Chapter 1
Introduction to Membrane Separation of Bioactive Compounds; Challenges and Opportunities

Roberto Castro-Muñoz and Seid Mahdi Jafari

Abstract Today, membrane technologies are emerging techniques as efficient protocols in multiple types of separation, including chemical compounds, solvents, biomolecules, salts, ions, among others. So far, it is likely that standard membrane-based technologies driven by pressure, such as microfiltration (MF), ultrafiltration (UF) and nanofiltration (NF), have been mainly explored in the separation of biologically active compounds and food ingredients from natural products. More emergently, fractionation and concentration of bioactive compounds, such as phenolic compounds from agro-food wastes and by-products, can also be done via membrane technologies. At this point, such technologies have been fully involved within valorization and recycling protocols of various by-products. Thus, the aim of this chapter is to provide a comprehensive overview of the main agro-food by-products processed by membrane technologies for the recovery of phenolic compounds, their derivatives of different molecular weight and some other compounds. An introduction is provided in terms of separation processes, molecule properties, membrane features and other interesting phenomena that occur during their extraction. To finalize, the current challenges of membrane technologies in bioactive separation are elucidated.

Keywords Bioactives · Membrane technologies · Wastes · Natural products · Food ingredients · Fractionation

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Nomenclature

MF	Microfiltration
UF	Ultrafiltration
NF	Nanofiltration
MWCO	Molecular weight cut-off
SG	steviol glycosides

1 Introduction

The food industry faces many issues during the production of the crucial ingredients for the fabrication of food products (Gibson et al. 2004). This becomes more challenging when there is a need of satisfying the production of bioactive compounds involved in food products. According to the literature, a bioactive compound comprises any kind of chemical found in small quantities in plants, vegetables and specific natural foods (such as fruits, nuts, oils, grains, among others), which must display any actions in the body that may foster good health in the consumers (Panić et al. 2019), for example, antioxidant (such as polyphenols and carotenoids) and non-antioxidant (such as phytosterols) compounds and dietary fiber have proved a significant role in health (Saura-Calixto and Goni 2009). Thanks to the high amount of bioactive compounds in natural products, scientists and food technicians are continuously exploring several types of sources to extract such bioactives and some other food ingredients (Conidi et al. 2020). To some extent, it is quite possible that the current available natural products may satisfy the current nutritive requirements in the manufacture of food products (Burdock et al. 2006), however, the main issue concerns to the right and suitable extraction protocol towards such molecules. To date, several extraction techniques and methods have been evaluated including hot-water extraction (Rao et al. 2012), solvent extraction, irradiation-assisted extraction, adsorption (Cerón-Montes et al. 2015; Valencia-Arredondo et al. 2020), ultrasound-assisted extraction (Malićanin et al. 2014), enzyme-assisted extraction (Galiano et al. 2019), pulsed-electric field (Gachovska et al. 2010) and supercritical fluid extraction (Chemat et al. 2020; Barba et al. 2015). Most of these methods have not released enough positive results associated to relevant factors, e.g. specific bioactives, such as phenolic compounds, antioxidants carotenoids, to mention just a few of them, tend to be thermolabile that imply their degradation/denaturation related to their low stability at high temperatures, long extraction periods and the necessity of solvents in such mentioned methodologies (Cassano and Conidi 2019; Garcia-Castello et al. 2010; Castro-Muñoz et al. 2016a). Therefore, new protocols and techniques are being proposed by the research community; in which membrane technologies are pointed out as promising methods since they own multiple advantages over conventional and emerging extraction techniques, such as low energy demand, high separation efficiency, possibility of scale-up, simple operating parameters, high productivity (i.e., permeate fluxes), and the absence of phase transition

(Castro-Muñoz et al. 2021; Díaz-Montes and Castro-Muñoz 2019). Together with these advantages, the inherent properties of the membranes facilitate the separation of bioactive and food ingredients from natural systems. However, the application of membrane technologies does not only rely on molecule's extraction from natural sources, their role has also been directed towards the agro-food waste valorization (Castro-Muñoz et al. 2020a; Cassano et al. 2015a; Ochando-Pulido and Martínez-Férez 2017; Roselló-Soto et al. 2015). Food waste generation is a result of raisin global economic development, which collaborates to discard food and by-products that still contain nutritive agents and is most often related to the behavior of retailers, the foodservice sector, and consumers (Ong et al. 2018). Carmona-Cabello et al. (2018) highlighted the importance of new strategies in biorefineries based on food waste nutrients and their interactions to generate new sustainable feedstocks, as illustrated in Fig. 1.1.

In addition to this, it is well-known that the final waste disposal has become a major concern of food industries due its harmful impact on the environment. Until now, various methods have been potentially proposed attending such an issue, including dissolved air flotation, simple decantation, de-emulsification, coagulation and flocculation, enzymatic hydrolysis, fermentations, to mention just a few of them, in which their primary core is targeted to reduce the organic matter from aqueous waste stream (Maroušek et al. 2019; Ale et al. 2020) or to produce extensive valuable components (García-Depraect et al. 2021). At this point, membrane technologies, such as micro- (MF), ultra- (UF) and nano- (NF) filtration, have also assisted the treatment of food wastes (by-products, wastewaters, so on). As an outlook, it can be described that MF has been involved in macroscopic pre-treatment (MF), while UF and NF implementation has been mainly devoted to the selective separation, fractionation and concentration (Cassano and Conidi 2019; Cassano

