

ROLE OF GC–MS IN THE IDENTIFICATION AND QUANTIFICATION OF VOLATILE COMPOUNDS IN MEDICINAL PLANTS – A REVIEW

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ABSTRACT

Medicinal plants contain numerous volatile constituents, particularly monoterpenes, sesquiterpenes, oxygenated terpenoids, aldehydes, ketones, alcohols and esters. These compounds contribute to aroma, plant defense and several biological activities and can serve as chemical markers for authentication and quality control. Gas chromatography–mass spectrometry (GC–MS) is one of the most important analytical techniques for volatile compounds because gas chromatography separates components while mass spectrometry provides characteristic fragmentation patterns for identification. Identification is commonly supported by mass spectral library matching, retention indices and, when required, authentic reference standards. GC–MS is widely applied to essential-oil characterization, phytochemical profiling, chemotaxonomy, authentication, adulteration assessment and quantitative analysis. This review briefly discusses the principle,

instrumentation, sample preparation, identification, quantification, applications, advantages, limitations and future perspectives of GC–MS in medicinal plants.

KEYWORDS: GC–MS; medicinal plants; volatile compounds; essential oils; phytochemical profiling; retention index; quality control; quantification.

1. INTRODUCTION

Medicinal plants are important sources of chemically diverse natural products. Their volatile fraction contains compounds that may contribute to characteristic aroma, biological activity and plant–environment interactions. The composition of volatile constituents varies with species, plant part, geographical origin, climate, harvesting period, processing and storage. Reliable analysis is therefore essential for authentication and standardization. GC–MS combines the high separation capability of gas chromatography with the structural information of mass spectrometry and is particularly suitable for volatile and thermally stable compounds.

2. Volatile Compounds in Medicinal Plants

The volatile fraction of medicinal plants is commonly rich in monoterpenes and sesquiterpenes and their oxygenated derivatives. Other constituents include phenylpropanoids, aldehydes, ketones, alcohols and esters. Examples include α -pinene, limonene, linalool, menthol, eugenol, thymol, carvacrol, camphor and β -caryophyllene. Their relative concentrations can provide characteristic chemical fingerprints useful for comparing species, chemotypes and batches.

3. Principle of GC–MS

In GC, the sample is vaporized and transported through a capillary column by an inert carrier gas. Components are separated according to volatility and interactions with the stationary phase. After separation, compounds enter the mass spectrometer, where they are ionized, commonly by electron ionization, and fragmented. The resulting ions are separated according to mass-to-charge ratio (m/z), producing a mass spectrum. The spectrum can be compared with reference libraries for tentative identification.

4. Instrumentation

A typical GC–MS system consists of a carrier-gas supply, injector, temperature-controlled oven, capillary column, transfer line, ion source, mass analyzer and detector. Helium, hydrogen or nitrogen may be used as carrier gases. Temperature-programmed GC is commonly employed for complex essential oils because constituents may have a wide range of volatilities. Quadrupole, ion-trap and time-of-flight analyzers are among the mass analyzers used in GC–MS systems.

5. Sample Preparation

Sample preparation strongly affects the measured volatile profile. Common approaches include hydrodistillation, steam distillation, solvent extraction, headspace sampling and headspace-solid-phase microextraction (HS-SPME). Hydrodistillation is widely used for essential oils, whereas headspace and HS-SPME are useful for analyzing highly volatile compounds with limited sample manipulation. Extraction temperature, time, plant moisture, storage and fiber characteristics in SPME can influence recovery and reproducibility.

6. Identification of Volatile Compounds

GC–MS identification generally combines chromatographic and spectral information. Retention time alone is not sufficient because it can vary with column and operating conditions. Mass spectral library searching provides candidate identities, while retention indices provide an additional independent parameter. NIST maintains evaluated electron-ionization spectral and GC retention-index libraries that support compound identification.^[6] For stronger confirmation, the experimental mass spectrum and retention index may be compared with an authentic standard analyzed under the same conditions.

