Varian Cary Eclipse spectrometer will be used to measure the same two wavelengths under the same excitation and emission conditions and a ratio at the same wavelengths will be calculated using the Varian Cary Eclipse data. It is not expected or necessary that the ratio be exactly the same. Efficiencies of gratings and other instrument conditions will vary from instrument to instrument and will, in fact, likely make this ratio different. The ratios from the BWE screening tool will be calibrated against a series of quinine sulfate concentrations. The Varian Cary Eclipse spectrometer will be calibrated in the same range with the same standards. If both instruments are linear in that range and track through zero when a zero concentration standard is analyzed, there will be a single value constant that can be used to multiply one result to correlate to the

other result. If the relationship is not linear, the correlation can be a more complex relationship, but if proper zeroing is performed on each instrument and the calibration concentration ranges are the same, then the linearity of the BWE screening tool and the Varian Cary Eclipse spectrometer can be directly compared through a relationship established based on the standard curves analyzed on each instrument.

The Varian Cary Eclipse spectrometer will be operated according to the procedures described in the Varian instrument manuals and any deviations will be documented. Specific operating procedures relevant to the Varian Cary Eclipse spectrometer measurements will depend on the BWE screening technology being evaluated and how it measures CDOM. For example, if only two wavelengths need to be measured, the Varian Cary Eclipse individual wavelength software package will be used rather than the scanning package software. The PMT voltage will be set to provide counts in the 0-1000 range for the strongest anticipated fluorescence. A standard operating procedure (SOP) for the Varian Cary Eclipse spectrometer reference measurement will be prepared for each technology tested. Each SOP will include instructions for:

- Setting the Varian Cary Eclipse spectrometer parameters so that the spectrometer out performs the technology being tested. Long enough integration time constants will be used to generate signal-to-noise greater than the technology produces.
- Validating all instrument parameters and storing the parameters used in a method file.
- Calibrating intensity.
- Properly cleaning and checking visual transmittance of cells between measurements.
- Checking sample-related instability and span consistency by analyzing calibration check samples and positive control samples during each sample set.

The quinine sulfate solution used for calibration will be verified by a second source standard or analysis of two independently prepared solutions from the same lot at least once during the tests.

B5 QUALITY CONTROL

Steps will be taken to maintain the quality of data collected during this verification test. This will include analyzing specific quality control samples (QCS) at a regular frequency. QCSs will include negative controls, positive controls, and calibration checks. Negative control

samples will be used to help ensure that no sources of contamination are introduced in the sample handling and analysis procedures. The positive control samples and calibration checks will indicate to the operator whether or not the technology is functioning properly. The vendor will provide the approximate endpoint that should result upon analysis of the positive control and calibration check by their technology. QCSs producing results that do not meet the anticipated results specified by the vendor will be reanalyzed and corrective action taken if needed to ensure that test sample results are not affected. Corrective actions may include reanalyzing samples to verify that the technology has been operated properly, conducting maintenance, or recalibrating. Positive and negative controls will be analyzed at a frequency of approximately 5 % based on the total number of test samples (see Table 5) for the BWE screening tools. More frequent negative controls may be analyzed using the Varian Cary Eclipse spectrometer to check cell cleanliness and instrument zero. Calibration checks will be analyzed at a minimum after every nine measurements of PT or environmental samples with the BWE screening tool and at a minimum as 5% of the total number of test samples for the Varian Cary Eclipse spectrometer.

B6 INSTRUMENT/EQUIPMENT TESTING, INSPECTION, AND MAINTENANCE

The equipment used by Battelle (i.e., instrumentation used for CDOM reference measurements, balances or pipettes used to prepare standards, etc.) will be tested, inspected, and maintained as per the standard operating procedures of Battelle and/or the manufacturer's recommendations so as to meet the performance requirements established in this document. When technical staff operate and maintain technologies undergoing testing, those activities will follow directions provided by the technology vendor.

B7 CALIBRATION/VERIFICATION OF TEST PROCEDURES

The Varian Cary Eclipse spectrometer used for reference analyses will be calibrated as appropriate before any samples are analyzed and recalibrated as needed based on the results of any calibration check samples.

Technologies undergoing testing will be calibrated initially by the respective technology vendor prior to shipping the technology to Battelle or during training, and will be repeated according to direction from the vendor. Calibration checks will be performed after every 9 measurements during verification testing for the BWE screening tools. In the event that

recalibration is necessary, the recalibration will be carried out following the vendor's instructions. All calibrations will be documented as appropriate by the technical staff.

