**Experiment: 6**

**Color Test for Ketoses and Pentoses**

**1/Seliwanoff's Resorcinol Test:**

\*This test is used to distinguish between aldoses and ketoses.

\*Ketohexoses form considerably more furfural derivatives (about 20 to 25 times) and at a faster rate than aldohexoses.

\*Resorcinol, condenses with the higher concentration of the furfural derivatives formed from the ketohexoses but not with the lower concentration formed from the aldohexoses. Consequently, the resorcinol test can be used to identify ketohexoses.

\*At very high concentrations the aldohexoses will give positive test.

\*Also, upon continued boiling, aldoses will give a red color with resorcinol reagent because of their gradual conversion to ketoses by the HCL.

\*This test can be used to detect the presence of fructose (example of hexose) in either free or combined state. Sucrose give a positive test. Pentoses react in this test to give a blue to green product.

\*Due to the interference of glucose, it must not be present in amounts greater than 2%. Concentration of HCL must not be more than 12%.

\*The reaction (red color) and the precipitate must be observed after not more than 26-30 seconds boiling.

Ketose + resorcinol red complex

Aldose + resorcinol light yellow to faintly pink color

Pentose + resorcinol blue to green color

**Procedure:**

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| 1%xylose | 1%Sorbose | 1%Sucrose | 1%Glucose | 1%Fructose | Water |
| 2 drops | 2 drops | 2 drops | 2 drops | 2 drops | 2 drops |
| Resorcinol Reagent | | | | | |
| 5 ml | 5 ml | 5 ml | 5 ml | 5 ml | 5 ml |
| Incubate in boiling water bath for 1 min.  See the color and continue incubation to 4 mins. See change in color. | | | | | |

**2/Bial's Orcinol Test:**

\*It is a simple, rapid qualitative test for pentoses.

\*Bial's gives a green color with the furfural from dehydrated pentoses.

\*Bial's reagent consist of: orcinol, HCL and ferric chloride. (Ferric chloride increase the sensitivity of the test).

\*Compounds containing pentoses and uronic acid, give a blue-green product when heated with a strong non-oxidizing acid in the presence of orcinol and FeCL3.

\*It can be used for quantitative assay of pentoses (e.g:Ribonuclic acid) in the absence of interfering substances.

Pentose + heat & HCL furfural

Furfural + orcinol blue-green color

**Procedure:**

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| 1%starch | 1%lactose | 1%fructose | 1%glucose | 1%xylose | water |
| 1 drop | 1 drop | 1 drop | 1 drop | 1 drop | 1 drop |
| Bial's Reagent | | | | | |
| 3 ml | 3 ml | 3 ml | 3 ml | 3 ml | 3 ml |
| Incubate in boiling water bath for 3-5 mins.  Observe the change in color formed | | | | | |

Blue-Green color product = positive

**Iodine Test for Polysaccharide:**

\*Iodine forms colored adsorption complexes with polysaccharides.

\*Starch gives a blue color with iodine, while glycogen gives red-brown colors.

\*This iodine color is due to coordination complex between the helically coiled polysaccharide chains and the iodine centrally located within the helix.

\*The iodine can be removed by extraction with ethanol or by reduction with sodium thiosulphate. So, the iodine is very loosely bound in the complex and that is still in the oxidized state.

Starch + Iodine blue to black color

Dextrin + Iodine red to violet color

Glycogen + Iodine red to brown color

\*Agar and xylan suspended particles will absorb the iodine to give blue to purple colors.

**Procedure:**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **1%cellulose** | **1%dextrin** | **1%glycogen** | **1%starch** | **water** |
| 2 ml | 2 ml | 2 ml | 2 ml | 2 ml |
| **Iodine solution** | | | | |
| 2 drops | 2 drops | 2 drops | 2 drops | 2 drops |
| Note the color developed | | | | |