Test Project

Chemical Laboratory Technology

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Introduction

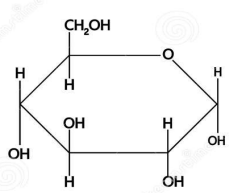
The determination of the content of impurities with reducing capacity at trace levels in purified disaccharide matrices is based on a volumetric complexometric method.

The analysis consists of two consecutive stages:

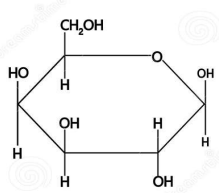
1. Heterogeneous Redox Reaction: Species with free carbonyl groups react with a known excess of alkaline cupric solution (stabilized with sodium tartrate and carbonate), proportionally reducing the soluble cooper ions to an insoluble precipitate of cuprous oxide.
2. Complexometric Titration: The remaining fraction of cooper ions that did not participate in the reduction reaction is quantified by titration with a standardized solution of ethylenediaminetetraacetic acid, using a visual color change metallochromic indicator system.

To prepare the calibration curve, select from the reagent cabinet the bottle of reagent that will reproduce the reducing capacity of the sample. Standard to be Selected: The appropriate reagent for preparing the stock solution must be a substance whose molecular structure corresponds to a monomer (or equimolar mixture of monomers) that has a free anomeric carbon with an unsubstituted hemiacetal or hemiketal hydroxyl group -OH. In an alkaline aqueous medium, this configuration allows the reversible opening of the ring to form the open-chain structure, exposing a carbonyl functional group (aldehyde in aldohexoses or α-hydroxyketone in ketohexoses). This free carbonyl group acts as a reducing agent capable of donating electrons and reducing dissolved cooper to cuprous oxide.

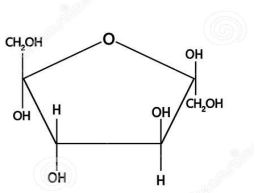
The following are the structural formulas for reducing and non-reducing sugars.



Glucose

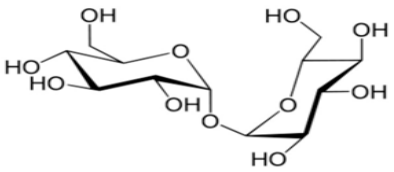


Galactose

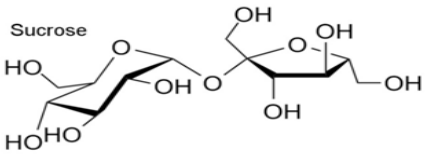


Fructose

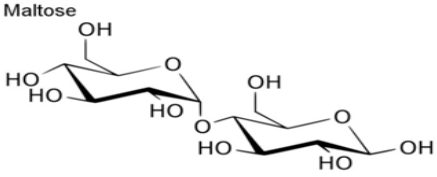
Trehalose



Sucrose



Maltose



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Description of project and tasks

- Planning
- Preparation of solutions.
- Solutions standardzation.
- Sample analysis.
- Certified Standard Material (CSM) analysis.
- Data processing.
- Conclusion.

The duration of this module is 4 hours.

Instructions to the Competitor

Planning

In your planning, present:

1. The volumes of solutions to be prepared, and the justifications for such decision.
2. A list or table with all reagents that will be used during the task, their respective principal Globally Harmonized System (GHS) code, and their protective actions**. You must show this list to the Experts before starting the experimental work.**

