Measuring Total Alkalinity and Hardness

(SM 2320B and SM 2340C as adapted from Hach kit instruction manual)

1. SCOPE AND APPLICATION

    1.1. These methods are based on Standard Methods 2320B and 2340C, as adapted from the HACH Alkalinity and Hardness Kit instruction manual. They are used to measure alkalinity and hardness in aqueous solutions.

    1.2. Measuring Limits: Hardness is generally not measured in samples having salinity of >1 parts per thousand (ppth) or conductivity > 5,000 µmhos-cm.

    1.3. The Bioassay laboratory is not NELAC certified for these analyses. See section 5.1.

2. SUMMARY OF METHOD

    2.1. Sample is measured into titration bottles. Reagents are added and mixed. Samples are then titrated with appropriate titrant until color change is complete. Volume of titrant used is converted to alkalinity or hardness as mg/L calcium carbonate ($CaCO_3$).

    2.2. Interferences:

        2.2.1. For alkalinity, interferences include, but are not limited to: soaps, oily matter, suspended solids, or precipitates.

        2.2.2. For hardness, interferences include, but are not limited to: metals (e.g. copper, iron, zinc), orthophosphate, and highly buffered samples.

    2.3. Definitions:

        2.3.1. **Alkalinity** — an aqueous solution's acid-neutralizing capacity

        2.3.2. **Hardness** — the amount of dissolved calcium and magnesium in the water

3. EQUIPMENT AND SUPPLIES

    3.1. 10 mL volumetric pipettes

    3.2. 2 mL volumetric pipettes

    3.3. Magnetic stir plate

    3.4. 25 mL Titration bottles (2 per sample)

    3.5. Magnetic stir-bar (1 per bottle)

    3.6. Scissors

    3.7. Computer with access to the Laboratory Information Management System (LIMS)

    3.8. Parameter Check logbook

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3.9. *Bioassay Sample Log-in* Logbook (when applicable)

3.10. *Organism Log-in* logbook (when applicable)

3.11. *Bioassay Bench Sheets* logbooks (when applicable)

4. REAGENTS AND STANDARDS

4.1. Alkalinity

4.1.1. Bromcresol Green-Methyl-Red Indicator powder pillows

4.1.2. Phenolphthalein Indicator Solution

4.1.3. Standard Sulfuric Acid

4.1.4. Alkalinity standard (100 mg/L), Orion® brand, 1000 mg/L

4.1.4.1. To make 100 mg/L standard using a 1000 mg/L standard, dilute the 1000 mg/L standard 9 to 1 with DI water (see Section 7).

4.1.4.2. Ex. 500mL is the typical volume made for the 100mg/L standard, consisting of 50mL of the 1000 mg/L standard solution and 450mL of DI water.

4.2. Hardness

4.2.1. ManVer II hardness powder pillows

4.2.2. Hardness I buffer

4.2.3. TitraVer titrating solution

4.2.4. Hardness standard, 100 mg/L as $CaCO_3$

5. METHODS

5.1. Results of alkalinity and hardness measurements performed on samples in the Bioassay laboratory **are not reportable for compliance purposes.** The results are used to determine potential causes of observed toxicity.

5.2. A Quality Control check is required to be performed prior to titrating samples once daily. Refer to Section 7.1-7.2 for method. The following steps (5.2-5.9) are done on the standard solution for the Quality Control check (as if it was a sample).

5.3. For each sample, add a magnetic stir-bar to 2 clean titration bottles (2 reps for each analyte being measured).

5.4. Pipette 10 mL of sample into each bottle.

5.5. Place titration bottles on the magnetic stir-plate and set stir-plate on low/medium speed.

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5.5.1. Up to four bottles can fit on the stir-plate; however, it is not required that all 4 be used at one time. It is the analyst's choice.

5.6. Add reagents.

5.6.1. **Alkalinity:** Add one drop of Phenolphthalein Indicator Solution to each alkalinity titration bottle. If the sample does not turn pink, proceed to Step 5.6.

5.6.1.1. If the sample turns pink:

5.6.1.1.1. Use the 2 mL pipet to titrate (slowly add) with standard sulfuric acid until the color disappears.

5.6.1.1.2. Multiply the volume of standard sulfuric acid used by 100. This gives the Phenolphthalein Alkalinity in mg/L (ppm) Calcium Carbonate. Initial volume is in milliliters. For example, if the pipet shows that 1 mL was used to titrate the solution, the value would be 100 mg/L, calculated from: 1 mL x 100 = 100 mg/L. Likewise, if 1.3 mL was used to titrate, the value would be 130 mg/L, calculated from: 1.3 mL x 100 = 130 mg/L.

5.6.1.1.3. Note the volumes (mL) of standard sulfuric. This result is added to the alkalinity determined in Step 5.8.

5.6.2. **Hardness:** Add 3 drops of Hardness I to each hardness titration bottle.

5.7. Use the scissors to open appropriate indicator powder pillows (given in next step). Add the contents of one indicator pillow per corresponding bottle.

5.7.1. **Alkalinity:** Bromcresol Green-Methyl-Red indicator powder; sample should turn green/blue-green

5.7.2. **Hardness:** ManVer II indicator powder; sample should turn pink/red

5.7.2.1. In samples with low hardness and low ionic strength (e.g. water from blackwater swamps), it may require two powder pillows to develop sufficient color to perform the titration.

