

Techniques Used in Chromatography and their Applications

Johnny Geurdes*

Department of Chemical Engineering, University of Jimma Institute of Technology, Jimma, Ethiopia

DESCRIPTION

Chromatography is a procedure for separating components present in a mixture. The mixture is dissolved in a substance called the mobile phase, which carries it through a second matter called the stationary phase. In this, the separation is made between two phases: the mobile phase and the stationary phase. Chromatography is mainly used to help catch criminals. It is used in programmes like CSI and gas chromatography, which are used to analyse blood and cloth samples, helping to identify criminals and bring them to integrity.

Types of chromatography

- HPLC
- Column chromatography
- Ion-exchange chromatography
- Gel-permeation (molecular sieve) chromatography
- Affinity chromatography
- Paper chromatography
- Thin-layer chromatography
- Gas chromatography
- Dye-ligand chromatography

HPLC stands for High Performance Liquid Chromatography. It is an analytical technique used to separate, identify, or quantify each component in a mixture. It is forced through at high pressures of up to 400 atmospheres. The chapter number of Hplc is 621. The components present in the hplc are the reservoir, pump, sample components, column components, reservoir, and detector. The types of detectors used in HPLC are UV detector, PDA detector, Fluorescence detector, conductivity detector, light scattering detector, and refractive index detector.

Applications of HPLC

The below following are the applications of HPLC:

- Analysis of drugs

- Analysis of synthetic polymers
- Analysis of pollutants in environmental analytics
- Determination of drugs in biological matrices
- Isolation of valuable products
- Product purity and quality control of industrial products and fine chemicals.
- Separation and purification of biopolymers such as enzymes or nucleic acids.
- Water purification

The procedure involved in HPLC is that the pump is positioned in the most upper stream of the liquid chromatography system and generates a flow of eluent from the solvent reservoir into the system. An injector is placed above the pump. The modest method is to use a syringe, and the sample is introduced into the flow of eluent. The commonly used technique is the injection method, which is based on sampling loops. The process of separation is performed inside the column. The analytes get separated inside the column, whereas a detector is used to detect the separation. The composition of the eluent is consistent, which indicates that there are no analytes present. The presence of analytes leads to changes in the composition of the eluent that the detector uses to measure these differences. This difference is observed as a form of an electronic signal. There are different types of detectors available on the market. The change in eluent detected by a detector is generated in the form of an electronic signal, and thus it is still not visible to our eyes. The calibration standard used for HPLC calibration is the caffeine standard because it is durable and standard.

Column chromatography is mainly used in LC, GC, Ion exchange chromatography, and chiral chromatography. The columns are generally made of borosilicate glass, stainless steel or acrylic glass. It is reverse phase chromatography if the mobile phase is polar and the stationary phase is non-polar; normal phase chromatography if the mobile phase is non-polar and the stationary phase is polar. Chiral chromatography is used to determine the content of chiral isomers in either normal or reverse phase, mobile phase varies, then it is known as a "gradient run." When it remains constant, then it is an isocratic run.

Correspondence to: Johnny Geurdes, Department of Chemical Engineering, University of Jimma Institute of Technology, Jimma, Ethiopia, E-mail: JohnnyGeurdes@calsu.us

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