

## Modern Approaches for Extracting Plant Bioactive Compounds to Enhance Food Security

Sumit Bhausahab Urhe\*, Abhinav Dubey, Guru P. N., Shrikrishna Nishani

*ICAR-Central Institute of Post-Harvest Engineering and Technology (CIPHET), Ludhiana-141004, Punjab, India*

---

---

### Abstract

Recent advancements in extraction techniques have revolutionized the extraction of plant bioactive compounds. Novel technologies such as Microwave Assisted Extraction (MAE), Supercritical Fluid Extraction (SCF), Pulsed Electric Field Extraction (PEF), Ultrasonication Extraction (UAE), Enzyme Assisted Extraction (EAE), High Pressure Process Extraction (HPP), and Cloud Point Extraction (CPE) are gaining traction. Celebrated for their green efficacy and cost-effectiveness, these methods are preferable over traditional techniques due to low solvent consumption, reduced extraction times, high purity of extracts, and non-thermal processing. Their non-toxic, environmentally friendly nature aligns well with sustainable practices. MAE uses microwave energy to heat solvents in contact with samples, enhancing extraction. SCF employs supercritical fluids, often carbon dioxide, for efficient dissolution and extraction of target compounds. PEF applies high voltage bursts to plant tissues, disrupting cell membranes and releasing bioactive substances. UAE uses ultrasonic waves to create cavitation in the solvent, promoting compound extraction. EAE leverages specific enzymes to break down cell walls, enhancing bioactive release. HPP uses high pressure to disrupt cell structures, extracting compounds without thermal degradation. CPE separates compounds based on their solubility in a surfactant solution at a specific temperature. These innovative techniques are particularly valuable in the food and pharmaceutical sectors, enabling efficient extraction of bioactive compounds to enhance food security and develop health-promoting products. This chapter aims to highlight the potential applications of these advanced methods in food processing, emphasizing their role in producing high-quality, bioactive-rich extracts.

---

---

### Introduction

Food and pharmaceuticals are important research areas with wide industrial applications. They need the application of novel, innovative extraction techniques to obtain bioactive compounds or phytochemicals. Techniques which are efficient, cheap, non-thermal and environmentally friendly are gaining popularity in the food industry. To furnish the growing

\*Corresponding author's e-mail: [sumit.urhe@icar.gov.in](mailto:sumit.urhe@icar.gov.in)

Enhancing Crop Resilience: Advances in Climate Smart Crop Production Technologies. Anjani Kumar, Rameswar Prasad Sah, Basana Gowda et al. (Eds). © 2024, BIOTICA.

needs the researchers found many extraction technologies and their potential relevance in food and pharmaceutical industry. Traditional extraction processes include grinding, maceration, soaking, preheating, Soxhlet extraction etc. They have limitations to use by its high solvent consumption, longer extraction time, impurity of extract, heat sensitivity and environment impact. The non-thermal novel extraction technologies can overcome the conventional in all these parameters for selection.

Extraction of bioactive compounds from plant sources will be efficient by using microwave assisted extraction, pulsed field extraction, enzyme assisted extraction, supercritical fluid extraction, ultra sonication and high-pressure extraction technologies. They increased mass transfer rates by cell wall disruption, molecular rotation, high diffusion coefficients, low heat generation and minimum time utilization are key factors in fastening the extraction process in novel techniques. Phytochemicals, functional ingredients, bioactive compounds, and animal source compounds are extracted industrially. The knowledge of using these methods are agglomerated in this review, an attempt made to emphasize the process parameters, selectivity of extraction process to different agricultural materials has been given. The advantages of these methods are manifold. They provide a non-thermal, non-toxic approach to extraction, significantly reducing environmental impact compared to traditional techniques. This green efficacy is crucial as the food industry increasingly prioritizes sustainability and safety. These techniques can efficiently extract a wide range of bioactive compounds from both plant and animal origins, offering high purity and potent extracts essential for enhancing food products.

The vast potential of these modern extraction methods is particularly notable in the food and pharmaceutical sectors, where they can help develop healthier, bioactive-rich products. By improving the efficiency and effectiveness of bioactive compound extraction, these advanced technologies play a pivotal role in bolstering food security, ensuring a more sustainable and health-conscious future. In this chapter we highlighted these techniques for extraction of plant bioactive compounds.

## **Conventional extraction techniques**

Bioactive compounds can be extracted from plant materials using conventional extraction techniques. Most of these methods rely on the extraction power of the various solvents used, as well as the use of heat and/or mixing. The three most common conventional extraction techniques for bioactive compounds are Soxhlet, maceration, and hydro-distillation.

### **1. Soxhlet extraction**

The Soxhlet extractor was invented by German chemist Franz Ritter Von Soxhlet (1879). It was designed primarily for lipid extraction, but it is no longer restricted to this purpose. It has been widely used to extract valuable bioactive compounds from a variety of natural sources. It is used as a model for comparing new extraction methods. A thimble is usually filled with a

small amount of dry sample. The thimble is then placed in a distillation flask containing the solvent of interest. A syphon is used to aspirate the thimble-holder solution after it has reached an overflow level. Siphon returns the solution to the distillation flask. The extracted solutes are carried into the bulk liquid by this solution. The solute remains in the distillation flask while the solvent returns to the plant's solid bed. The procedure is repeated until the extraction is finished. However, this technique necessitates a longer extraction time as well as a large amount of solvent (Heleno et al., 2016).

## **2. Maceration**

Maceration has been used for a long time in the homemade preparation of tonic. It quickly became a popular and low-cost method of obtaining essential oils and bioactive compounds. Maceration is the process of grinding the sample into smaller particles in order to increase the surface area for a good solvent mixture. Maceration typically consists of several steps for small-scale extraction. First, plant materials are ground into small particles to increase surface area for proper solvent mixing. Second, in the maceration process, a suitable solvent known as menstruum is added to a closed vessel. Finally, the liquid is strained, but the marc, the solid residue of this extraction process, is pressed to recover a large amount of occluded solutions. Filtration is used to separate impurities from the strained and press out liquids. Occasional shaking in maceration aids extraction in two ways: (a) it increases diffusion, and (b) it removes concentrated solution from the sample surface, allowing new solvent to enter the menstruum and increase extraction yield. This method has long been used to obtain essential oils and bioactive compounds (Azmir et al., 2013).

