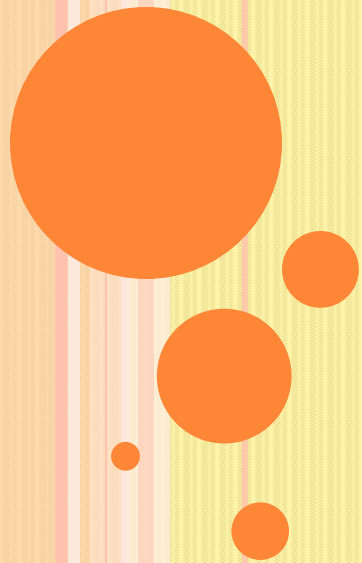




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# **ELECTROPHORESIS**



# INTRODUCTION

Electrophoresis, is a physical method of analysis which involves separation of the compounds that are capable of acquiring electric in conducting electrodes.

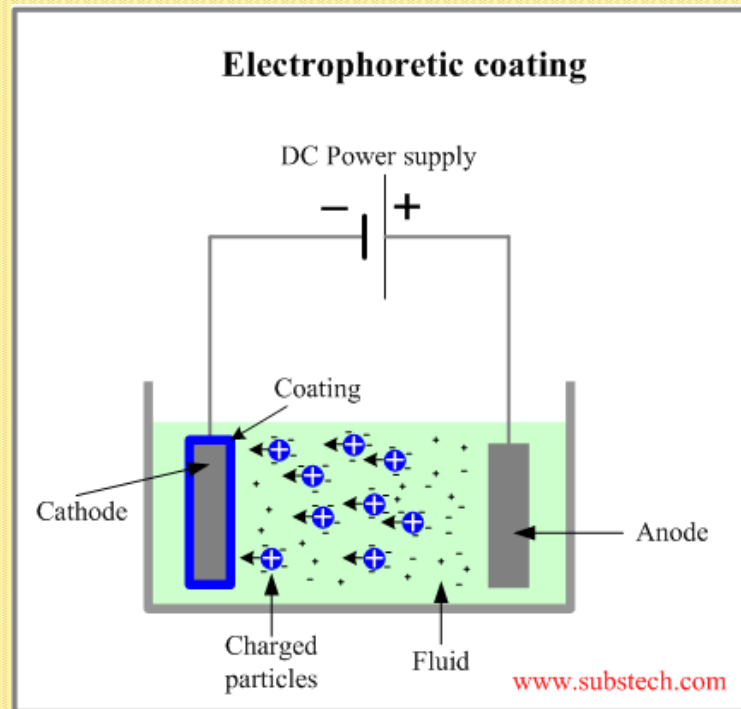
## DEFINITION

Electrophoresis may be defined as the migration of the charged particle through a solution under the influence of an external electrical field.



# PRINCIPLE

When any charged ion or molecules migrate when placed in a electric field the rate of migration depends upon its charge, shape, size and their applied current.



# TYPES OF ELECTROPHORESIS

## ❖ Free Electrophoresis

- \*Micro Electrophoresis

- \*Moving Boundary Electrophoresis

## ❖ Zone Electrophoresis

- \*Paper Electrophoresis

- \*Cellulose Electrophoresis

- \*Gel Electrophoresis

## ❖ Specialized Electrophoresis

- \*Two dimensional electrophoresis

- \*Immuno Electrophoresis

- \*Discontinuous Electrophoresis

- \*Gradient Electrophoresis

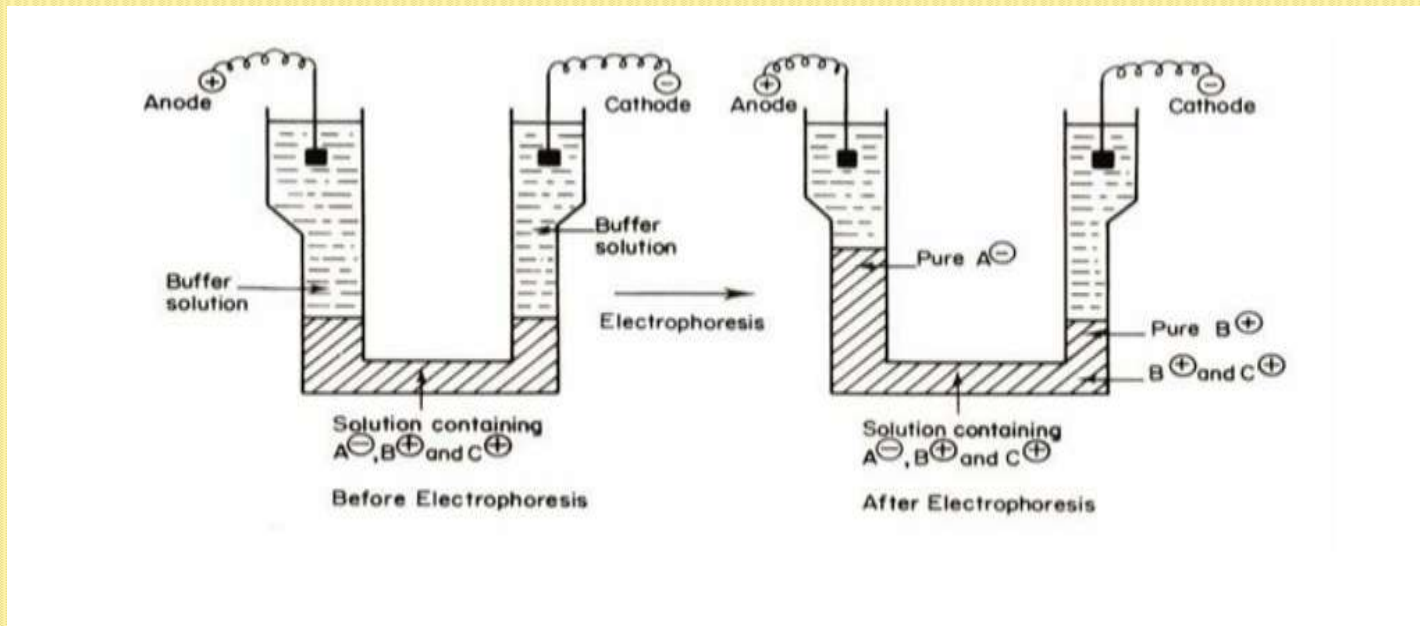


- \*High Voltage Electrophoresis
- \*Pulsed Field gel Electrophoresis
- \*Electrophoresis on cellular gel

