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Review Paper

Application of Chromatographic Techniques in Pharmaceutical and Biomedical Research

Dr. Girendra Kumar Gautam[1], Amit Rathore[2*], Deepak Kumar[3], Shikha Rathi[4]

[134]Shri Ram College of Pharmacy, Muzaffarnagar, Uttar Pradesh, India
[2] Department of Pharmaceutical Sciences, Gurukula Kangri (Deemed to be University), Haridwar, Uttarakhand, India

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ABSTRACT

Chromatographic techniques play a central role in pharmaceutical and biomedical research by enabling the reliable separation, identification, and quantification of complex chemical and biological components. This review discusses the application of major chromatographic methods, including thin-layer chromatography (TLC), gas chromatography (GC), high-performance liquid chromatography (HPLC), and ultra-high-performance liquid chromatography (UHPLC), with particular emphasis on their relevance to drug development and biomedical investigations. These techniques are widely employed at different stages of pharmaceutical research, such as drug discovery, formulation development, quality control, and stability studies. In biomedical research, chromatography contributes significantly to the analysis of biological fluids, metabolites, proteins, and biomarkers, supporting both clinical diagnostics and pharmacokinetic studies. The integration of chromatographic techniques with advanced detection systems, such as mass spectrometry and diode array detectors, has further enhanced analytical sensitivity, selectivity, and accuracy. This review also highlights recent methodological improvements that address challenges related to sample complexity, trace-level detection, and regulatory compliance. By examining current applications and practical considerations, the paper aims to provide a clear understanding of how chromatographic techniques continue to support innovation in pharmaceutical sciences and biomedical research. The continued development of efficient, robust, and environmentally conscious chromatographic methods is expected to further expand their role in ensuring drug safety, therapeutic efficacy, and reliable biomedical analysis.

INTRODUCTION

***Corresponding Author:** Amit Rathore
Address: *Department of Pharmaceutical Sciences, Gurukula Kangri (Deemed to be University), Haridwar, Uttarakhand, India.*
Email : amitrathore00730@gmail.com

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Chromatography has become one of the most indispensable analytical tools in pharmaceutical and biomedical research due to its ability to separate, identify, and quantify components present in complex mixtures. The increasing demand for safe, effective, and high-quality pharmaceutical products, along with advances in biomedical science, has placed strong emphasis on reliable analytical techniques. Among these, chromatographic methods stand out because of their versatility, sensitivity, and wide applicability across different stages of research and development.In pharmaceutical sciences, chromatography is routinely employed from the early phases of drug discovery to the final stages of quality control and regulatory compliance. During drug development, it supports the characterization of active pharmaceutical ingredients, excipients, impurities, and degradation products. Accurate separation and quantification are essential to ensure product consistency, stability, and therapeutic efficacy. Regulatory authorities across the world also require validated chromatographic methods as part of standard documentation, further underlining their critical role in pharmaceutical analysis.Biomedical research similarly relies heavily on chromatographic techniques for the analysis of biological samples such as blood, urine, tissues, and cell extracts. These samples often contain complex matrices with analytes present at very low concentrations, making selective and sensitive analytical approaches necessary. Chromatography enables researchers to study metabolic pathways, identify biomarkers, and evaluate drug–biomolecule interactions. Its application in pharmacokinetic and bioavailability studies has been particularly significant, as it allows precise monitoring of drug absorption, distribution, metabolism, and excretion.Over the years, chromatographic techniques have evolved considerably in response to growing analytical challenges. Traditional methods such as thin-layer chromatography and gas chromatography continue to be used for specific applications, while liquid chromatography, particularly high-performance liquid chromatography, has become the most widely adopted technique in pharmaceutical and biomedical laboratories. Improvements in column technology, mobile phase composition, and detection systems have led to enhanced resolution, reduced analysis time, and improved reproducibility. The development of ultra-high-performance liquid chromatography represents a further step toward higher efficiency and better analytical performance.The coupling of chromatography with advanced detection techniques has significantly expanded its capabilities. Hyphenated systems, such as liquid chromatography–mass spectrometry and gas chromatography–mass spectrometry, combine the separation power of chromatography with the structural identification strength of spectrometric methods. These integrated approaches have become essential in trace-level analysis, impurity profiling, and the identification of unknown compounds in both pharmaceutical formulations and biological samples.This review aims to provide a comprehensive overview of chromatographic techniques and their applications in pharmaceutical and biomedical research. By discussing fundamental principles, major techniques, and recent developments, the paper highlights the continuing importance of chromatography in supporting scientific innovation, ensuring drug safety, and advancing biomedical knowledge.

