



e-ISSN: 2319-8753 | p-ISSN: 2320-6710

IJIRSET

International Journal of Innovative Research in
SCIENCE | ENGINEERING | TECHNOLOGY

INTERNATIONAL JOURNAL OF INNOVATIVE RESEARCH

IN SCIENCE | ENGINEERING | TECHNOLOGY

Volume 10, Issue 1, January 2021

ISSN INTERNATIONAL
STANDARD
SERIAL
NUMBER
INDIA

Impact Factor: 7.512

9940 572 462

6381 907 438

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A Review on Extraction, Characterization and Structural Determination of *Aerva lanata*

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ABSTRACT: The study of valuable plant medicine was first described precisely in the basic literature Charak Samhita of Ayurveda medicine in India. "*Aerva lanata* (Linn) Juss. Ex Schult" is an annual herb of family Amaranthaceae. This plant are not only medicinally importance but are significantly serving several other scientific purposes. Calorimetric determination do not provide accurate data and have to determine as approximate. For investigation of plant aerva lanata many methods are used for Extraction, Characterization and structural determination. Separation techniques like (HPLC, TLC, GC, CC, OPLC, HPTLC) provides excellent and qualitative informations. For spectral analysis (UV-Visible, FTIR, NMR, Mass spectroscopy) gives standardized structure and good for identification of compounds. The common challenges in the characterization and structural determination of medicinal plant *Aerva lanata* are discussed. The main aim of this article review is focus on comprehensive view of methods of analytical methodologies of medicinal plant *Aerva lanata*.

KEYWORDS: *Aerva lanata*, Extraction, Characterization, whole plant, Structural Determination

I. INTRODUCTION

Natural medicines not only provide the primary health-care needs for the majority of the population in developing countries but also have attracted more and more attention in the developed countries due to the high health care cost and low or no side effect. More than 80% of the world's population relies on traditional medicine for their primary healthcare needs, most of which involve the use of plant extracts [1]. Medicinal plants are the richest bio-resource of drugs of traditional systems of medicine, nutraceuticals, food supplements, pharmaceutical intermediates, chemical entities for crude. This review paper takes a comprehensive look at the different extraction methods of the bioactive natural products. It mostly focused on the analytical methodologies, which includes the extraction methods, isolation and characterization of the natural products present in the plant extracts. The medicinal plants and products, minerals has been primary and important source of healthcare in fight against various physical health problems of 85 percent of rural people including poor in urban area [2]. The importance and role of these traditional healthcare systems will not diminish in the future because they are both culturally viable, safer to health and expected to remain affordable while the modern healthcare service is both limited and expensive [3].

II. EXTRACTION METHODS

Extraction is the separation of medicinally active portions of plant using selective and standard procedures. It is the crucial first step in the analysis of medicinal plants, because it is necessary to extract the desired chemical components from the plant materials for further separation and characterization [4]. There are several extractions methods for the separation of natural product from plants present.