7. Quantification of Volatile Compounds

Volatile constituents may be reported as relative percentages based on chromatographic peak areas or determined quantitatively using calibration standards. Relative composition is useful for describing essential-oil profiles, but peak-area percentage does not necessarily represent the actual concentration of each compound. Absolute quantification requires calibration, preferably using an internal standard or suitable reference standards. Important validation characteristics include linearity, accuracy, precision, limit of detection, limit of quantification and recovery.

8. Applications in Medicinal Plants

GC–MS has broad applications in medicinal-plant research. It is used for essential-oil characterization, volatile fingerprinting, chemotype differentiation, botanical authentication, quality control, adulteration assessment and investigation of bioactive volatile constituents. GC–MS profiles have also supported chemotaxonomic studies, particularly in aromatic plant families such as Lamiaceae. Differences in major constituents can help distinguish species or chemotypes, although chemical data should complement rather than replace botanical identification.

9. Advantages

Major advantages of GC–MS include good chromatographic separation, sensitive detection of many volatile constituents, structural information from mass spectra, extensive spectral libraries, small sample requirements and suitability for complex mixtures. The technique can provide both qualitative fingerprints and quantitative measurements when appropriately validated. Its combination of retention behavior and mass spectra makes it especially valuable for essential-oil and medicinal-plant analysis.

10. Limitations

GC–MS is mainly suitable for volatile and thermally stable compounds. Thermolabile compounds may degrade during injection or chromatographic separation. Co-elution can complicate spectral interpretation, and structurally similar compounds or stereoisomers may require additional confirmation. Differences in extraction procedures, columns and temperature programs can also produce different profiles. Therefore, standardized sample preparation, retention-index information and appropriate reference standards are important for reliable interpretation.

11. Future Perspectives

Advanced approaches such as comprehensive two-dimensional GC–MS (GC×GC–MS), high-resolution mass spectrometry, automated HS-SPME, chemometrics and metabolomics are expanding the capabilities of volatile analysis. Chemometric tools can assist in classification of species, geographical origins and chemotypes and may support adulteration assessment. The development of larger spectral and retention-index databases is also improving identification confidence.

12. Importance of GC–MS in Pharmaceutical Analysis of Medicinal Plants

The pharmaceutical importance of medicinal plants depends not only on the presence of biologically active constituents but also on the consistency and authenticity of the raw material. Volatile constituents can be used as chemical markers because their profiles may vary between species, chemotypes, geographical regions and processing conditions. GC–MS provides a practical approach for establishing these profiles. In pharmaceutical analysis, a characteristic GC–MS fingerprint can be compared between authenticated reference material and test samples to detect unusual changes or possible substitution. Such chemical information can complement pharmacognostical and physicochemical tests and can strengthen the overall quality-control strategy.

13. Extraction Techniques and Their Influence on GC–MS Results

The extraction technique determines which volatile constituents are recovered and therefore directly influences the GC–MS chromatogram. Hydrodistillation and steam distillation are commonly used to obtain essential oils, while headspace techniques analyze the volatile fraction released from the sample without necessarily isolating an oil. HS-SPME is particularly useful when only a small amount of sample is available or when solvent-free extraction is preferred. Different temperatures and extraction times can change the relative abundance of volatile compounds. Consequently, comparisons between samples should be performed using standardized extraction procedures. Recent studies continue to emphasize that extraction conditions are an important source of variation in volatile profiles.

14. Chromatographic Conditions for Volatile-Compound Analysis

Selection of GC conditions is important for obtaining good resolution. Column polarity, column length, internal diameter, film thickness, carrier-gas flow, injector temperature and oven-temperature program influence retention and peak separation. Non-polar columns are often used for essential-oil profiling, whereas more polar stationary phases may improve separation of selected oxygenated compounds. Temperature programming is commonly preferred for complex samples because it allows early-eluting volatile compounds and later-eluting sesquiterpenes to be analyzed in a single run. Optimization should aim for adequate resolution while maintaining reasonable analysis time.