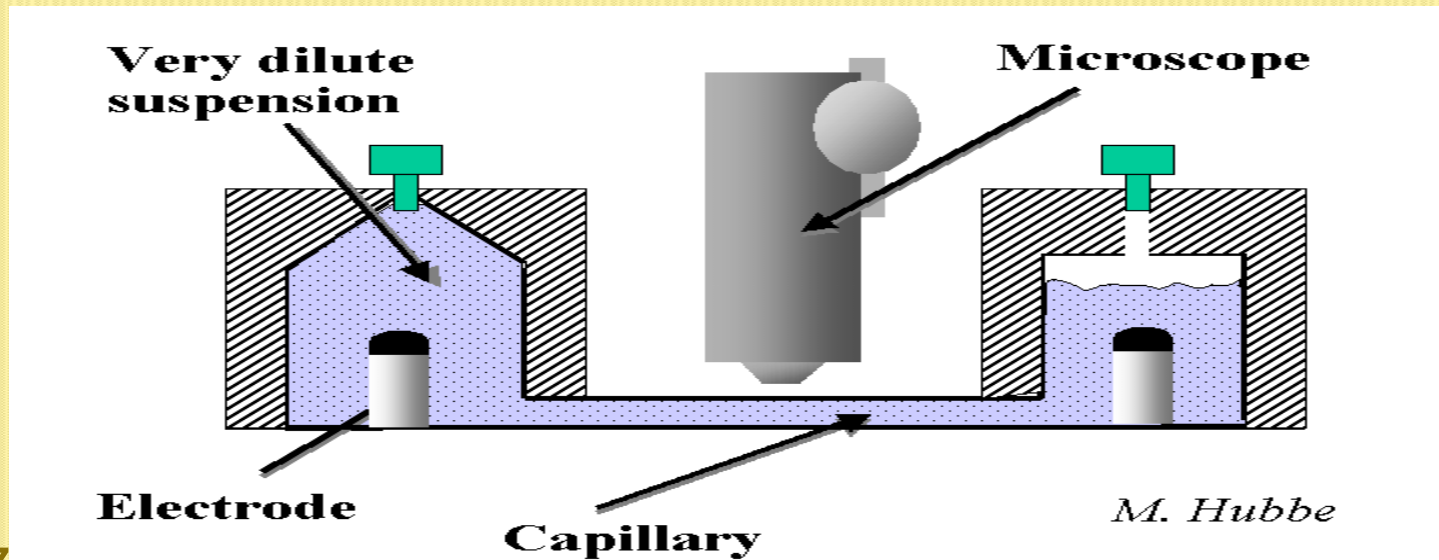
❖ Free Electrophoresis

Movement of small molecules with electric field in a microscope. EG: RBC , WBC, Platelets.

Moving Boundary Electrophoresis



# Micro Electrophoresis



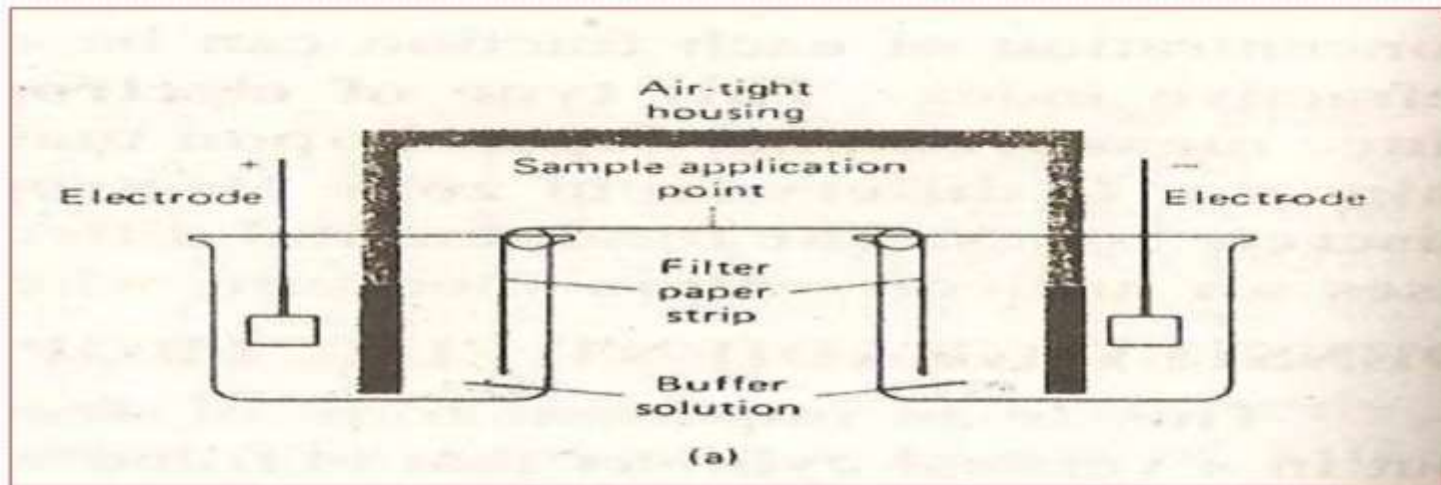
## ❖ Zone electrophoresis

It involves the migration of the charged particle on the supporting media. Paper, Cellulose acetate membrane, Starch gel, Poly acryl amide. Components separated are distributed into discrete zone on the support media.



