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GENERAL TECHNIQUES OF EXTRACTION OF ACTIVE CONSTITUENTS IN HERBAL DRUGS

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ABSTRACT

Medicinal plants provide unlimited opportunities for new leads due to their innumerable chemical diversity. Extraction is the initial step to separate the desired natural products from the raw materials. Extraction methods include solvent extraction, distillation method, pressing and sublimation according to the extraction principle. The conventional extraction methods, including maceration, percolation and reflux extraction, usually use organic solvents and require a large volume of solvents and long extraction time. Some modern or greener extraction methods such as super critical fluid extraction (SFC), pressurized liquid extraction (PLE) and microwave assisted extraction (MAE), have also been applied in natural products extraction.

KEYWORDS: maceration, percolation, super critical fluid extraction (SFC), pressurized liquid extraction (PLE), microwave assisted extraction (MAE).

INTRODUCTION

Medicinal plants are those plants rich in secondary metabolites and are potential source of drugs. These secondary metabolites include alkaloids, glycosides, coumarins, flavonoids, steroids. Extraction is the first step to separate the desired natural products from the raw materials. Extraction methods include solvent extraction, distillation method, pressing and sublimation according to the extraction principle. The conventional extraction methods, including maceration, percolation and reflux extraction, usually use organic solvents and require a large volume of solvents and long extraction time.

Some modern or greener extraction methods such as super critical fluid extraction (SFC), pressurized liquid extraction (PLE) and microwave assisted extraction (MAE), have also been applied in natural products extraction, and they offer some advantages such as lower organic solvent consumption, shorter extraction time and higher selectivity.[1]

Solvent extraction

Solvent extraction is the most widely used method. The extraction of natural products progresses through the following stages:

(1) solvent penetrates into the solid matrix

(2) solute dissolves in the solvents

(3) solute is diffused out of the solid matrix

(4) extracted solutes are collected.

Any factor enhancing the diffusivity and solubility in the above steps will facilitate the extraction. The properties of the extraction solvent, the particle size of the raw materials, the solvent-to-solid ration, the extraction temperature and the extraction duration will affect the extraction efficiency.

The selection of the solvent is critical for solvent extraction. Selectivity, solubility, cost and safety should be considered in selection of solvents. Solvents with a polarity value near to the polarity of the solute are likely to perform better and vice versa. Alcohols are universal solvents in solvent extraction for phytochemical investigation.

Generally, the finer the particle size, the better result the extraction achieves. Too fine particle size, however, will cost the excessive absorption of solute in solid and difficulty in subsequent filtration. High temperatures increase the solubility and diffusion. Temperatures that too high, however, may cause solvents to be lost, leading to extracts of undesirable impurities and the decomposition of thermolabile components.

The extraction efficiency enhances with the increase in extraction duration in a certain time range.[1,2]

Simple extraction method

If the desired ingredients are fat-soluble, we can use an organic solvent such as benzene, chloroform or ether with water conducting liquid-liquid extraction to remove water-soluble ingredients such as sugars, inorganic salts. If the desired ingredients are hydrophilic

substance, the water solution can be extracted with the weak lipophilic solvents such as ethyl acetate, butanol, pentanol acetate. Sometimes we can add a small amount of methanol or ethanol in chloroform or methylene chloride to extract. We often use PH gradient extraction method in the separation of alkaloids, so that making strong alkaline alkaloids and weak alkaline alkaloids to achieve initial separation. The extraction and separation of herbal to achieve active ingredients by liquid – liquid extraction method, often according to the differences of the nature of active ingredients or in coexistence of impurities, have one or a certain type of component distribution coefficient changed significantly with some methods. PH gradient extraction method is also based on a component can form salt or be free under a certain PH, changed the distribution coefficient of the composition in a solvent system and separate from the other component.