15. Mass Spectral Interpretation

The mass spectrum is an important chemical fingerprint of a volatile compound. Electron ionization commonly produces reproducible fragmentation patterns, which is one reason EI libraries are extensively used in GC–MS. The analyst should examine the quality of the chromatographic peak before interpreting its spectrum. Poorly resolved or co-eluting peaks may produce mixed spectra and misleading library matches. A high similarity score is therefore best regarded as evidence supporting an identification rather than automatic proof of identity. Comparison of the observed spectrum with library spectra, retention-index data and, when necessary, authentic standards provides a more reliable identification strategy.

16. Retention Indices in Compound Identification

Retention indices are numerical values that describe the chromatographic retention behavior of compounds relative to reference hydrocarbons. They are particularly useful in essential-oil

analysis because many terpenes have similar electron-ionization spectra. The use of retention indices together with mass spectra can reduce the probability of incorrect assignments. NIST maintains a large GC retention-index collection integrated with its mass-spectral resources; the current NIST26 release contains more than 527,000 retention-index values covering more than 216,000 compounds. However, retention indices depend on column characteristics and experimental conditions, so values should be compared with appropriate literature or database data obtained under compatible conditions.

17. Quantitative Approaches

Quantification can be performed using external calibration, internal-standard calibration or response-factor approaches. In an external calibration method, several concentrations of a reference compound are analyzed and a calibration curve is prepared. In the internal-standard approach, a known amount of a suitable compound is added to standards and samples and the analyte response is expressed relative to the internal standard. Internal standards can improve precision by compensating for variations in injection and instrumental response. For regulatory or pharmaceutical applications, validated quantitative procedures are preferable to simple area normalization.

18. Method Validation for GC–MS

Validation demonstrates that a GC–MS method is suitable for its intended purpose. Specificity is important when a target compound is measured in a complex plant matrix. Linearity evaluates the relationship between concentration and response over the selected range. Accuracy may be assessed using recovery studies, while precision is evaluated using replicate measurements. Detection and quantification limits are particularly relevant when minor constituents are measured. Robustness evaluates whether small deliberate changes in analytical conditions significantly affect the result. A recent review of GC–MS method validation for volatile compounds emphasizes parameters such as linearity, precision, accuracy, LOD and LOQ as important components of reliable analysis.

19. Chemometric Analysis of GC–MS Data

GC–MS analysis of medicinal plants can generate large datasets containing numerous peaks. Chemometric methods convert these complex datasets into useful patterns. Principal component analysis (PCA) can visualize similarities and differences between samples, while hierarchical cluster analysis (HCA) can group samples according to their chemical composition. Partial least-squares methods may be used for classification or prediction when

appropriate datasets are available. Recent work on plant essential oils has demonstrated that GC–MS data combined with PCA can reveal chemical markers responsible for separation of species or groups. These approaches can be useful in authentication, chemotype differentiation and quality assessment.

20. GC–MS for Authentication and Adulteration Detection

Authentication of medicinal plants is essential because substitution with another species or dilution of an essential oil can alter chemical composition and potentially affect product quality. GC–MS can compare the volatile fingerprint of an unknown sample with a reference profile. Changes in the abundance of characteristic compounds or appearance of unexpected peaks may indicate differences in botanical source, processing or adulteration. However, a GC–MS fingerprint should normally be interpreted together with botanical, microscopic, physicochemical and other analytical evidence. Modern reviews of essential-oil quality assessment identify GC–MS and related chromatographic techniques as important tools for chemical profiling and authenticity evaluation.

