



Phytochemical Study and Isolation of Phytoconstituents from Cassia Fistula

¹Mr. Mahesh P. Junghare, ²Mr. Laxman S. Junghare, ³Mr. Yogesh V. Junghare, ⁴Mr. Akash B. Borkar

³Mr. Dnyaneshwar R. Bajad

¹HOD, ²Principal, ^{3,4&5} of Lecturer

Late Ramraoji Zanak College Of Pharmacy And Research Center, Kenwad, Ta. Risod, Dist. Washim

Abstract : Cassia fistula Linn., commonly known as the golden shower tree, is a medicinal plant widely used in traditional systems of medicine for its laxative, antimicrobial, antioxidant, and anti-inflammatory properties. The present study focuses on the phytochemical investigation and isolation of major phytoconstituents from different parts of Cassia fistula, including pods, leaves, and bark. Preliminary phytochemical screening of various solvent extracts (petroleum ether, chloroform, ethyl acetate, methanol, and aqueous) revealed the presence of secondary metabolites such as flavonoids, anthraquinones, tannins, saponins, terpenoids, glycosides, and phenolic compounds. Methanolic extracts showed a rich concentration of phenolics and flavonoids.

I. INTRODUCTION

Medicinal plants have garnered significant attention due to their vast and largely untapped reservoirs of chemical substances possessing considerable therapeutic potential, establishing them as invaluable sources for biomedicine ([Mwangi et al., 2021](#)). Among these, *Cassia fistula* Linn., commonly known as the golden shower tree, stands out as a prominent species recognized for its extensive pharmacological properties and traditional medicinal uses across various cultures ([Tanveer et al., 2019](#)). Originating from South Asian continents, *C. fistula* is cultivated globally for its rich content of phytoconstituents, including triterpenes, sugars, rhein, and potassium ([Christopher et al., 2023](#)). Various parts of the plant, including leaves, stems, roots, pods, and fruits, contain a diverse array of phytochemicals such as flavonoids, tannins, saponins, and anthraquinones, which contribute to its recognized antimicrobial, anti-inflammatory, and antidiabetic activities ([Ajayi et al., 2023](#)) ([Singh et al., 2013](#)). These diverse phytochemicals, encompassing alkaloids, phenolics, steroids, and glycosides, are individually or collectively responsible for the plant's therapeutic efficacy ([Shaikh & Patil, 2020](#)). The identification and isolation of these bioactive compounds are crucial for understanding their mechanisms of action and developing novel pharmaceutical agents ([Shaikh & Patil, 2020](#)). This comprehensive review will therefore detail the phytochemical investigation of *Cassia fistula*, elucidating the diverse classes of secondary metabolites present and their corresponding therapeutic relevance ([Shaikh & Patil, 2020](#)) ([Omer et al., 2022](#)). The subsequent sections will explore the methodologies employed for phytochemical extraction and isolation, critically evaluate the identified compounds, and discuss their potential applications in modern medicine, thereby advancing the understanding of *C. fistula*'s pharmacological profile. Specifically, this investigation delves into the influence of solvent polarity on extraction efficiency and the comprehensive phytochemical profiles obtainable from *Cassia fistula* ([Jayashika, 2025](#)).

Literature Review :

Further investigation into the phytochemical profile of *C. fistula* is warranted to isolate and characterize novel compounds responsible for its diverse medicinal applications, thereby validating its ethnomedicinal claims through modern pharmacological studies.

