



Practical Experiment Instruction Sheet

EXPERIMENT TITLE : To determine Rf value & identify Phenyl Alanin and glycine mixture by paper chromatography

EXPERIMENT NO. :10 MIT(T)/BSH/Engg. Chemistry Lab/ Engg. Chemistry /Manual No

Class: F.Y. BTech.

DEPARTMENT: Basic Sciences & Humanity

LABORATORY : Engg. Chemistry

Location:-

PART:

PAGE:

Aim:- To determine Rf value & identify Phenyl Alanin and glycine mixture by paper chromatography

Apparatus:- Blotting paper / filter paper, beaker, pencil, clip, scissor, rod

Chemicals : Solvent (water, acetone, alcohol) & ink mixture, Phenyl alanin, glycine, etc.

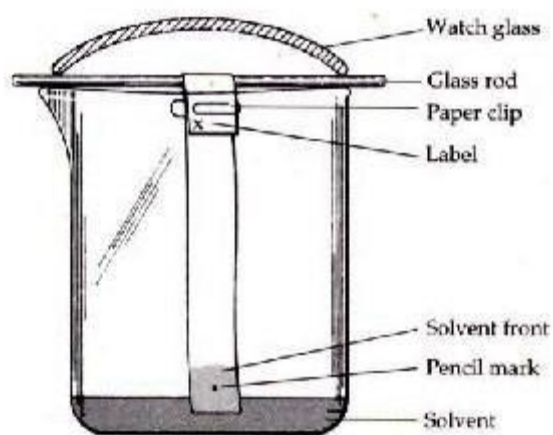
Theory:-

Chromatography is a analytical method of separating a mixture of components into individual components through equilibrium distribution between two phases paper chromatography is one of the types of chromatography procedures which runs on a piece of specialized paper. It is a planer chromatography system where in a filter paper acts as a stationary phase on which the separation of compounds occurs. The principle involved is partition chromatography where in the substances are distributed between liquid phases one phase is the water (solvent) which is held in the pores of the filter paper used & other is the mobile phase over the paper. The compounds in the mixture get separated due to difference in their affinity towards water (in stationary phase) & mobile phase solvents during the movement of mobile phase under the capillary action of pores in the paper. The principle can also be adsorption chromatography between solid & liquid phases, where in the stationary phase is the solid surface of paper and the liquid phase is of mobile phase most of the applications of paper chromatography works on the principle of partition chromatography (partition between liquid phases) The movement of substance / components relative to the solvent is expressed in terms of Rf value ie, migration parameter.

Procedure :-

1. Cut a rectangular piece of chromatography (filter) paper make sure that chromatography paper will fit into the beaker otherwise cut it down to a smaller size
2. Take a pencil and scale & draw a horizontal line with pencil about 2-3 cm away from the tip of paper put a drop of the ink(phenyl alanin and glycine) mixture in the middle of the line with the help of a capillary tube.
3. Allow the drop to dry. Take chromatographic chamber & pour solvent in it fold one end of the filter paper strip & (staple) clip it.
4. The lowering spot should remain about 1cm above the solvent level.
5. Leave the chromatographic chamber undisturbed for some time we can observe, as the solvent moves through the paper, it spreads the different pigments of the mixture to various distance when the solvent rises about 3/4 th up the strip.
6. Remove the strip carefully & let it dry mark the solvent front & different colour spot from original line calculate the Rf value of different pigments (colours) using following formula.

Rf = Distance travelled by component from original line / Distance travelled by the solvent from the original line



Observation Table :

Sr. No.	Component	Dist. Travelled by component	Dist. Travelled by solvent front	Rf
1	Red			
2	Blue			
3	Yellow			

Result:

1. Rf value of Red Pigment is found to be _____
2. Rf value of Blue Pigment is found to be _____
3. Rf value of Yellow Pigment is found to be _____