Manitoba Great Lakes (MBGL) Sampling Protocols

Centre for Earth Observation Science

Document Control

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1.1	C. Herbert	Working Copy	2018 May 15	Versioned according to Alfresco
1.2	C. Herbert	Working Copy	2018 June 15	Modified methods according to D. Capelle and added more detail to document
1.3	C. Herbert	Working Copy	2019 October 10	Modified sample processing schema and cleaning methods
1.4	K. Rodgers	Working Copy	2021 July 27	Minor edits, removed a few of Tim's lab methods and added aP and CDOM lab methods, added Appendix for Detection Limits

Document Location

A hardcopy of the document can be found in the Lab 489 document cupboard.
The digital copy can be found at:

https://cwincloud.cc.umanitoba.ca/data_mgmt/latextemplates/-/blob/master/MBGL_Protocols/MBGL_FW_protocols.pdf

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Herbert, C., Kamula, M., Rodgers, K. (2021). CanWIN Freshwater Sampling Protocols, Version 1.4. Center for Earth Observation Science. University of Manitoba.

Other Supporting Documents

Papakyriakou, Soloway and Capelle 2021. Keeyask Water Sample Protocols. pp. 10.

The digital copy can be found at:

https://cwincloud.cc.umanitoba.ca/MBGL/lab/-/blob/master/Protocols/Keeyask%20Water%20Sample%20Protocols-2021_7_15.docx

Contact Ashley Soloway at ashleydawn.soloway@umanitoba.ca for more information on protocols.

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1. Sample Collection Gear

Below are listed recommendations concerning samplers, sample containers and special sampling techniques and precautions. Each method listed in this protocol document may also have its own special sample container, technique and precautions.

Generally, 500 mL of water is required for complete analysis.

1.1 Sample Techniques and Equipment

1.1.1 Samplers

A 2 L van Dorn or Kemmer type sampler constructed of clear plastic is an excellent general purpose instrument. The sampler is lowered into the water column to the depth required. A metal trigger is then released, which closes the two open ends of the sampler. As the sampler is transparent, the sampler permits viewing samplers prior to transfer to sample bottles, which is essential when sampling near the sediment-water interface. Transparent tubing attached to the sampler and closed with a clamp allows users to release controlled amounts of water into the sample container.

The standard sample bottle used for MBGL sample collection is the 500 mL clear HDPE bottle-either provided by the Freshwater Institute or from the CEOS MBGL lab.

Samples can also be collected directly from a clean tap or spigot. Clean tubing with a clamp is attached to the spigot to enable sample collection directly from the tap.

Samples taken from a bridge are taken using a stainless steel sample bottle holder that holds the 500 mL botttles. A sample line is attached to the holder and the user ties off the line to the bridge before lowering sampler into the water. A small piece of plastic tygon tubing holds the sample bottle in place in the sampler.

1.1.2 Sample Bottles

500 mL to 2 L clear or brown high density polyethylene (HDPE) screw-cap bottles are used to collect water samples in the field (without the use of a sampler) or to transfer a large volume of sampler water to a second container.

1.1.2.1 Cleaning

Before use, sample bottles should have arrived from the University of Manitoba (UM) pre-cleaned. Any bottles used for sample collection should be cleaned to the following standard:

1. Polyethylene sample bottles are scrubbed once with hot water
2. Bottles are placed in 10% - 20% hydrochloric acid for a minimum of 24 hours
3. Bottles are then removed and rinsed three times (3) with distilled water
4. Bottles are filled with distilled water, capped and left to soak for 24 hours
5. After 24 hours, dump water from bottles and check for contamination (ex. visible particles, discolouration of bottles).
6. If clean, bottles are set to dry in drying rack, and **within 48 h** are put in the appropriate storage location

1.1.2.2 Glassware Cleaning

1. Glassware are placed in 10% - 20% hydrochloric acid for a minimum of 24 hours

2. Glassware are then removed and rinsed three times (3) with distilled water

3. Set glassware to dry in rack and **within 48 h** are put in the appropriate storage location

1.1.3 Sample Techniques

When collecting a sample directly from a water body or *in situ* sample line, rinse sample bottle three (3) times before filling with the sample. Leave $\frac{1}{4}$ inch headspace to allow for shaking of the sample. If collecting the sample in the field, rinse water should be dumped **downstream** of where the final sample is taken.

1.2 Special Precautions

1.2.1 All Samples

Water must not freeze at any time during or after sample collection except where stated in method.

1.2.2 Nutrients

When sampling for nutrients, contamination is a concern. Cleaned sample bottles must be kept closed and stored in a relatively dust free environment until used. Care must be taken to not place hands or any other contaminants on or near the mouth of the sample bottle or lid. Examples of contaminants include perspiration from field personnel, smoking or plant pollen.

1.2.3 Algal Toxins

Samples for algal toxins are taken using two methods.

Method 1 Label whirlpak bag with date, station id, whole water and time. Subsample from Nutrient bottle. Shake bottle vigorously, pour 250-500 mL into whirlpak and recap Nutrient bottle. Fold over top of Whirlpak bag 2 – 3 times, then, holding both bag end twist ties, whirl bag around to seal (with air). Tie twist ties together and freeze bag.

Method 2 Take a second sample using zooplankton net and same method as for zooplankton. Label Whirlpak bag with Date, station id, zooplankton net depth and number of rinses. Seal Whirlpak bag using same method as Method 1. Freeze sample.

1.2.4 Decontaminating Gear for Aquatic Invasive Species (AIS)

The full Aquatic Invasive Species Regulation under The Water Protection Act(C.C.S.M. c. W65) document can be found here.

The method we suggest to have the least damaging and most efficient to manage the equipment is the bleach method.

1. Obtain Bleach 5.25%(household sodium hypochloride)

2. Create dilution with a ratio of 100 ml bleach to 1 L of water
3. Completely submerge item for 30 minutes
4. Rinse thoroughly with potable water and wipe down with a completely dry cloth

Other methods for decontaminating gear, refer to Aquatic Invasive Species Regulations, pg. 25.

2. Guidelines for Sample Handling and Processing

Samples should be processed within 4 hours after collection if possible. Prior to processing, sample container should be stored in a cool, dark place around 4 °C (ex. a fridge). Quick processing time permits analytical distinction between dissolved and particulate phases of the sample and minimises changes in sample composition arising from biological uptake and excretion (?, ?).

Figure 2.1 illustrates a sampling schema which divides the sample collection and distribution within this project.

