SAP NERR Nutrient Metadata
Months and year the documentation covers
Latest Update: January 31, 2022

I. Data Set and Research Descriptors

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2) Research objectives

The nutrient monitoring program is designed upon spatial deployment across a wide variety of marsh types with differing fresh and marine water mixing. These differing dynamics allow scientists and researchers to select from both a wide variety of research sites as well as tailor research programs to specific tidal dynamics and utilize the Reserves SWMP data acquisitions to the maximum extent. Additionally, from a long-term trend perspective the variety of marsh types and hydrology being monitored will allow for a better understanding of the different effects of sea-level rise upon marsh type. Due to a lack of residential development and very low human activity within the watersheds of the sites, they serve as a proxy for reference conditions with the various marsh and associated hydrology types for the creeks and river stations. All of the sites selected have very little anthropogenic nutrient influences. The following brief descriptions are associated with each nutrient monitoring site. For more detail please refer to the site descriptors located under section (4) of this document and/ or contact the Research Coordinator at the SAP NERR for detailed information of any/all sites.

Lower Duplin: Located at the mouth of the Duplin River with large, rapid and near-complete hydraulic exchange with Doboy Sound within each diurnal cycle. Lower Duplin is typical of a high salinity, well mixed estuary site.

Hunt Dock: Located on the upper Duplin River with relatively high hydraulic retention requiring an estimated 6-7 diurnal events to complete a total hydraulic exchange. Rainfall may drop salinity precipitously in the basin depending on tidal height, duration and volume of precipitation.

Cabretta Creek: Located on the eastern side of Sapelo Island with direct exchange with the Atlantic Ocean. Cabretta Creek is typical of high salinity, high oceanic exchange and near complete hydraulic exchange with each diurnal event. Cabretta Creek is extremely buffered from rainfall (event driven) fluctuations in salinity.

Dean Creek: Located on the southern end of Sapelo Island is the primary drainage of the inter-dune (located amid primary and secondary dune systems) meadow. This site is highly susceptible to very high salinity fluctuations associated with rainfall events on both seasonal and short–term, event-driven scales. Tidal exchange is complete at each diurnal event and exchange water genesis is the Doboy Sound.

The Duplin River is a tidal basin with no freshwater influence within its headwaters apart from surficial aquifer weeping from the perched lens of water associated with Sapelo Island. This nutrient monitoring effort is tied into the Georgia Coastal Ecosystems, Long-Term Ecological Research (GCE-LTER) initiative and the University of Georgia Marine Extension Service water quality database whose collection and analysis of the water samples facilitates the database. This long-term data set is being developed to provide information on estuarine water mixing within the well-studied Duplin River basin in addition to providing a long-term characterization of water quality as related to nutrient loading within the Duplin River.

a) The Monthly Grab Sampling Program focuses on documentation of baseline reference nutrient trends within a wide array of local marsh systems with differing hydrology.

b) The Diel Sampling Program focuses on short-term temporal variability over a lunar tidal cycle.

3) Research methods

a) Monthly Grab Sampling Program

In January 2017, analysis of the samples switched from the locally available University of Georiga Marine Extension Servies, to the Virginia Institute of Marine Science (VIMS) laboratory. This necessitated overnight shipping on the same day of collection, and shortened the time frame for processing samples for shipment and analysis. Monthly grab samples were taken at four stations within the Duplin River estuary from January to December 2017. Bottom water samples were taken at the Lower Duplin (LD), Hunt Dock (HD), Cabretta Creek (CA) and Dean Creek (DC) stations using a Niskin style sampling bottle. All grab samples were taken sequentially in duplicate beginning near the time the last diel sample was collected by the ISCO sampler (this time corresponds to low tide at the end of the tidal cycle). Chronological collection times for each of the four sites vary. At the time of sample collection, latitude, longitude, time and depth were recorded. The depths at the Cabretta and Dean Creek sites were estimated as sampling took place from a bridge. Samples collected were immediately placed on ice, in the dark and taken back to Sapelo Island NERR Laboratory to be processed prior to shipping to the Virginia Institute of Marine Sciences laboratory for analysis.