Addition with polar solvent water "wash" the extraction of lipophilic solvent to remove mixed polar impurities, or with a lipophilic solvent “wash the lipophilic impurities of water extraction. These can be used repeatedly extraction method to complete.[3]

Continuous extraction method

Use of a continuous extractor, overcome the problems of using a separating funnel to extract many times. This instrument uses the different specific gravities of two solvents stratify naturally and dispersed droplets pass through the continuous phase solvent to occur mass transfer. The specific methods of operation is adding herbal aqueous solvent in the tube after solvent extraction, it can automatically flow into the heater, evaporation into the gas, after met the condenser to condense into the liquid, then extracted, and so endless cycle. This method is simple and can avoid the emulsifying, because the two-phases are flowing conditions to meet countercurrent and always maintain a large density difference, the extraction process can be carried out continuously, so the amount of solvent is lesser and the efficiency of extraction much higher.[4]

Solid Phase Extraction method

Solid Phase Extraction method is a solid liquid extraction technique. Here the compounds are suspended or dissolved in a liquid mixture and separated from other compounds according to their chemical and physical properties. This is according to the different strength of the interaction of extracted components and the other components of the sample at a fixed filler to make them separate from each other. It uses a solid -phase extraction column, usually is polypropylene column, also are glass or stainless steel column, add the extractant into the

column. Commonly used octadecyl silane chemically bonded silica or phenyl alkyl bonded silica, Now there are a cyano group, an amino or other special group filler to supply, upper and lower ends are covered with glass sand core or other porous filter. Add the sample into the column, make it flow through the solid phase extraction agent, the extracted sample was retained into the extraction agent, the solvent and other substances which are retained difficultly to outflows from the column, and then further using appropriate detergents to elute these unwanted components. Finally, using the eluent to elute the sample which is in the extraction column to obtain the desired compounds. For example, using SPE to measure the determination of caffeine content in coffee: make the coffee solution passing the extraction column which is filled C18 bonded silica, after the sample flowing through the column bed, with a certain volume of water to rinse firstly, and then dried under reduced pressure, finally with chloroform eluting the adsorbed caffeine for content determination. The advantage of this method is simple equipment, easy operation, fast speed, and can avoid emulsification that the simple extraction could cause, and the resulted extraction is no need to dry, it is suitable for the separation of trace components. SPE depend on the different separation purposes selecting different solid adsorbents and elution solvent.[5,6]

Maceration

In this process, the whole or coarsely powdered crude drug is placed in a stoppered container with the solvent and allowed to stand at room temperature for a period of at least 3 days with frequent agitation until the soluble matter has dissolved. The mixture then is strained, the marc (the damp solid material) is pressed, and the combined liquids are clarified by filtration or decantation after standing.

Infusion

Fresh infusions are prepared by macerating the crude drug for a short period of time with cold or boiling water. These are dilute solutions of the readily soluble constituents of crude drugs.[7]

Digestion

This is a form of maceration in which gentle heat is used during the process of extraction. It is used when moderately elevated temperature is not objectionable. The solvent efficiency of the menstruum is thereby increased.

Decoction

In this process, the crude drug is boiled in a specified volume of water for a defined time; it is then cooled and strained or filtered. This procedure is suitable for extracting water-soluble, heat stable constituents. This process is typically used in preparation of Ayurvedic extracts called “quath” or “kawath”. The starting ratio of crude drug to water is fixed, the volume is then brought down to one-fourth its original volume by boiling during the extraction procedure. Then, the concentrated extract is filtered and used as such or processed further.

Percolation

This is the procedure used most frequently to extract active ingredients in the preparation of tinctures and fluid extracts. A percolator (a narrow, cone-shaped vessel open at both ends) is generally used. The solid ingredients are moistened with an appropriate amount of the specified menstruum and allowed to stand for approximately 4 h in a well closed container, after which the mass is packed and the top of the percolator is closed. Additional menstruum is added to form a shallow layer above the mass, and the mixture is allowed to macerate in the closed percolator for 24 h. The outlet of the percolator then is opened and the liquid contained therein is allowed to drip slowly. Additional menstrum is added as required, until the percolate measures about three-quarters of the required volume of the finished product. The marc is then pressed and the expressed liquid is added to the percolate. Sufficient menstruum is added to produce the required volume, and the mixed liquid is clarified by filtration or by standing following by decanting.[7-9]

