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Semester VI (CHB-601) - II
Separation of molecules by chromatographic techniques

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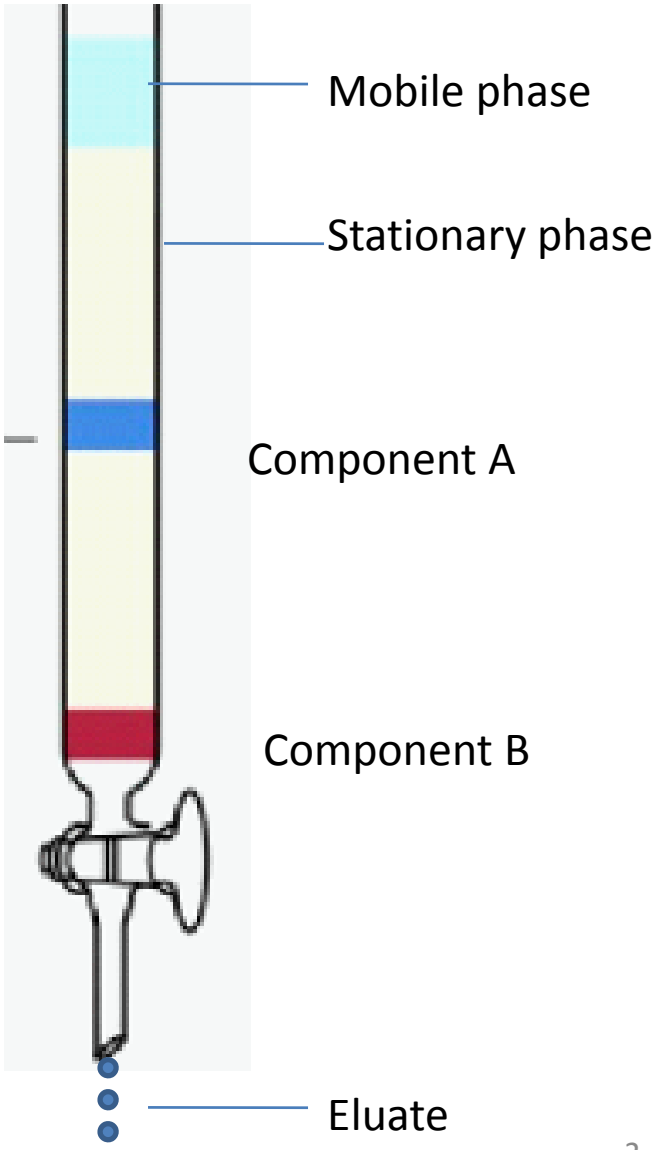
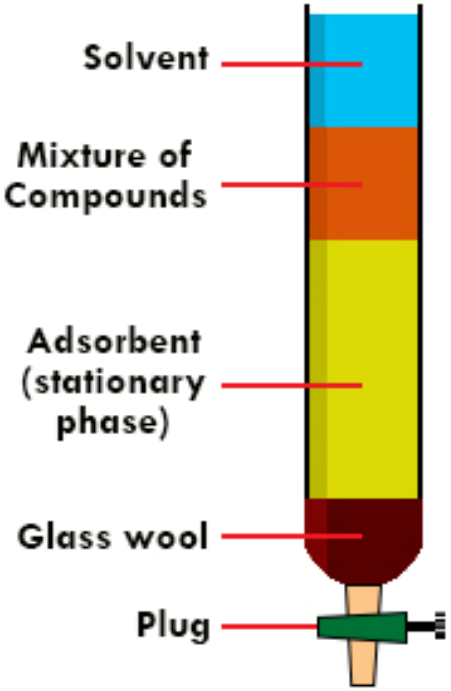
Elution in Column chromatography

