COMPOSITIONAL ANALYSIS METHOD IN POLYSACCHARIDE

SUBMITTED BY-

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INTRODUCTION

- Polysaccharides or complex carbohydrates are generally very large molecular weight molecules also composed of monosaccharide chains.

- Polysaccharide is complicated as they contain variety of bondings.

POLYSACCHARIDE

1. RESISTANCE STARCH POLYSACCHARIDE

2. NONRESISTANCE STARCH POLY SACCHARIDE

1. RESISTANCE STARCH POLYSACCHARIDE

- There are enzyme resistance starch consisting of structure like starch granule physical encloser of starch these resistance starch are analysed by DMSO treatment.

POLYSACCHARIDE

2. NONRESISTANCE STARCH POLYSACCHARIDE

- Sample is treated to remove free suger by enzymetic hydrolysis.

- The uncharged non-starch polysaccharide are precipitated with ethanol (8o % v/v)(liquid to liquid) watched and dry NSP is hydrolysed by two method .

a) Sequencial treatment with dilute acid (non cellulose polysaccharide.

b) 12M H_2SO_4 / 12 M acid (cellulose polysaccharide).

COMPOSITIONAL ANALYSIS METHOD OF POLYSACCHARIDES

POLYSACCHARIDE

PROCEDURE	APPLICATION	LIMITATION	REFERENCE
1. STARCH			
COLORIMETERY	Some serial food.	Need way careful calibration.	Fraser Brandon Bravo & Holmes 1956.
Dilute acid hydrolysis using a general sugar Method.	Highly refined food	Interference from low NSP.	South gat 1997.
Dilute acid hydrolysis and glucose specific Method.	Food low in glucose.	Presence of glucance.	South gat 1997 , DNA in 1978.

COMPOSITIONAL ANALYSIS METHOD OF POLYSACCHARIDES

POLYSACCHARIDE

PROCEDURE	APPLICATION	LIMITATION	REFERENCE
Enzymetic hydrolysis & glucose specific method.	All foods.	Choice of enzyme.	Champ in 1992.
Rapidally digestable starch and slowly digestable starch	Digestable food.	Choice of condition.	Cumming in 1992.

COMPOSITIONAL ANALYSIS METHOD OF POLYSACCHARIDES

POLYSACCHARIDE

PROCEDURE	APPLICATION	LIMITATION	REFERENCE
2. NON STARCH Enzymetic hydrolysis and removel of Starch.	All foods	Acid hydrolysis of NSP, GLC, HPLC seperation of component resistance.	Englust etal 1994.

