Florida National Marine Sanctuary Survey Field Sampling Protocol (last modified 12/2/2020)

A. Field Sampling Preparation and Checklist

SERC performs water-column sampling and measurement of the physical parameters listed in Table 1.0. Preceding a sampling event, the field personnel are required to ensure that all supplies and equipment are prepared for the sampling event. Table 2.0 lists the field sampling equipment used for sampling each matrix, while the following list is an equipment checklist prepared for the sampling team.

Water sampling equipment may include plastic sample containers, syringes, filter holders, Niskin Bottle samplers, buckets, YSI multi-parameter data loggers, Seabird© Conductivity, Temperature and Depth (CTD) profilers, and LaMotte turbidity meters.

TABLE 1.0

SERC Sampling Capabilities

Parameter Group	Sample Source
Field Parameters Temperature Salinity/Conductivity Dissolved Oxygen	Surface Water/Water Column (Seabird SBE-19 or SBE-19plus seacat profiler)
PAR	Surface/Water Column (LiCor PAR irradiance Spherical Quantum Sensor) model# 193SA
CDOM	Surface/Water Column (WETLabs ECO-CDOM Sensor)
Turbidity	Surface Water/Bottom Water (LaMotte 2020 WE Turbidity meter)
Field Parameters Salinity/Conductivity	Surface Water (YSI EXO2 multi-parameter logger)

Table. 2.0. Field Sampling Equipment

Field Sampling Equipment	Construction	Use	Parameter Groups	Notes
Syringes	140 ml plastic (Latex-free)	Collection	Soluble nutrients, Suspended matter, Chlorophyll	None
Bottles	60 ml plastic (HDPE)	Collection	Soluble nutrients	Filtered samples only
Bottles	125 ml plastic (HDPE)	Collection	Total nutrients	Non-filtered
Micro-centrifuge Tubes	2.0 ml plastic (HDPE)	Filter storage	Suspended matter, Chlorophyll-a	Chlorophyll-a preserve with acetone
Wide-mouth Amber Bottle	250 ml plastic (HDPE)	Storage	Suspended matter, Chlorophyll-a	Dark storage for Chlorophyll-a containers
Bucket	2-5 gallon plastic (LDPE)	Collection	All surface parameters	None
Niskin Sampler (model 1010)	2.5 L Teflon coated PVC	Collection	All surface/bottom parameters	None
In-line Filter Holders	2.5 cm plastic (Gellman)	Filtration	Soluble nutrients, Suspended matter, Chlorophyll-a	Attach to end of syringe
Filters	2.5 cm Whitman GF/F glass fiber	Filtration	Soluble nutrients, Suspended matter, Chlorophyll-a	Use filter forceps to place into and sample tube
Acetone	90%, ACS grade	Preservation	Chlorophyll-a	Store in a 60 ml HDPE bottle in a Ziploc bag
Disposable pipettes	Plastic (polyethylene)	Dispensing	Chlorophyll-a	Used to dispense 90% acetone
Forceps	Metal	Handling	Chlorophyll-a	None
DI water	1 L plastic (HDPE) 250 ml plastic (HDPE) squeeze bottle	Equipment blanks Rinsing	All parameters	None

KimTech Kimwipes®	Cellulose	Cuvette wipes	Turbidity	Used to clean Turbidity cuvettes
Field Sampling Equipment	**Construction**	**Use**	**Parameter Groups**	**Notes**
Coolers	Plastic	Transportation	All parameters	None
Field datasheets	Field data sheets Instrument calibration sheets Field equipment checklist	Documentation	All parameters	None
Pens	Waterproof	Documentation	All parameters	None
Labeling tape	Waterproof	Documentation	All parameters	None
Ice		Preservation	Total and Soluable nutrients, Chlorophyll-a	Need sufficient quantity to ensure even temperature distribution
DI water	1 L plastic (HDPE) 250 ml plastic (HDPE) squeeze bottle	Equipment blanks Rinsing	All parameters	None
Site charts	Waterproof	Reference	All parameters	None
Field instruments	Seabird SBE-19 or SBE19plus CTD YSI EXO2 Mult-parameter Data Logger LaMotte 2020 WE Turbidity Meter	Field measurements	All parameters Salinity Only Turbidity Only	Data Collection Salinity check for laboratory analysis Data Collection

Field Equipment Checklist:

Water Sampling Equipment

- LIMS Labeled sample bottles (narrow-mouth plastic)
    - 60 ml plastic (HDPE)
    - 125 ml plastic (HDPE)
- 140 ml clean plastic syringes (extra for backup)
- 2.0 ml Micro-centrifuge tubes pre-labeled
- 250 ml Wide-mouth amber bottle for dark storage
- 2.5 cm Gellman in-line filter holders pre-loaded with 2.5 cm Whatman GF/F glass fiber filters
- 1 box 2.5 cm Whitman GF/F glass fiber filters
- 2 boxes of Kimwipes® for cleaning turbidity cuvettes
- 2 Filter forceps
- 2-5 gallon plastic bucket with rope
- 3 Niskin Bottle water samplers (surface, bottom, and a backup)
- DI water (4) 1L for equipment blanks and rinsing

Field Measurement Equipment

- Seabird SBE19 CTD multi-parameter data logger\profiler
- Seabird SBE19*plus* CTD multi-parameter data logger\profiler
- YSI EXO2 multi-parameter data logger\profiler with handheld display
- LaMotte 2020 WE Turbidity meter

Sample Preservation

- 90% Acetone
- Disposable polyethylene pipettes
- Ice
- Coolers
- NIST – calibrated thermometer

Boat Supplies

- Depth finder
- GPS Chartplotter (and 1-2 additional GPS handheld units)
- VHF Radio (and additional handheld VHF Radio)
- Cell phone
- Satellite phone (if traveling outside cell phone coverage area)
- Hard copy with site list and waypoints of sampling locations
- Coast Guard required equipment and safety gear
- Charts
- Tool box

Field Equipment Checklist (continued)

Miscellaneous Equipment

- Clipboard with waterproof field data and calibration sheets
- Waterproof Pens
- Waterproof label tape
- Deionized water squeeze bottle (filled)
- Watch
- QAP plan (available in the field for reference)
- Blank Water\Control Water

1. Field Sampling Equipment Calibration

a. Seabird SBE19 CTD multi-parameter data logger\profiler Seabird SBE19plus CTD multi-parameter data logger\profiler

The SEB 19 and SBE19*plus* are designed to measure conductivity, temperature, and pressure in both marine and freshwater environments at depths up to 7000 meters. These instruments operate in a profile mode for acquiring vertical profiles. The SBE 19 and 19*plus* run continuously, sampling at 4 scans per second (4 Hz) and averaging the scan to log a reading every 0.5 second. These instruments are self-powered and self-contained and includes conductivity and temperature sensors and a precision, semiconductor, strain gauge pressure sensor. Nine D-sized alkaline batteries provide 60 hours of operation in profile mode and these are replaced prior to each sample survey. Setup, diagnostics, and data extraction are performed without opening the housing through a data cable. Data output is in raw data form or engineering units (.hex or XML).

Specifications: (based on factory specifications sheet)

Conductivity (S/m)
- Measurement Range: 0 to 9
- Initial Accuracy: 0.0005
- Typical Stability: 0.0003/month
- Resolution: 0.00005

Temperature (◦C)
- Measurement Range: -5 to +35
- Initial Accuracy: 0.005
- Typical Stability: 0.0002/month
- Resolution: 0.0001

Pressure Strain Gauge (m)
- Measurement Range: 0 to 20/100/35/600/1000/2000/3500/7000 meters.
- Initial Accuracy: 0.1% of full-scale range
- Typical Stability: 0.1% of full-scale range/year
- Resolution: 0.002% of full-scale range

The SBE 19 and 19*plus* are returned to the factory every 1000 hours of use or less to perform a calibration check. The criteria for calibration are based on the amount of sensor drift between diagnostic checks. If the sensor has drifted outside the range of "Typical Stability", the sensor is then calibrated. If the sensor fails calibration it is replaced, and new calibration coefficients are generated*. Sea-Bird sensors are calibrated by subjecting them to known physical conditions and measuring the sensor responses.* Coefficients are then computed, which may be used with appropriate algorithms to obtain engineering units. The conductivity, temperature, and pressure sensors on the SBE 19 and SBE 19*plus* are supplied fully calibrated, with coefficients printed on their respective Calibration Certificates and stored in the instrument configuration (.xmlcon or .con) file. Calibration reports and associated .con files are generated by the factory and saved electronically for reference. Factory calibration reports of sensor drift are evaluated to determine if the drift between calibration events is within FDEP SOP verification acceptance criteria in (Table FT 1000-1). If any of the sensors have drifted outside the criteria, the data collected with that instrument will be evaluated to determine if a qualifier is needed. A "J" qualifier and comment would then be attached to each data point for which the instrument was not within the accepted criteria.