Processing each sample:
Using filter towers (acid-washed towers with a 0.45 μm polycarbonate filter for nutrient filtering and clean towers with a GF/F filter for chlorophyll filtering), a small amount of sample was used to rinse the nutrient filter tower equipped with a filter and then the filtrate was discarded. The tower was then filled to the 250 mL mark. The chlorophyll tower with the GF/F filter was also filled to the 250 mL mark (or 500 mL mark if a larger filtration apparatus was used) and the towers were connected by small piece(s) of tubing. The vacuum pump was turned on to pull the sample through each filter and then the vacuum was released. The nutrient sample tower was disconnected and an acid-washed 250 mL polypropylene bottle was rinsed and filled with the

filtrate. Space was left in the sample bottle for expansion during freezing at approximately –18°C. If the first 250 or 500 milliliters went through the chlorophyll filter easily, the filtrate was discarded and an additional 50, 100, 250 or 500 milliliters was filtered, depending on suspended sediment load, to concentrate the sample onto the filter. The chlorophyll filter was then removed with tweezers and placed face up in a petri dish, wrapped in aluminum foil and labeled with the volume filtered and sample information. The chlorophyll filter towers were rinsed between replicate grabs with distilled water and the nutrient filter tower was acid-washed and DI water rinsed between samples.

Note: Enterococci were reported for HD and LD grab samples only, likely due an interest in oyster aquaculture in the Duplin River. These samples were not filtered or preserved.

b) Diel Sampling Program

As of November 2013, Reserve staff have been conducting all field work associated with this project. The recommended procedures for diel scheduling and sampling are as follows:
WWW Tide and Current Predictor for Wolf Island, South End was used to estimate low tide. As close to an early, low, neap tide as possible was selected each month for sampling. The ISCO sampler was deployed at the Lower Duplin (LD) site on the day previous to the grab sampling date chosen for that particular month with the sample line suction tube placed 1.5 feet below the surface of the water. The ISCO sampler collected the first diel sample as close as possible to the low tide predicted for the following day and continued collecting samples every two hours for the next 24 hours, representing a full tidal cycle and a total of 13 samples, ending at low tide near to the time when grab sampling began. The ISCO was turned off at the end of the collection period and the samples were secured with caps upon arriving at the site. The samples were filter processed in the laboratory by SINERR personnel. The filtration process for the diel samples follows the same process as for grab samples described above, with the exception of chlorophyll filtering. High-density polypropylene bottles were used to store the samples after filtration. Polypropylene bottles and filter towers were soaked in 10% HCl in preparation for the fieldwork, and then triple rinsed with distilled water. A squeeze bottle was used to acid wash (then rinse with distilled water) beakers and filter towers between filtering of each sample.

4) Site location and character

The Sapelo Island National Estuarine Research Reserve is located on the Southeastern Atlantic coast of the United States in McIntosh County, Georgia. The study area encompasses the Duplin River estuary, a tidally flushed drainage system flowing into Doboy Sound from the north and two inland creeks, Cabretta and Dean Creek. The Duplin River watershed occupies most of the Reserve, which also contains various forest types, sand dunes, a section of ocean beach and minor developed areas. The Duplin River estuary covers 3,300 acres between Sapelo Island and the mainland in McIntosh County. It drains a tidal bay and an extensive network of salt marshes about 6 miles long, into which there is little upland run-off. Diverse estuarine wetlands provide extensive and complex habitat types for fish and wildlife. The island contains several small, interior brackish and freshwater marshes fed by surficial aquifer expression (interdune meadow of Nannygoat beach: south end) and anthropogenic upland ditches and dikes produced in the early 19th century (north end). The upland forests are composed of several diverse habitats including long leaf pine/slash pine forests, climax maritime forests, small amounts of pond cypress bays and naturally regenerated loblolly pine forests which are timbered on a 70 year selectively cut harvest rotation. There are no current studies on pollutants in this area. Sapelo Island is typically considered a pristine environment, with minimal pollutant input.

All Sapelo Island NERR historical nutrient/pigment monitoring stations:

Station Code	SWMP Status	Station Name	Location	Active Dates	Reason Decommissioned	Notes
sapcanut	P	Cabretta Creek	31° 26' 37.32 N, 81° 14' 23.64 W	08/01/2004-current	NA	NA
sapdcnut	P	Dean Creek	31° 23' 22.56 N, 81° 16' 44.04 W	05/01/2004 - current	NA	NA
saphdnut	P	Hunt Dock	31° 28' 42.96 N, 81° 16' 23.16 W	03/01/2002 current	NA	NA
sapldnut	P	Lower Duplin	31° 25' 4.59 N, 81° 17' 45.77 W	01/01/2002 - current	NA	NA
sapfdnut	P	Flume Dock	31° 28' 58.08 N, 81° 16' 3.00 W	03/01/2002 - 08/01/2004	Discontinued historic sampling program	
sapmlnut	P	Marsh Landing	31° 25' 3.72 N, 81° 17' 45.96 W	01/01/2002 - 01/01/2004	Discontinued historic sampling program	

Active Sapelo Island NERR nutrient/pigment and water quality monitoring stations:

Latitude and Longitude-
Lower Duplin: Lat: 31º 25’ 4” N, Long: 81º 17’ 46” W
Hunt Dock: Lat: 31º 28’ 43” N, Long: 81º 16’ 23” W
Cabretta Creek: Lat: 31º 26’ 37.3” N, Long: 81º 14’ 23.7” W
Dean Creek: Lat: 31º 23’ 22.5” N, Long: 81º 16’ 44.2” W

Water Quality site descriptions-
Salinities at all Duplin River sites vary according to localized rainfall and associated runoff. The upper Duplin River site (Hunt Dock) experiences slightly lower salinities associated with rainfall events (2 -3 ppt) as compared to the lower Duplin River site. Average salinities range from 15 ppt to 30 ppt depending on seasonal or event rainfall. Average tidal range of diurnal tidal cycle is approximately 2.5 meters twice daily. Due to high turbidity, all Duplin River sites are lacking any persistent submerged aquatic vegetation and have an unconsolidated sandy/mud bottom (soft sediment) typical of southeastern near-ocean estuaries. Marsh sediments are relatively pristine and free of pollutants based on sediment analysis conducted in 1996 by C. Alexander, Skidaway Institue of Oceanography. The watershed is dominated by oceanic tidal influences associated with Doboy Sound. Depths are as follows: Lower Duplin (LD) ranges from 1.5 meters to 6.0 meters depending on tide, and the Hunt Dock site maximum depth is 4.27 meters.

Cabretta Creek is fed directly from waters of the Atlantic Ocean. Cabretta Creek experiences a maximum tidal range of approximately 4.3 meters. Average mean low water depth at the sample site is approximately 3.25 meters. Salinity ranges, with exception to major, long-term precipitation events, from 15-36 ppt, seasonally. The station is located on a small (one-lane), wooden, roadway bridge spanning Cabretta Creek, located on the island’s extreme eastern side. The benthos is composed primarily of sand substrate with small, intertidal oyster reef conglomerate communities. Adjacent to the site is extensive, intertidal, bank stabilization (armoring) in the form of woven rip-rap fencing and granite rocks. This manipulation is slowly becoming stabilized via oyster reef community colonization. The adjacent marshes are dominated by *Spartina alterniflora* with occasional *Juncus*

romerianus in the nearby fringe community habitat. The creek has very little adjacent uplands due to: 1) the low elevational gradient and 2) the area's geologically recent accretion genesis (Holocene) resulting in sandy soils; of which neither condition allows for extensive floral colonization or stabilization.

The Dean Creek site is located on a recently rebuilt steel bridge spanning Dean Creek, in close proximity to the adjacent Nannygoat Beach causeway. Dean Creek is a small tidal basin fed from the waters of Doboy Sound, which is located on Sapelo Island's south end. With exception to short duration local or long duration regional precipitation events, the creek's salinity normally ranges between 20 and 30 ppt. The benthic community consists of a sandy-mud substrate with occasional small, intertidal oyster reef community and mean tidal amplitude of approximately 2.4 m. Average mean low water depth at the sample site is approximately 1 meter, but fluctuates due to bank erosion. The small creek feeds approximately 150 acres of *Spartina alterniflora* dominated salt marsh, which is interspersed with small 0.5-1 acre hammocks and salt pans. Fringe community components range from Loblolly pine forests with a sub-canopy of Yaupon holly to Wax myrtle and Sable Palm.

5) Coded variable definitions

LD = Lower Duplin; HD = Hunt Dock; CA = Cabretta Creek; DC = Dean Creek.

Each individual sample is given a three-part name code in addition to other codes. The three-part name code, "sapldnut" for example, gives the reserve name (sap = Sapelo), station name (LD = Lower Duplin, etc.), and SWMP program code (nut = nutrient monitoring program).

Sampling Site codes:
sapldnut – Sapelo Island nutrient data for Lower Duplin
saphdnut – Sapelo Island nutrient data for Hunt Dock
sapcanut – Sapelo Island nutrient data for Cabretta Creek
sapdcnut – Sapelo Island nutrient data for Dean Creek

The monitoring program codes are set as "1" to indicate grab samples and "2" to indicate diel samples. Replicates are also given specific codes. Grab samples in which replicate field samples are taken utilize a "1" for the first sample and a "2" for the second sample. Only one sample is taken at each interval with the ISCO sampler.

6) Data collection period

Diel sampling for 2017 began at 06:30:00 on January 18, 2017 at the Lower Duplin site. Grab sampling commenced on January 19, 2017 for all sites. Start times for each site are as follows: 10:23:00 at the Hunt Dock site, 10:42:00 at the Lower Duplin site, 10:05:00 at the Cabretta site, and 09:45 at the Dean Creek site. The final sample data for 2017 was November 16, 2017.

