Microcystins in Brackish Water or Seawater
Sample Preparation

1. Intended Use
For the preparation of brackish water or seawater samples.

2. Sensitivity
0.263 ppb in brackish water or seawater

3. Materials and Reagents Provided (in ABRAXIS® Seawater Clean Up Kit, 45-test, PN 529912)
Disposable 5 ¾" glass Pasteur pipettes
Disposable 9" glass Pasteur pipettes
Glass wool
Pasteur pipette bulb
ABRAXIS® Microcystins-ADDA Seawater Sample Treatment Solution
ABRAXIS® Microcystins-ADDA Seawater Sample Clean-Up Resin

4. Additional Materials Required (not provided)
12x75mm test tubes
20 mL glass vials with Teflon-lined caps
Deionized or distilled water
4 mL glass vials with Teflon-lined caps
Scoopula
Micropipettes with disposable plastic tips
Vortex mixer
ABRAXIS® Microcystins-ADDA ELISA Kit (PN 520011)

5. Notes and Precautions
This procedure is intended for use with brackish water or seawater samples.

6. Column Preparation Procedure
6.1. Place a small amount of glass wool into the top of a 5 ¾" glass Pasteur pipette. Using a 9" glass Pasteur pipette, push the glass wool into to the bottom of the 5 ¾" pipette to form the base of the column. The depth of the glass wool should be approximately 5 mm. Place the column into a 12x75 mm test tube.

6.2. Each column will require approximately 1.5 g of ABRAXIS® Seawater Sample Clean-Up Resin. Calculate and add the appropriate amount of ABRAXIS® Microcystins-ADDA Seawater Sample Clean-Up Resin to a 20 mL glass vial.

6.3. Add distilled or deionized water at an approximately 2:1 ratio to the ABRAXIS® Microcystins- ADDA Seawater Sample Clean-Up Resin (for example, 10 mL of deionized or distilled water per 5 g of Resin). Shake or vortex.

6.4. Pipette the Resin in water solution into the column using the 9" Pasteur pipette. Avoid the formation of air bubbles in the column bed by keeping the tip of the pipette at the surface of the bed being created. Fill the column to the indentation approximately 2 cm from the top of the pipette. This will create an approximately 8 cm column.

6.5. Allow the deionized or distilled water to drain from the column. Lift the tip of the column at least 1 cm above the surface of the water in the tube. Place the pipette bulb against the top of the column (do not attach the bulb to the column) and push the remaining water out of the column. Avoid allowing the tip of the column to come into contact with the water in the tube to prevent aspiration of water back into the column.

7. Sample Clean-Up

7.1. Add 1 mL of brackish water or seawater sample to a clean, appropriately labelled 4 mL glass vial. Add 50 µL of ABRAXIS® Microcystins-ADDA Seawater Sample Treatment Solution. Vortex.

7.2. Add 375 µL of the treated brackish water or seawater sample, from step 7.1, to the top of the column. Allow the sample to drain through the column and collect in clean empty vial.

7.3. Add a second 375 µL aliquot of the treated sample to the column. Allow to drain through the column into the same vial from 7.2.

7.4. Lift the tip of the column at least 1 cm above the surface of the sample in the vial. Place the pipette bulb against the top of the column (do not attach the bulb to the column) and push the remaining sample out of the column. Avoid allowing the tip of the column to come into contact with the sample in the vial to prevent aspiration of the sample back into the column.

7.5. Lower the column back into the vial. Add 500 µL of distilled or deionized water to the top of the column. Allow the rinse to drain through the column and collect with the sample.

7.6. Lift the tip of the column at least 1 cm above the surface of the sample/rinse in the vial. Place the pipette bulb against the top of the column (do not attach the bulb to the column) and push the remaining rinse out of the column. Avoid allowing the tip of the column to come into contact with the sample in the vial to prevent aspiration of the sample back into the column.

7.7. Remove the column and discard (columns are single use only). Cap vial and vortex. The sample can then be analyzed using the ABRAXIS® Microcystins-ADDA ELISA Kit.

8. Evaluation of Results

The Microcystins concentration in samples is determined by multiplying the ELISA results by a factor of 1.75. Samples showing a concentration lower than standard 1 (0.15 ppb) should be reported as containing < 0.263 ppb of Microcystins. Samples showing a higher concentration than standard 5 (5.0 ppb) can be reported as containing > 8.75 ppb of Microcystins or diluted further and re-analyzed to obtain an accurate quantitative result.

9. Performance Data

Recovery

Samples containing various concentrations of seawater were spiked with Microcystin-LR, Microcystin-LA, or Microcystin-RR, prepared as described above, and then analyzed using the ABRAXIS® Microcystins-ADDA Assay. Average recovery for Microcystin-LR was 91.7%. Average recovery for Microcystin-LA was 92.1%. Average recovery for Microcystin-RR was 98.9%.

7. For ordering or technical assistance contact

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