## **3. Hydrodistillation**

Bioactive substances and essential oils are traditionally extracted from plants via hydro-distillation. It can be done prior to the dehydration of plant materials and does not require the use of organic solvents. Water distillation, water and steam distillation, and direct steam distillation are the three different kinds of hydro-distillation (Vankar, 2004). In the process of hydro-distillation, the plant materials are first packed into a still compartment, and then enough water is added and brought to a boil. Another option is to introduce direct steam directly into the plant sample. The key influencing variables that liberate the bioactive chemicals from plant tissue are hot water and steam. The vaporised combination of water and oil is condensed by indirect water cooling. Oil and bioactive chemicals automatically separate from the water in a separator as the condensed mixture flows from the condenser there (Silva et al., 2015). Three primary physicochemical processes-hydro-diffusion, hydrolysis, and thermal decomposition-are involved in hydro-distillation. A high temperature for extraction may cause some volatile components to evaporate. This flaw restricts its application to the extraction of thermolabile compounds. With hydro-distillation, both volatile and non-volatile organic molecules can be

physically removed and separated in a single step. Azeotropic distillation is used to extract the volatile organic components from the matrix before they are condensed, collected, and sorted in a Florentine flask. In the boiling water that is in contact with the matrix inside the alembic, the soluble non-volatile organic molecules are removed. However, hydro-distillation requires a lot of energy and takes a long time (Petigny et al., 2014). Hydro-distillation is performed with distilled water and is used to extract the volatile fraction in foods; this method usually takes 6–8 h and organic solvents are not involved. This technique involves three main physicochemical processes: hydro-diffusion, hydrolysis, and decomposition by heat. High temperatures during extraction can degrade compounds, which limits the use of this technique (Wu et al., 2015).

### **Selection of solvents for extraction**

Since solvents with various polarities are required for identification and isolation, the efficacy of conventional extraction techniques depends on the solvent of choice as well as the polarity of the component. Chemicals have different polarities; hence it is challenging to create a single extraction technique that is effective for all compounds. Low toxicity, a low boiling point, rapid mass transfer, preservation action, and the inability to cause the complex extract to dissociate are all characteristics of a good solvent. Examples of some extracted bioactive compounds by different solvents are listed in Table 1.

**Table 1: Solvents used for bioactive compound extraction**

<b>Solvent</b>	<b>Bioactive compound</b>
Water	Anthocyanins, Tannins, Saponins and Terpenoids
Ethanol	Tannins, Polyphenols, Flavanol, Terpenoids, Alkaloids, Tyrosinase, Polyphenols, Catechins, Punicalagin and Flavonoids
Methanol	Anthocyanin, Terpenoids, Saponins, Tannins, Flavones and Polyphenols
Chloroform	Terpenoids and Flavonoids
Dichloromethane	Terpenoids
Ether	Alkaloids and Terpenoids
Acetone	Flavonoids

(Source: Cowan (1999), Azmir et al., 2013)

Organic solvents (liquid-liquid/liquid-solid) are commonly used to extract bioactive compounds from fruit residues using various conventional methods such as maceration, Soxhlet extraction, and hydro-distillation, which all use heat and/or agitation. However, these techniques have drawbacks such as the use of high purity solvents, high cost and potential toxicity, low selectivity in extraction, long extraction times, and thermal decomposition of heat-labile

compounds (Banerjee et al., 2017). As an alternative, several researchers have investigated emerging extraction techniques such as Pressurized Liquid Extraction (PLE), Microwave Assisted Extraction (MAE), Supercritical Fluid Extraction (SFE), Enzyme assisted extraction, Pulsed Electric Fields (PEF), High pressure extraction, Cloud point extraction (CPE), and based on enzymes that include non-thermal processes, microwaves, and ultrasound.

### **Modern methods of extraction:**

#### **1. Microwave Assisted Extraction (MAE):**

The use of microwave energy to enhance the extraction of phytochemicals has been gaining prime importance. Its applications overcome traditional methods which are lacking extraction time and efficiency, also keeping quality of thermo labile bioactive compounds. Basically, the microwave assisted technique uses an application of certain capacities of microwaves radiations to improve structural changes in cell wall, heating, porosity changes that facilitate easy leach out of compounds from plant matrix. The compounds are extracted in the solvents *viz*, ethanol, water, or methanol then that mixture was evaporated to obtain clean phytochemicals.

MAE compound tyrosinase from black mulberry gives higher yields at 500W in 10 minutes of time. Phytophenols from grape marcs were extracted in 48% ethanol as solvent in MAE (Garrido et al., 2019). Maximum yield obtained of total anthocyanin content (TAC) from grape seed was obtained in 9.3 min of extraction time at 48°C. Punicalagin from pomegranate were extracted, the optimal solvent/solid ratio was 60mL/g at 600W microwave power. The MAE reduced use of volume of organic solvents (Alupului, 2012). Extraction of bioactive compounds from *Glycine max*, peel of *Punicagranatum*, *Hippophaerhamnoides*, *Radix astragalial* also reported (Baydar et al., 2006). MAE was efficiently used for extraction of essential oils from peels of *Citrus* spp., *Origanumvulgare*, *Menthapipenta*, and menthol mint (Razzaghi et al., 2019).

