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Phytochemical fingerprinting techniques for the authentication of medicinal plants

[1]Hamad Al- Yazidi, [2]Hana Al- Romaihi
[1, 2] Department of Pharmacy, Qatar Rehabilitation Institute, Hamad Medical Corporation, Doha, Qatar

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Abstract
Phytochemical fingerprinting is a crucial technique for the authentication and quality control of medicinal plants. This method involves the identification and characterization of bioactive compounds unique to specific plant species. As the use of medicinal plants in healthcare continues to grow, ensuring their authenticity and purity becomes increasingly important. This article reviews the various phytochemical fingerprinting techniques employed in the authentication of medicinal plants, highlighting their principles, applications, and advantages.

Keywords: Phytochemical fingerprinting, authentication, quality control

Introduction
Medicinal plants have been used for centuries across various cultures for their therapeutic properties. With the growing interest in natural remedies and alternative medicine, the demand for medicinal plants has surged. However, issues such as adulteration, substitution, and misidentification pose significant challenges to their safe and effective use. Phytochemical fingerprinting has emerged as a reliable method for authenticating medicinal plants, ensuring their quality, safety, and efficacy.
Phytochemical fingerprinting involves creating a profile of the chemical constituents of a plant. This profile serves as a unique identifier, akin to a fingerprint, that can distinguish one plant species from another. Techniques such as chromatography, spectroscopy, and molecular analysis are commonly used to generate these fingerprints. By comparing the phytochemical profiles of samples with reference standards, it is possible to authenticate the plant material and detect any adulteration or substitution.

Objective
The main objective of this article is to review the various phytochemical fingerprinting techniques used for the authentication of medicinal plants, ensuring their quality, safety, and efficacy.

Chromatographic Techniques
Chromatography is a cornerstone technique in phytochemical fingerprinting, extensively used for its precision, reproducibility, and ability to handle complex mixtures. It separates compounds based on their interactions with a stationary phase and a mobile phase, producing distinctive profiles that can be used to identify and authenticate medicinal plants. High-Performance Liquid Chromatography (HPLC) is one of the most powerful tools for phytochemical analysis. It separates and quantifies non-volatile and thermally labile compounds, making it ideal for analyzing diverse phytochemicals in medicinal plants. In HPLC, a liquid sample passes through a column packed with a solid stationary phase under high pressure, separating compounds that elute at different times, known as retention times. This separation is monitored by a detector, producing a chromatogram that represents the unique phytochemical fingerprint of the sample. HPLC is widely used for authenticating medicinal plants by comparing chromatographic profiles with reference standards, ensuring the consistency and quality of plant materials. For instance, the analysis of flavonoids, alkaloids, and glycosides in plants like Ginkgo biloba, Hypericum perforatum (St. John's wort), and Panax ginseng can be effectively performed using HPLC. The technique offers high resolution and sensitivity, capable of detecting phytochemicals at very low concentrations, and can be coupled with various detectors such as UV-Vis, fluorescence, and mass spectrometry (LC-MS), enhancing its analytical capabilities.
Gas Chromatography (GC) is another pivotal technique in phytochemical fingerprinting, particularly for analyzing volatile and semi-volatile compounds. In GC, the sample is vaporized and carried through a column coated with a stationary phase using an inert gas. Compounds are separated based on their volatility and interactions with the stationary phase, resulting in distinct retention times. The separated compounds are detected by a flame ionization detector (FID) or mass spectrometry (GC-MS), producing a chromatogram. GC is widely used to analyze essential oils, terpenes, and other volatile phytochemicals. For example, the authentication of lavender oil, eucalyptus oil, and other essential oils relies on GC to identify their characteristic compounds and detect adulteration by identifying synthetic additives or substitutions with cheaper oils. GC offers excellent separation efficiency and sensitivity for volatile compounds, and when coupled with mass spectrometry, it provides detailed structural information, enabling precise identification of phytochemicals.
Thin Layer Chromatography (TLC) is a simpler and more cost-effective chromatographic technique used for preliminary screening and fingerprinting of phytochemicals. In TLC, a small amount of sample is applied onto a plate coated with a thin layer of stationary phase (usually silica gel). The plate is then developed in a solvent system, causing the compounds to separate based on their affinity for the stationary phase and solubility in the mobile phase. The separated compounds appear as distinct spots, which can be visualized using UV light or chemical reagents. TLC

is used for the rapid screening of phytochemicals in medicinal plants, such as alkaloids, flavonoids, and saponins. It is particularly useful for detecting adulteration and assessing the overall quality of plant materials. For example, the TLC profiling of secondary metabolites in plants like Centella asiatica (Gotu kola) and Echinacea purpurea provides a quick and reliable method for their authentication. TLC is simple, inexpensive, and requires minimal sample preparation, allowing the simultaneous analysis of multiple samples and providing a visual representation of the phytochemical profile. Although less sensitive than HPLC or GC, TLC is effective for qualitative analysis and preliminary screening.