1. Principles and Classification of Chromatographic Techniques

Chromatography is an analytical separation technique based on the differential distribution of components between two phases: a stationary phase and a mobile phase. The separation occurs due to variations in physicochemical properties such as polarity, molecular size, charge, and affinity of analytes toward the stationary phase. As the mobile phase moves through or across the stationary phase, individual components migrate at different rates, resulting in effective separation.

The efficiency of chromatographic separation is governed by several key parameters, including the nature of the stationary phase, the composition and flow rate of the mobile phase, temperature, and interactions between analyte molecules and the chromatographic system. These interactions may involve adsorption, partitioning, ion exchange, or molecular sieving, depending on the type of chromatography employed. The choice of chromatographic technique is therefore dictated by the chemical nature of the analyte and the analytical objective.

1.1 Fundamental Principles of Chromatographic Separation

The core principle of chromatography lies in the equilibrium established between the stationary and mobile phases. When a mixture is introduced into the system, its components distribute themselves between the two phases according to their respective affinities. Compounds with stronger interactions with the stationary phase exhibit slower migration, while those with higher affinity for the mobile phase move more rapidly. This differential migration leads to spatial or temporal separation of analytes.Resolution, selectivity, and retention are essential parameters defining chromatographic performance. Resolution reflects the degree of separation between adjacent peaks, selectivity represents the ability of the system to distinguish between different analytes, and retention describes the time or distance a compound remains within the chromatographic system. Optimization of these parameters is critical for achieving accurate and reproducible results in pharmaceutical and biomedical analyses.

1.2 Classification Based on Mobile and Stationary Phases

Chromatographic techniques can be broadly classified according to the physical state of the mobile and stationary phases. In gas chromatography, the mobile phase is an inert gas, while the stationary phase is a liquid or solid supported on an inert matrix. Liquid chromatography employs a liquid mobile phase, which may be aqueous, organic, or a mixture of solvents, combined with a solid or chemically bonded stationary phase.Based on the nature of the stationary phase, chromatography may also be categorized into adsorption chromatography, partition chromatography, ion-exchange chromatography, and size-exclusion chromatography. Each of these techniques exploits a specific interaction mechanism, enabling selective separation of analytes from complex mixtures.

1.3 Planar and Column Chromatographic Techniques

Chromatographic methods are further classified into planar and column techniques. Planar chromatography, such as thin-layer chromatography and paper chromatography, involves a stationary phase distributed on a flat surface. These methods are widely used for qualitative analysis, reaction monitoring, and preliminary screening due to their simplicity and low cost.Column chromatography, in contrast, employs a stationary phase packed within a cylindrical column, allowing continuous flow of the mobile phase. Techniques such as high-performance liquid chromatography and gas chromatography fall under this category and offer superior resolution, sensitivity, and quantitative accuracy. Column-based methods are therefore preferred in advanced pharmaceutical and biomedical research.

1.4 Normal Phase and Reversed Phase Chromatography

Another important classification is based on the relative polarity of the stationary and mobile phases. In normal phase chromatography, the stationary phase is polar, typically silica or alumina, while the mobile phase is non-polar. Separation in this mode is primarily driven by polarity differences among analytes.Reversed phase chromatography, the most widely used mode in pharmaceutical analysis, employs a non-polar stationary phase and a polar mobile phase. This arrangement provides enhanced reproducibility, better peak shape, and compatibility with aqueous biological samples, making it particularly suitable for drug analysis, bioanalysis, and metabolite profiling.

