

ION EXCHANGE CHROMATOGRAPHY

Ion exchange (IEX) chromatography is a technique that is commonly used in biomolecule purification. It involves the separation of molecules on the basis of their charge.

This technique exploits the interaction between charged molecules in a sample and oppositely charged moieties in the stationary phase of the chromatography matrix. The principle of separation is thus by reversible exchange of ions between the target ions present in the sample solution to the ions present on ion exchangers.

Two types of ion exchange separation is possible - cation exchange and anion exchange. In anion exchange the stationary phase is positively charged whilst in cation exchange it is negatively charged.

Principle of Ion Exchange Chromatography

IEX chromatography is used in the separation of charged biomolecules. The crude sample containing charged molecules is used as the liquid phase. When it passes through the chromatographic column, molecules bind to oppositely charged sites in the stationary phase.

The molecules separated on the basis of their charge are eluted using a solution of varying ionic strength. By passing such a solution through the column, highly selective separation of molecules according to their different charges takes place.

Key steps in the ion exchange chromatography procedure are listed below:

- An impure protein sample is loaded into the ion exchange chromatography column at a particular pH.
- Charged proteins will bind to the oppositely charged functional groups in the resin
- A salt gradient is used to elute separated proteins. At low salt concentrations, proteins having few charged groups are eluted and at higher salt concentrations, proteins with several charged groups are eluted.
- Unwanted proteins and impurities are removed by washing the column

The Applications of Ion Exchange Chromatography

Ion exchange is the most widely used chromatographic method for the separation and purification of charged biomolecules such as polypeptides, proteins, polynucleotides, and

nucleic acids. Its widespread applicability, high capacity and simplicity, and its high resolution are the key reasons for its success as a separation method. Ion exchange chromatography is widely used in several industrial applications some of which are as follows:

- Separation and Purification of blood components such as albumin, recombinant growth factors and enzymes.
- Biotechnology - Analytical applications such as quality control and process monitoring
- Food and clinical research - to study wheat varieties and the correlation of proteinuria with different renal diseases.
- Fermentation - Cation exchange resins are used to monitor the fermentation process during β -galactosidase production.