Chromatographic techniques such as HPLC, GC, and TLC are indispensable tools for phytochemical fingerprinting and the authentication of medicinal plants. These methods provide detailed and reliable profiles of the chemical constituents, ensuring the quality, safety, and efficacy of plant materials. By employing these techniques, the authenticity of medicinal plants can be verified, adulteration can be detected, and the overall integrity of herbal products can be maintained. As the demand for medicinal plants continues to grow, chromatographic fingerprinting will play a vital role in supporting the development and regulation of herbal medicine

Spectroscopic Techniques

Spectroscopy is another vital set of techniques in phytochemical fingerprinting, used to identify and quantify compounds based on their interaction with light. These methods provide unique spectral signatures that can be used to authenticate medicinal plants by creating detailed profiles of their chemical constituents. Several spectroscopic techniques are particularly valuable in this context, including Ultraviolet-Visible (UV-Vis) Spectroscopy, Fourier Transform Infrared (FTIR) Spectroscopy, and Nuclear Magnetic Resonance (NMR) Spectroscopy.

Ultraviolet-Visible (UV-Vis) Spectroscopy measures the absorbance of UV or visible light by a sample. This technique identifies and quantifies phytochemicals based on their specific absorbance spectra. It is simple, rapid, and cost-effective, making it suitable for preliminary screening and fingerprinting of medicinal plants. For example, UV-Vis spectroscopy is used to analyze flavonoids and phenolic acids, which absorb light in the UV-visible range, providing a characteristic spectrum that can be compared with reference standards to confirm the identity of the plant material.

Fourier Transform Infrared (FTIR) Spectroscopy measures the absorption of infrared light by a sample, producing a spectrum that represents the molecular vibrations within the sample. FTIR is used to identify functional groups and provide information about the molecular structure of phytochemicals. This non-destructive technique requires minimal sample preparation and is suitable for the rapid authentication of medicinal plants. FTIR can be used to differentiate between species and detect adulteration by comparing the spectral fingerprints of samples with those of authentic reference materials. For instance, FTIR has been used to authenticate the essential oils of different plant species by identifying unique absorption bands corresponding to specific functional groups.

Nuclear Magnetic Resonance (NMR) Spectroscopy provides detailed information about the molecular structure and dynamics of compounds. It involves placing the sample in a magnetic field and measuring the interaction of nuclear spins with radiofrequency pulses. NMR is highly sensitive and can provide comprehensive fingerprints of complex mixtures, making it invaluable for the authentication of medicinal plants. NMR spectroscopy can identify and quantify multiple components simultaneously, providing a complete profile of the phytochemicals present. For example, NMR has been used to analyze the saponin content in ginseng and the alkaloid profile in various medicinal plants, ensuring their authenticity and quality.

Spectroscopic techniques like UV-Vis, FTIR, and NMR are essential tools in the phytochemical fingerprinting and authentication of medicinal plants. They provide detailed and reliable profiles of the chemical constituents, ensuring the quality, safety, and efficacy of plant materials. By employing these techniques, the authenticity of medicinal plants can be verified, adulteration can be detected, and the overall integrity of herbal products can be maintained. As the use of medicinal plants continues to grow, spectroscopic fingerprinting will play a crucial role in supporting the development and regulation of herbal medicine.

Molecular Techniques

Molecular techniques are essential for the authentication of medicinal plants, providing genetic-level insights that complement phytochemical fingerprinting. These methods analyze the genetic material of plants, offering precise identification and differentiation between species. The most prominent molecular techniques used in this field include DNA barcoding and Polymerase Chain Reaction (PCR)-based methods.

DNA Barcoding involves sequencing a short, standardized region of the plant's DNA and comparing it with a reference database. This technique is highly specific and can differentiate between closely related species. DNA barcoding typically targets regions such as the chloroplast gene matK or the nuclear ribosomal internal transcribed spacer (ITS) region. By comparing the sequence obtained from a sample with sequences in a barcode database, researchers can accurately identify the plant species. DNA barcoding is particularly useful for the authentication of medicinal plants that are morphologically similar or processed in a way that obscures their physical characteristics. For instance, DNA barcoding has been used to distinguish between different species of Echinacea and verify the authenticity of ginseng products.

Polymerase Chain Reaction (PCR) amplifies specific DNA sequences, enabling their detection and analysis. Various PCR-based techniques are employed for phytochemical fingerprinting, including Random Amplified Polymorphic DNA (RAPD) and Amplified Fragment Length Polymorphism (AFLP). These methods generate unique genetic fingerprints that can be used to authenticate medicinal plants and detect adulteration.