Table 1. Fundamental Principles Governing Chromatographic Separation

Principle	Description	Relevance in Pharmaceutical & Biomedical Research
Adsorption	Separation based on differential adsorption of analytes onto a solid stationary phase	Used in TLC and column chromatography for qualitative screening
Partition	Distribution of analytes between two immiscible liquid phases	Basis of HPLC and bioanalytical separations
Ion Exchange	Electrostatic interactions between charged analytes and oppositely charged stationary phase	Applied in protein, peptide, and nucleic acid analysis
Size Exclusion	Separation according to molecular size and shape	Widely used for biopolymers and macromolecules
Affinity	Specific biological interactions between analyte and ligand	Essential for biomarker and enzyme purification

Table 2. Classification of Chromatographic Techniques Based on Mobile and Stationary Phases

Type of Chromatography	Mobile Phase	Stationary Phase	Typical Applications
Gas Chromatography (GC)	Inert gas (He, N_2, H_2)	Liquid or solid coated support	Volatile drugs, residual solvents
Liquid Chromatography (LC)	Liquid solvents	Solid or bonded phase	Drug assays, impurity profiling
Supercritical Fluid Chromatography (SFC)	Supercritical CO_2	Solid stationary phase	Chiral separations, green analysis
Ion Exchange Chromatography	Buffered aqueous solution	Charged resin	Protein purification
Size Exclusion Chromatography	Aqueous or organic solvent	Porous gel matrix	Molecular weight determination

Table 3. Comparison of Planar and Column Chromatographic Techniques

Parameter	Planar Chromatography	Column Chromatography
Format	Flat stationary surface	Packed cylindrical column
Examples	TLC, Paper Chromatography	HPLC, GC
Resolution	Moderate	High
Quantitative Capability	Limited	Excellent
Application Scope	Preliminary screening	Advanced pharmaceutical analysis

Table 4. Distinction Between Normal Phase and Reversed Phase Chromatography

Feature	Normal Phase Chromatography	Reversed Phase Chromatography
Stationary Phase	Polar (silica, alumina)	Non-polar (C18, C8)
Mobile Phase	Non-polar solvents	Polar solvents
Separation Mechanism	Polarity-based interactions	Hydrophobic interactions
Reproducibility	Moderate	High
Pharmaceutical Use	Limited	Widely used

2. Chromatographic Techniques Used in Pharmaceutical Research

Chromatographic techniques form the analytical backbone of pharmaceutical research due to their precision, reliability, and adaptability to diverse drug molecules. These techniques are routinely employed for the identification, separation, purification, and quantification of active pharmaceutical ingredients (APIs), excipients, impurities, and degradation products. Selection of an appropriate chromatographic method depends on the physicochemical properties of the analyte, sensitivity requirements, and the intended stage of pharmaceutical development.

2.1 Thin Layer Chromatography (TLC)

Thin layer chromatography is one of the most widely used planar chromatographic techniques in pharmaceutical laboratories. It is primarily applied for rapid qualitative analysis, reaction monitoring, and preliminary identification of compounds. Despite its relatively lower resolution compared to column techniques, TLC remains valuable due to its simplicity, cost-effectiveness, and ability to analyze multiple samples simultaneously.

Fig 1 Thin Layer Chromatography

Table 5 Pharmaceutical Applications of Thin Layer Chromatography

Application Area	Purpose	Advantage
Raw material analysis	Identity confirmation	Rapid screening
Reaction monitoring	Progress assessment	Minimal solvent use
Herbal drug analysis	Phytochemical profiling	Multiple sample analysis
Impurity detection	Preliminary assessment	Low operational cost

2.2 Ultra-High Performance Liquid Chromatography (UHPLC)

Ultra-high performance liquid chromatography represents an advancement over conventional HPLC, utilizing columns packed with smaller particle sizes and operating at higher pressures. UHPLC enables faster analysis, improved resolution, and reduced solvent consumption, aligning well with high-throughput pharmaceutical research.

Table 6 Comparison of HPLC and UHPLC in Pharmaceutical Analysis

Parameter	HPLC	UHPLC
Particle size	3–5 µm	<2 µm
Analysis time	Moderate	Short
Resolution	High	Very high
Solvent consumption	Higher	Lower
Suitability	Routine analysis	High-throughput studies

2.3 Gas Chromatography (GC)

Gas chromatography is extensively used for the analysis of volatile and semi-volatile pharmaceutical compounds. It plays a crucial role in residual solvent analysis, impurity profiling, and detection of low-molecular-weight compounds. When coupled with sensitive detectors, GC provides excellent precision and sensitivity.

