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Microwave Assisted Extraction of Bioactive Compounds from Food: A Review

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Abstract The microwave-assisted extraction is also considered as a novel method for extracting soluble products into a fluid from a wide range of materials using microwave energy. Natural bioactive compounds are produced as secondary metabolites; include a broad diversity of structures and functionalities. Some of those compounds can be found in nature at high concentration such as polyphenols but others can only be found at very low levels, so that massive harvesting is needed to obtain sufficient amounts, and their structural diversity and complexity make chemical synthesis unprofitable. The inherent difficulties in screening and producing these compounds have led to the development of advanced technologies. The commonly used methods for their extraction are the conventional liquid–liquid or solid–liquid extraction and the advanced include pressurized-liquid extraction, subcritical and supercritical extractions, and microwave- and ultrasound-assisted extractions. These new technologies could provide in the next few years an innovative approach to increase the production of specific compounds for use as nutraceuticals or as ingredients in the design of functional foods. Advantages of microwave heating compared to conventional processing methods include energy-saving rapid heating rates and short processing times, deep penetration of the microwave energy (which allows heat to be generated efficiently without directly contacting the work-piece), instantaneous and precise electronic control, clean heating processes, and no generation of secondary waste. Microwave energy processes for heating, drying, and curing have been developed for numerous laboratory-scale investigations and, in some cases, have been commercialized. MAE is also recognized as a green technology because it reduces the use of organic solvent. This review is aimed to discuss this extraction technique along with their basic mechanism for extracting bioactive compounds from plant matrix.

Keywords Microwave synthesis, Natural bioactive compounds, Extraction

1. Introduction

The search for natural bioactive compounds (NBCs) with potential for the treatment and prevention of human diseases and to meet other needs is currently a key topic in many laboratories and industries. These compounds efficiently interact with proteins, DNA, and other biological molecules to produce a desired outcome, which could be exploited for designing natural products-derived therapeutic agents [1].

Nowadays, there is a marked trend in the food industry toward the development and manufacture of functional products. This new class of food products has seen great success in the market due to the growing of consumer interest for "healthy" food [2, 3]. NBCs are often synthesized in small quantities in nature and are present as conjugates or mixtures in extracts which require labor-intensive and time-consuming purification procedures [4]. Besides, the structural diversity and complexity of these molecules make their chemical synthesis unprofitable [4]. NBCs have been studied directly in their different natural matrices such as tea, olive oil, exotic fruits, plants, algae, microalgae, bacteria, and fungi. In this regard, an increasing number of scientific studies have been performed on these substances and their sources.

Recently, much attention has been directed to the recovery of bioactive compounds from different industrial food residues: leaves, peels, barriers, seeds, wood, culls, rinds, pits, pulp, press cakes, marc, malts, hops, hulls, husks, spent grain, carapace of crustaceans and shrimp, algae and other fish by products [5].

Traditional extraction techniques such as Soxhlet, solid–liquid extraction (SLE), or liquid liquid extraction (LLE) are characterized by high volumes of solvents and long extraction times. These techniques often produce low extraction yields of bioactive components and present low selectivity [6]. To overcome the limitations of these conventional extraction methods, non-conventional techniques, free of toxic solvents, have been introduced [7, 8].

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In addition, extraction processes for the separation of compounds have been developed to obtain highly purified products, rendering them useful in a wide range of applications. These technologies could provide an innovative approach to increase the production of the desired compounds.

Among them, microwave assisted extraction (MAE) has been accepted as a potential and powerful alternative for the extraction of bioactive compounds from food industrial residues. In addition, MAE technique possesses many advantages compared to other methods for the extraction of compounds such as bioactive compounds [9] saving in processing time and solvent, higher extraction rate, better products with lower cost, reduced energy consumption (up to 85-fold savings) and waste [10, 11]. In addition, MAE technique possesses many advantages compared to other methods for the extraction of compounds such as bioactive compounds [9] saving in processing time and solvent, higher extraction rate, better products with lower cost, reduced energy consumption (up to 85-fold savings) and waste generation [10, 11]. MAE is a method to extract soluble products into a fluid from a wide range of materials using microwave energy [12].