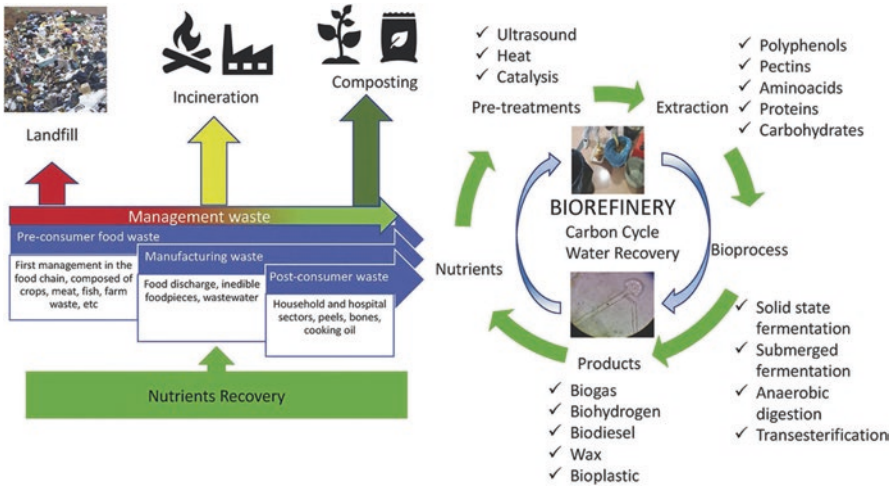

Fig. 1.1 General concept of food waste recovery based on its composition in biorefinery (Carmona-Cabello et al. 2018)

et al. 2015a; Galanakis 2015). Therefore, the main goal of this chapter is to release an overview of the bioactive compounds (mainly phenolic compounds) and some food ingredients that have been recovered from natural sources and food wastes by means of membrane-based technologies. This chapter also describes some basic principles of membrane technologies and their relevant phenomena appearing during the recovery of specific compounds, finalizing with the current challenges of membrane technologies in bioactive extraction.

2 Membrane Technologies: The Emerging Pathway for Recovering Bioactive Compounds

In principle, pressure-driven membrane technologies are generally differentiated by the membrane's molecular weight cut-off (MWCO), as represented in Fig. 1.2. Initially, the presence of a narrower pore size of the membrane will require a higher pressure demand. MF generally owns the larger pore size allowing to easily remove suspended particles, bacteria, and oil emulsion (Ochando-Pulido and Martínez-Férez 2017; Castro-Muñoz et al. 2015a), while UF is declared as one of the most efficient membrane technologies for the separation of proteins, sub-molecular organic groups, viruses, macromolecules (Russo et al. 2019; Castro-Muñoz and

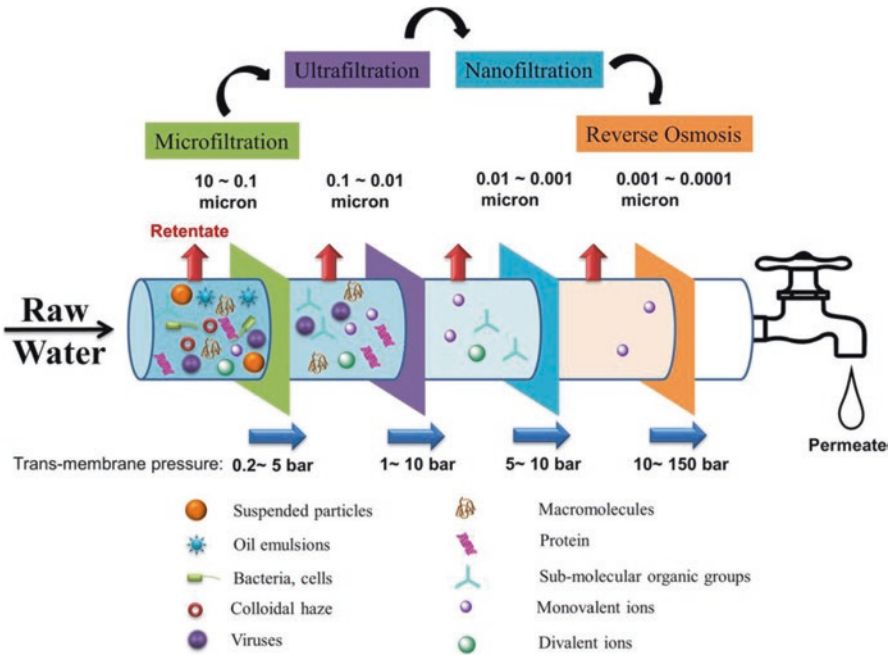

Fig. 1.2 Schematic drawing of pressure-driven membrane technologies and their role in separation (Liang et al. 2019)

Yañez-Fernandez 2015; Van Der Bruggen et al. 2003a). Evidently, the separation performance of UF becomes more efficient when membranes have tight pore sizes (Galanakis 2015; Cassano et al. 2018), it means, the UF membranes possess a MWCO in the range of 1–3 kDa, being able to effectively extract and thus concentrate low-molecular weight molecules (including anthocyanins, low molecular weight phenols, low molecular weight sugars, and peptides) (Castro-Muñoz et al. 2020a); these particular membranes are categorized in the molecular limit of the NF membranes.

Considered as the most selective technology for the fractionation and concentration of bioactives, NF technology (having pore size between 350–400 Da), together with tight UF membranes are the most recommended to achieve the recovery of low molecular weight polyphenols (Liang et al. 2019; Castro-Muñoz et al. 2019a). In the light of recovery bioactives from natural products, Table 1.1 summarizes the most extracted bioactives from natural sources using membrane technologies. It can be proved that these technologies exhibit acceptable recovery rates towards various bioactives; for instance, MF offers a rate from 47 up to ∼100% toward molecules with a molecular weight between 200 and 500 g/mol, such as anthocyanins, glutamine, isoproline, proline, betanin, isobetanin, sugars, and galacturonic acid and some phenolic compounds. Depending on the membrane's MWCO, UF can display rates between 44 and 99% of similar compounds, which have been mainly contained on the permeate side. It is important to note that some of these bioactives can be initiated to be retained by the membranes, and hence partially recovered on the retentate side. NF in turn collects mostly water on permeate side and concurrently concentrates bioactive molecules on the retentate side from 50 up to 99%. In addition to this, the application of membrane technologies has been extended to the extraction of specific food ingredients. This is the case of steviol glycosides (SGs) generally obtained from the *Stevia rebaudiana* plant (Žlabur et al. 2015). Such ingredients have recently increased their popularity since they can easily exceed the sweetening power of sucrose. Castro-Muñoz and co-workers (Castro-Muñoz et al. 2020b) have recently reviewed the current advanced in extracting SGs via membrane process. In principle, considering their molecular weight (oscillates about 318 g/mol) (Myint et al. 2020) and physiochemical properties, their extraction has been proposed using specific methods (such as solvent extraction, microwave-assisted extraction, supercritical fluid extraction, chromatographic techniques, etc) (Žlabur et al. 2015; Bursać Kovačević et al. 2018; Jaitak et al. 2009; Carbonell-Capella et al. 2017), however, their purification becomes challenging since they have exhibited specific bioactive properties (against diabetes mellitus, cancer, hypertension, gastroenteritis, cholesterol) (Ceunen and Geuns 2013), therefore, the research community has initiated to look for alternatives to preserve such bioactivity. By analyzing the literature data, Castro-Muñoz et al. stated that membranes can offer recovery yields from 25 to 80%. They also found out that the recovery rate strongly depends on various factors, such as operating parameters (transmembrane pressure, feed flow, temperature, etc.), intrinsic properties of the membranes and pre-treatment steps. However, the highest efficiencies were noticed using integrated membrane processes. For instance, Díaz-Montes et al. (2020a) demonstrated that a