Up to five particulate fractions may be obtained from the sample, depending on the project. Particulate fractions may include chlorophyll, particulate suspended solids (TSS/LOI/Tripton), particulate phosphorous, particulate carbon and nitrogen and aP. The filtrate is used for cation analysis, dissolved nutrient elements, major anions and dissolved solids.

Dissolved and particulate phases are defined as what either passes through or is retained on a pre-ignited (16 hours at 500 °C) Whatman 42.5 cm GF/C glass fibre filter with 1.2 µm particle retention size. For certain sample analysis (e.g. CDOM), samples may be filtered through a GF/F glass fibre filter, which has a particle retention size of 0.7 µm.

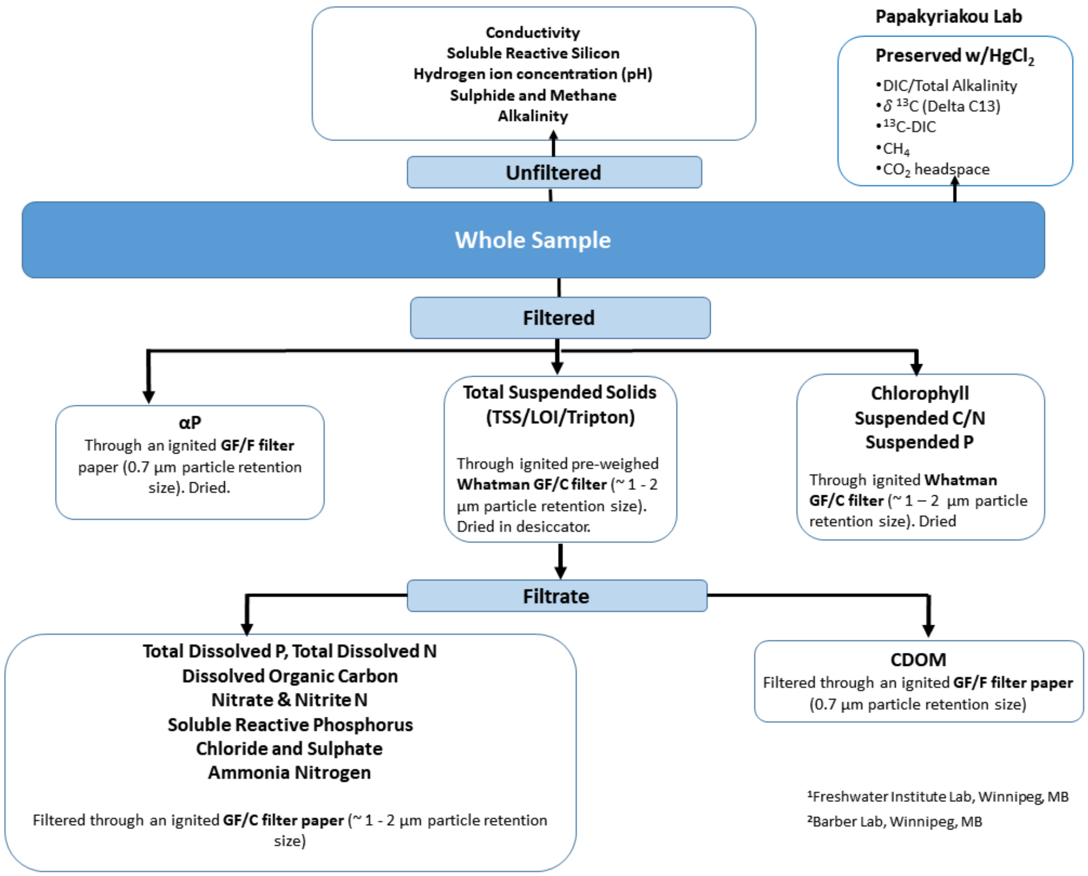

Figure 2.1: Sample processing schema

3. Sample Collection

Whole water may be collected at weekly, bi-weekly or monthly intervals. Samples will be processed according to Table 3.1 and stored according to Table 4.1. The collection code (Table 3.2) identifies which bottle the sample can be taken from. The volume of sample needed for sample analysis is also included in Table 3.1 (not including rinse volume). Detailed protocols for sample processing are found starting in Sample Processing and Storage. The table can be modified according to specific project needs.

Table 3.1: Sample Collection Table

Parameter	**Collection Code**	**Final volume collected** (mL)	**Pre-processing storage conditions** (°C)	**Pre-processing storage container**
		Papakyriakou Lab		
Dissolved inorganic carbon (DIC), Total Alkalinity (TA) in Papakyriakou lab, $_{18}$O	B	12	Cold (4 °C) and dark	B
Dissolved inorganic carbon 13 ($_{13}$C-DIC) Papakyriakou Lab	G	30	Cold (4 °C) and dark	G
Ch_4	H	60	Cold (4 °C) and dark	H

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Parameter	Collection Code	Final volume collected (mL)	Pre-processing storage conditions (°C)	Pre-processing storage container
		Barber Lab		
Algal Toxins	A	250 - 500	($-20\,^{\circ}$C)	F
Algae	A	20	Cold (4 °C) and dark	A
CDOM Barber Lab	A	10 - 100	Cold (4 °C) and dark	A
		Wang Lab		
Total and Methyl Mercury	E	750 (250 * 3)	Cold (4 °C) and dark	E
Dissolved Organic Carbon (DOC)	A	4	Cold (4 °C) and dark	A
		Ehns & Mundy Lab		
aP	A	500 - 2000	Cold (4 °C) and dark	A
CDOM	A	500 - 2000	Cold (4 °C) and dark	A
		Freshwater Institute		
Alkalinity	A	500 – 2000	Cold (4 °C) and dark	A
Conductivity	A	500 – 2000	Cold (4 °C) and dark	A
Dissolved organic carbon (DOC)	A	500 – 2000	Cold (4 °C) and dark	A
Major cations (Ca,Mg,K)	A	500 – 2000	Cold (4 °C) and dark	A

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Parameter	**Collection Code**	**Final volume collected** (mL)	**Pre-processing storage conditions** (°C)	**Pre-processing storage container**
pH	A	500 – 2000	Cold (4 °C) and dark	A
Salinity	A	500 – 2000	Cold (4 °C) and dark	A
Nutrient and Chlorophyll, Particulate and dissolved (carbon, nitrogen, phosphorous), suspended sediments (TSS, LOI, Tripton)	A	500 – 2000	Cold (4 °C) and dark	A

Table 3.2: Sample collection type and code

Collection Code	**Collection Type**
A	500 mL, 1 L or 2 L HDPE screw cap bottle
B	12 mL exetainer vial
C	GF/F Filtrate
D	1 L HDPE screw cap bottle
E	250 mL glass container
F	1 000 mL Whirlpak bag
G	30 mL amber glass bottle
H	60 mL glass serum bottle with rubber stopper and aluminum crimp seal

4. Sample Processing and Storage

Table 4.1 is a modified list from ? (?) and provides guidelines for the treatment and storage of samples. Unless otherwise stated in the specific sample protocol, all samples should be refrigerated (at approximately 4 °C) after processing. Table 4.2 lists the final storage container. Filter papers should be placed in their Petri dishes and frozen at either -20 °C or -80 °C. Unprocessed samples and filtrate will deteriorate at varying rates depending on the parameter (i.e. nutrients faster than major cations and anions), and the concentration (i.e. trace levels faster than major levels).

