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Basic Principle of Plants Bioactive Compounds Extraction Using Various Extraction Methods

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ABSTRACT

Bioactive compounds are also defined as secondary metabolites that are present in plants. Bioactive compounds are present in a wide variety of plant materials and can be classified into several types, such as terpenoids, alkaloids, nitrogen-containing compounds, organosulfur compounds, and phenolics. Bioactive chemicals of natural origin are often found in small amounts. This occurrence has resulted in the development of more complex extraction methods. Therefore, it is crucial to carefully choose a suitable extraction technique to effectively obtain the desired bioactive components. The objective of this article is to present a comprehensive review of the several extraction techniques used to obtain bioactive compounds from plants. The extraction methods include several kinds of procedures, including both conventional and modern approaches.

Keywords: Bioactive compounds, Conventional methods, Extraction, Modern methods

Introduction

Plant bioactive chemicals are key sources for culinary, nutraceutical, cosmetic, and pharmaceutical product development (Yusoff et al., 2022). Bioactive substances are generally secondary metabolites found in plants, several primary metabolites have recently been identified as bioactive compounds (Banozic et al., 2020). Carbohydrates, amino acids, and proteins are key metabolites that are important in the development and maturation of plant tissues. Secondary metabolites are created during the developmental cycle to help plants survive and overcome natural hurdles (Azmir et al., 2013). They provide a protective role in plants against both biotic and abiotic stress. Bioactive chemicals provide numerous benefits to our bodies that help to preserve our health. They have served as a method of disease prevention.

A diverse range of plant materials contains bioactive molecules, which can be categorized into several types such as terpenoids, alkaloids, nitrogen-containing compounds, organosulfur compounds, and phenolics (Altemimi et al., 2017). The majority of bioactive compounds found in nature are produced in limited quantities and are typically obtained as mixtures in extracts. Considering the limited quantities of bioactive compounds, it is essential to enhance the production process to maximize yield and explore cost-effective alternative sources (Azmir et al., 2013; Banožić et al., 2020; Cvjetko Bubalo et al., 2018). The very first step in the process involves extraction, which allows for the separation of the required natural compounds from the raw materials. The process of extraction is affected by the solubility of the active components in combination with other solutes, various compounds found in the plant matrix, and the choice of solvent used to dissolve the active ingredients (Berk, 2018). Bioactive compounds are produced in various amounts and usually available in small

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quantities in many parts of plants, including leaves, roots, barks, tubers, woods, gums or oleoresin, exudates, fruits, figs, flowers, rhizomes, berries, twigs, and the entire plant. Therefore, it is important to carefully choose the appropriate extraction method in order to optimize the extraction yield from the plant matrix (Joana Gil-Chávez et al., 2013; Tiwari, 2015). The extraction efficiency is influenced by various parameters, including extraction technique, plant component matrix properties, extraction solvent, temperature, pressure, and extraction time (Drosou et al., 2015). This article discusses several extraction techniques that can be applied to obtain bioactive compounds in plant matrix components.

Material and Methods

The methodology employed in the preparation of this article is a narrative review of relevant literature. The literature included in this study comprises articles sourced from both national and international publications. The literature utilized in this study was sourced from various database portals, such as ResearchGate, ScienceDirect, PubMed, and Google Scholar. The keywords applied for finding research papers contain a combination of the phrases bioactive compound, extraction, conventional extraction, modern extraction, and emerging technologies. The screening process involved the evaluation of both full-text articles and abstracts to identify outcomes of the research that matched the specified criteria. The search results that are considered suitable are further examined and compared with other results.

Results and Discussion

Conventional extraction methods

Maceration

Maceration is one of the oldest extraction methods for medicinal preparation. It is regarded as a popular and affordable method of obtaining natural goods from plant material. In this procedure, the solvent will be added after the powdered solid components are placed in a closed vessel Figure 1. It is permitted to stand for a long period (ranging from hours to days) with shaking periodically. A sufficient amount of time is allowed for the solvent to pass through the cell wall and solubilize the ingredients contained in the plant. Only molecular diffusion is used in the process. After a sufficient period has passed, the liquid is separated and the solid residue is pressed to collect as much solvent as possible (Rasul, 2018).