This comprehensive review aims to consolidate current knowledge regarding the phytochemical constituents isolated from *Cassia fistula* and their associated biological activities, providing a foundation for future drug discovery and development ([Ajayi et al., 2023](#)) ([Rao et al., 2023](#)). Furthermore, understanding the intricate mechanisms through which these compounds exert their effects could pave the way for the development of new therapeutic agents against various human ailments, particularly in the context of emerging antimicrobial resistance ([Chakraborty et al., 2022](#)). The exploration of natural compounds from medicinal plants, such as *C. fistula*, offers a promising avenue for discovering new antimicrobial agents, given that herbal drugs often serve as safer and time-tested sources for human therapeutic use ([S. et al., 2024](#)). For instance, other *Cassia* species, such as *Cassia alata*, contain potent phytochemicals like kaempferol, apigenin, quercetin, rhein, and rutin, which are crucial for wound healing processes ([Keng et al., 2024](#)). Similarly, the leaves of *C. fistula* contain a wide variety of antioxidants, including terpenoids, flavonoids, alkaloids, and phenolic compounds, contributing to its medicinal properties ([Naseer et al., 2020](#)). Additionally, the therapeutic potential of *Cassia* species extends to antimicrobial, antioxidant, and anti-diabetic activities, attributable to their rich phenolic and flavonoid content ([Alshehri et al., 2022](#)). Specifically, plant-based antibacterial compounds like coumarins exhibit significant activity against *Staphylococcus aureus*, while berberine is potent against Gram-positive bacteria, including methicillin-resistant *Staphylococcus aureus* ([Imon et al., 2023](#)).

Methodology

The subsequent sections detail the systematic approaches employed for the extraction, isolation, and structural elucidation of these bioactive compounds.

This involves qualitative and quantitative phytochemical screening using established protocols, such as those described by Rahman et al., to detect key classes like phenolics, tannins, and cardiac glycosides ([Imon et al., 2023](#)).

Further analyses often incorporate advanced chromatographic techniques such as High-Performance Liquid Chromatography and Gas Chromatography-Mass Spectrometry for precise identification and quantification of individual compounds ([Zibae et al., 2023](#)). These methods enable a detailed exploration of the plant's chemical complexity, facilitating the correlation between specific phytoconstituents and their observed pharmacological effects ([Hafez et al., 2019](#)). Such detailed characterization is crucial for validating the traditional uses of *Cassia fistula* and for exploring its potential in developing new phytomedicines, especially given the rising interest in natural compounds for treating metabolic syndromes ([Alshehri et al., 2022](#)) and infectious diseases ([Mohammed et al., 2023](#)). Specifically, recent breakthroughs in phytochemical and pharmacological research of genus *Senna*, which includes *Cassia*, underscore the necessity of further isolating active constituents and elucidating their structures to validate traditional medicinal claims ([Diab, 2024](#)). This integrated approach, combining traditional knowledge with modern analytical techniques, offers a powerful strategy for identifying novel therapeutic agents from *Cassia fistula* ([Imon et al., 2023](#)) ([Gebrehiwot et al., 2024](#)). The emphasis on such studies is particularly relevant today, as global interest in natural medicines continues to surge, necessitating rigorous research into plants like *Cassia fistula* to fully comprehend their therapeutic potential and mechanisms of action ([Thaker et al., 2023](#)). The intricate interplay between these identified phytochemicals and their synergistic effects often contributes to the holistic therapeutic efficacy observed in traditional medicine, warranting a deeper exploration into their combined biological activities ([Alsheikh et al., 2020](#)). Moreover, the advancements in analytical techniques, including LC-MS and NMR, significantly expedite the identification and characterization of these intricate compounds, bridging traditional knowledge with modern pharmacological understanding ([Atanassova et al., 2025](#)) ([Chihomvu et al., 2024](#)). This rigorous analytical approach, complemented by computational techniques like molecular docking and QSAR modeling, facilitates the prediction of potential targets and guides experimental validation efforts ([Chihomvu et al., 2024](#)). Advanced spectroscopic methods, including High-Performance Liquid Chromatography and Gas Chromatography-Mass Spectrometry, are predominantly employed for both the analysis and characterization of metabolites based on their specific chemical compositions ([Mohsin et al., 2024](#)). High-resolution mass spectrometry, particularly Quadrupole Time-of-Flight, offers detailed information regarding accurate masses and chemical formulas of