Appendix A lists the laboratories that will be analyzing the samples. Each sample description also states in which lab the analysis is conducted. Different labs may use varying methods of analysis for the same sample parameter, which may require different volumes of water or preservative. There are two main labs where samples taken for the freshwater sampling program at CEOS are currently shipped, either in-house within CEOS (F. Wang, T. Papakyriakou or D. Barber lab analysis), or at the Freshwater Institute, Department of Fisheries and Oceans Winnipeg, MB. Methods listed in this manual are specific to these labs and are not meant to be a definitive guide to freshwater sample analysis.

Table 4.1: Guidelines for the treatment and storage of samples.

Analysis	**Max. Storage duration prior to processing**	**Processing method**	**Final storage Vol.** (mL) **& container**	**Storage conditions after processing**	**Storage time after processing**
		Papakyriakou Lab			
Dissolved inorganic carbon (DIC), Total Alkalinity (TA) Papakyriakou method	24 hours	Whole water from A or sample equipment. Preserve with 20 µL $HgCl_2$	12 * 2 vials/parameter Cr	Ziploc bag in cupboard – Papakyriakou side Lab 486	
Dissolved inorganic carbon 13 ($_{13}$C-DIC)	24 hours	Whole water from A or sample equipment. Preserve with 1 mL $HgCl_2$	30, K	Ziploc bag in cupboard – Papakyriakou side Lab 486	
Ch_4		Whole water from A or sample equipment. Preserve with 0.1 mL $HgCl_2$		Ziploc bag in cupboard – Papakyriakou side Lab 486	
Total and Methyl Mercury	4 hours		250 (3), K	4°C, dark	
Oxygen 18 (^{18}O)	4 hours	Fill to overflow 3x, then fill to overflow and cap. Seal with parafilm	12 * 2 vials	Ziploc bag in cupboard – Papakyriakou side Lab 486	

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Analysis	Max. Storage duration prior to processing	Processing method	Final storage Vol. (mL) & container	Storage conditions after processing	Storage time after processing
		Barber Lab			
Algal Toxins	24-72 hours	Whole water from sample location	F	Frozen, then freeze-dried	Indefinitely
Algae	4 hours	Whole water from A (Table 3) with Lugols Iodine added	20, B	Room temperature – 466 or 566 Lab	Indefinitely
CDOM Barber Lab	24 hours	Whole water filtered through either 0.2 µm filter OR 42.5 mm GF/F filter	Varies, G or J	-20 °C to -80 °C	Indefinitely
		Wang Lab			
Dissolved Organic Carbon (DOC) Wang Lab	7 days	Whole water filtered from A. Add 50 µL 2 M HCl	4 – 10, L	4 °C	Indefinitely
		Freshwater Institute			
Alkalinity	7 days	Whole water from A – no processing	varies, A or N	4 °C, dark	indefinitely
Conductivity	7 days	Whole water from A – no processing	varies, A or N	4 °C, dark	indefinitely

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Analysis	**Max. Storage duration prior to processing**	**Processing method**	**Final storage Vol.** (mL) **& container**	**Storage conditions after processing**	**Storage time after processing**
Dissolved organic carbon (DOC)		F	175, D	4 °C	
Major cations (Ca,Mg,K)	7 days	Whole water filtered from A	175, D	4 °C	
Particulate Nutrients and Chlorophyll, Suspended carbon/nitrogen and phosphorous	7 days	Whole water filtered from A onto GF/C (H)	Varies, H	-20 °C	Indefinitely
Particulate solids (TSS/LOI/Tripton)	7 days	Whole water filtered from A onto GF/C (I)	Varies, I	-20 °C	Indefinitely
Dissolved Nutrients: Nitrogen and phosphorous	7 days	Whole water filtered from A (Table 3) using a GF/C (H)	175, D	4 °C, dark	72 hours
pH	7 days	Whole water from A (Table 3) – No processing	varies, A or N	4 °C, dark	1 week
Salinity	7 days	Whole water from A (Table 3) – No processing	varies, A or N	4 °C, dark	1 week

Table 4.2: Sample processing and storage container codes.

Collection Code	Collection Type
A	0.5 - 2 L HDPE screw cap bottle from Table 3
B	20 mL glass scintillation vial
C	12 mL vial from Table 3
D	175 mL screw cap HDPE bottle
E	GF/C filtrate from A, Table 3
F	GF/C filtrate from A, Table 3
G	Pre-ignited GF/F 42.5 mm filter paper
H	Pre-ignited GF/C 42.5 mm filter paper
I	Pre-ignited and Pre-weighed GF/C 42.5 mm filter paper
J	GF/F Swinex filter paper
K	30 mL amber glass bottle
L	5-10 mL glass vials with Teflon lids
M	60 mL glass serum bottle with rubber stopper and aluminum crimp seal
N	60 mL screw cap HDPE bottle

5. Sampling and Laboratory Methods

5.1 Filtering

Version 1.2

25 April 2019

Reference: C. Herbert, 2019

5.1.1 Method Principle

1. Samples preserved in the dark at 4 °C can be kept for up to 7 days before processing for dissolved and particulate nutrients.

5.1.2 Precautions

1. Before use, bottles should have been cleaned at the University of Manitoba (UM), CEOS lab as per Section 1 Sample Collection Gear, of this manual, or bottles should have arrived from the Freshwater Institute (FWI), Department of Fisheries and Oceans, Winnipeg, pre-cleaned.