Figure 1. Cold maceration apparatus

Maceration consists of three main phases. First, plant materials are ground into a fine powder. This enables good material and solvent interaction. A specified solvent is introduced in a sealed vessel after grinding. The remaining solid from this extraction procedure is pressed to recover a significant amount of occluded solutions after the liquid has been strained off. Periodic shaking improves

extraction throughout the maceration phase by enhancing diffusion and removing concentrated solution from the sample surface, which introduces additional solvent to the menstruum and increases extraction yield (Azmir et al., 2013).

Maceration extraction, as a method for obtaining plant extracts, presents several advantages. Firstly, it results in a higher oil yield compared to other extraction techniques. Additionally, the components of the volatile oil extracted through maceration are less prone to hydrolysis and polymerization. The control of wetness at the bottom of the extraction still becomes crucial for managing hydrolysis, while the thermal conductivity of the still walls impacts polymerization. Moreover, if refluxing is appropriately regulated, the loss of polar compounds is minimized, ensuring a more comprehensive extraction of valuable components. Furthermore, the quality of oil produced through maceration is more reproducible in comparison to steam and water distillation methods. Notably, maceration extraction is distinguished by its cost-effectiveness and environmental friendliness, as it does not necessitate the use of organic solvents.

However, the maceration extraction method is not without its drawbacks. Firstly, complete extraction of the plant material is not always achievable. Additionally, the direct contact of the plant material near the bottom of the still with the furnace fire may lead to charing, imparting an undesirable odor to the essential oil. Prolonged exposure to hot water during maceration can also cause hydrolysis of certain constituents, such as esters, potentially altering the chemical composition of the extracted oil. The challenging aspect of heat control in maceration may result in variable distillation rates, affecting the consistency of the extracted compounds. Furthermore, the process demands a greater number of stills, more space, and increased fuel consumption, making it potentially uneconomical in comparison to other extraction methods.

Percolation

Percolation is widely recognized as the most common technique employed in the preparation of fluid extracts, including tinctures. Percolation refers to the process of gradually passing a liquid through a solid medium in a droplet-by-droplet process. During the process of percolation, the solvent, often ethyl alcohol, is passed through the plant material at a slow rate. This results in the progressive accumulation of phytochemicals inside the solvent, which is then driven downwards by the addition of fresh solvent from the top (Azwanida, 2015).

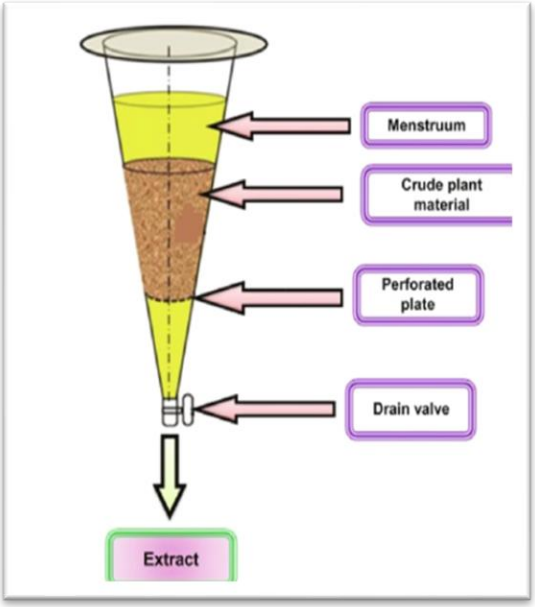

Figure 2. Percolator (Alara et al., 2021)

Percolation is a method of extraction frequently used for pharmaceutical substances that are expensive and particularly for active components that are sensitive to heat. Percolation is a technique that works on the continuous movement of the solvent through a static bed containing the raw botanical substance, to extract the soluble bioactive constituents contained there. A percolator (Figure 2) is the term used to refer to this apparatus; it is a slender, conical-shaped vessel with holes at both ends (Manousi et al., 2019).

