

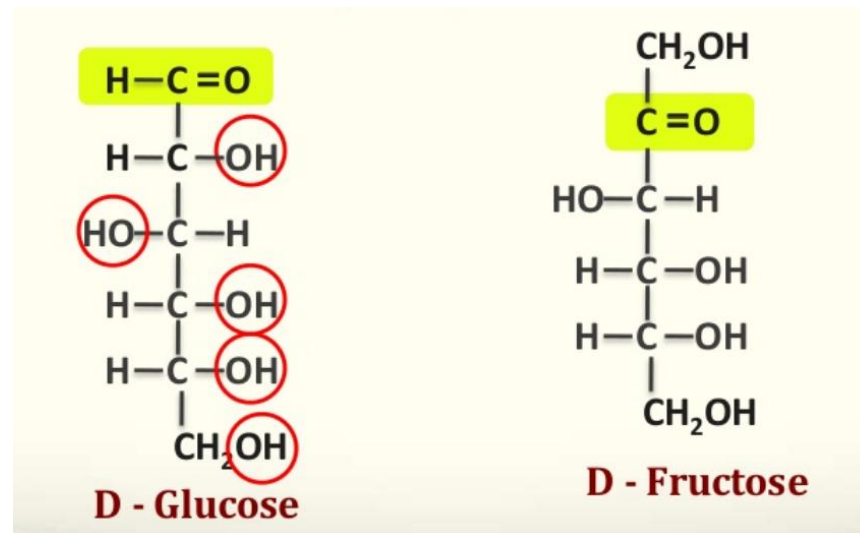
FE 271 FOOD CHEMISTRY LABORATORY

EXPERIMENT 4 - CARBOHYDRATES



CARBOHYDRATES

- Carbohydrates are organic compounds made up of carbon, hydrogen & oxygen
- Carbohydrates are polyhydroxy aldehydes or polyhydroxy ketones and their derivatives
- The word carbohydrates is derived from their general formula $[C(H_2O)]_n$ that makes them seem to be “hydrates of carbon.”
- Carbohydrates occur in many plant & animal tissues as well as microorganisms



Importance of Carbohydrates

- Distributed widely in nature
- Key intermediates of metabolism (sugars)
- Structural components of plants (cellulose)
- Central to materials of industrial products: paper, fibers
- Key component of food sources: sugars, flour, vegetable fiber



Classification of Carbohydrates

| | | | | | |
|-------------------------|--|--------------------------------|--|--------------------------------|------|
| Complexity | Simple Carbohydrates monosaccharides | | Complex Carbohydrates disaccharides, oligosaccharides & polysaccharides | | |
| Size | Tetrose C4 sugars | Pentose C5 sugars | Hexose C6 sugars | Heptose C7 sugars | etc. |
| C=O Function | Aldose sugars having an aldehyde function or an acetal equivalent. Ketose sugars having a ketone function or an acetal equivalent. | | | | |
| Reactivity | Reducing sugars oxidized by Tollens' reagent (or Benedict's or Fehling's reagents). Non-reducing sugars not oxidized by Tollens' or other reagents. | | | | |

Classification of Carbohydrates

| Monosaccharide | | Oligosaccharide | | | Polysaccharide | |
|-------------------------|------------------------|-----------------|----------------|------------------|---------------------|----------------------|
| Functional group | Number of carbon atoms | Di-saccharide | Tri-saccharide | Tetra-saccharide | Homopoly-saccharide | Hetropoly-saccharide |
| Aldoses e.g Glucose | Trioses | Maltose | Raffinose | Stachyose | Starch | Hyaluronic acid |
| | Tetroses | Lactose | | | Dextrin | Heparin |
| | Pentoses | Sucrose | | | Glycogen | Chondroitin sulfate |
| | Hexoses | | | | Cellulose | Dermatan Sulfate |
| Ketoses e.g Fructose | Heptoses | | | Inulin | Keratan Sulfate | |

Monosaccharides are sub divide into different groups

- Depending upon the functional

- ➔ Aldoses (CHO) or

- ➔ Ketoses (C=O)

- Depending upon the number of carbon atoms

- ➔ Trioses (3C)

- ➔ Tetroses (4C)

- ➔ Pentoses (5C)

- ➔ Hexoses (6C) and

- ➔ Heptoses (7C)

