# Chlorophyll *a* Filtering Procedure

State of Wisconsin Department of Natural Resources Lake Monitoring Standard Operating Procedures June 2021



Wisconsin DNR, Water Quality Bureau Monitoring Program EGAD #3200-2021-06

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EGAD # 3200-2021-06

#### Chlorophyll a Filtering Procedure

#### Scope

This procedure describes the method for filtering surface water for determining the chlorophyll *a* concentration.

# Why Sample Chlorophyll a

Chlorophyll is the green molecule in plant cells that carries out the bulk of energy fixation in the process of photosynthesis. Chlorophyll a is a measure of the amount of algae growing in a waterbody.

## Safety Protocols

WDNR field method safety protocols must be used when sampling. Use a stable boat for sampling and wear a personal flotation device (PFD). A two-person crew is recommended. Ensure that the anchor rope does not become fouled in the propeller. Collecting samples in cold weather carries the risk of hypothermia and collecting samples in hot weather carries the risk of dehydration and heat stroke.

Prepare with appropriate clothing, blankets, sun protection, and drinking water. Carry a fire extinguisher, cellular phone or portable radio, and a first aid kit that includes materials for cleaning wounds (antibacterial soap and clean water or ethyl alcohol).

## Equipment

- Membrane filtration apparatus
- Millipore SM 5.0 µm membrane filters (SMWP 04700, 47 mm diameter, 100/pack). Millipore, Bedford, MA 01730, 800-646-5476
- Vacuum source (hand or electric pump)
- 1000 mL filtration flask
- 250 mL graduated cylinder
- Tubing

- Wash bottle
- Deionized water
- Filter forceps
- 15-mL plastic centrifuge tube from the Wisconsin State Laboratory of Hygiene (SLH) with label sticker
- Zip lock bag (3" by 5")
- Plastic gloves

# **Collection Procedures**

Collect the samples using the procedures given in the <u>Water Chemistry Sampling Procedures for Lakes</u>. Be sure to store collected sample on ice and out of direct sunlight.

# Sample Handling and Filtration

This procedure should be conducted under low light as soon as possible (same day preferred or within 48 hours of sample collection if shipping to the SLH for filtration). If indoors, dim the lights and close the blinds. If outdoors, work in the shade.

- 1. Place all equipment on your work area.
- 2. Insert the bottom part of the filtering cup (membrane filtration apparatus) into the 1000-mL filtration flask. Wet the stopper first to get a good seal.
- 3. Put on gloves.



- 4. Attach the plastic tubing of the vacuum pump to the spout of the filtration flask.
- 5. Pick up one small membrane filter with the filter forceps and place it on the center of the filter base (the black screen). Note that filters are white, and divider sheets are blue -- be careful to use a filter! Squirt a small amount of deionized (DI) water on the filter to keep it in place. Do not touch the filter with your fingers while removing it from the bag or when placing it on the screen.
- 6. Carefully place the cup on top of the filter base. It is magnetic. Be sure that the filter does not move!
- 7. Determine the approximate volume of water to filter for the chlorophyll analysis (refer to Table 1). Match the Secchi depth against the volume of water that should be filtered.
- 8. Gently mix the sample by turning the sealed bottle upside down several times. Before the sample settles, pour sample into the graduated cylinder, and record the volume. You will need to record the total volume of water filtered on the lab slip. The volume measured in the filtration cup is not accurate enough, so be sure to use a graduated cylinder.
- 9. Pour the water from the graduated cylinder into the filter apparatus. Rinse the graduated cylinder with DI water and pour into the filter apparatus to be sure no algae were left behind.
- 10. Squeeze the hand pump or turn on the vacuum pump to move the water through the filter. Once in a while, gently swirl the flask to ensure that algae do not stick to the side of the filter cup.
- 11. Rinse the filter cup walls with DI water.
- 12. When you are finished filtering, separate the top cup from the filter base.
- 13. Using the forceps, fold the filter paper in half so that the algae are on the inside. Do not touch the algae with your fingers or the forceps!
- 14. Fold the filter paper in half again and place the filter into the 15-mL centrifuge tube.
- 15. Fill out the chlorophyll sticker or label the tube with the sample information. Attach the sticker directly to the test tube. **INCLUDE THE VOLUME FILTERED!**
- 16. Place the test tube in the zip lock bag.
- 17. Write the volume of water filtered on the lab slip.
- 18. Freeze and mail frozen samples to SLH.

#### Table 1: Volume to filter based on Secchi depth<sup>1</sup>

Secchi Depth	Volume to Filter		
< 1ft	50 mL		
1-1.5 ft	100 mL		
>1.5 ft	200 mL		

<sup>1</sup>Note: These are approximate guidelines – filter more if needed but do not clog the filter.

## Documentation

Prior to sample, generate the Inorganic Surface Water and Microbiology lab slip (SLH form 4800-024) from SWIMS, which will include the items listed below. Verify that the labslip details are correct and record the **volume of sample filtered**. Verify and/or record the following data on your labslip:

- Person(s) sampling
- Lake name
- WBIC
- Date
- Time
- Sample Location/Description
- WAMS ID
- Project ID

- Station ID
- Weather conditions
- Water surface conditions
- Volume of sample filtered
- Water conditions affecting observations (i.e., algae, turbidity, etc.)
- User perception of water quality
- Visual estimate of water level

# **Disinfection and Decontamination**

Avoid or prevent invasive species contamination by following DNR field equipment disinfection protocols. Clean all equipment between lakes and at the end of the day to prevent the spread of invasive species. The boat, anchor, and any other equipment used in lake water must be cleaned using current DNR standard protocol disinfection procedures:

• <u>Boat, Gear and Equipment Decontamination and Disinfection Manual Code 9183.1</u> (https://dnr.wisconsin.gov/topic/Invasives/disinfection.html)

## References

WI DNR Field Procedures Manual Intranet Edition, Part B: Collection Procedures 1004 Chlorophyll Sample Filtration. Arneson, Ron. WDNR. 1989.

## Updates and Tracking

Version No.	Date	Title	Section	Author	Approval
	1989	WI DNR Field Procedures Manual Intranet Edition Part B: Collection Procedures 1004 Chlorophyll Sample Filtration		Ron Arneson	
1	2021	Chlorophyll a Filtering		Catherine Hein	Hein