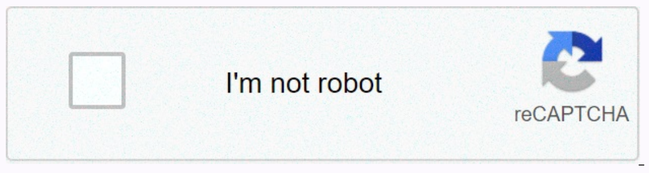

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Qualitative test for carbohydrates pdf

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Quality tests for carbohydrate potato consist of different carbohydrates such as starch, sugar reduction, etc. Difficulties are encountered in the qualitative and quantitative analysis of samples containing mixtures of carbohydrates, in particular sugars, due to their structural and chemical similarity, as well as in relation to their stereoisomers. During biochemical investigations it may be due to the fact that it is necessary to determine whether a given sample, in particular a purified preparation, consists of carbohydrates or not. Several rapid tests are available in the presence or absence of sugar or carbohydrates in a sample. These tests are based on specific color reactions that are typical for their group and are described below. For laboratory practice, it may be recommended to perform these tests with the individual rather than sugar mixture. Use of sugar solution of different concentrations (0.1-1%) during these experiments would also provide valuable information on the sensitivity of these tests. The types of carbohydrates detected by these tests are: Name of test Application Molisch's Test General Test for Carbohydrates Anthone Test General Test for Carbohydrates Iodine Test For Sweets (starch, Glycogen) Test barfoed To distinguish between mono-saccharides by reducing the test of seliwanoff diasaccharides for fehling's ketones Test To reduce sugars Bendict test to reduce sugars Test picric acid To reduce sugars Test of Bial for pennies 1) Principle MOLISCH TEST This is a general test for all carbohydrates. Conc. H2SO4 moisturizes glycoside bonds to produce monosaccharides which in the presence of acid dehydrate to form furfural and its derivatives. These products react with sulphonized a-naphthol to give a purple band. Polysaccharides and glycoproteins also give a Reaction. Reaction reagents 1. Sync this is the first time that we've had H2SO4 2. α-mothball: 5% (w/v) in ethanol (fresh preparation) Process and observations Add 2-3 drops of α-moththalol solution to 2 ml of the test solution. Very soft pipette 1ml conc. H2SO4 along the side if the test tube so that the two distinct layers are formed. Carefully observe any color change at the intersection of two layers. The appearance of the purpose color indicates indicates carbohydrates in the sample preparation or test solution. Precautions 1. a solution of mothballs is unstable and must be prepared fresh. 2. Conc. H2SO4 shall be along the sides of the test tubes causing minimal disturbance to the contents of the tube. 2) Anthrone TEST Principle Anthrone reaction is another general test for carbohydrates. In this furfural produced reacts with anthrony to give the blue green colored complex. Reaction materials and reagents Boiling the water bath. Conc. H2SO4 0.2% (w/v) anthronium solution Process and observations Add 0.5 - 1 ml of the test solution to about 2 ml of anthronium reagent and mix well. Notice if the color changes to blue green. If not, examine the tubes again by keeping them in a boiling water bath for 10 minutes. 3) IODINE TEST Principle Iodine forms colored adsorption assemblies with polysakahaids. Starch gives blue color with iodine, while glycogen reacts to form reddish brown complex. Therefore, it is useful, convenient and rapid testing for the detection of amylase, amylopectin and glycogen. Reagents Iodine solution: Prepare 0.005N iodine solution in 3% potassium iodine solution (w/v). 1% Glucose, sucrose, starch, glycogen, cellulose test solutions, etc. Procedure and observations Take 1 ml of the sample extract or test solution in a test tube. Add 4 - 5 drops of iodine solution to it and mix the contents gently. Notice if a colored product has been formed. Note the color of the product. 