# Paper Electrophoresis

This technique is useful for the separation of small charged molecules such as amino acids and small proteins. A strip of filter paper is moistened with buffer and the ends of the strip are immersed into buffer reservoirs containing the electrodes.

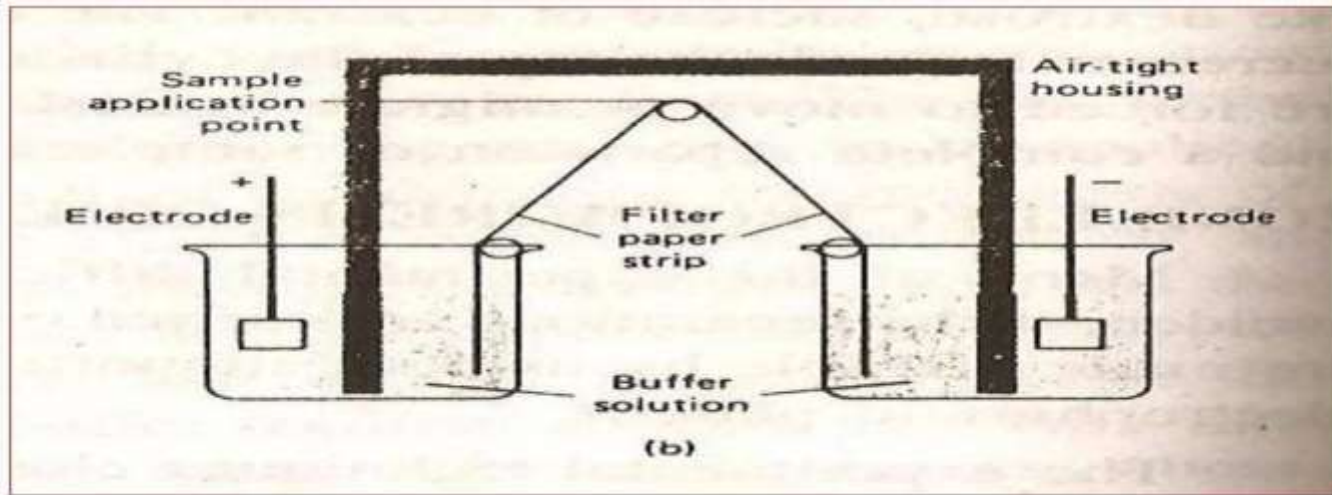


**Principle:** To separate any ionic substances





➤ Vertical paper electrophoresis:



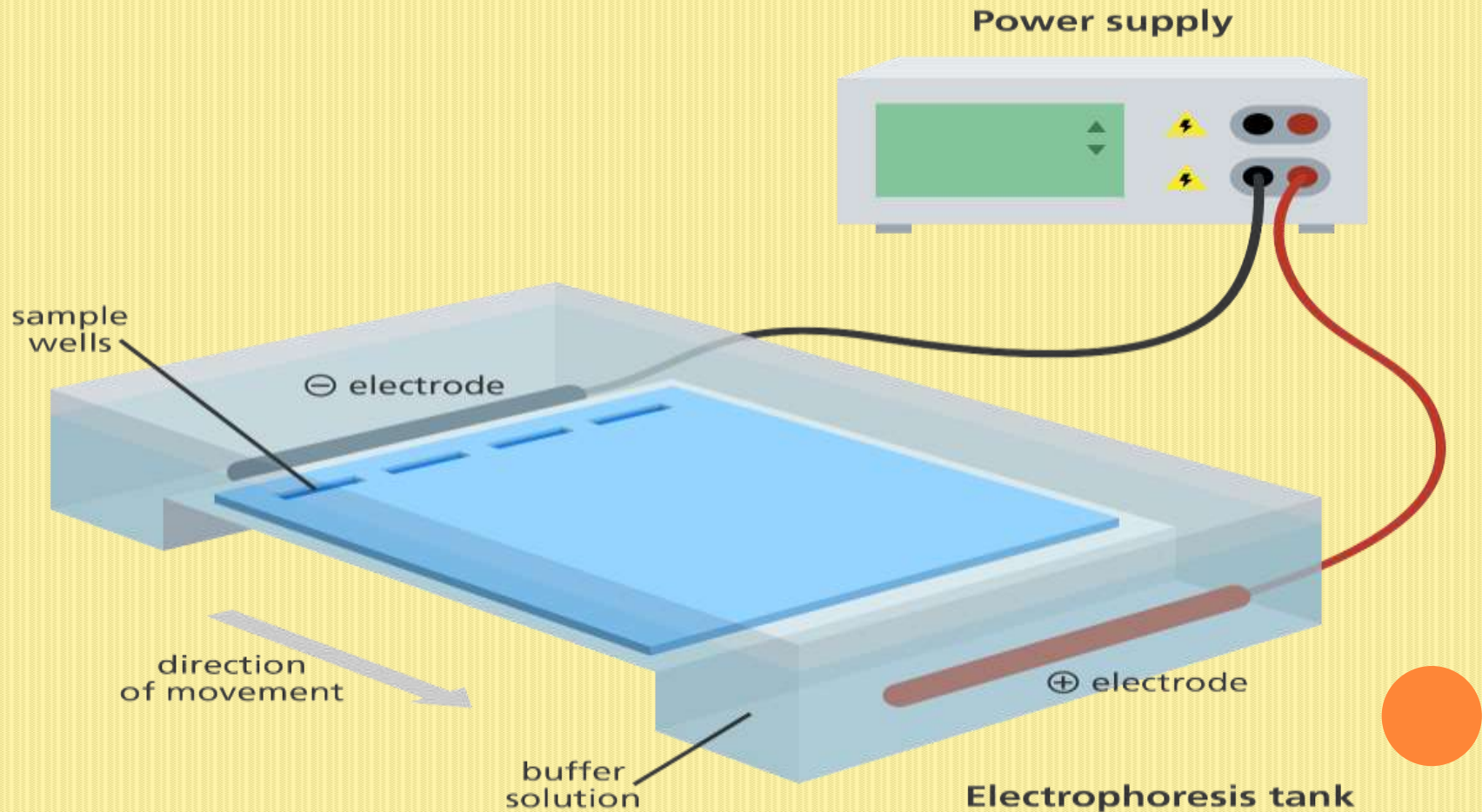
### Cellulose acetate Electrophoresis

cellulose acetate is an acetate salt of cellulose produced by treating cotton with acetic acid using sulphuric acid as a catalyst. Migration take place on the buffer film on the surface of the cellulose plate or membrane.