Interest in Microwave-Assisted Extraction (MAE) has increased significantly over the past 5-10 years in particular medicinal plant research, as a result of its inherent advantages as special heating mechanism, moderate capital cost and its good performance under atmospheric conditions [13, 14].

Manabe et al. [15] used microwave energy to extract pectin from mandarin orange peels. They found that by using microwave energy, they could extract about 5% more pectin in 15 min than could be extracted by conventional methods in 60 min.

More recently Wang et al. [16] optimized the operating conditions of pectin extraction assisted by microwave. They concluded that the application of microwaves in the extraction of pectin from dried apple pomace dramatically reduced the extraction time. The extracted pectin after purification could be added to different food products, both to enhance the nutritional value and improve food texture in jams, fillings, sweets, among others. Considering that tomato is one of the most consumed vegetable product, the amount of industrial waste is an huge quantity. Furthermore, the industrial tomato pomace is reach of for this reason El-Malah et al. [17] decided to compare ultrasound and microwave extraction in order to find the best extraction yield and antioxidant activity. They concluded that MAE was an optimum method to extract lycopene in a cheap and eco-friendly way. Peanut skins are another low-value by-product of peanut blanching and roasting operations [17]. They have been shown to contain significant levels of phenolic compounds with demonstrated antioxidant properties. Microwave hydro diffusion and gravity (MHG) is a recently developed technology with massive potential for variety of extractive applications, for instance, the extraction of essential oil has been executed from rosemary leaves [18, 19], Spearmint (*Mentha spicata* L.) and Pennyroyal (*Mentha pulegiom* L.) plant [20], and from citrus peel [18, 19]. The qualitative and quantitative studies of bioactive compounds from plant materials mostly rely on the selection of proper extraction method [21, 22].

2. Microwave Irradiation

Microwaves (MW) are a form of non-ionizing electromagnetic energy at frequencies ranging from 300 MHz to 300 GHz. This energy is transmitted as waves, which can penetrate in biomaterials and interact with polar molecules into materials, such as water to generate heat [23]. Microwave energy acts directly on molecules by ionic conduction and dipole rotation and thus only polar materials can be heated based on their dielectric constant.

Microwaves (MW) have the wavelengths between 1 mm and 1 m depending on the frequencies between 0.3 and 300 GHz. In electromagnetic radiation spectrum, the microwave radiation region is located between infrared radiation and radio waves [24]. In order to prevent any interference with cellular phone frequencies and telecommunications, the frequency of 2.45 GHz, which corresponds to a wavelength of 12.25 cm, is used for all domestic microwave ovens and chemical syntheses. The microwave heating is caused by the interaction of the electric field of radiation with the matter that can affect molecular actions, such as dipole rotations or ions migration. But, they cannot ionize the atoms crossed or change the molecular structure due to their low energy content [25]. Actually, it has a selective heating function. In other words, non-polar molecules are inert in the microwave electric fields while polar molecules with high dielectric constant and low molecular weight can selectively absorb microwave energy [26]. This selective heating of certain compounds may lead to the formation of microzones with a temperature much higher than the overall recorded temperature of the reaction bulk mixture. These high temperature microzones are called “hot spots” [27]. Thus a rapid enhancement of temperature is produced which may lead to an increase in the acceleration of chemical reaction rate [25]. For chemical reactions such as transestrification, that use microwaves as a means of heating, the polar molecules may bring more advantages [28].