5.1.3 Sample Considerations

1. Keep sample bottle in the fridge at 4 °C until filtration.

5.1.4 Lab for Analysis

1. Samples for nutrients and chlorophyll analysis are sent to the FWI for analysis. Total suspended solid (TSS) samples are analyzed in the CEOS Barber lab. Separate samples for dissolved organic carbon (DOC) are analyzed in the Wang lab at CEOS. A complete list of sample types and labs for analysis can be found in Table 4.1

5.1.5 Equipment/Apparatus

1. Two (2)-empty petri dishes for Suspended Carbon/Nitrogen (Susp C/N) blanks **NOTE: If filtering in Lab 566 (CEOS), you only need to process one (1) blank filter paper for Susp C/N (i.e. you do not need to filter distilled water through)**
2. One (1)-empty petri dish for Suspended Phosphorous (SuspP) blank
3. Filter unit (either glass or magnetic unit)
4. Tweezers
5. Distilled water (DROW) bottle
6. Filtration stand and clamps to hold glass flask and sample bottle **in field**
7. Ground glass 500 mL bottle to filter sample into
8. 1 L glass overflow flask and associated tubing/stopper
9. Vacuum line (lab), electric air pump (Namao) or hand pump (Waterhen)

5.1.5.1 For each sample

i. One (1) empty Petri dish

ii. One (1) 175 mL anion bottle

iii. One (1) pre-combusted GF/C filter for Susp C/N

iv. One (1) pre-combusted GF/C filter for SuspP

v. One (1) pre-combusted GF/C filter for Chlorophyll-a

vi. One (1) pre-combusted, pre-weighed GF/C filter for TSS

vii. One (1) pre-combusted, GF/F filter for αP

viii. One (1) 4 mL vial for DOC (Wang lab), cleaned with 10% - 20% HCL and combusted at 500 °C, lids dried at 65 °C

ix. One (1) 60 mL amber glass vial for CDOM (Ehns lab), cleaned with 10% - 20% HCL and combusted at 500 °C, lids dried at 65 °C

x. One (1) 20 mL glass scintillation vial w/ Lugols preservative for algae

5.1.5.2 For each sample day (blanks)

i. Three (3) empty Petri dishes

ii. One (1) pre-combusted filter GF/C for blank Susp C/N for 100 mL Distilled water filtration

iii. One (1) pre-combusted GF/C filter paper for Susp C/N – No water filtered onto it

iv. One (1) pre-combusted GF/C filter paper for SuspP – No water filtered onto it

5.1.5.3 Check Samples

i. Every **10** samples make a check sample which is a duplicate of the whole water sample you are processing

ii. One (1) blank Petri dish

iii. One (1) pre-combusted GF/C filter paper for Susp C/N

5.1.6 Method

1. At end of day fill out CEOS Lab log and FWI Chemistry sample ID sheets and CEOS filter log book
   (a) There will be one FWI chem sheet for each blank type (ex. SuspC/N, SuspP) and one for all samples of same analysis parameters
2. Label petri dishes and bottles
   (a) At the beginning of each filter day, one (1) blank sample will be filtered using pre-combusted filter papers for suspended C/N. Label petri dish and filter 100 mL distilled water onto the filter paper. **NOTE: If filtering in Lab 566 skip this step**
   (b) At the beginning of each filter day, one (1) blank pre-combusted filter paper for suspended C/N will be removed from the filter paper container and placed in a clean petri dish. Label with 0 volume.
   (c) At the beginning of each filter day, one (1) blank pre-combusted filter paper for suspended P will be removed from the filter paper container and placed in a clean petri dish. Label with 0 volume.

(d) For each sample, there will be one (1) pre-combusted GF/C filter paper for each of your particulate parameters. They can include all or some of: suspended C/N, suspended P and chlorophyll.

(e) For each sample, there will be one (1) pre-combusted,pre-weighed GF/C filter paper for total suspended solids (TSS).

(f) For each sample, there will be one (1) pre-combusted GF/F filter paper for αP.

(g) For each sample there is one (1) 175 mL pre-cleaned anion bottle

(h) For each sample there is one (1) 4 mL glass vial acid washed and pre-combusted for DOC.

(i) For each sample there is one (1) 60 mL amber glass vial acid washed and pre-combusted for CDOM

3. All Petri dishes are labelled with sample ID and type (either C/N or Chl,), the date, and after the sample is filtered onto the paper, the volume.
4. Anion and DOC bottles and vials are labelled with the sample ID, Type and date.
5. Algae scintillation vials are labelled with Sample ID, type, date, time, whole or net sample. Lids are also labelled with Sample ID, date and depth (0 m)
6. Place aluminum foil onto surface used to process samples. Wipe foil with ethanol (95-99%).
7. Rinse forceps with DI water

5.1.6.1 Filtering Blanks

At the beginning of each sample day, blank filters must be processed. To do this:

1. Ensure the filter unit is attached to the 1 L flask.
2. Place one GF/C filter paper onto the bottom of the magnetic filter unit, **grid side down**, then replace the cup top.
3. Filter the blank samples first. For each parameter, pour 100 mL of distilled water onto the filter paper and place in appropriate petri dish (ex. Susp C/N).

4. Remove one filter paper and place in clean petri dish Label as blank, 0 mL, Susp C/N, and another for Susp P

5.1.6.2 Filtering Samples

Between each different sample station or sample depth, filter unit must be rinsed. To do this, rinse empty filter unit and attached filtration flask with 50 mL of distilled water. Detach filter unit (ensure you do not touch the inside of the unit or the filter top) and swirl water in flask. Empty flask and reattach unit. Then shake sample and pour a small amount (10 - 50 mL depending on amount of sample you have) into filter unit, ensuring you cover sides of unit. Again detach filter funnel, swirl liquid in flask and empty out.

To filter a sample:

1. Remove top of filter unit
2. Place suspended **C/N**, GF/C filter paper onto filter unit
3. Replace cup, shake sample vigorously and pour 100 mL into graduated cylinder. Turn on vacuum unit, or begin hand pumping. Pour sample onto filter paper. **Turn off vacuum**.
   - **NOTE:** The time between shaking sample, pouring into the graduated cylinder and pouring into filtration unit should be **AS SMALL AS POSSIBLE** to minimize particulate settling
4. Remove top of filter funnel unit. **Place upside down on cleaned surface, ensuring you do not touch the inside of the unit.** Remove filter paper, place filter into Suspended C/N Petri dish.
   - **NOTE:** After filtering the first sample, you will remove the bottom of the filter unit, place on clean surface ensuring filter surface does not touch anything, and swirl and empty filter flask. This ensures filtrate for analysis is not contaminated by previous sample.
5. Place top of filter unit back into filter flask. Repeat Steps 2 - 4 to filter for Susp P, Chlorophyll, and TSS. Must used a pre-weighed GF/C filter paper
6. YOU MUST NOW USE THE POLYETHYLENE GLOVES.
7. Remove paper, place filter into corresponding petri dish

8. Remove filter base and mix filtrate thoroughly in flask, rinse anion bottle 3 times and pour sample into anion bottle. Cap bottle.
9. Rinse DOC vial three times then fill vial, replace cap until time to add 50 µL HCL.
10. You can now remove the polyethylene gloves.
11. **If filtering for αP or CDOM:** place **GF/F** filter onto magentic unit and filter as per the previous samples (e.g. SuspP, SuspC/N)
    - Place αP filter into petri dish
    - Remove filter base and mix filtrate thoroughly in flask, rinse CDOM bottle 3 times and pour sample into bottle. Cap.