Table 1.1 Bioactive compounds extracted from natural products via membrane processes

Bioactive compound	Natural source	Recovery rate	Technology	Membrane characteristics (MWCO/material/configuration)	Ref.
Antioxidant compounds	Orange juice	~98.4%	UF	15 kDa/PVDF/tubular	Cassano (2003)
	Lemon juice	~98.4%			
	Carrot juice	~98.4%			
Ascorbic acid	Kiwifruit juice	~99.5%	UF	15 kDa/PVDF/tubular	Cassano et al. (2004)
Aroma compounds (methyl butanoate, ethyl butanoate, methyl benzoate, ethyl benzoate, 3-hexen-1-ol, (E)-hexen-1-ol, 1-hexanol, 1-octen-3-ol)		N.R	UF	15 kDa/PVDF/tubular	
Betacyanins	Cactus pear juice	~63.8%	UF	10 kDa/PSF/hollow fiber	Cassano et al. (2007)
Betaxanthins		~93.5%			
Ascorbic acid		62.4%			
Citric acid		62.4%			
Glutamic acid		65.5%			
Pectins	Kiwifruit juice	60.0%	UF	30 kDa/cellulose/flat sheet	Cassano et al. (2008)
Folic acid		95.6%			
Citric acid		98.0%			
Ascorbic acid		96.4%			
Glutamic acid		~100.0%			
Total polyphenols		86.4%			
Total phenolics	Clementine mandarin juice	83.6%	UF	N.R/PSF/hollow fiber	Cassano et al. (2009)
		91.7%		N.R./PEEKWC/hollow fiber	

Flavonoids	Cactus pear juice	58.2%	UF	200 kDa/PVDF/flat sheet	Cassano et al. (2010)
Total phenolics		93.8%			
Ascorbic acid		72.4%			
Betacyanins		100.0%			
Betaxanthins		77.0%			
Proteins		100.0%			
Ascorbic acid	Pomegranate juice	69.1%	UF	10% rejection dextran 68,800 MW/PEEK/hollow fiber	Cassano et al. (2011a)
Total polyphenols (cathechin)		83.4%		10% rejection dextran 68,800 MW/PEEK/hollow fiber	
Malic acid		95.7%		10% rejection dextran 68,800 MW/PEEK/hollow fiber	
Citric acid		98.6%		10% rejection dextran 68,800 MW/PEEK/hollow fiber	
Betalains	Purple cactus pear juice	100.0%	UF	100 kDa/PSF/hollow fiber	Castro-Muñoz et al. (2015b)
Anthocyanins	Blue corn extract	~78.0%	UF	5 kDa/regenerated cellulose/ spiral wound	Cerón-Montes et al. (2015)
Hesperidin	Lemon juice	~65.8%	UF	N.R./PVDF/flat sheet	Chornomaz et al. (2013)

(continued)

Table 1.1 (continued)

Bioactive compound	Natural source	Recovery rate	Technology	Membrane characteristics (MWCO/material/configuration)	Ref.
Anthocyanins	Roselle extract	~100.0%	UF	5 kDa/PES/flat sheet	Cisse et al. (2011a)
		~90.0%	UF	1 kDa/composite polyamide/flat sheet	
		~90.0%		2 kDa/thin film/flat sheet	
		~80.0%		20 kDa/PES/flat sheet	
		~60.0%		50 kDa/PES/flat sheet	
		~90.0%	NF	N.R./composite/flat sheet	
		~100.0%		0.2–0.4 kDa/polyamide thin-film composite/flat sheet	
		~100.0%		N.R./PES/flat sheet	
		~100.0%		0.15–0.3 kDa/polyamide PSF thin-film/flat sheet	
		~100.0%		N.R./cross linked polyamide composite/flat sheet	
		~100.0%		N.R./composite/flat sheet	
Vitamin C	Roselle extract	~95.0%	MF	0.2 μm/ceramic/tubular	Cisse et al. (2011b)
Anthocyanins		~98.4%			

Total polyphenols	Bergamot juice	93.7%	UF	100 kDa/PSF/hollow fiber	Conidi et al. (2011)
Narirutin		99.2%	NF	450 Da/TiO_2/Monotubular	
Naringin		95.3%			
Hesperidin		91.7%			
Neohesperidin		96.3%			
Malic acid		99.2%	UF	100 kDa/PSF/hollow fiber	
Ascorbic acid		87.5%			
Citric acid		99.7%			
Total polyphenols	Pomegranate juice	92.0%	UF	150 kDa/cellulose triacetate/ hollow fiber	Conidi et al. (2017)
Cyanidin 3,5-O-diglucoside		90.1%			
Cyanidin 3-O-diglucoside		93.1%			
Delphinidin 3-O-glucoside		82.0%			
Pelargolidin 3,5-O-diglucoside		85.1%			
Anthocyanins		89.7%	NF	2 kDa/thin film composite/ flat sheet	
Polyphenols		86.0%			
Sucrose	Pineapple juice	74.2%	UF	100 kDa/PSF/flat sheet	de Carvalho et al. (2008)
Vitamin C	Passion fruit juice	~84.3%	MF	0.3 μm/polyamide/hollow fiber	De Oliveira (2012)
Galacturonic acid		~59.5%			
Chlorogenic acid, Cynarin, Apigenin-7-O-glucoside	Artichoke extract	>85.0%	NF	400 Da/PES/spiral wound	Cassano et al. (2015b)

(continued)

Table 1.1 (continued)

