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Review

Opuntia (Cactaceae) plant compounds, biological activities and prospects – A comprehensive review

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ABSTRACT

Opuntia species are utilized as local medicinal interventions for chronic diseases and as food sources mainly because they possess nutritional properties and biological activities. The *Opuntia* plant is distributed worldwide and has great economic potential. Differences in *Opuntia* species phytochemical composition exist between wild and domesticated species, and within species. *Opuntia* aerial and underground parts exhibit beneficial properties due to their phenolic content, other antioxidants (for example ascorbate), pigments (carotenoids, betalains), and other unidentified components. This work comprehensively reviews the phytochemical composition of the different aerial and underground plant parts of *Opuntia* species. The applications of *Opuntia* compounds and their biological activities are also discussed. Other topical aspects covered include *Opuntia* spp. taurine composition, *Opuntia* side effects, *Opuntia* by-products valorisation and the role of *Opuntia* spp. in tackling antimicrobial resistance. Although biological activities have been extensively researched, much less information is available on reaction mechanisms, herbal mixtures toxicology and commercialisation prospects – aspects which should be considered for future research in this area.

1. Introduction

Bioactive compounds are compounds with nutritional benefits (Kris-Etherton et al., 2002; Kudanga, Nemadziva, & Le Roes-Hill, 2017), and are usually categorized into phenolic and non-phenolic compounds and pigments (Martins et al., 2011). Industrial applications of these bioactive compounds are increasing. Apart from their application in the pharmaceutical industries, bioactive compounds are being used in the food industry for the production of nutraceuticals (Gil-Chávez et al., 2013), novel food formulations (juices, beverages, jams, sweetners) (Gurrieri et al., 2000; Pawar, Killedar, & Dhuri, 2017) and for animal feed supplementation (Ennouri, Fetoui, Bourret, Zeghal, & Attia, 2006). The *Opuntia* plant is an interesting source of plant bioactive compounds/products (Gaballah, Embaby, Hamed, & El-Samahy, 2016). It belongs to the dicotyledonous angiosperm Cactaceae family of which about 1500 species of cactus are known (Butera et al., 2002). Apart from bioactive compounds, other *Opuntia* products have commercial value in the production of juices, alcoholic beverages, jams and natural liquid sweeteners (Abdel-Hameed, Nagaty, Salman, & Bazaid, 2014; Gurrieri et al., 2000; Lee, Kim, Kim, & Jang, 2002; Pawar et al., 2017). They are also used in agrochemicals, cosmetics, in chemical industries (Guaadaoui, Benaicha, Elmajdoub, Bellaoui, & Hamal, 2014), and in wastewater treatments (Zito et al., 2013).

Opuntia species are capable of growth in almost all climates, for example, arid, temperate and tropical climates (Butera et al., 2002; Lallouche, Boutekrabt, & Hadjkouider, 2015) (Table 1) and their bioactive compound profiles change with species, cultivar, and climate conditions (Paiva, de Souza, Costa, Santos, & Coelho, 2016). There is therefore an opportunity for the discovery of novel compounds from different *Opuntia* cultivars.

In addition, the current global trend in growth of antimicrobial resistance cannot be overemphasized. Microbial sources of antimicrobial compounds are depleting, hence the search for novel antimicrobials from plants such as *Opuntia* (Aremu, Amoo, Ndhlala, Finnie, & Van Staden, 2011; Valtierra-Rodríguez, Heredia, García, & Sánchez, 2010). Novel antimicrobials/compounds with biological activities may be present in both extractable and hydrolysable (non-extractable) polyphenol plant fractions which are generally referred to as macromolecular antioxidants (MAs). In the usual extractive processes, these fractions are ignored compared to the extractable polyphenol fractions (Arranz, Saura-Calixto, Shaha, & Kroon, 2009).

Macromolecular antioxidants (MAs) contribute to total biological activity of food matrices (Bensadón, Hervert-Hernández, Sáyago-Ayerdi, & Goñi, 2010). While extractable compounds of *Opuntia* species

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Table 1
Selected *Opuntia* spp. and their locations.

Species	Locations
Subgenus *Opuntia*	
Opuntia basilaris	Southwest United States (US) and northwest Mexico
Opuntia chlorotica	Native to southwest US, and the Sonoran and Mojave deserts
Opuntia engelmannii	Mexico
Opuntia ficus-indica	Originally in south-central Mexico; cultivated in warm parts of the world for its edible fruit
Opuntia fragilis	Northern Great Plains and as far west as British Columbia, also found in the southern Great Plains
Opuntia humifusa, Opuntia compressa var. humifusa	Throughout the US, east of the Great Plains and into southern Ontario
Opuntia leucotricha	Mountains of Central Mexico
Opuntia macrocentra	Southwest US and Northern Mexico
Opuntia macrorhiza	Throughout the Great Plains except for the northernmost areas (not found in North Dakota), and extending sporadically eastward as far as Kentucky
Opuntia ficus-indica	Spain, Italy
Opuntia microdasys	Mexico (Hidalgo)
Opuntia ficus-indica (prickly and spineless), *Opuntia stricta, Opuntia macrorhiza, Opuntia microdasys*	Algeria, Tunisia, Morocco, Kenya, Egypt, South Africa (Northern, Eastern and Southern Africa)
Opuntia dillenii, Opuntia ficus-indica, Opuntia humifusa	India, Iran, Korea, Saudi Arabia
Opuntia santa-rita	Texas, Arizona and Northern Mexico
Opuntia stricta	Coastal regions
Opuntia polyacantha	
Opuntia phaeacantha	
Opuntia lindheimeri, Opuntia engelmanii var. *lindheimeri*	
Opuntia littoralis	
Opuntia erinacea	
Opuntia pusilla	

Compiled from Griffiths, 1915; MacMahon & Wagner, 1985; Stiling, Rossi, & Gordon, 2000; Bobich & Nobel, 2001; Van Sittert, 2002; Goettsch & Hernández, 2005.

and their association with nutrition, health and chronic disease have been discussed in earlier reviews (Osuna-Martínez, Reyes-Esparza, & Rodríguez-Fragoso, 2014), there have been no reviews on the biological activities of *Opuntia* MA fractions; *Opuntia* taurine amino acid content; as well as potential risks associated with the use of *Opuntia* extracts/herbal mixtures. This work comprehensively reviews these topics and elaborates on novel applications of the *Opuntia* plant and its by-products, at subsistent and industrial levels. Current status of *Opuntia* species in helping to fight the ever-growing global trend of antimicrobial resistance is also discussed together with future commercialisation prospects.

2. *Opuntia* plant: locations of growth and general compound profile

Opuntia ficus-indica is a slow growing perennial shrub which can grow up to 3-5 m high. Some arid growth locations for *Opuntia* species include Saudi Arabia, Egypt, Pakistan, Middle East, Algeria, Iran, Libya, Mexico, South Africa, Spain (Feugang, Konarski, Zou, Stintzing, & Zou, 2006; Moussa-Ayoub et al., 2014). Semi-arid/steppe regions include South Asia, India, New Zealand, Australia, North America and North Africa (Feugang et al., 2006). The temperate regions where the plant may be found include Tunisia (Ayadi, Abdelmaksoud, Ennouri, & Attia, 2009), Korea (Jun, Cha, Yang, Choi, & Kim, 2013), Portugal, Belgium, Bulgaria (Feugang et al., 2006); while tropical countries include Brazil (Paiva et al., 2011), Ethiopia (Chiteva & Wairagu, 2013), Kenya (Kunyanga, Vellingiri, & Imungi, 2014), and the Mediterranean country of Morocco (Castellanos-Santiago & Yahia, 2008). Some *Opuntia* species and their locations of growth are shown in Table 1, but most species are concentrated in Mexico (from which it is native) and the Americas. Some *Opuntia* species have spread to some African and Asian regions and show significant biodiversity within and across species (Caruso, Currò, Las Casas, La Malfa, & Gentile, 2010). This biodiversity determines the component profile of each species, as well as other factors such as growth location, prevailing climatic conditions and soil conditions, among others (Ammar, Ennouri, Khemakhem, Yangui, & Attia, 2012; Butera et al., 2002).

Cactus (*Opuntia*) plants contain carotenoids, amino acids, vitamins C and E, fibres and antioxidant phenol components, for example, betalains (Table 2b) and flavonoids. These and other phytochemical groups such as phenolic acids, sterols, esters, coumarins, terpenoids, and alkaloids bring about several health benefits such as hypoglycaemic (Paiz et al., 2010; Piattelli, Minale, & Prota, 1965; Stintzing, Schieber, & Carle, 2001; Strack, Vogt, & Schliemann, 2003), and antioxidant activities (Osorio-Esquivel, Álvarez, Dorantes-Álvarez, & Giusti, 2011). Certain water-soluble nitrogenous *Opuntia* phytochemical compounds/pigments known as betalains are classified as betacyanins (red to purple coloured), and betaxanthins (yellow and orange coloured) (Hendry & Houghton, 1996). Betalains also serve as chemo-systematic markers as members of the Caryophyllales family (Khan & Giridhar, 2015; Slimen, Mabrouk, Hanène, Najar, & Abderrabba, 2017). *Opuntia ficus-indica* and other species also contain an array of flavonoid compounds (most notable are kaempferol and quercetin) and their derivatives (De Leo, Abreu, Pawlowska, Cioni, & Braca, 2010). Esters, alkaloids, phenolic acids, essential fatty acids (Ramadan & Mörsel, 2003b), polysaccharides, sugars (Ammar et al., 2012; Benayad, Martinez-Villaluenga, Friasa, Gomez-Cordoves, & Es-Safi, 2014; Yeddes, Chérif, & Ayadi, 2014; Yeddes, Chérif, Guyot, Baron, & Trabelsi-Ayadi, 2014) as well as alkanes, carotenoids, amino acids, sterols, among others, can be found in *Opuntia* species. (Jiang, Li, Chen, Min, & Lou, 2006; Manzano et al., 2017). The profiles of phenolic and non-phenolic components reported to be present in *Opuntia* species are presented in Tables 2a and 2b.

3. Cactus flower-associated compounds

Opuntia flowers come in various colours but colour progression during flowering begins from white through yellow to red/orange/pink/peach/cream. Flowers are auxillary, large, bisexual and possess tepals arranged spirally (Heuzé & Tran, 2017). The *Opuntia ficus-indica* flowers are considered as by-products and are usually cast-off after fruit separation (Benayad et al., 2014; Yeddes, Chérif, Guyot, et al., 2014).