Diel Sampling: Start/End, Dates/Times				
Location	**Start Date**	**Start Time**	**End Date**	**End Time**
LD	1/18/2017	6:30	1/19/2017	6:30
LD	3/7/2017	10:30	3/8/2017	10:30
LD	4/4/2017	9:30	4/5/2017	9:30
LD	5/16/2017	9:00	5/17/2017	9:00

LD	6/21/2017	11:30	6/22/2017	11:30
LD	7/18/2017	10:30	7/19/2017	10:30
LD	8/15/2017	9:00	8/16/2017	9:00
LD	9/27/2017	9:00	9/28/2017	9:00
LD	10/25/2017	6:30	10/26/2017	6:30
LD	11/15/2017	10:00	11/16/2017	10:00

Grab Sampling:
Dates; Start and End Times

Date	CA Start	CA End	DC Start	DC End	LD Start	LD End	HD Start	HD End
1/19/2017	10:05	10:06	9:45	9:46	10:42	10:46	10:23	10:25
3/8/2017	10:15	10:17	10:45	10:46	10:31	10:35	9:50	9:53
4/5/2017	10:28	10:31	11:05	11:08	10:42	10:44	10:15	10:17
5/17/2017	9:58	10:00	9:31	9:32	10:50	10:53	10:20	10:23
6/22/2017	10:05	10:07	9:39	9:41	11:05	11:08	10:21	10:23
7/19/2017	9:50	9:52	9:25	9:27	10:45	10:47	10:15	10:17
8/16/2017	10:15	10:14	9:50	9:52	10:55	10:57	10:32	10:34
9/28/2017	9:40	9:42	9:21	9:23	10:29	10:32	9:55	9:59
10/26/2017	10:05	10:07	9:22	9:25	11:01	11:03	10:40	10:43
11/16/2017	10:21	10:23	9:44	9:45	11:09	11:11	10:48	10:50

7) Associated researchers and projects

As part of the SWMP long-term monitoring program, SAP NERR also monitors Meteorological and Water Quality data which may be correlated with this Nutrient dataset. These data are available from the Research Coordinator or online at www.nerrsdata.org.

For a complete viewing of associated projects visit the following website and search the collaborators links:
http://gce-lter.marsci.uga.edu/lter/
http://www.uga.edu/marine_advisory/

8) Distribution

NOAA retains the right to analyze, synthesize and publish summaries of the NERRS System-wide Monitoring Program data. The NERRS retains the right to be fully credited for having collected and process the data. Following academic courtesy standards, the NERR site where the data were collected should be contacted and fully acknowledged in any subsequent publications in which any part of the data are used. The data set enclosed within this package/transmission is only as good as the quality assurance and quality control procedures outlined by the enclosed metadata reporting statement. The user bears all responsibility for its subsequent use/misuse in any further analyses or comparisons. The Federal government does not assume liability to the Recipient or third persons, nor will the Federal government reimburse

or indemnify the Recipient for its liability due to any losses resulting in any way from the use of this data.

Requested citation format:
NOAA National Estuarine Research Reserve System (NERRS). System-wide Monitoring Program. Data accessed from the NOAA NERRS Centralized Data Management Office website: www.nerrsdata.org; *accessed* 12 October 2016.

NERR nutrient data and metadata can be obtained from the Research Coordinator at the individual NERR site (please see Principal investigators and contact persons), from the Data Manager at the Centralized Data Management Office (please see personnel directory under the general information link on the CDMO home page) and online at the CDMO home page www.nerrsdata.org. Data are available in comma separated version format.

II. Physical Structure Descriptors

9) Entry verification

Nutrient data are entered into a Microsoft Excel worksheet and processed using the NutrientQAQC Excel macro. The NutrientQAQC macro sets up the data worksheet, metadata worksheets, and MDL worksheet; adds chosen parameters and facilitates data entry; allows the user to set the number of significant figures to be reported for each parameter and rounds using banker's rounding rules; allows the user to input MDL values and then automatically flags/codes measured values below MDL and inserts the MDL; calculates parameters chosen by the user and automatically flags/codes for component values below MDL, negative calculated values, and missing data; allows the user to apply QAQC flags and codes to the data; produces summary statistics; graphs selected parameters for review; and exports the resulting data file to the CDMO for tertiary QAQC and assimilation into the CDMO's authoritative online database.