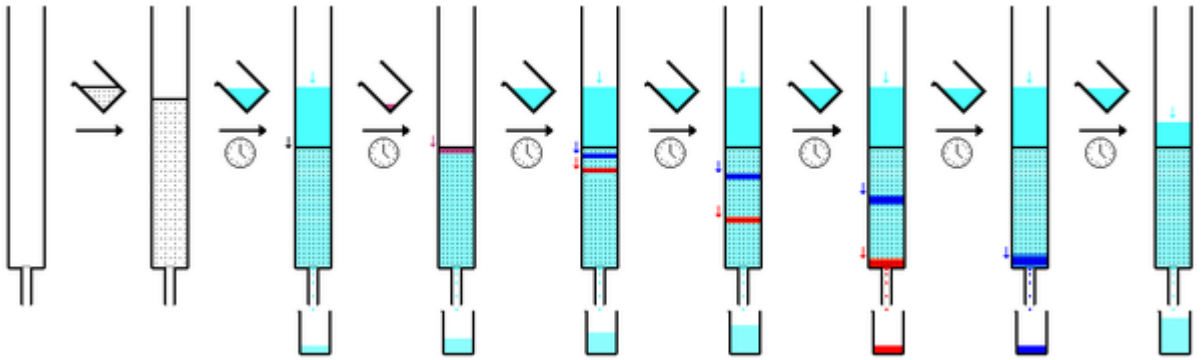
- Elution is a process in which solutes are washed through a stationary phase by the movement of a mobile phase.
- The mobile phase that exits the column is termed the eluate.
- An eluent is a solvent used to carry the components of a mixture through a stationary phase.



Elution in Column chromatography

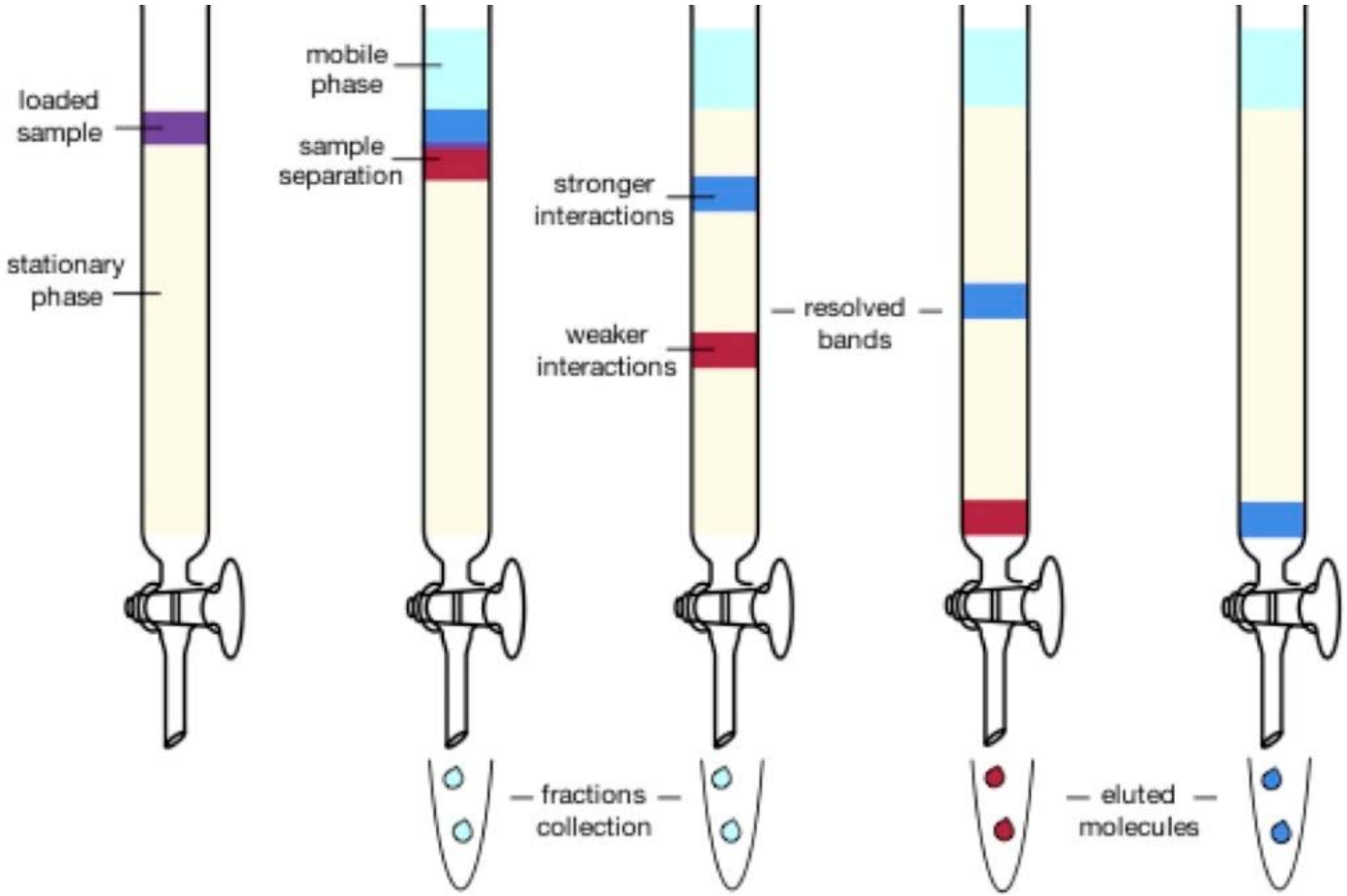
- Column consists of a narrow tubing packed with a stationary phase.





