# **Paper Electrophoresis**

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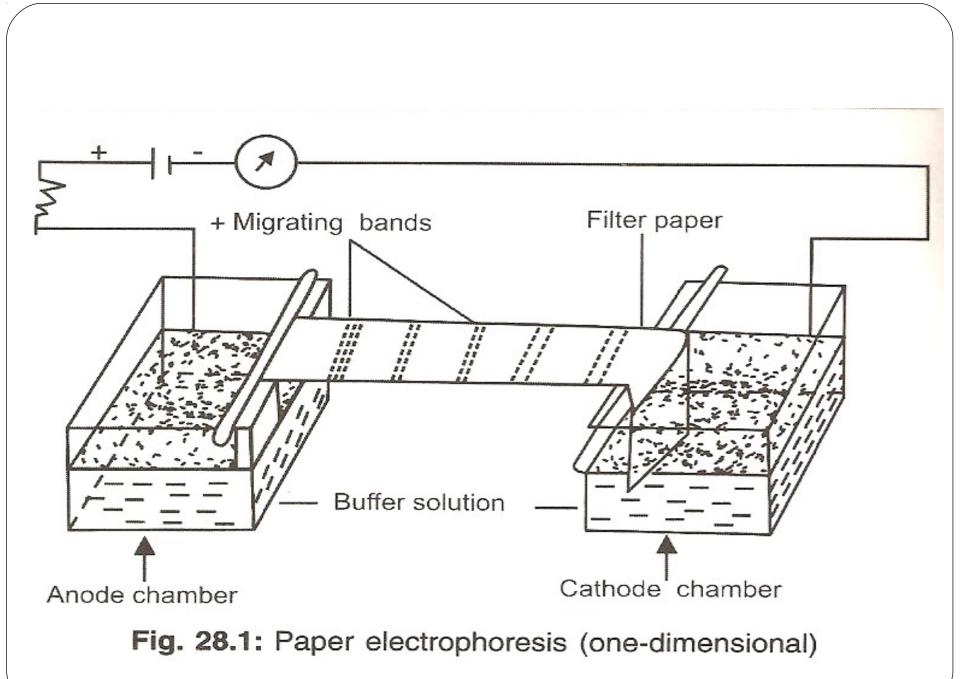
### Introduction

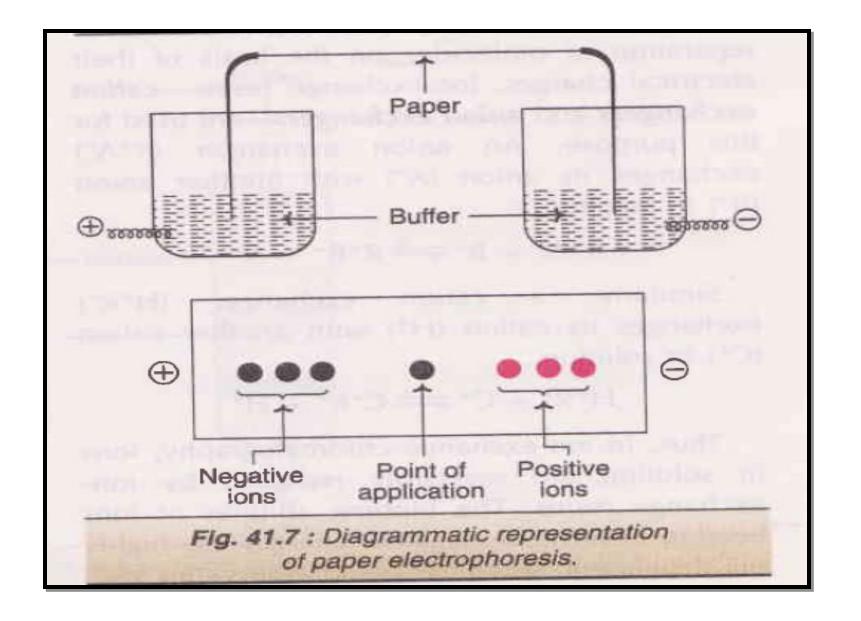
The technique of paper electrophoresis is simple and inexpensive and requires only micro quantities of plasma for separation.

#### \*The support medium is a filter paper

The electrophoresis apparatus in its simplest form consists of two troughs to contain buffer solution, through which electric current is passed.

Frequently used in isolating proteins, amino acids and oligopeptides.





#### Procedure

- 1) A long strip of filter paper is moistened with a suitable buffer solution of the desired p H and the sample is applied transversely across the central part of the strip.
- 2) Ends are fixed to dip in buffer solutions in two troughs fitted with electrodes.
- 3)Electric field of about 20 volts/cm is established.
- 4)The charged particles of sample migrate along the strip towards respective electrodes of opposite polarity, according to net charges, sizes and interactions with the solid matrix.

## Contd...

- 5)Homogeneous group of particles migrate as a separate band
- 6)The electrophoresis is carried out for 16-18 hours.
- 7) Separated Proteins are fixed to a solid support using a fixative such as Acetone or Methanol
- 8)Proteins are stained (**bromophenol blue**) to make them visible
- 9) The separated proteins appear as distinct bands10) Drawback-long time interval and blurring of margins

#### Observation

✤ The different fractions appear as blue coloured bands across the filter paper starting from the moving boundary backwards.

✤ If a quantitative estimation is required for each fraction, the bands may be carefully cut and eluted, or the bands may be scanned optically in a densitometer.

✤ In human plasma five different bands can be identified on paper electrophoresis