Bioactive compound	Natural source	Recovery rate	Technology	Membrane characteristics (MWCO/material/ configuration)	Ref.
Sinapic acid	Blood orange juice	94.2%	UF	100 kDa/PSF/hollow fiber	Destani et al. (2013)
p-Coumaric acid		89.9%			
Naringin		95.9%			
Hydroxybenzoic acid		95.0%			
Hesperidin		96.0%			
Ferulic acid		93.1%			
Epicatechin		95.6%			
Ellagic acid		94.7%			
Catechin hydrate		97. 0%			
Caffeic acid		94.9%			
Chlorogenic acid		99.6%			
Total phenolics	Castanea sativa leaves aqueous extract	92.1%	UF	5 kDa/modified PES/flat sheet	Díaz-Reinoso et al. (2011)
		82.5%		10 kDa/modified PES/flat sheet	
Soluble protein				5 kDa/modified PES/flat sheet	
		96.5%		5 kDa/modified PES/flat sheet	
		80.5%		10 kDa/modified PES/flat sheet	

Total phenols	Apple juice	45.8%	MF	0.45 μm/polyamide/flat sheet	Fuenmayor et al. (2014)
Malic acid		94.8%			
Fructose		76.8%			
Glucose		85.1%			
Sucrose		76.0%			
Ascorbic acid	Blood orange juice	90.7%	UF	15 kDa/PVDF/tubular	Galaverna et al. (2008)
Cyanidin-3-glucoside		97.7%			
Cyanidin-3-glucoside-6″-malonyl		97.1%			
Total anthocyanins		97.6%			
Sinapic acid		~100.0%			
Caffeic acid		~100.0%			
Ferulic acid		~100.0%			
p-Coumaric acid		~100.0%			
Narirutin		~100.0%			
Hesperidin		~100.0%			
Total polyphenols	Pineapple juice	92.8%	MF	0.1 μm/PSF/hollow fiber	Laorko et al. (2010)
		99.6%		0.2 μm/PSF/hollow fiber	
		75.0%	UF	30 kDa/PSF/hollow fiber	
		91.9%		100 kDa/PSF/hollow fiber	
Polyphenols	Propolis extract	92.8%	NF	98% rejection mg SO_4/ polyamide-polysulphone/ spiral module	Mello et al. (2010)
Flavonoids		~100.0%			

(continued)

Table 1.1 (continued)

Bioactive compound	Natural source	Recovery rate	Technology	Membrane characteristics (MWCO/material/configuration)	Ref.
Polyphenols	Blood orange juice	76.6%	UF	50 kDa/PSF/hollow fiber	Mondal et al. (2016)
		83.2%		100 kDa/PSF/hollow fiber	
		76.4%		50 kDa/polyacrylonitrile hollow fiber	
Anthocyanins		90.7%		50 kDa/PSF/hollow fiber	
		92.6%		100 kDa/PSF/hollow fiber	
		94.2%		50 kDa/polyacrylonitrile/hollow fiber	
Histidine	Cactus pear	100.0%	MF	0.2 μm/ceramic/N.R.	Moßhammer et al. (2006)
Glutamine		100.0%			
Isoproline		100.0%			
Proline		100.0%			
Betanin		100.0%			
Isobetanin		100.0%			
Gallic acid	Mate aqueous extract	75.8%	NF	150–300 Da/thin film/spiral-wound	Negrão Murakami et al. (2011)
3,4-Dihydroxybenzoic acid		94.7%			
Chlorogenic acid		89.5%			
4,5-Dicaffeoylquinic acid		~100.0%			

Chlorogenic acid	Apple juice	97.7%	UF	100 kDa/PES/cassette	Onsekizoglu et al. (2010)
Epicatechin		98.5%			
Phloridzin		99.3%			
Citric acid		88.9%			
Galacturonic acid		94.3%			
Malic acid		100.0%			
Quinic acid		96.5%			
Succinic acid		86.2%			
Fumaric acid		90.0%			
Trans-2-hexenal		79.4%			
Gallic acid	Pomegranate juice	87.4%	UF	30 kDa/PVDF/cassette	Onsekizoglu (2013)
Ellagic acid		84.6%			
Catechin		71.1%			
Chlorogenic acid		87.7%			
Caffeic acid		59.8%			
Citric acid		100.0%			
Malic acid		100.0%			
Quinic acid		92.0%			
Oxalic acid		99.6%			
Anthocyanins	Blueberry juice	47.6%	MF	0.45 μm/ZrO_2-TiO_2/tubular	Pizzolato et al. (2012)

(continued)

Table 1.1 (continued)

Bioactive compound	Natural source	Recovery rate	Technology	Membrane characteristics (MWCO/material/configuration)	Ref.
Total phenols	Blood orange juice	99.3%	UF	100 kDa/PSF/hollow fiber	Quist-Jensen et al. (2016)
Naringin		96.2%			
Hesperidin		96.8%			
Narirutin		95.8%			
Glycoside	Stevia extract	97.1%	MF	0.05 μm/Al_2O_3-TiO_2/tubular	Reis et al. (2009)
		93.7%		0.1 μm/Al_2O_3-TiO_2/tubular	
		94.7%		0.2 μm/Al_2O_3-TiO_2/tubular	
Phenols	Sideritis extract	83.7%	NF	500 Da/modified polyimide/flat sheet	Tylkowski et al. (2011)
		98.3%		400 Da/polyimide/flat sheet	
		99.2%		300 Da/modified polyimide/flat sheet	
Flavonoids		96.8%		500 Da/modified polyimide/flat sheet	
		98.8%		400 Da/polyimide/flat sheet	
		99.0%		300 Da/modified polyimide/flat sheet	
Anthocyanins	Raspberry	83.0%	MF	0.2 μm/ceramic/hollow fiber	Vladisavljević et al. (2013)
		56.9%	UF	30 kDa/PSF/hollow fiber	
Betalains	Xoconostle fruit juice	> 90.0%	UF	100 kDa/PSF/hollow fiber	Castro-Muñoz et al. (2018a)
Total polyphenols		> 90.0%			