phytochemicals, which is critical for their characterization prior to isolation ([Masota et al., 2023](#)). Furthermore, metabolomics, an advanced 'omic' technique, plays a crucial role in comprehensively analyzing phytochemicals and is increasingly being utilized for quality control of herbal drugs, allowing for a deeper understanding of the bioactive compounds present in plant samples ([Mukherjee et al., 2016](#)) ([Rani et al., 2023](#)). This holistic approach provides a comprehensive understanding of the plant's chemical diversity and its potential pharmacological applications ([Ahmad et al., 2022](#)) ([Gahlaut et al., 2013](#)). These advanced analytical techniques, often coupled with in silico methods such as molecular docking and virtual screening, enable researchers to predict and elucidate the interactions of plant-derived compounds with various biological targets, offering insights into their therapeutic potential or toxicological risks ([Singh et al., 2025](#)).

Results

The application of these sophisticated methods has yielded significant progress in identifying and quantifying numerous compounds within *Cassia fistula*, highlighting a diverse phytochemical landscape. For instance, the synergistic application of techniques like GC-MS and LC-MS, supported by NMR spectroscopy, has been instrumental in profiling the complex matrix of this species, revealing a wide array of metabolites including amino acids, sugars, organic acids, alkaloids, steroids, and terpenoids ([Singh et al., 2025](#)). Such comprehensive metabolomic studies, whether targeted or untargeted, involve meticulous sample processing, data acquisition, and sophisticated data analysis to fully characterize the chemical complexity ([Trifan et al., 2024](#)). These advanced analytical approaches enable the discovery of novel bioactive compounds, providing a deeper understanding of the plant's medicinal properties ([Cadena-Zamudio et al., 2022](#)). Many studies have leveraged various solvents for extraction and employed gas chromatography and liquid chromatography-mass spectrometry to profile a wide range of metabolites, including phenolic acids, flavonoids, terpenes, and alkaloids ([Singh et al., 2025](#)). The subsequent sections will elaborate on the specific compounds isolated and characterized from *Cassia fistula*, alongside their proposed biological activities and potential therapeutic applications. This foundational work is crucial for establishing a robust scientific basis for its traditional uses and paves the way for further pharmacological investigations into specific isolates. Further, these detailed phytochemical investigations often involve an initial screening using spectrometric and chromatographic methods to gather preliminary data on chemical and pharmacological activities, guiding the selection of biologically active plant extracts ([Konappa et al., 2020](#)). Additionally, the integration of artificial intelligence in phytochemical research is increasingly pivotal, offering novel approaches for the elucidation of novel potential phytochemicals and enabling the efficient analysis of high-dimensional 'omics' data ([Varghese et al., 2025](#)). The utilization of advanced analytical techniques, such as hyphenated platforms combining chromatography with spectroscopy, allows for the accurate identification, distribution, and quantification of phytochemicals, facilitating the de novo discovery of constituents in biogenic materials ([Gashaw et al., 2025](#)).

Discussion

These sophisticated approaches are critical for overcoming the challenges associated with the structural elucidation of complex natural products and for advancing our understanding of their therapeutic mechanisms ([Varghese et al., 2025](#)). The inherent complexity and variability of botanical extracts necessitate the employment of such advanced techniques to ensure accurate separation and detection of bioactive compounds ([Wu et al., 2012](#)). High-resolution mass spectrometry provides detailed insights into these plant substances, identifying their location within plant tissues, while nuclear magnetic resonance spectroscopy facilitates compound structure elucidation without complex setups ([Shehu et al., 2025](#)). Furthermore, advanced metabolomics techniques are becoming indispensable for comprehensively annotating the constituents of plant natural products, thereby aiding in screening, drug discovery, and quality control of phytomedicines ([Salem et al., 2020](#)). Despite significant advancements, challenges remain in fully elucidating the global plant metabolome due to the structural diversity and complexity of specialized metabolites, especially alkaloids, and the inadequate metabolomics infrastructure ([Tsugawa et al., 2021](#)). This underscores the need for continued innovation in analytical methodologies and computational tools to achieve a more complete understanding of the phytochemical cosmos ([Tsugawa et al., 2021](#)). However, recent technological strides, particularly in artificial intelligence, have begun to address these limitations by enabling high-throughput data processing and interpretation, thus accelerating the discovery and characterization of novel phytochemicals ([Varghese et al., 2025](#)).