Before inserting plant material into the percolator, it is important to shred it thoroughly, ensuring that the particles are not excessively small. The presence of excessively small particles can introduce difficulties with the process of separating the fine particles from the extraction solvent. As a result, the extracted substance would exhibit contamination, along with a formation of residual matter at the base of the percolator (Azwanida, 2015).

The selection of the extraction solvent is based on the chemical characteristics of the secondary metabolites. A common solvent mixture in extraction processes is composed of water and alcohol. This combination enhances the efficiency of extraction due to the hydrating properties of water, which facilitates the interaction with plant cell walls. Additionally, the chemical similarity between alcohol and the active components extracted from the plant material further contributes to the effectiveness of this solvent mixture. For instance, the extraction of phenolics, specifically epicatechin, was conducted using a 70% ethanol solution, whereas petroleum ether was employed for the extraction of antioxidants, such as phenols and flavonoids (Bitwell et al., 2023).

The percolation method is similar to maceration as it involves the insertion of a finely ground substance into a closed system, followed by the slow addition of solvent from the top layer to the bottom portion. Filtration is unnecessary in this context as the percolator devices are fitted with filters designed to selectively allow the flow of solvents carrying the extract. The challenges associated with the percolation method have similarities to those encountered in the maceration method, including the requirement of large solvent volumes, and time-consuming (Alara et al., 2021).

The advantage of the percolation method is that it can provide a higher extract yield, which means this method can extract secondary metabolites more effectively (Verawati et al., 2017). Zhang et al. (2018), conducted a comparative analysis of the percolation and refluxing extraction techniques for the extraction of *Undaria pinnatifda*. They discovered that the concentration of the primary constituent, fucoxanthin, obtained through the percolation extraction technique was higher compared to that obtained through the refluxing method. However, there was no statistically significant difference observed in the overall amount of extract obtained between the two methods. Research by Verawati et al. (2017) also showed similar results where the percolation method produced a higher yield of bay leaf (*Syzygium polyanthum*) extract compared to the maceration and soxhletation methods. Apart from that, the phenolic content produced from the percolation method was also higher (103,911 mg/g) compared to the maceration method (69,764 mg/g) and soxhletation (72,800 mg/g). The reason for this result is perhaps attributed to the thermolability of the phenolic compounds present in bay leaves. Consequently, the application of heat during the soxhletation process leads to the breakdown and subsequent destruction of these phenolic compounds.

Soxhlet extraction

The Soxhlet extraction method is a liquid-liquid extraction technique that uses solvents such as ethanol, alcohol, n-hexane, and others. This method of extraction is commonly employed for the isolation of substances with limited solubility, where the impurities contained are insoluble in the chosen solvent. Figure 3 illustrates the components of a Soxhlet extractor. The Soxhlet extraction technique involves a cyclic filtration procedure aimed at achieving optimal results while minimizing solvent (Anam et al., 2014). According to Kadji et al. (2013), the Soxhlet extraction method yielded a higher quantity in comparison to maceration. This is because heat treatment can increase the ability of the solvent to extract compounds that are insoluble at room temperature conditions, as well as the

maximum withdrawal of compounds by the solvent which is always circulating in the process of contact with the simplicia, thereby providing an increase in yield.