4) Barfoed test principle This test is used to distinguish monosaccharides from the reduction of disaccharides. Monosaccharides usually react in about 1 - 2 minutes, while discharides that reduce take much longer between 7 - 12 minutes to hydrolyse and then react with the reagent. Brick red color is taken in this test which is due to the formation of oxide cuprous. Reaction (CH3COO)2Cu2 + H2O ® 2CH3COOH + Cu(OH)2 Cupric Cupric Cupric Hydroxide Cu(OH)2 ® CuO+H Materials and reagents Boiled hydrosyphlegic Reagents Barfoed: Dissolve 13,3 g copper acid in 200 ml of water and add 1,8 ml of glacial acid to it. Procedure and observations Take 2 ml of Barfoed solution in a test tube and add 1 ml of sample solution to it. Keep the test tubes in a boiling water bath. A quick boiling water bath should be used to achieve reliable results. Look for the formation of red brick color and also note the time taken for its appearance. 5) SELIANOFI TEST Principle This test is used to distinguish aldses from ketoses. Ketoses undergo dehydration to give furfural derivatives, which are then with resorkinol to form a red cluster. Prolonged heating will hydrolyse disaccharides and other monosaccharides will also eventually give color. Reaction materials and reagents Boiling water bath The Seliwanoff reagent: 0.05% (w/v) resorkinol in 3 HCl Process and observations Add 1ml of the test solution to 2 ml of Seliwanoff reagent and heat in a boiling water bath for 1 minute. Note for the appearance of a deep red color. Color. means that the sample solution contains keto sugar. 6) The fehling test of the Fehling test principle is a specific and highly sensitive one for the detection of reducting sugars. The formation of yellow or red ppt of cuprous oxide indicates the presence of reducting sugars. Rochelle salt acts as the 30th factor in this reaction. Reaction materials and reagents Boiling the water bath. Fehling A solution: Dissolve 35 g CuSO4.5H2O in water and the volume is at 500 ml. Fehling B solution: Dissolve 120 g KOH and 173 g of Na-K tartaric (Rochelle salt) in water and volume in 500 ml. Fehling reagent: Mix equal volumes of Fehling solution A and B. These solutions should be mixed immediately before use. Procedure and observations Add 1 ml of Fehling reagent (reagent No 4) to 1 ml of the test solution. Mix well and place the test tubes in a strongly boiling water bath. Watch out for the formation of red ppt of cuprous oxide that would indicate the presence of sugar reduction in the solution. 7) Benedict Benedict's test principle of testing is more convenient and this reagent at more stable. In this method sodium citrate acts as a late agent. The presence of reducting sugars results in the formation of red ppt cuprous oxide. Reaction materials and reagents Boiling the water bath. Benedict's reagents: Dissolve 173 g of sodium icrate and 100 g of Anhydrous Na2CO3 in 600 ml of hot H2O. Dilute to 800 ml with water. Dissolve 17.3 g CuSO4.H2O in 100 ml of hot water. Cool and dilute to 100 ml. Slowly add reagent No.2 to reagent No.3 by continuous stirring. The final volume in 1 L. Procedure and observations Add 0,5 - 1 ml of the test solution or sample extract to 2 ml of Benedict reagent (reagent No 4). Keep the test tubes in an intensely boiling water bath. Observe for the formation of red precipitations whose appearance indicates the presence of reducting sugars in the given or sample extract. 8) Picric Acid Testing Authority is another test to detect sugar reduction. Reducting sugars react with picric acid to form a red picramic acid color. Reaction materials and reagents Boiling the water bath. Saturated bitter acid: Dissolve 13 g picric acid in distilled water, boil and cool. 10% Na2CO3. Procedure and observations Add 1 ml of saturated picric acid to 1 ml of sample solution followed by 0.5 ml 10% Na2CO3. Heat the test tubes in a boiling water bath. The appearance of red indicates the presence of reducting sugars in the sample solution. 9) Trial principle of Bial This test is useful for the determination of pentose sugars. The reaction is due to the formation of furfuralis in the medium condensed with orquinol in the presence of iron ions to give a blue-green color complex which is soluble in butyl alcohol. Reaction materials and reagents Boiling water bath Dissolve 1,5 g orquinol in 100 ml conc. HCl and add 20-30 drops of 10 % iron chloride solution. Procedure and observations In 2 ml of Bial Bial add 4-5 drops of test solution and heat in a boiling water bath. Notice about the formation of the blue-green band color. Complex.