Table 7 Applications of Gas Chromatography in Pharmaceuticals

Analyte Type	GC Application	Regulatory Importance
Residual solvents	Quantification	ICH compliance
Volatile impurities	Detection	Toxicity control
Low molecular drugs	Assay	Quality assurance
Environmental contaminants	Monitoring	Safety evaluation

Fig2 Gas Chromatography

2.4 Ion Exchange Chromatography

Ion exchange chromatography separates compounds based on their charge properties. It is particularly important in the analysis and purification of ionic drugs, peptides, and biopharmaceutical products. This technique is widely used in protein purification and characterization.

Table 8 Ion Exchange Chromatography in Pharmaceutical Research

Sample Type	Separation Basis	Application
Proteins	Net charge	Purification
Peptides	Ionic interaction	Characterization
Nucleic acids	Charge density	Isolation
Ionic drugs	pKa differences	Analytical separation

2.5 Size Exclusion Chromatography (SEC)

Size exclusion chromatography separates molecules based on their hydrodynamic volume without chemical interaction with the stationary phase. It is particularly useful for analyzing macromolecules such as polymers, proteins, and biopharmaceutical formulations.

Table 9 Pharmaceutical Uses of Size Exclusion Chromatography

Application	Purpose	Benefit
Molecular weight determination	Polymer analysis	Non-destructive
Protein aggregation studies	Stability assessment	High reliability
Biopharmaceutical characterization	Quality evaluation	Minimal interaction

3. Role of Chromatography in Drug Discovery and Development

Chromatographic techniques play a central role in drug discovery and development by enabling the efficient separation, identification, purification, and quantification of chemical entities at every stage of the pharmaceutical pipeline. From the initial screening of candidate molecules to the final evaluation of drug stability and safety, chromatography provides reliable analytical support essential for informed decision-making and regulatory compliance.

Table 10 Chromatographic Techniques Used in Lead Identification

Technique	Purpose	Contribution to Drug Discovery
TLC	Preliminary screening	Rapid detection of bioactive compounds
HPLC	Compound purification	Isolation of pure lead molecules
LC–MS	Molecular characterization	Confirmation of molecular mass
Chiral chromatography	Enantiomer separation	Selection of pharmacologically active isomer

3.1 Application in Lead Optimization

Once lead compounds are identified, structural modifications are introduced to improve potency, selectivity, and pharmacokinetic properties. Chromatography supports structure–activity relationship studies by enabling comparative analysis of closely related analogues. Accurate quantification and impurity assessment ensure reliable evaluation of optimized candidates.

Table 11 Role of Chromatography in Lead Optimization

Parameter Evaluated	Chromatographic Method	Analytical Outcome

Purity assessment	HPLC	Accurate quantification
Stereochemical analysis	Chiral HPLC	Enantiomeric purity
Degradation behavior	Stability-indicating HPLC	Chemical robustness
Metabolic profiling	LC–MS	Identification of metabolites

3.2 Role in Formulation and Process Development

Chromatography assists in evaluating drug–excipient compatibility, optimizing manufacturing processes, and ensuring consistency of drug formulations. It enables detection of process-related impurities and degradation products that may arise during scale-up and storage.

Table 12 Chromatographic Role in Formulation and Process Development

Development Aspect	Chromatographic Technique	Outcome
Drug–excipient compatibility	HPLC	Stability confirmation
Process impurity analysis	HPLC, GC	Quality assurance
Content uniformity	HPLC	Dosage accuracy
Stability testing	Stability-indicating HPLC	Shelf-life determination

4. Applications in Pharmaceutical Quality Control and Regulatory Compliance

Pharmaceutical quality control (QC) is a critical component of drug manufacturing that ensures the identity, purity, strength, and safety of pharmaceutical products. Chromatographic techniques form the analytical cornerstone of QC laboratories due to their high precision, sensitivity, and reproducibility. Regulatory authorities worldwide mandate the use of validated chromatographic methods to ensure consistent product quality and patient safety throughout the product lifecycle.

4.1 Role in Assay and Content Uniformity Testing

Chromatographic methods, particularly high-performance liquid chromatography, are routinely employed for quantitative estimation of active pharmaceutical ingredients in bulk drugs and finished dosage forms. These analyses ensure that each unit contains the correct amount of drug substance within acceptable limits, thereby guaranteeing therapeutic efficacy.