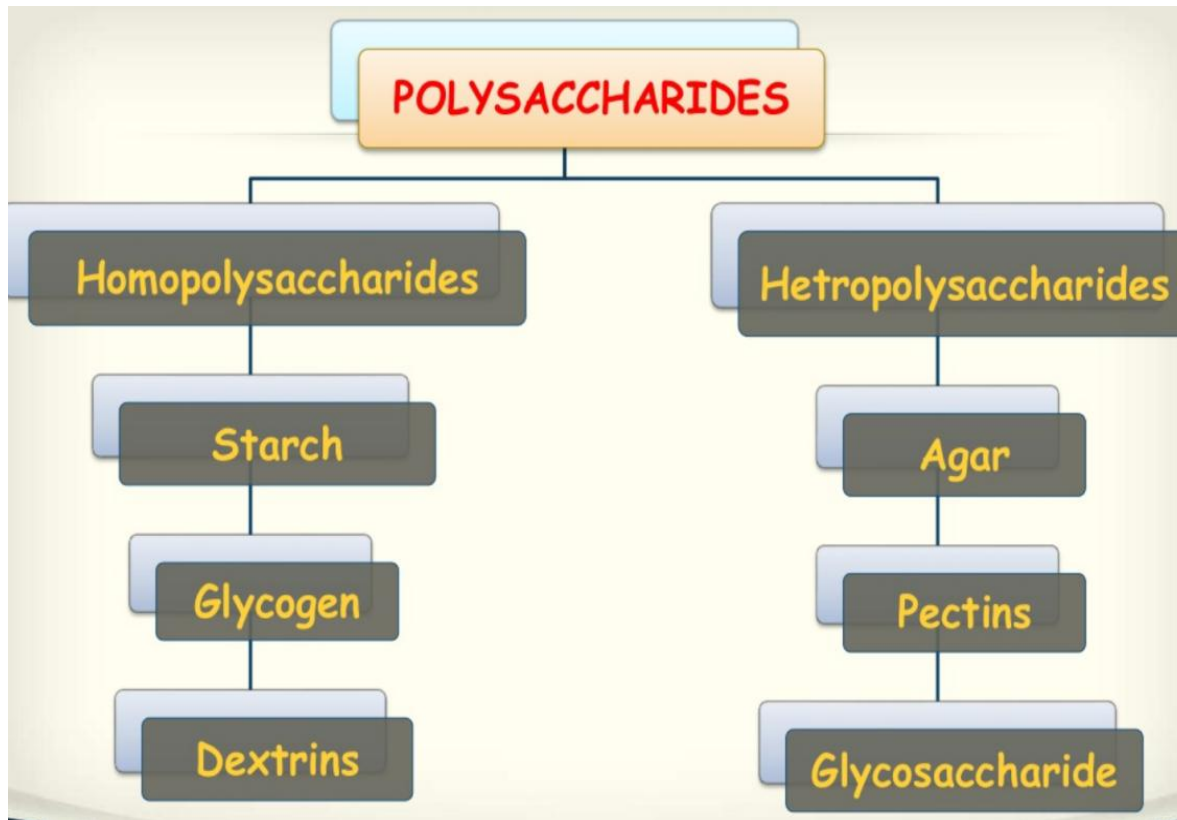
Oligosaccharides

- Contains 2 to 10 monosaccharides
- They are subdivided based on the number of monosaccharide units.

| Classification of Oligosaccharides | | | |
|------------------------------------|--------|------------|----------------------------------|
| | No "C" | Examples | Type of monosaccharide |
| Disaccharides | 2 | Maltose | Glucose + Glucose |
| | | Lactose | Glucose + Galactose |
| | | Sucrose | Glucose + Fructose |
| Trisaccharides | 3 | Raffinose | Glu + Fruc + Galactose |
| Tetra saccharides | 4 | Stachyose | 2 Galactose + Glucose + Fructose |
| Penta saccharides | 5 | Verbascose | 3 Galactose + Glucose + Fructose |

