

# CHAPTER 8

## PLANKTON

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### **Disclaimer**

Methodologies for assessing ALU and other regulatory bioassessments based on plankton have not been developed for Texas waters. Before conducting any biological monitoring activities using plankton, it is imperative to coordinate this work with the TCEQ and the TPWD. As methodologies and metrics become established, this manual will be updated to reflect those changes.

### **Non-Wadable Streams, Rivers, Lakes, Reservoirs, and Bays**

This chapter describes methods the TCEQ recommends for the collection and assessment of plankton assemblages. Plankton are free floating, mostly microscopic, plants, animals and bacteria. They generally cannot swim; instead, plankton are transported by tides and currents.

Plankton assemblages may include either phytoplankton (algae)—tiny single-celled plants—or zooplankton—free-floating animals. This chapter describes sampling methods for each assemblage.

The purpose of plankton sampling is to collect data that can be used to assess water quality trends and compare water quality between sites. Phytoplankton may also be sampled during a harmful algal bloom (HAB) such as one that resulted in a fish kill. Special handling may be required for red-tide, golden-alga, or cyanobacteria blooms. For HAB monitoring, contact the TPWD before collecting any samples. The TPWD—<[www.tpwd.state.tx.us/landwater/water/environmentconcerns/hab/](http://www.tpwd.state.tx.us/landwater/water/environmentconcerns/hab/)>—is responsible for investigating and researching the causes of HABs.

Plankton sampling is not part of routine monitoring and would only be done as part of a special study. Special studies require an approved QAPP or QAP prior to sampling. If a study is in progress, refer to the study's QAPP or QAP for details. If you are developing a QAPP or QAP for a special study, contact appropriate SWQM personnel for assistance.

### ***Phytoplankton-Collection Methods***

Phytoplankton can be collected either by a grab sample or with a plankton net. Surface grab samples are generally sufficient when sampling an algal bloom for presence of HABs. When the purpose of the sample collection is to document the entire phytoplankton community, collect integrated samples through the euphotic zone. For certain studies, phytoplankton net tows may be desired. When sampling phytoplankton with a net, its size and mesh size are important.

### **Collecting Samples from a Harmful Algal Bloom**

For algal-bloom samples (red tide, golden alga, cyanobacteria, or unknown blooms) surface grab samples are often sufficient when the sampler comes across a bloom unexpectedly in the field. If this happens, collect a surface sample (0.3 m depth) in a clean 500 mL container. Some algal

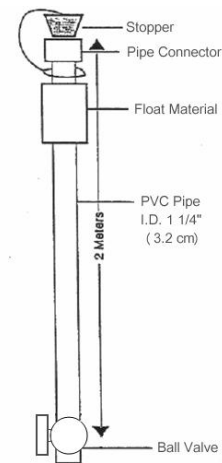
blooms may be harmful or contain skin irritants. Wear gloves to collect the sample and avoid skin and eye contact or ingestion. Contact the TWPD for instructions on preserving the sample, or refer to the section below on sample preservation.

## Collecting Grab Samples of Phytoplankton

Phytoplankton grab samples can be collected using methods developed by the EPA for the 2007 and 2012 National Lakes Assessment (U.S. EPA 2012). This method uses an integrated sampler based on a design by the Minnesota Pollution Control Agency; any similar device that allows a quantitative sample to be collected evenly through the water column of the euphotic zone should provide acceptable results.

The integrated water sampler is a PVC tube 2 m long with an inside diameter of 1.24 inches (3.2 cm) fitted with a stopper plug at the upper end and a valve that opens and closes the bottom end. The tube can be made in two sections with a screw coupler in the middle for easier storage and transportation. This device allows collection of a 1 L integrated grab sample from the upper two meters of the water column within the euphotic zone.

1. Estimate the depth of the euphotic zone by doubling the Secchi-disk depth at the site.
2. If the euphotic zone is greater than 2 m, sample the top 2 meters of the water body. If the euphotic zone is less than 2 meters, sample only to the depth of the euphotic zone—two times the Secchi-disk measurement. See Chapter 3 of Volume 1 (RG-415) for details on Secchi-disk measurements.
3. Rinse the sampler by opening both ends and submerging it in the lake three times.
4. With the valve open and the stopper off, slowly lower the sampler into the water as vertically as possible until the upper end is just below the surface or you have reached the depth of the euphotic zone if it is less than 2 meters.
5. Cap the sampler and slowly raise it to the surface, closing the valve before you pull it out of the water.
6. Empty the sample into a clean 4 L container by removing the stopper and slowly opening the valve. A large funnel, rinsed three times with ambient water, simplifies the sample transfer.
7. Repeat four times, each time dispensing the sample into the 4L container.
8. Once the four grab samples are composited into the 4L container, mix it thoroughly and pour 1 L into a clean container for phytoplankton processing.
9. Preserve samples as described in “Sample Preservation,” below.



