

EXPERIMENT # 06

7-12-2020

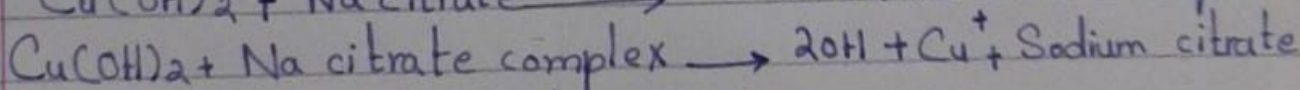
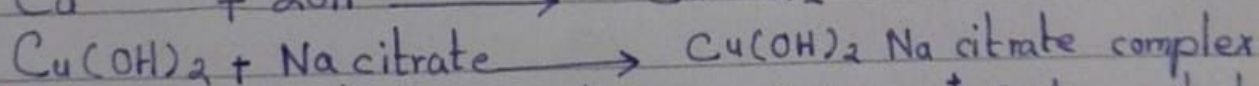
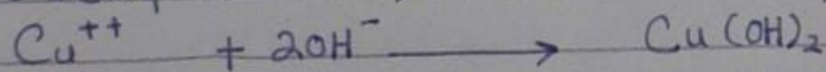
Title:- Test for Reducing Sugars

Benedict's Test (To detect presence of reducing sugars).

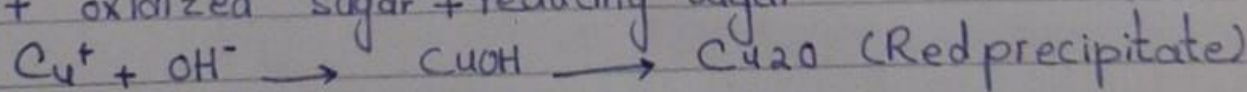
It is a qualitative and semi-qualitative test for reducing carbohydrates. Benedict test is more specific than Fehling's test. Moreover in benedict's test, uric acid and creatinine does not interfere its results.

Principle :-

Benedict's principle is similar to Fehling's test, the reducing sugar can reduce cupric ions to the cuprous ion, which is the basis of Fehling's and Benedict's test. The cupric hydroxide is then reduced to cuprous oxide on heating with reducing carbohydrates.



+ oxidized sugar + reducing sugar



Cuprous hydroxide during the process of heating

is converted to red cuprous oxide, which precipitates immediately. The precipitation of cuprous hydroxide is avoided by sodium citrate. In Benedict's test alkaline medium is provided by sodium carbonate. Note that ultimate quantity cuprous oxide produced at the end of reaction depends upon the amount of reducing sugars present in the sample used. As benedict reagent has blue color the final color is the mixture of blue color of Benedict's reagent and red color of Cu_2O . It may vary from green to brick red depending upon the concentration of reducing sugar. If sample contains the reducing sugar more than 2% the final color of solution will remain red because production of red cuprous oxide will not affect the already present color of solution.

Reagents :-

Benedict's reagent consists of :-

- Copper Sulphate
- Sodium Citrate
- Anhydrous sodium carbonate.

Benedict's reagent is prepared by dissolving 173g of sodium citrate and 90g of anhydrous sodium carbonate in about 750ml of distilled water. Slightly heat to dissolve the content and filter the solution. Dissolve separately 17.3g of CuSO_4 in about 100ml of water and is then

added to the solution of sodium citrate and sodium carbonate with continuous stirring. Finally the volume of solution is made up to 1000 ml with water.

Procedure :-

Take 5ml of benedict reagent in a test tube and add 8 drops of given sample solution. Mix and boil over a flame or in a boiling water bath for 2 minute and cool the solution. Appearance of green, yellow, orange or red precipitate indicates that carbohydrate is reducing one. This is semi-qualitative test. It gives the correct information if the solutions are taken in correct proportion and procedure is followed strictly. The approximate concentration of carbohydrate can be judge from the color of precipitate.

Fallacies :-

The test give false positive test in the presence of some non-carbohydrate reducing agents i.e- ascorbic acids or end products of certain drugs like aspirin.

Clinical Applications :-

This is widely employed test for detection of glucose in urine.

It is commonly used for preliminary screening for hyperglycemia and for monitoring the effect of treatment.

Observation :-

Test type	Precipitate Appears	Time taken
Benedict's Test	Green, yellow orange or red precipitate	2 mins

Results:-

- 1- Reducing sugars are confirmed.
- 2- So, Benedict's test is a reducing sugar identifying test.

Colour of the precipitate	Approx. Conc. of CHO (gm%)
---------------------------	----------------------------

- | | |
|----------------|--------------------|
| 1- Light green | traces |
| 2- Green | (+) or upto 0.5 |
| 3- Yellow | (++) or upto 1 |
| 4- Orange | (+++) or upto 1.5 |
| 5- Red | (++++) or upto 2 |
| 6- Brick red | Above 2 |

Important Point :-

After 2% we cannot estimate the amount of reducing sugar in the given solution that is why this test is called semi-qualitative test.