Polysaccharides

- Contains many (more than 10) sugar units



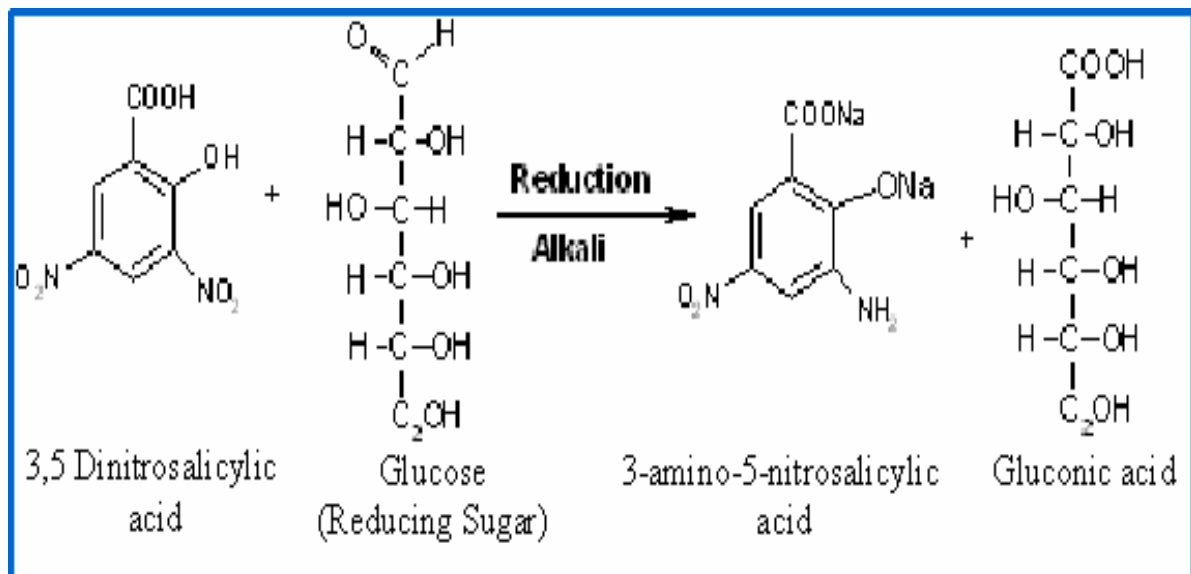
What is the aim of experiment?

- To determine **sugar content** of different food products by using **DNS method**.



PRINCIPLE OF DNS METHOD

- DNS method tests for the presence of free carbonyl group ($C=O$), the so-called **reducing sugars**. This involves the oxidation of the aldehyde functional group present in, for example, glucose and the ketone functional group in fructose.
- When alkaline solution of 3,5-dinitrosalicylic acid (DNS) reacts with reducing sugars, it is reduced to 3-amino-5-nitrosalicylic acid with orange color.



Reagents and Equipments

Reagents:

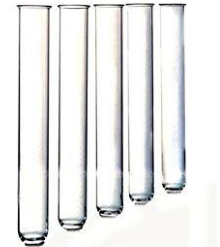
1. Standard Glucose Solution for calibration curve
2. Dinitro salicylic acid reagent
3. HCl (37 %)
4. 5 N KOH

Equipments:

1. Test tubes
2. Pipettes
3. Spectrophotometer capable of measuring absorbance in the 540 nm region.
4. Cuvettes for spectrophotometer
5. Water bath (100°C)



Spectrophotometer



Test tubes



Cuvettes



Water bath

PROCEDURE

1. Prepare the samples in an appropriate dilution.

Tomato paste: Take 0.5 g tomato paste and dilute to 100 mL with distilled water (5000 mg/L).

Jam: Take 0.25 g jam and dilute to 100 mL with distilled water (2500 mg/L).

Peach Juice: Take 1 g peach juice and dilute to 100 mL with distilled water (10000 mg/L).

Cola: Take 1 g cola and dilute to 100 mL with distilled water (10000 mg/L).