There are fewer reports on *Opuntia* flower compounds compared to the fruits. Phenolics and betalain pigments form the major component of cactus flowers (Tables 2a, 2b) (Arcoleo, Ruccia, & Cusmano, 1961; Clark, Brown, & Mays, 1980; Clark & Parfitt, 1980; Shabir & Zaman, 1968). The betalamic acid core of betalains binds to different amino/imino groups to generate betaxanthins and betacyanins (Slimen et al.,

Table 2a
Phenolic and non-phenolic compounds in cactus plant parts.

Major Compounds	Cacti part	References
Gallic acid	Flower	Ammar et al., 2015; Clark & Parfitt, 1980, Clark et al., 1980; De Leo et al., 2010; Yeddes, Chérif, & Ayadi, 2014, Yeddes, Chérif, Guyot, et al., 2014
Quercetin 3-*O*-Rutinoside		
Quercetin 7-*O*-Rutinoside		
Kaempferol 3-*O*-Rutinoside		
Quercetin 3-*O*-Glucoside		
Isorhamnetin 3-*O*-Robinobioside		
Isorhamnetin 3-*O*-Galactoside		
Isorhamnetin 3-*O*-Rutinoside		
Isorhamnetin 3-*O*-Glucoside		
Isorhamnetin 7-*O*-rutinoside		Ammar et al., 2015; Benayad et al., 2014; De Leo et al., 2010
Isorhamnetin 3-O-rhamnosyl 7-O-rutinoside		
Kaempferol 3-*O*-arabinoside		
Rhamnetin 3-*O*-rutinoside		Merina et al., 2011
Other compounds		
Terpenes: (limonene, linalool, germacrene D, aromadendrene, squalene)		*Ammar* et al., *2012*
Esters and alkaloids (ethyl linoleate)		
Sterols		
Carboxylic acids (linoleic, palmitic, octanoic, butanedioic, pentanedioic)		
Phenolic acids: Hydroxycinnamic acid derivatives:		
(5-Hydroxyferulic acid-rhamnosidehexoside; Caffeoyl		
methoxycinnamoyl quinic acid; 1,4-Diferuloyl syringic acid; 4-p-Coumaroyl caffeic acid; 1,5-Dicaffeoyl		
ferulic acid; 1,4-Syringicferuloyl 4-coumaroyl caffeic acid)		
Flavonoids	Whole fruit/ pulp	Bensadón et al., 2010; Cha et al., 2013; Fernández-López et al., 2010; Mabrouki et al., 2015; Osorio-Esquivel et al., 2011
Rutin		
Myricetin		
Quercetin		
Isorhamnetin		
Kaempferol		
Luteolin		
Isorhamnetin glycosides		
Kaempferol glucosides		
Catechin		
Phyllocactin		Osorio-Esquivel et al., 2011; Tesoriere et al., 2004
Protocatechuic acid		
4-hydroxybenzoic acid,		Galati et al., 2003; Serra et al., 2013
Ferulic acid		
Vanillic acid (also isovanillic acid)		
Trans-coumaric and trans-cinnamic acids		
Syringic, Fukic, piscidic, and eucomic acids		
Betalains		Khatabi et al., 2013
Taxifolin		
Orientin		
Vitexin		
Volatile organic compounds (VOCs) - 2-decanynoic acid, γ terpinene, linalool, α farnesene etc.		Zrira, Petretto, Saidi, Salaris, & Pintore, 2016
Other compounds		
Carotenoids [Lutein- 5,6- epoxide, (all-E)-b-criptoxanthin, (all-E)-a-carotene, (all-E)-b-carotene, (9Z)-b-carotene, lycopene].		Feugang et al., 2006; Tesoriere et al., 2005
Ascorbic acid and Tocopherols		
Amino acids (taurine, cystine)		
Biothiols (glutathione)		
Phenolic acid	Seed	Chougui et al., 2013; Tlili, Bargougui, et al., 2011
Feruloyl-sucrose isomer 1		
Feruloyl-sucrose isomer 2		
Sinapoyl-diglucoside		
Flavonoids		
Tannins		
Fatty acids (linoleic, oleic, palmitic, stearic, vaccenic); sterols, vitamin E		Ghazi et al., 2013; Tlili, Bargougui, et al., 2011; Zakynthinos & Varzakas, 2016
Total phenolic acid	Peel/Skin	Kuti, 2004; Moussa-Ayoub, El-Samahy, Kroh, & Rohn, 2011, Moussa-Ayoub, El-Samahy, Rohn, & Kroh, 2011; Ndhlala et al., 2007
Flavonoids – Kaempferol, Quercetin, Isorhamnetin, Isorhamnetin glucosides		
Gallic acid, Rutin, Catechin, Epicatechin, Vanillin		Guzman-Maldonado et al., 2010
17-decarboxybetanin, betanin		Abou-Elella & Ali, 2014
Xanthophylls		Cano et al., 2017; Yahia et al., 2010;
Terpene alcohols		Koubaa, Ktata, Bouaziz, et al., 2015
Fatty acids; Minerals (Fe, Mn, Mg, Ca, Zn); Glucose, fructose		El-Salid, Nagib, Rahman, & Deraz, 2011; Guzman-Maldonado et al., 2010
Polysaccharides		Koubaa, Ktata, Bouaziz, et al., 2015
Phospholipids and glycolipids		Ramadan & Mörsel, 2003a; Ramadan & Mörsel, 2003b
Phytoestrogens		Diaz-Vela et al., 2013; Elleuch et al., 2011; Federici et al., 2009

(*continued on next page*)

Table 2a (*continued*)

Major Compounds	Cacti part	References
Phenolic acids - Gallic acid, Coumaric acid, 3,4-dihydroxybenzoic, 4-hydroxybenzoic; Ferulic, Salicylic, Vanillic Syringic, Synaptic acids; Protocatechuic acid etc.	Cladode	Bensadón et al., 2010; Gallegos-Infante et al., 2009; Guevara-Figueroa et al., 2010; Valente et al., 2010; Wright & Setzer, 2014
Flavonoid - Isorhamnetin, Quercetin, Kaempferol, Isoquercetin, Nicotiflorin, Rutin, Narcissin		Avila-Nava et al., 2014; Gallegos-Infante et al., 2009; Ginestra et al., 2009; Guevara-Figueroa et al., 2010;
Terpenoid volatiles (cis-linalool oxide, trans-linalool oxide)		
Fatty acids and tocopherols		Chahdoura et al., 2014
Other components		
Alkanes (heptadecane)		Avila-Nava et al., 2014; Betancourt-Domínguez, Hernández-Pérez, García-Saucedo, Cruz-Hernández, & Paredes-López, 2006
Ascorbic acid		
β-carotene, Lutein		
alpha-pyrones, opuntiol, opuntiosides		Qiu, Zhao, Doude, Xu, & Liu, 2007; Siddiqui et al., 2016
Pectic polysaccharides		Panico et al., 2007; Yang et al., 2008
Lignans		Rocchetti et al., 2018)
Flavonoids	Stem	Jiang et al., 2006; Manzano et al., 2017
Glycosides		
Coumarins		
Terpenoids:		De Las Heras & Hortelano, 2009; Jamal, Yaacob, & Din, 2009;
Triterpenes, Labdane-type diterpenes		Manzano et al., 2017
Tannins		Manzano et al., 2017
Polysaccharides		
Fatty acids (azelaic acid, palmitelaidic acid)		
C_{29}-5β-sterols (Taraxerol)		Jiang et al., 2006

2017). Studies have also shown the presence of unidentified isorhamnetin glycoside (Arcoleo, Bellino, & Ruccia, 1962); as well as esters, sterols and fatty acids (Arcoleo, Ruccia, & Natoli, 1966) and flavonoids (Chahdoura et al., 2014; Yeddes, Chérif, & Ayadi, 2014; Yeddes, Chérif, Guyot, et al., 2014). More recently, quercetin (Yeddes, Chérif, & Ayadi, 2014, Yeddes, Chérif, Guyot, et al., 2014), isorhamnetin 3-*O*-robinobioside (most predominant component), and other volatiles such as (E)-3-hexen-1-ol, 1-hexanol, nonanal, 2-ethyl hexyl acetate, tetrahydrolavandulol, have been identified in *Opuntia* flowers (Table 2a) (De Leo et al., 2010). Volatile compounds from aqueous distillates have been derived from *Opuntia macrorhiza*, *Opuntia lindheimeri*, and *Opuntia microdasys* (Bergaoui et al., 2007). Other non-phenolics such as sugars; α-, β-, γ-, and δ-tocopherols; organic acids; fatty acids [saturated, monounsaturated and polyunsaturated fatty acids (PUFAs)] and hydroxycinnamoyls (Table 2a) have also been quantified in almost all the flowering stages (Chahdoura et al., 2014). The total amount of flavonoids and other phenols in *Opuntia* flowers have been shown to vary with extraction solvent used, developmental flower stage/phase, and processing methodologies employed (Ammar et al., 2012; Cevallos, Byrne, Okie, & Cisneros, 2006; Lee & Kader, 2000).

The use of gas chromatography and mass spectrometry in the analysis of Tunisian *Opuntia ficus-indica* (L.) and *Opuntia stricta* (Haw.) flowers at different developmental stages (vegetative phase, initial, full and post-flowering phases) also showed significant variations in extract compound profiles (Ammar et al., 2012). Furane aromatic compound was found in *Opuntia ficus-indica* budding stage but not in *Opuntia stricta*, while hexadecanoique carboxylic acid was found in all flower developmental stages of both species. Nonadecane hydrocarbon was also reported to be found in all flowering stages of *Opuntia stricta* only (Ammar et al., 2012). Some of the major compounds were alcohols, esters and terpenes. Polar solvents have been shown to be most efficient in polyphenol extraction of *O. ficus-indica* flowers (Ammar et al., 2015). According to the aurthors, the methanolic/soxhlet extract (EC_{50} of 160 μg/mL) and aqueous extracts (EC_{50} of 200 μg/mL) showed more phenolic compound types and best antioxidant capacity as measured by their ability to reduce Fe^{3+} to Fe^{2+}. Compared to the maceration method of extraction, the Soxhlet method was more efficacious and demonstrated the presence of thermostable polyphenols which contributed significantly to the antioxidant activity of *O. ficus-indica* flowers. Generally, higher extract yields were observed for Soxhlet compared to the maceration processing method. Similarly, Benayad et al. (2014) demonstrated that the green and economic processing method of accelerated solvent extraction (ASE) gave improved yields/expression of flavonoids and phenols than the maceration method.