**Figure 8.1.** Integrated water sampler.

The remainder of the composite sample can be used for accompanying chlorophyll *a*, nutrient, or other analyses if required by the study plan. If the euphotic zone is less than 2 m, more than four

grabs may be required, depending on the total volume needed for the phytoplankton sample and accompanying sample analyses.

## **Collecting Phytoplankton Samples with a Net**

Two types of net samples can be collected—either vertical or horizontal tows. Vertical tows can be made from a pier or other fixed location or from a stationary boat. Horizontal tows are made by slowly towing the plankton net from a moving boat or towing the length of a pier or bulkhead. Plankton net samples are not as quantitative as integrated or composite grab samples, but can be used to sample the phytoplankton community that is captured. Plankton nets are available in many different sizes and mesh sizes. Use one with a mesh size less than or equal to the size of the smallest phytoplankton cells expected in the sample.

1. Using a plankton net with a mesh size appropriate for the desired community, collect at least two net tows per station. In general, use a mesh size of 20 to 80  $\mu\text{m}$  for microplankton, and 2 to 20  $\mu\text{m}$  for nanoplankton.
2. Vertical tows:
  - Estimate the euphotic zone by calculating a depth of two times the Secchi-disk measurement.
  - Lower a clean plankton net with bucket attached to the lower end of the euphotic zone. In a shallow water body where the euphotic zone is equal to the depth, sample 0.5 m from the bottom to avoid disturbing bottom sediments.
  - Keeping the net line as vertical as possible, raise the net through the water column at about 0.3 m/sec. Proceed to step 4.
3. Horizontal tows:
  - Lower the net until it is completely submerged.
  - Let out a sufficient amount of line to allow the net to be towed beneath the surface.
  - Tow the net slowly for the desired distance. All samples in the study should be collected over the same distance. Once that distance is covered, pull the net slowly out of the water so that water flows out through the net mesh and not out the mouth of the net. Proceed to step 4.
4. Rinse the plankton on the net surface down into the bucket. Either:
  - hold the net upright and dunk it several times into the water, up to the mouth, or
  - splash water on the outside of the net and the plankton will be washed down to the bucket.
5. Disconnect the bucket and rinse it into a clean, waterproof container using a rinse bottle filled with ambient water that has been filtered through the plankton net.

### ***High Phytoplankton Abundance—Composite Grab Samples***

Some water bodies may be eutrophic and have very high phytoplankton abundance. If plankton are too abundant for the sample to drain freely through the net, but a net sample is preferred over

an integrated grab sample, it is acceptable to discard the net sample and collect a composite from water samples collected from specified depths with a Kemmerer or Van Dorn sampler.

1. Collect samples from the following 3 depths.
  - a. 0.3 m below the surface
  - b. midway between the surface and the lower end of the euphotic zone *or* midway between the surface and the bottom of the water body
  - c. the lower end of the euphotic zone *or* 0.5 m above the bottom

Additional samples may be collected as specified in the study plan, for example, every 3 meters in deeper water bodies.

2. Pour each water sample through the plankton net into the attached bucket.
3. Rinse the outside of the net with ambient water to wash the organisms down into the attached bucket.
4. Rinse the bucket into a clean container using a rinse bottle filled with ambient water that has been filtered through the plankton net.
5. Disconnect the bucket and rinse the sample into a clean waterproof container using a rinse bottle filled with the prefiltered ambient water.

## Sample Preservation

Do not use preservatives for samples that are going to be analyzed immediately unless instructed otherwise by the laboratory that will be analyzing them. Preservatives tend to distort the organisms and make them more difficult to identify. Instead, collect samples in an amber bottle (if available), wrap a bottle in foil, or use whatever method is available to exclude as much light as possible. Wrap the sample in wet paper to keep it as cool as possible, but do not place it directly on ice.

Several algal preservatives are available, each having its advantages and disadvantages. If in doubt, contact the laboratory or person responsible for identifying the plankton for guidance. The most commonly used preservative for phytoplankton is Lugol's solution (5 to 10 percent); however, formaldehyde (3 to 5 percent) or glutaraldehyde (2 percent) may be used.

Add approximately 0.5 to 1.0 mL of Lugol's for every 100 mL of sample. The fixed sample will be light brownish, the color of weak tea or brandy. Lugol's solution is degraded by light, so both its container and preserved water samples should be stored in dark containers or using some other method to exclude light.

### *Recipe for Lugol's Solution*

Prepare Lugol's solution by dissolving 20 g potassium iodide and 10 g iodine crystals in 200 mL distilled water containing 20 mL glacial acetic acid.

## Safety

Label the secondary container with solution components. Be sure to add the acid to water. Wear safety glasses and latex gloves when preparing this solution.