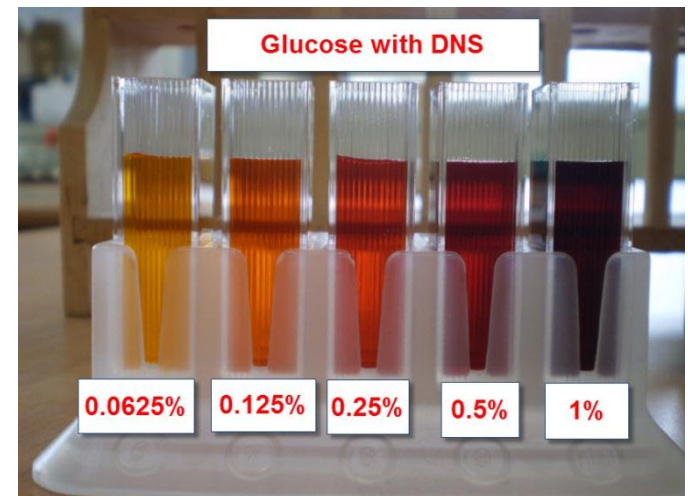
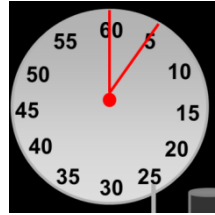
Honey: Take 0.25 g honey and dilute to 100 mL with distilled water (2500 mg/L).

2. Homogenize by mixing.

3. Filter the solution to remove precipitates.

PROCEDURE

4. To each tubes, add 1 ml of sample.
5. Add 1 drop of 37% HCl acid.
6. Put it in the oven for 5 min at 90°C.
7. Add 3 drops of 5N KOH solution to neutralize the acid.
8. Add 3 mL DNS reagent to all the test tubes and mix well
9. Place the tubes in a boiling water for 5 minutes
10. Cool the tubes to room temperature and add 6 ml distilled water
11. Measure the absorbance at 540 nm by using spectrophotometer.

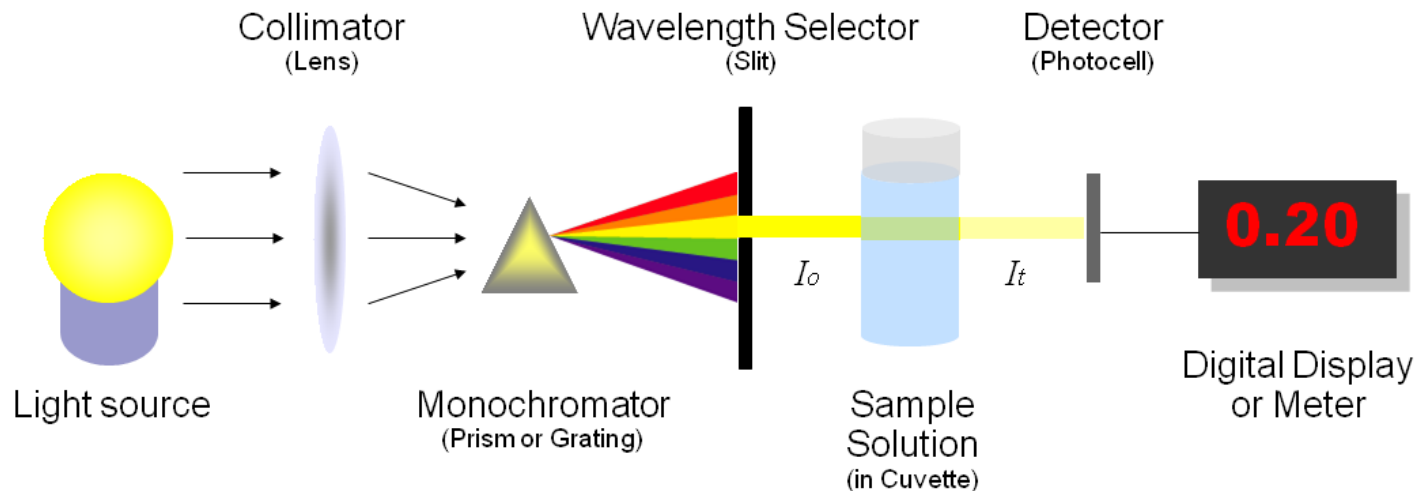


SPECTROPHOTOMETER

Figure illustrates the basic structure of spectrophotometers. It consists of a light source, a collimator, a monochromator, a wavelength selector, a cuvette for sample solution, a photoelectric detector, and a digital display or a meter. Detailed mechanism is described below.

Spectrometer: It produces a desired range of wavelength of light. First a collimator (lens) transmits a straight beam of light (photons) that passes through a monochromator (prism) to split it into several component wavelengths (spectrum). Then a wavelength selector (slit) transmits only the desired wavelengths, as shown in Figure.