Hot Continuous Extraction

In this method, the finely ground crude drug is placed in a porous bag or “thimble” made of strong filter paper, which is placed in chamber E of the Soxhlet apparatus. The extracting solvent in flask A is heated, and its vapors condense in condenser D. The condensed extractant drips into the thimble containing the crude drug, and extracts it by contact. When the level of liquid in chamber E rises to the top of siphon tube C, the liquid contents of chamber E siphon into flask A. This process is continuous and is carried out until a drop of solvent from the siphon tube does not leave residue when evaporated. The advantage of this method, compared to previously described methods, is that large amounts of drug can be extracted with a much smaller quantity of solvent. This effects tremendous economy in terms of time, energy and consequently financial inputs. At small scale, it is employed as a batch

process only, but it becomes much more economical and viable when converted into a continuous extraction procedure on medium or large scale.[10]

Aqueous Alcoholic Extraction by Fermentation

Some medicinal preparations of Ayurveda adopt the technique of fermentation for extracting the active principles. The extraction procedure involves soaking the crude drug, in the form of either a powder or a decoction (kasaya), for a specified period of time, during which it undergoes fermentation and generates alcohol which facilitates the extraction of the active constituents contained in the plant material. The alcohol thus generated also serves as a preservative. If the fermentation is to be carried out in an earthen vessel, it should not be new: water should first be boiled in the vessel. In large-scale manufacture, wooden vats, porcelain jars or metal vessels are used in place of earthen vessels. Examples are like karpurasava, kanakasava, dasmularista.[11]

Counter-current Extraction

In counter-current extraction (CCE), wet raw material is pulverized using toothed disc disintegrators to produce a fine slurry. In this process, the material to be extracted is moved in one direction (generally in the form of a fine slurry) within a cylindrical extractor where it comes in contact with extraction solvent. The further the starting material moves, the more concentrated the extract becomes. Complete extraction is thus possible when the quantities of solvent and material and their flow rates are optimized. The process is highly efficient, requiring little time and posing no risk from high temperature. Finally, sufficiently concentrated extract comes out at one end of the extractor while the marc (practically free of visible solvent) falls out from the other end.

This extraction process has significant advantages:

i) A unit quantity of the plant material can be extracted with much smaller volume of solvent as compared to other methods like maceration, decoction, percolation.

ii) CCE is commonly done at room temperature, which spares the thermolabile constituents from exposure to heat which is employed in most other techniques.

iii) As the pulverization of the drug is done under wet conditions, the heat generated during comminution is neutralized by water. This again spares the thermolabile constituents from exposure to heat.

iv) The extraction procedure has been rated to be more efficient and effective than continuous hot extraction.[12]