5.1.6.3 Preserving DOC Samples

1. Put back on the Polyethylene gloves
2. Add 50 µL 2 M HCl to vial
3. Recap sample, seal with parafilm and store in labelled Ziploc bag in fridge.

5.1.6.4 Cleanup

1. Ensure all samples are labelled with the Station ID, Sample type, date and volume.
2. Place all Petri dish lids under Petri dishes and place all Petri dishes into desiccator
3. When dry, replace lids on Petri dishes and group according to sample type (C/N etc.) and in date order. Tape together. Place each sample type into a Ziploc bag, and place all bags into one larger bag. Label with Project Name, PI Names, To/From dates (e.g. LWpg, Herbert/Pap, May 17, 2016 – June 1, 2016) and place in freezer until shipment to CEOS or the FWI
4. Rinse used sample bottles and glassware once with hot water and scrub. Then place in a 10% - 20 % hydrochloric acid tub and leave to soak for a minimum of 24 hours. If HCl is not available, rinse with distilled or deionized water three times.

5. Replace all sample bottles in fridge. Whole water must also be shipped to FWI for **alkalinity, pH and conductivity** analysis.
6. After glassware and bottles have finished soaking in HCL tub, remove using rubber gloves, rinse 3 times with hot water, scrubbing after first rinse. Rinse 3 times with distilled water. Re-fill bottles with distilled water and cap. Leave overnight to soak. After 24 hours, empty bottles, check for any particulates, and, if clean, place upside down to dry. When dry replace bottles in appropriate storage location.

5.2 Nutrient Sampling

Version 1.1

06 June 2018

Reference: (?, ?)

5.2.1 Method Principle

1. Samples preserved in the dark at 4 °C can be kept for up to 7 days before processing for dissolved and particulate nutrients.

5.2.2 Precautions

1. Before use, bottles should have been cleaned at the University of Manitoba (UM), CEOS lab as per Section 1 Sample Collection Gear, of this manual, or bottles should have arrived from the Freshwater Institute (FWI), Department of Fisheries and Oceans, Winnipeg, pre-cleaned.

5.2.3 Sample Considerations

1. Keep sample bottle in the fridge at 4 °C until shipping.

5.2.4 Lab for Analysis

1. Samples for nutrient analysis are processed in the CEOS Barber lab and sent to the FWI for analysis.

5.2.5 Equipment/Apparatus

1. 500 mL, 1 or 2 L clear or brown HDPE screw cap bottle

5.2.6 Method

1. When collecting a sample directly from a water body or in situ sample line, rinse sample bottle three times before overfilling with the sample. If collecting the sample in the field, rinse water should be dumped **downstream** of where the final sample is taken

2. **When sampling**, unroll sample line **over the side of the ship into the water**, and two meters longer than **sample** depth
3. Attach line to **railing of vessel** or seat leg in smaller vessels with loose knot.
4. If using VanDorn/Kemmerer with tubing on end, ensure tubing line is closed when lowering to collect sample.
5. Lower sampler into water to desired depth, send trigger down and pull sampler up slowly.
6. Rinse sample bottle three (3) times with sample water as per point 1.
7. Fill sample bottle to $\frac{1}{4}$ from top. If sampling by hand dip, insert bottle into water upside down then tilt upwards to fill bottle. Make sure you fill bottle underneath the surface of water.
8. Cap sample bottle and shake vigourously. **DO NOT** touch inside of cap or sample bottle rim with bare hands to avoid contamination.
9. Unscrew sample bottle lid and subsample for algae Section 5.3
10. Re-cap sample and place bottle in fridge at 4 °C until shipping or processing.

5.3 Preserved Algal Sample

Version 1.0

17 April 2017

5.3.1 Method Principle

1. Whole water samples can be sub-sampled and preserved with Acid Lugols Iodine for algal identification.

5.3.2 Precautions

1. Lugols Iodine is non-toxic but will stain clothing. Vials are pre-filled with the Iodine solution. Do not overfill vials.

5.3.3 Sample Considerations

1. Water must not freeze at any time during or after sample collection

5.3.4 Lab for Analysis

1. Samples for nutrient analysis are processed in the CEOS Barber lab and sent to the FWI for analysis.

5.3.5 Equipment/Apparatus

1. One (1) - 20 mL glass scintillation vial pre-filled with about 0.5 mL of Lugols preservative for algal sample collection.

5.3.6 Method

1. Label glass vial with the sample ID, sample type, date and time
2. Shake nutrient sample bottle vigorously. Fill glass vial $\frac{3}{4}$ full.
3. Cap vial and store in sample box in the dark.

5.4 Total Suspended Solids/Loss Of Ignition/Tripton

Version 1.0

06 June 2018

Reference: Freshwater Institute Analytical Unit, Method FWI-TSS, 2008

5.4.1 Method Principle

1. A homogenous sample is filtered through a pre-weighed Whatman GF/C filter. The residue retained on the filter is dried to a constant weight at 104±3 °C. The increase in the weight of the filter is assumed to be the total suspended solids.

5.4.2 Precautions

1. Be careful of hot oven.

5.4.3 Sample Considerations

1. Water must not freeze at any time during or after sample collection

5.4.4 Detection Limits

1. A detection limit of $2.5\,\mathrm{mg\,L^{-1}}$ has been established as the goal for the TSS test. This translates to 0.25 mg sediment collected from 100 mL water

5.4.5 Lab for Analysis

1. Samples are analyzed in Barber Lab, Room 486

5.4.6 Equipment/Apparatus

1. One (1) pre-weighed and pre-combusted GF/C filter paper
2. One (1) aluminum weigh tray stamped with a unique number

5.4.7 Method

5.4.7.1 Method Blanks

A filter (preweighed) is taken through the filtration procedure but distilled water is passed through the filter and then treated as a sample

5.4.7.2 Quality Control

1. Calibrated weights used to check balance calibration.
2. Use the same balance to weigh samples as was used to pre-weigh the filters.
3. Balances are calibrated to 17025:2005 standards.
4. Check weights are used to control balance reproducibility.
5. Every 10–20 samples (approximately) a method blank filtration and weighing is done to monitor contamination. Every 15 samples is a good target frequency.
6. Every 10–20 samples (approximately) a **duplicate weighing**, both tare and gross weight, are done to monitor reproducibility. Every 15 samples is a good target frequency. Duplicate weighings may not differ by more than 0.03 mg.
7. Filters, tares, gross weights, method blanks and reference samples, are dried and weighed repeatedly to a reproducible weight, within 0.04 mg for filters $\leq$ 100 mg and 0.06 mg for filters >100 mg.

