CHROMATOGRAPHY

From the Greek word CHROMA, this means 'COLOR' and 'GRAPHEIN', which means to write or writing.
Hence the literal meaning of Chromatography is "COLOR WRITING"

Definition

Chromatography can be defined as a laboratory technique used for separating components of a mixture, in which the various components to be separated are distributed between two phases. One of this is called the stationary phase and the other is called the mobile phase
The mobile phase moves over the stationary phase in a definite direction to facilitate separation.

The components of the mixture redistribute themselves between these two phases.

In chromatography the stationary phase is usually a solid or liquid, and the mobile phase can be a gas, liquid or super-critical fluid. Chromatography is probably the most powerful and versatile technique available to the modern analyst. In a single step process it separates a mixture into its individual components and simultaneously provides a qualitative estimate of each component.

HISTORY OF CHROMATOGRAPHY

The first scientist to recognize chromatography as an efficient method of separation was the Russian botanist MIKHAIL TSWETT in 1903. Mikhail employed the technique to separate various pigments such as Chlorophyll, carotenoids and Xanthophyll by passing a solution of these compounds through a glass column packed with finely divided calcium carbonate. This technique was later named Liquid- Solid adsorption Chromatography. Although color has little to do with modern chromatography the name has persisted.

The technique as described by Tswett was largely ignored until the late 1930's and early 1940's when Martin and Synge introduced liquid-liquid chromatography, they further recommended replacing the liquid

Column Packing

Dry packing:

Here the adsorbent is added through a funnel into the column, when the adsorbent is even at the top, a filter paper or any material is used to keep the surface flat. The mobile phase for the separation is then added continuously until equilibrium is attained.

Note that the level of mobile phase should be slightly above that of the stationary phase.

Slurry packing

A slurry of the adsorbent is made in the mobile phase to be used for the separation, and then introduced into the column, until an even surface of adsorbent is obtained

Wet packing

An appropriate volume of the solvent is placed in the column, using a funnel and then the adsorbent is gradually introduced into the column, with constant tapping to ensure uniformity of adsorbent at the top

Note that in all three methods of packing, the solvent or mobile phase should be at the top of the adsorbent.

In column chromatography the mixture to be separated is introduced with the mobile phase at the top of the adsorbent in the column. The mobile phase cause a gradual downward migration of the components of the mixture, and the bands are separated and subsequently collected.

Column Qualities

Chromatographic tubes are usually made up of glass and are of various types. They are intended to support the adsorbent and are designed to permit control of solvent input and effluent collection. Dimensions for chromatographic tubes are usually variable but the length should be at least ten times the internal diameter.