Conclusion

The continuous integration of genomics, transcriptomics, and advanced metabolomics, including spatial multi-omics techniques like MALDI/DESI-MS, is vital for linking novel metabolites to their genetic origins and understanding their roles in plant phenotypes (Tsugawa et al., 2021). Such approaches allow for a deeper understanding of biosynthetic pathways and the evolutionary trajectory of specialized metabolites within diverse plant lineages (Uckele et al., 2021). These advanced multi-omics strategies, particularly when combined with machine learning and comparative genomics, significantly enhance our ability to prioritize and characterize unknown mass features identified through metabolomics, ultimately accelerating natural product discovery (Tsugawa et al., 2021) (Wu et al., 2023) (Wang et al., 2024). Moreover, advancements in high-throughput sequencing and emerging techniques like single-cell and spatial omics are providing unprecedented insights into cell-specific and spatially resolved biosynthetic dynamics (Wang et al., 2024). The increasing number of sequenced plant genomes also facilitates comparative genomics and phylogenetic analyses to pinpoint phytochemical diversity hotspots (Tsugawa et al., 2021). This systematic integration can lead to a more comprehensive mapping of the plant kingdom's metabolic potential, thereby guiding targeted searches for novel bioactive compounds (Tsugawa et al., 2021). Furthermore, phylogenomics-based metabolite structure prediction offers a promising avenue for identifying functional phytochemical diversity beyond well-characterized biodiversity hotspots (Tsugawa et al., 2021).

References

- Ahmad, S., Katiyar, C. K., Ulrich-Merzenich, G., & Mukherjee, P. K. (2022). Editorial: Metabolomics and Ethnopharmacology in the Development of Herbal and Traditional Medicine. *Frontiers in Pharmacology*, 13. <https://doi.org/10.3389/fphar.2022.851023>
- Ajayi, V. A., Adebayo, T. E., & Lateef, A. (2023). Novel multitasking gold nanoparticles biosynthesized by *Cassia fistula*: antifungal, anti- obesity, anti-diabetic, and anti-ulcer activities. *Research Square (Research Square)*. <https://doi.org/10.21203/rs.3.rs-3590139/v1>
- Alshehri, M. M., Quispe, C., Herrera-Bravo, J., Sharifi-Rad, J., Tütüncü, S., Aydar, E. F., Topkaya, C., Mertdinç, Z., Özçelik, B., Aital, M., Kumar, N. V. A., Lapava, N., Rajković, J., Ertani, A., Nicola, S., Semwal, P., Painuli, S., González-Contreras, C., Martorell, M., ... Cho, W. C. (2022). A Review of Recent Studies on the Antioxidant and Anti-Infectious Properties of Senna Plants [Review of *A Review of Recent Studies on the Antioxidant and Anti-Infectious Properties of Senna Plants*]. *Oxidative Medicine and Cellular Longevity*, 2022, 1. Hindawi Publishing Corporation. <https://doi.org/10.1155/2022/6025900>
- Alsheikh, H. M. A., Sultan, I., Kumar, V., Rather, I. A., Al-Sheikh, H., Jan, A. T., & Haq, Q. M. R. (2020). Plant-Based Phytochemicals as Possible Alternative to Antibiotics in Combating Bacterial Drug Resistance [Review of *Plant-Based Phytochemicals as Possible Alternative to Antibiotics in Combating Bacterial Drug Resistance*]. *Antibiotics*, 9(8), 480. Multidisciplinary Digital Publishing Institute. <https://doi.org/10.3390/antibiotics9080480>
- Atanassova, M., Zahnit, W., Messaoudi, M., Rebiai, A., Benchikha, N., & Sawicka, B. (2025). *Analytical Techniques for Phytochemical Analysis and Functional Investigation* (p. 151). https://doi.org/10.1007/978-981-96-2790-5_6
- Cadena-Zamudio, J. D., Monribot-Villanueva, J. L., Pérez-Torres, C.-A., Alatorre-Cobos, F., Jiménez-Moraila, B., Guerrero-Analco, J. A., & Ibarra-Laclette, E. (2022). The use of ecological analytical tools as an unconventional approach for untargeted metabolomics data analysis: the case of *Cecropia obtusifolia* and its adaptive responses to nitrate starvation. *Functional & Integrative Genomics*, 22(6), 1467. <https://doi.org/10.1007/s10142-022-00904-1>
- Chakraborty, A. K., Saha, S., Poria, K., Samanta, T., Gautam, S., & Mukhopadhyay, J. (2022). A saponin-polybromophenol antibiotic (CU1) from *Cassia fistula* Bark Against Multi-Drug Resistant Bacteria Targeting RNA polymerase. *Current Research in Pharmacology and Drug Discovery*, 3, 100090. <https://doi.org/10.1016/j.crphar.2022.100090>
- Chihomvu, P., Ganesan, A., Gibbons, S., Woollard, K., & Hayes, M. A. (2024). Phytochemicals in Drug Discovery—A Confluence of Tradition and Innovation [Review of *Phytochemicals in Drug Discovery*—