Table13 Chromatographic Techniques Used in Assay and Content Uniformity

Quality Parameter	Chromatographic Technique	Regulatory Significance
API assay	HPLC	Confirms labeled strength
Content uniformity	HPLC	Ensures dose consistency
Blend uniformity	HPLC	Prevents dosage variation
Finished product testing	HPLC, UHPLC	Batch release approval

4.2 Impurity Profiling and Related Substances Analysis

Detection and quantification of impurities are essential for pharmaceutical safety and regulatory compliance. Chromatography enables effective separation of process-related, degradation-related, and residual impurities at trace levels. Regulatory guidelines require comprehensive impurity profiling to minimize toxicological risk.

Table14 Chromatographic Methods for Impurity Analysis

Impurity Type	Analytical Technique	Purpose
Organic impurities	HPLC	Structural differentiation
Volatile impurities	GC	Residual solvent analysis
Inorganic impurities	Ion chromatography	Elemental purity
Degradation products	Stability-indicating HPLC	Shelf-life determination

4.3 Residual Solvent Analysis

Residual solvents originating from manufacturing processes pose potential health risks and must be strictly controlled. Gas chromatography is the preferred technique for residual solvent analysis due to its sensitivity toward volatile compounds. Regulatory authorities specify permissible limits and analytical requirements for solvent quantification.

Table15 Application of GC in Residual Solvent Analysis

Solvent Class	Examples	Analytical Technique
Class I	Benzene	GC
Class II	Methanol, acetonitrile	GC
Class III	Ethanol, acetone	GC
Process solvents	Various	GC-FID

4.4 Stability Testing and Shelf-Life Determination

Chromatographic techniques are indispensable in stability studies conducted under various environmental conditions. Stability-indicating chromatographic methods help identify degradation pathways and ensure that the pharmaceutical product maintains its quality throughout its intended shelf life.

Table16 Application of GC in Residual Solvent Analysis

Stability Study Type	Chromatographic Method	Outcome
Accelerated stability	HPLC	Degradation profiling
Long-term stability	HPLC	Shelf-life assignment
Photostability	HPLC-DAD	Light-induced degradation
Stress testing	HPLC	Method specificity

4.5 Compliance with Good Manufacturing Practices (GMP)

Chromatographic analyses support GMP compliance by ensuring consistent manufacturing processes, controlled impurity levels, and reproducible product quality. Routine chromatographic monitoring is an integral part of in-process control and batch release testing.

5. Chromatographic Applications in Biomedical Research

Chromatographic techniques play a crucial role in biomedical research by enabling the precise analysis of complex biological systems. Biological samples such as blood, urine, tissues, and cell extracts contain a wide range of endogenous and exogenous compounds that require highly selective and sensitive analytical approaches. Chromatography provides an effective platform for the separation, identification, and quantification of biomolecules, drugs, metabolites, and biomarkers, thereby supporting disease diagnosis, therapeutic monitoring, and biomedical discovery.

Table17 Chromatographic Techniques for Biomolecule Analysis

Biomolecule Type	Chromatographic Technique	Biomedical Application
Proteins	Ion exchange chromatography	Purification and characterization
Peptides	Reversed-phase HPLC	Sequence analysis
Nucleic acids	Size exclusion chromatography	Molecular size determination
Lipids	HPLC	Lipid profiling

5.1 Role in Proteomics and Peptide Mapping

Proteomics research relies heavily on chromatography for protein separation, purification, and peptide mapping. High-resolution liquid chromatography enables the separation of complex protein digests prior to mass spectrometric analysis. This approach facilitates protein identification, post-translational modification analysis, and comparative proteomic studies.

Table18 Chromatographic Applications in Proteomics

Analytical Objective	Technique Used	Research Outcome
Protein separation	HPLC	High-resolution profiling
Peptide mapping	Reversed-phase HPLC	Structural characterization
Post-translational modification analysis	LC–MS	Functional insights
Biomarker discovery	Multidimensional chromatography	Disease association

5.2 Applications in Metabolomics and Lipidomics

Metabolomics and lipidomics aim to comprehensively analyze small molecules involved in metabolic processes. Chromatographic separation is essential for resolving structurally similar metabolites and lipids prior to detection. These studies contribute to the identification of disease-related metabolic alterations and therapeutic targets.