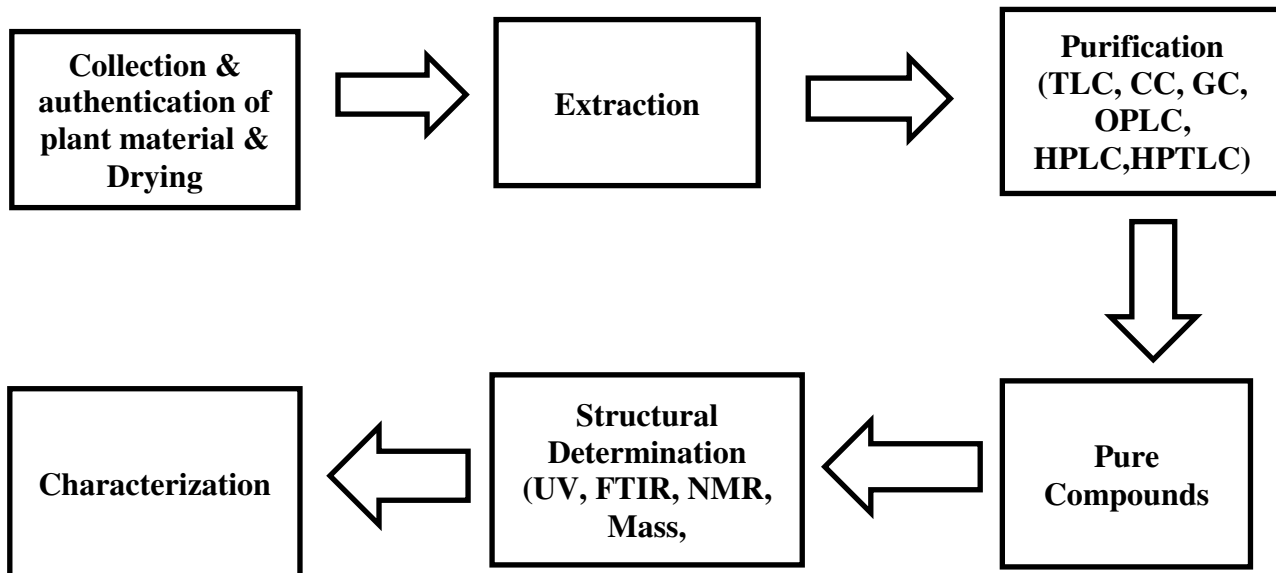


Fig. 1. A Brief Summary in Extraction, Isolation and Characterization of Natural Products from Medicinal Plant *Aerva lanata*

1) Soxhlet Extraction

Named after ‘Franz Ritter von Soxhlet’, a German agricultural chemist, it is the best method for the continuous extraction of a solid by a hot solvent [5]. Soxhlet apparatus is a specialized glass refluxing unit mainly used for organic solvent extractions. The powdered solid material is placed in a thimble made up of filter paper and is placed inside the soxhlet apparatus. The apparatus is fitted to a round bottomed (RB) flask containing the solvent and to a reflux condenser. The solvent in the RB flask is boiled gently, the vapor passes up through the side tube, condensed by the condenser and falls into the thimble containing the material and slowly fills the soxhlet. When the solvent reaches the top of the attached tube it siphons over into the flask, thus removes the portion of the substance, which it has extracted.

2) Ultrasonic Extraction

In this process, natural compounds are liberated from the plant tissues by high frequency sound, which damage the cell wall. Ultrasound assisted extraction can be used with mixtures of immiscible solvents such as hexane with methanol/water. The process creates heat so that heat labile compounds may decompose [6]. In such cases the extraction container is placed in ice bath to reduce the temperature.

3) Solid Phase Extraction (SPE)

This rapid, economical and sensitive technique uses different types of cartridges and disks, with a variety of sorbents, where the solute molecules are preferentially attached over the stationary phase. Sample preparation and concentration can be achieved in a single step [7]. Normal phase, reverse phase and ion exchange solid phase extraction units are available.

III. SEPARATION AND CHARACTERIZATION METHODS

A. Thin layer chromatography

TLC is the most commonly used planar chromatographic method in natural product research. This is the easiest and cheapest technique and can be applied in the analysis, isolation and setting the parameters for column chromatography [4]. Usually, silica or alumina (more polar) is used as the stationary phase and organic solvents (less polar) are used as the mobile phase. This situation is categorized as normal phase chromatography. In contrast to this, reverse phase TLC is available, in which stationary phase is alkyl bonded silica or alumina (less polar) and mobile phase is polar solvent like water, alcohol etc.

B. Column Chromatography (CC)

Column chromatography is the most effective technique used in separation of crude plant extracts into its components in pure form. This is a preparative chromatographic method and the stationary phase (silica gel) is packed in a column and then the mobile phase (eluent) is passed through the column after loading the extracts on the top of the stationary phase. The mobile phase carries the natural products present in the mixture at different rate based on their affinities to the stationary and mobile phase. Separated compounds can be collected along with the mobile phase [4].

C. High Performance Liquid Chromatography (HPLC)

It is a versatile, robust, and widely used technique for the isolation of natural products. HPLC is an analytical technique for the separation and determination of organic and inorganic solutes in any samples especially biological, pharmaceutical, food, environmental, industrial etc. Currently, this technique is gaining popularity among various analytical techniques as the main choice for fingerprinting study for the quality control of medicinal plants. In order to identify any compound by HPLC, a detector must first be selected, The extent or degree of separation is mostly determined by the choice of stationary phase and mobile phase. Modern HPLC uses a non-polar solid phase, like C18 and a polar liquid phase, generally a mixture of water and another solvent. High pressure up to 400 bars is required to elute the analyte through column before they pass through a diode array detector (DAD). A DAD measures the absorption spectra of the analytes to aid in their identification. HPLC is useful for compounds that cannot be vaporized or that decompose under high temperature and it provides a good complement to gas chromatography for detection of compounds [8].

D. High Performance thin Layer Chromatography (HPTLC)

It is a planar chromatography where separation of natural compounds is achieved on high performance layers with detection and data acquisition. These high performance layers are pre-coated plates coated with a sorbent of particle size 5-7 microns and a layer thickness of 150-200 microns. The reduction in thickness of layer and particle size results in increasing the plate efficiency as well as nature of separation [9]. HPTLC plates are substantially more expensive (4- to 6-times more) than normal plates but are an efficient alternative when high sensitivity, accuracy and precision are required in situations demanding high performance [10].