A Confluence of Tradition and Innovation]. *International Journal of Molecular Sciences*, 25(16), 8792. Multidisciplinary Digital Publishing Institute. <https://doi.org/10.3390/ijms25168792>

9. Christopher, I., Sounderraajan, A., Murugesan, V., Indu, S., Periyasamy, V., & Manikkam, R. (2023). Molecular docking analysis of triterpenoids from *Cassia fistula* with breast cancer targets. *Bioinformation*, 19(11), 1067. <https://doi.org/10.6026/973206300191067>
10. Diab, M. (2024). Review Article on Biological Activities and Chemical Constituents of Genus *Senna*. *SINAI International Scientific Journal.*, 1(1), 73. <https://doi.org/10.21608/sisj.2024.371239>
11. Gahlaut, A., Shirolkar, A., Hooda, V., & Dabur, R. (2013). A rapid and simple approach to discriminate various extracts of *Saraca asoca* [Roxb.], De. Wild using UPLC-QTOFMS and multivariate analysis. *Journal of Pharmacy Research*, 7(2), 143. <https://doi.org/10.1016/j.jopr.2013.03.005>
12. Gashaw, A. D., Desta, M. A., & Yaya, E. E. (2025). A comprehensive review-current development in spectroscopic and chromatographic techniques for natural product analysis. *Results in Chemistry*, 16, 102341. <https://doi.org/10.1016/j.rechem.2025.102341>
13. Gebrehiwot, H., Ensermu, U., Dekebo, A., Endale, M., & Hunsen, M. (2024). Exploring the medicinal potential of *Senna siamea* roots: an integrated study of antibacterial and antioxidant activities, phytochemical analysis, ADMET profiling, and molecular docking insights. *Applied Biological Chemistry*, 67(1). <https://doi.org/10.1186/s13765-024-00899-2>
14. Hafez, S. A., Othman, S., Ibrahim, H. A., Seida, A. A., & Ayoub, N. (2019). Chemical Constituents and Biological Activities of *Cassia* Genus: Review. *Archives of Pharmaceutical Sciences Ain Shams University*, 3(2), 195. <https://doi.org/10.21608/aps.2019.15746.1008>
15. Imon, R. R., Talukder, Md. E. K., Akhter, S., Islam, Md. S., Ahammad, F., Anis-Ul-Haque, K. M., Moniruzzaman, Md., Afroze, M., Khan, M., Jamal, M. A. H. M., Wani, T. A., Uddin, M. J., & Rahman, Md. M. (2023). Natural defense against multi-drug resistant *Pseudomonas aeruginosa*: *Cassia occidentalis* L. in vitro and in silico antibacterial activity. *RSC Advances*, 13(41), 28773. <https://doi.org/10.1039/d3ra03923d>