A Skalar Continuous Flow Analyzer with Flow Access software is used to analyze nutrient concentrations. The instrument is calibrated daily for each parameter to be tested using a series of working standards. Once the calibration run is complete and satisfactory (r >/= 0.99500 up to 1.0000), the samples are set up for analysis. A set of mid-range check standards is used before the sample run, after approximately every 10 samples and at the end of the run to ensure the instrument is in control. The check standards must remain within ± 10% of their original value during the entire run. Also, a blank sample is run and then spiked with each analyte to a known concentration, which must come out within ± 10% as well. An external standard independent of calibration standards is processed with each set of samples. Once the run is complete, the raw data is reviewed on the computer attached to the Skalar Continuous Flow Analyzer with Flow Access software, and the timing is checked to ensure proper integration of sample peaks. Once this is completed, the data is exported via network to another computer. The data file for each month is saved and the results are copied into a comprehensive file with all results and downloaded in the laboratory's Laboratory Information Management System (LIMS).

10) Parameter titles and variable names by category

Required NOAA/NERRS System-wide Monitoring Program nutrient parameters are denoted by an asterisks "*".

Data Category	Parameter	Variable Name	Units of Measure

Phosphorus and Nitrogen:

*Orthophosphate	PO4F	mg/L as P
*Ammonium, Filtered	NH4F	mg/L as N
*Nitrite, Filtered	NO2F	mg/L as N
*Nitrate, Filtered	NO3F	mg/L as N
*Nitrite + Nitrate, Filtered	NO23F	mg/L as N
Dissolved Inorganic Nitrogen	DIN	mg/L as N

Plant Pigments:

*Chlorophyll a	CHLA_N	µg/L
Phaeophytin	PHEA	µg/L

Microbial:

Enterococci	ENTERO_MPN	MPN/100ml

Notes:
1. Time is coded based on a 2400 clock and is referenced to Standard Time.
2. Reserves have the option of measuring either NO2 and NO3 or they may substitute NO23 for individual analyses if they can show that NO2 is a minor component relative to NO3.

11) Measured or calculated laboratory parameters

a) **Parameters measured directly**

Nitrogen species:	NH4F, NO2F, NO23F
Phosphorus species:	PO4F
Other:	CHLA, PHEA, ENTERO_MPN

b) **Calculated parameters**

NO3F	NO23F-NO2F
DIN	NO23F+NH4F

12) Limits of detection

Minimum detection limits in 2017 were determined using the "Federal Register". This evaluation is made yearly from a sample obtained from the Chesapeake Bay at low concentrations, or alternatively, a standard made up in synthetic seawater may be acceptable. The standard should be made at a concentration of 1-5 times the expected MDL. The sample is filtered in-house as required into separate pre-cleaned containers for specific parameters. Particulates are filtered onto appropriate filters. The first 7 replicates are used for the MDL determination. The standard deviation from the seven results is calculated and then multiplied by the "student t" value (3.143 or rounded to 3 for seven replicates).

Parameter	Variable	Mean Conc. mg/L as N/P	Std. Dev.	MDL mg/L as N/P	Dates in use	Date Revisited
Ammonium	NH4F	0.047	0.001	0.0056	01/01/2017 – 12/31/2017	8/17/2017
Nitrite	NO2F	0.139	0.001	0.0016	01/01/2017 – 12/31/2017	8/17/2017
Nitrite + Nitrate	NO23F	0.126	0.001	0.0047	01/01/2017 – 12/31/2017	8/17/2017
Orthophosphate	PO4F	0.087	0.001	0.0020	01/01/2017 – 12/31/2017	8/17/2017

Chl-a	CHLA	0.7987 µg/L	0.0094	0.50 µg/L	01/01/2017 – 12/31/2017	8/17/2017
Phaeophytin	PHEA	4.0642 µg/L	0.1389	0.50 µg/L	01/01/2017 – 12/31/2017	8/17/2017

Enterococcus is a test for the presence and quantified based on how many wells are positive. If one well is positive the normal detection limit would be 1 MPN, but the method for marine samples is to only use 10 mls in the analysis (instead of 100ml), so basically a 1:10 dilution. Thus 1 MPN becomes 10 MPN for detection.

13) Laboratory methods

a) Parameter: NH4F

Method Reference: Standard Methods 4500-NH_3 H.
U.S. EPA 1983. USEPA-600/4-79-020. Method 350.1.

Method Descriptor: Samples were filtered with a 0.45 µm membrane filter and subjected to hypochlorite, which in the presence of phenol, catalytic amounts of nitroprusside and excess hypochlorite, yields indophenol blue, which measured at 630 nm is proportional to the original ammonia concentration.

Preservation Method: Samples are filtered and stored frozen (-20°C).
Holding Time: 28 days

b) Parameter: NO23F

Method Reference: Standard Methods 4500-NO_3 F.
U.S. EPA 1974. Method 353.2.

Method Descriptor: Samples were filtered with 0.45 µm polycarbonate filters. Filtered sample is subjected to cadmium reduction column to reduce nitrate to nitrite. The sample nitrite is then determined by diatizing with sulfanilamide and coupling with N-(1-napthyl)-ethylenediamine dihydrochloride to form a highly colored azo dye which is measured at 520 nm and is proportional to the original nitrate + nitrite concentration. The NO2F concentration (below) is subtracted from this result to give NO3F.