Auxiliary Sensors:

SBE 43 Oxygen Sensor (mg/L)

Measurement Range: 0.00 – 20.00 mg/L
Initial Accuracy: 2% of saturation
Typical Stability: 0.5% per 1000 hours (with clean membrane)

The SBE 43 is calibrated at six different temperature points ranging from 2 – 30 °C. At each temperature point oxygen/nitrogen is diffused into the bath and three different oxygen points are taken (7, 4, and 1 ml/L) and the results are displayed in the calibration report.

ECO-CDOM sensor (mg/mˆ3) WET labs sensor

Measurement Range: 0.00 – 50.00 mg/mˆ3 or 0 – 500 ppb
Excitation: 370 nm
Emission: 460 nm
Sensitivity: 0.09 ppb QS

Note: This sensor is delivered from WET Labs calibrated and must returned to WET Labs for calibration. After calibration Seabird adds the calibration coefficients to the CTD software.

LiCor (LI-193) Spherical Quantum Sensor – The LI-193 measures PAR underwater from all directions at depths up to 350 meters. It uses a diffusive sphere to direct light through glass optical filters to the silicon photodiode. The filters create nearly uniform sensitivity to

light between 400 and 700 nm, which closely corresponds to light used by most terrestrial and aquatic plants and algae.

Specifications:

Absolute Calibration: ± 5% in air traceable to NIST
Sensitivity: Typically, 7 µA per 1,000 µmol s^{-1} m^{-2} in water
Linearity: Maximum deviation of 1% i[tp 10,000 µmol s^{-1} m^{-2}
Stability: < ± 2% change over a 1-year period
Response Time: 10 µS
Angular Response: < ± 4% error up to ± 90˚ from normal axis
Operating Temperature Range: - 40 ˚C to 65 ˚C
Detector: High stability silicon photovoltaic detector (blue enhanced)

Note: This sensor is delivered from LiCor calibrated and must be returned to LiCor for calibration. After calibration Seabird adds the calibration coefficients to the CTD software.

b. YSI EXO2 multi-parameter data sonde –

A Multiparameter Water Quality Sonde with 7 sensor ports (including a central wiper port) and optional depth sensor, is used to take surface water quality readings. Salinity readings are calculated from conductivity and temperature sensors.

Sensor Specifications

Conductivity (mS/cm)
- Range: 0 to 200 mS/cm
- Accuracy: (0 to 100) ± 0.5% of reading or 0.001 mS/cm
  (100 to 200) ± 1% of reading
- Response: T63<2 sec
- Resolution: 0.0001 to 0.01 mS/cm (range dependant)

Temperature (˚C)
- Range: -5 to 35 ˚C
  35 to 50 ˚C
- Accuracy: ± 0.01 ˚C2
  ± 0.05 ˚C2
- Response: T63<1 sec
- Resolution: 0.001 ˚C

Salinity (ppt)
- Range: 0 to 70 ppt
- Accuracy: ± 1.0% of reading or 0.1 ppt
- Response: T63 < 2 sec
- Resolution: 0.01 ppt

The sonde is calibrated in the laboratory following FDEP FT-series SOP criteria.

Note: Sonde and standards are equilibrated to room temperature prior to calibration.