B8 INSPECTION/ACCEPTANCE OF SUPPLIES AND CONSUMABLES

In general, this verification test relies on the materials and equipment provided by the vendors. Battelle will provide the following equipment and materials needed for the preparation of the performance test, environmental and QC samples:

- Burdick and Jackson HPLC grade water, or equivalent
- various laboratory supplies necessary for accurate preparation of the test samples and subsequent dilutions (i.e., volumetric pipets; pipet bulbs; Eppendorf micro pipettes and pipette tips, or equivalent; volumetric flasks; disposable pipets; balances; etc.)
- reference standards with a known level of purity for target analytes - NIST traceable, or equivalent if available
- glass sample containers
- equipment and consumables for the Varian Cary Eclipse spectrometer
- personal protective equipment

All materials, supplies, and consumables will be ordered by the Verification Test Coordinator or designee. Where possible, Battelle will rely on material and consumable sources that have been used previously without problems for ETV verification testing. Battelle will also rely on previous experience or recommendations from EPA advisors, stakeholders, test collaborators, subcontractors, or technology vendors. Upon receipt of any supplies or consumables, the Verification Test Coordinator or designee will visually inspect and ensure that the materials received are those that were ordered and that there are no visual signs of damage that could compromise the suitability of the materials. If damaged or inappropriate goods are received, they will be returned or disposed of and arrangements will be made to receive replacement materials. Certificates of analysis (COA) or other documentation of analytical purity will be checked for all reagents and standards to ensure suitability for the verification test and a copy will be stored with the test files. Where possible, reagents (i.e., HPLC grade water, sulfuric acid, etc.) used to prepare standards will be checked for background fluorescence prior to their use. Unsuitable materials will be returned or disposed of and arrangements for the receipt of replacement materials will be made.

B9 NON-DIRECT MEASUREMENTS

No non-direct measurements will be used during this verification test.

B10 DATA MANAGEMENT

Various types of data will be acquired and recorded electronically or manually by Battelle during the verification test. Table 2 summarizes the types of data to be recorded. All maintenance activities, repairs, calibrations, and operator observations relevant to the technology operation will be documented by technical staff in LRBs or on data sheets. Results from the reference methods, including raw data, analyses, and final results, will be compiled by Battelle.

Records received by or generated by any technical staff during the verification test will be reviewed by a Battelle staff member within two weeks of generation or receipt, before the records are used to calculate, evaluate, or report verification results. If a Battelle staff member generated the record, this review will be performed by a Battelle technical staff member involved in the verification test, but not the staff member who originally generated the record. The review will be documented by the person performing the review by adding his/her initials and date to the hard copy of the record being reviewed. In addition, any calculations performed by technical staff will be spot-checked by Battelle QA and/or technical staff to ensure that calculations are performed correctly. Calculations to be checked include any statistical calculations described in this test/QA plan. The data obtained from this verification test will be compiled and reported independently for each technology. Results for technologies from different vendors will not be compared with each other.

Among the QA activities conducted by Battelle QA staff will be an audit of data quality. This audit will consist of a review by the Battelle Quality Manager (or his designee) of at least 10% of the test data. During the course of any such audit, the Battelle Quality Manager will inform the technical staff of any findings and any need for immediate corrective action. If serious data quality problems exist, the Battelle Quality Manager will request that Battelle's AMS Center Manager issue a stop work order. Once the assessment report has been prepared, the Verification Test Coordinator will ensure that a response is provided for each adverse finding or potential problem, and will implement any necessary follow-up corrective action. The Battelle Quality Manager will ensure that follow-up corrective action has been taken.

SECTION C
ASSESSMENT AND OVERSIGHT

C1 ASSESSMENTS AND RESPONSE ACTIONS

Every effort will be made in this verification test to anticipate and resolve potential problems before the quality of performance is compromised. One of the major objectives of this test/QA plan is to establish mechanisms necessary to ensure this. Internal quality control measures described in this test/QA plan, which is peer reviewed by a panel of outside experts, implemented by the technical staff and monitored by the Verification Test Coordinator, will give information on data quality on a day-to-day basis. The responsibility for interpreting the results of these checks and resolving any potential problems resides with the Verification Test Coordinator. Technical staff have the responsibility to identify problems that could affect data quality or the ability to use the data. Any problems that are identified will be reported to the Verification Test Coordinator, who will work with the Battelle Quality Manager to resolve any issues. Action will be taken to control the problem, identify a solution to the problem, and minimize losses and correct data, where possible. Independent of any EPA QA activities, Battelle will be responsible for ensuring that the following audits are conducted as part of this verification test.