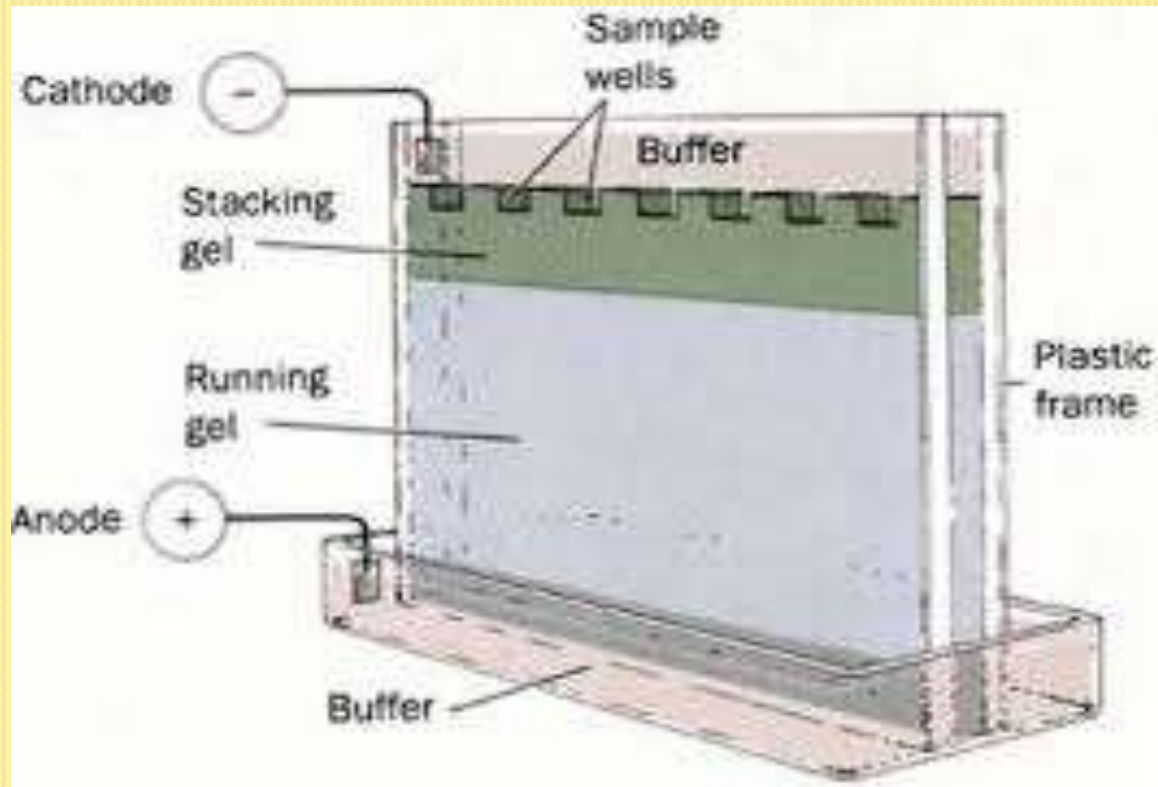


# Gel Electrophoresis

Gel Electrophoresis is a method for separation and analysis of macromolecules and their fragments based on their size and charge.



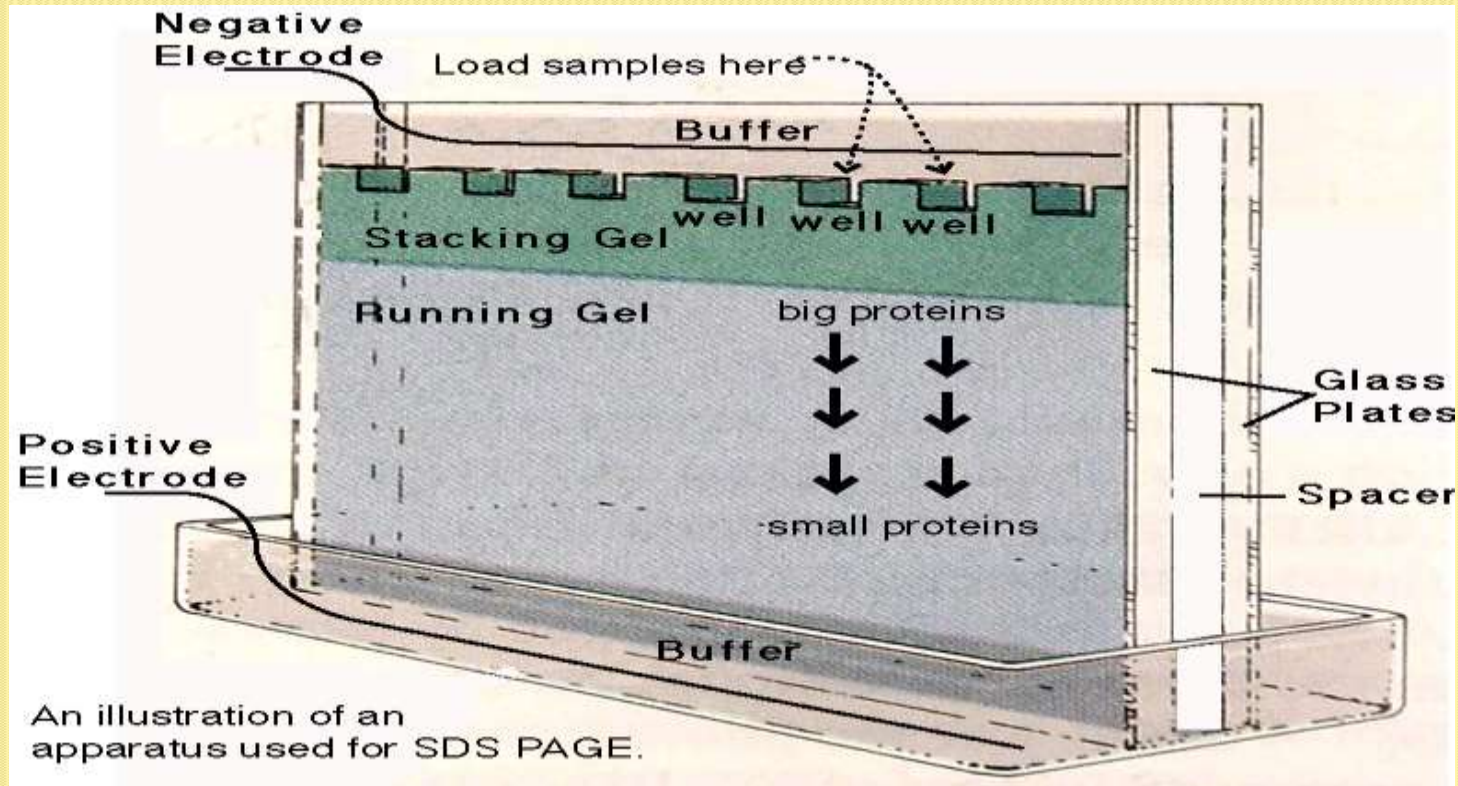
➤ PAGE(Poly Acrylamide Gel Electrophoresis) :



