limitations of paper chromatography technique: Paper chromatography

has some limitations such as:

- Semi-quantitative in nature.
- Overlapping of spots for components with close Rf values.
- Higher concentrations of components leading to streaking rather than welldefined spots.
- Errors in Rf calculations due to uneven flow of the solvent front, which can be caused by various factors such as:
 - Running out of solvent at the bottom of the chamber.
 - Uneven cutting of the filter paper.
 - Unevenness of the bottom of the development chamber.
- · Improper sample spotting, including:
 - Spotting below the marked line, resulting in dipping into the solvent.
 - Accidental dipping of the spot into the solvent while inserting the paper into the solvent chamber.

Ion Exchange Chromatography (IEC):

Ion exchange chromatography is a process by which a mixture of similar charged ions can be separated by using an ion exchange resin which exchanges ions according to their relative affinities. Ion exchange chromatography is a type of adsorption chromatography. There is a **REVERSIBLE EXCHANGE OF**SIMILAR CHARGED IONS. Mostly similar charged ions like cations and anions can be conveniently separated by this technique. Many drugs and pharmaceutical agents are weakly or strongly acidic or basic in nature. Hence a mixture of similar charged substances can also be separated into pure components.

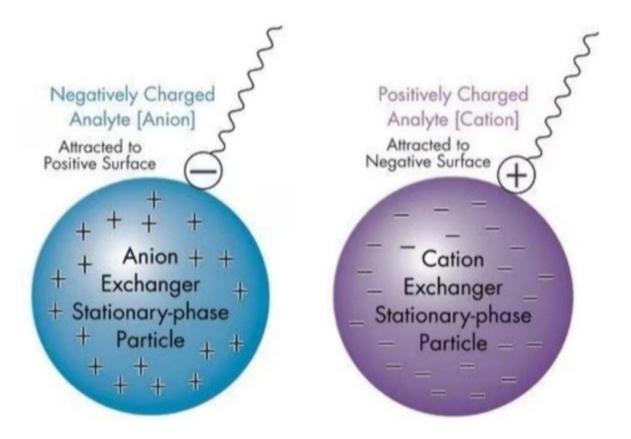


Figure 3: Ion Exchange Chromatography

The principle of this chromatographic technique based on exchange of ions between charges of stationary phase and ions of opposite charge in mobile phase. Soluble mixture to be separated, allowed to pass through a column of ion exchange resins which are insoluble polymer containing ionic group. Two types of ion exchanger are existing:

- 1) Natural e.g. certain clay minerals.
- 2) Synthetic such as; cellulose, dextran, acrylamide polymer etc.

Ion exchange resins are classified as:

- Cations exchanger.
- Anion exchanger.

CATION EXCHANGE:

The separation of cations using cation exchange resin. The cations (X⁺) to be separated are present in solution and exchanges for similar ions present in cation exchange resin (R⁻H⁺), a solid matrix. the exchange can be represented by the following equation:

$$X^{+}_{(l)} + R^{-}H^{+}_{(s)} \longrightarrow X^{+}R^{-}_{(s)} + H^{+}_{(l)}$$

The cations by the solid matrix of ion resin can be eluted by using buffers of different and hence of cations call be.

ANION EXCHANGE:

Separation of anion using anion exchange resin can be carried out. The anions to be separated are present in solution and for similar ions present in anion exchange resin, a solid matrix the can be represented by the following equation:

$$X^{-}_{(l)} + R^{+}Cl^{-}_{(s)} \longrightarrow X^{-}R^{+}_{(s)} + Cl^{-}_{(l)}$$

The anions retained by the solid matrix of ion exchange resin can be eluted by using buffers of different strength and hence separation of anions can be effected.

Resin type	Functional group
 1- <u>Cation Resin</u> A. Strong cation exchanger B. Weak cation exchanger 	-SO3H (sulphonic group) -COOH (carboxylic group)
 2- <u>Anion Resin</u> A. Strong Base anion exchanger B. Weak Base anion exchanger 	-NH ₄ ⁺ (quaternary ammonium group) -NH ₂ (amine group)

The properties of Ion- Exchange resin should have following requirements:

- 1) It must be stable.
- 2) It should be insoluble in common solvents.
- 3) It should have a degree of cross linking.
- 4) The swollen resin must be denser than water.
- 5) It must contain no. of ion exchange group.