Total polyphenols	Pomegranate juice	74.9%	UF	2500 kg Mol^{-1}/PSF/hollow fiber	Cassano et al. (2015c)
		67.3%		2500 kg Mol^{-1}/PEEKWC/ hollow fiber	
Flavonoids		75.8%		2500 kg Mol^{-1}/PSF/hollow fiber	
		66.5%		2500 kg Mol^{-1}/PEEKWC/ hollow fiber	
Total polyphenols	Pomegranate juice	73.5%	UF	15 kDa/stainless/tubular	Baklouti et al. (2012)
Total polyphenols	Strawberry juice	97%	NF	150–300 Da/PVDF/tubular	Arend et al. (2017)
Anthocyanins		99%			
Total polyphenols	Apple juice	48.2%	UF	100 kDa/PSF/flat sheet	Domingues et al. (2014)
		44.3%	UF	50 kDa/PES/flat sheet	Gulec et al. (2017)
		63.2%		30 kDa/regenerated cellulose/flat sheet	
Total polyphenols	Apple juice	59.2%	UF	100 kDa/modified PSF/flat sheet	Gulec et al. (2018)

(continued)

Table 1.1 (continued)

Bioactive compound	Natural source	Recovery rate	Technology	Membrane characteristics (MWCO/material/ configuration)	Ref.
Cyanindin-3-glycoside	Jussara fruit juice	93.6%	NF	180 Da/polyamide thin film composite/flat sheet	Vieira et al. (2018)
		97.8%		340 Da/polyamide thin film composite/flat sheet	
		89.9%		150–300 Da/polyamide thin film composite/flat sheet	
		78.8%		150–300 Da/polyamide thin film composite/flat sheet	
		79.9%		400 Da/PES/flat sheet	
		50.9%		1000 Da/PES/flat sheet	

two-step UF processes (implying 100/1 kDa membranes) was effective enough to extract rebaudioside A, as depicted in Fig. 1.3.

More specifically, the authors reported that the UF100 membrane unit worked mostly to remove total solids (ca. 42%) and carbohydrates (ca. 41%) from the crude aqueous extract, while tight UF1 membrane unit recovered about 93% of the initial rebaudioside A.

In the light of integrated membrane processes, Valencia-Arredondo et al. (2020) implemented, for the first time, an integrated membrane-adsorption protocol for anthocyanin extraction from red cabbage. In a first approach, the acidified extract, containing 32 mg cyanidin-3-glucoside per milliliter (mg ECyn-3-glu•L^{-1}), was processed using membrane technologies, such as MF and UF followed by adsorption processes, producing an enriched anthocyanin concentrate with 3221 mg ECyn-3-glu•L^{-1}. Secondly, the pigments were completely fractionated by molecular exclusion chromatography, reverse-phase vacuum liquid chromatography and semi-preparative chromatography, purifying di-acylated cyanidin. The latter is recognized among the most valuable water-soluble pigments. Its importance comprises as food additive for use in manufacturing purple-colored jam, confectionaries, and

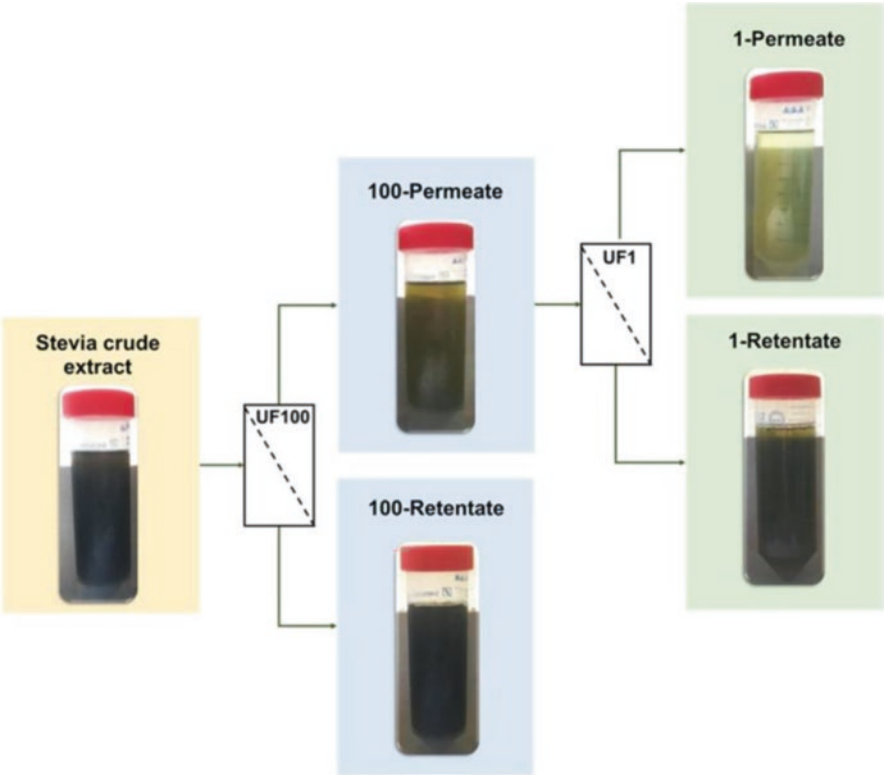

Fig. 1.3 Integrated ultrafiltration process implemented for the aqueous extraction of SGs (Díaz-Montes et al. 2020a)

beverages. It is important to mention that acylated anthocyanins are used in the food industry since they display high stability over nonacylated anthocyanins (Khoo et al. 2017). This approach opened a new window of exploration since more efficient integration processes are needed to satisfy the current demand for natural ingredients and colorants for food formulations (Carunchia et al. 2015). With the aim of extracting another food additive (like dextran) from complex aqueous systems, Díaz-Montes et al. (Díaz-Montes et al. 2020b) developed an integrated microdiafiltration-MF process for dextran extraction from fermentation broth. The usage of dextran regards as stabilizer and moisturizer (Heinze et al. 2006). Its synthesis is primarily performed via facultative bacteria *Leuconostoc mesenteroides*, cultivated in a sucrose enriched medium (Aman et al. 2012). The typical production of dextran implies separation and extraction stages, such as precipitation with polar solvents (e.g., ethanol and methanol) due to its insoluble properties in such alcohols (Vettori et al. 2011). At this point, Díaz-Montes et al. pointed out this integrated membrane approach can simultaneously extract and recover of dextran from the fermentation broth. Additionally, the authors compared the microfiltration-mediated extraction with a conventional solvent extraction protocol in terms of product yield, and physicochemical properties of the dextran. Interestingly, a successful extraction with a final yield (∼22%) was acquired using the membrane stages and resulted in less ethanol use for the final dextran precipitation, saving about 75% ethanol compared with normal ethanol precipitation.