- The sonde can be connected to the PC via the “wet-mate” connector cable or Bluetooth connection. To “awaken” sonde via Bluetooth, use the sonde magnet.
- Once connected to the PC, open the KOR EXO software and select the *Calibration Menu*, a list of the installed sensors will appear, select the sensor you are calibrating.
- Rinse the probe with analyte-free water and then with the appropriate standard solution.
  Note: When selecting the appropriate standard solution select a standard that best represents the conditions in the field.
- Place the sonde probes inside calibration cup with the appropriate standard solution *Note: some sensors require a two-point calibration.*
- Selecting the parameter will initiate the probe’s calibration in the standard. Allow the sensor readings to stabilize.
- Enter the standard value, the reading should match the standard value when stabilized.
  Note: EXO sensors have SmartQC© technology, the sensor will read Green, Yellow or Red after stabilizing.
  A Green score means the sensor is calibrated properly and all parameters used to assess its performance state are within factory-defined limits. A Yellow score means that the sensor will still perform within factory-defined limits, but that during calibration enough of an adjustment was required to suggest that the sensor is drifting from those limits or may soon require some adjustments. A Red score means that the sensor is not performing within factory-specified limits or a component of the sensor is due to be replaced.
- Once stable, confirm probe is reading within the acceptable margin of error and Select **Apply** to accept the calibration point. Repeat for each calibration point and select **Complete** when all points have been calibrated.
- A calibration summary report is generated with QC score, and the report is saved for records. If a calibration error occurs, repeat the calibration, or preform a factory reset (see manual for details).

- After the Initial Calibration (IC) a Continuing calibration verification (CCV) is preformed, and the sonde is placed in two different standards that bracket the expected range of sample measurements and the values are recorded. These values must meet the calibration acceptance criteria listed in FDEP SOP table FT-1000-1).

Tank Test – A Continuing Calibration Verification (CCV) is performed with each sample survey. Prior to each sample survey all three calibrated instruments (Seabird SBE19, *SBE19plus,* and YSI EXO2) are placed in a large, clean 55-gallon calibration chamber with known physical conditions. The salinity of the tank is adjusted using Instant Ocean and the salinity is verified using the calibrated EXO2 sonde. An aerator is used to fully saturate the water and a certified NIST thermometer is used to determine accurate temperature. The internal clocks of all three instruments are synchronized.
Note: Instrument clocks are synced using the United States Naval Observatory Master Clock (USNO).
The instruments are programmed to log data every second and allowed to run for approximately 30 minutes. The resulting data is then analyzed for consistency between instruments as well as to verify they are reading within FDEP SOP acceptance criteria (see Table FT1000-1). If any of the instruments do not meet this criterion, they are not used in the sampling survey. A post survey verification is preformed after the sample survey to ensure instruments were working properly during the survey. If the instrument fails post verification, a "J" qualifier and comment would then be attached to each data point for which the instrument was not within the accepted criteria. The tank test results, both pre and post, are saved as an excel file and stored for reference.

c. **LaMotte Turbidity 2020 WE turbidity meter –** is a portable, microprocessor controlled, direct reading nephelometer. Turbidity is measured directly by either EPA Method 180.1 or ISO Method 7027. The 2020 uses a multi-detector micro optical configuration that assures long term stability of calibrations, high precision and accuracy and low detections limits. [from manual] EPA turbidity uses a tungsten filament light source that meets or exceeds EPA specifications.

Specifications:

Range: 0 – 4000 NTU/FNU, 1-10, 500 ASBC, 0-150 EBC
Resolution: 0.01 NTU/FNU 10.00-10.99
0.1 NTU/FNU 11.00-109.9
1.0 NTU/FNU 110-4000
Accuracy: From 0-2.5 NTU (accuracy is ±0.05 NTU
From 2.0-100 NTU/FNU (accuracy is ±2%)
From 100 NTU (accuracy is ±3%)
Detection limit: 0.05 NTU/FNU
Reproducibility: 0.02 NTU/FNU or 1%
Light Source: Tungsten (EPA) complies with EPA 180.1. Standard 860 LED (ISO) complies with ISO 7027

The meter is calibrated prior to each sampling survey using factory standards: 0.0, 1.0, and 10.0 NTU and the results are recorded on the calibration sheet along with the following information: date, initials of technician, and lot# of standards used. Calibration checks using (0.0, 1.0, 10.0 NTU) in "run" mode, are performed at the beginning and end of each sample day and the results recorded on the field sheets. Batteries are re-charged after each sampling event and sample cuvettes are rinsed with DI water and air dried.

