

**Standard Operating Procedure for
Phytoplankton Sample
Collection and Preservation**

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1.0 Scope and Application

This Standard Operating Procedure describes the sampling and preservation of phytoplankton samples taken for the GLNPO open water Great Lakes surveys.

2.0 Summary of Method

Phytoplankton samples are created from a composite of water samples taken at discrete depths (surface, 5M, 10M, and 20M) with the rosette. Aliquots from each depth are combined, and approximately 1 L of the composite sample is preserved with Lugol's Solution for analysis at the CRL.

3.0 Safety and Waste Handling

Preservation of the phytoplankton samples with Lugol's solution must take place in a hood, and gloves and safety glasses should be worn.

4.0 Equipment and Supplies

960 mL plastic sample bottle
1 gallon cubitainer
Repipetter with 10 mL delivery capability
Distilled or super Q H₂O
Glacial Acetic Acid
I₂
KI
Hotplate
1L Flask
Opaque 1L container
Magnetic Stirring Bar
Glass funnel

5.0 Reagents

- 5.1 Lugol's Solution: Prepare at least one week prior to survey
 - 5.1.1 Using a Mettler balance or equivalent, measure 100 g KI and 50 g of I₂. Cover the I₂ reagent with tinfoil as it is light sensitive and will evaporate.
 - 5.1.2 Combine 900 mL Super Q H₂O and dry chemicals in a large flask. This should be performed in a fume hood.
 - 5.1.3 Add a magnetic stir bar and place on hotplate equipped with stirring action.

- 5.1.4 Warm slightly while stirring to facilitate dissolution of the dry chemicals. *Do Not Boil!*
- 5.1.5 In about an hour, once the solution is completely dissolved, pour into an opaque container using a glass funnel. Add 100 mL Glacial Acetic Acid to container and cap tightly. Invert several times to mix solution.
- 5.1.6 Label container with date, contents, and pH (usually around 2.4).

6.0 Sample Collection and Preservation

Note: Steps 6.1-6.4 are generally done by the ship contractor or EPA personnel. GLAS contract personnel will conduct this task when requested.

- 6.1 Remove 1 L of water from each of the Niskin bottles on the rosette from 20M, 10M, 5M and 1M, and add them to a 1 gallon cubitainer. This is the composite sample.
- 6.2 Mix the sample by gently turning the cubitainer over several times.
- 6.3 Pour approximately 1 L of the sample into the plastic sample bottle which has been labeled with station, sample number and survey.
- 6.4 In the Biology lab add Lugol's solution (5.1) to make the concentration 1%. If the sample nearly fills the entire sample container, add 10 mL of Lugol's solution to the sample. If less sample has been added to the container, adjust the volume of Lugol's solution that is added to achieve a 1% preservative concentration.
- 6.5 Samples must be stored in the dark and under refrigeration. Store the sample in the area designated by the sample coordinator.