Analytical Procedures

Analysis of VERSENE™ Chelating Agents

Safety Precautions
Before using any of the chelating agents in analytical or other procedures, study the Safety and Handling section for suggested minimal precautions for safe handling of the chelating agents. Note also that certain reagents used may pose a serious health hazard in handling. It is important that appropriate safe handling procedures are followed. Users should obtain complete safety and handling recommendations from suppliers.

Oxalate Titration for Chelation Value
The oxalate titration for chelation value is an accurate and convenient method for determining chelation value of many chelant products. It is the industry standard method for chelating agents in North America. Characteristics of the calcium oxalate endpoint titration include:

- The chelation of calcium occurs quantitatively and quite definitely in a 1:1 molar ratio with VERSENE™ 100, VERSENEX™ 80, VERSENOIL™ 120, and other EDTA-, DTPA- and HEDTA-based products;
- The materials used and apparatus needed are likely to be found in any laboratory;
- The solutions used are quite stable and may be kept for long periods of time.

Principle
The total chelation value is determined by titrating with a solution of calcium salt in the presence of oxalate ion at a pH of 11.0 or slightly higher. The chelating agent will complex the calcium until an excess of calcium is present; this end point is indicated by the appearance of the white calcium oxalate precipitate.
Reagents

1. Ammonium Oxalate Monohydrate, 3.0% solution. Dissolve approximately 30g of ACS reagent grade (calcium-free) ammonium oxalate monohydrate in a liter of distilled water. Prepared solutions of ammonium oxalate are also available from laboratory chemical suppliers.

2. Sodium Hydroxide, 50%. Dissolve about 500g of reagent-grade sodium hydroxide in 500ml of distilled water. Store in a polyethylene bottle. Also commercially available from a variety of laboratory chemical suppliers.

3. Calcium Chloride, 0.5M standard solution. Dissolve approximately 73.5g of reagent-grade calcium chloride dihydrate and dilute to one liter with distilled water. Prepared solutions of calcium chloride are also available from various laboratory chemical suppliers. The solution is then standardized with dry, purified ethylenediaminetetraacetic acid (J.T. Baker, Ultrex EDTA, or equivalent).

Standardization of calcium chloride: To the nearest 0.1 milligram, weigh a 2.5-3.0g of dry, purified EDTA into a 200ml beaker or other suitable titration vessel. Add 80ml of distilled water and sufficient 50% sodium hydroxide to raise the pH to 11-12. Add 20ml 3.0% ammonium oxalate monohydrate, then titrate with the calcium chloride until the first faint permanent turbidity is reached. Check pH; if it is less than 11, add enough 50% sodium hydroxide to raise the pH to above 11. If turbidity disappears, continue titration to the endpoint. For standardization purposes, it is recommended to take the average value of three or more replicate analyses.

Calculation

\[
\text{Calculation} \quad \text{grams std. grade EDTA acid} \quad \frac{\text{Molarity} = \text{ml CaCl}_2 \text{, titrant solution}}{\times 3.422} \times 100
\]

Procedure

To the nearest milligram, weigh a sample containing approximately 10 millimoles of chelating agent (for example, 10g of VERSENE™ 100 product) into a clean 200ml beaker or other suitable titration vessel. Add 85ml distilled water and 20ml of 3% ammonium oxalate monohydrate solution. Titrate the sample to the first faint permanent turbidity with standardized calcium chloride solution. The pH of the titration should be checked after the endpoint has been reached, using a pH meter or pH indicator paper. If the pH is below 11, add sodium hydroxide solution to raise the pH above 11. After the pH has been adjusted, the titration should be completed if the precipitate has dissolved.

Calculation

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\text{Calculation} \quad (\text{ml CaCl}_2 \text{ sol}) \times (\text{Molarity of CaCl}_2 \text{ sol}) \times 100 = \text{mg CaCO}_3/\text{gram of chelating agent = chelation value}
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Ferric Chloride Titration for Active Content

The ferric chloride titration also may be used to determine the active content. Since the chelation of Fe occurs in a 1:1 stoichiometry with most chelating agents, including NTA, the chelation value results of a ferric chloride titration may differ from the chelation value results of a calcium chloride titration. The ferric chloride titration may be adapted to determine the chelation value of unchelated VERSENE™, VERSENEX™, VERSENSOL™, and NTA products. This method is standard in Europe.

Principle

The active content is determined by titrating the chelating agent with a solution of ferric salt using a redox electrode to detect the endpoint at the steepest mV/ml inflection. A glycine buffer is used to keep the pH in the range of 2.8 to 3.0. The use of an automatic titrator is recommended to facilitate the

Reagents

1. Glycine buffer solution. Mix approximately 150g of reagent-grade glycine in about 500ml of deionized water in a 1500ml beaker. While stirring on a magnetic stirrer, slowly add about 75ml concentrated hydrochloric acid aqueous solution. Add deionized water to about 900ml volume and cool to room temperature. Then adjust the pH to about 2.9 using a pH meter and 50% sodium hydroxide solution. Dilute to a final volume of one liter.