Chromatographic separation

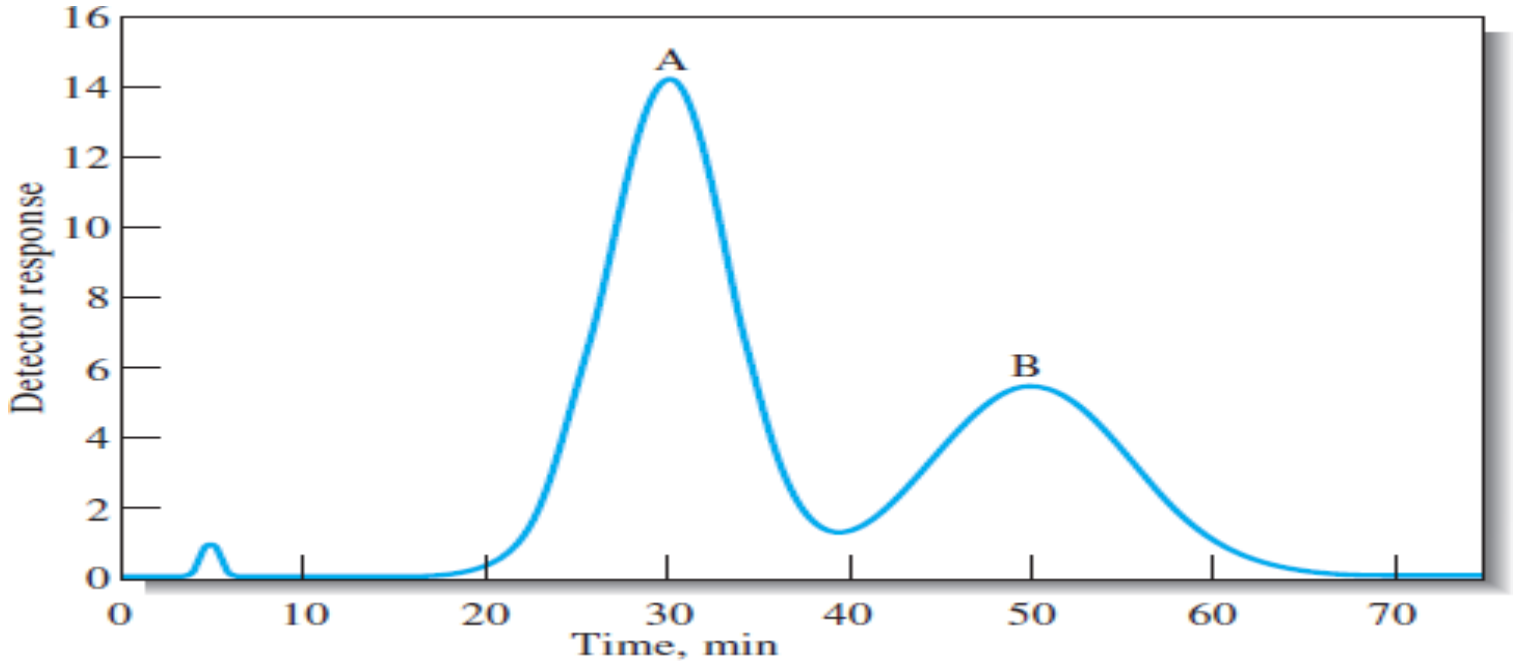
- In the mobile phase, components of the sample are uniquely drawn to the stationary phase and thus enter this phase at different times