#### **2. Super Critical Fluid Extraction (SCF):**

Bioactive compounds or essential oils are extracted from plant materials by means of solvents by increasing their pressure and temperature to critical stage. The critical stage where either temperature or pressure of the solvent does not come under liquid or gas stage. Carbon dioxide at its critical stage (pressure = 7.38MPa, temperature = 31.1°C) can be used in supercritical fluid extraction technique for plant materials. Super critical CO<sub>2</sub> is widely applicable in extraction as it is odourless, colourless, non-inflammable, pure, safe and economic solvent. Higher diffusive rates of SC CO<sub>2</sub> leach out compounds or oils from matrix very efficiently, that minimizes the residual percentages.

The polar compounds are poorly extracted by SC SO<sub>2</sub> as its lipophilic property make unsuitable for it. This can be eliminated by adding a small amount of co-solvent like ethanol or water (less than 5%w/w) this moves

extraction process into two-phases. SC CO<sub>2</sub> as time consuming process, but its preconditioned materials can reduce time and increases extraction efficiency. The reduced particle size, higher surface area exposed, lower bed density of material, viscosity, solvent power, facilitate easy penetration and diffusion (Silva et al., 2016). The applications of SC CO<sub>2</sub> are widespread in food analysis, environmental, pharmaceutical and polymer sciences. Earlier this technology was used for decaffeinated coffee preparations, polyphenols extraction from herbs and grape seed, catechin from grape seed, flavonoids from citrus paradise. Sunflower oil, palm kernel oil, poppy seed oil was efficiently extracted by use of SCF extraction process.

### **3. Pulsed Electric Field (PEF) extraction:**

In the pulsed electric field extraction process principle of electric field to separate dipole molecules according to charge was applied to disrupt the cell structure to facilitate extraction. The treatment improves diffusion process, permeability which increases the rate of extraction in given exposure time. The application of electric field given to specimen of plant part, where two electrodes are used to hold that part, the design are available in batch or continuous modes. The critical electric potential nearly of 1V required to create repulsion in molecule carrying charge. The performance of treatment varies according to number of pulses, specific energy, temperature, and material characteristics. Minimum time exposure to electric pulses benefits heat sensible compounds to retain quality in solid liquid phase extraction. The moderate level of 500 and 1000V/cm electric field for 10<sup>-4</sup> to 10<sup>-2</sup> s time will damage the cell membranes and tissue (Fincan & Dejmek, 2002). The pulsed electric field of 2.7kV/cm was used as pretreatment in protein extract from beer (Ganeva et al., 1999). However higher intensities of electric field intensity above 10kV/cm were rarely applied in PEF technique (Parniakov et al., 2014). Moreover, in assisted extraction process limit use of low intensities industrially. To overcome the researchers found that higher intensities exceeding 10kV/cm were better for extraction of bovine serum albumin. High intensity pulsed electric fields ranging between 20 and 80 kV/cm are involved in food processing applications.

Potential applications of PEF extraction are widespread for varied compounds. The saccharides from plant, animal and microorganisms sources are extracted in solvents by PEF (He et al., 2020). High intensity PEF works at room temperature with minimum exposure time and in low solvent use. However, other methods like SC CO<sub>2</sub>, alkali extraction took more time and they cause damage to heat sensible compounds in comparison to PEF extraction process (Cardenas-Toro et al., 2015). Economically important bioactive compounds such as DNA, betulin, anthocyanins, chondroitin sulphate, quercetin, epigallocatechin, proteins, amino acids etc. were previously been extracted with this technology. The extraction of bioactive compounds from food wastes can be recovered by PEF extraction process (Han et al., 2020). Citrus peel oil, pomegranate peel, eggshell calcium and essential oils are efficiently extracted by this method (Yan et al., 2017).

#### **4. Ultrasonication Extraction (UAE):**

Ultrasound waves include mechanical vibrations, with frequency greater than 20kHz. The technique specified that the acoustic cavitation in UAE results agitation of the cell walls, particles size reduction and amplification of exposure between solvent and targeted compounds. Mechanism of Ultrasonication: 1) US transducer converts electrical energy into sound energy. 2) Sound waves create small bubbles in liquid foods. 3) The numerous formed bubbles collapse and each creates high temperature and pressure 4) this causes destruction of cell walls and surge the discharge of intracellular components into the solvent. The process by which bubbles form, grow and collapse is known as cavitation. It was manifested that ultrasound can stimulate the extraction of organic compounds contained with the plant tissues. The main reason for choosing hybrid extraction like ultrasonication, that it has high extraction efficiency feasible to standardize heat transfer increased and good quality and lower use of solvent.

In the ultrasonicated extraction of lycopene from tomato the optimized conditions of extraction time (45.6min), temperature (47.6°C), and 74:4:1 v/w (ratio of solvent to freeze-dried tomato sample) achieved extraction yield of trans-lycopene by 75.93% compared to optimized conventional method (Eh et al., 2012). Extraction of pectin from the custard apple peel using ultrasound technique obtained maximum yield of 8.93%with optimized conditions of liquid-solid ratio (23.52mL/g, ultrasonication time 18.04min, and temperature 63.22°C and solution pH 2.36. Custard apple peel pectin rich in galacturonic acid, high molecular weight, good in emulsifying property, antioxidants obtained by using this process.

#### **5. Enzyme Assisted Extraction (EAE):**

Enzyme assisted extraction is an emerging approach among green extraction techniques, and plays a significant role in the food and pharmaceutical industry (Soquetta et al., 2018). The enzymes are used to ameliorate the extraction process by hydrolyzing matrix of the plant cell wall. Enzymatic reactions degenerate the cell wall and expansion of cell permeability. This technique depends on temperature, pH, time, particle size of plant material and solvent used (Poojari et al., 2017). The most frequently used enzymes for extraction of bioactive compounds from food materials are cellulase, protease, hemicellulose, xylase, palygalacturonase,  $\alpha$ -amylase, neutrase,  $\beta$ -glucosidase and pectinesterase (Jeong et al., 2014). EAE is noted as an environmentally safe technique because enzymes are organic materials and low preparation concentration used. The bioactive compounds extracted from enzyme aided extraction exhibit superior quality, precise, consumes lower energy and time compared to conventional extraction techniques.