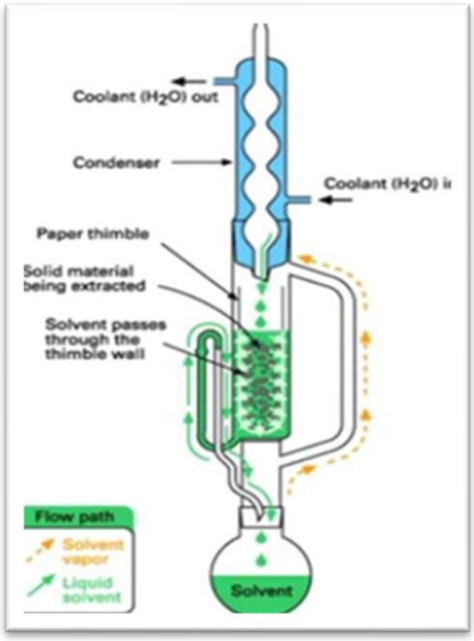

Figure 3. Soxhlet extractor

The Soxhlet process has several advantages in oil extraction, such as increased oil production, minimal solvent usage, quicker extraction time (Pratama et al., 2017), and higher yield compared to maceration (Kadji et al., 2013). The Soxhlet extraction method has a weakness in that the continuous application of heat during the extraction process can potentially cause damage to the solute or other components with poor heat resistance (Tiwari et al., 2011). Additionally, the suitability of the Soxhlet extraction method is limited by the requirement for dry, ground solids as the ideal sample, and numerous factors must be taken into consideration when employing this technique (Amid et al., 2010). Furthermore, the solvent used in the extraction system must possess a high level of purity. According to (Azwanida, 2015), the Soxhlet extraction approach is well recognized as having negative environmental implications and can potentially contribute to pollution issues when compared to the supercritical fluid extraction method.

Novel extraction methods

Supercritical fluid extraction (SFE)

Supercritical fluid extraction (SFE) is a technologically advanced extraction method that employs supercritical fluid as a solvent, providing numerous benefits compared to conventional extraction techniques. The extraction approach being discussed has promise due to its ability to produce extracts with better purity (Moncada et al., 2016). Supercritical fluids have properties similar to gases, facilitating their diffusion into the matrix and enabling access to target chemical compounds. Simultaneously, their liquid-like attributes offer significant dissolving ability. Furthermore, the diffusivity, density, surface tension, and viscosity of supercritical fluids can be altered through the manipulation of operating parameters, such as temperature and pressure. Consequently, the characteristics of this supercritical fluid offer benefits in regulating the extraction procedure (Salleh et al., 2014). Another advantage of SFE is its ability to use a smaller quantity of sample compared to other extraction techniques. In contrast to the typical sample quantities of 20-100 g used in conventional

extraction methods, the application of Supercritical Fluid Extraction (SFE) requires a significantly lower sample size of 0.5-1.5 g (Ayre et al., 2013).

The supercritical fluids used as solvents in supercritical fluid extraction (SFE) include carbon dioxide, ammonia, hydrocarbons like propane and butane (Escobar et al., 2020), and nitrogen (Khaw et al., 2017). Carbon dioxide (CO2) is often used as a supercritical solvent for the extraction of natural chemicals due to its advantageous properties. It has attributes such as being colorless, odorless, non-toxic, non-flammable, safe, highly pure, and affordable, making it an ideal option for this method. Furthermore, it is important to note that carbon dioxide (CO2) has a comparatively lower critical point in comparison to other supercritical solvents, with a critical pressure (Pc) of 7.38 MPa and a critical temperature (Tc) of 31.1◦C. This characteristic makes CO2 an advantageous choice as it allows for the prevention of oxidation and thermal degradation by operating at lower pressures and temperatures (Babova et al., 2016). The main drawback of carbon dioxide (CO2) is its relatively low polarity, which results in reduced efficiency in extracting polar molecules. To solve this issue, the addition of polar solvents as co-solvents has been proposed (Escobae et al., 2016). The polarity of carbon dioxide (CO2) can be enhanced by using co-solvents, such as water or ethanol (5% w/w) (Campardelli et al., 2015).

The extraction process of SFE technology using carbon dioxide (CO2) includes three primary stages. (I) To sustain a liquid state, carbon dioxide (CO2) is initially pressurized at around 50 bar and cooled to a temperature below 5 C. System pressure is efficiently managed through the use of either a basic regulator or a back pressure regulator. Next, carbon dioxide (CO2) is pumped into the heating zone, resulting in the change of CO2 into a supercritical state. The extraction vessel is subsequently filled with carbon dioxide at supercritical conditions, causing fast diffusion of the gas into the solid matrix and dissolution of the target material. (III) The dissolved material is transferred from the extraction vessel to the separator at lower pressure conditions, facilitating the decomposition of the material. In addition, supercritical fluids can be cooled and recycled (Geeta et al., 2020).