PAGE is a technique widely used in biochemistry, Forensic chemistry, Genetics, Molecular biology and Biotechnology to separate biological macromolecules.



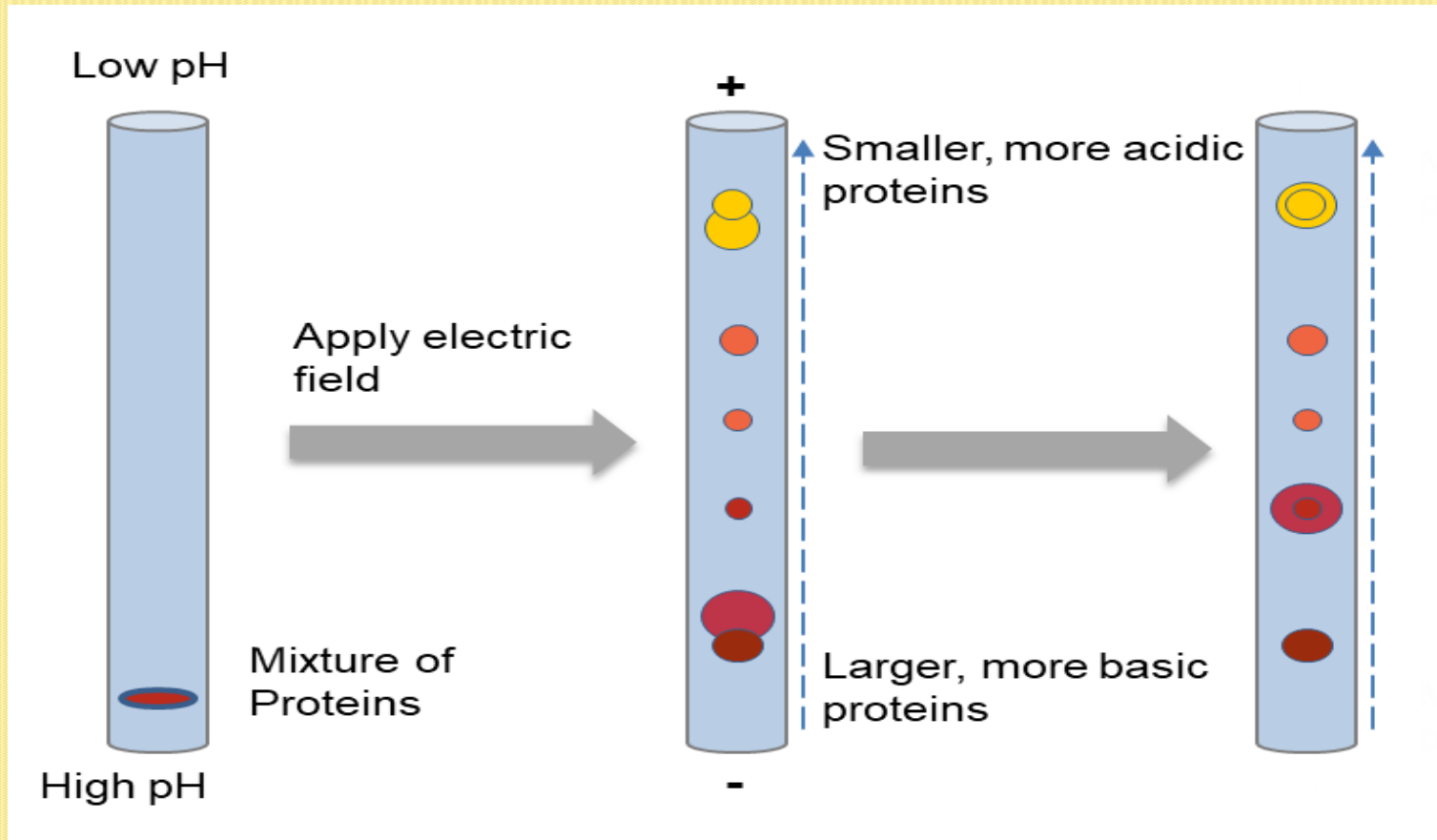
➤ SDS-PAGE Electrophoresis:



SDS-PAGE the use of sodium dodecyl sulfate poly acrylamide gel largely eliminates the influence of the structure and charge and proteins.