Apart from recovery of bioactive compounds and food ingredients from natural sources, membrane technologies are contributing to the treatment of the primary by-products and wastes from food industries. Initially, these technologies were implemented for the organic matter elimination from aqueous streams, nevertheless, such a role has shifted to the valorization of agricultural waste enriched in a wide amount of bioactive molecules (Castro et al. 2018). For instance, Table 1.2 enlists the main bioactive compounds reclaimed from food wastes using membrane processes. Hydroxytyrosol, protocatechuic acid, caffeic acid, tyrosol and p-cumaric acid are among the most recovered bioactives, while olive mill wastewaters (OMW) have been found as the most investigated food waste over the last 20 years (Rahmanian et al. 2014; Conidi et al. 2014a; Galanakis et al. 2016; Cassano et al. 2016a). Russo (2007) was most probably the pioneering scientist proposing a membrane steps for the fractionation phenolic compounds from raw OMW extracts. Herein, MF and UF processes were used and produced permeates containing phenolic fractions, such as hydroxytyrosol (134,879–266,679 ppm), tyrosol (7968–11,218 ppm,) oleuropein (7765–26,698 ppm), caffeic acid (10,570–21,982 ppm) and protocatechuic acid (8871–22,601 ppm), among others. Since olive and its derived products are fundamental part of the Mediterranean diet (Bendini et al. 2007), it is obvious that by-products derived from olive processing are among the most produced wastes but also relevant source of nutraceutical molecules. The author also utilized NF and Reverse Osmosis (RO) unit operations for the fractionation and concentration of the phenolics, respectively (Russo 2007).

Unfortunately, most of the researches have concluded that the fouling phenomenon is a critical parameter when dealing with the long-term operation and stable

Table 1.2 Bioactive compounds extracted from food wastes and by-products via membrane processes

Recovered bioactive	Agro-food waste	Membrane processes	MWCO/material/configuration	Ref.
Phenolic compounds	Olive mill wastewaters	UF	30 kDa/Polyethersulfone/flat sheet	Garcia-Ivars et al. (2015)
	Winery effluents	MF	0.5 μm/PVDF/flat sheet	Giacobbo et al. (2015)
	Winery effluents	MF	0.2 μm/PVDF/hollow fiber	Giacobbo et al. (2017)
	Orange press liquor	UF	100 kDa/Polysulphone/hollow fiber	Ruby Figueroa et al. (2011) and Ruby-Figueroa et al. (2012)
Phenolic compounds	Nixtamalization wastewaters	Integrated membrane process:		Castro-Muñoz and Yañez-Fernandez (2015) and Castro-Muñoz et al. (2016b)
		MF	0.2 μm/Polysulfone/hollow fiber	
		UF	100 kDa/Polysulfone/hollow fiber	
		UF	1 kDa/Polysulfone/hollow fiber	
Phenolic compounds	Olive mill wastewaters	NF	200 Da/polymeric/spiral wound	Paraskeva et al. (2007)
Phenolic compounds	Grape seeds	UF	0.22 μm/cellulose acetate/flat sheet	Nawaz et al. (2006)
Phenolic compounds	Fermented grape pomace	UF	1000 Da/thin-film/spiral wound	Díaz-Reinoso et al. (2009) and Díaz-Reinoso et al. (2010)
		UF	1000 Da/ceramic (titania)/tubular	
		NF	250 Da/polyamide-polysulfone/spiral wound	
		NF	350 Da/polyamide-polysulfone/spiral wound	
		NF	150–300 Da/thin-film/spiral wound	

(continued)

Table 1.2 (continued)

Recovered bioactive	Agro-food waste	Membrane processes	MWCO/material/configuration	Ref.
Hydroxytyrosol, protocatechuic acid, caffeic acid, tyrosol, p-cumaric acid	Olive mill wastewaters	MF	0.2 μm/polypropylene/tubular	Cassano et al. (2011b)
		UF	4 kDa/polyethersulphone/flat sheet	
			5 kDa/regenerated cellulose/flat sheet	
			10 kDa/regenerated cellulose/flat sheet	
			10 kDa/Polyethersulphone/flat sheet	
Hydroxycinnamic acids, o-diphenols	Winery sludge from red grapes	UF	100 kDa/Polysulfone/flat sheet	Galanakis et al. (2013)
			20 kDa/Polysulfone/flat sheet	
			1 kDa/composite fluoropolymer/flat sheet	
3,4-DHPEA, p-HPEA, 3,4-DHPEA-EDA, verbascoside, and total phenols	Olive mill wastewater	Integrated membrane process:		Servili et al. (2011)
		MF	0.3 μm/polypropylene/tubular	
		UF	7 kDa/polyamide-polysulfone/spiral wound	
p-cumaric	Olive mill wastewaters	Integrated membrane process:		Conidi et al. (2014a)
		MF	0.2 μm/Polyvinylidenefluoride/flat sheet	
		UF	30 kDa/Polysulphone/hollow fiber	

Chlorogenic acid, Cynarin, Apigenin-7-O-glucoside	Artichoke wastewaters	Integrated membrane process:		Conidi et al. (2014b)
		UF	50 kDa/Polysulfone/hollow fiber	
		NF	400 Da/Polyethersulfone/spiral wound	
		NF	150–300 Da/polyamide/spiral wound	
	Artichoke wastewaters	NF	400 Da/Polyethersulphone/spiral wound	Cassano et al. (2015a)
Gallic acid, chlorogenic acid and epigallocatechin gallate	Residues from mate tree	NF	150–300 Da/thin-film/spiral wound	Prudêncio et al. (2012)
Free low MW polyphenols, hydroxytyrosol, procatechuic acid, tyrosol, oleuropein, tyrosol, caffeic acid	Olive mill wastewaters	UF	1 kDa/Polyethersulphone/spiral wound	Russo (2007)
Proanthocyanidins	Defatted milled grape seeds	UF	200 kDa/Polyvinylidenefluoride/tubular	Santamaría et al. (2002)
Hydroxytyrosol, procatechin acid, catechol, tyrosol, caffeic acid, p-cumaric acid and rutin.	Olive mill wastewaters	Integrated membrane process:		Cassano et al. (2013)
		UF	0.02 μm/Polyvinylidenefluoride/hollow fiber	
		UF	1 kDa/composite fluoropolymer/flat sheet	
		NF	Salt rejection >97%/thin-film/spiral wound	