2. Sampling Equipment and Cleaning Procedures

Water sampling equipment such as Niskin samplers will be used to collect water samples from the appropriate depths. Additional water sampling equipment include 140 ml plastic piston syringes, 2.5 cm plastic (Gellman) in-line filter holders with 2.5 cm Whatman GF/F glass fiber filters. A Seabird SBE19 and SBE19*Plus* CTD data logger/profiler and YSI EXO2 multi-parameter sonde will be used to collect physio-chemical parameters in the field. Immediately following a sampling trip, all reusable field sampling and measurement equipment is cleaned in the laboratory according to the following conditions:

1. Wash all surfaces thoroughly with fresh water.
2. Rinse thoroughly with distilled water.
3. Let air dry completely, or dry with Kimwipes®.
4. Place equipment in field storage boxes or in plastic bags for storage and transport to the field.

a. **Filter holders** used for the collection of suspended matter and chlorophyll-a samples are rinsed three times with distilled water, then allowed to air dry. Clean dry filter holders are kept in a clean plastic bag until next sampling event.

b. **Data Loggers:**

i. **Seabird SBE 19 and SBE *19plus CTD* loggers/profilers –** the pumping mechanisms is flushed with tap water and then DI water after use. Battery volts and memory are checked prior to each sample day. Batteries are replaced when the voltage is below 11 volts (full is 13.1 volts). The memory is cleared at the end of each survey.

ii. **EXO2 logger –**The sonde and probes are rinsed with tap water and then DI water and stored back in the calibration cup with a small amount of water.

B. Field Measurements

The Field Manager or Crew Chief is responsible for all corrective actions in the field during sampling campaigns. To ensure collection of undisturbed samples, the boat is advanced toward a sampling station from the downstream direction whenever possible.

Field measurements at each station include temperature, conductivity/salinity, dissolved oxygen, PAR, turbidity, and depth. At each site a cast of all these parameters will be logged in a surface to bottom profile from the boat. Water column profiles collecting the above parameters every 0.5 second are preformed using Sea-Bird model SBE-19 or SEB 19*plus* CTD data loggers. All users must be properly trained in the use of this instrument, and a copy of the instrument manual taken out into the field during each sampling trip for reference.

1. <u>CTD Cast:</u>
   The magnetic switch is turned to ON position. At this point, the instrumentation will start collecting data. The time and station number are recorded on the field data sheet. The CTD is placed overboard on the sun-lighted side of the boat, at approximately 1-2 meters depth. Once the pump turns on the davit operator will allow the CTD to “soak” 3 minutes to allow the instrument to completely flush with ambient water. The CTD is raised to verify the pump is activated and then submerged again, making sure the PAR sensor is just below the surface; then will begin the slow steady decent to the seafloor. When the CTD reaches the bottom, the davit operator starts the ascent back to the surface. The CTD is recovered, the pumping mechanism is verified and the CTD is turned to the OFF position and secured to the deck

2. <u>EXO2:</u>
   The EXO2 is lowered over the side of the boat at the level of sample collection (within 0.5 m of the surface), and the readings are allowed to stabilize. The salinity reading is recorded on the field sheet and the sonde is removed from the

water and rinsed with analyte-free water and returned to storage cup. *Note: A single salinity reading is taken to note salinity ranges for laboratory analysis, these readings are NOT used in the final report.*

3. LaMotte Turbidity meter:
   A small portion of the unfiltered sample water (125 ml bottle), is used to rinse a clean sample cuvette three times. The cuvette is then filled to the "fill line" and wiped clean with a lint-free Kimwipe®. The sample cuvette is then placed in the Turbidity chamber aligning the index notch with the index arrow, and the reading is recorded on the field sheet. *Note: Both surface and bottom samples are read. (See below in Field Sampling techniques)*

C. Field Sampling Techniques

Water samples are collected using either the actual sample container or Niskin sampler from either side of the boat, as in the case for a small boat, or from a sampling platform, as in the case of the large vessel. It is important that samples are collected away from the boat's engine. Only those persons trained in the proper method of sample collection from a boat or vessel will conduct the sampling.

Three types of water samples are collected: suspended matter samples for chlorophyll-*a* determination, filtered water samples for determination of dissolved nutrients, and unfiltered samples for determination of turbidity, total phosphorus, total nitrogen, total silicate, and total organic carbon. Water samples are collected according to the following procedures:

1. Unfiltered Samples

a. Surface samples:

- Samples are collected by removing the container cap of a125 ml plastic HDPE bottle and slowly submerge the container, opening first, into the water.
- The sample bottle is inverted so the opening is upright and pointing upstream into the oncoming direction of water flow. The sample container is then rinsed three times with sample water prior to filling, allowing water to run slowly into the container until filled. *Note: Rinsate is collected in a small bucket and then discarded after sampling is completed.*
- The sample container is then returned quickly to the surface.
- A small portion of this sample is used to measure turbidity using a LaMotte 2020 WE turbidity meter (see below procedure). The remainder of the sample is stored in a cooler on ice.