C1.1 Performance Evaluation Audits

A Performance Evaluation (PE) audit will be conducted to assess the quality of the reference method measurements made in this verification test. The reference method PE audit will be performed by supplying an independent, NIST-traceable solid standard, if available, or at a minimum a second quinine sulfate standard solution prepared from a different source of quinine sulfate than that used in verification testing. The PE audit samples will be analyzed in the same manner as all other samples and the analytical results for the PE audit samples will be compared to the nominal concentration. The target criterion for this PE audit is agreement of the analytical result within 3% of the nominal concentration, after the data have been adjusted for the Overall Instrument Sensitivity based on the observation of the naphthalene-anthracene standard. If the PE audit result does not meet the target criterion, the PE audit will be repeated. If the outlying results persist, the source of error will be investigated and corrective action taken as

necessary until successful PE audit results are obtained. This audit will be performed once prior to the start of the test, and will be the responsibility of the Verification Test Coordinator or designee.

C1.2 Technical Systems Audits

The Battelle Quality Manager or designee will perform a TSA at least once during this verification test. The purpose of this audit is to ensure that the verification test is being performed in accordance with the AMS Center QMP[1], this test/QA plan, any published reference methods, and any Standard Operating Procedures (SOPs) used. In the TSA, the Battelle Quality Manager, or a designee, may review the reference methods used, compare actual test procedures to those specified or referenced in this plan, and review data acquisition and handling procedures. In the TSA, the Battelle Quality Manager will tour the laboratory where verification and reference testing are taking place, inspect sample COC documentation, and review technology-specific record books. He or she will also check standard certifications and technology data acquisition procedures, and may confer with the technology vendors and technical staff. A TSA report will be prepared, including a statement of findings and the actions taken to address any adverse findings. The EPA AMS Center Quality Manager will receive a copy of Battelle's TSA report. At EPA's discretion, EPA QA staff may also conduct an independent on-site TSA during the verification test. The TSA findings will be communicated to technical staff at the time of the audit and documented in a TSA report.

C1.3 Data Quality Audits

The Battelle Quality Manager or his designee will audit at least 10% of the verification data acquired in the verification test. The Battelle Quality Manager will trace the data from initial acquisition, through reduction and statistical comparisons, to final reporting. All calculations performed on the data undergoing the audit will be checked.

C1.4 QA/QC Reporting

Each assessment and audit will be documented in accordance with Section 3.3.4 of the AMS Center QMP.[1] The results of the technical systems audit will be submitted to EPA. Assessment reports will include the following:

- Identification of any adverse findings or potential problems
- Response to adverse findings or potential problems
- Recommendations for resolving problems
- Confirmation that solutions have been implemented and are effective
- Citation of any noteworthy practices that may be of use to others.

C2 REPORTS TO MANAGEMENT

The Battelle Quality Manager, during the course of any assessment or audit, will identify to the technical staff performing experimental activities any immediate corrective action that should be taken. If serious quality problems exist, the Battelle Quality Manager is authorized to request that Battelle's AMS Center Manager issue a stop work order. Once the assessment report has been prepared, the Verification Test Coordinator will ensure that a response is provided for each adverse finding or potential problem and will implement any necessary follow-up corrective action. The Battelle Quality Manager will ensure that follow-up corrective action has been taken. The test/QA plan and final report are reviewed by EPA AMS Center QA staff and EPA AMS Center program management staff. Upon final review and approval, both documents will then be posted on the ETV website (www.epa.gov/etv).

SECTION D
DATA VALIDATION AND USABILITY

D1 DATA REVIEW, VALIDATION, AND VERIFICATION REQUIREMENTS

The key data review requirements for the verification test are stated in Section B10 of this test/QA plan. In general, the data review requirements specify that the data generated during this test will be reviewed by a Battelle technical staff member within two weeks of data generation. The reviewer will be familiar with the technical aspects of the verification test, but will not be the person who generated the data. This process will serve both as the data review and the data verification, and will ensure that data have been recorded, transmitted, and processed properly. Furthermore, this process will ensure that the BWE screening tool data and the reference method data are collected under appropriate testing conditions and that the reference method data meet the reference method specifications.

The data validation requirements for this test involve a data quality assessment relative to the DQIs and audit acceptance criteria specified for this test. The DQIs listed in Section A 7.1 and B5 will be used to validate the data quality. The QA audits described within Section C of this document, including the performance evaluation audit and data quality audit, are designed to validate the data quality.