(continued)

Table 1.2 (continued)

Recovered bioactive	Agro-food waste	Membrane processes	MWCO/material/configuration	Ref.
Isoflavones (aglycone and glucoside)	Soy processing waste	UF	1 kDa/regenerated cellulose/spiral wound	Xu et al. (2004)
Hydroxytyrosol, procatechin acid, tyrosol, caffeic acid, p-cumaric acid, oleuropein and some other low MW polyphenols.	Olive mill wastewaters	Integrated membrane process:		Garcia-Castello et al. (2010)
		UF	200 nm/Al_2O_3/tubular	
		NF	578 Da/Polyethersulphone/spiral wound	
Hydroxycinnamic acids and flavonols.	Olive mill wastewaters	UF	100 kDa/Polysulfone/spiral wound	Galanakis et al. (2010)
		UF	25 kDa/Polysulfone/spiral wound	
		UF	10 kDa/Polyethersulfone/spiral wound	
		UF	2 kDa/Polyethersulfone/spiral wound	
		NF	120 Da/Polypiperazine/spiral wound	
Anthocyanins, flavonoids	Orange press liquor	NF	180 Da/polyamide-polysulfone/spiral wound	Conidi et al. (2012)
		NF	300 Da/Polypiperazine amide thin-film composite/spiral wound	
		NF	400 Da/Polyethersulfone/spiral wound	
		NF	1000 Da/Polyethersulfone/spiral wound	
Anthocyanins (cyanidin-3-glucoside chloride, myrtillin chloride and peonidin-3-glucoside chloride), flavanones	Orange press liquor	NF	Na_2SO_4 rejection >25–50%/Polyethersulfone/spiral wound	Cassano et al. (2014)

Chlorogenic acid, Apigenin-7-O-glucoside	Artichoke wastewaters	NF	200–300 Da/polyamide/spiral wound	Conidi et al. (2015)
Oligosaccharides	Enzymatic by-product	NF	1000 Da/polyamide/spiral wound	Córdova et al. (2016)
		NF	400 Da/Polyethersulfone/spiral wound	
		NF	1000 Da/Polyethersulfone/spiral wound	
Carbohydrates	Nixtamalization wastewaters	UF	100 kDa/Polysulfone/hollow fiber	Castro-Muñoz et al. (2015c)
Oligosaccharides	Artichoke extract	Integrated membrane process:		Machado et al. (2016)
		MF	0.20 μm/Polyvinylidenefluoride/flat sheet	
		NF	150–300 Da/polyamide/tubular	
	Grape marc	Integrated membrane process:		Zagklis and Paraskeva (2015)
		UF	Pore size 100 nm/ceramic (zirconia)/tubular	
		NF	470 Da/polyamide/spiral wound	
Catechol, hydroxytyrosol, tyrosol, caffeic acid, and vanillic acid	Grape marc	Integrated membrane process:		Bazzarelli et al. (2016)
		MF	Pore size 140 nm/TiO_2/tubular	
		NF	$MgSO_4$ rejection 96%/cross-linked polyimide/spiral wound	

performance of the processes. Therefore, the concept of integrated membrane processes has also been used in these applications to reduced membrane fouling. By prepending UF and NF membranes. Cassano et al. (2013) fractionated OMWs obtaining a concentrated fraction enriched with phenolic substances (ca. 960 mg L^{-1}), which was suggested for food, cosmetic and pharmaceutical applications according to the presence of hydroxytyrosol, tyrosol, caffeic acid, p-cumaric acid, catechol and protocatechuic acid. In this work, the authors proposed a narrow pore size membrane, which in contribution with the nature of the phenolics, reached an excellent recovery. It is documented that phenolic compounds possess aromatic rings and aliphatic chains producing a hydrophobic profile increasing their volume, while concurrently attract water molecules allowing the volume increase of the polyphenols, and thus restricting their permeation due to the "polarity resistance" phenomenon (Galanakis 2015).

The winemaking is another food processing sector that produces large quantities of wastes, including grape seeds, fermented grape pomaces, lees and liquors. Díaz-Reinoso and co-workers (Díaz-Reinoso et al. 2009) recovered antioxidants from liquors. At this point, UF and NF membranes with narrow pore size were able to concentrate phenolic fractions between 0.615–1.09 mg L^{-1} from initial concentration of 0.173 mg L^{-1} in extracts. Artichoke wastewaters (AWs) are also important agro-food by-products, which have been a target of study for fractionation via integrated membrane process (Conidi et al. 2014b). In this waste, cynarin, chlorogenic acid and apigenin-7-O-glucoside were the primary molecules obtained with concentrations of 412, 612 and 400 mg L^{-1}, respectively. After evaluating their bioactivity, the complex of polyphenols displayed high antioxidant properties (ca. 40 mM Trolox).

Ultimately, an integrated membrane process was designed to extract bioactive compound compounds from Nixtamalization wastewaters (NWs) (Castro-Muñoz and Yañez-Fernandez 2015), recognized as typical by-product from the tortilla processing in America (Castro-Muñoz et al. 2017). To sum up, MF and tight UF membranes were capable to separate a phenol content of 951 mg L^{-1}. By analyzing the reported development works, this chapter has evidenced that UF and NF technologies can easily recover low molecular weight bioactives (such as phenolic compounds) from various wastewaters (Conidi et al. 2020; Cassano and Conidi 2019; Castro-Muñoz and Ruby-Figueroa 2019). Unfortunately, membrane technologies are still facing specific issues to consolidate their applications. The following section is devoted to the challenges and important factors in the framework of membrane technologies.