Extraction of pectin from apple pomace through enzyme assisted extraction, increase the higher pectin efficiency and yield of 19.8% was obtained after treatment with endo-1, 4-xylanase at pH 5.0, temperature of 40°C, and solid-liquidratio 1g/15ml. Higher amount of protein and phenols contained

pectin isolated with endo-xylanase (4.38% and 1.34% respectively) and exhibits good emulsifying properties. The phenolics extracted from grape seed through enzyme assisted extraction obtained highest yield with optimized conditions at 48°C for 2.4 hr, pH 3.5 and enzyme dosage of 20mg/g. EAE is more efficient method for the extraction of phenols from grape seed compared to UAE method. This technique not only releases larger amount of phenolic but also exhibits free radical scavenging activity.

## **6. High pressure extraction (HPP):**

High pressure is a non-thermal technique which plays a significant role in the extraction of bioactive compounds from food materials, inactivates the enzyme and also used as preservative technology in the food industry. HPP is also called high hydrostatic pressure processing or ultra-high pressure and it is acknowledged by US-FDA as an ecofriendly extraction method (Vorobiev and Lebovka, 2006). It operates at a high pressure of 100–600MPa and low temperature up to 60°C, to extract rapidly with low volumes of organic solvents and furnish recoveries like other technologies. The increase in pressure leads to a decrease in volume of liquid food (15% volume reduction in application of 600 MPa pressure). High pressure treatments lead to breakdown of the plant tissues, cell wall, membranes, and organelles, which boost the mass transfer rate of solvent into the material and the soluble constituents into the solvents. HPP can increase mass transfer rates by altering the concentration gradient and diffusivity, causing breakdown of the plant cell membrane and its permeability increases and improves the penetration of solvent into the cells. This advanced technology helps in shortening extraction time and hence, cost effectiveness. Higher extraction yields, requires lower energy requirements, prevents thermal degradation, and retains bioactivity of extracted compounds. Bioactive compounds extracted from aronia berry puree through high pressure processing at the pressure of 400 and 600 MPa for 5min lead to preserve the phenolic and anthocyanin content (Yuan et al., 2018). Total phenols can be extracted from sour cherry by using HPP extraction method at 500 MPa pressure.

## **7. Cloud Point Extraction (CPE)**

The CPE is a novel technology for extraction of functional components as it comes under non-conventional green technology. No requirement of sophisticated equipment and skilled labor are additional features of it. Hydrophobic bioactive compounds like phenolic compounds, carotenoids, essential oils, essential fatty acids, and water insoluble vitamins can be extracted through this method using non-ionic surfactants. Apart from this it has wide applications in the food processing industry like separation and purification of proteins, sample preparation method for analysis of food constituents.

In CPE a hydrophobic core is developed towards the center of the micelle when the mixture of micelle (surfactant rich phase), and sample is heated beyond



the cloud point temperature of the surfactant. The concentration of surfactant should be more than the CMC (Critical micellar concentration). Two isotropic phases comprising of co-acervate phase (dense phase or surfactant rich phase), and bulk phase (lean phase or aqueous phase) are formed. Bioactive compounds isolated from these different phases are quantified through any analytical technique. There are different factors that affect the cloud point extraction process like cloud point temperature, time of extraction, concentration of surfactant, nature of surfactants used, solvent-to-material ratio, presence of additives and physicochemical properties of solutes. It has tremendous applications in extraction of bioactive compounds from food processing by-products like extraction of lycopene from tomato peel, thymol from *ajwain* seeds (*Trachysper mumammī*), phenols and carotenoids from red-flesh orange juice and olive mill wastewater. However, CPE can also be used as an integrative approach for concentration of extracted bioactive compounds with other extraction techniques like ultrasound assisted extraction of alkaloids and polyphenols from mulberry leaf and extraction of chlorophyll from spinach leave using solid liquid extraction (Leite et al., 2018). The following table explains the different extraction techniques used for extraction of bioactive compounds from plants sources. These modern methods of extraction was successfully applied to different commodities for extraction of bioactive as shown in Table 2.

**Table 2. Modern methods of extraction for various bioactive compounds from agricultural commodities**

Method of extraction	Extracted Compound	Raw material	Silent Findings	Reference
MAE	Tyrosinase	Black mulberry	Optimum extract at 500W, 35% ethanol, 10min.	Koyu et al., 2017
MAE	Polyphenols	Grape marc	48% ethanol, 10 min, and 1.77 g sample	Garrido et al., 2019
MAE	Catechins	Grape seed	94% ethanol, 170°C temperature and 55 min	Xiong et al., 2020
MAE	Total anthocyanin content	Saffron Tepals	48°C temperature, 9.3 min, 77.5 ml solvent/g saffron tepals	Garavand et al., 2019
MAE	Punicalgin	Pomegranate	50% aqueous ethanol, 60/1 ml/g solvent : solid ratio, 600W microwave power	Kaderides et al., 2019

Method of extraction	Extracted Compound	Raw material	Silent Findings	Reference
MAE	Flavonoids	Pomegr- anate peel	4.26% yield achieved at 60% ethanol, 40:1 solvent : solid ratio for 3 min time	Huang et al., 2017
PEF	Isoflavon- oids	Soybeans	50 pulse of 1.3kV/cm gives 20% higher isoflavonoids in comparison to referent sample	Guderjan et al., 2005
PEF	Tocopherols and polyphenols	Rapeseed	5kV/cm at 60 pulses and 7kV/cm for 120 pulses increases polyphenolic content and tocopherols	Guderjan et al., 2007
PEF	Total polyphenols	Tomato fruits	44% increase in polyphenols content under 30 pulses at 1.2kV/cm	Vallverdu-Queralt et al., 2012
SFE	Polyacetylenes	Bidens- pilosa	Cabon dioxide at 40°C and 250 bar at 15g/min for 240 min.	Cortes-Rojas et al., 2013
SFE	Flavonoids	Mentha spicata	CO <sub>2</sub> as solvent at 60°C, 200bar, 15g/min for 60 min	Bimakr et al., 2011
SFE	Carotenoids	Bactrisg- asipaes	CO <sub>2</sub> as solvent at 40°C, 300bar, 3L/min for 91 min	Espinosa-Pardo et al., 2014
SFE	Triterpene (melianone)	<i>Melia azedarach</i> L.	CO <sub>2</sub> at 60°C, 250bar 2g/min for 150min.	Scapinello et al., 2014
SFE	Curcumin	Turmeric rhizomes ( <i>curcuma longa</i> L.)	CO <sub>2</sub> at 60°C and 400bar, ethanol at 25°C and water at 60°C.	Martinez-Correa et al., 2017
SFE	Apple seed oil	Apple seed	191g extract/kg CO <sub>2</sub> at 1300 bar and at 63°C	Montanes et al., 2018