Extraction using supercritical carbon dioxide has high selectivity. According to research by Syukriah and Azizi (2014), extraction of *Quercus infectoria* galls using Soxhlet method produces a higher extract yield than SC-CO2 extraction. However, the extract with SC-CO2 for 2 hours had a higher total phenolic compound content (143.75 mg GAE/g) compared to Soxhlet extraction using 70% methanol (112.28 mg GAE/g) which required an extraction time of 6 hours. This shows that even though it produces a lower yield, extraction using SC-CO2 has higher selectivity compared to Soxhlet extraction so that the targeted compound can be obtained with high purity.

Pressurized liquid extraction (PLE)

Pressurized Liquid Extraction (PLE) is a technique employed for liquid extraction, where higher temperatures and medium to high pressure are utilized to enhance and improve the extraction procedure. The solvent employed in Pressurized Liquid Extraction (PLE) has a temperature and pressure that are below the critical temperature and pressure values. The objective of this attempt is to sustain the solvency of the substance in a fluid form. The solvent utilized in pressurized liquid extraction (PLE) is pumped into the system with the assistance of a pump. In addition, a pump is used to facilitate the discharge of the extract following the completion of the extraction process. The extraction process takes place within an extraction cell that is placed in an oven. The functioning of the oven involves the regulation of temperature and pressure necessary for the extraction process. The outcomes of the extraction process are gathered within a container designated as the collection bottle, which is positioned at the end point of the extraction system (Patel et al., 2019).

The procedure for extracting the analyte from semisolid and solid samples in pressurized liquid extraction (PLE) is as follows:

1. The sample (consisting of the analyte to be extracted and the matrix) should be moistened using an extraction solvent.

2. The process of desorption involves the release of chemicals from the matrix, which may or may not involve the breakdown of chemical bonds.
3. The component should be dissolved in the extraction solvent.
4. The dispersion of the chemical from the matrix
5. Diffusion occurs as solute particles penetrate other solvent layers surrounding the matrix, and eventually reach the bulk solvent.

The PLE extraction process is influenced by various parameters, such as temperature, pressure, extraction duration, and solvent-sample ratio. As the temperature rises, there is a decrease in the surface tension and viscosity of the solvent, followed by an increase in the diffusivity of the solvent. The observed modifications in solvent characteristics as temperature increases facilitate enhanced mass transfer rates and improved wetting of the sample. Furthermore, the process of transferring the analyte from the matrix to the solvent, known as desorption, is facilitated by higher temperatures due to the decreased intermolecular interactions that hold the analyte and matrix together. If the extraction is conducted at the appropriate moment, it will result in an extensive and quicker process (Alvarez-Rivera et al., 2020).

The pressure in the PLE process has an impact on the characteristics of the solvent as long as the solvent is maintained in a liquid state. High pressure will wet the sample matrix, thereby increasing extraction efficiency. The pressure usually used is 5-15 Mpa unless solvent saturation pressure is used. The solvent-sample ratio in the PLE process must be as small as possible to avoid dilution of the extract, but at the same time large enough to provide optimal extraction results. The solvents usually used in the PLE extraction method are ethanol, ethyl acetate, ethyl lactate, or d-limonene, because they have been recognized as safe (GRAS) and are considered more environmentally friendly solvents. The choice of solvent in the extraction process is considered based on the nature of the compound to be extracted (Alvarez-Rivera et al., 2020).

Research conducted by Rahmawati (2018) shows that the applications of Pressurized Liquid Extraction have been carried out include the extraction of phenolic components from microalgae and cyanobacteria (Escobar et al., 2017), extraction of saponins and fatty acids from Ziziphusjujuba, determination of 8 rhizome and root components of Curcuma longa. This is supported by the statement from (Alvarez-Rivera et al., 2020) that Pressurized Liquid Extraction can be applied for the extraction of several bioactive compounds such as polyphenols, terpenoids, lipids, and essential oils. In addition, this extraction method is commonly used in essential oil processing, carotenoid extraction, and analysis of pesticides, metals, drug residues, and poisons.