Preservation Method: Samples are filtered and stored frozen (-20°C).
Holding Time: 28 days

c) Parameter: NO2F

Method Reference: Standard Methods 4500-NO_3 F.
U.S. EPA 1974. Method 353.2.

Method Descriptor: Samples were filtered with 0.45 µm polycarbonate filters. Nitrite in a filtered sample is measured by closing off the cadmium reduction column so that the nitrate is not converted and the sample follows through the same chemistry as with NO3F to yield the original nitrite concentration.
Preservation Method: Samples are filtered and stored frozen (-20°C).
Holding Time: 28 days

d) Parameter: NO3F

Method Reference: Standard Methods 4500-NO_3 F.
U.S. EPA 1974. Method 353.2.

Method Descriptor: Nitrate is calculated from NO23F minus NO2F results.
Preservation Method: Samples filtered and stored frozen (-20°C).
Holding Time: Nitrate is calculated from NO23F minus NO2F results.

e) Parameter: DIN

Method: DIN is calculated by adding the NH4F and NO23F results together.

f) Parameter: PO4F

Method Reference: Standard Methods 4500-P E.
U.S. EPA 1978. Method 365.1.

Method Descriptor: Samples were filtered with 0.45 μm polycarbonate filters. Filtered sample is subjected to ammonium molybdate and antimony potassium tartrate under acidic conditions to form a yellow complex. This complex is reduced with ascorbic acid to form a blue complex, which absorbs light at 880 nm. The absorbance is proportional to the concentration of orthophosphate in the sample.
Preservation Method: Samples are filtered and stored frozen (-20°C).
Holding Time: 28 days

g) Parameter: CHLA_N and PHEA

Method Reference: EPA 445 REV 1.2

Method Descriptor: Suspended sediment and other material in a water sample is concentrated onto a 47 mm GF/F filter under low vacuum. The sample is stored in a petri dish wrapped in aluminum foil in an airtight plastic bag kept on ice while in the field. The samples are then kept frozen and in the dark until analysis. The acetone extraction method is used to extract the chlorophyll over 2-24 hours and a fluorometer is used to obtain readings, which are calculated into a final result. The sample is then acidified and read again for pheophytin.
Preservation Method: Filters are stored frozen (-20°C).
Holding Time: 28 days

h) Parameter: ENTERO_MPN

Method Reference: Determination of Enterococcus in Marine and Freshwater by Enterolert™

Method Descriptor: The Enterolert™ system produced by the IDEXX Laboratories, Inc., uses 4-methylumbelliferyl-β-glucuronide as the defined substrate nutrient indicator. This compound when hydrolyzed exhibits fluorescence at 365nm wavelength. Non-enterococci groups are suppressed and cannot metabolize the substrate nutrient indicator. The Quanti-tray system developed by IDEXX Laboratories, Inc., is used for enumeration of the enterococcus present in the water sample. The Enterolert™ system used for marine and estuarine samples has a reporting limit of 10 MPN when 10 milliliters of sample are diluted to 100 milliliters with sterile, nonbuffered, oxidant free water as the procedural notes require. The Enterolert detects enterococci at 1 cfu (colony forming unit) per 100 ml sample.
Preservation Method: None
Holding Time: <24 hours

14) Field and Laboratory QAQC programs

a) **Precision**

i) **Field variability** – Field replicates are successive grab samples. Duplicate grabs are collected. Samples are filtered and placed on ice before the next sample is grabbed (usually about 10 minutes between grabs).

ii) **Laboratory variability** – All samples are analyzed in duplicates.

iii) **Inter-organizational splits** – Samples were analyzed by one lab.

b) Quality Control

i) The laboratory is required to operate a formal quality control (QC) program. The minimum requirements of this program consist of an initial demonstration of laboratory capability and the continued analysis of laboratory reagent blanks, field duplicates (where applicable) and quality control samples as a continuing check on performance. The laboratory will maintain performance records that define the quality of the data generated.

ii) Initial Daily Calibration - Correlation coefficient (r): The correlation coefficient must be 0.995 or better for the calibration curve to be used. Calibration standards should bracket the range of samples analyzed.

iii) Laboratory Control Sample (LCS) – Certified reference material - The laboratory must analyze an ammonia certified LCS to verify the accuracy of the initial calibration. Alternatively, a material from a second-source or lot that is traceable to a national standard may be used.

