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REVIEW

Recent Advances on Application of Ultrasound and Pulsed Electric Field Technologies in the Extraction of Bioactives from Agro-Industrial By-products

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Abstract Agro-industrial by-products are rich sources of natural bioactive compounds and their valorisation is a must for global food sustainability. Along with conventional techniques, numerous novel methods have been developed and optimised to facilitate extraction of the bioactives in sustainable and efficient manner. This review summarises the recent advances in the application of novel extraction technologies for various classes of bioactive molecules from agro-industrial by-products with special emphasis on two emerging techniques, i.e. ultrasound- and pulsed electric field-assisted extraction. These two technologies have shown promising extraction efficacy with reduced usage of extraction solvents, thus saving time and cost. The mechanism through which these techniques aid extraction, the various parameters affecting their efficacy, integration with other novel techniques, and promising applications for by-product valorisation are discussed.

Keywords Food processing by-products · Phytochemicals · Novel extraction techniques · Ultrasound-assisted extraction (UAE) · Pulsed electric field (PEF)

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Introduction

Agricultural produce, such as cereal grains, fruits and vegetables, herbs and spices, nuts, etc., is a rich source of non-nutritive health-promoting compounds besides nutrients for growth and development. Many of these compounds are secondary metabolites (phytochemicals) produced by the plants' defence mechanism or primary metabolites, which in addition to their nutritive value confer health protecting effects in humans (Joana Gil-Chávez et al. 2013). Apart from being consumed fresh, a number of processed products such as frozen or minimally processed produce, juices, nectars, pulps, concentrates, ketchups/sauces, pickles, jam/jellies/marmalades, soups, crisps/chips and flakes are popular among consumers because of their convenience. Industrial primary processing of plant foods generates in millions of tonnes of by-products annually. Classic examples of processing by-products are damaged raw materials, seeds, peels/skins, husk/hulls/cobs, brans, oilseed cakes, spent grains, molasses, etc., which account for approximately 190 million tonnes per year worldwide (FAO 2013), resulting in significant financial burden to the processors and cause environmental concerns. Several studies have revealed that these by-products are rich sources of non-nutritive but biologically active compounds, thus providing a valid rationale for their recovery from agro-industrial by-products. A variety of bioactive compounds such as phenolics, carotenoids, vitamins and dietary fibre derived from different agro-industrial by-products have been reviewed previously (Schieber et al. 2001; Ayala-Zavala et al. 2011; Balasundram et al. 2006; Larrauri 1999). Whilst knowledge of the hidden potential of these agro-industrial food by-products has been well known for over a decade, yet they are routinely utilised as animal feed or as fertilisers (Schieber et al. 2001). Efficient utilisation of by-products, a low-cost raw material could help to fulfil the growing demand

for natural food ingredients and products in functional foods and nutraceuticals. A primary reason behind under-exploitation of by-products appears to be the flaws of extraction/purification techniques that include labour intensive, time consuming, and non-environmental friendly extraction and purification techniques of target compounds (Joana Gil-Chávez et al. 2013). Thus, the feasibility of realising the potential economic benefits of agricultural by-products could be improved by application of energy efficient, rapid, inexpensive, environmentally friendly, extraction techniques (Ayala-Zavala et al. 2011; Schieber et al. 2001).

There is also a growing realisation that whilst traditional solid–liquid extraction (SLE) techniques can be used to recover bioactive compounds from food processing by-products, they are often time consuming, expensive and un-sustainable. In the past two decades, many other techniques have been investigated including ultrasound-assisted extraction (UAE), pulsed electric field (PEF), pulsed light (PL), high voltage energy discharge (HVED), microwave-assisted extraction (MAE), supercritical fluid extraction (SFE), pressurised liquid extraction (PLE), high hydrostatic pressure extraction (HHP/HPE), enzyme-assisted extraction (EAE) and instant controlled pressure drop-assisted extraction (DIC) (Galanakis 2013; Joana Gil-Chávez et al. 2013, Hossain et al. 2015). These innovative extraction technologies have many advantages compared to traditional approaches (SLE, Soxhlet extraction and distillation) in terms of shorter extraction time at ambient or low temperatures whilst reduce the usage of organic solvents, higher yield and good quality extract (Azmir et al. 2013).

Wijngaard et al. (2012) have reviewed the effectiveness of novel techniques to extract bioactive compounds from by-products with special attention to PLE, SFE and MAE. However, no detailed review for by-products utilisation for recovery of various bioactive compounds using UAE and PEF currently exists. Therefore, the main aim of this review is to provide a recent update on the non-traditional extraction techniques with particular attention to UAE and PEF technologies. Some of the applications of UAE and PEF for extracting various classes of bioactives from diverse group of agro-industrial by-products are also discussed.

Prevalent Bioactives from Agro-Industrial By-products

A variety of bioactive compounds from agro-industrial by-products has been subject of a number of reviews. The commonly found bioactive compounds from various agro-industrial by-products and their biological properties are summarised in Table 1. These include the following: (i) bioactive polysaccharides, (ii) bioactive peptides, (iii) unsaturated fatty acids, (iv) carotenoids, (v) steroids, (vi) vitamins, (vii) pigments, (viii) essential oils, (ix) alkaloids and (x) phenolics. Generally, fruits and vegetables wastes are the most widely investigated substrates for the extraction of phenolic compounds and dietary fibres, whereas cereal crops and mushroom by-products have served as sources of bioactive polysaccharides (Cheung 2013; Wu et al. 2004), and pulses and oilseed crops residues for bioactive peptides and sterols, respectively (Galanakis 2012). For example, apple by-products have been reported to contain a variety of valuable compounds including pectin (a carbohydrate widely used as gelling and stabilising agent), low-molecular-weight polyphenols (flavonols, flavanols), organic acids such as malic acid, monosaccharides (fructose, glucose and sorbitol), cuticular waxes, highly unsaturated fatty oil, carotenoids, tocopherols and high molecular weight condensed polyphenols (Kammerer et al. 2014).

Polysaccharides are widely distributed in nature and they function as structural and storage components such as cellulose, hemicellulose (arabinoxylans), pectin, inulin, chitin/chitosan and β-glucans (Table 1). They are also abundantly present in a variety of agro-industrial by-products such as hulls, husks, pods, peels, stipes of mushroom, spent grains, shells, stems, seeds, stalks, bran and press cakes. Additionally, polysaccharides have shown various bioactive properties such as enhanced mineral absorption, antioxidative, antibacterial functions (Ren et al. 2014), stimulation of the immune system (Smiderle et al. 2011), regulation of lipid metabolism, appetite suppression, body fat reduction and attenuation of oxidative stress (Anderson et al. 1994). Oligosaccharides are low-molecular-weight polysaccharides present as storage carbohydrates and can also be derived from chemo-enzymatic treatment of a polysaccharide-rich by-product. Fructose oligosaccharides (FOS) are present in fruit and vegetable by-products, especially onion wastes (discarded onions) and impart prebiotic effects (Roldan-Marin et al. 2009). Raffinose, α-galacto-oligosaccharides and galactosyl-cyclitols are present in pulse by-products such as hulls, meal and soybean whey and have been purported to induce changes in colon micro biota (Dinoto et al. 2006). For example, inulin and FOS have been shown to improve bowel functionality (Kleessen et al. 1997). In addition, beneficial roles of processing derived oligosaccharides such as xylooligosaccharides from lingo-cellulosic biomass have been reported by Samanta et al. (2015). Some of the reported bioactive sugar alcohols showing functional properties are mannitols from yeast extract displaying diuretic activity (Zhao et al. 2009) and sorbitols from dried fruits and prunes, which confer a laxative effect (Stacewicz-Sapuntzakis et al. 2001).