Other biological activities have also been reported for *Opuntia* flowers. It has been demonstrated that *Opuntia* flower infusions possess *in vitro* and *in vivo* diuretic activity which was linked to their potassium content (Galati, Tripodo, Trovato, Miceli, & Monforte, 2002). Interestingly, research on the polyphenolic profile of Moroccan *O. ficus-indica* flowers showed that the flowers could be used as a starter ingredient for preparation of nutraceuticals/infusions (Benayad et al., 2014; Hossain, Barry-Ryan, Martin-Diana, & Brunton, 2011). The presence of flavonoids in extracts have also been associated with antiinflammatory (Benayad et al., 2014), anti-atherosclerotic (Fuhrman, Volkova, Coleman, & Aviram, 2005), and anti-angiogenic (Piao, Mori, Satoh, Sugita, & Tokunaga, 2006) bioactivities. Recent research has shown that *Opuntia* flower methanolic and mucilage extracts [*in vitro* effective concentration (EC_{50}) of DPPH radical scavenging reported at 200 and 230 μg/mL, respectively] have shown complete wound healing capacities on day thirteen of treatment (Ammar et al., 2015). Cactus pear flowers are rich in natural bioactive polyphenolic components which could find use in food industry, as well as in pharmaceutical and cosmeceutical preparations. These functions give them added value and reduce their environmental impact (Benayad et al., 2014; Merina, Kesavan, & Sulochana, 2011).

Generally, the biological activities of major phytochemical groups in *Opuntia* species plant parts are closely linked with structure of compounds within the groups, working singly or synergistically within extracts (Cai, Sun, Xing, Luo, & Corke, 2006). These phytochemicals are secondary matabolites whose structure consist of one or more hydroxyl groups linked to aromatic rings (Sant'Anna, Gurak, Marczak, & Tessaro, 2013). Most phenolic/bioactive molecules belong to tannins, stilbenes, phenolic acids, coumarins, quinones, flavonoids/flavonoid glycosides and lignans. Generally, antioxidant activity and other pharmacological and physiological functions are associated with these compound structures, where the number and position of hydroxyl (OH^-) groups and presence of substituents such as methoxy and attached glucose molecules may either reduce or enhance biological activities. Tannins (polymeric phenols with multiple number of hydroxyl groups) and *ortho*-dihydroxy groups have been shown to have strongest bioactivites compared to quinones and lignans (Cai et al., 2006).

Study has shown absence of antioxidant activity in cinnamic and benzoic acids without hydroxyl groups (Cai et al., 2006). In addition,

Table 2b
Betalains (betaxanthins and betacyanins) associated with *Opuntia* species

	Betaxanthin pigments	Special characteristics		Attached amino/imino groups	*Opuntia* species	References
		[M + H]*	kmax (nm)			
1	Portulacaxanthin I	*325, 309	483	R3 = hydroxyproline	*Opuntia ficus-indica*	Castellanos-Santiago & Yahia, 2008; Khan & Giridhar, 2015
2	Portulacaxanthin III	269	470	R3 = glycine	*O. ficus-indica*	Castellanos-Santiago & Yahia, 2008; Strack et al., 2003
3	Muscaaurin	349	472	R3 = histidine	*O. robusta, O. ficus-indica, O. megacantha, O. robusta Wendl, O. robusta, O. streptacantha*	Castellanos-Santiago & Yahia, 2008; Stintzing et al., 2005
4	Indicaxanthin	309	*260,305, 485	R3 = proline	*Lemaire, O. ficus-indica, O. megacantha; O. albi-carpa*	Castellanos-Santiago & Yahia, 2008; Stintzing et al., 2001; Stintzing, Schieber, & Carle, 2002
5	(S)-serine-betaxanthin	299	468	R3 = serine	*O. ficus-indica*	Castellanos-Santiago & Yahia, 2008; Strack et al., 2003
6	(S)-valine-betaxanthin	311	470	R3 = valine	*O. ficus-indica*	Khan & Giridhar, 2015; Strack et al., 2003
7	(S)-isoleucine-betaxanthin	325	470	R3 = isoleucine	*O. ficus-indica*	Khan & Giridhar, 2015; Strack et al., 2003
8	γ-Aminobutyric acid-Bx	297	451	R = butyric acid	*Opuntia* spp.	Khan & Giridhar, 2015; Strack et al., 2003
9	Methionine-betaxanthin	*343, 299	477	R3 = methionine	*Opuntia* spp.	Khan & Giridhar, 2015; Strack et al., 2003
10	(S)- Phenylalaine-betaxanthin	359	472	R3 = phenylalanine	*O. ficus-indica*	Khan & Giridhar, 2015; Strack et al., 2003
11	Vulgaxanthin I	340	470	R3 = glutamine	*O. robusta Wendl, O. ficus-indica; O. streptacantha; O. beta vulgaris*	Piattelli et al., 1965; Strack et al., 2003
12	Vulgaxanthin II	341	469	R = glutamic acid	*O. bergeriana; O. ficus indica; O. alba-carba; O. robusta and O.* spp	Piattelli et al., 1965; Stintzing et al., 2005
13	Vulgaxanthin III	326	470	R = asparagine	*O. streptacantha; O. beta vulgaris; O. alba-carba; O. robusta; O.* spp	Kugler, Stintzing, & Carle, 2004; Stintzing et al., 2005
14	Vulgaxanthin IV	325	470	R3 = leucine	*O. streptacantha; O. beta vulgaris; O. alba-carba; O. robusta Wendl, O. ficus-indica*	Kugler et al., 2004; Castellanos-Santiago & Yahia, 2008; Strack et al., 2003
15	Miraxanthin II	–	477	R3 = aspartic acid	*O. bergeriana; O. ficus indica*	Piattelli et al., 1965; Stintzing et al., 2001
	Betacyanin pigments					
16	Betanin	551.389, 149	537	R1 = R2 = H	*O. robusta Wendl, O. robusta, O. streptacantha, O. ficus-indica, O. megacantha, O. albi-carpa, O. xoconostle*	Osorio-Esquivel et al., 2011; Castellanos-Santiago & Yahia, 2008; Stintzing et al., 2005
17	Iso-betanin	–	–	R1 = R2 = H	*O. robusta Wendl, O. robusta, O. streptacantha, Lemaire, O. ficus-indica, O. xoconostle*	Osorio-Esquivel et al., 2011; Castellanos-Santiago & Yahia, 2008; Stintzing et al., 2005

– = data not available.
* kmax values differ based on the solvent and instrument.

weak antiradical activity was reported for monohydroxybenzoic acids, but methoxy groups (-OCH_3) could improve the activity of the monohydroxybenzoates. Syringic acid (4-OH, 3,5-OCH_3) and vanillic acid (4-OH, 3-OCH_3) had higher activity than the monohydroxybenzoates without methoxy groups. Syringic acid was even more active than protocatechuic acid (dihydroxy). This suggested that the methoxy groups in hydroxybenzoic acid molecules might enhance hydrogen-donating capacity and radical scavenging activity (Natella, Nardini, Felice, & Scaccini, 1999). Generally, electron donating groups such as methoxy groups tend to enhance antioxidant capacity while electron withdrawaing groups have an opposite effect.

Flavonoid antioxidative and cardioprotective activites arise from the ability to inhibit lipid peroxidation, chelate redox-active metals, and inhibit processes involving reactive oxygen species. They do this by electron transfer (Tsao, 2010), activating antioxidant enzymes (Elliott, Scheiber, Thomas, & Pardini, 1992), and inhibiting oxidases (Cos et al., 1998). Anti-inflammatory activity is brought about by the inhibition of inflammatory pathway and enzymes involved, and induction of nitric oxide synthase and cyclooxygenase-2 (COX-2) enzymes (Togna et al., 2014). The induction of cell apoptosis by bioactive compounds is related to anticancer activity where proapoptotic protein expression is elevated. Chromatin condensation, mitochondria-transmembrane-depolarization potential decrease and cellular shrinkage in cancer cells are some of the reported anticancer mechanisms of action (Lin et al., 2014). Phytosterols act by reducing intestinal absorption of cholesterols (Bartnikowska, 2009). They are also able to elevate antioxidant enzymes levels and induce cell death (Soodabeh, Azadeh, Ahmad, & Mohammad, 2014).

4. Cactus (*Opuntia*) seeds related components

Opuntia seeds refer to the small hard part of the cactus pear fruit which consists of the embryo and its food source, protected within a seed testa/coat. It is a mature plant ovule which may be sown and is capable of germination and growth into a whole new plant. *Opuntia* seeds may exhibit induced (by environmental factors) or natural dormancy (Podda, Santo, Leone, Mayoral, & Bacchetta, 2017; Reyes-Agüero & Valiente-Banuet, 2006).

Opuntia plant seeds are a good but overlooked source of natural bioactive phytochemicals with potential food, pharmaceutical and industrial applications (Romero, Tovar, Ramo, & Motilva, 2003; Tlili et al., 2009). They could serve as potential mine fields for generation of oleochemicals, which could reduce dependence on petrochemicals or supplement them in the future (Falade et al., 2008). Cactus seed compounds include fatty acids (Ennouri et al., 2006; Tlili, Bargougui, Elfalleh, Triki, & Nasri, 2011), and vitamins from *Opuntia microdasys* (Lhem.) and *Opuntia macrorhiza* (Engelm.) (Chahdoura et al., 2015). Sugars and organic acids such as oxalic, quinic and malic acids (Chahdoura et al., 2015; Chougui et al., 2013), have also been identified from *Opuntia* seeds (Table 2a). In addition, presence of sterols (β-sitosterol, campesterol, stigmasterol and fucosterol) have been reported in Moroccan *Opuntia dillenii* and *Opuntia ficus-indica*. Oil extraction is usually achieved with supercritical carbon (IV) oxide extraction, or soxhlet extraction method using petroleum ether, hexane or chloroform: methanol (2:1 v/v) at varied lengths of time (Ghazi, Ramdani, Fauconnier, El Mahi, & Cheikh, 2013).

