Dechlorination Procedure using Sodium Thiosulfate

1. SCOPE AND APPLICATION

1.1. This method is based on Standard Methods Online Edition (1997), Method 4500-Cl. It can be used to remove chlorine from bioassay, algal assay, microbiology and any other aqueous samples that need to be dechlorinated.

2. SUMMARY OF METHOD

2.1. Residual chlorine is measured. Based on the concentration of residual chlorine, the volume of sodium thiosulfate needed to dechlorinate the sample is calculated. Sodium thiosulfate is added to the sample. The residual chlorine is measured in the "dechlorinated" sample. Additional manipulations are performed if: the chlorine residual is > 0.1 mg/L after dechlorination in bioassay samples; and, greater than the MDL in other samples listed in 1.1.

2.2. The sample should have been warmed to 20.0 – 25.0 ˚C for the total residual chlorine (TRC) measurement. Use a thermometer to confirm the temperature is in the appropriate range. The TRC of the sample should have already been measured per SOP TA-06.22. The result should have been recorded in the sample bench sheet (usually the bioassay bench sheet/facility logbook) and in the chlorine logbook section of LIMS (Laboratory Information Management System) SOP TA-06.42.

3. EQUIPMENT AND SUPPLIES

3.1. 4 L Erlenmeyer flask, one each

3.2. 2 L Graduated cylinder, one each

3.3. Thermometer (glass or digital), minimum 0-30 °C range (Calibrated per SOP TA-06.13 or SOP LB-035)

3.4. Hach pocket colorimeter™ or colorimeter™ II chlorine test kits (S/N 05110C042002)

3.5. 1 mL and 2 mL Disposable pipets

3.6. 2.5cm Magnetic stir-bar

3.7. Magnetic stirrer

3.8. Magnetic stir-bar retriever

3.9. Laboratory calculator

3.10. Computer with access to LIMS

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4. REAGENTS AND STANDARDS

4.1. Chlorinated sample

4.2. Sodium thiosulfate ($Na_2S_2O_35H_2O$), 0.025 N, ACS grade

5. METHOD

5.1. After the TRC has been measured, use the following formula to determine the amount of sodium thiosulfate to add to the total volume of sample: **X = [(TRC) × (total volume) / 886.25]**

5.1.1. X = volume in mL of 0.025N sodium thiosulfate to add to sample

5.1.2. Total volume = volume of sample to be dechlorinated in milliliters

5.1.3. 886.25 µg/L is a dechlorination constant

5.1.3.1. Constant 886.25 µg/L is specific to the 0.0250N sodium thiosulfate.

5.1.4. **Example**: TRC = 0.5 mg/L, 2500 ml total volume of sample to be dechlorinated would require 1.4 mL of 0.025N sodium thiosulfate. This is 0.56 mL per L of sample.

5.1.4.1. X = [(TRC) × (total volume) / 886.25]

X = [(0.5 mg/L) (2500 ml) / 886.25 µg/L]

X = (1.25 mg mL/L) / 886.25 µg/L

X = (0.0014 mg mL/ µg) (1000 µg/mg)

X = 1.4mL

5.1.4.2. 1.4 mL is per 2500 mL, but it's needed to be used per L for application.

1.4 mL/2.5L)

= 0.56 mL/L

5.2. Use the graduated cylinder to add the desired volume of sample to the 4-L flask.

5.2.1. Note: If the volume of sample is smaller, a smaller flask can be used.

5.3. Put the stir bar in the 4-L flask and place both on the magnetic stirrer.

5.4. Turn the magnetic stirrer on and adjust to medium speed.

5.4.1. Set the speed fast enough to stir the sample without aerating it.

5.5. Use the 2-mL sterile disposable pipet to add the calculated volume of 0.025 N $Na_2S_2O_4$ to the flask. Keep track of the volume added to the flask as the total volume of sodium thiosulfate added to the sample (expressed as mL of sodium thiosulfate per L of sample) will have to be recorded on the *Bioassay Benchsheets*.

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5.5.1. To determine the mL of sodium thiosulfate per liter of sample, divide the total mL of sodium thiosulfate by the volume (in liters) of sample.

5.6. Use the Hach chlorine test kit to determine the total residual chlorine in the dechlorinated sample. Record the result on the *Bioassay Bench sheets* and in the *LIMS Logbook TOX_CHLORINE.*

5.7. If the TRC of the dechlorinated sample is less than 0.1 mg/L (ideally <0.03 mg/L), the sample is ready to be used (go to Step 5.8). If the TRC is > 0.1 mg/L, check the original calculation and then (if calculation is correct) repeat Step 5.1, Step 5.5 and Step 5.6. DO NOT ADD more than three times the original calculated volume (if correct) of sodium thiosulfate to the sample.