The drying and weighing procedures described here apply to both the **initial taring** of the filters and subsequent **weighing of filter with sample**.

5.4.7.3 Sample Blanks, Reference Samples and Duplicates

When taring filters one in every 18 filters, 15 sample filters and 3 quality control filters, is labeled as **Method Blank**, one is labeled as **Ref Sample** and one is labeled **Duplicate**. Record the information on the Tare side of the weighing sheet and label the Petri dishes appropriately. Each weighed filter is placed in a Petri dish which is labeled as above and numbered consecutively to identify the filter for future reference. Record this number on both the tare and gross sides of the weighing sheet to ensure that tare and gross weights are recorded on the same worksheet.

5.4.7.4 Drying the Filters

Caution. Be sure that in the manipulation of the tared (preweighed) filters none of the filter disc material is broken off and no sample material lost.

1. When drying the filters match the sample number on the aluminium foil weigh tray with the sample id in the sample id column on the sample worksheet and record the alpha-numeric sample number and the volume filtered in the appropriate spaces. Verify the data.
2. Dry in oven at 104±3 °C for approximately 16 hours or overnight (see Note 1 below).
3. Record oven temperature on sample sheet.
4. After drying place each filter, in order, in desiccator. Desiccate and cool filters for 20–30 minutes.

Note 1 To avoid sample loss, handle filters gently, with forceps only, and only by the edges. Do not leave samples in drying oven for more than 24 hours, because the filters can become brittle and may break apart with loss of sample.

5.4.7.5 Weighing the Filters

This section discusses in detail the weighing of the filters. The **Perkin-Elmer AD-6 and the Sartorius** balances are discussed in detail. These balances are readily available to the lab but this is not intended to limit use to these balances.

Balances that meet calibration requirements are suitable for use.

5.4.7.6 Recording Weighing Data

The filter weighing record consists of two parts, a tare weight, (Tare) TSS-07, and a gross weight, (Gross) TSS-07, or later versions. When determining the tare weights enter the filter number in the appropriate space. When done enter the filter numbers into the corresponding spaces on the gross weight form. This will ensure that the data for that filter remains on the same sheet.

5.4.7.7 Precautions

1. Be sure the balance is in a stable environment free of vibrations.

2. The relative humidity should be between 40% and 60%.
3. The sample filter discs should be in thermal equilibrium with the balance.

5.4.7.8 Using the Sartorius Model BP211D

1. Turn on the balance at least 45 minutes before using.
2. Be sure balance is level.
3. Use the tare to bring the balance reading to zero.
4. Check balance with the check weight. If not correct re-tare the balance and repeat. If the correct weight is not obtained do not proceed but determine the cause.
5. Place empty aluminum tray that has been in the oven with the samples onto weigh scale. Tare tray.
6. Check zero before each weighing.
7. Transfer filter from original tray to sample tray in weigh scale.
8. Weigh sample filters, record weights.

5.5 Dissolved Organic Carbon

Version 1.1

06 June 2018

5.5.1 Method Principle

1. Method as per Wang Lab, CEOS.

5.5.2 Precautions

1. Use Polyethylene gloves when handling sample.

5.5.3 Sample Considerations

1. Water must not freeze at any time during or after sample collection

5.5.4 Lab for Analysis

1. Wang lab at CEOS

5.5.5 Equipment/Apparatus

1. One (1) 4 mL pre-combusted (at 500 °C for 5 hours) glass vial
2. 2 M HCl
3. Eppendorf pipette (0 - 100 µL) and tips

5.5.6 Method

1. Process as per MBGL sample collection and processing method.
2. After all samples have been processed, line up the DOC vials.
3. **PUT ON POLYETHYLENE GLOVES**
4. Using ependorf pipette, place a new tip on the end. DO NOT touch the pipette tip end with bare hands.
5. Pipette 50 µL of 2 M HCl into pipette.

6. Inject into DOC vial.
7. Place all DOC vials into a Ziploc bag, label with appropriate survey name, date and researcher.

5.6 ^{13}C – DIC (Dissolved Inorganic Carbon)

Version 1.1

06 June 2018

5.6.1 Method Principle

1. Method as per Papakyriakou Lab, CEOS.

5.6.2 Precautions

1.

5.6.3 Sample Considerations

1. Water must not freeze at any time during or after sample collection
2. Avoid trapping air bubbles and ice crystals inside of sample bottles during sample collection

5.6.4 Lab for Analysis

1.

5.6.5 Equipment/Apparatus

1. 30 mL amber glass serum bottle with conical screw cap (Figure 5.1)
2. $HgCl_2$
3. parafilm
4. Eppendorf pipette (0 - 100 µL)

5.6.6 Method

1. If sampling from the sample line, input directly into the sample bottle. Otherwise, use flexible tubing to fill sample bottles directly from the sampler.
2. Pinch tubing to stop flow, then insert all the way to bottom of bottle.

3. Release pressure on the tubing slowly, so bottle fills without entraining air bubbles.
4. Once bottle is completely full, allow water to flow until the bottle has been 'flushed' with 3x its volume. Slowly remove the tubing without entraining air bubbles (pinch it right before the tubing is removed).
5. Screw on the cap (no air bubbles).
6. Store all water samples at 4 °C in the dark for sample processing within 24-48 hours.

5.6.7 Sample Processing

1. Prepare a workspace by laying down an absorbent pad on the bench to work on.
2. Put on nitrile gloves. Have paper towel/Kimwipes nearby.
3. Unscrew caps add 20 µL $HgCl_2$ solution to each bottle, using pipette or 1 mL syringe
4. Re-cap
5. Place preserved samples inside clean Ziploc bags in Lab 486 cupboard. Write the sample date, location, and 'PRESERVED with $HgCl_2$' clearly visible on the outside of the bag (e.g. Keeyask – June 1, 2018 – PRESERVED WITH $HgCl_2$

Figure 5.1: ^{13}C – DIC bottle.

