Aim 30	Two Dimensional Chromatography	
Introduction		

The principle, reagents and procedure, precautions are same as in experiment No. 13.

This chromatography uses two solvents instead of one. Only One solvent is used at a time. In this, firstly the mixture of compounds are separated in and then paper is taken out left it to dry, turned this to 90° angle and after that use second solvent to separate again. Take out paper when the solvent has enclosed with more than $\frac{3}{4}$ of the area of the paper. Identification of compounds can be done by comparing the R_f values of unknown with the known under the same conditions. The only disadvantage of this method is that only one spot of sample or standards can be applied on one filter paper sheet.

Solvent - 1 (Butanol : Glacial acetic : Water) 12:3:5

Solvent - 2 (Phenol - Water), 25 ml of water is added to 100 g phenol.

Procedure

As in the experiment No. 13.

The spots which are not visible by one dimensional chromatography can be resolved by two dimensional chromatography, this is the main advantage of this method. Different R_f values can be obtained by running the developing chromatogram again in second solvent system.