

Carbohydrates

Introduction

Because carbohydrates are so important to humans, much research has been done concerning their properties and synthesis. As a result, there are several useful tests that are well known and easy to use. In this experiment, you will perform the following tests on a variety of known and unknown sugars to learn more about them.

Benedict's Test

Most sugars, especially monosaccharides, are reducing sugars. This means that the aldehyde functional group of a carbohydrate molecule is easily oxidized into a carboxylic acid, and thus reduce other compounds around them. An easy test for this is with Benedict's reagent, which is an aqueous solution containing copper (II) ions (Cu^{2+} ions) that produce a blue colored solution. If the copper $2+$ ions are reduced to $1+$ ions, the aqueous solution will change from blue to green and then red. Most polysaccharides are immune to this test since the aldehyde functional group is sacrificed to connect the individual units. Even some disaccharides are not reducing sugars, although most are.

Tollen's Test

This test is similar to Benedict's. (A third test, called Fehling's test, is also similar.) Aldehydes are again oxidized, this time by using a solution containing Ag^+ ions. The silver ions are reduced to silver metal, coating the side of the test tube with a "silver mirror." The same limitations regarding polysaccharides apply.

Starch-Iodine Test

Starch is a polysaccharide made up of many thousands of glucose units. When iodine is added to starch, a deep blue-black color appears. In this way, the presence (or absence) of starch can be easily detected.

Procedure

Place about 200 mL of water into a 400-mL beaker and bring it to a boil on a hot plate. This will serve as your hot water bath for the various tests.

Part 1: Reducing Sugars via Benedict's Test

Label 10 test tubes (1 through 10) and add 10 drops of the solutions listed to the respective test tubes.

#1	water	#6	1% lactose
#2	1% glucose	#7	1% starch
#3	1% fructose	#8	1% honey
#4	1% galactose	#9	1% saccharin
#5	1% sucrose	#10	1% Nutrasweet

Now add 2 mL of Benedict's reagent to each test tube and place the test tubes in the hot water bath for five minutes. Observe and record the color changes that occur.

Part 2: Reducing Sugars via Tollen's Test

The Tollen's reagent will have to be prepared immediately prior to use. To do so, add 1 mL of Tollen's solution A to 1 mL of Tollen's solution B. A large black precipitate should appear. Dissolve this precipitate by adding enough 10% ammonium hydroxide (NH_4OH) to make the solution clear (shaking will help enormously).

Since the Tollen's test is difficult to clean, perform this test only on a fresh sample of glucose and sucrose. Put a few drops of each sample in two separate test tubes, and add a few drops of bis-2-ethoxyethyl ether, which is used as the solvent instead of water. Now slowly add about half the Tollen's reagent you prepared to each test tube and shake well. The reaction may take a few minutes.

Part 3: Starch Test

Label 4 test tubes (1 through 4) and place 1 mL of the following solutions in each test tube.

#1	1% starch
#2	1% glucose
#3	1% fructose
#4	1% sucrose

Add 2 drops of iodine to each test tube and observe any color changes that occur.

After all color changes have been observed, add 1 mL of 6 M HCl to test tubes #1 and #4, and place the test tubes in a boiling-water bath. Allow to boil for 20 minutes. Note any color changes. At the end of 20 minutes, add 3 mL of Benedict's reagent and put it back in the hot water bath for a few minutes. Again, note any color changes.

Waste

Place all liquid waste from each test in the correspondingly labeled waste container. For example, pour the liquid in the Benedict's test tubes into the Benedict's waste container.

Hazards

Take extra care when using the 6 M HCl. This concentrated acid will burn the skin, so wash your hands after use. Keep your goggles on at all times to protect your eyes.

Pre-lab Questions

1. Test tube #1 contains only water. What is the purpose of testing water?
2. Briefly summarize the expected color changes that correspond with a positive result for each of the three tests.
3. For the ten samples used in Part 1, identify each as a monosaccharide, disaccharide, or polysaccharide. (Look up the structures as needed.)

Data

What data do you need to record? Refer back to the procedure and set up your own data section!

Post-lab Questions

1. Write chemical reactions for each of the monosaccharides in Part 1 that acted as reducing sugars to give positive results for the Benedict's test. (Use Fischer projections.)
2. Did all of the disaccharides used in Part 1 act as reducing sugars? Discuss the structural features of the disaccharides in the context of your results.
3. How did the Tollen's test compare to the Benedict's test? For example, were the results for each sample the same in both tests? Was one test harder to perform than the other? Were the results more or less ambiguous for a particular test?
4. Which of the samples in Part 3 underwent a color change? Were the results as you expected? Explain.
5. What did the 6 M HCl do to the starch? Use the results you obtained from the iodine starch test and the Benedict's test to support your answer.