3. Heating Mechanism

Microwave heating (MWH) is a sub-category of dielectric heating at certain frequencies. MWH is a non-contact energy transfer process from electromagnetic energy into thermal energy, which indicates fast heating rate, if the electromagnetic energy is efficiently absorbed by the treated materials, under low microwave (MW) radiation. Conventional heat transfer methods, such as conduction, convection and radiation, need to overcome the heat transfer barrier and usually take a longer time to reach the desired temperature for the target materials in comparison to MW heating. The electromagnetic wave can be launched from an emitter and guided to the target. The mechanism of dielectric heating relies on dipolar and interfacial polarization effects. The electric field component of MW causes polar molecules (e.g. water) to rotate and try to align in both permanent and induced dipoles at certain frequency called dipolar polarization. Friction and collision generated by the increased molecular rotation and moving result in heat loss and MW heating [29]. It is believed that kinetic energy associated with vibrational, rotational and translational movement of valance electrons is responsible for the generation of thermal energy [30]. It was also reported that energy is dissipated in the form of heat called Max-well-Wagner polarization (interfacial polarization effects) [29, 31].

In general, the higher the induced polarity, the greater the influence of MW, which indicates selective heating, uniform and volumetric heating characteristics of MWH. Based on the heating mechanism, MWH has the advantages of reducing the raw material pretreatment requirements, a substantial increase of product value, making it an efficient way of biomass energy usage [32].

Dielectric lost tangent parameter (DLTP) quantifies a dielectric material's inherent dissipation of electromagnetic energy into heat, expressed by the ratio of dielectric loss factor (imaginary permittivity) to dielectric constant (relative real permittivity), where dielectric constant specifies the amount of electromagnetic incident energy reflected and absorbed by the treated material and the dielectric loss factor corresponds to the amount of electric energy dissipated in the form of heat within the material. DLTP is used to characterize the MW energy absorption and is helpful in studying the MWH processes. Most solid wastes show relatively low MW absorption when subjected to MW irradiation. These materials cannot be heated to the desired temperature owing to their inability to absorb sufficient amount of MW energy [33]. These problems can be solved by addition of MWAs into the heating system. The chemical structure, shape and size of microwave absorbers (MWAs) determine the number and intensity of the tiny micro-plasma (small or electric arcs) spots generated during MWH [30].

4. Principles and Applications of Microwave-Assisted Extraction

Extraction is the first step of any medicinal or functional plant study, plays a significant and crucial role on the final result and outcome. Extraction methods are sometimes referred as ‘‘sample preparation techniques’’. The most common factors affecting extraction processes are matrix properties of the plant part, solvent, temperature, pressure and time [34]. Due to economics and environmental issues, food and chemical industries are facing the challenge of using new technologies in order to reduce energy consumption and CO_2 emissions [35].

The fundamentals of the microwave extraction (MAE) process are different from those of conventional methods (solid–liquid or simply extraction) because the extraction occurs as the result of changes in the cell structure caused by electromagnetic waves. In MAE, the process acceleration and high extraction yield may be the result of a synergistic combination of two transport phenomena: heat and mass gradients working in the same direction [36]. On the other hand, in conventional extractions the mass transfer occurs from inside to the outside, although the heat transfer occurs from the outside to the inside of the substrate (Fig. 1). In addition, although in conventional extraction the heat is transferred from the heating medium to the interior of the sample, in MAE the heat is dissipated volumetrically inside the irradiated medium. During the extraction process, the rate of recovery of the extract is not a linear function of time: the concentration of solute inside the solid varies, leading to a no stationary or unsteady condition. A series of phenomenological steps must occur during the period of interaction between the solid-containing particle and the solvent effectuating the separation, including (1) penetration of the solvent into the solid matrix; (2) solubilization and/or breakdown of components; (3) transport of the solute out of the solid matrix; (4) migration of the extracted solute from the external surface of the solid into the bulk solution; (5) movement of the extract with respect to the solid; and (6) separation and discharge of the extract and solid [37].

