

Exercise 20

Aim: To detect the presence of carbohydrates like glucose, sucrose and starch.

Principle: Carbohydrates with the free aldehyde or ketone groups (i.e., simple sugars) reduce copper sulphate of Benedict and Fehling's reagent to cuprous oxide forming a yellow or brownish-red coloured precipitate depending on the concentration of sugar.

Colour	Approximate amount of reducing sugar
No change of blue colour	Absence of reducing sugar
Blue changes to green precipitate	0.1-0.5 % of reducing sugar
Blue changes to yellow precipitate	to 0.5 to 1.0% of reducing sugar
Blue changes to orange-red precipitate	to 1.0-2.0 % of reducing sugar
Blue changes to brick-red precipitate	Over 2.0 % of reducing sugar

Requirements: Glasswares - test tubes, funnel, beaker; Chemicals - Benedict's solution, Fehling's solutions A and B; concentrated HCl, saturated solution of NaOH, dilute iodine solution; Equipments - water bath; Miscellaneous - test tube holder, test tube stand, spirit lamp.

Sample for test: Fruit juice of apples/banana/leaves of onion/sugar cane extract, milk, etc.

Preparation of reagents

- (i) Benedict's reagent: Mix 173g of sodium citrate and 100g of anhydrous sodium carbonate in 600mL of water in a beaker and warm gently (solution A). Dissolve 17.3g of hydrated CuSO_4 in 100mL of distilled water (solution B). Add solution B to solution A with constant stirring. Cool and transfer to a one litre flask and make the volume upto the mark with water.
- (ii) Fehling's reagent A: Dissolve 6.93g of copper sulphate in 100mL of distilled water.
- (iii) Fehling's reagent B: 20g of KOH and 34.6g of sodium-potassium tartarate (Rochelle's salt) dissolved in 100 mL of distilled water.

A. Test for glucose

Procedure

Take small pieces of banana, apple and grapes. Crush them separately and strain their juices in different test tubes. Take milk in another test tube. Cut

fresh leaves of onion bulb into small pieces and boil for 2 to 3 minutes in a test tube and filter it.

(a) Benedict's test

- Take 2 mL of juice (fruit extract) / milk / onion leaf juice in a test tube.
- Add 2 mL of Benedict's reagent to it and boil. Direct heating of test tube should not be done. It should be carried out in a water bath at the boiling point of water.
- The colour changes from blue to green and finally to orange or brick red indicating the presence of simple sugar (glucose).

Benedict's Test

Name of the food items	Colour of the Precipitate	Inference drawn
1. Apple juice	1. _____	1. _____
2. Banana extract	2. _____	2. _____
3. Grapes juice	3. _____	3. _____
4. Onion juice	4. _____	4. _____
5. Milk	5. _____	5. _____

(b) Fehling's test

- Mix equal volume of Fehling's solution A and B (1-2mL) in a test tube.
- Add equal volume of the fruit juice or onion leaf juice or milk and place it in water-bath at the boiling of water.
- The initial blue colour turns green to yellow and finally a brick-red precipitate is formed.

Fehling's Test

Name of the food items	Colour of the Precipitate	Inference drawn
1. Apple juice	1. _____	1. _____
2. Banana extract	2. _____	2. _____
3. Grapes juice	3. _____	3. _____
4. Onion juice	4. _____	4. _____
5. Milk	5. _____	5. _____

B. Test for Sucrose

Procedure

Collect sugarcane juice or cut sugarbeet into small pieces. Crush them and strain their juice.

(a) Benedict's and Fehling's test

- Perform Benedict's and Fehling's test with cane sugar or sugar beet juice as described above.
- No change of colour indicates the absence of simple sugars (i.e., monosachharides).

(b) Hydrolysis test

- Take 10 mL of sample juice in a beaker, add few drops of concentrated HCl and boil.
- After cooling, add 4 mL of saturated solution of NaOH or Na_2CO_3 to neutralize the solution. Now divide the solution into two parts. Transfer one part in test tube 'A' and the other in test tube 'B'.
- Perform Benedict's test with sample in test tube 'A'. Do you observe any change in colour? If there is a change, it indicates the presence of simple sugar or monosaccharides.
- Perform Fehling's test with sample in test tube 'B'. Do you observe any change in colour? If there is change, it indicates the presence of simple sugar or monosaccharides.

C. Test for starch (Iodine test)

Principle: Iodine specifically makes a blue-coloured complex with starch.

Procedure

- Add 1 or 2 drops of dilute iodine solution to 2 to 3mL of extract.
- A blue-black colour shows the presence of starch.
- The blue colour disappears on heating and reappears on cooling.

Discussion

The composition of blue coloured substance is not well defined. This may be an adsorption complex of starch with iodine rather than a definite compound.

Questions

1. What is the use of HCl in the test for carbohydrate?
2. Why does the blue colour disappear on boiling and reappear on cooling in the test for starch?
3. Sucrose solution gives a negative test with Benedict's test. Why?
4. Will Iodine test give a positive result with glucose, fructose or sucrose solution?
5. Why are monosaccharides called reducing sugars?

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