E. Optimum performance laminar chromatography (OPLC)

It is a new concept in parallel chromatography; OPLC combines the advantages of both TLC and HPTLC. OPLC is both an analytical and preparative tool, suitable for research and quality control laboratories. It is a powerful liquid chromatography separation technique that combines the user- friendly interface and resolution of HPLC with the capacity of flash chromatography and multi dimensionality of TLC. The basis of OPLC is similar to that of other chromatographic techniques in that a pump is used to force a liquid mobile phase through a stationary phase, such as silica. The OPLC columnhousing structure allows flat planar columns to be used in the same way as cylindrical glass or stainless steel ones. The flat column is pressurized up to 50 bars and mobile phase is forced through it at constant linear velocity via a

solvent delivery pump [11].

F. Gas Chromatography (GC)

It is an analytical technique for separating compounds based primarily on their volatilities. GC provides both qualitative and quantitative information for individual compounds present in a sample. The gas phase is flowing and the liquid phase is stationary. The rate of migration for the chemical species is determined through its distribution in the gas phase. For example, a species that distributes itself 100% into gas phase will migrate at the same rate as the flowing gas, whereas, a species that distributes itself 100% into stationary phase will not migrate at all. Species that distribute themselves partly in both phases will migrate at an intermediate rate [11]. Gas chromatography involves a sample being vaporized and injected onto the head of the chromatographic column. The sample is then transported through the column by the flow of inert, gaseous mobile phase. The column itself contains a liquid stationary phase, which is adsorbed onto the surface of an inert solid.

IV. STRUCTURE DETERMINATION

Determination of the structure of natural products uses data from a wide range of spectroscopic techniques such as UV-Visible, Infrared (IR), Nuclear Magnetic Resonance (NMR) and Mass spectroscopy. The basic principle of spectroscopy is passing electromagnetic radiation through an organic compound that absorbs some of the radiation, but not all. By measuring the amount of absorption of electromagnetic radiation, a spectrum can be produced. The spectra are specific to certain bonds in a compound. Depending on these spectra, the structure of the natural compound can be identified. Scientists mainly use spectra produced from either three or four regions—Ultraviolet (UV), Visible, Infrared (IR), Radio frequency (FTIR), and electron beam for structural clarification [12].

A. UV – Visible Spectroscopy

UV-visible spectroscopy can be used for identification of certain classes of compounds in both pure and biological mixtures. Preferentially, UV-visible spectroscopy can be used for quantitative analysis because aromatic molecules are powerful chromophores in the UV range. Natural compounds can be determined by using UV-visible spectroscopy [13]. Moreover, spectroscopic UV-Vis techniques were found to be less selective and give information on the composition of the total polyphenol content. This technique is not time-consuming, and presents reduced cost compared to other techniques [14].

B. Fourier Transform Infrared Spectroscopy (FTIR)

Fourier-transform infrared spectroscopy is a valuable tool for the identification of functional groups present in the plant extract. It helps for identification and structure determination of the molecule [14]. It is a high-resolution analytical tool to identify the chemical constituents and elucidate the structural compounds. FTIR offers a rapid and nondestructive investigation to fingerprint herbal extracts or powders.

C. Nuclear Magnetic Resonance Spectroscopy (NMR)

Nuclear Magnetic Resonance Spectroscopy gives physical, chemical and biological properties of matter. One dimensional technique is routinely used but the complicated structure of the molecules could be achieved through two dimensional NMR techniques. Solid state NMR spectroscopy is used for the determination of molecular structure of solids. Radiolabeled ^{13}C NMR is used to identify the types of carbon present in the compound. ^1H -NMR is used to find out types of hydrogen present in the compound and to find out how the hydrogen atoms are connected [11].

D. Mass Spectroscopy

Mass spectrometry is a powerful analytical technique for the identification of unknown compounds, quantification of known compounds and to elucidate the structure and chemical properties of molecules. Through MS spectrum, the molecular weight of sample can be determined. This method is mostly employed for the structural elucidation of organic compounds, for peptide or oligonucleotide sequencing and for monitoring the existence of previously characterized compounds in complex mixtures with a high specificity by defining both the molecular weight and a diagnostic fragment of the molecule simultaneously [11].

V. CONCLUSION

From the above studies we can conclude that bioactive compounds occurring in plant material consist of mixtures, their purification and characterization methods still create problems. The current investigation demonstrates that the separation of medicinal plants creates a big challenge for the process of identification of components present in plants. Most of them have to be purified by the several chromatographic techniques for identification and characterization. We can use various other methods for to isolate bioactive compounds.

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