COMPOSITIONAL METHOD

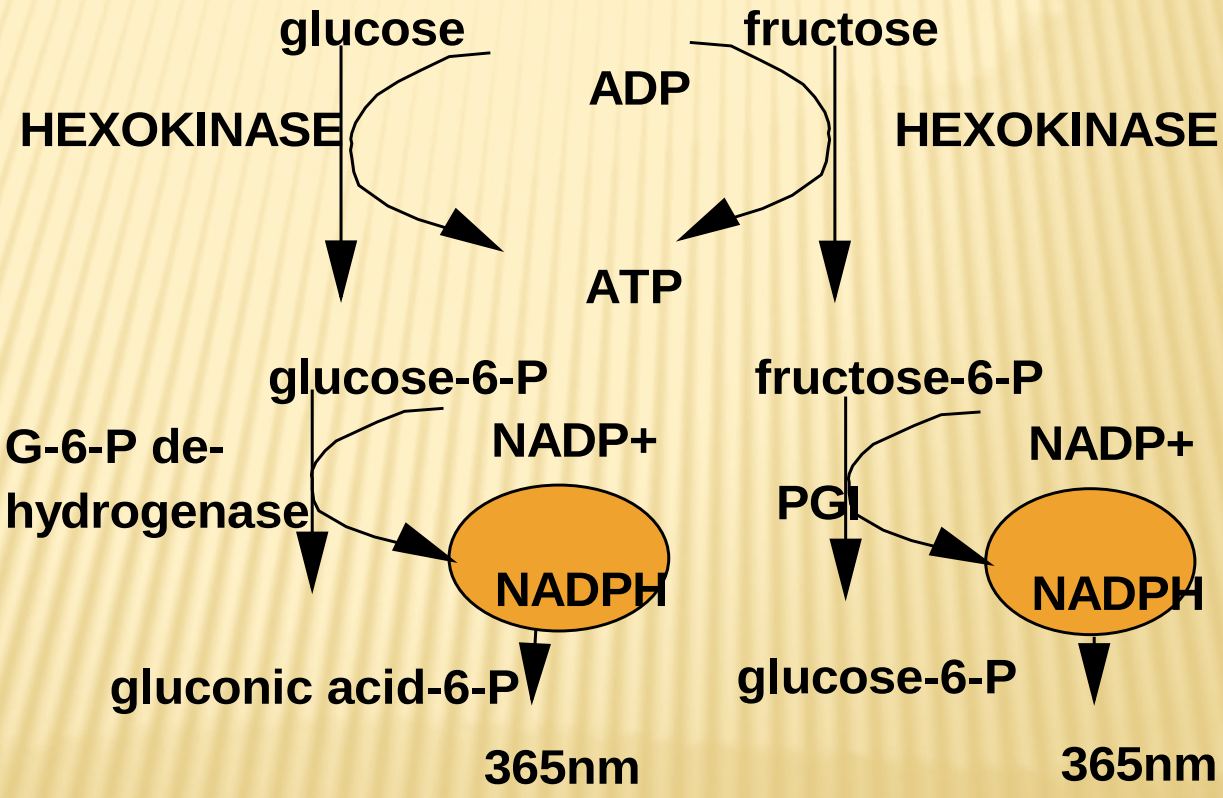
Compositional analysis method

1. Enzymetic treatment method
2. Hydrolysis method
3. Remolval of lipid and protein

1. ENZYMETIC TREATMENT

- Enzymes are large proteins produced by living cells, plants and other organisms.
- All living organisms require enzymes for growth, and for production and utilization of energy.
- Enzymes are biological catalysts

ENZYMATIC DETERMINATION OF STARCH OR OTHER SIMPLE SUGAR



PGI = phosphoglucoisomerase

FIG 1- ENZYMATIC DETERMINATION OF STARCH.

COMPOSITIONAL METHOD

PRINCIPLE

Starch is hydrolyzed into glucose units by enzymatic conversion

D-glucose can then be quantified by enzymatic methods

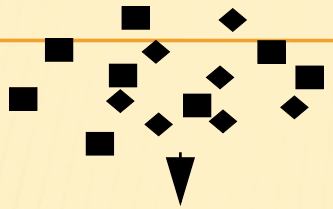
2. HYDROLYSIS METHOD

- Sucrose hydrolysis occurs quite frequently.
- Sucrose inverts or hydrolyzes to form 1 molecule of glucose and 1 of fructose from the heat of processing and natural organic acids.
- Results in changes to sweetness

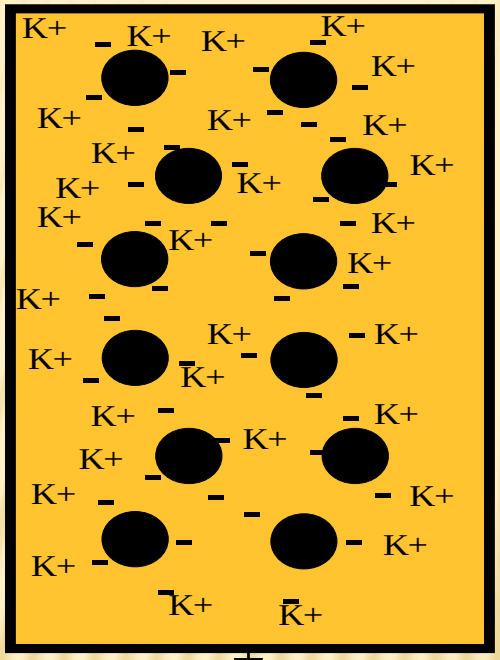
COMPOSITIONAL METHOD

CATION EXCHANGE

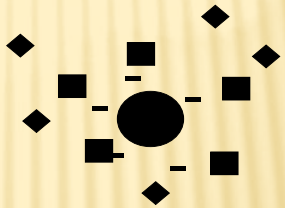
solute molecules



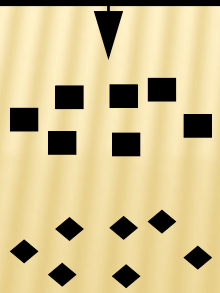
Stationary Phase Contains Anions… "Anion Exchange Resin"



The square molecules have greater affinity for the stationary phase because they are + charged



K+ is the counter ion and is exchanged for the + charged solute molecules



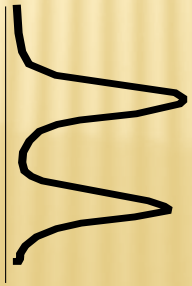


FIG 2- HYDROLYSIS METHOD

Key points:

Stationary phase
Mobile phase
Charges
Ions
Counter-ions
Binding
Elution

3. REMOVAL OF LIPID AND PROTEIN

- Remember that acids result in hydrolysis reactions with some sugars.
- Don't want any changes to the sugar during analysis ie. glucose and fructose suddenly appearing in your sample.
- Nice sample clean-up step, gets rid of "trash" and other charged particles that could interfere with analysis

ANALYTICAL METHOD

ANALYTICAL METHOD IN POLYSACCHARIDE

1. GLC (GAS LIQUID CHROMATOGRAPHY
2. HPLC (HIGH PERFORMANCE LIQUID CHROMATIGRAPHY.)
3. COLORIMETRY

1. GLC (GAS LIQUID CHROMAYTOGRAPHY

A form of chromatography in which the mobile phase is a gas and the stationary phase is a liquid, usually on small beads packed in a long column.

ANALYTICAL METHOD

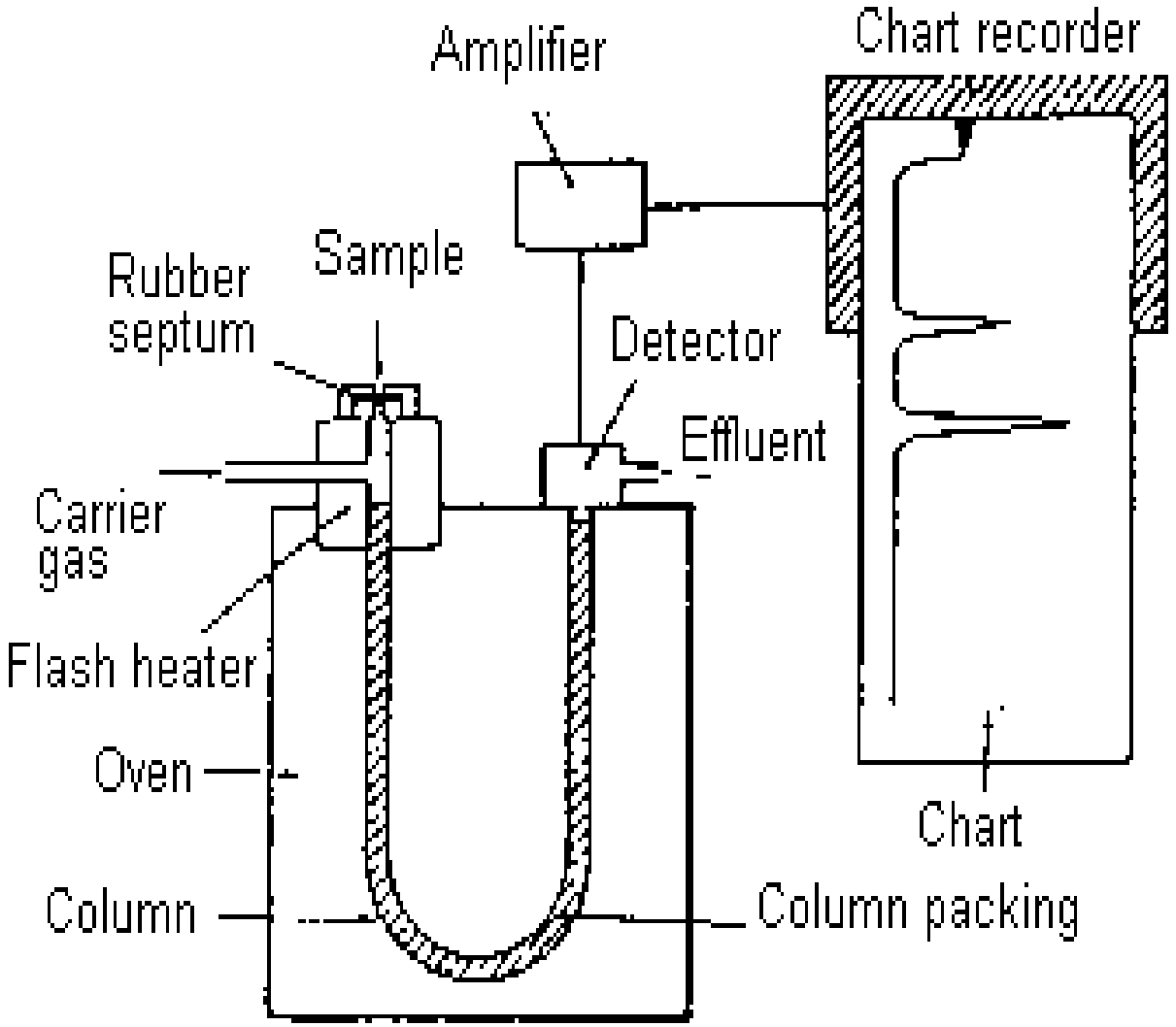


FIG- 3 - GAS LIQUID CHROMATOGRAPHY

ANALYTICAL METHOD

2. HPLC (HIGH PERFORMANCE LIQUID CHROMATOGRAPHY)

- HPLC methods are non-destructive.
- Stationary phase (usually a non-ionic resin).
- Mobile phase is usually 100% water.
- Compounds elute based on size and affinity to stationary phase
  Common sugars:
  - Sucrose
  - Glucose
  - Fructose
  - Maltose
  - Lactose

ANALYTICAL METHOD

HPLC Detectors for CHO Analysis

TYPES OF DETECTORS

- Refractive Index : Measures the changes in refractive index of a solution coming out of and HPLC column .
- Can be applied to many carbohydrates.
- Limitations: It is sensitive to changes in flow, pressure, temperature, and generally requires high CHO concentrations.