<b>Method of extraction</b>	<b>Extracted Compound</b>	<b>Raw material</b>	<b>Silent Findings</b>	<b>Reference</b>
USE	Pectin	Pomegrate peel	20kHz frequency, 61.9°C temp, pH of 1.2 and liquid to solid ratio 17:1 gives 23.87% yield	Moorthy et al., 2015
USE	Pectin	Tomato waste	37kHz frequency, time 15 min	Grassino et al., 2016
USE	Dietary fibre	Banana bract	Power intensity 50%, temperature 80oC, time 10 min, liquid to solid ratio 25:1 to achieve 71% yield in NaOH solvent	Begum et al., 2019
USE	Tartaric acid and malic acid	Grape seed	Power intensity of 30 and 70 % at 20°C & 50°C for 5 and 15 min in water and methanol solvents	Palma et al., 2002
USE	Phenolics/ antioxidants /flavonoids	Mango peel	Temp of 30°C, time 20 min, liquid to solid ratio 25:1 at 20kHz in ethanol solvent	Guadalini et al., 2019
USE	Phenolics/ flavonoids	Soybean germ	LSR 6.25:1 temperature of 45oC, power 60-80W at 19-40kHz frequency	Gravotto et al., 2008
CPE	Phenolic compounds	Carica papaya leaves	A total of 60.1% total phenolic content with a DPPH free radical scavenging activity of 81.9% was recovered in the micelle-rich phase of the 10% (w/w) Pluronic L-61	Rodrigues et al., 2019

<b>Method of extraction</b>	<b>Extracted Compound</b>	<b>Raw material</b>	<b>Silent Findings</b>	<b>Reference</b>
CPE	Phenolic compounds	Table olive processing waste-waters	The most appropriate conditions were found to be 10% of Genapol X-080 (w/v), a pH value of 2, a temperature of 70 °C and an equilibrium time of 30 min. With a yield of 68%	Kiai et al., 2018
CPE	Thymol	Ajwain seed	Maximum extraction efficiency of thymol Was achieved with 30% (v/v) of SPAN 80 surfactant, 45 min of heating at 65 °C.	Chatterjee et al., 2017
CPE	B-carotene	Commercial fruit Juice	A detection limit of 0.01 mg/L of beta-carotene (3SB/m), A coefficient of determination of 0.998 and a linear range of 0.04-10 mg/L were obtained.	Safdarian et al., 2021
CPE	Phenols and Flavonoids	Pomegranate peel	The total yield obtained for total phenols was 205.2 mg of GAE/g while for total flavonoids it was 60.05 mg of QE/g of pomegranate peel powder.	More et al., 2019
EAE	Lycopene	Tomato	Enzyme aided extraction increases lycopene extraction yield by 132 µg/g (198%) when treated by cellulose(3%w/w) and 108 µg/g (224%) in case of pectinase (0.5%w/w) enzyme treatment.	Choudhari and Ananthanarayan, 2007

Method of extraction	Extracted Compound	Raw material	Silent Findings	Reference
EAE	Polyphenols	Sweet cherry ( <i>Prunus avium</i> L.) pomace	Optimal extraction with Depol, Promod and Pectinase enzymes obtained at temperature 70°C and a pH of 10.0, extraction time was 40 min for Depol and Promod enzymes and 18.4 min for Pectinase enzyme while the optimum enzyme concentration was 140 µL of Promod /g of sample, 90 µL of Depol/g of sample, and 2 µL of Pectinase/g of sample	Domin-guez et al., 2021
EAE	Gingerol, oleoresin	Ginger	Higher yields of oleoresin (20–21%) and gingerol (10.1–12.2%) on pre-treatment of ginger with cellulase, amylase, pectinase and viscozyme enzymes at 0.5% concentration followed by extraction with acetone.	Manasa et al., 2013
EAE	Essential oils	Bay leaves ( <i>Laurus nobilis</i> L.)	Pretreatment of enzymes namely cellulase, hemicellulase, xylanase and the ternary mixture of them resulted increase in 243, 227, 240.54 and 0.48% increase in the essential oil yields in samples	Boulila et al., 2015
EAE	Glucose	Grape-fruit peel waste	5mg pectinase/g peel dry matter and 2 mg cellulase/g peel dry matter were the lowest loadings to yield the most glucose at optimum pH of 4.8.	Wilkins et al., 2007

### **Future thrust:**

In the realm of future research endeavors, the integration of advanced extraction techniques emerges as a compelling avenue for augmenting efficiency and yield. The synergistic amalgamation of methodologies such as Microwave Assisted Extraction (MAE) with Ultrasonication Extraction (UAE) or Supercritical Fluid Extraction (SCF) with Enzyme Assisted Extraction (EAE) holds substantial promise for optimizing the extraction process. Furthermore, the exploration of nanoencapsulation and delivery systems for bioactive compounds extracted through these techniques presents a fertile ground for enhancing bioavailability and stability, thereby unlocking novel prospects for bolstering functional properties in both food and pharmaceutical domains.