Table 19 Chromatography in Metabolomics and Lipidomics

Study Area	Sample Type	Chromatographic Method
Metabolomics	Plasma, urine	HPLC, LC–MS
Lipidomics	Tissue extracts	HPLC
Pathway analysis	Biological fluids	GC–MS
Biomarker identification	Serum	LC–MS/MS

5.3 Contribution to Translational and Personalized Medicine

Chromatography supports translational research by bridging laboratory findings with clinical application. Personalized medicine approaches rely on chromatographic analysis of patient-specific biomarkers and drug response profiles, enabling tailored therapeutic strategies.

6. Bioanalytical Chromatography

Bioanalytical chromatography is a specialized application of chromatographic techniques focused on the quantitative and qualitative analysis of drugs, metabolites, and endogenous compounds in biological matrices. These matrices, including plasma, serum, urine, saliva, and tissues, present significant analytical challenges due to their complex composition. Chromatography, particularly when combined with sensitive detection systems, provides the selectivity, accuracy, and reproducibility required for reliable bioanalytical measurements.

6.1 Role in Quantification of Drugs and Metabolites

Accurate quantification of drugs and their metabolites in biological samples is essential for evaluating pharmacokinetics, bioavailability, and therapeutic efficacy. Liquid chromatography is widely preferred due to its compatibility with aqueous biological matrices and its ability to separate structurally diverse compounds. Rigorous method validation ensures reliability and regulatory acceptance of bioanalytical data.

Table 20 Chromatographic Techniques Used in Drug and Metabolite Quantification

Sample Matrix	Analyte Type	Chromatographic Technique
Plasma	Parent drug	HPLC
Serum	Metabolites	LC–MS
Urine	Excretion products	GC, LC–MS
Tissue homogenates	Drug residues	LC–MS/MS

6.2 Application in Pharmacokinetic and Pharmacodynamic Studies

Bioanalytical chromatography is central to pharmacokinetic and pharmacodynamic evaluations, where time-dependent changes in drug concentration are measured following administration. These studies provide critical information on absorption, distribution, metabolism, and elimination, which guides dose optimization and clinical trial design.

Table 21 Chromatographic Techniques Used in Drug and Metabolite Quantification

Study Parameter	Biological Matrix	Analytical Technique
Absorption	Plasma	LC–MS/MS
Distribution	Tissue samples	HPLC
Metabolism	Liver microsomes	LC–MS
Elimination	Urine, feces	GC

6.3 Therapeutic Drug Monitoring (TDM)

Therapeutic drug monitoring relies on bioanalytical chromatography to maintain drug concentrations within the therapeutic window, particularly for drugs with narrow safety margins. Chromatographic methods provide the sensitivity and specificity necessary to support individualized patient care and minimize adverse effects.

Table 21 Chromatographic Techniques Used in Drug and Metabolite Quantification

Drug Class	Clinical Importance	Analytical Method
Antiepileptics	Narrow therapeutic index	HPLC
Immunosuppressants	Dose optimization	LC–MS/MS
Antibiotics	Resistance prevention	HPLC
Anticancer agents	Toxicity control	LC–MS

7. Hyphenated Chromatographic Techniques

Hyphenated chromatographic techniques combine the separation efficiency of chromatography with the structural and quantitative capabilities of advanced detection systems. These integrated analytical platforms have significantly enhanced pharmaceutical and biomedical research by enabling simultaneous separation, identification, and quantification of complex mixtures. The coupling of chromatographic systems with spectroscopic or spectrometric detectors has improved sensitivity, selectivity, and analytical confidence, particularly in trace-level and complex biological analyses.

7.1 Liquid Chromatography–Mass Spectrometry (LC–MS and LC–MS/MS)

Liquid chromatography–mass spectrometry is one of the most widely used hyphenated techniques in pharmaceutical and biomedical research. The chromatographic component provides effective separation of analytes, while mass spectrometry offers molecular weight determination and structural information. LC–MS/MS, with its enhanced sensitivity and specificity, is extensively applied in bioanalysis, pharmacokinetic studies, metabolite identification, and biomarker discovery.LC–MS-based methods are particularly advantageous for analyzing thermally labile and non-volatile compounds, making them suitable for drugs, metabolites, peptides, and endogenous biomolecules. The technique supports high-throughput analysis and meets regulatory expectations for bioanalytical method validation.

