REVIEW

Functional properties and technological applications of vegetables leaves

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Summary

Recently, there has been a global interest in natural sources of phytochemicals as antioxidants and functional food ingredients. Phenolic compounds form a large group of secondary metabolites and the presence of these compounds in vegetable leaves has been related to sensory and health-promoting properties. Considering that a major part of vegetable leaves is discarded in spite of its high nutritional value, this literature review aimed to group the most recent studies, which evaluated the therapeutic and functional aspects and the technological applications of leaves. Moreover, the roles of phenolic compounds in the functional properties and in the sensory characteristics are also discussed. Studies evaluating the main technologies in the reuse of vegetable leaves are presented as well. Finally, the main challenges of the use of the leaves in the food industry are discussed. The search for new applications of by-products is an alternative to valorize these materials, in order to provide new opportunities of use for the food and pharmaceutical industries.

Keywords

bioactive compound; by-product; leaf; technological applications; toxicology

Phenolic compounds form a large group of bioactive chemicals of different biological functions. These compounds are secondary metabolites of plants and comprise a wide variety of molecules that have a polyphenol structure, i.e. contain several hydroxyl groups on aromatic rings [1, 2].

Vegetable leaves have been recognized as potential sources of phenolic compounds, with high bioavailability, compared to many marketable leafy vegetables, such as brassicas and lettuce [3]. These compounds present antioxidant properties, acting as reducing or metal-chelating agents, hydrogen donors and singlet oxygen quenchers. For this reason, they are related to healthy diets and to the prevention of several chronic diseases [4].

Leaves are considered alternative sources of vegetables due to their resistance, tolerance and production yield [5, 6]. Moreover, they are used to increase the nutritional value and help in the malnutrition treatment [7]. In addition to being used dehydrated in forms of teas (dried) [8], leaves have also been used as flours (dried and powdered) [9], composing bakery products [10], beverages [11], and even as matrices for extraction of bioactive compounds [11, 12]. The phytochemicals isolated from leaves have been used in pharmaceutical industry and also as natural additives to preserve and/or enrich food products in food industry [13].

Therefore, the knowledge on various phenolic compounds and other nutrients may contribute to the development of functional foods or pharmaceutical products using leaves from different oleraceous as natural ingredients [14]. Functional properties of these materials can be evaluated by the determination of some parameters such as antioxidant capacity, contents of phenolic compounds, as well as antihypertensive and antihyperglycemic activities [15–17]. This work conducts an overview about the main phenolic compounds present in vegetable leaves and discusses the health-promoting and technological properties of these compounds in leaves. For this purpose, the main therapeutic effects, functional properties and the technological feasibility are discussed, as well

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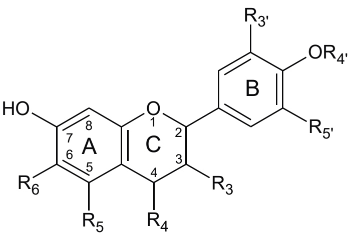

Fig. 1. Basic structure of flavonoids [20].

as the challenges for the use of leaves in the food industry and in the human diet.

Phenolic compounds in vegetables

The consumption of leaves was shown to be an alternative for combating malnutrition in the human diet and in the animal feed [18]. Furthermore, these materials are sustainable sources of bioactive compounds (i.e. polyphenolic compounds) that could be used for different applications such as food additives, functional ingredients and nutraceuticals [19].

Polyphenolic compounds are a large group of secondary metabolites widely distributed in plants, divided into two major subgroups: flavonoids and phenolic acids [20]. Flavonoids (Fig. 1) comprise a group of over 4000 aromatic plant compounds, among which anthocyanins, proanthocyanidins, flavonols and catechins are the major ones (Fig. 2). They are low molecular weight compounds, which chemical structure consists of two aromatic rings, called ring A and B, joined by three carbons forming a heterocyclic ring, called ring C [22].

Phenolic acids (Fig. 3) are characterized by a benzene ring, a carboxylic grouping and one or more groups of hydroxyl and/ or methoxyl in the molecule, responsible for antioxidant capacity in vegetables [24]. They include hydroxycinnamic acids (e.g. caffeic or ferulic acid conjugates, sinapic acid) and hydroxybenzoic acids (e.g. benzoic, gentisic or *p*-anisic acids) [3, 25, 26]. These compounds are important in pharmaceutical and food industries and provide health benefits for human, acting as antioxidants in vegetables [27, 28]. This activity is associated with high adsorption and neutralization ability of free radicals, which extinguishes singlet and triplet oxygen, or peroxides in decomposition [29].

