Experiment 36

The Calcium Content of Milk

Problem
How can we determine the calcium content of milk?

Introduction
Calcium has long been known to be necessary for a healthy diet. Dairy products are touted as an excellent source of calcium. In this experiment you will determine the amount of calcium in milk by gravimetric titration with EDTA. The calcium concentration in the milk will be determined by finding the mass of the EDTA solution needed to titrate a known mass of milk to a blue endpoint. This process is similar to the method often used to find the hardness of water which is caused primarily by magnesium and calcium ions.

EDTA (ethylenediamine tetraacetic acid) is a large molecule that has a strong attraction for metallic ions like $\text{Ca}^{2+}$, the form of calcium in milk. The calcium ions and the EDTA combine in a 1:1 mole ratio to form a large complex ion called a “chelate.” Chelates, such as the hemoglobin contains the $\text{Fe}^{2+}$ ions and is found in your blood cells, are common in nature.

In this titration, you will carefully weigh a pipet full of EDTA solution, then add the EDTA dropwise to a known mass of milk. An indicator will be added to the milk to tell you when all the calcium ions have been removed from the milk. The indicator that you will use, hydroxy naphthol blue, changes from red, when calcium is present, to blue, when all of the calcium ions have been removed from the solution. As you add the EDTA, the calcium ions will be used up, causing the indicator to change color. The blue end point appears gradually, so you may want to run a preliminary trial just to observe the color change before you do your quantitative measurements. Because the indicator only works when the system is basic, you will add a drop of 6-molar sodium hydroxide solution to each of your titration samples.

Remember that red and blue make purple; the mixture will be purple when both the calcium-containing form and the calcium-free form of the indicator are present in about equal amounts. This will occur before the blue endpoint.

Using the mass of EDTA in your solution and the mass of the milk, you will be able to determine the number of milligrams of calcium in 240 mL of milk (about one cup).

Prelaboratory Assignment
✓ Read the Introduction and Procedure before you begin.
✓ Answer the Prelaboratory Questions.
    1. Why would you expect the calcium in milk to be present as calcium ions, rather than as elemental calcium?
2. According to one source, one cup (~240 mL) of whole milk contains 291 mg of calcium. Use this to calculate the mass percent of calcium in whole milk. Assume the density of milk is 1.0 g/mL.

3. Would you expect the density of milk to be affected by its fat content? Explain. Would increased fat content raise or lower the density? Explain.

Materials

Apparatus
- Milligram balance
- Thin-stem pipets (3)
- 24-well test plate
- Plastic toothpick for stirring
- Safety goggles
- Lab apron
- Gloves (optional)

Reagents
- Milk: whole, 2%, skim
- EDTA solution, 1.00%
- 6 M NaOH
- Hydroxy naphthol blue indicator

Note: Small beakers or flasks (e.g., 10-20 mL) may be substituted for the 24-well plate.

Safety

1. Sodium hydroxide is caustic, even in small quantities. Clean up spills with large amounts of water.
2. Hydroxy naphthol blue and EDTA are considered irritants; avoid contact with your skin. You may want to wear gloves to protect your hands if you have sensitive skin.
3. Safety goggles and a lab apron must be worn at all times when working in the laboratory.

Procedure

1. Label three pipets: EDTA, NaOH and Milk; fill each with the appropriate solution.

2. Weigh the milk pipet and the EDTA pipet and record their masses in a Data Table. The mass of the NaOH pipet is not needed.

3. Add 15 to 20 drops of milk to one well of your 24 well plate. Reweigh the milk pipet and record its mass in your Data Table.

4. Add one drop of 6M NaOH to the milk in the well plate.

5. Add a very small amount of the indicator to the mixture in the well plate. Only a few crystals are needed. Stir the mixture. It should have a red to rose color. If you think the color is too light, add a few more crystals, but if you use too much, results will be inconsistent.

6. Add EDTA solution a drop at a time to the mixture in the well plate. As you add the EDTA, the calcium ions will be tied up, removing them from the milk solution and causing the color to change, first to purple, then to blue. Record the mass of the EDTA pipet after the solution has turned blue.

7. Carry out two additional trials. If the results do not show good agreement, run additional trials, as needed. If you have not achieved consistent results after 5 trials, consult your teacher.
8. **Optional** Determine the density of milk in grams per milliliter, then use this experimental value in your calculations rather than assuming that 240 mL of milk has a mass of 240 g. You must report data and show calculations.

**Cleaning Up**

1. The 6M sodium hydroxide, NaOH, is a strong base, so should be handled with care. Aside from that, there are no environmental concerns connected with this experiment.
2. Depending on the fat content of the milk used, it may take some effort to clean the well plate. The higher the fat content of the milk, the more oily the mixture and the more effort you will need to get the wells clean. Wash everything well with soap and water and return it to the location specified by your teacher.
3. Wash your hands thoroughly before leaving the laboratory.

**Analysis and Conclusions**

Complete the **Analysis and Conclusions** section for this experiment either on your Report Sheet or in your lab report as directed by your teacher.

1. The concentration of the EDTA solution has been adjusted so that exactly 1 gram of EDTA solution will tie up exactly 1.08 mg of calcium. Calculate the number of milligrams of calcium present in each of your three titration samples. Show your work for the first titration. If you carried out more than three titrations, base all your calculations on the three that show the best agreement.
2. Use your results from the first calculation to determine the number of milligrams of calcium in 1 cup (~240 mL) of milk. Report both the individual values for the three samples and an average value. Assume that skim milk has the same density as water. Show your work for trial 1.
3. Calculate the individual deviations from the average for each trial. Calculate the average deviation for your three trials.
4. When an average deviation is known for a series of analyses, the results of the analysis are generally given in the form: (average value) ± (average deviation). Report the concentration of calcium in milk with the average deviation.
5. Calculate the percent deviation for your experimental results. Show your work.
6. The USRDA for calcium is 1200 mg per day. What fraction of the daily requirement would one cup of milk provide, based on your average value?
7. According to the carton label, one cup of milk provides 35% of the USRDA for calcium. What is your percentage error, assuming the carton value is correct?
8. Women over the age of 50 and men over age of 65 need about 50% more calcium than younger adults. Why is this?
9. A student purchased a calcium supplement tablet which contained calcium carbonate. The student placed the tablet in water for several hours. Addition of NaOH and the indicator gave a blue solution. What does this indicate? Does this test show that the supplement is not giving the student any useful calcium? Explain your reasoning. (**Hint:** Consider solubility rules.)