Moreover, the tailored refinement of extraction protocols tailored to specific bioactive compounds heralds an intriguing frontier for scientific inquiry. By meticulously elucidating the intricate chemical properties and structural nuances of target compounds, researchers can ingeniously engineer extraction processes geared towards maximal yield while minimizing energy expenditure and environmental impact, thereby catalyzing the transition towards more sustainable extraction methodologies. Concomitantly, addressing the formidable challenges inherent in scaling up and industrially implementing these advanced extraction techniques is paramount for unleashing their transformative potential. The development of strategic frameworks to surmount hurdles such as cost, scalability, and regulatory compliance stands poised to streamline the widespread adoption of these methods in the expansive domains of large-scale food and pharmaceutical production, thereby fostering consequential integration within industrial frameworks. Furthermore, the stringent focus on establishing robust quality control paradigms and standardization protocols assumes pivotal importance in ensuring the reproducibility and fidelity of extracted compounds, thereby engendering unwavering trust and credibility in the utility of advanced extraction techniques across multifarious industrial sectors.

### **Conclusion:**

In conclusion, the chapter delves into the transformative impact of advanced extraction techniques on obtaining bioactive compounds from natural sources. These methods, including Microwave Assisted Extraction, Supercritical Fluid Extraction, and Ultrasonication Extraction, offer unparalleled efficiency, sustainability, and versatility compared to traditional approaches. By harnessing innovative principles such as enzyme assistance and high-pressure processing, these techniques facilitate the extraction of a diverse array of compounds with enhanced purity and bioactivity. Moreover, their eco-friendly nature aligns with the growing demand for sustainable practices in the food and pharmaceutical industries. Looking ahead, the integration of these techniques, exploration of underutilized sources, and advancements in nanoencapsulation hold promise for

unlocking new frontiers in functional food and pharmaceutical development. However, challenges remain in scaling up these processes and ensuring standardization and quality control in industrial settings. Nevertheless, with continued research and collaboration, these advanced extraction methods are poised to revolutionize the production of high-quality, bioactive-rich extracts, contributing to improved health outcomes and sustainable development.

## Reference

- Alupului, A., Calinescu, I., & Lavric, V. (2012). Microwave extraction of active principles from medicinal plants. *UPB Science Bulletin, Series B*, 74(2), 129-142.
- Azmir, J., Zaidul, I. S. M., Rahman, M. M., Sharif, K. M., Mohamed, A., Sahena, F., & Omar, A. K. M. (2013). Techniques for extraction of bioactive compounds from plant materials: A review. *Journal of Food Engineering*, 117(4), 426–436.
- Banerjee, J., Singh, R., Vijayaraghavan, R., MacFarlane, D., Patti, A. F., & Arora, A. (2017). Bioactives from fruit processing wastes: Green approaches to valuable chemicals. *Food chemistry*, 225, 10-22.
- Baydar, N. G., Sagdic, O., Ozkan, G., & Cetin, S. (2006). Determination of antibacterial effects and total phenolic contents of grape (*Vitis vinifera* L.) seed extracts. *International journal of food science & technology*, 41(7), 799-804.
- Begum, Y. A., & Dekka, S. C. (2019). Effect of processing on structural, thermal, and physicochemical properties of dietary fiber of culinary banana bracts. *Journal of Food Processing and Preservation*, 43(12), e14256.
- Bimakr, M., Rahman, R. A., Taip, F. S., Ganjloo, A., Salleh, L. M., Selamat, J., ... & Zaidul, I. S. M. (2011). Comparison of different extraction methods for the extraction of major bioactive flavonoid compounds from spearmint (*Mentha spicata* L.) leaves. *Food and bioproducts processing*, 89(1), 67-72.
- Boulila, A., Hassen, I., Haouari, L., Mejri, F., Amor, I. B., Casabianca, H., & Hosni, K. (2015). Enzyme-assisted extraction of bioactive compounds from bay leaves (*Laurus nobilis* L.). *Industrial Crops and Products*, 74, 485-493.
- Cardenas-Toro, F. P., Alcázar-Alay, S. C., Coutinho, J. P., Godoy, H. T., Forster-Carneiro, T., & Meireles, M. A. A. (2015). Pressurized liquid extraction and low-pressure solvent extraction of carotenoids from pressed palm fiber: experimental and economical evaluation. *Food and Bioproducts Processing*, 94, 90-100.
- Chatterjee, S., Jain, A., & De, S. (2017). Effect of different operating conditions in cloud point assisted extraction of thymol from Ajwain (*Trachyspermum Ammi* L.) seeds and recovery using solvent. *Journal of food science and technology*, 54(13), 4353-4361.
- Choudhari, S. M., & Ananthanarayan, L. (2007). Enzyme aided extraction of lycopene from tomato tissues. *Food chemistry*, 102(1), 77-81.