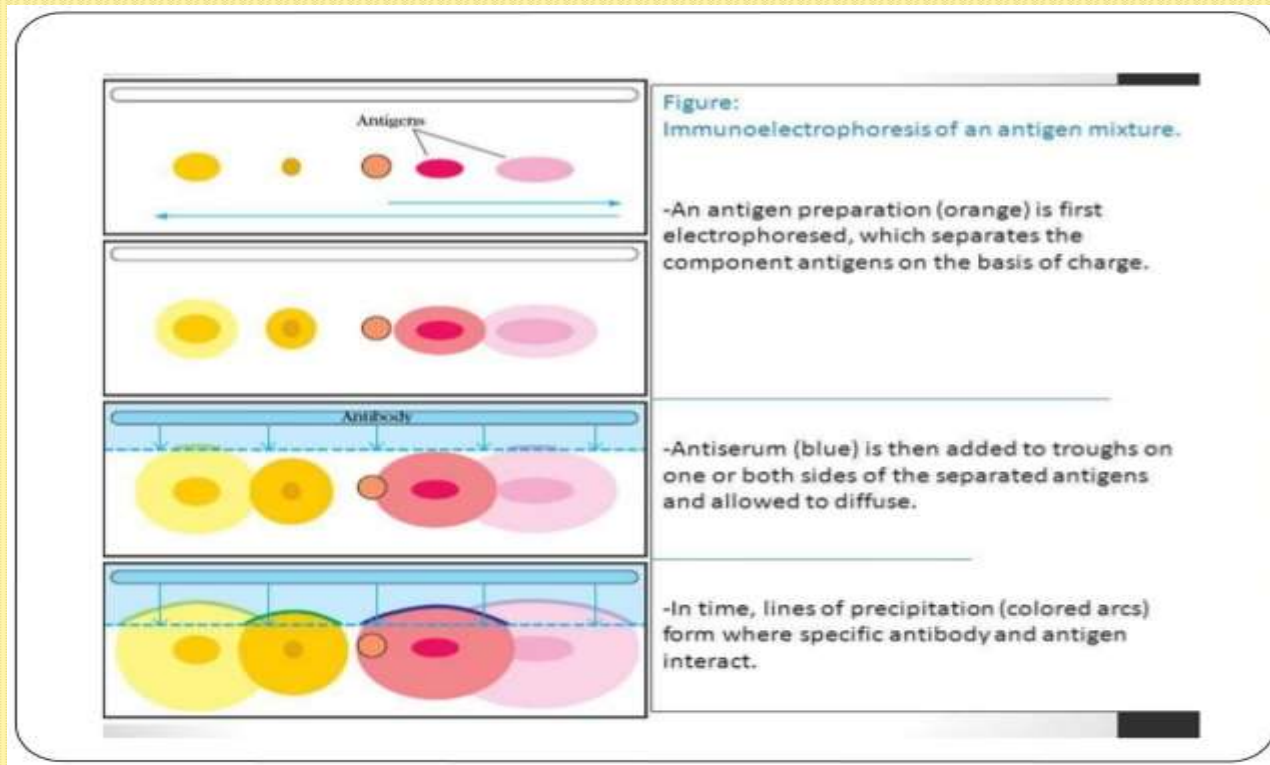


- ❖ Specialized Electrophoresis
- Two dimensional Electrophoresis

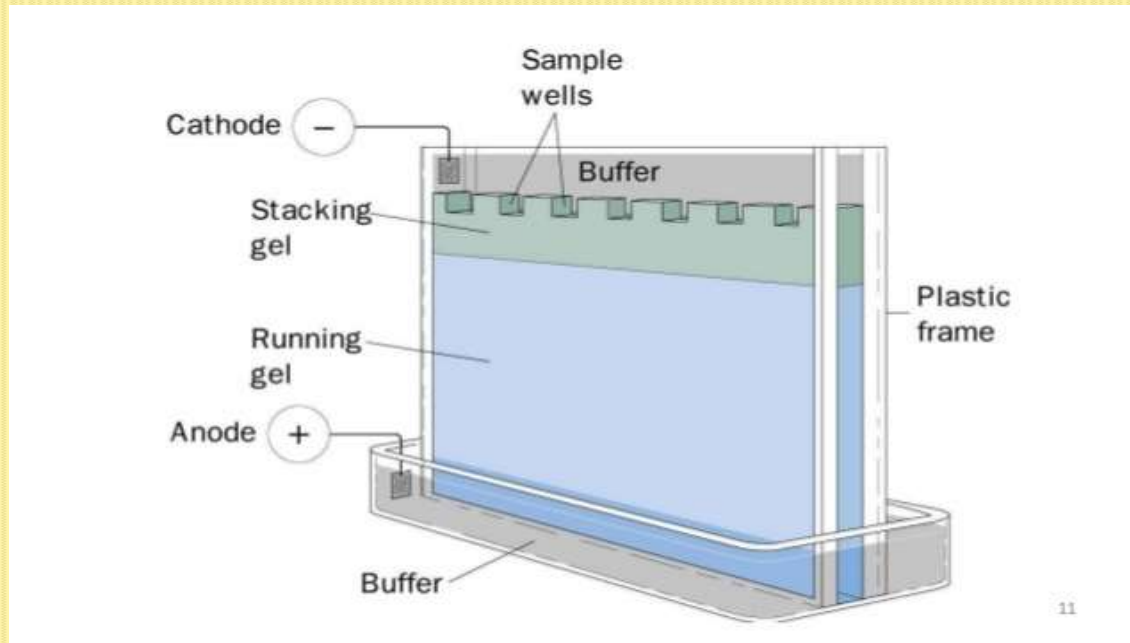


## ➤ Immuno Electrophoresis

immuno Electrophoresis refer to precipitation agar under an electric field. It is a process of combination of immuno-diffusion and electrophoresis. An antigen mixture is first separated into component part by electrophoresis and then tested by double immuno-diffusion.