Bioactive peptides are obtained by enzymatic hydrolysis of dietary proteins, which are otherwise usually inactive. In recent years, several studies have been conducted on the production and isolation of bioactive peptides from dietary proteins of plant origin (Gangopadhyay et al. 2016). However,

Table 1 Different types of plant bioactives from by-products and their bioactivities

Major class	Sub-class	Bioactive compounds	By-product source	Bioactivity/property	References
Carbohydrates	Oligosaccharides	Xylooligosaccharides from xylans, raffinose	Corn stalks and cob, wheat straw, sugarcane bagasse	Prebiotic	(Samanta et al. 2015)
	Polysaccharides	β-glucan (glucose polymer), chitins (N-acetyl glucosamine), pectin, arabinoxylan, inulin, dietary fibres	Brewer's spent grain, mushroom stalks, citrus and apple pomace, crop residues, bran, hulls, peel, chicory roots	Dietary fibre, antioxidant, antitumor, immunomodulatory and hypoglycaemic	(Fu et al. 2006)
	Sugar alcohols	Mannitol, sorbitol	Olive leaves	Sweetener, laxative, used in diabetic food products	(Ghoreishi and Shahrestani 2009)
Proteins	Peptides	Protein hydrolysates, peptide derivatives, lectin	Rice bran and meal, soybean meal, mushroom stalks	Antioxidant, dipeptidyl peptidase IV inhibitory for type 2 diabetes, anticancer	(Xu et al. 2011; Kannan et al. 2010; Hatanaka et al. 2012; Rayaprolu et al. 2013; Bandyopadhyay et al. 2008)
Unsaturated fatty acids	PUFA	Omega-3-fatty acids (DHA, EPA)	Oil seed cakes, fruits and vegetable seed oils, bran oil	Lowering LDL, foetal brain development	(Fromm et al. 2012a; Hanmoungjai et al. 2001; Ayala-Zavala et al. 2011; Joana Gil-Chávez et al. 2013
	Fatty alcohol	Policosanols	Wheat straw, germ, and bran	Lowering LDL and increasing HDL	(Joana Gil-Chávez et al. 2013)
Carotenoids	Xanthophylls (oxygenated)	Astaxanthin, zeaxanthin, lutein	Papaya, peaches, prunes and squash, lettuce wastes	Antioxidant, food colouring, photo-protectants	(Alzate et al. 2013; O'Connell et al. 2007; Stahl and Sies 2005)
	Carotenes (non-oxygenated)	α,β,γ carotenes, lycopene	Tomato by-products, carrot press cake, banana peel, mango peel, citrus peel	Pro-vitamin A activity, protective against cardiovascular, coronary heart diseases, and cancer	(Wijngaard et al. 2012)
Steroids	Sterols and stanols	β-sitosterol, campesterol, stigmasterol, stigmastanol, brassicasterol, ergosterol	Mushrooms, maize hull	LDL cholesterol lowering	(Moreau et al. 2000; Gil-Ramírez et al. 2013)
Vitamins		Ascorbic acid (Vit. C), tocopherols (Vit. E)	Pomegranate peel, wheat germ	Antioxidant	(Ayala-Zavala et al. 2011; Joana Gil-Chávez et al. 2013)
Pigments		Curcumin, chlorophyll, betalains	Turmeric skin, beet root skin	Antimicrobial	(Schieber et al. 2001; Loginova et al. 2011; Delgado-Pelayo et al. 2014)
Alkaloids		Glycoalkaloids: α-solanine, α-chaconine and aglycones: solanidine and demissidine	Potato peel	Dose dependent anti-cancerous and anti-inflammatory	(Hossain et al. 2014)
Phenolics	Simple phenols, Benzoquinones (C6)	Catechol, resorcinol	Argan oil	Cardiprotective	(Charrouf and Guillaume 2007)
	Hydroxybenzoic acid (C6-C1)	Gallic acid, protocatechuic acid, salicylic acid, vanillic acid, syringic acid	Citrus peel	Antioxidant	(Ma et al. 2009)
	Hydroxycinnamic acid (C6-C3), coumarins	Caffeic acid, ferrulic acid, coumaric acids, sinapic acid and their esters with quinic acid, e.g. chlorogenic acid	Potato peel, red beet peel, citrus peel, coffee by-products	Antioxidant	(Schieber et al. 2001; Bocco et al. 1998)
	Xanthonoids (C6-C1-C6)	Xanthones, e.g. Mangiferin	Mangosteen by-products	Antidiabetic, anti- HIV, anticancer, antioxidant activity and immunomodulatory	(Shan et al. 2011)
	Stillbenoids, anthraquinones (C6-C2-C6)	Resveratrol	Grape by-products	Antioxidant, anticoagulant	(Olas and Wachowicz 2005)
	Flavonoids, isoflavonoids, chalconoids (C6-C3-C6)	Flavonols, e.g. quercetin, myricetin, rutin (glycoside form) Flavones, e.g. apigenin Isoflavones, e.g. daidzein Flavanols, e.g. catechins, epicatechin Flavanones, e.g. naringenin, hesperetin, eriodictyol (aglycones)	Red onion waste, radish leaves, fennel leaves, apple pomace, white grape pomace and seed citrus by-products	Antioxidants	(Schieber et al. 2001; Giannuzzo et al. 2003; Wijngaard et al. 2012)
		Anthocyanidine (aglycones), Anthocyanine (glycosidic form), e.g. pelargonidin, cyanidin, peonidin, delphinidin, petunidin and malvidin	Red grape by-products, banana peel, winery by-products	Antioxidants	(Schieber et al. 2001)
		Chalcones, e.g. phloridzin	Apple pomace and seed	Antidiabetic	(Fromm et al. 2012b)
	Lignans (C6-C3)2 (phytoestrogens)	Pinoresinol, secoisolariciresinol	Flax seed, sesame seed, brassica veggies, brewer's spent grain	Antioxidant, anti-inflammatory	(Korkina et al. 2011)
	Lignin (C6-C3)n	Cross-linked with plant polysaccharides like hemicellulose, cellulose	Brewer's spent grain, flex hull	Dietary supplements	(Mussatto et al. 2007)
	Tannin/flavolans (C6-C3-C6)n (condensed form)	Proanthocyanidin (polyflavonoids) and gallotannins (polyflavanols)	Skins and seeds of exotic fruits, tea	Anti-nutritional and toxic factors	(Ayala-Zavala et al. 2011)

only a small number of studies have been reported which outlines bioactive peptides from agro by-products in comparison with better protein sources such as dairy-derived protein fractions (Li et al. 2013; Xu et al. 2011; Hatanaka et al. 2012; Rayaprolu et al. 2013; Bandyopadhyay et al. 2008). Kannan et al. (2010) isolated a novel penta-peptide (Glu-Gln-Arg-Pro-Arg) with a molecular mass of 684.37 Da from heat stabilised defatted rice bran. This peptide showed inhibition against proliferation of colon, breast, lung and liver cancer cells. Some high value proteins, in spite of low lysine content, from sunflower press cake have been reported because these proteins are low in anti-nutritional factors and devoid of toxic substances (González-Pérez and Vereijken 2007). In another study, pea protein isolate from beach pea was analysed for its nutritive and functional characteristics. The protein isolates were reported as ideal protein due to adequate percent ratios of essential to total amino acids (above 36%) with more than 80% in vitro digestibly (Chavan et al. 2001).

Polyunsaturated fatty acid (PUFA) such as docohexaenoic acid (DHA, C22:6) and eicosapentaenoic acid (EPA C20:5), which are of great importance for cardiovascular health because of their ability to lower the LDL cholesterol level (Fernandez and West 2005) and for foetal brain development (Swanson et al. 2012), are also present in appreciable quantities in many plant food processed by-products. Grape seed oil is rich in unsaturated fatty acids particularly alpha-linolenic acid, a precursor of EPA and DHA (Ayala-Zavala et al. 2011). Apple seeds are reported to be rich in highly unsaturated fatty oil, PUFA (Fromm et al. 2012a).

Carotenoids (tetraterpenoids/octa-isoprene molecules with or without oxygen) are fat-soluble organic pigments of biological importance due to their health-promoting activities like provitamin A activity, photo protective activity, preventing retinal degeneration and skin protectant and antioxidant activity (Stahl and Sies 2005). By-products from tomato processing industries have been exploited to recover lycopene and β-carotene having high antioxidant activity (Baysal et al. 2000). Citrus peel oil (limonene-a cyclic terpene) is a good source of essential oil and has antimicrobial activity (Bourgou et al. 2012).

Polyphenols comprise of diverse groups of phytochemicals ranging from benzoquinones to tannins (Table 1). These phenolic compounds are unevenly distributed in plant tissues, for instance flavonoids and phenolic acids in the outer layers of the skin, attractant anthocyanins in aleurone cells and deterrent tannins in seed coats and hulls. Polyphenols are present either in non-glycosylated form or as glycosides and/or associated with various organic acids and/or complex polymerised molecules with high molecular weights as in tannins (Kammerer et al. 2014). Several phenolic compounds have been identified in various agricultural by-products such as fruits and vegetables discards (Peschel et al. 2006), peanut hull (Francisco and Resurreccion 2009), rice bran (Pourali et al. 2010) and coffee by-products (Murthy and Naidu 2010). Generally, by-products, such as peel, seed, seed coats and hulls, contain higher phenolic compounds than the bulk edible parts (Balasundram et al. 2006; Ayala-Zavala et al. 2011). Some of the phenolic compounds possessing a C_3 side chain as in eugenol (phenylpropanoids) from clove are referred as essential oils and possess antimicrobial activities (Cowan 1999).

Emerging Technologies for the Extraction of Bioactives

Extraction of bioactive compounds from a variety of substrates is a determinant factor dictating the feasibility of utilisation of by-products. In view of the diversity of target compounds, plant species, their location and interactions in plant matrix, it is essential to adopt an appropriate extraction technique (Pinelo et al. 2006). The success of extraction is a function of the mass transfer action of diffusion and permeation phenomenon of solvent and solute (Huang et al. 2013). Prior to extraction, macroscopic pretreatment such as wet milling, thermal and/or vacuum concentration, mechanical pressing, freeze drying, centrifugation and microfiltration and molecule separation like alcohol precipitation, ultrafiltration, isoelectric solubilisation-precipitation, extrusions, have been reported to play important role in recovery of high-added value components from food wastes (Galanakis 2012).