2. Sodium Hydroxide, 1 Normal solution. This reagent is used for pH adjustment and does not need to be standardized.

3. Hydrochloric Acid, 1 Normal solution. This reagent is used for pH adjustment and does not need to be standardized.

4. Ferric Chloride, 0.05M: Dissolve approximately 13.52g of reagent-grade ferric chloride hexahydrate in one liter of deionized water. A small amount of HCl may be added to avoid precipitate formation. This solution must be standardized against a pure sample of EDTA.

Standardization of ferric chloride: To the nearest 0.1 milligram, weigh 0.08 to 0.12g of dry, ultrapure EDTA into a suitable titration vessel. Add 40 to 80ml of deionized water and about 2ml 1 Normal sodium hydroxide solution and dissolve all of the EDTA. Add 15ml glycine buffer and, if needed, add 1 Normal HCl and/or 1 Normal NaOH to obtain a pH of 2.8 to 3.0 as measured with a calibrated pH meter. Titrate with the ferric chloride solution to the steepest mV/ml inflection. For standardization purposes, it is recommended to take the average value of three or more replicate analyses.
Calculation
grams std. grade EDTA acid
Molarity = \text{ml FeCl}_3 \text{ titrant solution} \times 3.422

Procedure
To the nearest milligram, weigh a sample containing approximately 0.5 to 1 millimole of chelating agent (for example, 0.5 to 1.0g VERSENE™ 100 product) into a suitable titration vessel. Add 40 to 80ml of distilled water and 15ml pH 2.9 glycine buffer solution. Using a calibrated pH meter, add 1 Normal sodium hydroxide and/or 1 Normal hydrochloric acid as needed to make sure the pH is in the 2.8 to 3.0 range. Titrate using standardized 0.05M ferric chloride solution to the steepest inflection point. There may be more than one inflection point, so proceed until the mV/ml response begins to level out. Titrate more slowly near the inflection points. The total chelation value is calculated from the steepest mV/ml inflection point (normally the last inflection point if more than one is present). The use of an automatic titrator is recommended.

Calculation
\frac{(\text{ml FeCl}_3 \text{ solution}) \times (\text{molarity of FeCl}_3) \times \text{molecular weight of chelant active content}(%\text{wt})}{(\text{grams of sample}) \times 10}

Determination of Very Low Concentrations of Chelating Agent
Special situations may require analysis for the chelating agent at concentrations of less than 25ppm. Dow laboratories have developed a satisfactory method utilizing zirconium and xylenol orange. An advantage of the method is that small amounts of heavy metals do not interfere.

Principle
When a chelating agent is added to a strongly acidic solution of zirconium-xylenol orange complex, a portion of the zirconium stoichiometrically equivalent to the amount of chelating agent added is removed from the highly colored zirconium-xylenol orange complex. The decrease in absorbance at 535 millimicrons is proportional to the quantity of chelating agent needed.

Reagents
1. Chelating Agent Standard Solution A. Dissolve 0.744g of reagent disodium dihydrogen ethylenediaminetetraacetate in water and dilute to one liter. The concentration is 2 micromoles/ml.
3. Zirconium Standard Solution A. Dissolve 2.250g of zirconium oxychloride (ZrOC1\textsubscript{2}•8H\textsubscript{2}O) in 65ml of concentrated hydrochloric acid and dilute to one liter.
5. Xylenol Orange Reagent. Dissolve 0.8g of xylenol orange in 334ml of concentrated hydrochloric acid; add this to a solution of 100g of hydroxylamine hydrochloride in 300 to 400ml of water. This is diluted to one liter, allowed to stand overnight and is filtered.

Preparation of a Standard Curve
1. Pipet 5ml of zirconium standard solution B, and 5ml of xylenol orange reagent into each of six 50ml volumetric flasks. Prepare a color blank containing all reagents except the zirconium.
2. Pipet into the previously prepared volumetric flasks 0, 5, 10, 15, 20, and 25ml of the freshly prepared chelating agent standard solution B (equivalent to 0, 0.2, 0.4, 0.6, 0.8, and 1.0 micromoles of chelating agent); dilute to volume and mix well.
3. Allow the solutions to stand one hour, and determine the absorbance of each solution using a Beckman Model B Spectrophotometer or equivalent at 535 millimicrons and a cell length of 1.0cm. Use the color blank to zero the spectrophotometer.
4. Plot the absorbance against the micromoles of chelating agent.