Recently, clinical evidence has shown that polyunsaturated fatty acids (PUFAs) can help in managing stroke and coronary heart disease symptoms (Calder, 2008; Gupta, Sarkar, Baral, & Laskar, 2010). Interestingly, at 2 ml/kg body weight, *Opuntia* seed oil extracted without the use of solvent showed significant reduction in alloxan-induced death *in vivo* (Berraaouan et al., 2015). According to the authors, a 0.96 mg/mL IC_{50} of DPPH scavenging activity was reported to contribute to this effect. The seed oil achieved 86.2% inhibition compared to vitamin C which gave 97.12% DPPH inhibition after 90 min of mice treatment (Berraaouan et al., 2015). This reductive capacity can be attributed to vitamin K1, tocopherols, β-Carotene (Kamimura et al., 2013; Ramadan & Mörsel, 2003a), phenol compounds (Chougui et al., 2013), and UFAs in the seed extract (Suresh & Das, 2001). Increased antioxidant activities have also been recorded for supercritical (SC)-CO_2 extract (172.2 ± 11.9 µg GAE/g of oil) compared to the hexane fraction (76.0 ± 6.9 µg GAE/g of oil). This observation was related to release/expression of more polyphenols acting synergistically (Koubaa et al., 2017).

Analysis of *Opuntia* seeds have shown the presence of phenol −268.4 mg/100 g (Tlili, Elguizani, Nasri, Khaldi, & Triki, 2011) and 48–94 mg GAE/100g (Chougui et al., 2013), as well as flavonoid (1.5–2.8 mg QE/100g) and tannin (4.1–6.7 mg CE/100 g) (Chougui et al., 2013). Phenol composition of defatted extract from *Opuntia* seed correlated significantly with their antioxidant capacity. (Khoo, Azlan, Ismail, & Abas, 2012; Mondor et al., 2009). More than twenty compounds were detected with varying complexities at 330 nm after liquid chromatographic (LC) separation. Significant differences in antioxidant activity have also been recorded for ground seeds compared to whole prickly pear seeds which was attributed to their high total phenol composition (Chaalal, Louaileche, Touati, & Bey, 2013; Morales, Barros, Ramírez-Moreno, Santos-Buelga, & Ferreira, 2014). As an added function, prickly pear seed oil extracts have also shown anticorrosion capacity, inhibiting corrosion of mild steel in 1M HCl (Hmamou et al., 2012). Therefore, plant seeds such as *Opuntia* seeds which are found in abundance could be explored for their phenolic and non-phenolic compound composition, as well as for their biological activities which are related to the aromaticity of phenol compound structure, number of hydroxyl groups and nature of other chemical substituents present (Tlili, Elguizani, et al., 2011).

Furofuran-type lignans identified and purified from *Opuntia* seeds showed hepatoprotective activity (EC_{50}; 13.7–22.2 µM) (Kim, Yang, Kim, Kim, & Sung, 2017). Lignans bioactivity has been linked to their ability to precipitate proteins (Naumann et al., 2014), as well as their structural flexibility, polarity, and molecular weight (Hagerman, 2012). Lignans possess a dimeric phenylpropanoids core structural unit with attachment of various structural subunits and substituent such as hydroxyl, oxygen, methyl and other groups to produce different lignan compound groups. Nature of the substituting groups significantly affect biological property of lignans (de Souza et al., 2005).

In addition, supplementation of animal diets with 25 g/kg of *Opuntia ficus-indica* seed oil also reduced feed conversion efficiency (Ennouri et al., 2006). According to the authors, cholesterol, low and very low-density lipoproteins (LDL and VLDL) levels were also reduced due to the seed oil richness in tocopherol and phytosterols. Compared to the control group fed on standard diet, treatment mice groups fed on the seed oil enriched feed diet showed 22% reduction in serum glucose levels. However, there was no signifant difference in weight gain for both groups which was attributed to low uptake of oil from the lipid supplemented diet compared to the control mice group (Ennouri et al., 2006).

5. Cactus (*Opuntia*) peel/skin related compounds

Opuntia peel makes up 60% of the entire fruit (Ramadan & Mörsel, 2003b). Peels are the rinds of the *Opuntia* fruit which are often discarded after separation from the fruit and are regarded as a by-product (Ramadan & Mörsel, 2003b). The generation of high value food products with improved nutritional value through food processing methods such as fermentation applied to agro-industrial by-products (e.g. *Opuntia* peels/skin) will continually evolve as an economically viable option. The use of these food by-products as food themselves or supplements/additives to produce functional foods with improved health benefits is expanding (Lamsal & Faubion, 2009). As food ingredients, fruit peels can be considered a source of fibre for prebiotic preparations and antioxidants (Diaz-Vela, Totosaus, Cruz-Guerrero, & Pérez-Chabela, 2013; Rodrıguez, Jimenez, Fernndez-Bolanos, Guillen, & Heredia,

2006). Studies have shown that non-digestible oligosaccharides from plant by-products with prebiotic potential can undergo fermentative processes involving lactic acid bacteria (LAB) and bifidobacteria to produce new compounds, with enhanced health benefits (Diaz-Vela et al., 2013; Gibson & Roberfroid, 1995).

Opuntia peels contain fatty acids, carbohydrates, vitamins and antioxidants (Cerezal & Duarte, 2005; Ramadan & Mörsel, 2003b). They also have significant amounts of fibre (32.67%), ascorbic acid (87.82%) and pectin (14.25%). The antioxidant components identified include oleuro, pyrogallol, benzoic, 3-OH tyrosol, ellagic, chorogenic, protocatechuic acid, p-oh-benzoic, epicatechin, and gallic acid. When the antioxidant-rich powder of *Opuntia* peel was used in supplementation of wheat flour it improved bread nutrient composition, shelf life and reduced staling (Anwar & Sallam, 2016). Also, *in vivo* study with peel extracts used in hamster diet supplementation increased excretion of cholesterol and lowered liver cholesterol levels. Mechanisms of extracts hypocholesterolemic activities include inhibition of hepatic HMG-CoA activity, interference with cholesterol absorption in the intestine and bile acid trafficking and antioxidant activity (Milán-Noris, Chavez-Santoscoy, Olmos-Nakamura, Gutiérrez-Uribe, & Serna-Saldívar, 2016). These mechanisms work synergistically for coronary heart disease prevention (del Socorro Santos, Barba de la Rosa, Héliès-Toussaint, Guéraud, & Nègre-Salvayre, 2017). In addition to this, Diaz-Vela et al. (2013) showed that fermented flours processed from *Opuntia* peels had higher carbohydrate and fibre content, and significantly higher antioxidant activities (89% inhibition of ABTS and 2.6 trolox equivalent antioxidant capacity value) compared to pineapple peel flours (69% ABTS radical inhibition and 1.5 trolox equivalent antioxidant capacity value). Fermentation kinetics showed that *Pediococcus pentosaceus* was the best microorganism for the fermentation process (Diaz-Vela et al., 2013).

Opuntia ficus-indica (L.) Mill] peels contain considerable amounts of neutral glycolipids and phospholipids (Ramadan & Mörsel, 2003b). 17-Decarboxy betanin and betanin (Table 2b) (Abou-Elella & Ali, 2014); xantophylls [(all-E)-lutein, (all-E)-violaxanthin and (all-E)-zeaxanthin], hydrocarbon carotenes (belonging to two types of oxygenated carotenoid derivatives); and chlorophyll (Cano, Gómez-Maqueo, García-Cayuela, & Welti-Chanes, 2017; Yahia, Castellanos, & Mondragon-Jacobo, 2010), have also been identified. Flavonoid glycosides dominate the flavonoid profile of cactus peels (Ginestra et al., 2009; Moussa-Ayoub, El-Samahy, Kroh, & Rohn, 2011; Moussa-Ayoub, El-Samahy, Rohn, & Kroh, 2011). Spineless cultivars contain more flavonols than the spiny/prickly varieties, and prickly pear peels contain higher level of flavonoids when compared to the pulp (Yeddes, Chérif, Guyot, Sotin, & Ayadi, 2013). Polysaccharides have also been reported in *Opuntia stricta* Haw. peel extracts and exhibited significant antioxidant activity measured by 94% DDPH inhibition at 50 mg/ml concentration and 6.5 mg/ml IC_{50} value (Koubaa et al., 2015). Other bioactive components include phytoestrogens (Diaz-Vela et al., 2013; Elleuch et al., 2011) and terpene alcohols (Koubaa et al., 2015).

Anticarcinogenic and antihypertensive activities have also been reported (Federici, Fava, Kalogerakisc, & Mantzavinos, 2009; Schieber, Stintzing, & Carle, 2001). *Opuntia ficus-indica* fruit peel extracts have also exhibited anticancer activity in Ehrlich Ascites Carcinoma Cells (EACC). Their potency and total phenolic content were significantly related, with two purified compound fractions having cancer cell killing efficicency of 51.5–76.0 dead cells/100 μg/ml (Abou-Elella & Ali, 2014). By induction of hyperpolarization in cell membrane, *O. ficus-indica* extract phenolic composition significantly increased calcium levels within human Jurkat T-cell lines (Aires et al., 2004). The extent of molecular branching, chemical structure and composition, presence of sugar moieties and physicochemical attributes of these bioactive molecules, are responsible for the biological activites reported for *Opuntia* peel compounds/extracts/products (Koubaa, Ktata, Barba, et al., 2015).

The components and bioactivities of peel extracts may depend on the extraction method. Recently, ultrasound and pulsed electric field technologies have been successfully used to achieve greater cell denaturation to facilitate better recovery of the intracellular coloured compounds from red prickly pear fruit peels (Koubaa et al., 2016). At higher temperatures more phenolic compounds are extracted, but with a reduction in antioxidant activity. Optimal extraction methods for retention of phenolic and antioxidant activities was achieved at 80 °C with 45% ethanol after 2 h of extraction (Han et al., 2016; Jorge, De La Garza, Alejandro, Ruth, & Noé, 2013). The physicochemical properties of peel fibres can be exploited in the food industry to improve food product parameters such as shelf-life, sensory attribues, staling, and viscosity. Peels are cheap and readily available, and these factors enhance the potential for commercialisation. They may also be applied as fat or sugar substitutes to stabilise oxidative processes in foods, and as oil and water retention capacity enhancers (Anwar & Sallam, 2016; Elleuch et al., 2011). Preservation of margarine products may also be achieved by replacement of vitamin E with prickly pear peels (Chougui et al., 2015). Margarines containing peel extracts resisted oxidation better than vitamin E margarine. The peel extract did not affect the physicochemical or the microbiological properties of the margarine (Chougui et al., 2015). Therefore, *Opuntia* peels have great commercialisation prospects.