Traditional extraction methods suffer from many disadvantages. These include health hazards of toxic organic solvents, requiring large volume of solvents, long extraction time and high temperatures. Hence, many improved novel extraction methodologies have emerged in recent years. The principle, mechanism and process control parameters of some of the important novel extraction technologies are summarised in Table 2. The mode of action of these novel techniques are generally through enhanced mass transfer rate, due to increased cell permeabilisation and solvent diffusivity, caused by rupturing or degradation of cell membrane.

With respect to extraction of valuable components from plant food by-products by far the greatest numbers of studies have examined UAE and PEF processes. The subsequent sections examine the detailed use of UAE and PEF for extraction of valuable compounds with health-promoting properties from agro-industrial processing by-products. Various other effective contemporary extraction techniques include MAE, PLE or ASE, SCF, EAE, HHP/HPE, EAE and DIC.

Briefly, in microwave-assisted extraction (MAE), electromagnetic energy (frequency ranges from 300 MHz to 300 GHz) is generated through magnetrons of different power and converted to thermal energy. The induced energy is further transferred to the biological material via dipole rotation and ionic conduction. This thermal energy heats up the

Table 2 Various novel extraction technologies: principle and operational parameters

Novel extraction techniques	Principle/mechanism	Control parameters	References
Ultrasound-assisted extraction (UAE)	Acoustic cavitation /cavitational dislodgement, micro-jetting and micro-streaming effects, disintegration of solid materials and disruption of cell walls	Frequency, amplitude, power, pressure, temperature, and viscosity of media	(Soria and Villamiel 2010; Vilkhu et al. 2008; Patist and Bates 2008)
Pulsed electric energy i. Pulsed electric field (PEF) ii. High voltage energy discharge (HVED) iii. Pulsed light (PL)	Electro-permeabilisation by electromechanical force, electroporation of cell membrane, increased mass transfer	Electric field intensity/input energy, pulse duration i.e. pulse width (μs) and number of pulses, pulse geometry	(Soliva-Fortuny et al. 2009; Knorr et al. 2001)
Microwave assisted extraction (MAE)	Conversion of electromagnetic waves into thermal energy, microwave heating without thermal gradient, evaporation of moisture creating high pressure on the cell wall leading to its and organelles' disruption	Magnetic field strength of magnetron, type of microwave device, microwave power, frequency and time, dielectric properties of sample and solvent, number of extraction cycles	(Routray and Orsat 2011; Kubrakova and Toropchenova 2008; Kadam et al. 2013)
Pressurised liquid extraction (PLE) or Accelerated solvent extraction (ASE) Subcritical water extraction (SWE)	Increased solubility and diffusion rate at elevated temperature (above boiling point) under pressurised condition, reduction in viscosity and surface tension of solvents, increased mass transfer	Temperature (in the range of 50–200 °C), Pressure (3.5–20 MPa), type of extraction solvent, temperature, static time, and number of cycles	(Wijngaard et al. 2012; Kadam et al. 2013)
Supercritical fluid extraction (SFE)	Increased density and reduced viscosity of extraction fluid at temperature and pressure above critical points, altered diffusivity, surface tension, heat capacity and thermal conductivity, increased penetration and mass transfer	Type of supercritical fluid (most commonly CO_2), used modifier (co-solvent), temperature, pressure, fluid flow rate and pressure control	(Sahena et al. 2009; Wijngaard et al. 2012)
High Hydrostatic pressure extraction (HHP/HPE)	Very high differential pressure, increased solvent inflow through micro channels, deprotonation of charged groups, disruption of salt bridges and hydrophobic bonds in cell membranes, cell wall disruption, increased permeability and decrease in resistance to diffusion, increase mass transfer	Magnitude of fluid pressure (mostly between 100 and 1000 MPa), operating time, extraction solvent, solid-to-liquid ratio, and pressure relief/decompression time	(Huang et al. 2013)
Enzyme assisted extraction (EAE)	Hydrolysis of cell wall materials and membranes, breaking down the linkages, catalysis of structural matrix, increasing cell wall permeability, higher extraction yields	Selection of enzymes, and its catalytic property, optimum treatment time, temperature and pH, enzyme concentration	(Puri et al. 2012)
Instant controlled pressure drop-assisted extraction (DIC)	Thermo-mechanical effects due to an abrupt pressure drop towards a vacuum in a short span, auto-vaporisation, expansion of matrix structure and breaking of the cell wall	Steam pressure, temperature, vacuum and processing time	(Ben Amor and Allaf 2009; Mounir et al. 2014)

moisture inside the cells and causes evaporation, producing a high pressure on the cell wall. The built up pressure inside the biomaterial modifies the physical properties of the tissues (cell wall and organelles disruption) improving the porosity of the biological matrix. This causes better penetration of extracting solvent through the matrix and improves yield of the desired compounds. The microwaves heat the matrix internally and externally without a thermal gradient, which is advantageous over solid–liquid extraction. Flórez et al. (2015) have reviewed the MAE in detail with regard to mechanism and its utilisation in the extraction of various plant bioactive molecules such as polysaccharides, lipids, proteins, phenolics and essential oils.

Another novel and important technique with improved extraction yield is pressurised liquid extraction (PLE) where a high pressure–temperature combination is used to enhance the extraction. It is also known as pressurised fluid/solvent extraction or accelerated fluid/solvent (ASE) extraction. PLE is referred as pressurised/subcritical water extraction (SWE) when water is used as solvent. The applied high pressure elevates the boiling point of the solvent, facilitating use of higher temperature for extraction without changing the original physical state of the solvent. This elevated temperature increases the solubility and reduces the viscosity and surface tension of the solvent and thus increases the mass transfer rate. Azmir et al. (2013) have recommended PLE for extraction of natural

bioactive products: isoflavones from soybean, terpenoids and sterols from tobacco, flavonoids from spinach and phenolic compounds from parsley flakes. Wijngaard et al. (2012) have summarised the application of PLE for efficient extraction of polyphenols from plant by-products such as procyanidins and anthocyanins from red grape pomace, flavonols from onion waste, phenolic acids from potato peels and polyphenols from apple pomace.

In regard to supercritical fluid extraction (SCF), Herrero et al. (2010) have extensively described its mechanism and application for extraction of various bioactives from plant by-products, i.e. caffeine from tea stalk, phytosterols from loquat seeds, polyphenols from pomegranate seeds and lycopenes from tomato waste.

Biologically assisted extraction or commonly known as enzyme-assisted extraction (EAE) is another novel and green extraction method in plant bioactives. The mechanism, benefits over chemical and physical extraction processes, and application of enzyme-assisted extraction (EAE) towards extraction of plant bioactives have been discussed and reviewed by Puri et al. (2012). The authors have enlisted a gamut of products obtained using EAE including oils and carotenoids, pectins, inulin, lignans, soluble fibres, phenolics, flavonoids and vanillin.

High hydrostatic pressure (HHP) or high pressure extraction (HPE) is another emerging technology for extraction purposes which works on pressure gradient principle. Very high pressure is applied in two stages: (i) below deformation limit that leads to extraction solvent inflow through micro channels of the cell wall and (ii) above deformation limit leading into cell deformation and cell wall damage causing increased permeability and decreased resistance for diffusion. At the final pressure relief stage, the pressure rapidly decreases causing cell expansion facilitating intracellular fluid outflow (Huang et al. 2013). Prasad et al. (2010) and Corrales et al. (2008) have showed the potential application of HPE in extraction of phenolic compounds from longan fruit pericarp tissues and grape by-products, respectively. The authors have demonstrated the higher total phenolic content and antioxidant activity of the HPE extracts compared to conventional extracts.

Instant controlled pressure drop (DIC) extraction technology is based on combined thermo-mechanical effects arising due to an abrupt pressure drop towards a vacuum after a short-time/high-temperature and pressure treatment of the sample. The abrupt pressure drop causes instantaneous cooling of the products and thereby preventing thermal-induced degradation. This leads to expansion of matrix structure and breaking of the cell wall, thus causing auto-vaporisation of volatile compounds and improved mass transfer of desired compounds (Mounir et al. 2014). Allaf et al. (2013) and Ben Amor and Allaf (2009) have reported the use of DIC as an effective pretreatment tool for enhanced extraction of anthocyanins from hibiscus flower and essential oil from orange peel, respectively.