## ➤ Discontinuous disc gel electrophoresis



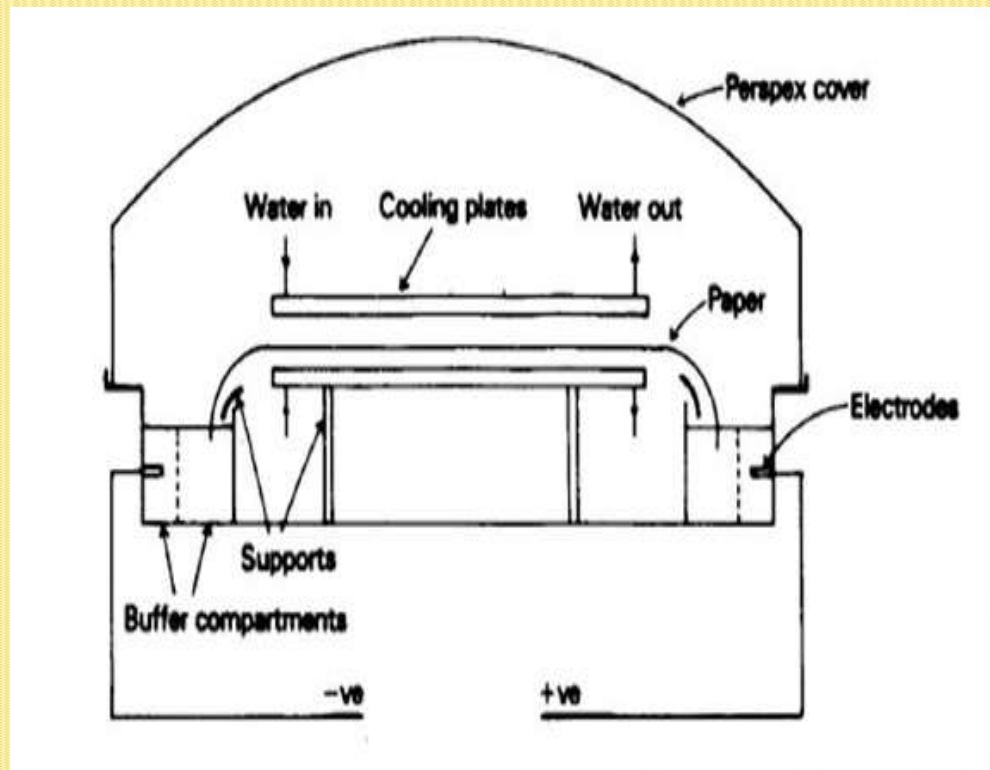
## ➤ Gradient Gel Electrophoresis

It is migrate macromolecule in electric field .



## ➤ High voltage electrophoresis

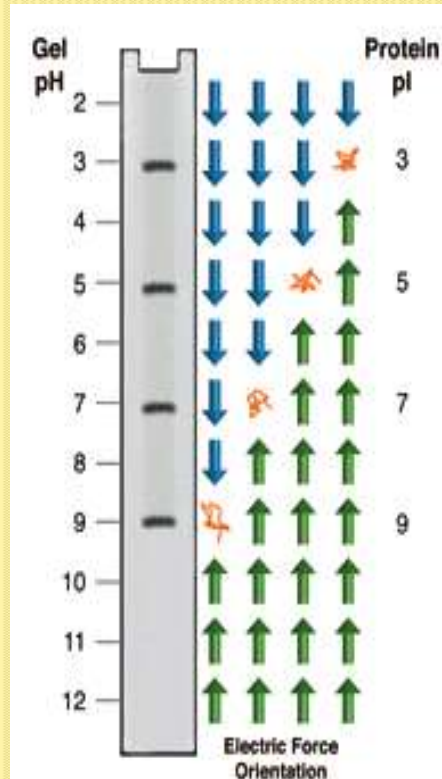
The separation of low molecular weight such as amino acids, peptides, sugar, purines , Pyrimidines, amine, Inorganic ions.