Pressurized Liquid Extraction (PLE) offers several advantages as an alternative to conventional techniques, such as boiling, Soxhlet extraction, and solid-liquid extraction. The application of the PLE (Pressurized Liquid Extraction) method has been seen to have a positive impact on extract yield and extraction time using rising temperature and pressure. The resulting extract contains a higher concentration of bioactive constituents. The process of PLE extraction enables the disintegration of plant cells, facilitating the extraction of bioactive constituents present in the cells, exceeding the efficacy of conventional extraction techniques. One drawback associated with the PLE approach relates to the utilization of high temperatures during the extraction procedure, which consequently leads to the release of numerous unwanted compounds (Wijngaard et al., 2012).

Microwave-assisted extraction (MAE)

Microwave-assisted extraction (MAE) is a recently developed technique that utilizes microwave energy to induce the heating of polar solvents in direct contact with solid samples. This method facilitates the partitioning of target chemicals between the sample and the solvent, resulting in reduced extraction time and decreased solvent usage. Additionally, it produces higher extraction rates and achieves better results at lower costs (Dahmoune et al., 2015). The illustration of microwave-assisted extraction (MAE) is displayed in Figure 4.

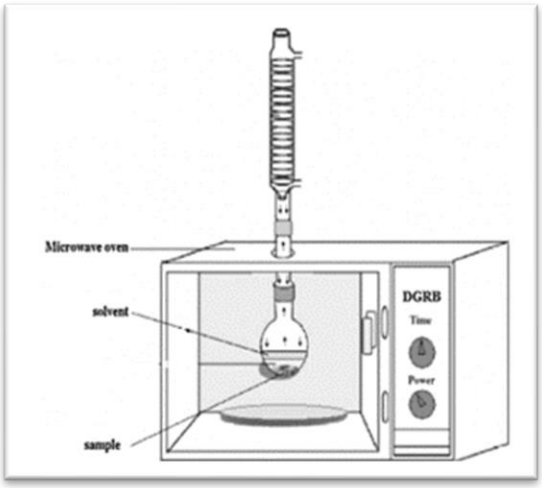

Figure 4. Microwave-assisted extraction unit (Thirugnanasambandham et al., 2015)

When microwave-assisted extraction is performed simultaneously. One notable factor is the rapid elevation in temperature, which leads to a decrease in emulsion viscosity and the breakdown of the outer coating of plant material. Consequently, this phenomenon enhances the rate of extraction. Another mechanism is molecule rotation, which works to neutralize the Zeta potential (5). This occurrence causes a reorganization of the electrical charges surrounding the molecules, leading to an increased mobility of ions and thus enhancing the effectiveness of the extraction process. Furthermore, certain process variables in microwave-assisted extraction (MAE), including the weight of the sample, microwave power, and extraction time, were found to have a substantial impact on the efficiency of the process. By optimizing these parameters, it is possible to greatly enhance the extraction yield (Thirugnanasambandham et al., 2015).

This extraction approach involves the utilization of microwave energy to induce heat through interactions with polar components, resulting in the conversion of electromagnetic energy into thermal energy. The dielectric constant and dissipation factor are crucial parameters in microwave procedures. These parameters play a significant role in determining the amount of power energy reflection at the air-sample interface and the degree of interaction with the sample. Hence, the selection of a solvent possessing a high extraction capacity and robust affinity towards both the matrix and the desired bioactive chemicals holds crucial significance. Solvents such as ethanol, methanol, and water possess the ability to absorb microwave energy as a result of their elevated dielectric constant and dielectric loss. In addition, the dielectric constant can be altered by combining various solvent mixes (Pimentel-Moral et al., 2018).