7.2 Gas Chromatography–Mass Spectrometry (GC–MS)

Gas chromatography–mass spectrometry is a powerful hyphenated technique used for the analysis of volatile and semi-volatile compounds. GC–MS provides excellent separation efficiency combined with reliable compound identification through mass spectral libraries. In pharmaceutical research, GC–MS is widely used for residual solvent analysis, impurity profiling, and toxicological screening.In biomedical research, GC–MS plays a crucial role in metabolomics, forensic analysis, and detection of environmental contaminants. The robustness and reproducibility of GC–MS make it a preferred technique for confirmatory analysis and regulatory compliance.

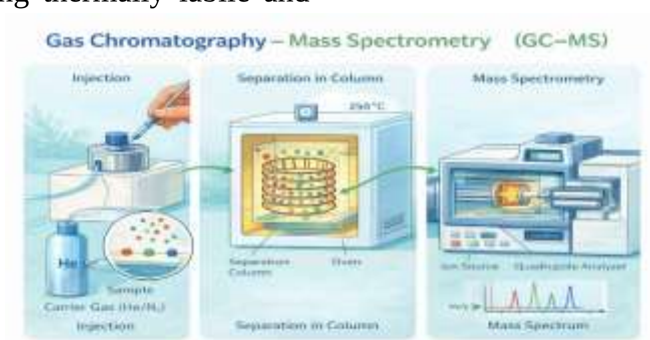

Table 23 Gas Chromatography – Mass Spectrometry (GC- MS)

8. Recent Advances in Chromatographic Technologies

Recent years have witnessed significant technological progress in chromatographic sciences, driven by the growing demand for higher sensitivity, faster analysis, improved resolution, and enhanced compatibility with complex biological and pharmaceutical matrices. These advances have substantially expanded the scope and efficiency of chromatography in both pharmaceutical and biomedical research.One of the most notable developments is the emergence of ultra-high-performance liquid chromatography (UHPLC). By employing columns packed with sub-2 µm particles and operating at higher pressures, UHPLC provides superior separation efficiency, sharper peak resolution, and reduced analysis time compared to conventional HPLC. This advancement has proven particularly valuable in high-throughput drug screening, impurity profiling, and metabolomic studies.Hyphenated chromatographic techniques have also undergone remarkable refinement. Advanced coupling of chromatography with mass spectrometry, such as LC–MS/MS and GC–MS/MS, has enabled highly selective and sensitive detection of analytes at trace levels. Improvements in ionization techniques, mass analyzers, and data acquisition systems have strengthened the reliability of structural elucidation and quantitative bioanalysis, especially in pharmacokinetic and toxicological investigations.Another important advancement is the development of novel stationary phases with enhanced selectivity. The introduction of core–shell particles, monolithic columns, and chemically modified stationary phases has improved mass transfer kinetics and separation efficiency. Chiral stationary phases have advanced enantioselective chromatography, supporting the analysis of stereoisomers that are critical in modern drug development.

CONCLUSION

Chromatographic techniques have become indispensable analytical tools in pharmaceutical and biomedical research due to their versatility, precision, and reliability. Throughout this review, the fundamental principles and classifications of chromatography have been discussed, highlighting how different techniques are tailored to address the complexity of pharmaceutical formulations and biological matrices. From conventional methods such as thin-layer chromatography and gas chromatography to advanced liquid chromatographic and hyphenated systems, chromatography continues to support all stages of drug discovery and development.The application of chromatographic techniques in pharmaceutical research extends from early-stage compound identification to formulation development, quality control, and regulatory compliance. In biomedical research, chromatography plays a central role in bioanalysis, therapeutic drug monitoring, metabolomics, and biomarker identification. The integration of chromatography with sensitive detection systems, particularly mass spectrometry, has greatly enhanced analytical selectivity and sensitivity, enabling accurate quantification of trace-level compounds in complex samples.Recent technological advancements, including ultra-high-performance liquid chromatography, novel stationary phases, green chromatographic approaches, and automated systems, have further strengthened analytical efficiency and sustainability. These innovations not only improve resolution and throughput but also align chromatographic practices with evolving regulatory and environmental expectations.In conclusion, chromatography remains a cornerstone of modern pharmaceutical and biomedical analysis. Continuous technological evolution and methodological refinement are expected to further expand its applications, ensuring robust analytical support for drug development, clinical research, and patient-centered healthcare.

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