Description and Applications of Ultrasound-Assisted Extraction

Ultrasound waves are high frequency (>20 kHz) sound waves beyond the threshold of human hearing. The basic principle of ultrasound-assisted extraction (UAE) is acoustic cavitation and micro-streaming. When high power ultrasound waves propagate through any medium, a sequence of compressions and rarefactions is induced in the molecules of the medium causing pressure alteration. The developed negative pressure during the rarefaction phase advances above tensile strength of the fluid causing the formation of cavitation bubbles from the gas nuclei of the medium. These bubbles grow over a number of cycles until they become unstable and finally violently collapse/implodes. This phenomenon of creation, expansion, and implosive collapse of bubbles in ultrasonicated medium is called acoustic cavitation phenomenon (Tiwari 2015). Figure 1 depicts the schematic representation of the acoustic cavitation mechanism. Since frequency is inversely proportional to the bubble size (Lorimer and Mason 1987), so in case of power ultrasound treatments, larger cavitation bubbles are formed. This implosion generates high temperature and pressure which in turn results into high sheer energy waves and turbulence causing combination of mechanical effect on the material (Soria and Villamiel 2010). It also develops strong micro-streaming currents that may alter the characteristic of the medium. These effects collectively cause disruption of cell wall resulting into greater diffusion and improved mass transfer rate causing better release of intracellular material. Thus, UAE can provide added benefit by increasing the extraction yield at lower temperatures and thus decreasing extraction time resulting in a better quality product (Mason et al. 1996). Additionally, commercial scale-up of UAE process is achievable with good return on capital investment (Patist and Bates 2008).

Two types of ultrasound equipment are most commonly used for extraction purposes, namely, ultrasonic water bath and ultrasonic probe system fitted with horn transducers. The factors affecting the efficiency of ultrasonic treatments can be divided into three categories: (1) process factors such as frequency, amplitude, power and treatment time; (2) product factors such as moisture content, particle size and compound of interest and (3) media factor such as pressure, temperature, viscosity and polarity of solvent (Wang and Weller 2006; Patist and Bates 2008). Ultrasonic intensity (UI in Watt/cm^2) is expressed as actual power output per surface area of probe, i.e. $4P/\pi D^2$, where P = ultrasonic power, i.e. amount of energy (Q = mCpΔT) consumed per unit time and D is diameter of probe (Tiwari et al. 2008).

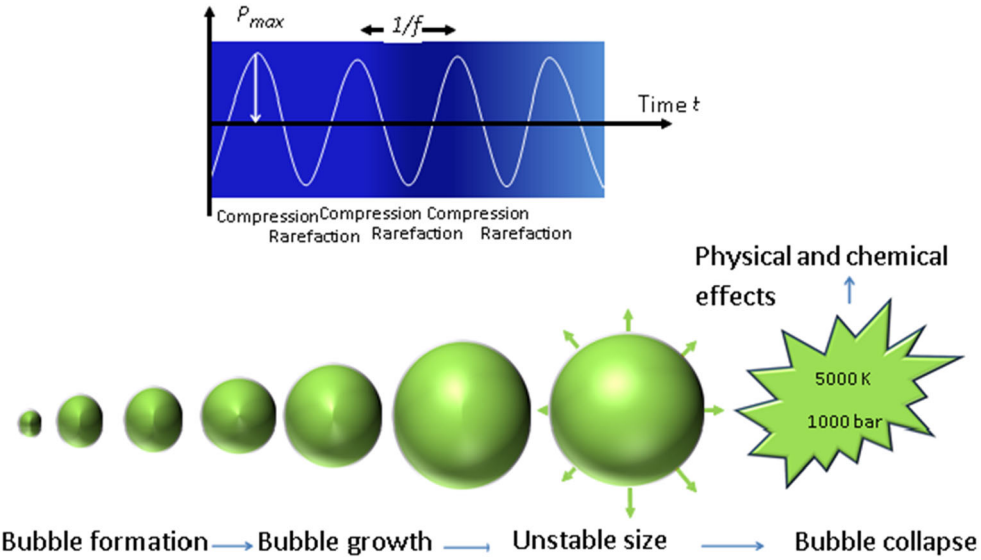

Fig. 1 Schematic representation of the acoustic cavitation mechanism

Optimal conditions (mostly response surface methodology (RSM)) for extraction of target bioactives from various agro-industrial by-products using UAE are outlined in Table 3. For example, Roselló-Soto et al. (2015) have shown that water extracts of olive kernel by-product yield approximately 1.5-fold to 2-fold higher proteins at 18 and 55 kJ/kg ultrasonic energy input, respectively. The extracts also had a higher level of carotenoids, chlorophylls (a and b), total phenolic content (TPC) and antioxidant capacity compared to untreated samples. Fu et al. (2006) have examined alkaline extraction of xyloglucan, a hemicellulose, from apple pomace using ultrasound-assisted extraction (operating power of 160 W) and found that the technique produced a comparable yield about three times faster than the traditional alkaline extraction method without UAE. The optimum ultrasonic extraction parameters (using RSM with central composite design (CCD)) were found to be liquid to solid ratio of 34.4:1 (*v*/*w*), 3.3 M potassium hydroxide concentration and an UAE time of 2.5 h. In another study, Minjares-Fuentes et al. (2014) observed 20% higher yields of pectins in UAE-pretreated (frequency 37 kHz, power 140 W and power density 0.05 W mL^{-1}) grape pomace compared to control (extracted at same extraction conditions with no ultrasonic treatment). The optimal extraction conditions (RSM with Box-Behnken design (BBD)) was found to be solid to solvent ratio 1:10 (*w*/*v*), temperature 75 °C, time 60 min and pH 2.0, using citric acid as solvent. In addition, pectins from UAE exhibited a higher average molecular weight. In contrast, Samaram et al. (2015) concluded that application of UAE could not increase the yield of papaya seed oil (rich in MUFA) compared to Soxhlet solvent extraction. They indicated that among different ultrasound extraction variables (namely time, temperature, power and solvent to sample ratio), the extraction time and extraction temperature were the most significant ($p < 0.05$) variables; however, the efficiency of oil extraction was increased at higher ultrasound power for longer time. Tian et al. (2013) observed that application of UAE at an elevated power (140 W) and temperature (40 °C) with high proportion of solvent to sample ratio (10 mL/g) resulted into higher oil recovery (25.11% yield) compared to conventional Soxhlet extraction (20.5% yield) from pomegranate seed. Yield % was calculated by using the formula (Wo/Ws × 100), where Wo is the weight of the extracted material (g) and Ws is the weight of the sample (g). Similarly, rice bran, a major by-product from rice milling industry, was used by Tabaraki and Nateghi (2011) to optimise the UAE parameters (RSM with CCD) for extraction of polyphenols and antioxidants. Maximum extraction yield of 19.83% (calculated by using the formula Wo/Ws × 100) was attained at 60 °C of UAE temperature, 87% ethanol concentration and 28 min of treatment time, whereas UAE temperature of 54 °C, extraction time of 40 min and 67% ethanol concentration resulted into optimal total phenols (6.21 mg GAE/gDw). The maximum antioxidant activity (FRAP) (54.14 μmol Fe^{2+}/gDw) and antiradical activity (DPPH) (52.83% inhibition) was achieved at 51 °C UAE temperature, 45 min treatment time and ethanol concentration of 65 and 67%, respectively. In another study, citrus peels were treated with ultrasound to obtain polyphenolic rich fractions (Ma et al. 2009). The authors observed that the yields of extracts, after UAE at 15 °C for 1 h, were significantly higher than those by conventional maceration treatment at 40 °C for 8 h. The UAE temperature above 40 °C for 20 min resulted into lower yields of phenolic acids. It was noted that caffeic, p-coumaric, ferulic and p-hydroxybenzoic acid

Table 3 Applications of ultrasound-assisted extraction (UAE) in extraction of bioactive compounds from agro-industrial by-products