Other important factors in the MAE process are the temperature and duration of extraction. Long contact with microwave radiation has been observed to potentially reduce the extraction yield due to the breakdown of the chemically active structures of polyphenols. The adjustment of time extraction has the potential to provide control over the exposure and enhance the yield of the extract. However, in the case that a longer duration for extraction is necessary, it is possible to extract the samples in a series of steps through repeated extraction cycles (Ameer et al., 2017). On the other hand, higher temperatures lead to an increase in solvent power as a result of reduced viscosity and surface tension, hence facilitating greater solvent penetration. Nevertheless, higher temperatures have been found to hurt the extraction yield due to the degradation of the molecular structure of bioactive chemicals. It is important to achieve a state of balance between the length of the extraction process and the temperature (Pimentel-Moral et al., 2018).

Pulsed electric field extraction (PEF)

Pulsed Electric Field (PEF), is a technique commonly referred to as electroporation. It involves the application of electrical voltage to cells, resulting in controlled cell wall disruption while preserving the integrity of the bioactive constituents present within the cell, including antioxidant chemicals and secondary metabolites (Rahmah et al., 2019). The application of Pulsed Electric Fields (PEF) treatment has demonstrated remarkable potential as a less severe and more effective alternative to traditional methods of cell disintegration. The application of a moderate intensity electric field (0.5-10 kV/cm) and low energy (1-10 kJ/kg) to plant tissue, in the form of short repetitive voltage pulses (typically ranging from a few microseconds to 1 millisecond), results in the permeability of cell membranes. This permeability enables the release of juice and valuable compounds from the intracellular areas. The application of pulsed electric field (PEF) treatment to food products has been found to have a non-thermal effect. This treatment has the potential to selectively alter the permeability of cellular membranes, specifically the tonoplast and plasma membrane while leaving the cell wall intact. This selective permeability improvement has been observed to improve the purity and yield of extracted substances from the food products (Bobinaitė et al., 2015).

The utilization of pulsed electric field extraction has been found to offer considerable improvements in both extraction yield and extraction time reduction. This is attributed to its ability to enhance mass transfer during the extraction process by disrupting membrane structures. The efficacy of PEF treatment is dependent upon various parameters, such as the strength of the electric field, the amount of energy input per unit mass, the number of pulses administered, and the temperature when the treatment is performed (Zhang et al., 2018).

Research by Bozinou et al. (2019), shows the extraction of *Moringa oleifera* leaf extraction using the PEF method with electric field strength (E) was set at 7 kV/cm, pulse duration (PD) 10 msec, pulse interval (PI) of 25 msec, extraction time for 40 minutes, and using room temperature has a higher total polyphenol compound than other extraction methods (ultrasound-assisted and microwave-assisted). Extraction using the PEF method had a total polyphenolic compound content of 38.24 mg GAE/g, while the ultrasound method only had 29.04 mg GAE/g, and the microwave method only had 36.59 mg GAE/g. According to the results, it can be concluded that the use of PEF technology shows considerable potential as an effective choice in comparison to other extraction techniques, such as microwave-assisted and ultrasound-assisted extractions, for the recovery of significant chemicals from plant samples. In addition, it is noteworthy that PEF technology has lower power requirements, resulting in reduced costs. Additionally, a significant advantage of PEF is its ability to prevent an increase in sample temperature during the extraction process. This characteristic is particularly beneficial for the extraction of sensitive compounds, enabling the manufacture of high-value extracts.

Ultrasound-assisted extraction (UAE)

Ultrasonic-assisted extraction (UAE), also referred to as ultrasonic extraction or sonication, involves the utilization of ultrasonic wave energy to facilitate the extraction process. The application of ultrasound in solvent-induced cavitation leads to enhanced solute dissolution, diffusion, and heat transfer. Consequently, the extraction efficiency is improved. Another benefit of the UAE is its ability to minimize solvent and energy usage, as well as decrease extraction temperature and time. This method is suitable for the extraction of thermolabile and unstable chemicals. Hence, it's frequently used in the process of extracting several categories of natural goods (Zhang et al., 2018). This method is a simple extraction technique that utilizes induced mechanical effects via the explosion of micro-sized bubbles. The illustration of the UAE technique is displayed in Figure 5. This process efficiently disrupts tissue, promoting the diffusion of bioactive compounds from the material into the solvent. In experimental procedures, it is typically necessary to employ ultrasonic waves within a frequency range of 20 to 2000 kHz to enhance the permeability of cell walls and induce the formation of cavitation (Alara et al., 2021).