b. *Bottom samples: (Niskin Bottle)*

- The site water depth is noted and the Niskin bottle is cocked and lowered to appropriate depth taking care not to disturb the sediments.
- The sample device is rinsed with ample amounts of site water prior to collecting the first sample.
- At the desired depth, the messenger weight is sent down to trip the closure mechanism, and the sample is slowly retrieved to the boat.
- Labeled sample bottles are rinsed three times with site water prior to filling via the discharge tube. *Note: Rinsate is collected in a small bucket and discarded after sample collection.*
- A small portion of the sample water is used to measure turbidity and the labeled sample bottles are then stored on ice.

2. Filtered Samples

a. Surface samples

- Samples are collected using a clean 140 ml syringe. The syringe is rinsed three times with ambient sample water and then filled beyond 140 ml.
- Excess water is discharged to ensure full 140 ml in syringe.
- A Gillman in-line filter holder containing a clean 2.5 cm Whitman GF/F glass fiber filter is placed on the end of the syringe.
- 10 ml of sample water is forced through the filter prior to rinsing sample bottle (60ml).
- The sample bottle is rinsed three times with a small amount of filtered water from the syringe.
- The sample bottle is then filled to the shoulder to allow for expansion during freezing.
- The sample is then placed on ice until return to shore, where it is placed upright in the freezer and freezing time is noted on field sheets.
- The remainder of the water in the syringe is forced through the filter along with a small amount of air to dry the filter. The filter is then collected for Chlorophyll-a analysis and placed in a pre-labeled micro-centrifuge tube with 1.5 ml of 90% acetone and placed in the dark on ice. The amount of water filtered is noted on field sheets.

b. Bottom samples (Niskin Bottle)

- Samples are collected using a clean 140 ml syringe via the discharge tube of the Niskin bottle. The syringe is rinsed three times with ambient sample water and then filled beyond 140 ml.
- Excess water is discharged to ensure full 140 ml in syringe.
- A Gillman in-line filter holder containing a clean 2.5 cm Whitman GF/F glass fiber filter is placed on the end of the syringe.
- 10 ml of sample water is forced through the filter prior to rinsing sample bottle (60ml).
- The sample bottle is rinsed three times with a small amount of filtered water from the syringe.
- The sample bottle is then filled to the shoulder to allow for expansion during freezing.
- The sample is then placed on ice until return to shore, where it is placed upright in the freezer and freezing time is noted on field sheets.

3. **Chlorophyll a sample:**
   Suspended matter samples collected for chlorophyll-a determination are preserved in the field by adding 1.5 ml of acetone to the HDPE micro-centrifuge tube with a disposable pipet. The chlorophyll-a samples are then stored in a cooler in the dark on ice. Equipment blanks for chlorophyll-a determination are preserved with the same amount of acetone and stored in the dark on ice. The acetone preservative used is of ACS reagent-grade or better; obtained fresh from the laboratory stocks on a daily basis and transported to the field in an HDPE bottle.

4. **Field Equipment Blank Checks and QC**
   SERC's field quality control includes the collection of duplicate samples at every 10^{th} sample site (10% of total samples collected). In addition, according to FDEP-FQ1000, both field blanks and equipment blanks are collected during each sample event. *Note: There are four "Surveys" per year (ie: K100) and there are approximately 10 "Sampling Events" or Sample days, within a Survey.*
   Field blanks and Equipment blanks are collected at the beginning and the end of the sampling event/day (C1-1 and C1-2). For Field blanks, analyte-free water is poured directly into the control sample container (unfiltered surface samples). For the equipment blank, analyte-free water is run through the equipment used for sampling such as the Niskin bottle (bottom samples) and the sample syringe (filtered and chlorophyll-a samples) and placed in appropriate bottles and preserved according to each analysis. The filter is saved and placed in a micro-centrifuge tube along with 1.5 mls of 90% acetone and serves as the chlorophyll-a blank. The collections of blank samples are recorded on the field data sheet along with the process times.

Field instrument checks are completed at the beginning of each sampling event, and at the end of the field-sampling event. Field equipment not functioning properly are not used to collect data until they are brought back to the laboratory for repair. Duplicate field equipment and probes are kept on hand during field sampling trips.

