

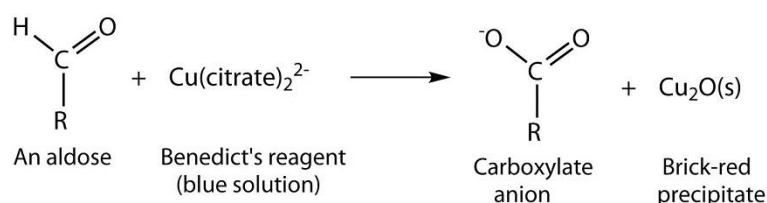
Benedict's Test : Principle, Reagent Preparation, and Procedure

Benedict's test is used as a simple test for reducing sugars. A reducing sugar is a carbohydrate possessing either a free aldehyde or free ketone functional group as part of its molecular structure. This includes all monosaccharides (eg. glucose, fructose, galactose) and many disaccharides, including lactose and maltose.

Benedict's test is most commonly used to test for the presence of glucose in urine. Glucose found to be present in urine is an indication of Diabetes mellitus.

Principle of Benedict's Test

Reducing sugars under alkaline condition tautomerise and form enediols. Enediols are powerful reducing agents. They can reduce cupric ions (Cu^{2+}) to cuprous form (Cu^+), which is responsible for the change in color of the reaction mixture.



This is the basis of Benedict's test. When the conditions are carefully controlled, the colouration developed and the amount of precipitate formed (Cuprous oxide) depends upon the amount of reducing sugars present.

Composition and Preparation of Benedict's reagent

One litre of Benedict's Solution can be prepared from 100 g of anhydrous sodium carbonate, 173 g of sodium citrate and 17.3 g of copper(II) sulfate pentahydrate.

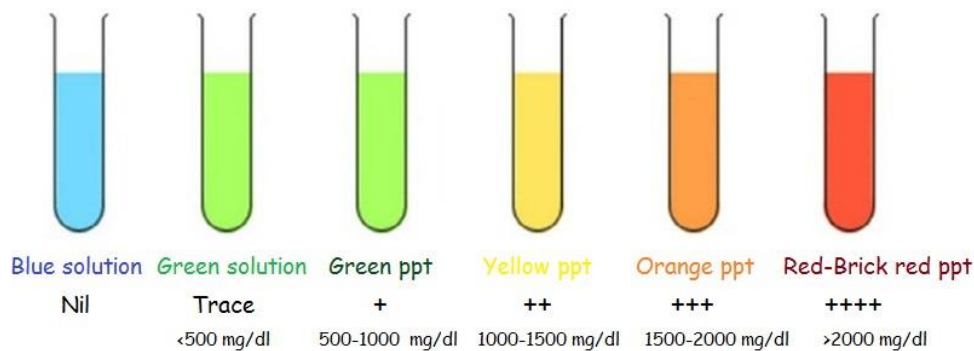
Constituent	Amount	Functions
Copper sulphate	17.3 gm	Furnishes cupric ions (Cu^{++})
Sodium carbonate	100 gm	Makes medium alkaline
Sodium citrate	173 gm	Complexes with the copper (II) ions so that they do not deteriorate to copper(I) ions during storage
Distilled water	Upto 1000 ml	Solvent

Quality Checking : Benedict's solution is blue in color. In order to check purity of Benedict's solution take 5 ml of Benedict's solution in test tube and heat it. If it does not change color, it means it is pure.

Procedure of Benedict's Test

1. Pipette 5 ml of Benedict's reagent in a test tube (20x150mm).
2. Add 8 drops of urine to the Benedict's reagent.
3. Heat carefully on a flame of a gas burner or place in a boiling water for 5-10 minutes.
4. Cool under tap water or by placing in a beaker containing tap water.
5. Observe the color change and precipitate formation and analyse the test result.

Result Interpretation of Benedict's Test



The colour of the mixture serves as a guide to the amount of sugar in the urine. Remove the tubes and examine the solution in each tube for precipitate and change of colour. Report the sugar concentration as follows:

Color	Approximate glucose mg/dl	Indication
Blue solution	Nil	
Green solution	<500 mg/dl	Trace
Green ppt	500-1000 mg/dl	+
Yellow ppt	1000-1500 mg/dl	++
Orange ppt	1500-2000 mg/dl	+++
Red to Brick red ppt	>2000 mg/dl	++++

False positive reactions may also be obtained if certain drugs are present, e.g. salicylates, penicillin, streptomycin, isoniazid, and p-aminosalicylic acid.