Microcystins and Cylindrospermopsin in Recreational Water
Sample Collection, Preparation, Storage and Transportation

1. Intended Use

For the collection, preparation, storage, and transportation of recreational water samples (lakes, ponds, reservoirs, etc.)to be analyzed using the ABRAXIS® Microcystins ELISA Kits (PN 520011, 520011OH, 520011SAES) and the ABRAXIS® Cylindrospermopsin ELISA Kit (522011).

Note: This guidance is not intended to replace local, state or federal requirements.

2. Sample Collection

Collect at least 100 mL of water sample and store in clear or amber **glass or polyethylene terephthalate (PETG)** sample containers.

Note: Samples to be tested for Cylindrospermopsin only can also be stored in high density polyethylene (HDPE), polycarbonate (PC), polypropylene (PP), or polystyrene (PS) containers, but the use of plastic collection and/or storagecontainers other than PETG for samples to be tested for Microcystins may result in adsorptive loss of Microcystins, producing inaccurate (falsely low) results.

3. Sample Storage/Transportation

Samples can be stored refrigerated for up to 5 days. If samples must be held for greater than 5 days, samples should be frozen. If samples are to be shipped, they should be shipped overnight, on ice.

4. Notes and Precautions

To prevent matrix interference during analysis, sample pH must be within the range of 5 – 11. Samples with pH levelsoutside of this range may produce inaccurate (falsely low) results and should be adjusted as necessary, using hydrochloric acid (HCl) or sodium hydroxide (NaOH), prior to analysis.

5. Sample Lysing

To determine total Microcystins or total Cylindrospermopsin concentration (free and cell-bound), samples must be lysed prior to analysis. Samples may be chemically lysed using ABRAXIS® QuikLyse™* (please see the ABRAXIS® QuikLyse™* users guide for additional information) as an alternative to the freeze/thaw method described below. (*ABRAXIS® QuikLyse™ reagentsmay be used in a method of U.S. Patent 9,739,777.)

Note: The use of sonication in cell lysing can negatively affect toxin concentrations, producing falsely low sample results.

To lyse samples using the freeze/thaw method:

5.1 Shake the sample thoroughly. Add 1 mL of sample to an appropriately labeled glass vial.

Note: Sample volume should be increased to 2 mL if using the Cyanotoxin Automated Assay System (CAAS) to ensure adequate sample volume for automated pipetting, or 10 mL if the sample requires filtration (see section 6 below).

5.2 Place the vial, lying on its side, in a freezer (< 0ºC) until completely frozen (approximately 1 hour,depending on freezer temperature).

Note: The vial should be placed on its side to allow for expansion of the water sample as it freezes, therefore decreasing the potential for vial breakage.

5.3 Remove the sample from the freezer and allow it to thaw completely (no visible ice crystals remaining in the sample). The sample may be placed in a room temperature or up to 37ºC water bath to thaw more rapidly.
5.4 Repeat steps 5.2 and 5.3 for an additional two cycles (for a total of three freeze/thaw cycles).
The sample is now ready to filter, if desired, or to analyze. Please see the appropriate user's guide for sample analysisprocedures.

6. Sample Filtration

Samples may be filtered prior to analysis using glass fiber filters. The use of alternate filter types (non-glass fiber filters) may produce inaccurate (falsely low) sample results, as Microcystins may bind to the filter material, removingit from the sample. Also, please note that some glass fiber filters are manufactured using a process, which may causeinterference that would result in inaccurate (falsely high) sample results. To avoid potential bias in sample results, use the filtration procedure described below.

Note: If determining total Microcystins or total Cylindrospermopsin concentration (free and cell-bound), samples must be lysed prior to filtration, (see section 5 above) to prevent the removal of cell-bound toxins, which would cause inaccurate (falsely low) results.

6.1 Shake the lysed sample thoroughly. Using a new disposable syringe, draw the sample into thesyringe to the 10 mL graduation line.
6.2 Place a new syringe filter on the tip of the syringe.
6.3 Depress the plunger until reaching the 5 mL graduation line, discarding the initial filtrate into thewaste container.
6.4 Fully depress the plunger, filtering the remaining sample into a clean, appropriately labeled glassvial.
The sample is now ready to analyze. Please see the appropriate user's guide for sample analysis procedures.

7. For ordering or technical assistance contact

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