Plant by-products	Bioactive compounds	UAE treatment conditions (E = Equipment, P = Power, F = Frequency, S = Solvent type, S/F = Solvent to feed ratio, t = Time, T = Temperature)	Yield (UAE versus SLE)	References
Litchi seeds	Polysaccharides	E: ultrasonic bath, P: 210 W, S: water, S/F: 15 mL/gDw, t: 45 min	3.39 ± 0.18 mg GE/gDw; SLE data not reported	(Chen et al. 2011)
Pomegranate seed	Seed oil	E: ultrasonic bath, P: 140 W, S: petroleum ether, S/F: 10 mL/gDw, t: 36 min, T: 40 °C	25.11 ± 0.08% (*w*/*w*) compared to 17.94% by SLE	(Tian et al. 2013; Eikani et al. 2012),
Pomegranate peel	i. Polyphenols ii. Polysaccharides	i. Polyphenol: E: ultrasonic bath, P: 140 W, F: 35 kHz, S: 70% aqueous ethanol, S/F: 50 mL/gDw, t: 30 min, T: 60 °C ii. Polysaccharide: E: ultrasonic bath, P: 148 W, F: 40 kHz, S: water, S/F: 24 mL/gDw, t: 63 min, T: 55 °C	Polyphenol: 45.4%. Polysaccharide: 13.658 ± 0.133% compared to 10.36% from SLE	(Tabaraki et al. 2012; Zhu et al. 2015b; Zhu and Liu 2013)
Rice bran	i. Polyphenols (TPC) ii. Flavonoid (TFC)	E: ultrasonic bath, P: 150 W, S: 50% ethanol, S/F: 10 mL/gDw, t: 60 min, T: 45 °C	i. TPC (mg GAE/gDw): 2.88 ± 0.14 compared to 2.71 ± 0.11 from SLE ii. TFC (mg QE/gDw): 1.56 ± 0.11 compared to 1.37 ± 0.13 from SLE	(Ghasemzadeh et al. 2015)
Sugar beet molasses	i. Polyphenols (TPC) ii. Anthocyanin (flavonoid)	i. Polyphenols: E: ultrasonic bath, P: 450 W, F: 35 kHz, S: 1.55 mol/L HCl, 57% ethanol (*v*/*v*), S/F: 30 mL/gDw, t: 73 min, T: 43 °C ii. Anthocyanin: E: ultrasonic bath, P: 450 W, F: 35 kHz, S: 1.72 mol/L HCl, 61% ethanol (*v*/*v*), S/F: 30 mL/gDw, t: 68 min, T: 41 °C	TPC: 17.39 ± 0.15 (mg GAE/gDw)Anthocyanin: 31.78 ± 0.56 (mg/100 g); SLE data not reported	(Chen et al. 2015b)
Wheat bran	Polyphenols (TPC)	E: ultrasonic bath, P: 250 W, F: 40 kHz, S: 64% aqueous ethanol, S/F: 20 mL/gDw, t: 25 min, T: 60 °C	TPC: 3.12 ± 0.03 mg GAE/gDw compared to 0.92 mg GAE/gDw by SLE	(Singh et al. 2012; Wang et al. 2008)
Papaya seed	Oil: predominantly MUFA	E: ultrasonic bath, P: 700 W, F: 40 kHz, S: n-Hexane, S/F: 7 mL/gDw, t: 38.5 min, T: 62.5 °C	Papaya seed oil: 23.3 ± 0.6%, compared to 30.4% by SLE	(Samaram et al. 2015)
Mushroom (*Agaricus bisporous*)	Polysaccharides	E: ultrasonic bath, P: 230 W, F: 40 kHz, S: water, S/F: 30 mL/gDw, t: 62 min, T: 70 °C	Yield: 6.02% (Mw- 158 kDa) compared to 2.36 ± 0.05% by SLE i.e. 155.08% increase	(Tian et al. 2012)
Grape seed	i. Polyphenols (TPC) and anthocyanins ii. Seed oil	i. TPC and Anthocyanins: E: ultrasonic probe, diameter: 13 mm, amplitude: 61.0 μm, P: 150 W, F: 20 kHz, S: methanol, S/F: 10 mL/gDw, t: 15 min, T: ambient (~23 °C) ii. Oil: E: ultrasonic probe, diameter: 13 mm, amplitude: 61.0 μm, P: 150 W, F: 20 kHz, S: n-hexane, S/F: 8 mL/gDw, t: 30 min, T: ambient (~23 °C)	i. TPC (mg GAE/gDw): 105.81 ± 3.21 compared to 89.45 ± 2.29 by conventional maceration; Anthocyanins (mg MAL/gDw): 2.20 ± 0.11 mg/mL, Maceration: 4.96 ± 0.21 ii. Oil: 14.08 ± 0.08% *w*/w compared to 14.64 ± 0.29 by SLE	(Da Porto et al. 2013)
Potato peel (Lady Rosetta var.)	Polyphenols	E: ultrasonic bath, P: 100 W, F: 33 kHz, S: 80% methanol, S/F: 10 mL/gDw, t: 900 min, T: ambient (~23 °C)	TPC (mg GAE/gDw): 7.67 ± 0.79 compared to 3.28 ± 0.07 by SLE	(Kumari et al. 2017)

Extraction yield% was calculated by the formula (Wo/Ws × 100), where Wo is the weight of the extracted material (g) and Ws is the weight of the sample (g)

SLE solid–liquid extraction, *gDw* gram dry weight, *GE* glucose equivalent, *MAL* malvidine, *GAE* gallic acid equivalent, *QE* quercetin equivalent

decreased significantly from 99.3 to 57.3, 168.9 to 136.6, 2224.5 to 1242.152 and 47.2 to 30.5 μg/gDw, respectively. Similarly, the TPC increased approximately by twofold when grape by-products were subject to ultrasonic extraction in ultrasonic bath of 35 kHz frequency at 70 °C for 1 h (Corrales et al. 2008). Application of UAE in extraction of anthocyanins from black chokeberry wastes with enhanced extraction rate was reported by Galván D'Alessandro et al. (2014)). The authors noted that ultrasound (100 W, 30.8 kHz)-assisted extraction of anthocyanins (3.15 mg cyanidin-3-glucoside equivalent per gram dry weight (CGE/gDw)) with water (1:40 solid to solvent ratio) at 20 °C temperature reduced the extraction time by threefold compared to unassisted extraction. However, a reduction of 20% extraction time was observed whilst extracting 9.58 mg CGE/gDw of anthocyanin at 70 °C in water. Various examples discussed above suggest that the UAE can be an effective technique in

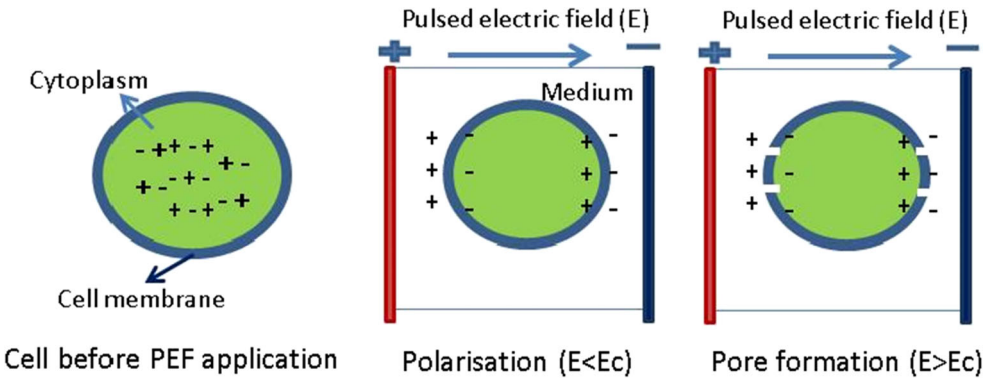

Fig. 2 Schematic representation of electroporation mechanism in biological cell membrane exposed to an electric field *E. Ec* critical electric field strength

valorisation of numerous agro-industrial by-products for a wide range of bioactive compounds.

Description and Applications of Pulsed Electric Field-Assisted Extraction

Pulsed electric field (PEF)-assisted extraction involves the application of short duration pulses (μs to ms) of moderate electric voltage (typically 0.5–20 kV/cm) to a substrate of choice placed between two electrodes. Using high electric voltage (5–50 kV/cm), the technology has been applied for preservation, enzyme and microbial inactivation purposes (Mohamed and Eissa 2012). Low to mild PEF treatment intensities are often considered an effective pretreatment method for enhancement of secondary metabolite extraction yields in cell cultures and plant systems (Balasa et al. 2011). The basic principle of PEF-assisted extraction is electroporation due to dielectric disruption of cell membrane (Zimmermann et al. 1974). It is assumed that cell membranes act like a capacitor with low dielectric constant having natural trans-membrane potential due to the presence of free charges of opposite polarities across the membrane. When the external electric field is applied, the trans-membrane potential is increased because of the accumulation of charges across the membrane. Subsequent exposure to electric field further increases the potential leading to electrostatic attraction between opposite charges across the membrane causing thinning of membrane. Breakdown of the membrane occurs if the critical breakdown voltage is reached by a further increase in the external field strength causing trans-membrane pore formation (Fig. 2). Permeabilisation can be reversible or irreversible depending on field strength, pulse duration and number of pulses. Breakdown is reversible if the pores are small in relation to the total membrane surface; however, reversible breakdown turns into irreversible breakdown if the size and number of pores become larger due to longer exposure time above critical field strengths. Cells remain viable in case of reversible electroporation whereas irreversible electroporation leads to mechanical destruction of the cell membrane and makes cells unviable. Efficiency of PEF treatment is often measured as cell disintegration index (Z) where '0' denotes intact cells and '1' indicates fully disintegrated cells (Knorr et al. 2001). Critical process factors in PEF application are electric field intensity, treatment time (t_{PEF} = number of pulses x pulse duration), pulse waveform (mainly exponential decaying, square wave, oscillatory, bipolar or instant reverse charges), conductivity, pH and ionic strength of the medium (Vega-Mercado et al. 1996), product geometry and size. Praporscic et al. (2005) demonstrated the effect of size and dimension of the sample on the efficiency of PEF treatment. They observed that the larger sized sugar beet slices (7 × 3 × 35 mm) yielded lowest juice (approx. 70%), whereas the highest yield (approx. 85%) was obtained from the smallest sized samples (1.5 × 1 × 35 mm).