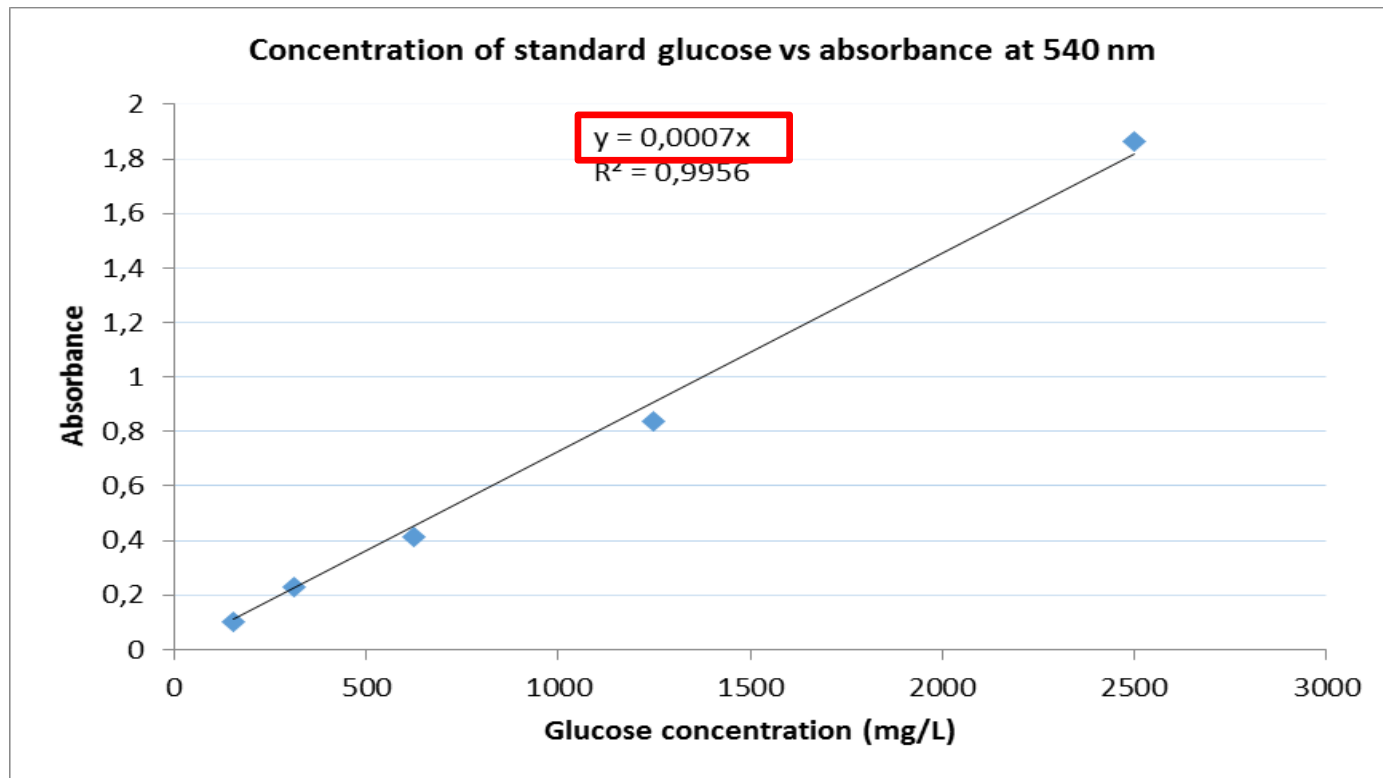
Photometer: After the desired range of wavelength of light passes through the solution of a sample in cuvette, the photometer detects the amount of photons that is absorbed and then sends a signal to a galvanometer or a digital display, as illustrated in Figure.



CALCULATION

Use the already prepared standard curve for the determination of the sugar content of your samples.

The following graph shows standard curve of glucose concentration. Absorbance readings were taken at 540 nm of 5 samples with known glucose concentration. The slope of this graph is used to calculate glucose concentration in unknown samples.



Discussion:

- Give a brief information about your sample (e.g. its composition)
- Compare your result with TSE or Codex. Explain if your result is in the range or not. If it is not in the legal range, explain the possible reasons of different finding.



POST LAB QUESTIONS

1. What is reducing sugar? Give examples for reducing and non-reducing sugars. How can you differentiate reducing and non-reducing sugars?
2. Explain the principle of DNS method with your own words.