RAPD uses short, random primers to amplify random segments of the genome, producing a set of DNA fragments that can be visualized on a gel. The pattern of these fragments is unique to each species, providing a genetic fingerprint. RAPD is relatively quick and simple, making it suitable for initial screening. However, its reproducibility can be influenced by experimental conditions.

AFLP is a more sophisticated technique that involves the digestion of genomic DNA with restriction enzymes,

followed by the selective amplification of a subset of these fragments. AFLP generates highly reproducible and informative fingerprints, allowing for the detailed genetic characterization of medicinal plants. This method is particularly useful for distinguishing between closely related species and assessing genetic diversity within plant populations.

Molecular techniques like DNA barcoding and PCR-based methods are powerful tools for the authentication and quality control of medicinal plants. They provide precise and reliable identification at the genetic level, ensuring the authenticity and purity of plant materials. These techniques can detect adulteration, substitution, and contamination, which are common issues in the herbal medicine industry. By integrating molecular techniques with traditional phytochemical fingerprinting, a comprehensive approach to the authentication and quality assurance of medicinal plants can be achieved.

In conclusion, molecular techniques play a crucial role in the phytochemical fingerprinting and authentication of medicinal plants. They offer genetic-level precision and reliability, complementing other analytical methods to ensure the quality, safety, and efficacy of herbal products. As the use of medicinal plants continues to grow, the integration of molecular techniques will be essential in supporting the development and regulation of herbal medicine.

Conclusion

Phytochemical fingerprinting techniques are indispensable tools for the authentication and quality control of medicinal plants. Chromatographic methods such as High-Performance Liquid Chromatography (HPLC), Gas Chromatography (GC), and Thin Layer Chromatography (TLC) provide detailed profiles of chemical constituents, ensuring the precise identification and purity of plant materials. Spectroscopic techniques, including Ultraviolet-Visible (UV-Vis) Spectroscopy, Fourier Transform Infrared (FTIR) Spectroscopy, and Nuclear Magnetic Resonance (NMR) Spectroscopy, offer unique spectral signatures that aid in the rapid and accurate authentication of medicinal plants. Molecular techniques like DNA barcoding and PCR-based methods add an additional layer of precision by providing genetic-level identification, crucial for distinguishing closely related species and detecting adulteration. The integration of these techniques creates a comprehensive approach to the authentication of medicinal plants, safeguarding their quality, safety, and efficacy. As the demand for herbal medicines continues to grow, the importance of reliable and accurate authentication methods becomes even more critical. These techniques not only help in verifying the authenticity of medicinal plants but also play a vital role in detecting contamination, adulteration, and substitution, which are prevalent issues in the herbal medicine industry.

References

1. Ram M, Abdin MZ, Khan MA, Jha P. HPTLC fingerprint analysis: a quality control for authentication of herbal phytochemicals. High-performance thin-layer chromatography (HPTLC); c2011. p. 105-116.
2. Saraf A, Saraf A, Chaturvedi A. Phytochemical analysis and chemical fingerprinting of seeds of *Abrus precatorius* L. Chemical Science Transactions. 2018;7(1):63-70.
3. Obradovič M, Krajšek SS, Dermastia M, Kreft S. A new method for the authentication of plant samples by analyzing fingerprint chromatograms. Phytochemical Analysis: An International Journal of Plant Chemical and Biochemical Techniques. 2007 Mar;18(2):123-132.
4. Yadav D, Reshi MS, Uthra C, Shrivastava S, Srivastava N, Narayana SK, *et al*. Botanical and chemical fingerprinting of medicinal roots of Justicia gendarussa burm f. Pharmacognosy research. 2017 Apr;9(2):208.
5. Dhami N, Mishra AD. Phytochemical variation: how to resolve the quality controversies of herbal medicinal products? Journal of herbal medicine. 2015 Jun 1;5(2):118-127.
6. Selvakumari E, Jenifer J, Priyadharshini S, Vinodhini R. Application of DNA fingerprinting for plant identification. JAIR. 2017 Mar;5(10):149-151.
7. Donno D, Beccaro GL, Mellano MG, Bonvegna L, Bounous G. *Castanea* spp. buds as a phytochemical source for herbal preparations: Botanical fingerprint for nutraceutical identification and functional food standardisation. Journal of the Science of Food and Agriculture. 2014 Nov;94(14):2863-2873.
8. Mohammed Abubakar B, Mohd Salleh F, Shamsir Omar MS, Wagiran A. DNA barcoding and chromatography fingerprints for the authentication of botanicals in herbal medicinal products. Evidence-Based Complementary and Alternative Medicine; c2017 Apr 27.