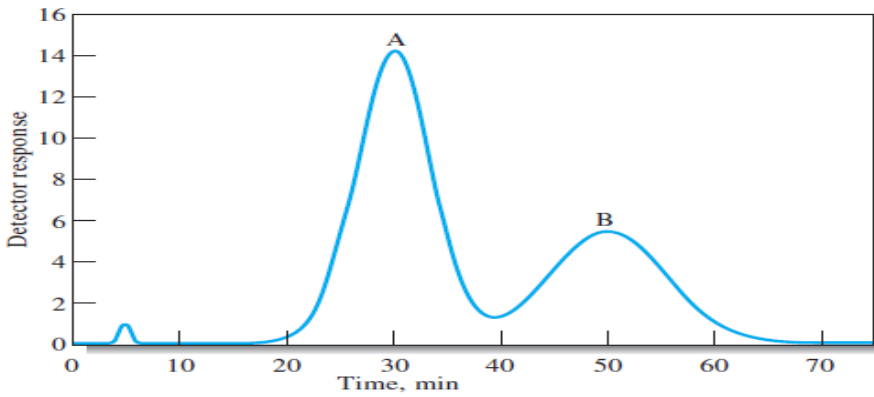
- Components of the sample are separated within the column

Chromatographic separation

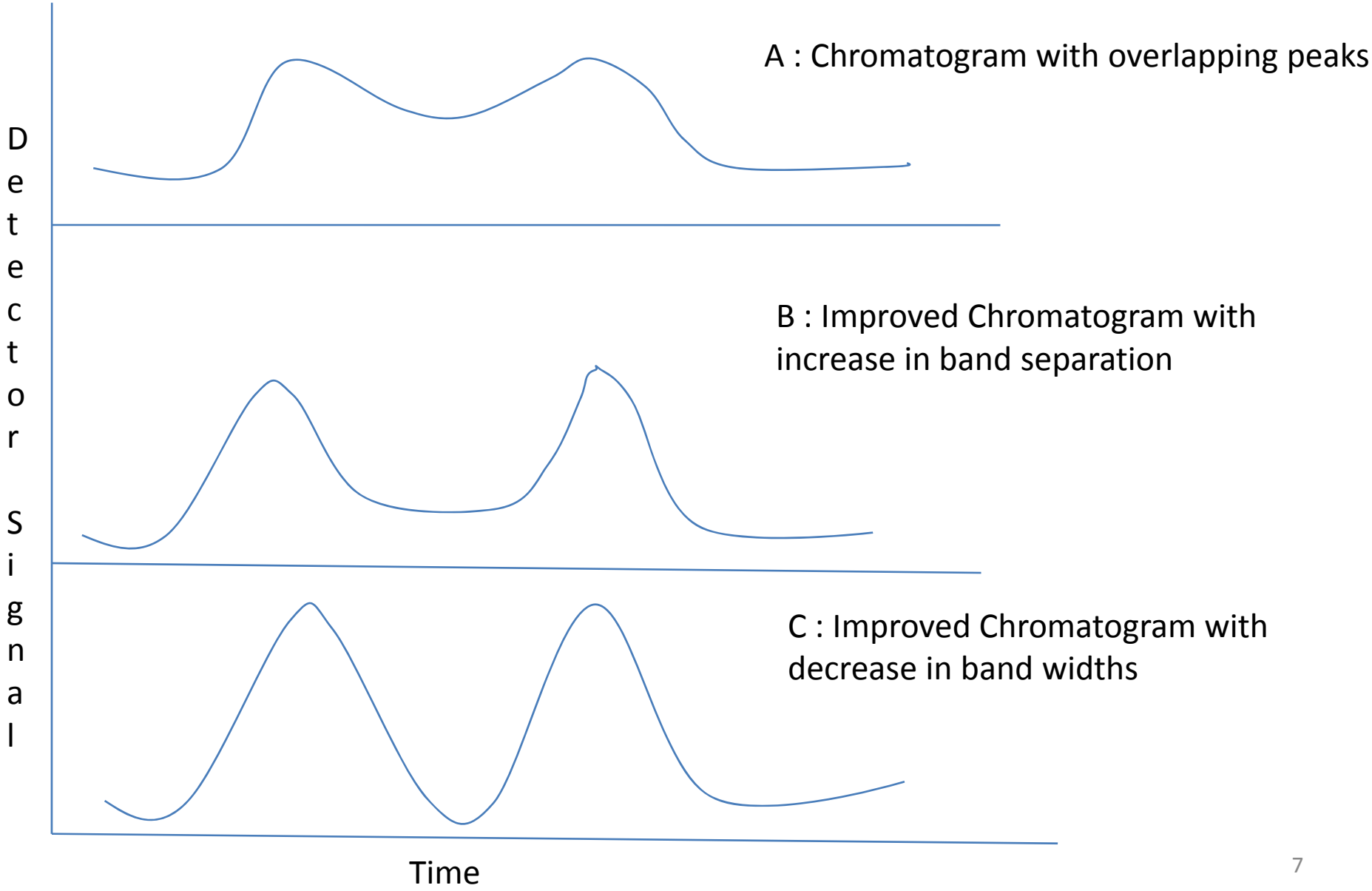
- Compounds used at the stationary phase reach the detector at unique times and produce a series of peaks along a time sequence