6. *Opuntia* cladodes compounds/produtcs, fingerprinting and biological activities

Cladodes are the spiny or spineless paddle-like, oblong (up to 70–80 cm), thick and succulent parts of the *Opuntia* plant with varying widths. They possess a waxy, water repellent epidermis and are capable of photosynthesis and asexual reproduction. Small bristles (glochidia) may be present with barbs in the areoles (Heuzé & Tran, 2017). Several studies have shown that *Opuntia* cladode compound profile changes with species type, post-harvest treatment, environmental conditions and plant age (Astello-García et al., 2015; Contreras-Padilla et al., 2011; Guevara-Figueroa et al., 2010). Nevertheless, some studies have demonstrated that different cladode cultivars from distinct locations have similar flavonol profiles which may be used in taxonomic identification/classification of *Opuntia* cultivars/products (Moussa-Ayoub et al., 2014). Only isorhamnetin aglycon (m/z 315) was detected which was in partial agreement with results published by Santos-Zea, Gutierrez-Uribe, and Serna-Saldivar (2011) and Ginestra et al. (2009) from cactus *O. ficus-indica* cladodes collected from Mexico and Italy respectively. Flavonoid glycosides such as isoquercitrin, kaempferol-3-*O*-rutinoside, isorhamnetin-3-*O*-glucoside and isorhamnetin-3-*O*-rutinoside were detected in wild and commercial *O. ficus-indica* cladodes cultivated in Mexico (same location and close phylogenetic proximity), but no kaempferol was detected (Guevara-Figueroa et al., 2010). Sugars, fatty acids, tocopherols and organic acids were reported in cladodes from *O. microdasys* and *O. macrorhiza* cladodes from Tunisia (Chahdoura et al., 2014). Eucomic acid, kaempferol 3-*O*-robinobioside-7-*O*-arabinofuranoside and isorhamnetin 3-*O*-rhamno-side-7-*O*-(rhamnosyl-hexoside) dominated the phenolic profile of *O. ficus-indica* (highly domesticated), *O. albi-carpa* (intermediate domestication), and *Opuntia hyptiacantha* and *O. streptacantha* (wildest). Lesser amounts of quercetin 3-*O*-rhamnosyl-(1-2)-[rhamnosyl-(1-6)]-glucoside) were reported in domesticated species, and kaempferol 3-*O*-arabinofuranoside in wild cultivars (Astello-García et al., 2015). In other studies, wild cladode varieties had similar or higher amounts of pigment compounds such as β-carotene, lutein and xanthophylls compared to commercial ones (Jun et al., 2013).

Cladode extracts of *O. ficus-indica* have been shown to lower cholesterol levels and have anti-ulcer and antiinflammatory activities. Aqueous extracts also improve wound healing (Laughton, Evans, Moroney, Hoult, & Halliwell, 1991). Nopal extracts containing quercetin, isorhamnetin, kaempferol and vitamin C have also shown *in vitro* (IC_{50} = 0.8 mg/mL) and *in vivo* (20% increase in plasma antioxidant activity after 3 days of subjects feeding) antioxidant activity (Avila-

Nava et al., 2014) and hepatoprotective activities (Hfaiedh et al., 2008; Zourgui, Golli, Bouaziz, Bacha, & Hassen, 2008). Antidiabetic and antioxidant activities of cladode extracts have been attributed to significant amounts of polysaccharides, as well as lignans, flavonols and phenolic acids in the cladode extracts (Rocchetti, Pellizzoni, Montesano, & Lucini, 2018; Valente et al., 2010; Yang et al., 2008). *Opuntia ficus-indica* nopal extracts rich in polysaccharides and polyphenols were also able to decrease the hydroxyl radical induced oxidation of linoleic acid and DNA (IC_{50} of 9.3 μg/mL, better than 3.2 μg/mL for ascorbic acid) (Lee et al., 2002; Panico et al., 2007), and may find use as surfactant and natural antioxidants in food and pharmaceutical industries. Mucilage, pectin and total pectic polysaccharide fractions of *Opuntia ficus-indica* cladodes were characterized and showed antioxidant activity (Bayar, Kriaa, & Kammoun, 2016). Cladode extract radical scavenging abilities may be linked to presence of phenolic antioxidants and vitamin content (Lee et al., 2002; Stintzing & Carle, 2005). Cladode cultivars have further demonstrated useful anticarcinogenic selenium-enriched chemotherapeutic properties and could serve as food providing advanced dietary selenopharmacology to help fight human diseases (Bañuelos et al., 2012).

Karachian *Opuntia dillenii* (Nagphana) cladodes have been shown to contain α-pyrones, and novel opuntiol and opuntioside (opuntiol glucosides) compounds (Siddiqui et al., 2016). Opuntiol and opuntioside have been successfully utilized in the reduction of pain, but opuntioside showed a more effective analgesic response (IC_{50} 26 ± 0.9 mg/kg). In the future design of novel analgesics, both compounds may serve as integral constituents (Siddiqui et al., 2016). *O. humifisa* cladode extract also protects against UVB-induced skin degeneration achieved by reducing associated enzymes gene expression (Park, Choi, Hong, Jung, & Suh, 2017). Recently, azelaic acid and labdane-type diterpene were detected in cactus cladodes (Manzano et al., 2017). Azelaic acid shows good antibacterial activity (Sieber & Hegel, 2014), and its antiinflammatory properties is hypothesized to be linked to increased expression and transcriptional activity of peroxisome proliferator-activated receptor gamma (PPARγ) (Mastrofrancesco et al., 2010). Diterpenes demonstrate broad biological effects (de las Heras & Hortelano, 2009). Bioactivities may be linked to terpenoids possession of isoprene structure with added elements such as oxygen and acetate units, as well as the branched, cyclic structure and the presence of hydroxyl groups (Ciocan & Bara, 2007). Palmitelaidic acid, a monosaturated fatty acid, was also detected and shown to regulate fatty acids metabolism for application in weight management (Kadegowda, Burns, Miller, & Duckett, 2013). It is interesting also that under stressed growth conditions, *Opuntia* cladodes can biologically accumulate compounds.

Algerian *O. engelmanii* and *O. streptacantha* cladode cultivars accumulated significant amounts of proline and soluble sugars which ensured osmotic adjustment under saline stress conditions (Lallouche et al., 2015). This phenomenon was also observed in other parts of the cactus plant, but to a lesser extent as salinity increased (Hadjadj, Djerroudi, & Bissati, 2011; Lallouche et al., 2015). This bioaccumulative potential may pave the way for the application of cladodes in atmospheric pollution. Recent research demonstrated that *Opuntia ficus-indica* cladodes accumulated lead when exposed to lead-bearing apatite particles over a four-month period (El Hayek, El Samrani, Lartiges, Kazpard, & Aigouy, 2017). *Opuntia* cladodes are also able to bioaccumulate inorganic constituents such as magnesium oxide, calcium carbonate (calcite), calcium oxalate monohydrate, calcium-magnesium bicarbonate, potassium chloride and potassium peroxydiphosphate [$K_4P_2O_8$] which have been identified using diffractive, microscopic, and spectrometric techniques (Contreras-Padilla, Rivera-Muñoz, Gutiérrez-Cortez, Del López, & Rodríguez-García, 2015).

Reports of the production of a natural, intense red pigmented material, nocheztli/cochineal/grana by a female insect (*Dactylopius coccus*) living on some *Opuntia* cladode hosts, has shown great prospect for future exploration and commercialisation. The natural red pigment is known as carminic acid. Grana is harmless to humans and has a very compatible and stable colouring power, thus making it globally acceptable for textile, food and other applications (Tovar, Pando-Moreno, & Garza, 2005). Grana export value has been recorded at $60–$112 per kilogram of dry grana and demand is increasing worldwide. Grana could replace harmful and mutagenic synthetic red colourings currently used in foods, cosmetics and medicines. Studies involving greenhouse production conditions, and winter compared to summer production, have shown potential to increase dry weight yield of grana (Aldama-Aguilera, Llanderal-Cázares, Soto-Hernández, & Castillo-Márquez, 2005; Tovar et al., 2005). Its economic importance has increased with increase in *Opuntia* cladode species and the introduction of the insect into other non-native regions. However, more work still needs to be done to appreciably motivate for large scale production of grana in more regions of the world (Tovar et al., 2005).

6.1. *Effect of processing conditions on Opuntia cladode compound profile and bioactivities*

Cladode processing conditions affect the amount and types of compound present. Studies have shown that a combination of 45 °C and 3 ms^{-1} air flow were optimal for the preservation of phenol, flavonoid, ascorbic acid and β-carotene (Medina-Torres et al., 2011). Boiling processes affected compound preservation of vitamins and sugars whereas lipids, proteins and fibre were well preserved. Decreased antioxidant activity (38.9 to 31.6 trolox equivalent for raw and boiled respectively) was linked to phytochemical losses due to processing technologies (Ramírez-Moreno, Córdoba-Díaz, de Cortes Sánchez-Mata, Díez-Marqués, & Goñi, 2013). Cladodes gel consistency and viscosity were reduced by heat treatment and this impacted the cladodes capacity to accumulate glucose and control the glycaemic response (Ramírez-Moreno et al., 2013). Jaramillo-Flores et al. (2003) identified cladode carotenoids where β-carotene was present in significant amounts. The Atlixco variety contained low levels of carotenoids, while the Milpa Alta variety showed 84% retention after heat processing. In contrast, reduced amounts of carotenoids were reported in other studies (Ayadi et al., 2009). Lactic acid fermentation of cladode pulp by a *Lactobacillus brevis* strain produced significant amounts of γ-amino butyric acid when compared with other LAB strains (*Lactobacillus rossiae, Lactobacillus plantarum* and *Pediococcus pentosaceus*). This fermentation improved the antiradical, antioxidant and immunomodulatory properties of the cladode pulp which was attributed to increase in carotenoid and vitamin C contents. This enhancement of bioactivity was confirmed using Caco-2/TC7 cells and fermented pulp demonstrated significant inhibition of related prostaglandin production (Filannino et al., 2016).

7. *Opuntia* fruit/pulp related compounds and biological activities

Fruits are the succulent, ellipsoid and edible part of the *Opuntia* plant. They are usually about 7 cm long but can have a wide range of colours which include green, white reddish, purple and yellow (Heuzé & Tran, 2017).