5.7 aP - Absorption by Particulates & aNAP - Absorption by Non-Algal Particulates

Version 1.0

06 July 2021

5.7.1 Method Principle

1. Ehns Method

5.7.2 Precautions

1. Do not keep samples in direct light
2. Analysis must be performed in a room with lights off or dimmed to protect samples from light
3. Max number of sample analyzed at one time is four (4)
4. Turn on spectrometer a minimum of 30 minutes prior to analysis

5.7.3 Sample Considerations

1. Water must not freeze at any time during or after sample collection
2. Allow spectrophotometer to warm up for a mininum of 30 minutes prior to analysis
3. Filtered samples (filter paper) must be stored in freezer and in the dark
4. Remove up to four (4) samples and thaw to room temperature prior to analysis
5. Use data sheet to check off completed steps

5.7.4 Lab for Analysis

1. Ehns & Mundy Lab, CEOS

5.7.5 Equipment/Apparatus

1. One (1) pre-combusted GF/F filter paper
2. One to four (1 -4) filter units (either glass or magnetic)
3. Forceps
4. Ruler
5. Kimwipes
6. Aluminium Foil
7. Beaker or Graduated Cylinder (reads 10 & 20 mL)
8. Methanol bottle (typically 95%)
9. Ethanol bottle (for cleaning forceps)
10. Milli-Q water bottle
11. Lambda 650S UV/Vis spectrophotometer (Lab 562)
12. Green lamps for illumination (Lab 562)
13. USB stick/drive for data backup

5.7.6 Method

More detailed protocols can be found on GitLab (here).

aP

1. Setup filtration units and methanol extraction to vacuum system.
2. Create designated folder on spectrophotometer computer for results.
3. Open PerkinElmer UV WinLab program, sign-in, and select method **aP**. Fill in Sample ID table with all control, blank, and sample information.
4. Press 'Autozero' before running any samples.
5. Run a control with no filter (air-air) in the chamber (a-a). Examine output graph to make sure y-limits are between 1.0 and -1.0, and the curve appears flat at the y-intercept of zero (0).

6. Moisten a blank GF/F with Milli-Q water (3-4 drops, do not soak filter paper) and clip onto jaw mount of the integrating sphere chamber. Run blank. Note: A blank should be run after every **5-6 samples**.
7. Make sure sample is thaw and moisten (3-4 drops) with Milli-Q water or sample water from site.
8. Place sample filter on jaw mount in the integrating sphere chamber. After scanning filter, rotate filter 90°and scan again.
9. Measure diameter (D_f) of biomass on filter with ruler.

aNAP

1. Place filter back into filtration unit.
2. Filter 10 mL of methanol. Close value once it has drained.
3. Pour 20 mL of methanol into filter cup. Do not drain. Cover filter cup with aluminum foil.
4. After 10 minutes, drain methanol. Close valued once it has drained.
5. Soak filter with 20 mL of methanol again for another 10 minutes. Cover filter cup with aluminum foil.
6. Drain methanol and place filter paper in the spectrophotometer. Moisten filter paper if it appears dry.
7. Analyze filter paper.
8. Rotate filter paper 90°and analyze again.
9. Check designated folder for all analyses. If not automatically save, manually export data from program.
10. Copy data from designated folder onto a USB.

5.8 CDOM - Coloured Dissolved Organic Matter

Version 1.0

08 July 2021

5.8.1 Method Principle

1. Ehns Method

5.8.2 Precautions

1. Do not keep samples in direct light
2. Analysis must be performed in a room with lights off or dimmed to protect samples from light
3. Turn on spectrometer a minimum of 30 minutes prior to analysis

5.8.3 Sample Considerations

1. Water must not freeze at any time during or after sample collection
2. Allow spectrophotometer to warm up for a mininum of 30 minutes prior to analysis
3. Remove samples from fridge and warm to room temperature (45 minutes), do not keep samples in direct light
4. Wear nitrile gloves when performing analysis
5. Cleaning cuvette between samples, rinse three times (3x) with a small amount of sample. For cleaning cuvette between sample and blank, rinse three times (3x) with Milli-Q water.
6. Export data after each run to ensure all files are backed up

5.8.4 Lab for Analysis

1. Ehns & Mundy Lab, CEOS

5.8.5 Equipment/Apparatus

1. 10 cm quartz cuvette
2. Methanol bottle (typically 95%)
3. Milli-Q water bottle
4. 10% HCL and small container for rinsing
5. Nitrile gloves
6. Paper towel
7. Forceps
8. Kimwipes
9. Lambda 650S UV/Vis spectrophotometer (Lab 562)
10. Green lamps for illumination (Lab 562)
11. USB stick/drive for data backup

5.8.6 Method

More detailed protocols can be found on GitLab (here).

1. Clean cuvette by first rinsing in 10% HCL bath, then rinse with methanol. After, use with Milli-Q water to rinse the outside and methanol again to rinse the inside of the cuvette. Thoroughly dry cuvette with Kimwipe.
2. Create designated folder on spectrophotometer computer for results.
3. Open PerkinElmer UV Winlab program, sign-in, and select method **DCOM**. Setup Sample ID table with all control, blank, and sample information.
4. Run a control with no filter (air-air)in the chamber (a-a). Examine output graph to make sure y-limits are between 1.0 and -1.0, and the curve appears flat at the y-intercept of zero (0). Note: A control should be run after every **six (6) samples**.
5. Press 'Autozero' before running any samples.
6. Gently fill cuvette with Milli-Q water to 100 mm line. Put on lid and wipe dry with a kimwipe. Place filled cuvette in holder of the integrated sphere

chamber, close chamber lid and run blank. Note: A blank should be run after every **three (3) samples**.

7. Empty cuvette and rinse three times (3x) with sample water. Fill cuvette with sample water to 100 mm line and dry with kimwipe. Place filled cuvette in holderof the integrated sphere chamber, close chamber lid and run blank.

8. Export data into designated folder then copy data to USB.

A. Analytical Laboratory List

Table A.1: Analytical Laboratory List.