Calculation
\frac{\text{micromoles EDTA found} \times \text{M.W. of EDTA}}{\text{ppm EDTA} = \frac{\text{gms sample}}{\text{mg EDTA}}}

Note: It is often convenient to dissolve and dilute the sample to volume in a volumetric flask, taking an aliquot for introduction into the 50ml flask used for color development. It is advisable to rerun the standard curve with each set of samples.
Other Analytical Procedures

Many colorimetric (Schwarzenbach) methods may be modified to determine the chelation value. These methods depend upon indicators such as Eriochrome Black T and Murexide, which undergo color changes in the presence of free metal ions. Details of analytical procedures for such methods are outlined in the literature. Some of the titration methods are reviewed in the reference books and bibliography, pages 5 to 6.

Other analytical techniques for chelants include high-performance liquid chromatography (HPLC) methods, spectroscopic methods, and electrochemical methods. A sampling of references to these techniques are included in the bibliography.

Use of VERSENE™ Chelating Agents in Analysis

General Metal Analysis

Because of their complexing ability, VERSENE™ Chelating Agents have wide use in analytical chemistry. Analytical methods for practically every metal are known. One such application is the rapid determination of water hardness which, because of frequent need, is outlined. The many other analytical methods based on EDTA cannot be covered here. However, detailed discussions of these methods are available in the reference books and bibliography, pages 5 to 6.

Standard Analysis for Water Hardness

The EDTA titration method to determine water hardness may be more convenient than calculating the hardness from separate calcium and magnesium analysis by atomic absorption or inductively coupled plasma spectrometry.

Principle

Azo dye indicators, such as Eriochrome Black T and Calmagite, form intensely colored wine-red complexes with magnesium and calcium. Thus, a solution containing only a very small amount of magnesium dye complex will show a distinct wine-red color. EDTA has a greater affinity for calcium and magnesium ions than the indicator dyes. When the last trace of hardness ions held by the dye is titrated with EDTA, the metal is extracted from the indicator complex to yield the color of the free dye (clear blue at a pH of 10.0). This color transition is more pronounced for Mg-dye than Ca-dye, so to ensure the sharpest endpoint for hardness determinations in samples containing little or no magnesium, a small amount of magnesium-EDTA chelate is added to the titration. The small amount of magnesium-EDTA chelate does not affect the hardness results since it is fully chelated and is thus neutral to the titration. EDTA will chelate calcium preferentially over magnesium, so during the course of the titration, magnesium ions will be available for the sharpest possible endpoint. When the last trace of wine-red magnesium-indicator complex has been titrated by EDTA, a sharp color transition from wine-red to clear blue is observed.

Reagents

1. Buffer solution. Dilute 125ml of concentrated ammonium hydroxide to approximately 800ml with deionized water. While stirring constantly, add 25ml concentrated hydrochloric acid solution, followed by 1 gram magnesium salt of EDTA. If needed, adjust the pH to 10.0 by dropwise addition of hydrochloric acid or ammonium hydroxide. Dilute to one liter. (The magnesium salt of EDTA is commercially available from a variety of laboratory chemical suppliers. Various commercially prepared hardness buffer solutions similar to the one described above are also available).

2. Standard Calcium Solution, 0.01 M. Weigh 1.000 gram anhydrous CaCO₃ (of primary standard grade) into a 125ml Erlenmeyer flask. Add about 20ml water and while swirling, add hydrochloric acid dropwise until all of the CaCO₃ just dissolves, allowing time for dissolution to occur between drops. Bring the volume up to about 50ml with distilled water and carefully boil the flask to expel carbon dioxide. Cool, quantitatively transfer the contents to a Class A 1-liter volumetric flask, and dilute to one liter. Calculate the exact molarity based on the amount of anhydrous, primary standard grade CaCO₃ weighed.

3. Eriochrome Black T indicator. Dissolve 0.5 grams of the dye in 100ml triethanolamine. Store in a brown bottle. (Calmagite indicator may also be used. Calmagite dye may be dissolved in water at about 0.1%).

4. Standard solution of VERSENE™ Chelating Agent, 0.01M. Dissolve 3.723g analytical reagent-grade disodium ethylenediaminetetraacetate dihydrate in distilled water and dilute to one liter. Standardize against the standard calcium solution, 0.01M, prepared above.

Procedure

Pipe 50.0ml of the water to be tested into a 125ml Erlenmeyer flask. Add 10ml of the buffer solution and 2-3 drops of Eriochrome Black T indicator solution. Titrate with standard solution of VERSENE™ to a color change from wine-red to clear blue.

Calculation

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\text{Hardness} = \frac{\text{ml std. EDTA for titration} \times \text{molarity EDTA solution} \times 1000}{\text{ml sample}}
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\text{Hardness (as mg CaCO}_3\text{ per liter)} = \text{ml sample}
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Source References and Analytical Bibliography

Analytical Source References

Analytical Bibliography
The following references represent a sample of the many different methods for the analysis of chelating agents and the use of chelating agents in analysis. This list does not imply any recommendations, but rather can serve as a starting point for further information on this voluminous subject.

Analytical Bibliography, continued