16. Jayashika, B. (2025). Solvent-Dependent Phytochemical Diversity in *Justicia tranquebariensis*: A Comparative Analysis of Methanol and Ethyl Acetate Extractions. *International Journal For Multidisciplinary Research*, 7(3). <https://doi.org/10.36948/ijfmr.2025.v07i03.43181>
17. Keng, J.-W., Lee, S.-K., Sang, S.-H., Liew, K. B., Teo, S., Mossadeq, W. M. S., Chow, S.-C., Akowuah, G. A., Lee, S.-K., Mai, C., & Chew, Y.-L. (2024). *Cassia alata* and Its Phytochemicals: A Promising Natural Strategy in Wound Recovery. *Sci*, 6(2), 34. <https://doi.org/10.3390/sci6020034>
18. Konappa, N., Udayashankar, A. C., Krishnamurthy, S., Pradeep, C. K., Chowdappa, S., & Jogaiah, S. (2020). GC-MS analysis of phytoconstituents from *Amomum nilgircum* and molecular docking interactions of bioactive serverogenin acetate with target proteins. *Scientific Reports*, 10(1). <https://doi.org/10.1038/s41598-020-73442-0>
19. Masota, N. E., Zehe, M., Vogg, G., Ohlsen, K., Meinel, L., & Holzgrabe, U. (2023). Searching for new agents against Enterobacteriaceae from nature: approaches, potential plant species, isolated compounds, and their respective properties. *Phytochemistry Reviews*, 23(3), 863. <https://doi.org/10.1007/s11101-023-09902-y>
20. Mohammed, B. S., Sutramay, P., Ahmadi, S., Fathima, S., Askani, S., Jambiga, P. C., Thumma, R., Dharavath, S. B., & Taduri, S. (2023). PHYTOCHEMICAL SCREENING AND ANTI-BACTERIAL ACTIVITY OF *ERYTHRINA VARIEGATA* LEAF, STEM AND ROOT EXTRACTS. *Journal of Plant Development*, 30(1), 77. <https://doi.org/10.47743/jpd.2023.30.1.927>
21. Mohsin, H. F., Eshak, Z., Sultan, S., Ahmad, W., Amini, M. H., & Ashraf, K. (2024). GCMS and LCMS/MS Based Analysis for the Identification and Characterization of Bioactive Metabolites of Seaweed *Kappaphycus striatus* as an Anti-hypertensive and Anti-hypercholesterolemic Agent. *Indian Journal of Pharmaceutical Education and Research*, 58(4), 1287. <https://doi.org/10.5530/ijper.58.4.141>
22. Mukherjee, P. K., Harwansh, R. K., Bahadur, S., Biswas, S., Kuchibhatla, L. N., Tetali, S. D., & Raghavendra, A. S. (2016). Metabolomics of Medicinal Plants - A Versatile Tool for Standardization of

Herbal Products and Quality Evaluation of Ayurvedic Formulations. *Current Science*, 111(10), 1624. <https://doi.org/10.18520/cs/v111/i10/1624-1630>