Furthermore, phenolics are important and decisive for sensory and nutritional quality of fruits, vegetables and other plants [30], and may also affect positively or negatively the characteristics of food with impacts on colour, flavour and astringency [31]. For example, polyphenolic compounds (mainly tannins) form complexes with salivary proteins, and this interaction plays an important role

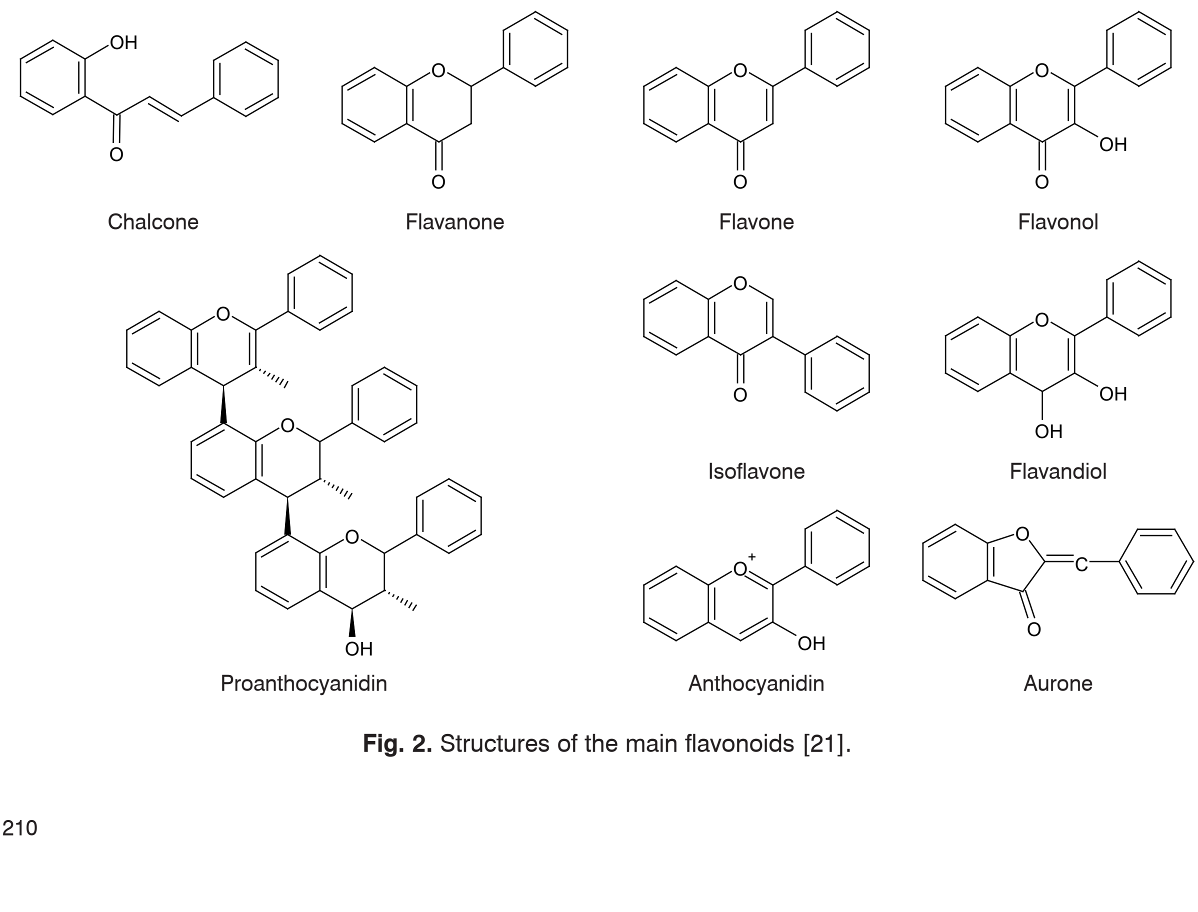

Fig. 2. Structures of the main flavonoids [21].

in the sensation of astringency, due to delubrication of oral surfaces [28, 32]. This interaction may form a layer acting as a water barrier and produces a mouth-drying sensation [33].