- Cortes-Rojas, D. F., Chagas-Paula, D. A., Da Costa, F. B., Souza, C. R., & Oliveira, W. P. (2013). Bioactive compounds in *Bidens pilosa* L. populations: a key step in the standardization of phytopharmaceutical preparations. *Revista Brasileira de Farmacognosia*, 23(1), 28-35.
- Domínguez-Rodríguez, G., Marina, M. L., & Plaza, M. (2021). Enzyme-assisted extraction of bioactive non-extractable polyphenols from sweet cherry (*Prunus avium* L.) pomace. *Food Chemistry*, 339, 12808.
- Eh, A. L. S., & Teoh, S. G. (2012). Novel modified ultrasonication technique for the extraction of lycopene from tomatoes. *Ultrasonics sonochemistry*, 19(1), 151-159.
- Espinosa-Pardo, F. A., Martinez, J., & Martinez-Correa, H. A. (2014). Extraction of bioactive compounds from peach palm pulp (*Bactris gasipaes*) using supercritical CO<sub>2</sub>. *The Journal of Supercritical Fluids*, 93, 2-6.
- Fincan, M., & Dejmek, P. (2002). In situ visualization of the effect of a pulsed electric field on plant tissue. *Journal of food engineering*, 55(3), 223-230.
- Ganeva, V., & Galutzov, B. (1999). Electropulsation as an alternative method for protein extraction from yeast. *FEMS Microbiology letters*, 174(2), 279-284.
- Garavand, F., Rahaee, S., Vahedikia, N., & Jafari, S. M. (2019). Different techniques for extraction and micro/nanoencapsulation of saffron bioactive ingredients. *Trends in Food Science & Technology*, 89, 26-44.
- Garrido, T., Gizdavic-Nikolaidis, M., Leceta, I., Urdanpilleta, M., Guerrero, P., de la Caba, K., & Kilmartin, P. A. (2019). Optimizing the extraction process of natural antioxidants from chardonnay grape marc using microwave-assisted extraction. *Waste Management*, 88, 110-117.
- Goldberg, E., Grant, J., Aliani, M., & Eskin, M. N. (2017). *10 methods for removing bitterness in functional foods and nutraceuticals*. USA: John Wiley & Sons, Inc.
- Grassino, A. N., Brnčić, M., Vikić-Topić, D., Roca, S., Dent, M., & Brnčić, S. R. (2016). Ultrasound assisted extraction and characterization of pectin from tomato waste. *Food chemistry*, 198, 93-100.
- Guandalini, B. B. V., Rodrigues, N. P., & Marczak, L. D. F. (2019). Sequential extraction of phenolics and pectin from mango peel assisted by ultrasound. *Food Research International*, 119, 455-461.
- Guderjan, M., Elez-Martínez, P., & Knorr, D. (2007). Application of pulsed electric fields at oil yield and content of functional food ingredients at the production of rapeseed oil. *Innovative Food Science & Emerging Technologies*, 8(1), 55-62.
- Guderjan, M., Töpfl, S., Angersbach, A., & Knorr, D. (2005). Impact of pulsed electric field treatment on the recovery and quality of plant oils. *Journal of Food Engineering*, 67(3), 281-287.
- Han, Z., Han, Y., Wang, J., Liu, Z., Buckow, R., & Cheng, J. (2020). Effects of pulsed electric field treatment on the preparation and



- physicochemical properties of porous corn starch derived from enzymolysis. *Journal of Food Processing and Preservation*, 44(3), e14353.
- He, G., Yin, Y., Yan, X., & Yu, Q. (2014). Optimisation extraction of chondroitin sulfate from fish bone by high intensity pulsed electric fields. *Food Chemistry*, 164, 205-210.
- Heleno, S. A., Diz, P., Prieto, M. A., Barros, L., Rodrigues, A., Barreiro, M. F., & Ferreira, I. C. (2016). Optimization of ultrasound-assisted extraction to obtain mycosterols from *Agaricus bisporus* L. by response surface methodology and comparison with conventional Soxhlet extraction. *Food Chemistry*, 197, 1054–1063.
- Huang, J., He, W., Yan, C., Du, X., & Shi, X. (2017). Microwave assisted extraction of flavonoids from pomegranate peel and its antioxidant activity. In *BIO Web of Conferences* (Vol. 8, p. 03008). EDP Sciences.
- Jeong, H. S., Kim, H. Y., Ahn, S. H., Oh, S. C., Yang, I., & Choi, I. G. (2014). Optimization of enzymatic hydrolysis conditions for extraction of pectin from rapeseed cake (*Brassica napus* L.) using commercial enzymes. *Food chemistry*, 157, 332-338.
- Kaderides, K., Papaoikonomou, L., Serafim, M., & Goula, A. M. (2019). Microwave-assisted extraction of phenolics from pomegranate peels: Optimization, kinetics, and comparison with ultrasounds extraction. *Chemical Engineering and Processing-Process Intensification*, 137, 1-11.
- Kiai, H., Raiti, J., El-Abbassi, A., & Hafidi, A. (2018). Recovery of phenolic compounds from table olive processing wastewaters using cloud point extraction method. *Journal of environmental chemical engineering*, 6(1), 1569-1575.
- Koyu, H., Kazan, A., Demir, S., Haznedaroglu, M. Z., & Yesil-Celiktas, O. (2018). Optimization of microwave assisted extraction of *Morus nigra* L. fruits maximizing tyrosinase inhibitory activity with isolation of bioactive constituents. *Food chemistry*, 248, 183-191.
- Leite, A. C., Ferreira, A. M., Morais, E. S., Khan, I., Freire, M. G., & Coutinho, J. A. (2018). Cloud point extraction of chlorophylls from spinach leaves using aqueous solutions of nonionic surfactants. *ACS sustainable chemistry & engineering*, 6(1), 590-599.
- Manasa, D., Srinivas, P., & Sowbhagya, H. B. (2013). Enzyme-assisted extraction of bioactive compounds from ginger (*Zingiber officinale* Roscoe). *Food Chemistry*, 139(1-4), 509-514.
- Green
- Soquetta, M. B., Terra, L. D. M., & Bastos, C. P. (2018). Green technologies for the extraction of bioactive compounds in fruits and vegetables. *CyTA-Journal of Food*, 16(1), 400-412.
- Martinez-Correa, H. A., Paula, J. T., Kayano, A. C. A., Queiroga, C. L., Magalhães, P. M., Costa, F. T., & Cabral, F. A. (2017). Composition and antimalarial activity of extracts of *Curcuma longa* L. obtained by a combination of extraction processes using supercritical

