

Extraction, Isolation and Characterisation of Bioactive Compounds

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Perspective

Because of the unrivalled abundance of chemical variety, natural products derived from medicinal plants, whether as pure compounds or standardised extracts, give limitless prospects for new therapeutic leads. Interest in edible plants has developed around the world as a result of a growing demand for chemical diversity in screening programmes and a desire to find therapeutic medications from natural sources. Bioactive substances can be found in botanicals and herbal medicines for medical use. The analytical approaches, which include the extraction, isolation, and characterisation of active components in botanicals and herbal medicines, are the subject of this research. Clinical trials aimed at determining the pharmacokinetics, bioavailability, efficacy, safety, and drug interactions of newly produced bioactive substances and their formulations (extracts) must be thoroughly assessed. Clinical trials are meticulously prepared to protect participants' health as well as to answer specific research questions by assessing both immediate and long-term adverse effects, as well as their outcomes, before the treatment is extensively used on patients. The application of conventional phytochemical screening tests, as well as chromatographic techniques such as HPLC and TLC, to the study of bioactive chemicals found in plant extracts.

Due to the obvious unprecedented abundance of chemical variety, natural products, such as plant extracts, either as pure chemicals or as standardised extracts, give limitless prospects for new drug discoveries. More than 80% of the world's population, according to the World Health Organization (WHO), rely on traditional medicine for their primary healthcare requirements. Men turned to ethnopharmacognosy as chemically generated medications started to cause side effects and microbial resistance. Thousands of phytochemicals derived from plants were discovered to be safe, effective, and have fewer side effects. Anticancer, antibacterial, antioxidant, antidiarrheal, analgesic, and wound healing properties have all been described. Many people say that certain natural or herbal products provide them with significant benefits. Clinical trials aimed at determining the pharmacokinetics, bioavailability, efficacy, safety, and drug interactions of newly produced bioactive substances and their formulations (extracts) must be thoroughly assessed. Clinical trials are meticulously prepared to protect participants' health as well as to answer specific research questions by assessing both immediate and long-term adverse effects, as well as their

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outcomes, before the treatment is extensively used on patients. The World Health Organization (WHO) estimates that nearly 20,000 medicinal plants exist in 91 countries, including 12 mega biodiversity countries. Extraction, pharmacological screening, isolation and characterization of bioactive compounds, toxicological evaluation, and clinical evaluation are the first steps in utilising biologically active compounds derived from plant resources.

Extraction, Identification and characterization

Extraction is the first and most important step in the analysis of medicinal plants because it is necessary to extract the desired chemical components from plant materials for further separation and characterization. The basic operation included steps such as pre-washing, drying of plant materials or freeze drying, grinding to obtain a homogeneous sample, and frequently improving the kinetics of analytic extraction as well as increasing sample surface contact with the solvent system. To ensure that potential active constituents are not lost, distorted, or destroyed during the extraction of plant samples, proper precautions must be taken. The specific nature of the bioactive compound being targeted heavily influences the solvent system selection. There are various solvent systems available for extracting bioactive compounds from natural products. Other modern extraction techniques with advantages include solid-phase micro-extraction, supercritical-fluid extraction, pressurized-liquid extraction, microwave-assisted extraction, solid-phase extraction, and surfactant-mediated techniques.

Because plant extracts are typically a mixture of different types of bioactive compounds or phytochemicals with varying polarities, their separation remains a significant challenge in the

identification and characterization of bioactive compounds. To obtain pure compounds, it is common practise to use a variety of separation techniques such as TLC, column chromatography, flash chromatography, Sephadex chromatography, and HPLC. The pure compounds are then used to determine structure and biological activity.

Chromatographic techniques

TLC (Thin-layer chromatography) is a simple, quick, and low-cost procedure that provides the researcher with an immediate answer as to how many components are present in a mixture. When the R_f of a compound is compared to the R_f of a known compound, TLC is used to support the identity of a compound in a mixture. Other tests include spraying phytochemical screening reagents, which cause colour changes based on the phytochemicals present in a plant extract, or viewing the plate under a UV light. Bio-autography is an effective method for determining bioactive compounds with antimicrobial activity in plant extracts. TLC

bioautographic methods combine chromatographic separation and in situ activity determination, making it possible to localise and isolate active constituents in a mixture.

HPLC (High Performance Liquid Chromatography) is a versatile, robust, and widely used technique for natural product isolation. Nowadays, this technique is attracting attention among various analytical techniques as the primary choice for fingerprinting research for herbal plant quality control. Chemical separations can be accomplished using HPLC by taking advantage of the fact that different compounds migrate at different rates given a specific column and mobile phase.

Even though bioactive compounds found in plant material are multi-component mixtures, their separation and determination continue to pose challenges. To isolate bioactive compounds, the majority of them must be purified using a combination of chromatographic techniques and other purification methods.