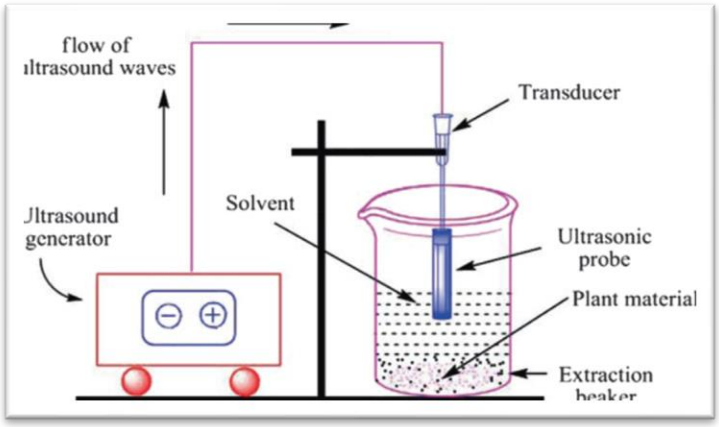

Figure 5. Pictorial representation of the UAE technique (Alara et al., 2021)

A study by Deng et al. (2017), showed that the UAE extract from fresh olives showed a higher extraction yield (7,01 mg/g) compared to the maceration method (5,18 mg/g). The UAE method was conducted using 22 mL/g of liquid-solid ratio, 47°C of extraction temperature, and 30 min of extraction time. Meanwhile, the maceration method was conducted using 24 mL/g of liquid-solid ratio, 50°C of extraction temperature, and 4.7 h of extraction time. This showed that the UAE method requires a smaller quantity of solvent, shorter extraction time, and lower extraction temperature.

Enzyme assisted extraction (EAE)

The Enzymatic Assisted Extraction (EAE) method depends on the utilization of enzymes that facilitate the hydrolysis of covalent bonds in the presence of water. This process causes the breakdown of cellular structures and enhances the material's permeability. Enzyme-assisted extraction can be employed as an independent technique or as a pre-processing step for traditional extraction methods. Several factors need to be taken into consideration, including particle size, time, pH, and temperature. Enzymatic reactions exhibit optimal efficiency when conducted in conditions characterized by lower temperatures, moderate pH levels, and shorter durations (often within a range of a few hours). Furthermore, these reactions do not necessitate the use of expensive machinery (Łubek-Nguyen et al., 2022). In this technique, the enzyme commonly used is carbohydrase such as cellulase (Reddy & Majumder, 2014).

The present study investigated the EAE of polysaccharides from the radix of *Astragalus membranaceus*. Multiple enzymes were employed, and the results revealed that glucose oxidase exhibited greater effectiveness in the extraction of polysaccharide compared to the other seven enzymes that were tested (amyloglucosidase, hemicellulase, bacterial amylase, fungal amylase, pectinase, cellulose, and vinozyme). The polysaccharide production had a significant rise of more than 250% when the EAE condition was employed with glucose oxidase, in comparison to the non-enzyme treated (Chen et al., 2014).

Conclusion

This review has examined several extraction techniques used to obtain the bioactive compounds from plant materials. The extraction techniques frequently used for the extraction of bioactive compounds from plant materials include maceration, percolation, and Soxhlet extraction. Despite the wide popularity of these procedures, they have several limitations. These include the recovery of only limited yields, the consumption of larger quantities of extraction solvents, longer extraction times, and an enormous accumulation of residues. These factors have prompted the development of alternative methodologies, such as supercritical fluid extraction (SFE), pressurized liquid extraction (PLE),

microwave-assisted extraction (MAE), ultrasound-assisted extraction (UAE), pulsed electric field extraction (PEF), and enzyme-assisted extraction (EAE) in order to address the limitations associated with traditional approaches.

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