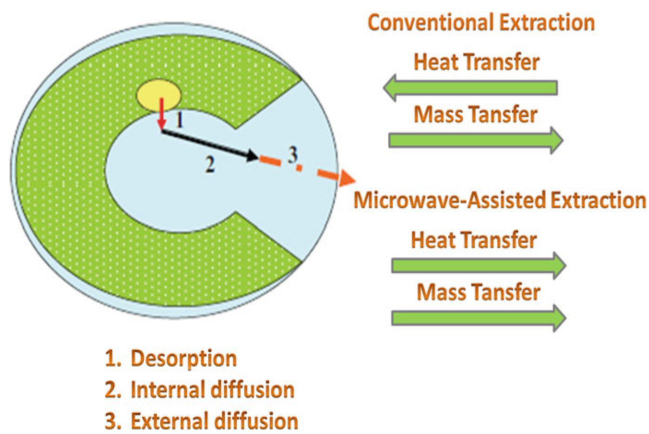

Figure 1. Conventional extraction [37]

In microwave-assisted extraction (MAE), microwave energy is used to heat solvents in contact with solid samples or liquid samples and to promote partition of the analytes from the sample matrix into the solvent (the extractant). The principle of heating using microwave is based upon its direct impacts on polar materials. Electromagnetic energy is converted to heat following ionic conduction and dipole rotation mechanisms [38]. During ionic conduction mechanism heat is generated because of the resistance of medium to flow ion. On the other hand, ions keep their direction along field signs which change frequently. This frequent change of directions results in collision between molecules and consequently generates heat.

The extraction mechanism of microwave assisted extraction is supposed to involve three sequential steps described by Alupului (2012): first, separation of solutes from active sites of sample matrix under increased temperature and pressure [39]; second, diffusion of solvent across sample matrix; third, release of solutes from sample matrix to solvent. Several advantages of MAE have been described by Cravottoa et al. (2008) such as quicker heating for the extraction of bioactive substances from plant materials [40]; reduced thermal gradients; reduced equipment size and increased extract yield. MAE can extract bioactive compounds more rapidly and a better recovery is possible than conventional extraction processes. It is a selective technique to extract organic and organometallic compounds that are more intact. MAE is also recognized as a green technology because it reduces the use of organic solvent [39].

Microwave energy is a non-ionizing radiation (frequency 0.3– 300 GHz) that causes molecular motion by migration of ions and rotation of dipoles. Microwaves are electromagnetic waves made up of two oscillating perpendicular fields: electrical and magnetic. This principle has been the basis for the development of microwave assisted extraction (MAE) which works heating the moisture inside the cells and evaporates, producing a high pressure on the cell wall. The pressure builds up inside the biomaterial which modifies the physical properties of the biological tissues (cell wall and organelles disrupter) improving the porosity of the biological matrix. This would allow better penetration of extracting solvent through the matrix and improved yield of the desired compounds [41]. Microwaves are used as information carriers or as energy vectors. This second application is the direct action of waves on material that is able to absorb a part of electromagnetic energy and to transform it into heat [42]. Thus, the principle of MAE is based on the direct effect of microwaves on molecules of the extraction system caused by two mechanisms, ionic conduction and dipole rotation [43]. The ionic conduction generates heat due to the resistance of the medium to ion flow. The migration of dissolved ions causes collisions between molecules because the direction of ions changes as many times as the field changes sign. The dipole rotation relates to the alternating movement of polar molecules, which try to line up with the electric field. Multiple collisions from this agitation of molecules generate energy release and therefore temperature increase [44]. Separation technologies, such as extraction, distillation and crystallization are promising areas of innovation which can promote the growth of sustainable processes in the chemical and food industries [45].

It should be noted that, unlike conventional forms of heating (convection and conduction), microwaves heat the system directly, leading to very short extraction times. Heat generation in the sample in the microwave field requires the presence of a dielectric compound. The greater the dielectric constant, the more thermal energy is released and the quicker the heating is for a given frequency. Advances in microwave-assisted extraction (MAE) have led in the development of various techniques such as compressed air microwave distillation (CAMD), vacuum microwave hydro distillation (VMHD), microwave hydro distillation (MWHD), solvent-free microwave extraction (SFME), microwave accelerated steam distillation (MASD), microwave by hydro diffusion and gravity (MHG) [45, 46]. Application of microwaves in separation and extraction processes has shown to reduce both extraction time and volume of solvent required, minimizing environmental impact by emitting less CO_2 in atmosphere [46, 47] and consuming only a fraction of the energy used in conventional extraction methods such as steam distillation, SD [46].