PEF has been extensively investigated as a non-thermal food processing, food preservation and microbial inactivation technique. On the other hand, use of PEF in the recovery of bioactive compounds from by-products is less explored but is an expanding field of study. PEF technology offers a great opportunity in the area of improved extraction of intracellular valuable compounds (Ade-Omowaye et al. 2001). It helps in permeabilisation of cell membranes, thus increasing the diffusivity of the intracellular substances and improving extraction efficiency by increased mass transfer rate. Table 4 details the PEF-assisted extraction studies applied for extraction of various bioactive compounds from plant by-products.

For instance, Boussetta et al. (2014) investigated the influence of PEF treatment in the range of 10–20 kV/cm electric field intensity and 1–10 ms treatment time (t_{PEF}) for the recovery of total polyphenols from rehydrated flaxseed hulls. Although the maximum amount of total polyphenols (2.7 times higher than control) was achieved at 20 kV/cm field intensity and 10 ms t_{PEF}, 4 ms t_{PEF} was chosen (2.3 times higher at this condition) from an optimum energy consumption point of view. Following 20 min of SLE extraction after PEF pretreatment under these conditions (20 kV/cm, t_{PEF} 4 ms), a fourfold increase in total polyphenol content was observed in the PEF-treated samples compared to the untreated sample. However, they have shown

Table 4 Applications of pulsed electric field (PEF) for extraction of bioactive compounds from agro-industrial by-products

Plant by-products	Bioactive compounds	Optimum treatment conditions (EF = electric field intensity, P = pulse width, N = no. of pulse, F = frequency, S = solvent, S/F = solvent to feed ratio, E = energy, t = treatment time, T-temperature)	Yield (PEF versus SLE)	References
Grape skin	Anthocyanins	PEF pretreatment in batch chambers followed by SLE: EF: 3 kV/cm, N: 30, F: 2 Hz, t: 15 s, E: 10 kJ/kg, SLE: S: 50% ethanol, S/F: 4.5, T: 70 °C, t: 60 min	Anthocyanin (mg Cy-3-glu eq./ gDw): PEF:14.05 ± 1.528; SLE: 7.93 ± 0.189 TPC (μmol GAE/gDw): PEF: ~350; SLE: ~220	(Corrales et al. 2008)
Orange peel	Polyphenols, flavonoids (naringin and hesperin)	PEF pretreatment followed by pressurisation: EF: 7 kV/cm, P: 3 μs, N: 20, F: 1 Hz, t: 60 μs, E: 3.77 J/kg	Polyphenol extraction yield increased 159%, antioxidant activity by 192%, naringin from 1 to 3.1 mg and hesperin from 1.3 to 4.6 mg/100 g fresh weight orange peel	(Luengo et al. 2013)
Potato peel	Steroidal alkaloids	PEF pretreatment in batch chamber followed by SLE: EF: 0.75 kV/cm, P: 3 μs, N: 200, F: 10 Hz, t: 600 μs, E: 18.5 J/kg. SLE: S/F:5 *w/v*, S: methanol and t: 60 min at 1700 rpm	PEF: 1.85 mg/gDw; SLE: 0.93 mg/gDw	(Hossain et al. 2015)
Flaxseed hulls	Polyphenols	PEF pretreatment in batch process: rehydration of hulls for 40 min in water with 20% ethanol and 0.3 mol/L NaOH (solid to liquid ratio 1:25 *w/v*); EF: 20 kV/cm, F: 0.33 Hz, E: 300 kJ/kg, P: 10 μs, N: 400, t: 4 ms	TPC (mgGAE/100 gDw): PEF ~120; SLE: ~30	(Boussetta et al. 2014)
Corn silk	Polysaccharides	PEF treatment continuous flow: EF: 30 kV/cm, P: 2 μs, t: 6 μs, S/F: 50 *w/v*, S: deionised water, flow rate: 25 mL/min	Polysaccharide: PEF-7.31%; SLE: 5.46%	(Zhao et al. 2011)
Mushroom	Polysaccharides, polyphenols, and proteins	PEF treatment with square bipolar pulses and continuous flow: suspension of 9% *w*/w using Milli-Q water as solvent, T: 20 °C, constant flow rate: 5.6 mL/s, EF: 38.4 kV/cm, t: 272 μs, residence times: 2.6 min, and T: 85 °C	Polysaccharide: PEF-97.72%; SLE-55.83%Polyphenol: PEF-50.85%; SLE-25.17%Protein: PEF-48.92%; SLE-44.75%	(Xue and Farid 2015)
Maize germ and hull	Phytosterol, germ oil	PEF pretreatment in batch chamber followed by SLE: EF: 0.6 kV/cm, N: 120, E: 0.62 kJ/kg. SLE conditions: S/F: 40 dw/v, S: hexane and t: 60 min at 150 rpm	Phytosterol: PEF: 1039 mg/100 g oil; SLE: 785 mg/100 g oil. Germ oil yield: PEF:43.7%; SLE: 23.2%	(Guderjan et al. 2005)

Extraction yield% was calculated by the formula (Wo/Ws × 100), where Wo is the weight of the extracted material (gDw) and Ws is the weight of the sample (gDw)

SLE solid–liquid extraction, *gDw* gram dry weight, *TE* Trolox equivalent, *GAE* gallic acid equivalent

that lower electric field intensity treatment resulted in lower polyphenol recovery. Impact of low intensity PEF treatments (0.25–1 kV/cm, 5–500 pulses of 3 μs pulse width with frequency of 10 Hz) on glycoalkaloid recovery from potato peel was studied by Hossain et al. (2015). When PEF treatment with field strength of 0.75 kV/cm and 600 μs was applied, maximum total steroidal alkaloid (glycoalkaloids and aglycone alkaloids) yield was obtained, which was 99.9% higher than the untreated sample. The authors also demonstrated that levels of aglycone alkaloids, i.e. solanidine and demissidine, exhibited an increasing trend with increase in electric field strength up to 0.75 kV/cm except at 1 kV/cm at any given treatment time. The maximum yield of solanidine (1.35 mg/gDw) and demissidine (0.26 mg/gDw) was achieved at electric field strength of 0.75 kV/cm with 1500 μs treatment time, which was 130.86 and 56.37% higher than the control, respectively. Interestingly, the authors suggested that the glycoalkaloids are more susceptible to PEF-induced degradation than their aglycones.

In another study, Parniakov et al. (2014) concluded that PEF pretreatment at electric field strength of 13.33 kV/cm in a batch process for 2720 s with 400 pulses prior to aqueous extraction from papaya peel waste caused significant enhancement in yield of proteins, carbohydrates and phenolic compounds even at moderate temperature of 50 °C and neutral pH compared to untreated samples.

The majority of PEF-assisted extraction studies from by-products have targeted the polyphenolic compounds; nevertheless, extraction of polysaccharides, proteins, isothiocyanates, phytosterols, steroidal alkaloids, seed and germ oil, etc. have also been investigated (Roselló-Soto et al. 2015; Sarkis et al. 2015; Parniakov et al. 2015). In recent years, research interests have grown towards the application of PEF for recovery of value-added products from by-products. In general, most of the research works have demonstrated advantages of the application of moderate electric field pulse technology either as pretreatment step or as continuous extraction system in the area of phytochemical extraction from by-products.

Integrated Novel Extraction Technologies in Combination to UAE and PEF

The integration of UAE and PEF-assisted extraction techniques with each other or with other novel extraction techniques like MAE, SFE, EAE and DIC with the objective to improve process efficiency is an area of current research interest. Whilst most of the combination studies adopt a sequential approach, very little is known about their simultaneous/coupled application. Additionally, there is limited literature on the use of combined novel extraction technologies with respect to bioactive recovery from agro-industrial by-products. Some of the examples describing the combination studies with UAE and/or PEF for extraction of bioactive compounds from agro-industrial wastes are outlined in Table 5.

Zhu et al. (2015a) investigated the combination of PEF and ohmic heating (pulsed ohmic heating) as a pretreatment step prior to solid–liquid extraction of inulin from chicory roots. Results showed that higher ohmic heating (55 °C) combined with electroporation (800 V/cm) causes more damage to chicory tissues ($Z = 0.9$ after $t_{PEF} = 0.3$ s) compared to low temperature ohmic heating (30 °C) at the same PEF strength ($Z = 0.75$ and $t_{PEF} = 2$ s). Moreover, the solute diffusivity D, for the different PEF treatments at constant temperature (30 °C), was found to be nearly the same for same values of Z. The authors also reported that preheated tissues at 50 and 70 °C had very low levels of cell disintegration in absence of electroporation. This shows that conventional heating or PEF treatment without ohmic heating applied separately is less effective than the pulsed ohmic heating treatment in combination with PEF for chicory tissue permeabilisation. Likewise, Praporscic et al. (2005) studied the combined effect of PEF with ohmic acid for the extraction of juice from sugar beet cuts. They reported enhanced recovery of juice up to 87.5% when ohmic heating at 60 °C for 10 min was followed by PEF treatment (600 V/cm, 400 pulses of 100 μs width and t_{PEF} of 0.04 s) compared to 77% yield with only ohmic heating.