One study has shown significant total amounts of vitamin C and polyphenol in *Opuntia humifusa* fruits compared to other *Opuntia* plant parts such as the root, cladodes and seeds (Hahm et al., 2015). Cactus pear fruits also contain significant quantities of minerals (Mabrouki, Zougari, Bendhifi, & Borgi, 2015), amino acids (Table 3) (Ammar et al., 2012), flavonoids (Hahm et al., 2015; Mabrouki et al., 2015; Osorio-Esquivel et al., 2011), and phenolic acids (e.g. ferulic acid, syringic acid and caffeic acid) (Cha et al., 2013; Osorio-Esquivel et al., 2011) (Table 2a). They also contain betalains such as betanin, isobetanin, betanidin, isobetanidin, and phyllocactin (Table 2b) (Osorio-Esquivel et al., 2011; Tesoriere, Allegra, Butera, & Livrea, 2004).

Betalains contribute to fruit colour and biological activity of the fruit pulp (Albano et al., 2015). Biological activity in betalain compounds have been linked to intrinsic structural features such as those

Table 3
Amino acids content (g/100 g) distribution in cactus (*O. ficus-indica*) plant parts

Amino acid	Cladode	Fruit	Seeds	References
Alanine	1.25	3.17	4.75	Feugang et al., 2006; Sawaya, Khalil, & Al-Mohammad, 1983
Arginine	5.01	1.11	6.63	
Asparagine	3.13	1.51	Trace	
Asparaginic acid	4.38	Trace	10.42	
Glutamic acid	5.43	2.40	21.68	
Glutamine	36.12	12.59	Trace	
Cystine	1.04	0.41	0.37	
Histidine	4.18	1.64	3.11	
Isoleucine	3.97	1.13	6.20	Ali, Alhaj, et al., 2014; El-Mostafa et al., 2014;
Leucine	2.71	0.75	9.94	
Lysine	5.22	0.63	6.79	Fernández-López et al., 2010;
Methionine	2.92	2.01	0.70	Feugang et al., 2006
Phenylalanine	3.55	0.85	5.25	Ali, Alhaj, et al., 2014
Serine	6.68	6.34	8.46	
Threonine	4.18	0.48	1.53	
Tyrosine	1.46	0.45	3.09	Ali, Alhaj, et al., 2014; Feugang et al., 2006
Tryptophane	1.04	0.46	Trace	
Valine	7.72	1.43	6.02	
α-Aminobutyric acid	Trace	0.04	Trace	
Carnosine	Trace	0.21	Trace	
Citrulline	Trace	0.59	Trace	Feugang et al., 2006; Sawaya et al., 1983
Ornithine	Trace	Trace	Trace	
Proline	Trace	46.00	Trace	
Taurine	Trace	15.79	Trace	Ali, Alhaj, et al., 2014; Fernández-López et al., 2010

attributed to the betalamic acid core of betalain strutures which seem to play a modified role in enhancing or reducing biological activity (Gandia-Herrero, Escribano, & García-Carmona, 2009). In a study by Cai, Sun, and Corke (2003), gomphrenin type betacyanins (EC_{50} = 3.7 μM) and betaxanthins (EC_{50} = 4.2 μM) demonstrated the strongest antioxidant activity, which was three times stronger than that for vitamin C (ascorbic acid) (EC_{50} = 13.9 μM). Antioxidant activity of the tested betalains also decreased in this order: simple gomphrenins > acylated gomphrenins > dopamine-betaxanthin > (S)-tryptophan-betaxanthin > 3-methoxytyramine-betaxanthin > betanin/isobetanin > celosianins > iresinins > amaranthine/iso-amaranthine (Cai et al., 2003). This proved that the number and position of hydroxyl groups and glycosylation in these pigmented compounds determine their antioxidative activites, as is the case with flavonoids, phenolic acids and other phenolic antioxidants found in *Opuntia* fruits. A similar study by Gandia-Herrero, Escribano, and Garia-Carmona (2016) supported this conclusion.

Opuntia fruit/pulp extracts possess antidiabetic, cardioprotective, neuroprotective, antiinflammatory, and hepatoprotective properties (Serra, Poejo, Matias, Bronze, & Duarte, 2013). Betanin in *Opuntia ficus-indica* fruits was reported to reduce chronic myeloid leukaemia cell spread (K562), and cell death was recorded at an inhibitory concentration (IC_{50}) of 40 mM betanin (Sreekanth et al., 2007). Cactus fruit ethyl acetate extracts containing flavonoids, trans taxifolin, and dihydrokaempferol also suppressed HeLa cervical carcinoma cell proliferation (at ≥100 μg/mL concentrations), while normal human BJ fibroblasts were unaffected, which suggests potential application as intervention for human cervical carcinoma management (Hahm et al., 2015). Taxifolin has been known to repress prostate cancer cell growth and inhibit HCT116 colorectal cancer cell growth (Woo et al., 2012; Zhang, Al Zaharna, Wong, Chiu, & Cheung, 2013). Reduction in the viability of hepatic (78.9–79.2%), prostate (61.2–75.2%) and colon (52.5–74.1%) cancer cells have been reported for prickly pear fruit coloured varieties - *Opuntia violaceae* (purple-red), *O. robusta* (green) and *O. rastrera* (purple) (Chavez-Santoscoy, Gutierrez-Uribe, & Serna-Saldívar, 2009). The antioxidant activity of prickly pear fruits is reported to be comparable with that of red oranges and grapes (Albano et al., 2015; Cano et al., 2017). Betalain antioxidant properties also evolve with fruit ripening. This has been demonstrated for *Opuntia megacantha* fruits of Argentinian origin (Cayupán, Ochoa, & Nazareno, 2011). *Opuntia ficus indica* juice rich in flavonoids and phenolic acid compounds also inhibited induced free radical chain reactions in rat erythrocytes (Alimi, Hfaiedh, Bouoni, Sakly, & Rhouma, 2013). Antioxidant activity have been reported for purified *Opuntia* fruit polysaccharides (Zhong, Jin, Lai, Lin, & Jiang, 2010). The molar ratio of the polysaccharide sugars differed from those reported earlier (Habibi, Heyraud, Mahrouz, & Vignon, 2004; Panico et al., 2007). The difference was attributed to different soil types/conditions and climates. Domestication gradients may also affect the content of bioactives and their biological activities (Astello-García et al., 2015).

Opuntia ficus-indica red, purple and orange-coloured fruits grown on agricultural drainage sediment were shown to possess improved and selenium-enriched chemotherapeutic properties, compared to those grown on normal soil (Bañuelos et al., 2012). Purple cultivars such as the Mexican semi-domesticated *Opuntia robusta* showed the highest total betalain, phenol, carotenoid and vitamin C contents, but with low antioxidant activity compared to other varieties. Yellow commercial *O. albi-carpa* variety had no betalains, low total phenolic content and low antioxidant activity (Yahia & Mondragon-Jacobo, 2011), which suggests the importance of these compounds as antioxidants. However, in some studies phenol content did not always correlate with good antioxidant activity (Chavez-Santoscoy et al., 2009).

Reports on *O. ficus-indica* fruits have shown major variation in total phenolics content. Phenol content of approximately 172 mg/kg (Chavez-Santoscoy et al., 2009) and 452 mg/kg (Díaz-Medina, Rodríguez-Rodríguez & Díaz-Romero, 2007) have been reported for juice from *Opuntia* spp. Higher levels of phenolic content have also been recorded in juice (Galati et al., 2003) and fruit pulp (Moussa-Ayoub, El-Samahy, Rohn, & Kroh, 2011). Moussa-Ayoub, El-Samahy, Rohn, and Kroh (2011) reported higher antioxidant activity for *O. macrorhiza* pulp compared to whole *O. ficus-indica* fruits. In other studies, the antioxidant activity and vitamin C content varied between different fruit samples. Purple *Opuntia ficus-indica* fruit varieties usually showed the highest antiradical ability (Bañuelos et al., 2012; Cayupán et al., 2011). However, Sumaya-Martinez et al. (2011) showed that antioxidant activities of differently coloured fruit cultivars (white and yellow) did not show great variance but had reduced activities compared to the purple and red variants. On the contrary, red cactus pears from Mexico had the highest ascorbic acid content/antioxidant activity compared to the purple variety (Abdel-Hameed et al., 2014; Stintzing et al., 2005). Nevertheless, the presence of betaxanthin in coloured cultivars was; purple > red > yellow > white pear (Stintzing et al., 2005).

In analysing fruit and fruit juice made from Moroccan *Opuntia megacantha*, El Kharrassi, Mazri, Benyahia, Benaouda, and Nasser (2016) showed that *Opuntia* fruit skin colour did not correlate with ascorbic acid concentration (Gurrieri et al., 2000; Kuti, 2004). The differences in these results were attributed to genotype, growth location and environmental contributors affecting the vitamin C content of the *Opuntia* cultivars (Gurrieri et al., 2000). However, in *Opuntia* variants studied, ascorbic acid concentrations were highest in red fruits than in yellow fruits (Kuti, 2004). *O. streptacantha* (red skinned cactus pear) carotenoid content was lower (14.6 mg/g fresh weight) than *O. stricta var. stricta* (yellow-skinned cactus pear) (23.7 mg/g fresh weight) (Kuti, 2004). Current research has shown that it is possible to concentrate the pigments and other bioactive fruit components such as betalains during ripening, compared to the very immature and under ripe cultivars days after anthesis (Pinedo-Espinoza et al., 2017).

A novel, neutral, and water-soluble (1 → 4)-α-d-glucan polysaccharide from *Opuntia ficus-indica* aqueous fruit extracts (from Ejfarah region of Libya) was reported by Ishurd et al. (2010). The polysaccharide was identified by gel-permeation and anion-exchange chromatographic techniques. A sincocin product [developed from extract mixtures from prickly pear fruit (*Opuntia engelmannii lindheimeri*), sumac *Rhus aromatica*, southern red oak and mangrove *Rhizophora mangle*] showed promise as an antinematodal product. While sincocin

was not effective in its anti-nematodal activity against *Meloidogyne incognita* and *Radopholus similis* compared to other chemical treatments (Chitwood, 2002; Sipes & Delate, 1996), it was an effective control of citrus, reniform and cyst nematodes (Banaszak & Jagusz, 1999).

In general, the biological capacities and nutraceutical benefits of *Opuntia* fruit species may be due to the synergistic effects of betalains, flavonoids and other biologically active components present (Stintzing et al., 2005). The highlighted applications suggest that *Opuntia* fruit/extracts have potential for industrial application in the food, agricultural and nutraceutical industries.