23. Mwangi, R. W., Macharia, J. M., Wagara, I. N., & Raposa, B. L. (2021). The medicinal properties of *Cassia fistula* L: A review [Review of *The medicinal properties of Cassia fistula L: A review*]. *Biomedicine & Pharmacotherapy*, 144, 112240. Elsevier BV. <https://doi.org/10.1016/j.biopha.2021.112240>
24. Naseer, M., Aslam, U., Khalid, B., & Chen, B. (2020). Green route to synthesize Zinc Oxide Nanoparticles using leaf extracts of *Cassia fistula* and *Melia azadarach* and their antibacterial potential. *Scientific Reports*, 10(1). <https://doi.org/10.1038/s41598-020-65949-3>
25. Omer, H. A. A., Caprioli, G., Abouelenein, D., Mustafa, A. M., Uba, A. I., Ak, G., Ozturk, R. B., Zengin, G., & Yagi, S. (2022). Phenolic Profile, Antioxidant and Enzyme Inhibitory Activities of Leaves from Two *Cassia* and Two *Senna* Species. *Molecules*, 27(17), 5590. <https://doi.org/10.3390/molecules27175590>
26. Rani, S., Puri, R., Boora, P., Qasim, A., Angmo, D., & Mehta, M. P. (2023). GC-MS, FT-IR, and ICP-MS Analysis of Bioactive Compounds in the Methanolic Leaves Extract of *Thalictrum foliolosum*. *Pharmacognosy Research*, 15(4), 705. <https://doi.org/10.5530/pres.15.4.075>
27. Rao, A. V., Shikha, K., Laura, J. S., & Dhanial, G. (2023). Qualitative Phytochemical Screening of Medicinal Plants Using Different Solvent Extracts. *Oriental Journal Of Chemistry*, 39(3), 621. <https://doi.org/10.13005/ojc/390312>
28. S., R. T., N., S. P., & Sarita, D. (2024). Antioxidant, urobactericidal, and antibiotic modulating activity of a parasitic medicinal plant: *Cuscuta reflexa* Roxb. *Journal of Medicinal Plants Research*, 18(4), 63. <https://doi.org/10.5897/jmpr2024.7341>
29. Salem, M. A., Souza, L. P. de, Serag, A., Fernie, A. R., Farag, M. A., Ezzat, S. M., & Alseekh, S. (2020). Metabolomics in the Context of Plant Natural Products Research: From Sample Preparation to Metabolite Analysis [Review of *Metabolomics in the Context of Plant Natural Products Research: From Sample Preparation to Metabolite Analysis*]. *Metabolites*, 10(1), 37. Multidisciplinary Digital Publishing Institute. <https://doi.org/10.3390/metabo10010037>
30. Shaikh, J. R., & Patil, M. (2020). Qualitative tests for preliminary phytochemical screening: An overview. *International Journal of Chemical Studies*, 8(2), 603. <https://doi.org/10.22271/chemi.2020.v8.i2i.8834>
31. Shehu, A., Pandey, P., & Kushwaha, R. (2025). *Advanced analytical techniques for the identification of plant derived bioactive compounds*. 2(1). <https://doi.org/10.70130/rcs.2025.0201004>
32. Singh, S., Singh, S., & Yadav, A. K. (2013). A Review on *Cassia* species: Pharmacological, Traditional and Medicinal Aspects in Various Countries [Review of *A Review on Cassia species: Pharmacological, Traditional and Medicinal Aspects in Various Countries*]. *American Journal of Phytomedicine and Clinical Therapeutics*, 1(3), 291. <http://pakacademicsearch.com/pdf-files/sci/268/291-312%20Vol%201%20Issue%203%20June%202013.pdf>
33. Singh, U., Almahasheer, H., Emwas, A. H., & Jaremko, M. (2025). Untargeted metabolic profiling of *Zygophyllum Coccineum* plant near the Red Sea for phytochemical investigation by GC-MS, LC-MS, and NMR. *Journal of Saudi Chemical Society*, 29(1). <https://doi.org/10.1007/s44442-025-00003-3>
34. Tanveer, S., Latif, A., Ashiq, K., & Bajwa, M. (2019). A COMPREHENSIVE REVIEW ON PHARMACOLOGICAL AND PHYTOCHEMICAL POTENTIAL OF *CASSIA FISTULA* LINN: A MAGICAL HERB [Review of *A COMPREHENSIVE REVIEW ON PHARMACOLOGICAL AND PHYTOCHEMICAL POTENTIAL OF CASSIA FISTULA LINN: A MAGICAL HERB*]. *International Journal of Biology Pharmacy and Allied Sciences*, 8(6). <https://doi.org/10.31032/ijbpas/2019/8.6.4734>
35. Thaker, K., Patoliya, J., Rabadiya, K., Reddy, N. R. R., & Joshi, R. (2023). *Senna (Cassia angustifolia Vahl): A comprehensive review of ethnopharmacology and phytochemistry* [Review of *Senna (Cassia angustifolia Vahl): A comprehensive review of ethnopharmacology and phytochemistry*]. *Pharmacological Research - Natural Products*, 1, 100003. Elsevier BV. <https://doi.org/10.1016/j.prenap.2023.100003>