Refractive Index Detector

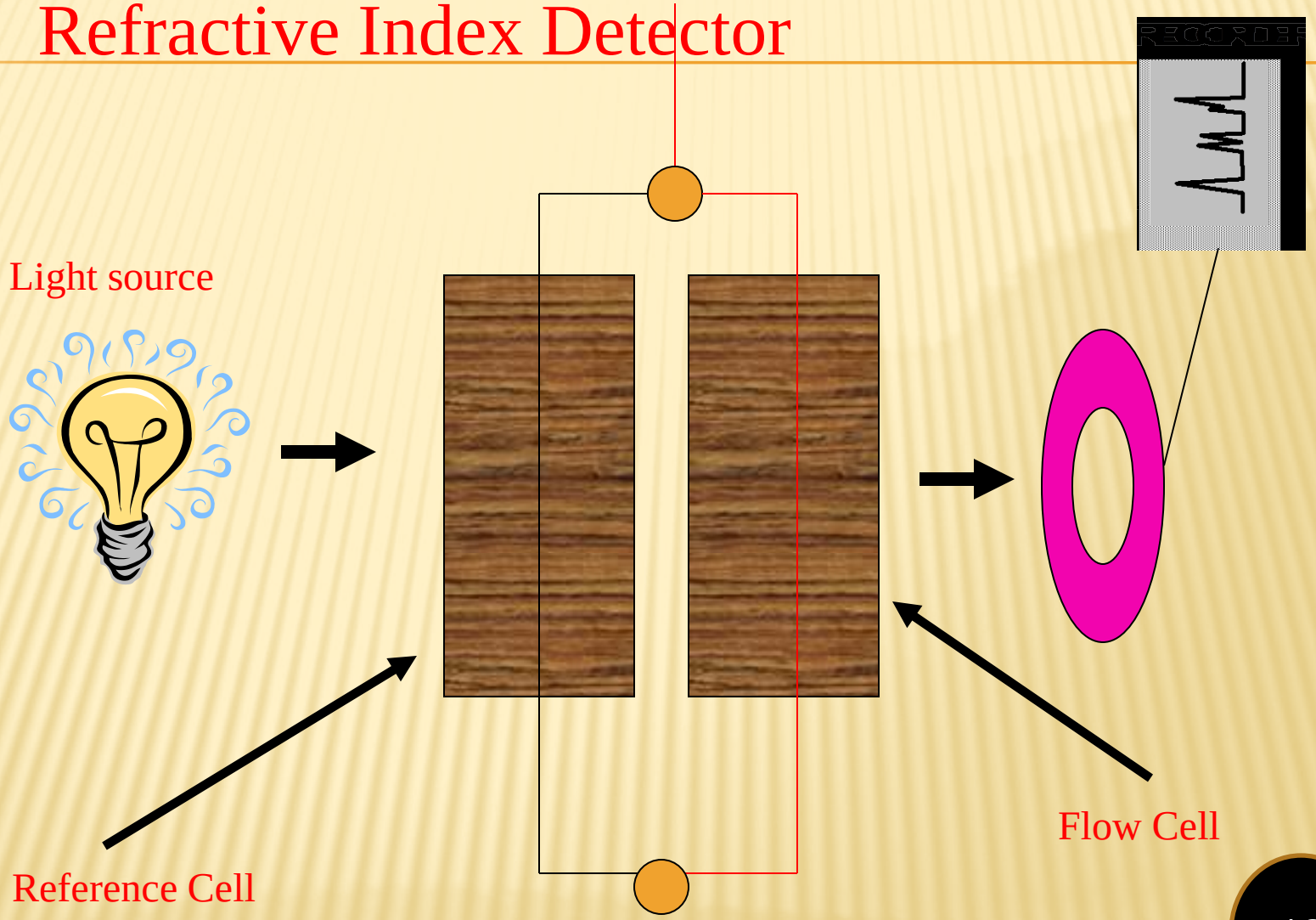


FIG -4- HPLC FROM REFRACTIVE INDEX DETECTOR

ANALYTICAL METHOD

3. COLORIMETRY

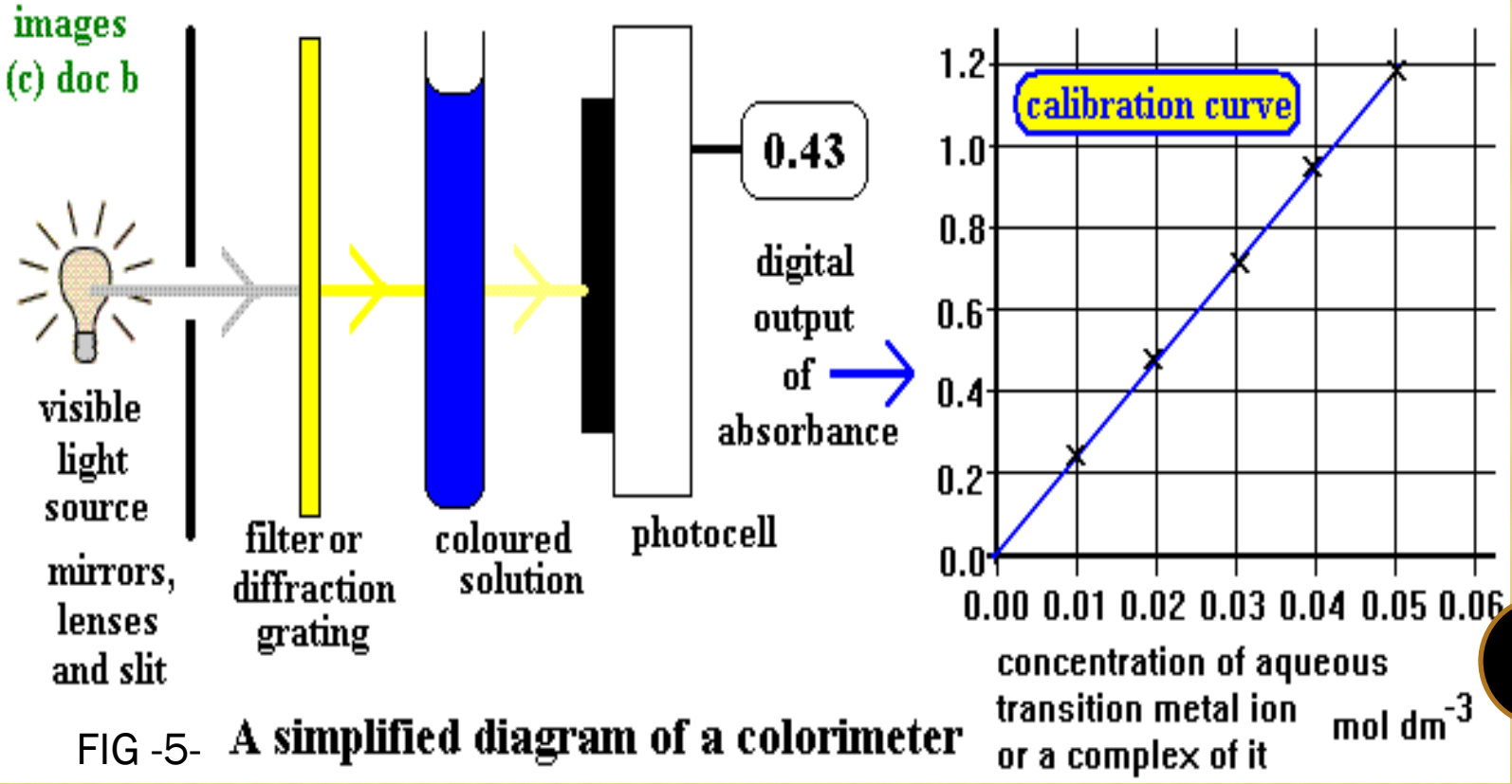


FIG -5- **A simplified diagram of a colorimeter**

SUMMARY

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CONCLUSION

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