iv) A CCV standard and blank are analyzed every 10 samples to assess drift throughout the run. If the CCV exceeds 10% of the true value, then a new calibration must be performed and the previous 10 samples reanalyzed. If the blank exceeds the lowest standard, then a new calibration must be performed and the previous 10 samples reanalyzed.

v) Duplicates and sample spikes must be performed for every 10 samples analyzed. Duplicate reproducibility should be $\leq$ 20% with the exception of samples which fall at or below the MDL or in the low end of the calibration curve at the lowest standard. In this instance, a subsequent sample falling in the proper range validates the run. Sample spikes should recover $\geq$ 80% of the spike added. If the sample does not recover the appropriate spike, the sample and spike should be repeated. If it again does not meet the QC requirements, the sample may be determined to have a matrix interference if all other sample spikes in the run have met the required QC acceptance limits. This sample will be reported to the client as having a matrix interference.

vi) A back calculation of calibration standards must be performed. The calibration standard must meet +/-10% of the true value criteria for each point except for standards falling at or below the PQL/MRL (approximately 3.18 MDL). Standards falling at or below the PQL must recover at +/-30% of the true value of the calibration standard.

vii) A QC sample spiked at 1-2 times the PQL/MRL (approximately 3.18 MDL) must be analyzed at least quarterly. The MRL should not exceed +/-20% of the back calculated standard.

15) QAQC flag definitions

QAQC flags provide documentation of the data and are applied to individual data points by insertion into the parameter's associated flag column (header preceded by an F_). QAQC flags are applied to the nutrient data during secondary QAQC to indicate data that are out of sensor range low (-4), rejected due to QAQC checks (-3), missing (-2), optional and were not

collected (-1), suspect (1), and that have been corrected (5). All remaining data are flagged as having passed initial QAQC checks (0) when the data are uploaded and assimilated into the CDMO ODIS as provisional plus data. The historical data flag (4) is used to indicate data that were submitted to the CDMO prior to the initiation of secondary QAQC flags and codes (and the use of the automated primary QAQC system for WQ and MET data). This flag is only present in historical data that are exported from the CDMO ODIS.

-4 Outside Low Sensor Range
-3 Data Rejected due to QAQC
-2 Missing Data
-1 Optional SWMP Supported Parameter
0 Data Passed Initial QAQC Checks
1 Suspect Data
4 Historical Data: Pre-Auto QAQC
5 Corrected Data

16) QAQC code definitions

QAQC codes are used in conjunction with QAQC flags to provide further documentation of the data and are also applied by insertion into the associated flag column. There are three (3) different code categories, general, sensor, and comment. General errors document general problems with the sample or sample collection, sensor errors document common sensor or parameter specific problems, and comment codes are used to further document conditions or a problem with the data. Only one general or sensor error and one comment code can be applied to a particular data point. However, a record flag column (F_Record) in the nutrient data allows multiple comment codes to be applied to the entire data record.

General errors

GCM	Calculated value could not be determined due to missing data
GCR	Calculated value could not be determined due to rejected data
GDM	Data missing or sample never collected
GQD	Data rejected due to QA/QC checks
GQS	Data suspect due to QA/QC checks
GSM	See metadata

Sensor errors

SBL	Value below minimum limit of method detection
SCB	Calculated value could not be determined due to a below MDL component
SCC	Calculation with this component resulted in a negative value
SNV	Calculated value is negative
SRD	Replicate values differ substantially
SUL	Value above upper limit of method detection

Parameter Comments

CAB	Algal bloom
CDR	Sample diluted and rerun
CHB	Sample held beyond specified holding time
CIP	Ice present in sample vicinity
CIF	Flotsam present in sample vicinity
CLE	Sample collected later/earlier than scheduled
CRE	Significant rain event

CSM	See metadata
CUS	Lab analysis from unpreserved sample

Record comments

CAB	Algal bloom
CHB	Sample held beyond specified holding time
CIP	Ice present in sample vicinity
CIF	Flotsam present in sample vicinity
CLE	Sample collected later/earlier than scheduled
CRE	Significant rain event
CSM	See metadata
CUS	Lab analysis from unpreserved sample

Cloud cover

CCL	clear (0-10%)
CSP	scattered to partly cloudy (10-50%)
CPB	partly to broken (50-90%)
COC	overcast (>90%)
CFY	foggy
CHY	hazy
CCC	cloud (no percentage)

Precipitation

PNP	none
PDR	drizzle
PLR	light rain
PHR	heavy rain
PSQ	squally
PFQ	frozen precipitation (sleet/snow/freezing rain)
PSR	mixed rain and snow

Tide stage

TSE	ebb tide
TSF	flood tide
TSH	high tide
TSL	low tide

Wave height

WH0	0 to <0.1 meters
WH1	0.1 to 0.3 meters
WH2	0.3 to 0.6 meters
WH3	0.6 to > 1.0 meters
WH4	1.0 to 1.3 meters
WH5	1.3 or greater meters