21. Biological and Pharmacological Relevance of Volatile Constituents

Volatile compounds are not merely analytical markers; many have biological significance. Monoterpenes, sesquiterpenes and oxygenated derivatives have been investigated for antimicrobial, antioxidant, anti-inflammatory and other biological activities. Essential oils can contain many compounds acting together, and their biological effects may depend on both major and minor constituents. Therefore, GC–MS profiling is often an important first step before biological evaluation. The identification of chemical markers can help researchers relate chemical composition to observed biological activity, although GC–MS identification alone does not establish pharmacological efficacy.

22. Reproducibility and Reporting of GC–MS Studies

For a GC–MS study to be reproducible, sufficient experimental information should be reported. This includes botanical authentication, plant part, collection location and time, sample condition, drying procedure, extraction method, extraction yield, instrument model, column specification, carrier gas, flow rate, injection conditions, oven program, ionization mode, scan range and library used. Reporting retention indices and the basis for compound identification is also important. Quantitative studies should describe standards, calibration procedures, internal standards and validation parameters. Complete reporting makes it easier

to compare results between laboratories and improves the scientific value of medicinal-plant GC–MS studies.

23. Comparative Overview of Common GC–MS Sample-Preparation Methods

Hydrodistillation is useful for essential-oil isolation and is relatively simple, but heating can modify thermolabile compounds. Headspace analysis is rapid and minimizes direct handling of the sample, making it useful for highly volatile components. HS-SPME provides solvent-free concentration of volatile compounds and can be automated, but fiber selectivity and extraction conditions must be controlled. Solvent extraction can recover a broader range of semi-volatile constituents, but solvent residues and matrix components may complicate analysis. Selection should therefore be based on the research objective rather than assuming that one extraction method is universally superior.

24. Current Developments and Future Scope

The analytical field is moving toward higher resolution, better identification confidence and more automated data processing. Comprehensive two-dimensional GC coupled with MS can improve separation of very complex volatile mixtures. High-resolution MS can provide accurate-mass information and additional structural evidence. Automated headspace and SPME systems can increase throughput and reproducibility. Chemometrics and machine-learning approaches may support plant authentication and classification when sufficiently large and well-characterized datasets are available. The expansion of evaluated spectral and retention-index databases is also expected to improve routine identification. The 2026 NIST release substantially expanded its EI and retention-index resources, illustrating the continuing development of reference-data infrastructure for GC–MS analysis.

25. Practical Workflow for a Medicinal-Plant GC–MS Study

A systematic study can be organized into the following stages: (1) botanical authentication and documentation; (2) collection and controlled storage of plant material; (3) selection and standardization of sample preparation; (4) extraction of volatile constituents; (5) optimization of GC conditions; (6) acquisition of mass spectra; (7) library searching; (8) retention-index comparison; (9) confirmation with standards where required; (10) quantitative analysis and validation; and (11) statistical or chemometric interpretation. Following a standardized workflow reduces analytical variation and improves confidence in conclusions.

26. Overall Significance

The major contribution of GC–MS to medicinal-plant research is its ability to transform a complex volatile mixture into a chemically interpretable profile. It can reveal both major and minor constituents, provide evidence for plant identity, support comparison of samples and assist in quality evaluation. When combined with appropriate sample preparation, retention indices, spectral libraries, reference standards and validation, GC–MS becomes more than a screening technique; it can form an important component of a comprehensive pharmaceutical analytical strategy.

CONCLUSION

GC–MS is a powerful and widely applicable technique for identifying and quantifying volatile compounds in medicinal plants. It combines efficient chromatographic separation with mass-spectral information and can support essential-oil characterization, phytochemical profiling, authentication, chemotaxonomy and quality control. Reliable identification is strengthened by combining mass spectral matching with retention indices and, where appropriate, authentic standards. Although limitations such as thermal degradation, co-elution and quantitative variability remain, advances in multidimensional GC, high-resolution MS, automated extraction and chemometrics are expected to further improve medicinal-plant volatile analysis.