8. *Opuntia* root compounds and biological activity

The *Opuntia plant* has a tap root-system with extensive horizontal/lateral roots which emerge in response to rainfall and abscise as soil dries (Heuzé & Tran, 2017). Roots occur at the upper layer of the soil where they function for efficient water and minerals absorption. A low root: shoot ratio (in terms of total plant biomass) is also observed for *Opuntia* species (Snyman, 2006). Roots are metabolically active but unexplored (Bais, Loyola-Vargas, Flores, & Vivanco, 2001). They are used in traditional pharmacopoeia and exude a wide variety of compounds into the rhizosphere. Some health benefits are associated with chemical compounds from plant roots (Flores, Vivanco, & Loyola-Vargas, 1999). For example, flavonoid and phenol-rich (57.56 mg GAE/g) *Opuntia ficus-indica* root extract helped to sustain the levels of *in vivo* antioxidant enzymes defence systems in rats and showed an 80% reduction in ethanol induced-ulcer lesion (up to 92.59% reduction in ulcer lesions) with gradual increase in dosage (Alimi et al., 2010). This phenol content was higher than that reported for swallow root (*Decalepis hamiltonii*) extract (34 mg GAE/g) which also showed anti-ulcerogenic activity (Naik, Smitha, Harish Nayaka, & Lakshman, 2007).

Root extracts showed moderate antioxidant activity. Aromadendrin rich ethyl acetate prickly pear root fraction has been effective in preventing diabetes by aldose reductase activity inhibition, glycation end products synthesis and α-glucosidase activity inhibition (Jeon et al., 2011). Polysaccharides-rich *Opuntia ficus-indica* root extracts also enhanced gastric healing, reduced gastric juice output and increased stomach mucus production. These antiulcerogenic activity (82.69 to 93.1% curative ratio after five and fifteen days extract treatment of Wistar rats at 400 mg/kg body weight) as well as other bioactivies mentioned, were due to synergism of antioxidant, anti-excretory and healing mechanisms (Alimi et al., 2013). However, research is still required to increase database on the knowledge of root-associated compounds and their biological activities, as well as studies into the dynamics of the *Opunita* plant root system distribution and development.

9. Techniques for the extraction of compounds from *Opuntia* residue

Pomaces/residues have been ignored as source of viable phytochemicals which are usually not released from plant matrices (Durazzo, Casale, Melini, Maiani, & Acquistucci, 2016; Gonzales et al., 2015). Therefore, extraction/separation techniques have been developed and utilized to characterize essentially valuable components from *Opuntia* residues which are not released from conventional extractive processes. Separation by adsorption using synthetic resins is usually applied for polyphenols recovery from plant crude extracts. This process is valuable in the food industry (Soto, Moure, Domingues, & Parajó, 2011). Low operation costs is one of its advantages as well as high adsorption capacities for different classes of compounds and simplicity in handling (Soto et al., 2011). Antioxidant and antiproliferative compounds (e.g. polyphenols and anthocyanins) have been recovered from apples and cherries through this process (Bobe, Wang, Seeram, Nair, & Bourquin, 2006; Schaefer, Baum, Eisenbrand, & Janzowski, 2006).

The use of alkaline hydrogen peroxide treatment (Vilela, Leão, Franca, & Oliveira, 2016); acid and alkaline hydrolysis and/or sonication; butanolysis and phloroglucinolysis (Arranz et al., 2009; Tarascou et al., 2010) have been efficient in the release of residue/fibre-bound antioxidants (macromolecular antioxidants - MA), also known as antioxidant dietary fibres (ADF) (Durazzo et al., 2016). Flavonols undergo degradation (to protocatechuic acid) when subjected to acidic hydrolysis (Moussa-Ayoub, El-Samahy, Rohn, & Kroh, 2011), and the extent of degradation depends on the concentration of acid utilized, nature of flavonol or plant material as well as hydrolytic temperature and treatment time (Häkkinen & Törrönen, 2000; Tura & Robards, 2002). Enzymatic hydrolysis has however been shown to be milder and more sensitive and yielded significant amounts of quercetin flavonol, and kaempferol compared to acidic hydrolysis. It also released undegraded sugars from flavonol glycosides to yield its corresponding aglycon (Bilyk, Cooper, & Sapers, 1984).

9.1. *Opuntia macromolecular antioxidant biological activities*

Macromolecular antioxidants (MA) are high molecular weight antioxidant compounds when compared with extractable phenols, and exhibit significant biological activites (Arranz et al., 2009; Cardador-Martínez, Jiménez-Martínez, & Sandoval, 2011). They are made up of hydrolysable polyphenols, tannins (hydrolysable and condensed tannins) (Cardador-Martínez et al., 2011), single phenolic acids and polymeric proanthocyanidins (Pérez-Jiménez & Saura-Calixto, 2015). MA are usually determined in non-extractable polyphenol (NEPP) and non-extractable proanthocyanidins (NEPAC) and expressed in acidified methanol and HCl/butanol (butanolysis) fractions, respectively. The depolymerised hydrolysates are then utilized in quantitative assays (Cheng, Xiangyan, Wenliang, Zhiqing, & Lina, 2013; Durazzo et al., 2016). *In vivo*, MAs are hypothesized to occur mainly in the colon where microbes ferment them to produce absorbable metabolites. MA exhibit different health-related properties which include antiproliferative, antioxidant, and gene expression modification (Tarascou et al., 2010). MA have been reported to constitute up to 50–100% of some fruits studied, and showed remarkable biological activities (Arranz et al., 2009; Tarascou et al., 2010).

Natural anticancer ingredients (ferulic acid, isorhamnetin derivatives, betalains) have been isolated from *Opuntia* fruit residues after hydroalcoholic solvents extraction and separation with macroporous resins (Amberlite XAD16 resin) (Serra et al., 2013). Juice residues from *O. robusta* and *O. ficus-indica* wild fruits from Portugal contained useful compounds which were responsible for inhibition of human colon carcinoma HT29 cell growth (ED_{50} of 4.6–6 mg/mL after 72 h of proliferation). Residue extracts resulted in an increase in reactive oxygen species (ROS) in carcinoma cells, indicating that the prooxidant effect of constituent compounds may induce cancer cell death (Serra et al., 2013). The combination of an extractive and resin concentration methods can be used to concentrate bioactive compounds for potential application in pharmarceutical and food industries. The antiproliferative effect was, however, slightly lower than those described by Hahm, Park, and Son (2010) using highly fractionated *O. humifusa* extracts (hexane, ethyl acetate extracts and water partitioned fractions) in a more sensitive cancer cell line (human glioblastoma cell line-U87MG cells). Compared to doxorubicin (a known anticancer compound), earlier studies on *Opuntia* extracts showed reduced inhibition of cancer cell proliferation. Nevertheless, a combination of these extracts with doxorubicin could improve anticancer efficacy and reduce chemoresistance (Serra et al., 2011).

Research has reported high antioxidant activity of MA extracts (52.22 ± 1.07 μmol trolox equivalent/g) even in the absence of some cladode MA compounds such as hydrolysable tannins and proanthocyanidins (Bensadón et al., 2010). Hydrolysed cladode extracts were more effective radical scavengers than the unhydrolysed fractions (Avila-Nava et al., 2014). Acid hydrolysis released polyphenols bound to or associated with the insoluble fibre as glycosides (Avila-Nava et al., 2014; Ginestra et al., 2009). Greek cactus pear fruit seed residue extract

also showed good antioxidant activity (69.5–95.1 mmol/kg) (Zakynthinos & Varzakas, 2016).

Other biological/gastrointestinal activities of antioxidants from non-*Opuntia* residues include antiinflammatory activity of grape proanthocyanidins (Tomás-Barberán & Andrés-Lacueva, 2012), and pomegranate ellagitannins (Larrosa et al., 2010). Non-*Opuntia* residue antioxidants also have beneficial microbial interactions [Cocoa flavanols (Tzounis et al., 2008)]; and antioxidant effects, that is, they inhibit oxidation of cholesterol and lipids (MUFA and PUFA), and proteins and vitamins (Tomás-Barberán & Andrés-Lacueva, 2012). They also interact with α-glucosidase, lipase and alpha-amylase enzymes (Tomás-Barberán & Andrés-Lacueva, 2012). Biological activities of *Opuntia* MAs could have potential applications as pill and capsule supplements; components of cosmetics, food ingredients (prebiotics etc.) and drug excipients. More research into MAs may provide new data on novel antioxidant compounds and their applications (Amoo et al., 2014; Aremu et al., 2014; Pérez-Jiménez, & Dı´az-Rubio, M. E., & Saura-Calixto, F., 2013). MA analyses and quantification in more *Opuntia*/plant species are still required in the compilation of a broader database. This would be useful in guiding future biological and nutritional research (Arranz et al., 2009).

10. Debate on the presence of taurine amino acid in the *Opuntia* plant

Taurine is an amino acid whose presence has been recorded in *Opuntia* species (Chauhan, Sheth, Jivani, Rathod, & Shah, 2010; McCarty, 1999; Piga, 2004; Stintzing & Carle, 2005). It is mainly found in seafoods, dairy and meat products, fungi and algae among others. Hence it is considered an unusual plant component. However, considerable quantities (323.6–572 mg/L) were observed in South African and Mexican *Opuntia* variants using reversed phase high performance liquid chromatographic (RP-HPLC) analysis with o-phthaldialdehyde (OPA) adduct/thiol identification method (Stintzing, Schieber, & Carle, 1999). Lower levels (8.0–11.2 mg/100 g) of taurine were reported by Tesoriere, Fazzari, Allegra, and Livrea (2005). Taurine content of species of yellow Sicilian fruit pulp and red-skinned cactus pear fruits from Spain has been reported to be within the range of 6.80–11.96 mg/100 g of fresh fruit tested (Fernández-López, Almela, Obón, & Castellar, 2010; Tesoriere et al., 2005). Studies have shown that the pulp of red and white cultivars contained lower levels of taurine (Stintzing et al., 1999; Tesoriere et al., 2005).