Preparation of solutions

1. **Murexide Solution:** Weigh 0.10 g of murexide, 0.04 g of methylene blue, and 8.0 g of sodium chloride. Transfer to a porcelain mortar and grind the mixture. This solution must be stored in a capped bottle and labeled according to the SAG rules.
2. **Alkaline Copper Solution:** For 1000 mL of this solution, weigh 25 g of anhydrous sodium carbonate and dissolve in a beaker. Weigh 25 g of sodium potassium tartrate tetrahydrate and dissolve in a beaker. Transfer these two (2) solutions to a 1000 mL volumetric flask containing approximately 400 mL of deionized water and 40 mL of 1.0 mol/L sodium hydroxide solution. Weigh 6 g of copper sulfate pentahydrate, dissolve in a beaker, and transfer quantitatively under continuous stirring to the same flask. Make up the volume with deionized water and homogenize. Estimate the consumption of this solution and prepare a sufficient volume. The excess volume of this solution must be stored and labeled according to the SAG rules.
3. **1 M Sodium Hydroxide Solution:** Estimate the consumption of this solution and prepare a sufficient volume.
4. **0.005 M EDTA Solution:** Estimate the consumption of this solution and prepare a sufficient volume.
5. **1% w/w reducing sugar solution:** Choose a reducing sugar in the module C cabinet and prepare a sufficient volume of this solution. Record the chosen reagent in your planning.
6. **0.05% w/w reducing sugar solution:** Prepare a sufficient volume of this solution from a dilution of the 1% w/w solution. Before filling the volumetric flask to the mark, add three (3) drops of 0.1% Phenolphthalein, and add 1 M NaOH until the color changes. Then, add one (1) or two (2) drops of 0.5 M HCl until the solution becomes colorless again. Estimate the consumption of this solution and prepare a sufficient volume.
7. **0.1% w/v Phenolphthalein Solution:** This solution will be ready in the module C cabinet.
8. **0.5 M Hydrochloric Acid Solution:** This solution will be ready in the module C cabinet.

Record in your planning the volumes of solutions you decided to prepare.

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Solutions standardization

Use the 0.05% reducing sugar solution that you prepared to make at least 5 dilutions with concentrations ranging between 0% and 0.05%. Prepare these dilutions directly in 125 mL Erlenmeyer flasks. The combined volume of water and the 0.5% reducing sugar solution must always be 5 mL. In each Erlenmeyer flask, you must also add and completely dissolve 5 g of Sucrose.

Add 5 mL of the alkaline copper solution and homogenize.

Place each Erlenmeyer flask in a boiling water bath and keep it there for 5 ± 1 minutes – show the Experts the moment you start and stop the stopwatch.

Remove each Erlenmeyer flask from the water bath and cool immediately.

Add about 0.1 g of the murexide solution and titrate with the 0.005 mol/L EDTA solution until the color changes from green to purple.

Record in your Word report:

1. A table with: The volumes of 0.05% solution and water used, the concentrations obtained after dilution, and the water bath times for each Erlenmeyer flask.
2. The calibration curve graph.

Sample analysis

Weigh 5 g of the crystal sugar sample, add 5 mL of water, and stir until completely dissolved.

Add 5 mL of the alkaline copper solution and homogenize.

Place each Erlenmeyer flask in a boiling water bath and keep it there for 5 ± 1 minutes – show the Experts the moment you start and stop the stopwatch.

Remove each Erlenmeyer flask from the water bath and cool immediately.

Add about 0.1 g of the murexide solution and titrate with the 0.005 mol/L EDTA solution until the color changes from green to purple.

Execute this analysis in triplicate

Certified Standard Material (CSM) analysis

Use the same procedure as the sample analysis.

Execute this analysis in triplicate.

Data processing

The calculations for:

1. Planning
2. Preparation of Solutions
3. Calibration curve
4. Reducing Sugars Concentration of the Sample
5. Method test analyzing the Certified Reference Material
6. Calculations to answer the task questions must be **identified and numbered using exactly these headings** and may be submitted either handwritten or in an Excel spreadsheet. For handwritten work, use the standardized sheets provided by the Experts. For Excel spreadsheets, use a separate tab for each of the six (6) headings and submit the .xlsx file within the allotted task time.

Conclusion

All text (measurement records, discussion, planning justifications, conclusions, answers to the task questions, etc.) must be submitted within the task duration in a text file saved in PDF format. It can be submitted in your native language. **Any other format will not be considered in the assessment.**

Task questions

1. Calculate the reducing sugars content in the crystal sugar sample (% w/w) and its respective coefficient of percentage variation.
2. Calculate the reducing sugars content in the Certified Reference Material (% w/w), the relative error compared to twe CRM specifications, and its respective coefficient of percentage variation.
3. Assuming the crystal sugar sample you received is representative of a batch of sugar classified as 3A, conclude whether the sample is approved. Write the text of an email addressed to the quality control supervisor informing them about the approval or rejection of the analyzed batch.

Other

List of annexes:

1. Safety data sheets for all chemical reagents in the Competition, in digital format.
2. Reference Material Certificate.
3. Sample analysis report.
4. Especificações do Açúcar Branco tipo 3A.
5. Basic infrastructure list for this module.

Check if you have received all these annexes and immediately inform the Experts if any are missing.

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