OC S 565: CHROMATOGRAPHIC TECHNIQUES FOR ORGANIC CHEMISTRY

Course Outcome

- The students will explain the fundamentals of chromatography.
- Acquire a critical knowledge of principle, analytical and applications of paper and thin layer chromatography in organic chemistry.
- Learn the principles, separation mechanisms and applications of column chromatography.
- Understand the theory, instrumentation and applications of GC and HPLC.
- Learn column chromatography, paper chromatography, thin layer chromatography, gas chromatography and High performance liquid chromatography.
- Illustrate the principles, separation mechanisms and applications of affinity chromatography.
- Demonstrate the principle of GFC techniques used in isolation, purification, identification and analysis of macromolecules.

UNIT-I **[12 Hours]**

Fundamentals of chromatography: Introduction, terms and parameters used in chromatography, classification of chromatographic methods, criteria for selection of stationary and mobile phase-nature of adsorbents, factors influencing the adsorbents, nature and types of mobile phases and stationary phases.

Paper Chromatography (PC): Definitions, theory and principle, techniques; one, two-dimensional and circular PC, mechanism of separation, structure of cellulose and types of paper, methodology, preparation of sample, choice of solvents, location of spots and measurements of RF value, factors affecting RF values, advantages and applications, Separation of amino acids and carbohydrates.

Thin Layer Chromatography (TLC): Definition, mechanism, efficiency of TL plates, methodology, selection of stationary and mobile phases, preparation of plates. Spotting, development, identification and detection, reproducibility of RF values, comparison of TLC with high performance thin-layer chromatography, paper chromatography and column chromatography. Qualitative and quantitative analysis.

UNIT-II **[12 Hours]**

Column Chromatography (CC): Construction and operation of column, choice of adsorbents and eluents, techniques of elution, methods of detection, analytical and industrial applications.

Gas chromatography (GC): Principle, comparison of GSC and GLC, instrumentation, columns pack and tubular, study of detectors-thermal conductivity. Flame ionization, electron capture and mass spectrometry. Factors affecting the separation, retention volume, retention time, applications.

High Pressure Liquid Chromatography (HPLC): Apparatus, pumps, column packing, characteristics of liquid chromatographic detectors-UV, IR, refractometer and fluorescences detectors, advantages and applications.

UNIT-III **[12 Hours]**

Ion Exchange Chromatography (IEC): Definitions, principle, requirements for ion exchange resin and its synthesis, types of ion-exchange resins, basic features of ion-exchange reactions, resin properties-ion-exchange capacity, resin selectivity and factors affecting the selectivity, applications of IEC in preparative, purification and recovery processes.

Gel Permiation Chromatography: size exclusion chromatography (Gel filtration) with special reference to separation of protein, carbohydrates and nucleic acids. Preparation of medium, column, determination of void volume, sample application, detectors. Affinity chromatography: Chromatographic matrix, ligand selection, linkage of ligands, absorbant derivatives. LC/MS, LC/MS-MS, GC/MS, GC/MS-MS for organic compound analysis.

References

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5. Quantitative Analysis, 5th edn., R. A. Dayand A. L. Underwood, Prentice-Hall (1998).
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9. Vogel's Textbook of Quantitative Chemical Analysis, 6th edn., J. Mendham, R. C. Denney, J. D. Barnes and M. J. K. Thomas, Pearson Education, New Delhi (2004).
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