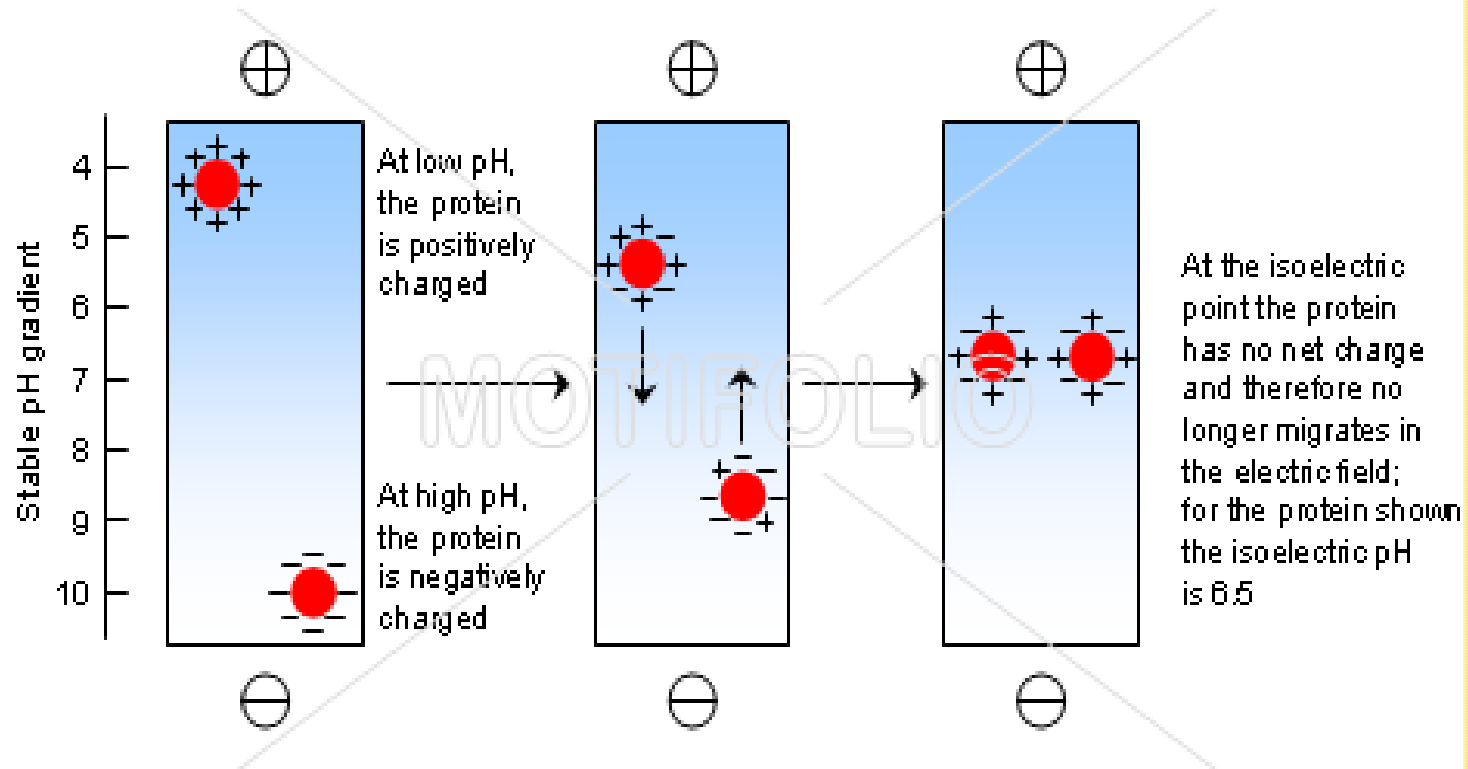


## ➤ Isoelectric Focussing

It is a technique for separating different molecules by differences in their isoelectric point (pI). It is a type of zone electrophoresis usually performed on proteins in a gel that takes advantage of fact that overall charge on the molecule of interest is a function of its surroundings.



# Separation of Protein Molecules by Isoelectric Focussing

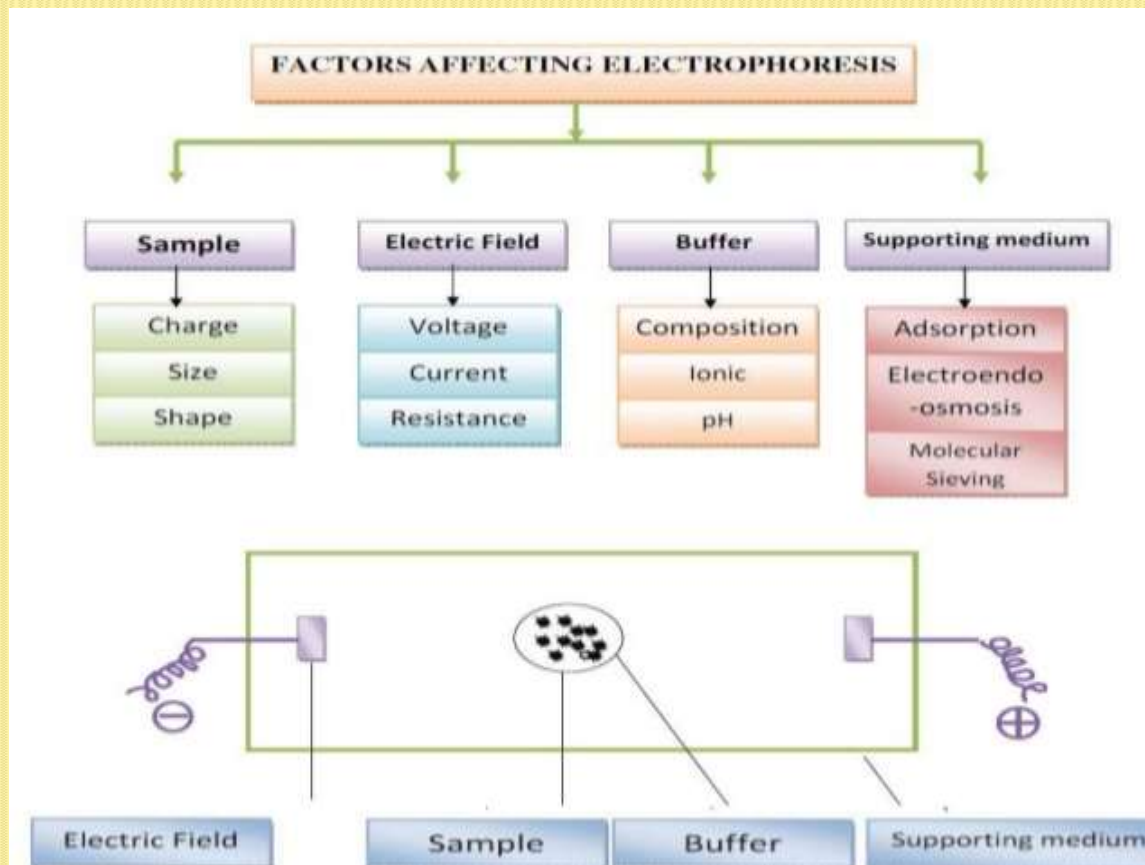


- Advantages of isoelectric focussing:
  - \*Versatility in Identification.
  - \*Accuracy of Results.
  - \*Low cost to use.
  
- Disadvantage of isoelectric focussing:
  - \*Toxicity.
  - \*Electrophoresis has limited sample analysis.
  - \*Electrophoresis measurement are not precise.
  - \*Only certain molecules can be visualised.



# FACTORS AFFECTING ELECTROPHORESIS MIGRATION

- It includes characteristics of the ion or molecule itself, the environment (buffer) in which the molecule or ions are being studied, and the applied electrical field.



# APPLICATION OF ELECTROPHORESIS

- ❑ DNA sequencing.
- ❑ Medical Research.
- ❑ Protein Research/Purification.
- ❑ Agricultural testing.
- ❑ Separation of organic acid, alkaloids, carbohydrates, amino acids, alcohols, phenols, nucleic acids, insulin.
- ❑ In food industry.



Thank  
& You