## Labeling Sample Containers

Attach a label to the outside of the container making sure the container is dry and wrap with clear tape to ensure that the label stays on the container. Labels must contain the following information.

- station number and location description
- date and time of collection
- preservative used
- name of each collector
- sample type (grab, integrated, net, net composite)
- sample volume
- container replicate number if needed (for example, *1 of 2* or *2 of 2*)

Additional labeling is necessary depending on the method of collection.

For net hauls include:

- number of hauls
- depth of hauls
- diameter of net mouth
- net mesh size

For Kemmerer (or Van Dorn) samples, subsequently filtered with a net and composited, include:

- volume of sampler (liters)
- sampling depths
- number of samples

## Sample Storage

Samples must be stored at a moderate temperature in the dark until analysis.

## Sample Processing

The purpose of samples collected for HAB or other blooms is to identify the bloom organisms and estimate the abundance of the organism under investigation—for example, *Karenia brevis* (red tide) or *Prymnesium parvum* (golden alga). These samples will usually be examined within 24 hours of collection in the event of an accompanying red-tide outbreak or fish kill unrelated to a planned study of the algal community. In those cases it is acceptable to only identify and enumerate the organisms of interest.

For other types of sampling, the purpose of microscopic examination of the phytoplankton sample is to inventory the algal community. For these samples as well as HAB samples, it is important to examine them within 24 hours of collection unless they have been chemically preserved in Lugol's solution or another chemical fixative. Preserved samples can be stored

indefinitely; however, pigmentation that may aid in identification will fade quickly, making it preferable to analyze them as soon as possible upon return to the laboratory.

Quantitative microscopic analyses of grab samples should follow the methods outlined in APHA 2012 (or the latest edition), Section 10200 F. “Phytoplankton Counting Techniques.”

## ***Zooplankton Collection Method***

Using a plankton net with a mesh size appropriate for the desired community, collect at least two vertical net tows per station. Use a Wisconsin-style plankton net with a mesh size of 50 to 243  $\mu\text{m}$ , depending on the size of organisms to be collected. Unless another study plan is proposed, sampling should follow protocols similar to those used in the 2007 and 2012 EPA National Lakes Studies as outlined below.

1. Determine the number of tows required to achieve a standard cumulative 5 m tow.
  - For lakes deeper than 7 m, take a 5 m tow.
  - For lakes with a depth less than 7 m, determine the number of tows that will be required to achieve the standard cumulative 5 m tow. For example, if the lake is 6 m deep, take two 2.5 m tows. Refer to Table 8.1 for the number and depth of tows.

**Table 8.1.** Number and depth of zooplankton tows.

<b>Water Body Depth (m)</b>	<b>Depth of Tow (m)</b>	<b>Number of Tows</b>
7+	5.0	1
4–6	2.5	2
2–3	1.0	5
1–2	0.5	10

2. Slowly lower the net over the side of the boat keeping it as vertical as possible until the correct depth is reached. It is helpful to mark the line attached to the net in increments of 0.5 m.
3. Retrieve the net by pulling it back to the surface at a steady rate of 0.3 m/s without stopping.
4. Once at the surface, slowly dip the net up and down in the water without submerging the net mouth to rinse the organisms into the bucket attached to the cod end of the net.
5. Splash or squirt ambient lake water against the outside of the net to rinse remaining organisms into the bucket. If necessary, rinse the insides of the net with deionized (DI) water only to avoid introducing additional organisms into the sample.
6. Remove the bucket from the net and set it into a small pail containing lake water and two Alka-Seltzer ( $\text{CO}_2$ ) tablets to narcotize the organisms in the sample. Be sure not to submerge the top of the collection bucket or sample loss will occur. Wait one minute or until all zooplankton movement has stopped.
7. Empty the collection bucket into a 125 mL sample container. Rinse the collection bucket with DI water until all zooplankton are rinsed into the sample container. Do not fill the container more than half full of sample and rinse water. If the sample and rinse water combined exceed half the sample container volume, use a second container to preserve the additional sample and label appropriately.

8. Fill the 125 mL sample container to the shoulder with 95 percent ethanol to preserve the zooplankton sample. To be effective, half the sample volume must contain preservative.

## **Labeling Sample Containers**

Attach a label to the outside of the container, making sure the container is dry and wrap with clear tape. Label the sample container with the following information and also include it in the field logbook.

- station number and location description
- date and time of collection
- sampling depths
- number of tows
- preservative used
- name of each collector
- sample type
- container replicate number if needed (for example, *1 of 2* or *2 of 2*)

## **Sample Storage**

The sample container must be stored in a cool, dark location until analyzed. It is not necessary to refrigerate preserved samples.

## **Sample Processing**

Quantitative microscopic analyses of grab samples should follow the methods outlined in APHA 2012 (or the latest edition), Section 10200 G, “Zooplankton Counting Techniques.”