Sample Parameter	**Lab for Analysis**
Preserved algae	CEOS, University of Manitoba
Algal Toxins	Environment and Climate Change Canada (Burlington)
Dissolved inorganic carbon/ Total Alkalinity	Papakyriakou
Dissolved inorganic carbon 13 (^{13}C-DIC)	Papakyriakou
Dissolved organic carbon	Wang Lab, CEOS
Dissolved organic carbon	Freshwater Institute, Department of Fisheries and Oceans
Major cations (Ca,Mg,K)	Freshwater Institute, Department of Fisheries and Oceans
CDOM	CEOS
Nutrients	Freshwater Institute, Department of Fisheries and Oceans
$Ch_4/N_2O/O_2/Ar$	Papakyriakou
Total and Methyl Mercury	Wang Lab, CEOS
Oxygen 18 (^{18}O)	Wang Lab, CEOS
Salinity	Freshwater Institute, Department of Fisheries and Oceans

B. Instructions to fill out Chemistry Forms for FWI

FRESHWATER INSTITUTE, WINNIPEG
ANALYTICAL CHEMISTRY UNIT, WATER CHEMISTRY LABORATORY

Submitted by____________________ Project Billing Code_______________ Date & time Received__________________ / ________

	WATER SAMPLE SOURCE DESCRIPTION					For Lab Use Only
	Date	Time	Location (4 digits max), subloc (3 max), station (6 max), depth (3 max) e.g. LWPG, SB, TRANS3, EPI	Pre proc. in field?	Chem Lab Field Bottle No.	Analyt. No.
1						
2						
3						
4						
5						
6						
7						
8						
9						
10						
11						
12						

Indicate requested analysses with an X -- please identify known contaminants or deviance from fresh water.

Request	Code	Analysis	Stored
	1, 2	Nitrate-N and Nitrite-N	
	3	Ammonia-N	
	4, 11	Suspended C and N	
	5	Total Dissolved N	
	6	Soluble Reactive P	
	7	Suspended P	
	8	Total Dissolved P	
	9	Dissolved Inorganic C	
	10	Dissolved Organic C	
	12	Chlorophyll - gross fluorescence	
	13	Chlorophylla - HPLC	
	14	Soluble Reactive Si	
	15,16	Chloride, Sulfate and Org acids	
	17	Total Suspended Solids	
	18	Total Suspended Iron	
	19	Conductivity	
	20	Sodium	
	21	Potassium	
	22	Calcium	
	23	Magnesium	
	24	Iron	
	25	Manganese	
	26	pH - equilibrated	
	27	Alkalinity	
	28		
		In Situ pH	
		In Situ Dissolved Inorganic C	
		Sulfide	
		Oxygen	
		Colour	

☐ Check if residual sample is to be returned, retained or directed elswhere when testing is done. Provide details in the comments section below.

If no instructions are provided samples will be disposed of when testing is done.

NB All samples are assumed to be in good order unless marked otherwise above, below or on obverse.

All samples submitted on this form but partially processed at an external site have been treated in accordance with the protocols required by the specific test methods used at the FWI lab. List exceptions in the comments section below.

Signed ________________________

Comments: Continue on obverse if required.

Figure B.1: Department of Fisheries and Oceans Freshwater Institute analysis request sheet.

C. Instructions to fill out CEOS Lab Form

CEOS, UManitoba Sample Intake & Analysis Form Version 1.0

CEOS, UNIVERSITY OF MANITOBA SAMPLE INTAKE & ANALYSIS FORM

Submitted by: ________________________ **Date & Time (24h) Received** _______________/___________________

	Sample Date	Sample Time	Sample ID	Location , Depth	Preproc. In Field	Bottle ID	Date sent to lab	Lab
1								
2								
3								
4								
5								
6								
7								
8								
9								
10								
11								
12								

Analysis	Lab	X for yes		Analysis	Lab	X for yes
Algal ID	Herbert/Kling			DOC	CEOS (Wang Lab)	
Algal Toxins	Environment Canada Burlington			Total & Methyl Mercury	CEOS (UCTEL)	
CDOM	CEOS			Nutrients/Chl/Ions	DFO (Freshwater Institute)	
DIC/Total Alkalinity	CEOS (Tim P.)			O_{18}	McGill Uni (Al Mucci)	
$\delta\,^{13}C$ (Delta C13)	McGill Uni (Al Mucci)			TSS/LOI/Tripton	CEOS (Barber Lab)	
^{13}C-DIC	CEOS (Tim P.)			CH_4	CEOS (Tim P.)	

Last edited June 14, 2018 by Claire Herbert

Figure C.1: CEOS Lab Form page 1 of 2.

CEOS, UManitoba Sample Intake & Analysis Form Version 1.0

REFERENCE KEY

Name	Definiton
Sample Date	Date sample was taken in field
Sample Time	Time sample was taken in field if known
Sample ID	Identification used for sample (**e.g**. Stn54)
Location Depth	Location of sample identifying water body type or area(ex. Lake Winnipeg, Red River, Keeyask Generating Station) Depth is depth in metres at which sample was taken
Preproc. In field	Was sampled processed in the field? (**i.e**. preserved from the original collection form). For example, if whole water was taken on Lake Winnipeg, and filtered in the field, the sample has been preprocessed before being sent back to the lab. If the sample must be processed or preserved for future analysis by the CEOS lab, sample was not preprocessed in field.
Bottle ID	Any bottle provided by the CEOS lab will have a permanent bottle identification number (**e.g.** 500-10)
Date sent to lab	Date sample was sent to lab for analysis. Sample could have multiple dates for multiple labs. Only relevant for samples sent outside of the CEOS (**e.g.** FWI).
Lab	Identifies to which lab the sample was sent. Only relevant for samples sent outside of the CEOS (**e.g.** FWI).

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Figure C.2: CEOS Lab Form page 2 of 2.

D. Short term laboratory storage and sample archive plan

Table D.1: Short-term Lab storage and sample archive plan.

Sample Type	**Short-term Storage Location (Lab No.)**	**Storage Duration**	**Long-term Archive Location (Lab No.)**
Text	Text	Text	Text
Text	Text	Text	Text
Text	Text	Text	Text
Text	Text	Text	Text
Text	Text	Text	Text
Text	Text	Text	Text
Text	Text	Text	Text

E. Variable Detection Limits

Table E.1: Variable Detection Limits.

Variable Name	Units	Detection Limit Value
NO3	μg/L	1.0
NO2	μg/L	0.5
NH4	μg/L	2.5
SuspN	μg/L	1.0
TDN	μg/L	1.5
SRP	μg/L	1.0
SuspP	μg/L	1.0
TDP	μg/L	1.0
DIC	μmole/L	10
DOC	μmole/L	10
SuspC	μg/L	7.0
Chla	ng	0.23
HPCL	ng	0.23
SRSI	μg/L	0.002
CL	mg/L	0.1
SO4	mg/L	0.2
TSS (FWI)	mg/L	0.2
TSS (MBGL)	mg/L	2.5
SusFe	mg/L	1.0
Cond	μS/cm @ 25°C	0.1

continued . . .

. . . continued

Variable Name	Units	Detection Limit Value
Na	mg/L	0.2
K	mg/L	0.2
Mg	mg/L	0.1
Ca	mg/L	0.2
Fe	mg/L	0.1
Mn	mg/L	0.1
pH	pH units	0.004
Alk	μeg/L	3.4