REFERENCES

1. Sadgrove NJ, Padilla-González GF, Phumthum M. Fundamental chemistry of essential oils and volatile organic compounds, methods of analysis and authentication. *Plants*. 2022; 11(6): 789.
2. Sparkman OD, Penton ZE, Kitson FG. *Gas Chromatography and Mass Spectrometry: A Practical Guide*. 2nd ed. Amsterdam: Academic Press; 2011.
3. Adams RP. *Identification of Essential Oil Components by Gas Chromatography/Mass Spectrometry*. 4th ed. Carol Stream, IL: Allured Publishing Corporation; 2007.
4. Belliardo F, Bicchi C, Cordero C, Liberto E, Rubiolo P, Sgorbini B. Headspace-solid-phase microextraction in the analysis of the volatile fraction of aromatic and medicinal plants. *J. Chromatogr. Sci.*, 2006; 44(7): 416–423.
5. Chandrasekar R, Sivagami B, Niharika K, Lavanya G, Hepsiba B, Kiran Kumari A, et al. GC-MS in quantification and characterization of phytoconstituents of herbal extracts: a review. *Pharmaceutical Research: Recent Advances and Trends*. 2024; 8: 155–178.

6. National Institute of Standards and Technology. NIST Standard Reference Database 1A: NIST/EPA/NIH Mass Spectral Library and NIST GC Retention Index Database. 2026.
7. McLafferty FW, Tureček F. Interpretation of Mass Spectra. 4th ed. Sausalito, CA: University Science Books; 1993.
8. Waseem R, Low KH. Advanced analytical techniques for the extraction and characterization of plant-derived essential oils by gas chromatography with mass spectrometry. *J. Sep. Sci.*, 2015; 38: 483–501.
9. Mondello L, Casilli A, Tranchida PQ, Dugo G, Dugo P. Evaluation of essential oils composition using both GC and MS fingerprints. *J. Chromatogr. A.*, 1999; 843: 269–276.
10. Snyder LR, Kirkland JJ, Dolan JW. Introduction to Modern Liquid Chromatography. 3rd ed. Hoboken, NJ: Wiley; 2010.
11. International Council for Harmonisation. ICH Q2(R2): Validation of Analytical Procedures. Geneva: ICH; 2023.
12. Chemotaxonomy, an efficient tool for medicinal plant identification: current trends and limitations. 2025.
13. Sadgrove NJ, Jones GL. A contemporary introduction to essential oils and volatile organic compounds: chemistry, analysis and authentication.
14. World Health Organization. WHO guidelines on good agricultural and collection practices (GACP) for medicinal plants. Geneva: World Health Organization; 2003.
15. Yang H, Huang X, Yang M, Zhang X, Tang F, Gao B, et al. Advanced analytical techniques for authenticity identification and quality evaluation in essential oils: a review. *Food Chem.*, 2024; 446: 139340.
16. Krause ST, Frey M. Extraction of essential oils and terpene volatiles from plants and identification by GC-MS-based techniques. *Methods Mol. Biol.*, 2025; 2895: 73–82.
17. Fauzan KAR, Bii MFW, Salsabila A, Rahma S, Wulandari D, Yanto A. Validation of gas chromatography-mass spectrometry (GC-MS) methods in the analysis of volatile compounds in natural materials. *Indonesia Natural Research Pharmaceutical Journal*. 2025; 10(2).
18. GC analyses of volatile constituents of plants from *Clausena* genus and computing the chemical variability by multivariate correlation analysis. *J. Herbs Spices Med. Plants*. 2025; 31(1).
19. Quality standards and pharmacological interventions of natural oils: current scenario and future perspectives. *Molecules*. 2023; 28: [article on natural-oil quality and chemical profiling].

20. Essential oils pharmacological activity: chemical markers, biogenesis, plant sources, and commercial products. *Process Biochem.*, 2024; 144: 112–132.