The application of PEF either as pretreatment prior to pressing or as combined treatment with pressing for extraction of juices from apple, sugar beet, carrot and spinach have been described by Vorobiev and Lebovka (2006). The authors noted increase in sugar beet juice yield with PEF pretreatment and combined PEF treatment with pressing (compressive pressure at 10 bars) by 62.3 and 82.4%, respectively, compared to pressing alone. Moreover, the qualities of the juice from PEF-treated sugar beet samples were better in terms of colour (less coloured), purity (no pectin contamination) and sugar concentration (high). Similar improvements were observed for carrot and spinach juice where total yield was improved to 38.3 and 60.6% with combined PEF treatment compared to 25.6 and 30% yield from conventional pressing process at 5 and 10 bars, respectively.

In another study, Pasquel Reátegui et al. (2014) investigated the use of supercritical CO_2 (SFE) at 15 MPa operating pressure and 40 °C coupled with an ultrasonic probe of 13 mm diameter (working power of 200 W) on the upper end of the supercritical fluid extraction cell. The combined treatment of SFE and ultrasonics enhanced the yield of phenolics by 30% from blackberry bagasse with a reduction in extraction time compared to uncoupled supercritical extraction. The authors also observed that using 10% water as a co-solvent at a working temperature of 60 °C resulted into the highest total phenolics (49.36 ± 0.27 mg GAE/gDw extract), ABTS antioxidant activity (154.98 ± 1.83 μmol TE/gDw extract) and anthocyanin recovery (6.594 ± 0.40 mg/gDw extract). The enhanced mass transfer with ultrasound coupling was attributed to structural changes of the substrate as observed by the scanning electron microscopy image analyser. It can be seen from the above-mentioned examples that the combined techniques can be more efficient than the techniques used in isolation. However, more such studies are required with different combinations to provide better understanding on the most effective combinations.

Some researchers have also examined the use of extraction techniques in sequence. For example, Liu et al. (2011) observed 21.39% increase in oil extraction efficiency from watermelon seeds when seeds were ultrasonicated at 547 W and 48 °C for 23 s prior to aqueous enzymatic extraction. Similarly, ultrasound-assisted-enzymatic extraction of arabinoxylan from wheat bran was investigated by Wang et al. (2014). The authors obtained an arabinoxylan yield of 3.12 ± 0.05 mg/gDw of destarched and deproteined wheat bran. Using the optimum raw material concentration, they

observed a 40% improvement in arabinoxylan yield for EAE with UAE in comparison to EAE alone.

Teh et al. (2015) used a PEF pretreatment in combination to UAE to extract polyphenols from defatted canola seed cake and optimised the process using RSM. The optimised conditions resulted in higher total phenolic content (as gallic acid equivalent (GAE)) and flavonoid content (as luteolin equivalent (LUE)) compared to conventional extraction methods. However, separate application of PEF or UAE alone was not conducted, making it difficult to judge the best possible novel technique either in isolation or in combination.

The effect of instant controlled pressure drop (DIC) combined with UAE for recovery of the antioxidant compounds hesperidin and naringin from orange peel was also evaluated by Allaf et al. (2013). The authors reported that the highest extraction yield of both naringin (~0.065 g/gDw) and hesperidin (~ 0.82 g/gDw) was recorded with DIC-pretreated samples combined with UAE using 80% ethanol as a solvent, and solid to solvent ratio of 1:20 at 40 °C for 60 min in ultrasonic bath operating at 25 kHz frequency with 150 W output power. The authors reported the order of extraction yield of naringin as well as hesperidin as DIC-UAE > DIC-SLE > UAE > SLE. They also noted that DIC pretreatment reduced the time to achieve 95% of the final extraction yield of naringin by an hour and a half compared to solid–liquid extraction and less than an hour with UAE.

In the above sections, the application of UAE and PEF along with their integration with each other or with other novel

Table 5 Applications of UAE and/or PEF in combination with other novel extraction technologies in bioactive recovery from by-products

Integrated novel extraction techniques	By-product materials	Interested compounds	Treatment conditions	Response	References
UAE (probe) coupled with MAE	Soybean germ	Germ oil	UAE/MAE Power: 50/100 W, 21 kHz frequency, 1:5 solid to solvent (Hexane) ratio, 45 °C, 1 h	Yield: 14.1% Dw compared to 10% with only MAE or 12.2% with UAE alone	(Cravotto et al. 2008)
SFE (CO_2) coupled with UAE (probe)	Blackberry bagasse	Total phenols and anthocyanins	15 MPa pressure at 40 °C and ultrasonic power of 200 W	Yield: 30% increase compared to uncoupled supercritical extraction	(Pasquel Reátegui et al. 2014)
PEF coupled with ohmic heating (POH)	Red grape pomace	Polyphenols	Field strength: 400 V/cm, t_{POH}: 5 s, t_{total} 20 s, 20–50 series of pulses (300 pulses of 100 μs width) with 1 s time interval, followed by diffusion with 30% ethanol in water at 50 °C for 1 h	Yield: 36% higher compared to untreated sample	(El Darra et al. 2013)
PEF pretreatment combined with UAE (bath)	Defatted canola seed cake	Polyphenols	Field strength: 1.1 kV/cm, 900 pulses of 20 μs width at 30 Hz frequency, 1:10 solid to solvent (10% ethanol) ratio and UAE with 200 W of ultrasonic power at 70 °C for 20 min	Total phenolic content: 2.6 g gallic acid equivalent (GAE)/100 g fresh weight	(Teh et al. 2015)
UAE (probe) pretreatment combined with SFE (CO_2)	Grape marc	Polyphenols	UAE power: 80 W at 20 kHz frequency for 4 min at 80 °C. SFE: 8 MPa pressure, at 40 °C with solvent flow rate of 6 kg/h CO_2 modified with 10% ethanol as co-solvent	TPC (mg GAE per 100 gDw) UAE alone: 2336 ± 10, SFE alone: 2736 ± 11, UAE + SFE: 3493 ± 61	(Da Porto et al. 2015)
DIC pretreatment combined with UAE (bath)	Orange peel	Phenolic compounds	UAE power: 150 W at 25 kHz frequency, solid to solvent (80% ethanol) ratio of 1:20 at 40 °C for 60 min in ultrasonic bath	Naringin (~ 0.065 g/gDw) and hesperidin (~ 0.82 g/g Dw)	(Allaf et al. 2013)
UAE (bath) pretreatment combined with EAE	Watermelon seed	Seed oil	UAE power: 547 W at 48 °C for 23 s followed by EAE with 2.63% Protex enzyme at 47.13 °C, pH 7.89 for 4.29 h with solid to solvent (water) ratio 1:4.35	Yield: 21.39% higher compared to non-ultrasonicated samples	(Liu et al. 2011)

Extraction yield% was calculated by the formula (Wo/Ws × 100), where Wo is the weight of the extracted material (g) and Ws is the weight of the sample (g)

extraction techniques with respect to by-product utilisation has been emphasised. However, studies in which direct comparison of UAE and PEF with respect to agro-industrial by-product valorisation are also of interest despite the fact that only a small number of such studies have been conducted. Notwithstanding this, Corrales et al. (2008) compared the effect of UAE (35 kHz, 70 °C, 1 min) and PEF pretreatment (3 kV/cm, 30 pulses, 10 kJ/kg, 70 °C, 15 s) with 1 h of diffusion in 50% ethanol at 1:4.5 solid to liquid ratio, on the extraction of total phenolics and anthocyanins from grape by-products. Although twofold increase in TPC was noted for both UAE and PEF-treated samples compared to untreated samples, no significant difference between UAE and PEF treatment was observed. On the other hand, PEF treatment was found to be more effective for anthocyanins extraction (1.8-fold increase); however, UAE treatment showed no significant increase. Additionally, PEF-treated samples showed 4.2-fold increase in antioxidant activity against only 1.6-fold increase for UAE samples when compared to untreated samples making PEF a better choice. In another study of UAE and PEF for extraction of protein and phenolic compounds from olive kernel (Roselló-Soto et al. 2015), 18 kJ/kg energy input for both UAE- and PEF-treated samples resulted into total phenolics of 100 mg GAE/L, whereas protein recovery was approximately 175 and 100 mg/L, respectively. Similarly, at highest energy input (109 kJ/kg), TPC content was 150 mg GAE/L for both UAE- and PEF-treated samples and protein recovery was ~250 and ~100 mg/L, respectively, showing UAE as more efficient extraction novel technique. Moreover, a previous work on protein and polyphenol extraction using UAE and PEF from vine shoot interestingly validated that PEF pretreatment (13.3 kV/cm, 0–1500 pulses, 10 μs pulse width, 0.5 Hz pulse frequency, 50 °C, 0–762 kJ/kg) was more efficient than UAE (ultrasonic probe of 14 mm diameter, 24 kHz, 400 W, 50 °C, 3 h diffusion, 0–3428 kJ/kg) (Rajha et al. 2014). A relative increase of 2.1 and 1.5 in total phenolics recovery was obtained at highest energy input of 762 and 3428 kJ/kg for PEF and UAE, respectively, demonstrating the much lower energy requirement by PEF treatment. Specifically, the content of individual phenolic compounds, i.e. kaempferol (0.156 mg/g), epicatechin (1.747 mg/g), resveratrol (0.032 mg/g) was higher in PEF-treated extracts compared to UAE extracts (kaempferol (0.097 mg/g), epicatechin (0.671 mg/g) and resveratrol (0.024 mg/g). Recently, Barba et al. (2015) demonstrated that DPPH antioxidant activity and total anthocyanins concentrations from fermented grape pomace were significantly higher with PEF treatment at cell disintegration index of $Z \geq 0.6$ compared to UAE. However, no significant difference was evident in case of TPC. They also reported that the energy consumption to achieve 1 mg of TPC or anthocyanin was always higher for UAE than PEF. From the above examples, it is apparent that both PEF and UAE has potential for intensified bioactive extraction; however, choosing between the two technology depends on various factors like type of substrate material, amount of energy input and quality of the extract.