3 Challenges in Membrane Technologies for Bioactive Compounds Separation

The challenges of these processes deal with their weakness and limitations during the separation. In this way, it is likely that the "purity restriction" is one of the limitations of membrane technologies since most of the streams do not present pure compounds; in other words, none of the streams usually contain a minor amount of untargeted molecules (Castro-Muñoz et al. 2018b). For instance, the permeate samples can contain a significant amount of molecules aimed to concentrate in the retentate side; this is due to the fact that the membrane selectivity is not infinite. Herein, it is worth noting that the storage and handling of the membranes are crucial to extend the initial physicochemical properties and self-life, for example, if the membrane is treated by the right cleaning procedures without modifying its structure can be reutilized as long as needed. Of course, there is another important matter, like membrane fouling, to maintain the original properties of the membranes. The "fouling" is identified as the key drawback of these technologies and thus the main challenge at obtaining a more feasible and stable process, however, this is an inherent phenomenon since it depends on the types of feed bulk to be treated, basically, it is directly related to the physicochemical composition of feed (Gule et al. 2016). In addition to such factors, the fouling also becomes dependent on the membrane material and configuration, as well as operating and fluid-dynamic parameters. Particularly, the fouling, especially non-reversible fouling, can majorly restrict the permeation rate through the membranes and thus limiting their use towards specific applications.

Conventional protocols to control and regulate membrane fouling involve preliminary treatments of the feed solutions, including particle sedimentation-decantation (Fukuda et al. 2014), centrifugation (Domingues et al. 2014), flocculation (Maroušek et al. 2019), enzymatic hydrolysis (Galiano et al. 2019), screening, along with membrane surface modification (Kucera 2019), hydrodynamic optimization of the membrane module and membrane cleaning with commercial chemical or enzymatic detergents. To date, there are plenty of commercially available enzymatic (such as Ultrasil® 62 and 53, Filzym® 161) and chemical (Ultrasil® 13, OptiClean™ A, Ultrasil® 10A, AMI Chemicals® AM-55) detergents that are usually used to hydrolyze most of the pollutants, including polysaccharides, proteins, polysaccharide-like, protein-like materials and humic substances (Nguyen et al. 2010). In the field of membrane engineering, researchers are strongly working on several developments to prevent the adhesion of organic and inorganic matter that is translated to biofouling on membranes. Here, the core application has been the modification of the physicochemical properties of membranes, such as hydrophilicity, membrane charge, and membrane surface (Pichardo-Romero et al. 2020; Buonomenna 2016). Basically, the manufacture of highly hydrophilic membranes is a promising alternative since they are less prone to matter incrustation. In this regard, the preparation of nanocomposite membranes using inorganic materials and

additives seems to be the most advanced way (Castro-Muñoz et al. 2019b; Akar et al. 2013; Vatanpour et al. 2012; Zinadini et al. 2014).

Furthermore, over the course of this chapter, it has been also noted that the use of integrated membrane system can significantly contribute to reduce the early-stage fouling in membranes; a typical integrated membrane process implies the design and arrangement of multiple membrane units in sequence, contributing to mitigate fouling phenomena in the subsequent membrane stages by prepending high pore size membranes (Steeneveldt et al. 2006). Classic cases of mitigation of membrane fouling using membranes have been evidenced in the fractionation of agro-food wastes, such as artichoke wastewaters (Conidi et al. 2014b; Castro-Muñoz et al. 2018c), artichoke brines (Cassano et al. 2016b), OMWs (Russo 2007; Cassano et al. 2013), Nixtamalization (Castro-Muñoz et al. 2015a, c, 2016b; Castro-Muñoz 2019) and cellulose alkaline by-products (Cassano et al. 2016c). It is known that the implementation of integrated membrane system may require more bioseparation steps to meet high recovery rates; nevertheless, the right selection of the membranes and sequence design can give a potential strategy for the fractionation of the food and waste systems.

When dealing with the fabrication of membranes, polymers are among the main materials used in membrane preparation at industrial level. Here, there are thermal, mechanical and chemical limitations in this kind of membranes. Polymeric membrane modules cannot offer an operation at high temperature conditions since polymers do not guarantee their physical integrity at temperatures over 90–100 °C. In addition to this, most of the polymers are susceptible to chemical degradation when treating strong acid and alkaline substances, resulting in a significant reduction in membrane life. Also, specific polymeric membranes have limited mechanical stability leading to a diminish in permeability at high pressures and potential membrane failure. In this sense, it is likely that inorganic membranes, based on alumina (Al_2O_3), titania (TiO_2), silica (SiO_2) and zirconia (ZrO_2), can overcome such limitations showing greatly improved chemical, mechanical and thermal stability in comparison with polymeric membranes (Majumdar et al. 2020; Castro-Muñoz et al. 2019c). Inorganic membranes are able to operate at temperatures as high as 500 °C, with extreme pH values and they are suitable to be subjected for cleaning with chemicals, organic solvents and hot water.

Finally, as in most of the downstream processes, the energy consumption-cost relationship is a critical factor when regarding the feasibility of processes. Theoretically, pressure-driven membrane processes have been considered as low energy consumption separation techniques (Mirza 2008; Van Der Bruggen et al. 2003b), which in turn can reduce the operating costs, however, membrane modules represent the major direct capital cost of all the unit membrane separation, followed by the devices investment and their maintenance, which indeed contribute greatly to overall process costs.

4 Conclusion

In this chapter, membrane technologies have been demonstrated their ability in separation functional bioactive compounds and food ingredients from natural sources, as well as their derivative products and wastes. At this point, UF and NF membranes can be efficiently used to separate, fractionate and concentrate bioactive compounds that, according to their biological activity, have potential applications in the food and pharmaceutical industries. When compared with conventional recovery processes, these membrane-based processes are economically viable not only in terms of recovery, but also since they do not need the application of external agents. Apart from natural sources, the production of bioactive solutes from wastes is both industrially sustainable and environmentally friendly alternative.

It is quite possible that R&D will pay attention on new implementations of NF technology as the emerging tool for the recovery and concentration of phenolic-based compounds. Importantly, when further purification is required, the implementation of alternative selective methodologies, such as osmotic distillation and adsorption processes, will be needed. To finalize, this chapter also denotes the main challenges of membrane technologies in terms of purity restrictions, chemical, mechanical and thermal stability and energy consumption-cost relationship, which should be analyzed by technicians before applying at any recovery stage. Likewise, such criteria greatly play an important role in the consolidation of such technologies.

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