5.7.1. The necessity of adding more than the original calculated volume of sodium thiosulfate to the sample is usually the result of interfering substances in the sample.

5.8. Record the total volume (expressed as mL/L) of sodium thiosulfate used to dechlorinate the sample on the bench sheet. Record the final TRC of the dechlorinated sample on the *Bioassay Bench sheets*.

5.8.1. Reminder: Tox Bioassays that require chlorine adjustment are accompanied by a screen test consisting of a sodium thiosulfate spiked control and 100% non-dechlorinated sample. AGP requires a sodium thiosulfate spiked blank and a non-dechlorinated sample to accompany the regular lab blank and dechlorinated sample. AGP blanks and bioassay controls are spiked with the same amount of sodium thiosulfate that was added to the sample.

5.9. Place the used pipet in the disposable glass container. Place dirty glassware on the "to be washed" cart.

6. DATA ARCHIVAL

6.1. Use a black, waterproof pen for all bench sheets and logbooks.

6.2. Record information in *Bioassay Bench Sheets* logbooks.

6.3. To correct raw data entries, place a single line through the incorrect entry, write the corrected entry near the error, date and initial the correction. Do not overwrite entries.

6.3.1. The corrected entry is written as near its appropriate space as possible or else in the comment box on the appropriate bench sheet.

6.3.2. If the correction requires an explanation, write a comment and reference the error. Initial and date the explanation. Do not attempt to put all information (correction and comments) into a small space.

6.4. Use sequential alphabetical annotation to identify errors and corresponding comments (use A for first error and use A for corresponding explanatory comment; use B for second error, etc.).

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7. QUALITY CONTROL

7.1. Refer to Laboratory Quality Manual for corrective action and handling out-of-control data.

7.2. If a problem occurs during the sample processing or testing, the following steps are performed: 1) Lab supervisor is notified. 2) Client is notified (client makes decision for additional testing). 3) Section administrator is notified. 4) A non-conformance report (NCR) is submitted describing the problem and the corrective action.

7.2.1. To create NCR: open LIMS and go to Laboratory/Administrative/Non-Conformance Report. Refer to https://floridadep.gov/dear/quality-assurance/content/dep-sops for sample collection and preservation, and the Laboratory Quality Manual for corrective action and handling out-of-control data.

8. SAFETY/HAZARDOUS WASTE MANAGEMENT

8.1. Proper laboratory protection, including lab hood or ventilation system, lab coat, gloves and safety glasses, is required when working with chemicals and unprocessed samples.

8.2. Refer to Laboratory Quality Manual for sample disposal.

8.3. Refer to the Laboratory Health and Safety Manual and Hazardous Waste Management.

9. REFERENCES

9.1. Standard Methods Online Edition (1997), Method 4500-Cl.

9.2. DEP SOP TA-06.05, Temperature Calibration and Measurement using YSI Model 58 Dissolved-Oxygen Meter.

9.3. DEP SOP LB-035, Calibration of Lab Thermometers Using a Constant Temperature Calibration Bath.

9.4. DEP SOP TA-06.13, Thermometer Calibration and Use.

9.5. DEP SOP TA-06.22, Measuring Total Residual Chlorine using the Hach® Pocket Colorimeter™ and Colorimeter™ II Test Kits.

9.6. Laboratory Quality Manual https://floridadep.gov/dear/florida-dep-laboratory/content/dep-laboratory-quality-assurance-manual-and-sops

9.7. Laboratory Health and Safety Manual and Hazardous Waste Management https://www.floridadep.net/resources/health-safety-manual

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APPENDIX OF CHANGES

12/21/2007	Added Scope and Application; Summary of Method and Appendix of Changes. Moved reference from title to Scope and Application.
11/18/2008	Added Reference section.
08/14/2013	Reformatted SOP.
02/11/2015	Formatted for 508 compliance.
08/17/2015	Updated links.
09/14/2016	Updated links. Added reference to SOP LB-035.
09/15/2017	Updated links.
08/30/2018	Updated links. Updated Colorimeter information. Changed font to Calibri
09/13/2018	Updated Colorimeter SOP title (M. Faircloth 09/13/2018)
08/01/2019	No significant changes.
09/02/2020	Updated sections with universal language. Rearranged equipment order.
05/03/2021	Updated links.
09/02/2022	Updated links.
09/01/2023	Added reminder for screen tests. Checked links.

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