Impact of UAE and PEF Extraction Techniques on Stability of Bioactives

The application of UAE and PEF at various stages of bioactive extraction from agro by-products is reviewed in the previous sections. The discussions were based primarily on the effect of these technologies on extraction efficiency and yield. There is however the potential of degradation effect of UAE and PEF treatments during extraction; a vast majority of the studies have mainly concentrated on the quantity of the compound rather than the type and structure of the new compounds formed by degradation (Rostagno et al. 2003; Chukwumah et al. 2009; Guderjan et al. 2005; Corrales et al. 2008). For example, Sun et al. (2011) observed that when dichloromethane was used as solvent whilst UAE extraction from mandarin peel, all-*trans*-β-carotene (a type of carotenoid) was absent compared to control (~3.8 μg/gDw) suggesting degradation of all-*trans*-β-carotene upon ultrasound application. However, authors did not report the type of degradation products formed during extraction. In contrast, the effect of novel processing techniques on the stability of health-promoting compounds in some food products (especially juices, puree and oils) have been studied and reviewed from a processing and preservation viewpoint. The most studied alterations in food product processing using ultrasound and PEF appear to be related to pigment compounds (anthocyanin, carotenoids), together with antioxidant activity, vitamin C content and fatty acid composition (Rawson et al. 2011; Pingret et al. 2013). Whilst very few fundamental studies elucidating the effect of UAE- and PEF-assisted extraction on the structure of the specific components from generic substrates have been conducted, the small number of studies examining the structural modification effect of ultrasound- and PEF-assisted extraction on the targeted compounds pertaining to agro by-products are outlined in Table 6. Furthermore, some studies using different model systems by employing ultrasound or PEF treatments on pure form of targeted bioactive compounds instead of food matrix have been conducted to examine the effect of sonication and PEF on the stability of bioactive compounds like carotenoids, phenolic acids and flavonoids.

The degradation of all-trans-lycopene via isomerisation during UAE extraction from red grape fruit has been reported by Xu and Pan (2013). The authors tentatively identified various isomers of all-trans-lycopene such as 9,13′-di-cis-, 9,13-di-cis-, 15-cis-, 13-cis- and 9-cis-lycopene isomers by HPLC-PAD. Ultrasonic extraction of oil from kiwi seed having nutritionally interesting fatty acid profile showed differences in the fatty acid composition compared to conventionally

Table 6 Effect of ultrasound and PEF-assisted extraction on targeted bioactive components of agro-industrial by-products

Novel technique/analytical method	Bioactive compound	By-product material	Effect on compound	References
UAE/GC	Fatty acid	Pomegranate seed	No evidence of degradation, increased recovery of punicic acid PUFA	(Tian et al. 2013)
UAE/UPLC-MS/MS	Glycoalkaloids	Potato peel	No degradation of steroidal alkaloids	(Hossain et al. 2014)
UAE/HPLC	Flavanone glycosides	Orange peel	No evidence of flavanone degradation	(Khan et al. 2010)
Ultrasonic treatment/FT-IR, GC-MS, and ^{13}C NMR	Beta-glucan depolymerisation	Mushroom/ Pseudoepicoccum cocos	No change in basic chemical structure except unfolding of polymer compound	(Chen et al. 2015a)
PEF/ HPLC-MS	(+)-catechin–acetaldehyde condensation	Wine ageing	No change in reaction products	(Zhao et al. 2013)
PEF and UAE/HPLC, LC-PDA-MS	Acetylated and non-acetylated anthocyanins	Grape skin	No evidence of degradation, increased stability of anthocyanins	(Corrales et al. 2008)
PEF/HPLC	Isoflavonoids (genistein and daidzein)	Soybean	Degradation after 1.8 kJ/kg energy input	(Guderjan et al. 2005)
UAE/HPLC-PDA	Inulin	Jerusalem artichoke tubers	Decreases the degree of polymerisation by UAE probe	(Lingyun et al. 2007)

GC gas chromatography, *HPLC* high-performance liquid chromatography, *UPLC* ultra-performance liquid chromatography, *MS* mass spectrometry, *MS/MS* tandem mass spectrometry, *FT-IR* Fourier transform infrared spectroscopy, *NMR* nuclear magnetic resonance spectroscopy, *PDA* photo diode array detector

extracted oil with the detection of marker lipid degradation compounds such as limonene (Z)-hept-2-enal and (2E, 4E)-deca-2,4-dienal (Cravotto et al. 2011). Zhao et al. (2006) examined the effect of ultrasonic treatment on the (all-E)-astaxanthin (a type of carotenoid) as a model compound and confirmed the degradation using HPLC analysis and UV/vis measurements into unidentified colourless compound. They also witnessed the detrimental effect of higher treatment time and ultrasonic power on the stability of the astaxanthin compound. Nevertheless, the study was conducted in model system rather than food matrix. Carail et al. (2015) studied the effect of high power ultrasound (20 kHz) on the stability of all-E-beta-carotene with respect to ultrasonic intensity, sonication time and temperature using a model system as well. The results showed the degradation effect in the order of sonication time > ultrasonic intensity > temperature. The authors also characterised the newly formed products using UPLC-MS/MS and tentatively identified four Z-isomers and seven β-apo-carotenals along with some unidentifiable oxygenated β-carotene derivatives. They also noted that degradation was more predominant in aqueous system compared to organic solvents. Another study investigating the effect of ultrasound treatment on seven phenolic acids in a model system revealed that the stability is not only dependent on type of phenolic acid but also on type of solvent and temperature of the system. Caffeic acid and sinapic acid were found to be more susceptible to degradation compared to protocatechuic acid, vanillic acid, p-hydroxybenzoic acid and p-coumaric acid. Ferulic was found to be the most stable. It was found that the degradation rate of caffeic acid was seven times faster at −5 °C compared to 25 °C. Analysis of the degradation product of caffeic acid and sinapic acid using HPLC-MS/MS and FT-IR indicated the occurrence of decarboxylation and polymerisation reactions leading to the presence of dimers; however, the structure of the degraded products was not investigated (Qiao et al. 2013). Luo et al. (2010) applied a PEF treatment on pure chitosan to conduct a study on physicochemical changes in chitosan structure. The result showed significant deformation in the chitosan granules with decrease in the molecular weight and reduction in crystallinity. Furthermore, the FT-IR spectra and UV absorption spectra confirmed that the carbonyl and carboxyl group bonds were weakened but no modification in chemical structure was observed.

It is evident from the above discussed facts that the nature of the stability of phytochemicals during ultrasound- or PEF-assisted extraction from plant matrices is poorly understood, although some studies on model system do exist. In particular, there is very little information on the type or structures of the newly degraded or regenerated products due to UAE or PEF treatments.

Conclusions and Future Trends

Agro-industrial by-products represent a potential source of natural food bioactives and the availability of economical and efficient extraction processes is a must to exploit the potential of low-cost by-products. Application of novel

extraction technologies (recommended as clean, green extraction technology) can be a good choice as these emerging techniques employ less solvent and can be used with GRAS solvents and development of industrial scale UAE equipment is possible. Applications of PEF-assisted extractions represent the potential to implement energy efficient processes to enhance the recovery of bioactive compounds. Notably, the application of moderate PEF requiring low energy inputs has been shown to improve the extractability of valuable bioactive compounds from different food matrices. However, if the potential of these novel techniques is to be fully exploited, a number of urgent knowledge gaps need to be filled. In particular, research should focus on optimising and standardising UAE and PEF conditions for each application for industrial uptake. To date, limited information in the area of the stability studies following UAE- and PEF-assisted extractions provides unique opportunity for further research on individual bioactive component. Use of mass spectrometry and NMR spectroscopy can be of great help in the stability research. More experimental studies on direct comparison of PEF against UAE extraction are necessary to establish the relative merits of these two techniques.

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