Despite earlier research results, recent research utilizing automated ion-exchange chromatography (IEC) reported the absence of taurine in commercially available juices and in different cultivars (red, white, orange) of prickly pears from South Africa, Italy and the Near East (Ali, Alhaj, Al-Khalifa, & Brückner, 2014; Ali, Al-Khalifa, & Brückner, 2014). Similary, in an earlier study, no taurine was detected in *Opuntia* fruits sourced from Egypt (Askar & El-Samahy, 1981). Instead considerable amounts of γ-aminobutyric acid (GABA) as opposed to taurine, were reported (Ali, Alhaj, et al., 2014; Askar & El-Samahy, 1981). GABA and proline amino acids were predominant in all the fruits tested (Ali, Alhaj, et al., 2014). It is relevant to note that amino acid assignments for IEC (taurine as an exception) were in line with HPLC data using OPA/thiol and gas chromatographic/mass spectrometric (GC-MS) techniques (Kugler, Graneis, Schreiter, Stintzing, & Carle, 2006; Stintzing et al., 1999). In more noteworthy studies, analysis of fresh plum juices by van Gorsel, Li, Kerbel, Smits, and Kader (1992) using IEC also proved taurine absence. However, aqueous extracts from the dehydrated plums were reported to contain low quantities of taurine, but retention time was used in the amino acid assignments. It is possible that the other analytical methods employed (OPA/thiol) may have affected results on *Opuntia* amino acid profiles (El-Mostafa et al., 2014).

Therefore, there seems to be a lack of convincing data on taurine presence in nutritional fruits/fruit juices. Recent studies suggest the use of taurine standard and more uniform or sensitive methods of analysis to increase accuracy of taurine content reports in *Opuntia* species and other fruits/plants. However, a number of other amino acids have been reported to be associated with various *Opuntia* species including alanine, methionine, valine, proline, and carnosine (Table 3).

Taurine is used in blood pressure regulation, as an ingredient in energy drinks (Wójcik, Koenig, Zeleniuch-Jacquote, Costa, & Chen, 2010). Its antioxidative activity has also been reported (Devamanoharan, Ali, & Varma, 1998; Wu, Wang, Fennessy, Redmond, & Bouchier-Hayes, 1999). Taurine is also suspected to be associated with modulating inflammatory responses (McCarty, 1999).

11. Potential side effects of *Opuntia* species extracts/compounds

Little information is available on *Opuntia* spp. toxicology and on adverse side effects of its compounds or extracts. However, from literature a low colonic obstruction has been attributed to the consumption of *Opuntia ficus-indica* seeds (Kleiner, Cohen, & Mares, 2002). Nausea, mild diarrhoea, increased stool volume and frequency, headaches and abdominal bloating, have also been reported in books on traditional folk medicine and case reports (De Smet, 2002; Gagnier, DeMelo, Boon, Rochon, & Bombardier, 2006). These effects were, however, not corroborated with results from scientific research. Although some health care professionals believe that herbal medicines are relatively safe because they are “natural”, there is little supporting data to back such an assumption. Although herbal mixtures may contain certain contaminants from any stage of the extraction/production process, side effects may also be linked to plant components and human factors (intrinsic) and non-plant related contaminant factors (extrinsic) (Gagnier et al., 2006).

There are possible indications of extreme sensitivity to any component of *Opuntia* as individual pharmacokinetics vary widely. Dermatitis has been reported to be a common adverse reaction to prickly pears (Pawar et al., 2017). The presence of opportunistic pathogens/microorganisms or microbial toxins could also affect the content of active components in herbal mixtures and result in adverse health effects. Therefore, plant-derived herbal interventions need to be treated with care, or as containing potentially toxic chemical compounds until proven otherwise. Some of these toxic chemical hazards have been listed in the Maryland National Library of Medicine (NLM) database for harmful substances (Osuna-Martínez et al., 2014).

12. *Opuntia* plant compounds and search for new antimicrobials

The rise of new infectious diseases and increased bacterial resistance to available antimicrobials, have necessitated plant-related research channelled towards identifying novel compounds with antimicrobial activity (Aremu et al., 2011; Silver & Bostian, 1993; Valtierra-Rodríguez et al., 2010). Antimicrobial compounds belong in a range of phenolic and non-phenolic classes such as betalains, polyphenols and phenolic acids (caffeic, cinnamic, catechol), quinones, flavones, flavonoids, flavonols, tannins, coumarins, lectins and polypeptides, alkaloids, terpenoids, essential oils, polyamines (spermidine), isothiocyanates, thiosulfinates, glucosides, polyacetylenes and acetylene compounds (Ciocan & Bara, 2007; Tapiero, Tew, Ba, & Mathe, 2002). The antimicrobial activities of *Opuntia* species extracts are attributed to the presence of quite a number of these compounds.

Bactericidal activity has been reported for Cardon Blanco (*O. streptacantha*), Jalpa and Real de Catorce (*O. ficus-indica*) nopal cacti cultivars against *Campylobacter jejuni* (1.1–12.5 mg/ml), *Clostridium perfringens* (0.8–16 mg/ml) and *Vibrio cholera* (4.4–30 mg/ml). The activities were partly ascribed to the significant amounts of total flavonoids and phenols present in the cultivars (Sánchez et al., 2014). *Opuntia ficus-indica* extracts in a wide array of solvents have shown activity against different bacterial strains such as *Staphylococcus aureus, Pseudomonas aeruginosa, Escherichia coli, Salmonella* spp., *Klebsiella pneumoniae, Citrobacter freundii* and *Streptococcus pneumoniae* (Shafiei,

Kariminik, & Hasanabadi, 2013; Wasnik & Tumane, 2016), and against *Bacillus subtilis* (Gnanakalai & Gopal, 2016). Terpenoids, glycosides, saponins, alkaloids and flavonoids were identified in the extracts. Acetone extract compared to n-hexane and petroleum ether extracts showed better antimicrobial activity (Wasnik & Tumane, 2016), while aqueous extracts of both stem and fruit (Gnanakalai & Gopal, 2016) showed the least antimicrobial activity which could be attributed to poor solubility of bioactive components in extraction solvents.

Opuntia flower extracts also showed *in vitro* antibacterial activity against *P. aeruginosa, S. aureus*, *B. subtilis*, and *E. coli*, and antifungal activity against *Candida lipolytica* and *Aspergillus niger*. Most important was the antibacterial activity which makes the extracts potentially suitable for food industry applications, for example, as food additives or preservatives (Ennouri, Ammar, Khemakhem, & Attia, 2014). Extracts of *Opuntia matudae* containing betalains inhibited the growth rate of *E. coli* O157:H7 (Hayek & Ibrahim, 2012). In addition, Ammar et al. (2015) reported marked antibacterial action (minimum inhibitory concentration - MIC of 25 ± 1.8 mm) of *Opuntia* flower methanol extract on *Listeria monocytogenes*.

Recently, first report of a 28.3 kDa protein with RNase activity purified from *Opuntia* cladode aqueous extract demonstrated antiviral action against cucumber and tobacco mosaic viruses (CMV and TMV) (MIC of 40 mg/mL). Antiviral activity was linked to interaction of the protein with RNase activity with viral nucleic acid (Rasoulpour, Afsharifar, Izadpanah, & Aminlari, 2017).

Broad spectrum antimicrobial activity of betalain-rich extracts against *Bacillus cereus*, *L. monocytogenes*, *Proteus vulgaris*, *Yersinia enterocolitica* and *Enterobacter aerogenes*, as well as against *Rhizoctonia solani* and *Candida albicans* yeasts (125–250 mg/ml), *Fusarium oxysporum* and *Aspergillus flavus* moulds (500 mg/ml), have also been reported (Tenore, Novellino, & Basile, 2012). The broad spectrum activity of the extracts was attributed to the adverse effect of bioactive components on microbial cell membrane integrity, function and structure (Canadanovic-Brunet et al., 2011). *Opuntia stricta* essential oil extracts containing mainly thymol (42.7%) also showed antimicrobial activity against important food and environment borne opportunistic bacterial pathogens such as *E. coli*, *B. cereus, P. aeruginosa* (1.25–2.5 mg/ml MICs) and yeast such as *C. albicans* (2.5 mg/ml MIC). The extracts of the *Opuntia* plant therefore have potential for commercialisation as novel drugs for use in antimicrobial therapy (Moosazadeh, Akhgar, & Kariminik, 2014).

Most plant compounds with antimicrobial properties are aromatic organic molecules (Ciocan & Bara, 2007). Phenols and flavonoids are aromatic, hydroxylated biologically active compounds. Presence of either phenols or aldehyde compounds in extracts reportedly show highest antibacterial activities compared to those containing terpene alcohols (Koubaa, Ktata, Bouaziz, et al., 2015; Naveed et al., 2013). The number and site of the hydroxyl groups on the aromatic molecules determine antimicrobial action. Usually the more hydroxyl groups present, the better the antimicrobial effect. Antimicrobial quinone compounds bind to extracellular and soluble proteins and disrupt the integrity of microbial cell membranes/wall to result in static or cidal effects. Similar antimicrobial mechanism is observed in coumarins where benzene and α-pyrone rings are bound together. Higher antimicrobial action was reported for polymeric phenols (with multiple number of hydroxyl groups) such as hydrolysable and condensed tannins which act by binding cell walls, inactivating microbial adhesins, enzymes, cell envelope and transport proteins, thereby preventing growth and protease activity (Ciocan & Bara, 2007).

13. Conclusion and directions for future research

Opuntia species contain a wide array of phenolic and non-phenolic constituents which singly or synergistically exert biological activities. This review discussed biological activities of compounds and extracts from cactus (*Opuntia*) plant parts. The taurine content, macromolecular antioxidants, and antimicrobial activity of *Opuntia* plants were also discussed. Data on MA fractions and biological activities are on the increase and will be important in the future. *Opuntia* extracts have potential for treatment of some chronic diseases such as cancer. Cactus compounds and/or extracts have also found their use as natural and alternative sources of food supplements, and have a wide range of applications for the food and other industries. However, more information is still required on the effective concentrations of known and novel *Opuntia* compounds. *Opuntia* taurine content reports also need uniformity regarding assay methods used to differentiate it from GABA. *Opuntia* plant metabolism/processing methods need to evolve for improved expression and optimization of bioactive compounds in extracts. In addition, more sophisticated methods are required for structural elucidation of potentially new and unidentified *Opuntia* compounds which may possess novel applications. Novel antibiotic agents to tackle antimicrobial resistance are yet to be developed and their mechanism(s) of action need to be studied. More focused research is required to add information on *Opuntia* extracts/herbal mixtures toxicology and their mechanism(s) of action to safeguard health. In the future, *Opuntia* will remain an interminable source of products with functions for food and other industries. Its capacity to contribute to subsistent and global food security, as well as to the health sector, cannot be overemphasized.

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Conflicts of interest

The authors of this manuscript declare that no conflict of interest exists.

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