Note: Two types of analyte-free water are produced in the laboratory: 1) deionized-distilled water and 2) double-deionized water. The deionized-distilled water is used for washing equipment and glassware, while the double-deionized water is used as reagent water.

Method: Tap water is first deionized using a Culligan system containing activated carbon and 2-mixed bed ion exchange beds followed by filtering through a 0.45 um polypropylene filter cartridge. The water is then either distilled through a Corning Mega-Pure 11 Liter Automatic Water Still to produce the deionized-distilled water or further deionized with a Barnstead model D8911 HN Ultrapure mixed bed deionization cartridge to produce the double-deionized water. Both types of water have proven to be analyte-free for the nutrients analyzed. The quality of this water is frequently checked with laboratory method and field equipment blanks. Containers used to store analyte-free water are kept dedicated to this use.

D. Sample Handling and Custody Requirements

1. Sample Preservation, Holding Times, and Sample Volume

Sample containers, sizes, preservatives, and maximum holding times, by parameter are included in Table 3. Note, there are two columns listing sample holding times prior to analysis. SERC-CAChE/NAL lab recognizes that samples should be analyzed as soon as possible after sample collection. Therefore, SERC has instituted its own maximum holding times as goals. The SERC holding times are almost always met; however, due to the intense sampling schedule of the Florida Keys National Marine Sanctuary Survey (FKNMS), the filtered samples are frozen and analyzed within the maximum holding time. If for any reason the samples are held beyond their maximum holding times, those samples are identified, and the data are flagged with a “Q” qualifier.

Table 3: Sample Preservation and Holding Times

Sample Type/Parameter	Container/Size	Preservation Method	FIU Holding Time (based on CAChE-NACF SOPs)	EPA Maximum Allowable Holding Times
Ammonia	Plastic, 60 ml	Filtered and iced in the field, Frozen upon return from field	48 hours after thawing, 28 days after collection SOP-004	28 days EPA350.1
Nitrite	Plastic, 60 ml	Filtered and iced in the field, Frozen upon return from field	48 hours after thawing, 28 days after collection SOP-004	28 days EPA353.4
Nitrate + Nitrite	Plastic, 60 ml	Filtered and iced in the field, Frozen upon return from field	48 hours after thawing, 28 days after collection SOP-004	28 days EPA353.4
Soluble Reactive Phosphate	Plastic, 60 ml	Filtered and iced in the field, Frozen upon return from field	48 hours after thawing, 28 days after collection SOP-004	28 days EPA365.1
Total Nitrogen	Plastic, 120 ml	Iced in field, refrigerated	Add HCl as preservative within 48 hours of sample submission, 28 days SOP-006	28 days ASTMD5176
Total Phosphorus	Plastic, 120 ml	Iced in field, refrigerated	Acid digest the samples within 48 hours of sample submission, 28 days SOP-008	28 days EPA365.1
Total Organic Carbon,	Plastic, 120 ml	Iced in field, refrigerated	28 days SOP-007	28 days NU-062-1.8
Turbidity	Plastic, 120 ml	Read in the Field within 15 min of collection	Within 15 minutes of collection SOP-FT1600	48 hours EPA 180.1
Chlorophyll-a	GF/F Glass Fiber Filter, 1.8 HDPE Micro-centrifuge tube	10% Acetone, Frozen at -15˚C, Stored in the dark	28 days* SOP-003 * The holding time in SOP-003 is 21 days. Data will not be flagged if analyzed between 21-28 days.	28 days EPA 445.0

2. Sample Custody

Sample custody is the responsibility of the sampling team and of the Chief Chemist. The sampling team is responsible for labelling, collecting, documenting in the field notebook, and transporting samples to the laboratory. The Chief Chemist is responsible for proper log-in and storage of the samples within the laboratory, and that the samples are analyzed within their appropriate holding times. Samples are not discarded until the analytical results are checked and approved by the Quality Assurance Officer. All documentation/logs are signed/initialed by appropriate personnel.

The SERC CAChE-NACF Laboratory is controlled by a Laboratory Information Management System (LIMS). Prior to leaving for the field, each sample container is labelled with a unique sample identification LIMS bar coded number, and colored tapes to differentiate surface and bottom samples.