D2 VALIDATION AND VERIFICATION METHODS

Data verification is conducted as part of the data review, as described in Section B10 for this test/QA plan. A visual inspection of handwritten data will be conducted to ensure that all entries were properly recorded or transcribed, and that any erroneous entries were properly noted (i.e., single line through the entry with an error code and the initials of the recorder and date of entry). Electronic data from the technologies and other instruments used during the test will be inspected to ensure proper transfer from the datalogging system. Data manually incorporated into spreadsheets for use in calculations will be checked against handwritten data to ensure that transcription errors have not occurred. All calculations used to transform the data will be reviewed to ensure the accuracy and the appropriateness of the calculations. Calculations performed manually will be reviewed and repeated using a handheld calculator or commercial software (e.g., Excel). Calculations performed using standard commercial office software (e.g.,

Excel) will be reviewed by inspecting the equations used in calculations and verifying selected calculations by handheld calculator. Calculations performed using specialized commercial software (i.e., for analytical instrumentation) will be reviewed by inspection and, when feasible, verified by handheld calculator, or standard commercial office software.

To ensure that the data generated from this test meet the goals of the test, a number of data validation procedures will be performed. Section C of this test/QA plan provides a description of the validation safeguards employed for this verification test. Data validation and verification efforts include the completion of QC activities and the performance of TSA and PE audits as described in Section C. The data from this test will be evaluated relative to the measurement DQIs described in Section A7.1 and B5, and the PE audit acceptance criteria given in Section C1.1 of this test/QA plan. Data failing to meet these criteria will be flagged in the data set and not used for evaluation of the technologies, unless these deviations are accompanied by descriptions of their potential impacts on the data quality.

A data quality audit will be conducted by the Battelle Quality Manager to ensure that data review, verification, and validation procedures were completed, and to assure the overall data quality.

D3 RECONCILIATION WITH USER REQUIREMENTS

The purpose of a verification test performed following this test/QA plan is to evaluate the performance of commercial technologies which screen for ballast water exchange by measuring CDOM. This test evaluates the CDOM measurement method capability only and is not a verification of whether or not the measurement quantitatively measures CDOM or detects ballast water exchange. This evaluation will include comparisons of the results from the technologies to results from standard reference techniques. To meet the requirements of the user community, the data obtained in such a verification test will include thorough documentation of the technology's performance during the verification test. The data review, verification, and validation procedures described above will assure that verification test data meet these requirements, are accurately presented in the verification reports generated from the test, and that data not meeting these requirements are appropriately flagged and discussed in the verification reports. Additionally, all data generated using the reference method, which are used to evaluate technology results during the verification test, should meet the QA requirements of any applicable standard operating procedures or instrumentation instruction manuals.

This test/QA plan and any resulting ETV verification report(s) generated following procedures described in this test/QA plan will be subjected to review by participating technology vendors, ETV AMS Center staff, test collaborators, EPA, and external expert peer reviewers. These reviews will assure that this test/QA plan, verification test(s) of BWE screening tools, and the resulting report(s) meet the needs of potential users and regulators. The final report(s) will be submitted to EPA in 508 compliant Adobe Portable Document Format (pdf) and subsequently posted on the ETV website.

SECTION E

REFERENCES

E1 REFERENCES

1. Quality Management Plan (QMP) for the ETV Advanced Monitoring Systems Center, U.S. EPA Environmental Technology Verification Program, prepared by Battelle, Columbus, Ohio, Version 6.0, November 2005.

2. "Environmental Technology Verification Program Quality Management Plan" (QMP), December 2002, EPA/600/R-03/021.

3. Hunt, Carlton D., Tanis, Deborah, Bruce, Elizabeth, and Taylor, Michael. In Press. Optical Signatures of Seawater and their Potential Use in Verification of At Sea Ballast Water Exchange *Marine Ecology and Progress Series.*

4. Murphy, K., Ruiz, G., Dunsmuir, W., Waite, T. 2006. Optimized Parameters for Fluorescence-Based Verification of Ballast Water Exchange by Ships. *Environ. Sci. Technol*., 40, 2357-2362.

5. Murphy, K., Boehme, J., Coble, P., Cullen, J., Field, P., Moore, W., Perry, E., Sherrell, R., and Ruiz, G. 2004. Verification of mid-ocean ballast water exchange using naturally occurring coastal tracers. *Marine Pollution Bulletin*, 48, 711-730.

6. ASTM E579-04, "Standard Test Method for Limit of Detection of Fluorescence of Quinine Sulfate in Solution," ASTM International.

7. *Code of Federal Regulations*, Title 40, Part 136, Appendix B, Definition and Procedure for the Determination of the Method Detection Limit-Revision 1.11.