36. Trifan, A., Wolfram, E., Skalicka-Woźniak, K., & Luca, S. V. (2024). Symphytum genus—from traditional medicine to modern uses: an update on phytochemistry, pharmacological activity, and safety. *Phytochemistry Reviews*. <https://doi.org/10.1007/s11101-024-09977-1>
37. Tsugawa, H., Rai, A., Saito, K., & Nakabayashi, R. (2021). Metabolomics and complementary techniques to investigate the plant phytochemical cosmos [Review of *Metabolomics and complementary techniques to investigate the plant phytochemical cosmos*]. *Natural Product Reports*, 38(10), 1729. Royal Society of Chemistry. <https://doi.org/10.1039/d1np00014d>
38. Uckele, K. A., Jahner, J. P., Tepe, E. J., Richards, L. A., Dyer, L. A., Ochsenrider, K. M., Philbin, C. S., Kato, M. J., Yamaguchi, L. F., Forister, M. L., Smilanich, A. M., Dodson, C. D., Jeffrey, C. S., & Parchman, T. L. (2021). Phytochemistry reflects different evolutionary history in traditional classes versus specialized structural motifs. *Scientific Reports*, 11(1). <https://doi.org/10.1038/s41598-021-96431-3>
39. Varghese, R., Shringi, H., Efferth, T., & Ramamoorthy, S. (2025). Artificial intelligence driven approaches in phytochemical research: trends and prospects. *Phytochemistry Reviews*. <https://doi.org/10.1007/s11101-025-10096-8>
40. Wang, M., Zhang, S., Li, R., & Zhao, Q. (2024). Unraveling the specialized metabolic pathways in medicinal plant genomes: a review [Review of *Unraveling the specialized metabolic pathways in medicinal plant genomes: a review*]. *Frontiers in Plant Science*, 15, 1459533. Frontiers Media. <https://doi.org/10.3389/fpls.2024.1459533>
41. Wu, H., Guo, J., Chen, S., Liu, X., Zhou, Y., Zhang, X., & Xu, X. (2012). Recent developments in qualitative and quantitative analysis of phytochemical constituents and their metabolites using liquid chromatography–mass spectrometry [Review of *Recent developments in qualitative and quantitative analysis of phytochemical constituents and their metabolites using liquid chromatography–mass spectrometry*]. *Journal of Pharmaceutical and Biomedical Analysis*, 72, 267. Elsevier BV. <https://doi.org/10.1016/j.jpba.2012.09.004>
42. Wu, M., Northen, T. R., & Ding, Y. (2023). Stressing the importance of plant specialized metabolites: omics-based approaches for discovering specialized metabolism in plant stress responses [Review of *Stressing the importance of plant specialized metabolites: omics-based approaches for discovering specialized metabolism in plant stress responses*]. *Frontiers in Plant Science*, 14. Frontiers Media. <https://doi.org/10.3389/fpls.2023.1272363>
43. Zibae, E., Javadi, B., Sobhani, Z., Akaberi, M., Farhadi, F., Amiri, M. S., Baharara, H., Sahebkar, A., & Emami, S. A. (2023). Cassia species: A review of traditional uses, phytochemistry and pharmacology [Review of *Cassia species: A review of traditional uses, phytochemistry and pharmacology*]. *Pharmacological Research - Modern Chinese Medicine*, 9, 100325. Elsevier BV. <https://doi.org/10.1016/j.prmcm.2023.100325>