- CO<sub>2</sub>, ethanol and water as solvents. *The Journal of Supercritical Fluids*, 119, 122-129.
- Montanes, F., Catchpole, O. J., Tallon, S., Mitchell, K. A., Scott, D., & Webby, R. F. (2018). Extraction of apple seed oil by supercritical carbon dioxide at pressures up to 1300 bar. *The Journal of Supercritical Fluids*, 141, 128-136.
- Moorthy, I. G., Maran, J. P., Muneeswari, S., Naganyashree, S., & Shivamathi, C. S. (2015). Response surface optimization of ultrasound assisted extraction of pectin from pomegranate peel. *International journal of biological macromolecules*, 72, 1323-1328.
- More, P. R., & Arya, S. S. (2019). A novel, green cloud point extraction and separation of phenols and flavonoids from pomegranate peel: An optimization study using RCCD. *Journal of Environmental Chemical Engineering*, 7(5), 103306.
- Palma, M., & Barroso, C. G. (2002). Ultrasound-assisted extraction and determination of tartaric and malic acids from grapes and winemaking by-products. *Analytica Chimica Acta*, 458(1), 119-130.
- Parniakov, O., Barba, F. J., Grimi, N., Lebovka, N., & Vorobiev, E. (2014). Impact of pulsed electric fields and high voltage electrical discharges on extraction of high-added value compounds from papaya peels. *Food Research International*, 65, 337-343.
- Petigny, L., Perino, S., Minuti, M., Visinoni, F., Wajsman, J., & Chemat, F. (2014). Simultaneous microwave extraction and separation of volatile and non-volatile organic compounds of boldo leaves. From lab to industrial scale. *International Journal of Molecular Sciences*, 15(5), 7183-7198.
- Poojary, M. M., Orlie, V., Passamonti, P., & Olsen, K. (2017). Enzyme-assisted extraction enhancing the umami taste amino acids recovery from several cultivated mushrooms. *Food Chemistry*, 234, 236-244.
- Razzaghi, S. E., Arabhosseini, A., Turk, M., Soubrat, T., Cendres, A., Kianmehr, M. H., & Chemat, F. (2019). Operational efficiencies of six microwave-based extraction methods for orange peel oil. *Journal of Food Engineering*, 241, 26-32.
- Rodrigues, L. G. G., Mazzutti, S., Vitali, L., Micke, G. A., & Ferreira, S. R. S. (2019). Recovery of bioactive phenolic compounds from papaya seeds agro industrial residue using subcritical water extraction. *Biocatalysis and Agricultural Biotechnology*, 22, 101367.
- Safdarian, M., Hashemi, P., & Ghiasvand, A. (2021). A fast and simple method for determination of  $\beta$ -carotene in commercial fruit juice by cloud point extraction-cold column trapping combined with UV-Vis spectrophotometry. *Food Chemistry*, 343, 128481.
- Scapinello, J., Oliveira, J. V., Ribeiros, M. L., Tomazelli Jr, O., Chiaradia, L. A., & Dal Magro, J. (2014). Effects of supercritical CO<sub>2</sub> extracts of *Melia azedarach* L. on the control of fall armyworm (*Spodoptera frugiperda*). *The Journal of Supercritical Fluids*, 93, 20-26.
- Silva, C.M., Zanqui, A.B., Gohara, A.K., Souza, A.H.P., Cardozo-Filho, L.,

- Visentainer, J.V., Chiavelli, L.U.R., Bittencourt, P.R.S., Silva, E.A., Matsushita, M., 2015. Compressed n-propane extraction of lipids and bioactive compounds from Perilla (*Perillafrutescens*). *J. Supercrit. Fluid* 102, 1–8.
- Silva, J. R., Cantelli, K. C., Soares, M. B., Tres, M. V., Oliveira, D., Meireles, & Mazutti, M. A. (2015). Enzymatic hydrolysis of non-treated sugarcane bagasse using pressurized liquefied petroleum gas with and without ultrasound assistance. *Renewable Energy*, 83, 674–679.
- Vallverdu-Queralt, A., Oms-Oliu, G., Odriozola-Serrano, I., Lamuela-Raventos, R. M., Martin-Belloso, O., & Elez-Martinez, P. (2012). Effects of pulsed electric fields on the bioactive compound content and antioxidant capacity of tomato fruit. *Journal of agricultural and food chemistry*, 60(12), 3126–3134.
- Vankar, P.S., 2004. Essential oils and fragrances from natural sources. *Resonance* 9(4), 30–41.
- Vorobiev, E., & Lebovka, N.I., 2006. Extraction of intercellular components by pulsed electric fields. In: Raso, J., Heinz, V. (Eds.), *Pulsed Electric Field Technology for the Food Industry: Fundamentals and Applications*. Springer, New York, pp. 153–194
- Wilkins, M. R., Widmer, W. W., Grohmann, K., & Cameron, R. G. (2007). Hydrolysis of grapefruit peel waste with cellulase and pectinase enzymes. *Bioresource technology*, 98(8), 1596–1601.
- Wu, C., Wang, F., Liu, J., Zou, Y., & Chen, X. (2015). A comparison of volatile fractions obtained from *Lonicera acanthoides* via different extraction processes: Ultrasound, microwave, Soxhlet extraction, hydrodistillation, and cold maceration. *Integrative Medicine Research*, 4(3), 171–177.
- Xiong, Y., Chen, M., Warner, R. D., & Fang, Z. (2020). Incorporating nisin and grape seed extract in chitosan-gelatine edible coating and its effect on cold storage of fresh pork. *Food Control*, 110, 107018.
- Yan, L. G., He, L., & Xi, J. (2017). High intensity pulsed electric field as an innovative technique for extraction of bioactive compounds—A review. *Critical reviews in food science and nutrition*, 57(13), 2877–2888.
- Yan, L. G., He, L., & Xi, J. (2017). High intensity pulsed electric field as an innovative technique for extraction of bioactive compounds—A review. *Critical reviews in food science and nutrition*, 57(13), 2877–2888.
- Yuan, B., Danao, M. G. C., Stratton, J. E., Weier, S. A., Weller, C. L., & Lu, M. (2018). High pressure processing (HPP) of aronia berry purée: Effects on physicochemical properties, microbial counts, bioactive compounds, and antioxidant capacities. *Innovative food science & emerging technologies*, 47, 249–255.