PAPER CHROMATOGRAPHY

- Chromatography is the technique For the separation of a mixture. This mixture is dissolved In a fluid called mobile phase, which carries it through a system On which is fixed A material called stationary phase.
- This technique is a type of partition chromatography in which the substances are distributed between two liquids, i.e., one is the stationary liquid (usually water) which is held in the fibers of the paper and called the stationary phase, the other is the moving liquid is the moving liquid or developing solvent and called the moving phase.
- Stationary phase : Network of cellulosic fibres (Paper)
- Mobile phase : liquid solvent (eg. Acetone, methanol etc.)

Material required:

- Fresh leaves of any plant (spineech)
- Whatman filter paper no. 1 as chromatographic paper
- Petroleaum ether, absolute Ethyl alcohol, acetone
- Test tubes, capillary tube, measuring cylinders, funnels,
- pestle-mortar, pencil, scale
- Chromatographic chamber etc.

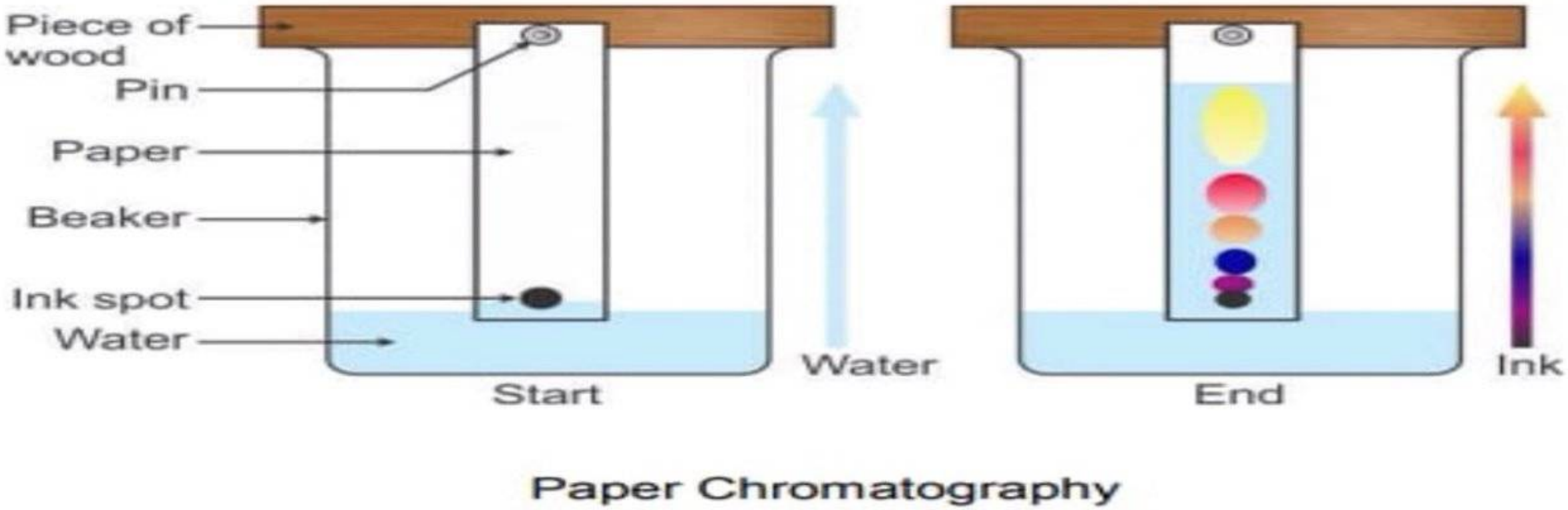
Method :

- Grind freshly harvested spineech leaves in ethyl alcohol in pestle-mortar.
- Take a chromatographic paper and draw a line with pencil above 1 cm from one end.
- Put a drop of plant extract on that line by capillary tube and allow it to dry. Repeat this step at least 30 times until the spot saturates.
- Fill the solvent petroleum ether acetone (9:1) in the jar and hang the paper strip into the jar.
- The bottom of the strip touches the solvent but should not exceed over the pencil line marked on it.
- Wait and watch the movement of solvent on the paper strip untill the solvent reaches to the top of the paper strip.

OBSERVATIONS

After 15 minutes the solvent reaches closer to the top of the paper strip with a band of pigments molecules travelled from bottom to the top along with the solvent and get separated from each other based on their molecular weights (relative density). Lighter molecules of carotenes show orange-red colour and move to the top (lighter in weight as compared to others), second to it is xanthophylls (yellow), third chlorophyll 'a' (blue green) and chlorophyll 'b' (yellow green), Determine the Rf value by following formula :

R_f = Distance(cm) travelled by solid form the base line/
Distance(cm) travelled by solvent from the base line



Paper Chromatography

CONCLUSIONS

Carotene pigments have the least molecular weight as compared with other pigments hence travelled much faster and accumulated on the top most position on the chromatogram followed by xanthophylls, chlorophyll 'a' and chlorophyll 'b'.

APPLICATIONS

- Paper chromatography is specially used for separation of mixture having polar and non polar compounds.
- For separation of amino acids.
- It is used to determine organic compound biochemical in urine etc.
- Some time used for evolution of inorganic compound like salt and complex.