Wind direction

N	from the north
NNE	from the north northeast
NE	from the northeast
ENE	from the east northeast
E	from the east
ESE	from the east southeast
SE	from the southeast
SSE	from the south southeast
S	from the south
SSW	from the south southwest
SW	from the southwest

WSW	from the west southwest
W	from the west
WNW	from the west northwest
NW	from the northwest
NNW	from the north northwest

Wind speed

WS0	0 to 1 knot
WS1	> 1 to 10 knots
WS2	> 10 to 20 knots
WS3	> 20 to 30 knots
WS4	> 30 to 40 knots
WS5	> 40 knots

17) Other remarks/notes

Data may be missing due to problems with sample collection or processing. Laboratories in the NERRS System submit data that are censored at a lower detection rate limit, called the Method Detection Limit or MDL. MDLs for specific parameters are listed in the Laboratory Methods and Detection Limits Section (Section II, Part 12) of this document. Concentrations that are less than this limit are censored with the use of a QAQC flag and code, and the reported value is the method detection limit itself rather than a measured value. For example, if the measured concentration of NO23F was 0.0005 mg/l as N (MDL=0.0008), the reported value would be 0.0008 and would be flagged as out of sensor range low (-4) and coded SBL. In addition, if any of the components used to calculate a variable are below the MDL, the calculated variable is removed and flagged/coded -4 SCB. If a calculated value is negative, it is rejected and all measured components are marked suspect. If additional information on MDL's or missing, suspect, or rejected data is needed, contact the Research Coordinator at the Reserve submitting the data.

Note: The way below MDL values are handled in the NERRS SWMP dataset was changed in November of 2011. Previously, below MDL data from 2007-2010 were also flagged/coded, but either reported as the measured value or a blank cell. Any 2007-2011 nutrient/pigment data downloaded from the CDMO prior to November of 2011 will reflect this difference.

Sample hold times for 2017: Nutrient and pigment samples are held at -20°C. NERRS SOP allows nutrient samples to be held for up to 28 days (CHLA for 30) at -20°C, plus allows for up to 5 days for collecting, processing, and shipping samples. Enterococci samples were to be processed within 24 hours of receipt by lab. Samples held beyond that time period are marked with an asterisk below, and are flagged suspect and coded CHB in the dataset. If measured values were below MDL, this resulted in <-4> [SBL] (CHB) flagging/coding.

Month	O-PO4	NH4	Nox	NO2	CHL/PHEA	ENTERO
01/18-19/2017	2/9-2/10/2017	2/9-2/10/2017	2/9-2/10/2017	2/9-2/10/2017	2/8/2017	1/20/2017
Feb - no samples	no samples	no samples	no samples	no samples	no samples	no samples
03/07-08/2017	3/23/2017	3/23/2017	3/23/2017	3/23/2017	3/21/2017	3/18/2017*
04/04-05/2017	5/2/2017	5/2/2017	5/2/2017	5/2/2017	5/1/2017	4/18/2017*
05/16-17/2017	6/8/2017	6/8/2017	6/8/2017	6/8/2017	6/1/2017	5/18/2017

06/21-22/2017	7/20/2017	7/20/2017	7/20/2017	7/20/2017	6/29/2017	6/23/2017
07/18-19/2017	8/16/2017	8/17/2017	8/16/2017	8/16/2017	7/31/2017	7/20/2017
08/15-16/2017	9/7/2017	9/7/2017	9/7/2017	9/7/2017	no samples	8/17/2017
09/27-28/2017	10/25-26/2017	10/25-26/2017	10/25-26/2017	10/25-26/2017	10/23/2017	9/29/2017
10/25-26/2017	11/6/2017	11/6/2017	11/6/2017	11/6/2017	11/16/2017	10/27/2017
11/15/-16/2017	12/6/2017	12/6/2017	12/6/2017	12/6/2017	1/8/2018	11/17/2017
Dec - no samples	no samples	no samples	no samples	no samples	no samples	no samples

Additional notes:

- CHLA/PHEA samples were only processed for grab samples in 2017. The diel pigment samples were lost in translating our protocol from handing raw water samples to the local UGA MAREX laboratory to shipping to VIMS, which required filtering the pigment samples in-house.
- Samples taken 2/23-24/2017 were received by the laboratory 3 days later on February 27, 2017 and the temperature of the samples was documented at 9.5°C. Due to the time lag and high temperature of the samples, the batch was rejected.
- August CHLA/PHEA grab samples were lost at the lab and sat out too long to be viable.
- Samples were not taken in December 2017, due to logistical and weather problems.