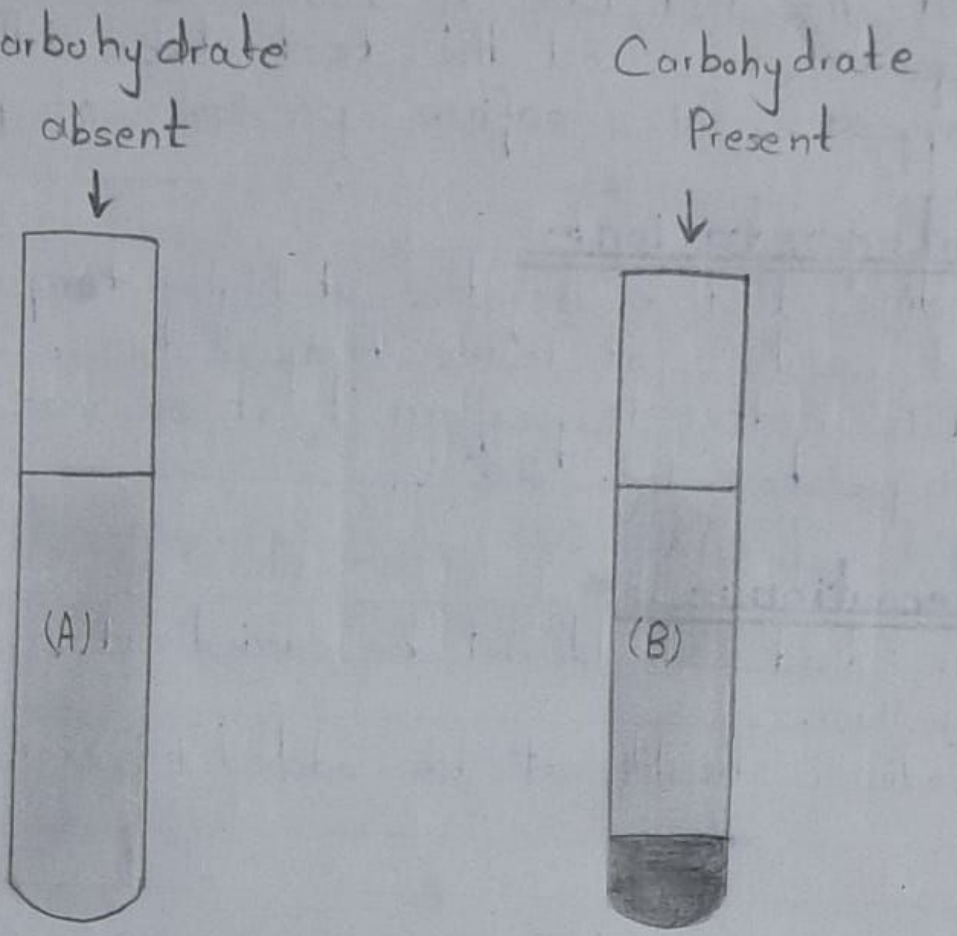

Barfoed's Test =

1- In Test tube A the solution remains Blue means that carbohydrate is absent.

2- In test tube B the red precipitates are formed means that mono or di-saccharides are present.

Faster => Monosaccharide is present

Slower => Disaccharide is present

EXPERIMENT # 04

23-Nov-2020

Title :-

Test to differentiate between Mono- and Disaccharides

3. Barfoed's Test (To differentiate monosaccharides and disaccharides)

To determine whether the reducing sugars is a monosaccharide or disaccharide. This test is basically mean to detect monosaccharides in acidic medium. It can also be used to distinguish between monosaccharide and disaccharide by controlling the time of heating. Barfoed's reagent, cupric acetate in acetic acid is slightly acidic and is balanced so that is can only be reduced by monosaccharides but not less powerful reducing sugars. Dissacharides may also react with this reagent; but the reaction is much slower when compared to monosaccharides.

Principle :-

This test differs from the Fehling's and Benedict's tests in aspects that the reduction of cupric ions is carried out in a mildly acidic medium. Aldoses and ketoses can reduce cupric ions even in acidic conditions. Some acidic medium is unfavorable for reduction, only the strongly reducing carbohydrates, i.e, monosaccharides, react very fast and give a positive test within 03 minutes. Disaccharides can

Observation :-

Monosaccharide	Faster + Positive result	Red precipitate 3-5 min
Disaccharide	Slower + Positive Result	Red Precipitate after 3-5min

also give this test positive provided they are boiled for sufficient time 3-5 minutes, enough to hydrolyze them in the presence of acidic medium. It is based on the reduction of copper (II) acetate to cuprous (I) oxide (Cu_2O), which forms a brick red-precipitate.

$$RCHO + 2Cu^{2+} + 2H_2O \longrightarrow RCOOH + Cu_2O\downarrow + 4H+$$

Reagents :-

Barfoeds reagent consists of

1. Copper acetate
2. Glacial acetic acid.

Barfoeds' reagent is prepared by dissolving 24 gm copper acetate in 400 ml of reagent, this add 25 ml of 8.5% glacial acetic acid solution. Stirr and cool the solution, add distilled water to make the volume of 500 ml.

Procedure :-

1. Take 2ml of Barfoed's reagent and 2ml of given solution in a test tube.
2. Mix the contents thoroughly. Note the time by your watch.
3. Place the test tube in a boiling water bath for 5 mins.
4. Remove the test tube from boiling water bath and cool under running tap water.

Result :-

1 - The formation of red precipitates in 03 minutes indicates that carbohydrate such as monosaccharide is present.

2 - If red precipitates appear after 3-5 mins, indicates that the carbohydrate such as disaccharide is present.

5. Note the appearance of precipitates, if precipitates donot appedr, put the test tube in the boiling water bath for 15 minutes. And note the appearance of precipitates.

Interpretation :-

- If the red precipitates appear at the bottom of test tube in 03 minutes. It indicates carbohydrate under test is a monosaccharide.
- If the red precipitates appear after 3-5 minutes of heating. It indicates that the carbohydrate under test is a disaccharide.

Precaution:-

- Chloride ions interfere with this test. Therefore, the test should not be carried out without containing chloride ions e.g, urine.