

CHROMATOGRAPHY

- The components of a mixture are separated based on differences in the rates at which they are carried through a fixed or stationary phase by a gaseous or liquid mobile phase.
- Chromatography is a widely used method for the separation, identification, and determination of the chemical components in complex mixtures.
- Components of a mixture are carried through the stationary phase by the flow of a mobile phase, and separations are based on differences in migration rates among the mobile-phase components.



- The **stationary phase** in chromatography is a phase that is fixed in place either in a column or on a planar surface.
- The **mobile phase** in chromatography is a phase that moves over or through the stationary phase carrying with it the analyte mixture. The mobile phase may be a gas, a liquid, or a supercritical fluid.



- In **column chromatography**, the stationary phase is held in a narrow tube, and the mobile phase is forced through the tube under pressure or by gravity.
- In **planar chromatography**, the stationary phase is supported on a flat plate or in the pores of a paper, and the mobile phase moves through the stationary phase by capillary action or under the influence of gravity.



- **Chromatographic methods fall into three categories based on the nature of the mobile phase:**
 - **Liquid,**
 - **Gas,**
 - **Supercritical fluid.**



General Classification	Specific Method	Stationary Phase
Gas Chromatography	Gas-liquid	Liquid adsorbed or bonded to a solid surface
	Gas-solid	Solid
Liquid Chromatography	Partition	Liquid adsorbed or bonded to a solid surface
	Adsorption	Solid
	Ion Exchange	Ion exchange resin
	Size exclusion	Liquid in interstices of a polymeric solid
	Affinity	Group specific
Supercritical fluid Chromatography		Liquid bonded to a solid surface



Elution in Column Chromatography

Elution is a process in which solutes are washed through a stationary phase by the movement of a mobile phase.

The mobile phase that exits the column is termed the **eluate**.

- A solution of the sample containing a mixture of A and B in the mobile phase is introduced at the head of the column as a narrow band at time t_0 .
- The two components distribute themselves between the mobile phase and the stationary phase.
- Elution then occurs by forcing the sample components through the column by continuously adding fresh mobile phase.



An **eluent** is a solvent used to carry the components of a mixture through a stationary phase.

- With the first introduction of fresh mobile phase, the eluent, the portion of the sample contained in the mobile phase moves down the column, where further partitioning between the mobile phase and the stationary phase occurs (time t_1).
- Partitioning between the fresh mobile phase and the stationary phase takes place simultaneously at the site of the original sample.
- Further additions of solvent carry solute molecules down the column in a continuous series of transfers between the two phases.



Chromatograms

- A **chromatogram** is a plot of some function of solute concentration versus elution time or elution volume.
- The positions of the peak maxima on the time axis can be used to identify the components of the sample.
- The peak areas provide a quantitative measure of the amount of each species.



- The **retention time**, t_R , is the time between injection of a sample and the appearance of a solute peak at the detector of a chromatographic column.
- The **dead time** t_M , is the time it takes for an unretained species to pass through a chromatographic column.
- The **retention factor**, k_A , for solute A is related to the rate at which A migrates through a column. It is the amount of time a solute spends in the stationary phase relative to the time it spends in mobile phase.
- The **selectivity factor**, α , for solutes A and B is defined as the ratio of the distribution constant of the more strongly retained solute (B) to the distribution constant for the less strongly held solute (A).
- The **resolution** of a chromatographic column is a quantitative measure of its ability to separate analytes A and B.



Applications of Chromatography

- Chromatography is a powerful and versatile tool for separating closely related chemical species.
- In addition, it can be used for the qualitative identification and quantitative determination of separated species.