5. Critical Analysis

5.1. Critical Analysis: Scenario of MAE of Botanicals

Search results depicted in Fig. 2 clearly showed a trend that each year after 2010, minimum 50% of the publications that appeared on microwave assisted extraction where based on extraction of bioactive(s) from terrestrial plants. Pearson coefficient value of 0.982 clearly indicates a linear trend (2005–2015) between the total numbers of articles published in Scopus indexed journals on MAE and the number of articles actually related to extraction of bioactive(s) from botanicals. This is quite understandable from the fact that application of microwaves, pressurized liquids or ionic liquids are relatively new technologies which is being applied for the extraction of plant based bioactive(s). Huge technological advancements related to natural product research has taken place but innovation pertaining to extraction of botanicals has been on the slower side. This is evident from the fact that when it comes to drug discovery from natural products for the management of different diseases, publication rate of research papers has always been high. Publications recorded in Scopus database on MAE of botanicals was compared with the intensity of research on ethnopharmacology of medicinal plants where basically crude extracts are prepared and evaluated for their possible activity on different disease models (*in vivo* and *in vitro*) and the results are depicted in Table 1.

Table 1. Comparison of publication records between MAE of botanicals and ethnophamacology research of medicinal plants [49]

Area of research on medicinal plants	Publications recorded in Scopus (2005–2015)
MAE of botanicals	1157
Antidiabetic	4731
Anticancer	4616
Hepatoprotective	2854
Anti-inflammatory	9626
Antibacterial	12022

Table 2. Different wide range of MAE extraction conditions for phenolics from leaves and fruits using open and closed vessel extraction system

Plant Part	Extraction solvent	Microwave System	Plant Name	Extraction conditions
				Power
Leaves	Ethanol	Open vessel	*Pistacia lentiscus* [50]	600W
		Close vessel	*Cyclocarya paliurus* [51]	Information not available
			Information not available 5 min *Centella asiatica* [52]	300W
	Ethanol: water	Open vessel	*Theobroma cacao* L [53].	100–200W
			Myrtus communis [54]	700W
			Highbush blueberry [55]	20%(maximum) 800 W
			Herbal tea [56]	500W
	Methanol	Open Vessel	*Prunus laurocerasus* [57]	307.6W
			Rosmarinus officinalis [58]	800 W
			Prunus laurocerasus [59]	300W
	Water	Close vessel	*Pestacia lentiscus* [60]	400W
			Pestacia lentiscus [61]	400W
		Close vessel	*Dalbergia odorifera* [62]	300W
Fruits	Acetone: Water	Open vessel	*Citrus sinensis* [63]	500W
	Ethanol: Water	Open vessel	*Vitis vinifera* [64]	140W
			Cherry pomace [65]	700W
			Crataegus pinnatifida [66]	400W
			Dimocarpus longan Lour [67].	500W
		Close vessel	*Malus domestica* [68]	735W
	Methanol	Open vessel	*Prunus laurocerasus* [59]	300W
			Prunus laurocerasus [69]	550W
	Water	Open vessel	*Lycium ruthenicum* [70]	350 s
			Dryopteris fragrans [71]	300W
			Camellia oleifera [72]	50%
			Punica granatum [73]	600W
		Close vessel	*Solenum melongena* [74]	150W
	Methanol: Water	Open vessel	*Prunus cerasus* [75]	400W
			Citrus mandarin [76]	152W