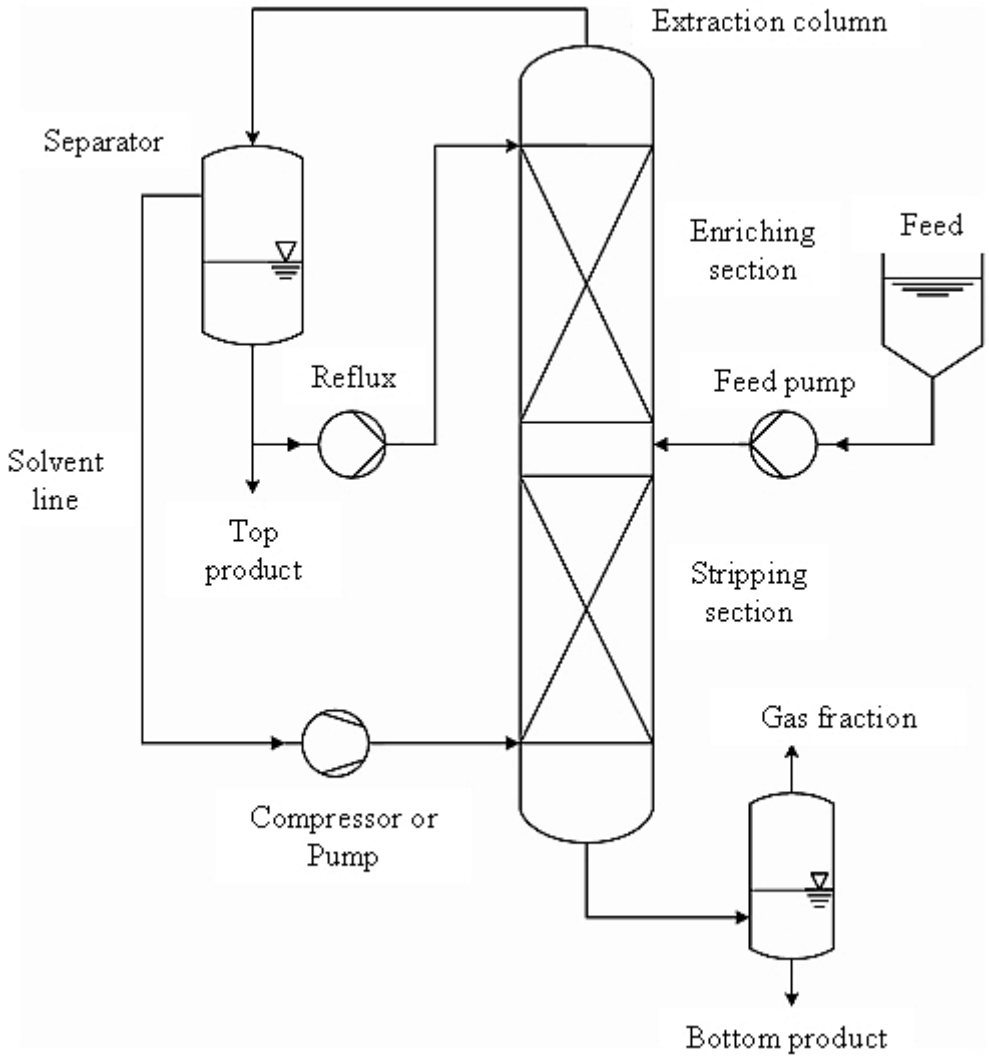

Figure 1: Counter-current extraction.

Ultrasound Extraction (Sonication)

The procedure involves the use of ultrasound with frequencies ranging from 20 kHz to 2000 kHz; this increases the permeability of cell walls and produces cavitation. Although the process is useful in some cases, like extraction of rauwolfia root, its large-scale application is limited due to the higher costs. One disadvantage of the procedure is the occasional but known deleterious effect of ultrasound energy (more than 20 kHz) on the active constituents of medicinal plants through formation of free radicals.[13]

Supercritical Fluid Extraction

Supercritical fluid extraction (SFE) is an alternative sample preparation method with general goals of reduced use of organic solvents and increased sample throughput. The factors to consider include temperature, pressure, sample volume, analyte collection, modifier

(cosolvent) addition, flow and pressure control, and restrictors. Generally, cylindrical extraction vessels are used for SFE and their performance is good beyond any doubt. The collection of the extracted analyte following SFE is another important step. There are many advantages to the use of CO2 as the extracting fluid. In addition to its favorable physical properties, carbon dioxide is inexpensive, safe and abundant. But while carbon dioxide is the preferred fluid for SFE, it possesses several polarity limitations. Solvent polarity is important when extracting polar solutes and when strong analyte-matrix interactions are present. Organic solvents are frequently added to the carbon dioxide extracting fluid to alleviate the polarity limitations. Of late, instead of carbon dioxide, argon is being used because it is inexpensive and more inert. The component recovery rates generally increase with increasing pressure or temperature: the highest recovery rates in case of argon are obtained at 500 atm and 150° C.

The extraction procedure have distinct advantages:

i) The extraction of constituents at low temperature, which strictly avoids damage from heat and some organic solvents.

ii) No solvent residues.

iii) Environmentally friendly extraction procedure.

The largest area of growth in the development of SFE has been the rapid expansion of its applications. SFE finds extensive application in the extraction of pesticides, environmental samples, foods and fragrances, essential oils, polymers and natural products.[14]

CONCLUSION

Medicinal plnts contain numerous phytoconstituents. But non standardized extraction procedures may degrade the phytoconstituents in plants and thus lead to variations and lack of reproducibility. Thus the best extraction technique has to be aopted for better results and pharmacological activity.

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