3. Field Custody

Field Data Sheets are used to record field information during sampling (Figure 1). Once QA checked, the sheets are kept in project-specific files. Entries into the field notebook include: name and number of sampling trip; date of sampling trip; general weather and water conditions (waves and tides); name of individuals in sampling team; location and number of sample; and sample collection start time and end times. Additional recorded information includes, turbidity readings (surface and bottom), surface salinity and the volume of water filtered. Each sheet is signed by the sampling team. If an error is made in the field notebook, corrections are made by drawing a line through the error and entering the correct information next to the error.

Fig. 1. Field Data Sheet

FIELD DATA SHEET (V.5) 15-row (revised 2020-07-01)

Weather Time: ____:____ Temp. ____° C / ____° F, humidity _____ %, cloud cover _____ %, wind spd _____ units _____ & direction _____ , sea height _____ & direction _____							Vessel used: _____ Depth calibrated at sea level (daily) _____	
Sampling Event		**Date of Collection**	**Names**				**Equipment Used/ Serial#**	**Equipment Calibration**
			sampled by:				CTD or YSI Serial #	Date:
			read by:				CTD or YSI Sonde #	hour: attach equipment calibration sheet
Station Number	**Equip. Rinsed**	**Start Time**	**Salinity (ppt)**	**Turbidity Surface (NTU)**	**Turbidity bottom (NTU)**	**Vol. filtered (ml)**	**End Time**	COMMENTS
		: :					: :	
		: :					: :	
		: :					: :	
		: :					: :	
		: :					: :	
		: :					: :	
		: :					: :	
		: :					: :	

4. Laboratory Custody

Legal chain-of-custody (COC) procedures are followed for samples collected as part of this project. Upon transport of the samples to the CAChE-NACF laboratory at FIU, the field sampling personnel log-in the samples on the COC form (Figure 2). All the sample bottles are inspected for integrity, proper documentation (labelling), and preservation (bagged in cooler with wet ice). Any samples bottles found broken, leaking, not properly marked, or not properly preserved are rejected from analysis, and noted on the bottom of the Sample Checklist.

Fig. 2. COC sheet

CAChE Nutrient Analysis Laboratory Core
OE 148 (Office)/VH-303 (Lab), University Park, Miami, FL 33199
305-348-3095 or 305-348-2836

MATRIX			
FW	Freshwater	S	Soil
SW	Saltwater	O	Other

SDG# **200722**

PROJECT NAME:	K100	**LIMS RECORD:**	AC84360-AC84565	**ACCOUNT NO.:**	800007547
DELIVERED BY:	Sandro Stumpf	**RECEIVED BY:**		**DATE AND TIME:**	

ITEM #	BOTTLE ID	SAMPLE UNIQUE ID	MATRIX	COLLECTION DATE and Time		# OF REPLIC.	ANALYSES REQUIRED* FILT. NUTR.	TP: (W)ater (S)oil	SI	TN	(T)OC (D)OC	CHL-a	TN/TC
1	AC84360	K216BIF	SW	7/7/2020			X	W	X	X	T		
2	AC84361	K216SIF	SW	7/7/2020	10:41		X	W	X	X	T	X	
3	AC84364	K218BIF	SW	7/7/2020			X	W	X	X	T		
4	AC84365	K218SIF	SW	7/7/2020	11:23		X	W	X	X	T	X	
5	AC84366	K219BIF	SW	7/7/2020			X	W	X	X	T		
6	AC84367	K219SIF	SW	7/7/2020	11:46		X	W	X	X	T	X	
7	AC84370	K222BIF	SW	7/7/2020	:		X	W	X	X	T		
8	AC84371	K222SIF	SW	7/7/2020	12:24		X	W	X	X	T	X	
9	AC84374	K224BIF	SW	7/7/2020	:	1	X	W	X	X	T		
10	AC84375	K224SIF	SW	7/7/2020	13:01	1	X	W	X	X	T	X	
11	AC84376	K225BIF	SW	7/7/2020	:		X	W	X	X	T		
12	AC84377	K225SIF	SW	7/7/2020	12:47		X	W	X	X	T	X	
13	AC84382	K228BIF	SW	7/7/2020	:		X	W	X	X	T		
14	AC84383	K228SIF	SW	7/7/2020	13:33		X	W	X	X	T	X	
15	AC84434	K264BIF	SW	7/7/2020	:		X	W	X	X	T		