5.8. Using a 2 mL pipette, accurately titrate the samples.

5.8.1. **Alkalinity:** Titrate with standard sulfuric acid until the color changes from green/blue-green to pink/red. Titrate slowly toward the endpoint, as it takes a little time for the reaction and resulting color change to take place.

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Tip: If it is difficult to determine the exact color break point, titrate until the color shift is complete. Then, subtract 0.1 mL from the volume of sulfuric acid used.

5.8.2. **Hardness:** Titrate with TitraVer Solution until the color changes from pink/red to blue. Titrate slowly toward the endpoint, as it takes a little time for the reaction and resulting color change to take place. If more than 4 mL of TitraVer is needed to reach the endpoint, the sample can be diluted; or the analyses can be stopped, and the result reported as > 400 mg/L hardness.

Tip: Just before the hardness titration endpoint, the solution will turn purple. It usually takes only one to two more drops of titrant to turn the solution blue (i.e., reach the endpoint).

5.9. Calculate the final alkalinity and hardness values. Be sure to correct for any dilutions by multiplying the result by the dilution factor.

5.9.1. **Alkalinity:** Multiply the volume of acid used by 100. Average the two results obtained for one sample. Add any phenolphthalein alkalinity determined in Step 5.5.1.1.2. This is the total alkalinity of the water in mg/L as $CaCO_3$.

5.9.2. **Hardness:** Multiply the volume of TitraVer used by 100. Average the two results obtained for one sample. This is the total hardness of the water in mg/L as $CaCO_3$.

5.9.2.1. **Example:** 0.89 mL x 100 = 89 mg/L

5.10. Record the volume of titrant used in *LIMS/Laboratory/Technical/Laboratory Logbook/TOX_ALK_HARD* for rep 1, 2 and the mean on their respective screens.

5.10.1. If the sample is facility water, record the results in the *Bioassay Sample Log-in* logbook in addition to LIMS. If the sample is organism holding water, record the numbers in the *Organism Log-in* logbook in addition to LIMS. All results will initially be recorded in the *Parameter Check* logbook before being transferred to LIMS.

5.10.2. Facility water and dilution water values will also need to be transcribed in the appropriate *Bioassay Bench Sheets* logbooks during testing.

5.11. Repeat Steps 5.2-5.9 for each sample.

5.12. At the sink, use the deionized (DI) water to rinse the titration bottles at least three times. Shake/drain residual water from titration bottles and allow to dry fully (or fully drain) before use with another sample.

6. DATA ARCHIVAL

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

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6.2. Make sure that the lot numbers or serial numbers of the standards are recorded in the logbook.

6.3. To correct raw data entries, place a single line through the incorrect or illegible 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 as possible to the error or 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.

6.3.3. 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.).

6.4. FDEP Laboratory Information Management System (LIMS).

6.5. Record information in appropriate logbooks (*Bioassay Sample Log-in* logbook, *Organism Log-in* logbook and *Bioassay Bench Sheets* logbooks).

7. QUALITY CONTROL

7.1. A quality control check is performed prior to titrating samples once daily when analysis is performed.

7.1.1. **Alkalinity:** Perform Steps 5.2.-5.9. using the 100 mg/L alkalinity standard as the sample. The standard result should be 100 mg/L (after subtraction of the alkalinity of the preparation water – see Note below). If the result differs from 100 by more than ±10%, prepare a second standard and test. Review the Hach manual for trouble-shooting instructions. Record in LIMS logbook TOX_ALK_HARD.

NOTE: When the alkalinity standard is prepared, the alkalinity of the deionized water (~10-15 mg/L as CaCO3) used to prepare the 100 mg/L standard is measured and recorded in LIMS logbook TOX_ALK_HARD.

7.1.2. **Hardness:** Perform Steps 5.2.-5.9. using the 100 mg/L hardness standard as the sample. The standard result should be 100 mg/L. If the result differs from 100 by more than ±10%, prepare a second standard and test. Review the Hach manual for trouble-shooting instructions. Record in LIMS logbook TOX_ALK_HARD.

7.2. Two alkalinity measurements are made per sample. The alkalinity procedure covers a range of 0-250 mg/L. If the results are >250 mg/L, dilute the sample with deionized water and perform the procedure again. Multiply the result by the dilution factor.

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7.3. Two hardness measurements are made per sample (see 1.2 for exclusions). When the hardness titration volume exceeds 4 ml (400 mg/L as CaCO3), stop the titration, and report the values as > 400 mg/L as CaCO3.

7.4. Titration bottles are thoroughly rinsed and dried before use.

7.5. A new, sterile pipette is used for each sample. Proper pipetting techniques are used to ensure the accuracy of the volumes being measured.

8. SAFERY/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 Assurance Manual for sample disposal.

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

8.4. Read MSDS information for reagent handling and disposal.

9. REFERENCES

9.1. Instruction Manual for HACH Alkalinity and Hardness Kit (located in B246)

9.2. Standard Methods 2320B

9.3. Standard Methods 2320C

9.4. FDEP Laboratory Quality Assurance Manual

9.5. FDEP Laboratory Health and Safety Manual

9.6. USGS Hardness of Water

9.7. MSDS safety sheets

APPENDIX OF SIGNIFICANT CHANGES

07/29/2013 Combined and reformatted SOPs TA-06.08-3.16 and TA-06.09-4.16.

01/30/2015 Formatted for 508 compliance.

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