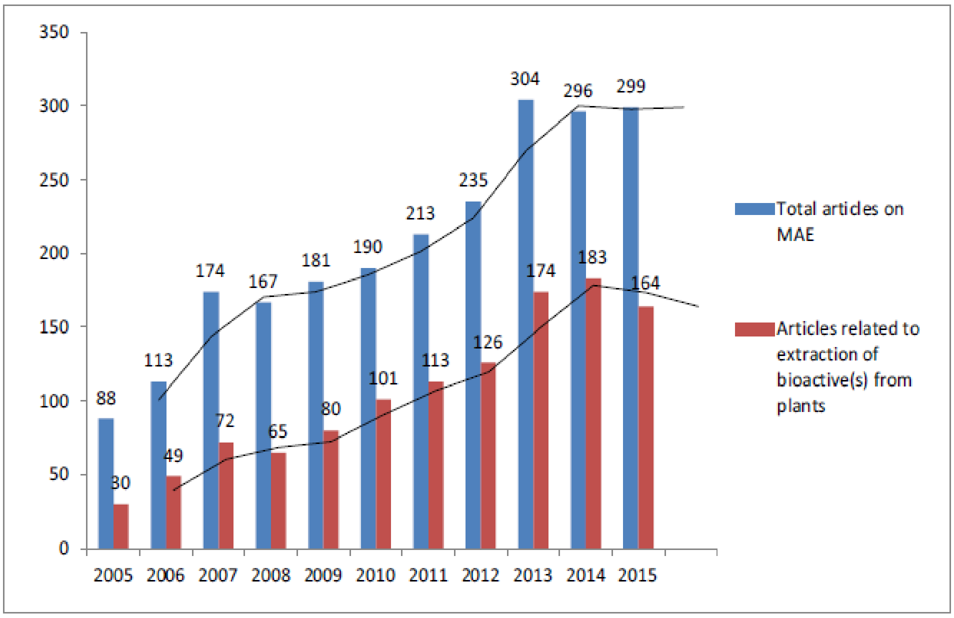

Figure 2. Search results depicting year wise the total number of publication that appeared on MAE and the number of publication which were related to MAE of bioactive (s) from plants. Database: Scopus, time range: 2005–2015

Today with so much advancement in chromatographic system where we can achieve high resolution of complex mixtures of almost every matrix and with detection limits up to few nanograms or even below, the whole analytical process for the detection of bioactive(s) can really fall apart if a suitable extraction method is not applied for the sample preparation prior to reaching the chromatographic system [48].

The disease area mentioned has been investigated on crude extract of medicinal plants. The word "extract" was included in the keywords to locate only those articles which are concerned with plants. As mentioned earlier a wide range of extraction conditions have been reported which makes it almost impossible to make a generalization on extraction conditions. Table 2 presented a glimpse of complexity involved in deciding the optimum microwave power and time for extraction of plant phenolics. However, this table contains a consolidated data of only 29 articles and the complexity shall increase further on studying more articles. Hardly any correlation could be framed.

6. Conclusions

As summarized, polar molecules under the microwave spectrum try to align themselves with the magnetic field lines, which are changed continuously. Thus the charged ion or molecular dipoles which interact with the changing magnetic field have to rotate rapidly. It results in the generation of energy in the form of heat due to molecular friction under MW irradiation. Transesterification process assisted by microwave technology is followed by a higher reaction temperature cites called hot spots. The review indicated that reaction in this situation is taken place in a temperature much higher than bulk temperature. Therefore, more efficient heating can be obtained compared to the usual conventional heating even more than sonochemical or supercritical reaction. This energy acts on the molecular scale [77]. From a technological point of view, recent batch microwave systems are smarter than earlier versions. These systems include libraries of predefined methods and can count the number of samples and recognize the vessel type loaded [78]. With this sequential approach, operators have the option to select an accurate temperature control for every sample or to handle different sample types with different extracting solvents, all within the same rack. In order to reach a batch-style approach, the main remaining hindrance is the sample-solvent separation after the extraction step. Most of the future fields of application are likely to focus on improving the flexibility of the latest introduced sequential systems. The capacity to control the extraction conditions for each sample should enable scientists to develop robust, reproducible methods for a diversity of matrices and chemicals. Overall, MAE appears to be an excellent alternative, as green extraction method, for the accurate determination of emerging organic pollutants, enabling its extension towards the regulatory environmental field.

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