- Retention time is determined by each component reaching the detector at a characteristic time
- Number of components in a sample is determined by the no of peaks
- Amount of given component in a sample is determined by the area under the peaks
- Identity of components can be determined by the given retention times



Chromatographic bands

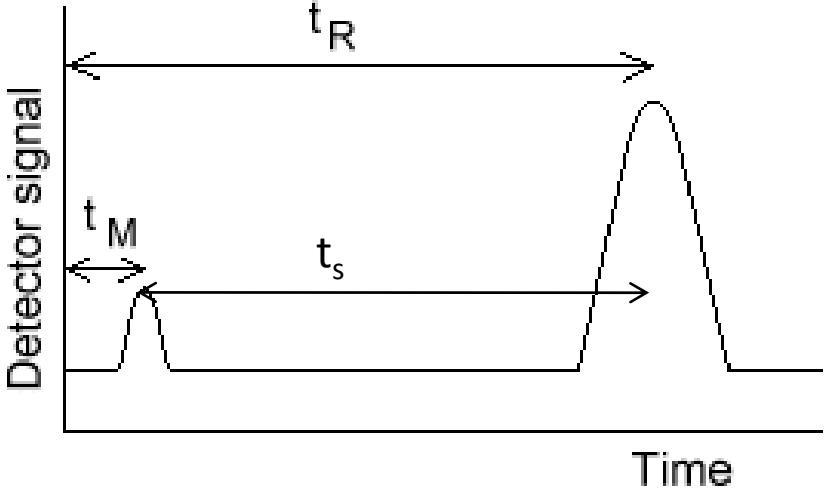


Theoretical Aspects of Chromatographic Bands

$$A_{\,mobile} \leftrightarrows A_{stationary}$$

A-analyte

Distribution Coefficient (K_c) $= \dfrac{(a_A)_S}{(a_A)_M} = \dfrac{(C_A)_S}{(C_A)_M} = \dfrac{C_S}{C_M}$



$t_R = t_S + t_M$

Average linear rate of solute migration

$$\bar{v} = \frac{L}{t_R}$$

Average linear velocity of mobile phase $u = \frac{L}{t_M}$

Average linear rate of solute migration $\bar{v} = \frac{L}{t_R}$

$\bar{v} =$ u x fraction of time solute spends in mobile phase

This fraction equals the average no of moles of solute in mobile phase at any instant divided by the total no. of moles of solute in column

$\bar{v} =$ u x moles of solute in mobile phase / total moles of solute

$$\bar{v} = u \times \frac{C_M V_M}{C_M V_M + C_S V_S} \qquad \bar{v} = u \times \frac{1}{1 + K_c V_S / V_M}$$

$$Retention\ factor\ k_A = K_A \frac{V_S}{V_M} \qquad \bar{v} = u \times \frac{1}{1 + k_A}$$