Particular structures including pigments (yellow, orange, red and blue), as anthocyanins, yellow flavanols and flavones are involved in food flavour [28]. In food processing, the conversion of anthocyanins to other compounds may decrease the stability of colour and increases their variety. Besides these characteristics, there are volatile polyphenols, as vanillin and eugenol, which provide strong odour to foods and are related to bitterness and adstringency [28].

Vegetable leaves and their functional properties and therapeutic effects

Several studies suggested therapeutic and functional effects of leaves of fruits and vegetables (Tab. 1). The health-promoting properties of leaves are related to the presence of phenolic compounds, which have attention due to their antioxidant properties [31]. Epidemiological studies revealed that these compounds provide significant protection against the development of several oxidative stress-associated diseases such as cardiovascular diseases, cancer, diabetes, infections, aging and asthma. Their antioxidant function is due to different mechanisms of action including inhibition of free radical formation, inhibition of free radical chain reaction, chelating free radical-producing metal ions and reducing the localized O^2 concentration by quenching O^{2-} radicals [43, 44].

Sweet potato leaves were reported to have a high content of total phenolic compounds, compared to major commercial leafy vegetables (Tab. 2), with emphasis on derivatives of caffeoylquinic [6] and chlorogenic acids [25] (Fig. 4). Yacon leaves were related to several phytochemical activities, such as antimicrobial and anti-inflammatory actions [34] and are associated with a considerable content of phenolic compounds, among which caffeic acid and gallic acid (Fig. 4) stand out as major phenolics [47].

The therapeutic benefits attributed to pitanga leaves are related to the presence of total polyphenols and total flavonoids [48]. Oszmiansky and Wojdyło [49] classified green and yellow leaves of cherries as sources of phenolics, especially polymeric procyanidins and chlorogenic acid. Extracts of strawberry leaves showed to be dominated by gallic acid derivatives and flavonol derivatives, which seem to be quite promising as antioxidants and anti-proliferative agents [50]. High contents of phenolic compounds were identified in leaves of blueberries [16], oranges [51] and pomegranate [37]. Papaya leaves are also known to contain a large number of biologically active metabolites [17] such as flavonoids and phenolic acids [52]. Rana et al. [11] reported that phenolics from apple leaves can be used for the treatment of inflammatory diseases and pathogenic infections. Persimmon leaves contain quercetin, kaempferol and their glycosides as the main flavonoids [53], and have been traditionally used for increasing cognitive function associated with the regulation of the antioxidative defense system [54] and for preventing chronic diseases [55]. Quercetin was shown to be an important polyphenol in guava leaves [38, 56, 57], related to antioxidant, antibacterial [58] and antifungal activities [59].

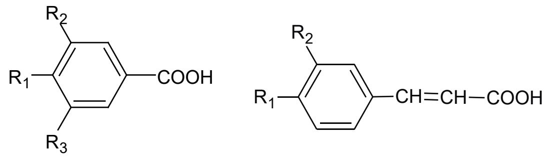

Fig. 3. Basic structures of phenolic acids [23].

Moringa oleifera leaves have been considered as excellent sources of natural antioxidants due to the content of several kinds of polyphenolic compounds [60] such as phenolics, flavonoids and flavonols [61]. Phenolic compounds such as chlorogenic acid, isoquercetin and astragalin were identified as major phytochemicals and, at a concentration of 100 $\mu g \cdot ml^{-1}$, aqueous and alcoholic extracts of *M. oleifera* leaves could significantly reduce relative amount of intracellular reactive oxygen species [41]. Leaves of moringa were also found to possess antifungal properties [62]. Four different polar extracts of soya leaves were reported to contain seven phenolic compounds, from which coumestrol was dominant and correlated to α-glucosidase inhibitory properties [63]. Leaves from olives [64] and cabbage [65] were reported to contain polyphenols and possess antioxidant potential. Some studies also revealed the significant potential of eucalyptus leaves as a source of phenolics and antioxidants, as well as an antifungal agent [66].

Moreover, leaves of several vegetable crops provide other important nutrients for human health. For example, leaves of sweet potatoes contain relevant levels of dietary fibre, carotenoids, vitamins and minerals [67]. Furthermore, they are important in the prevention of hypertension and/or atherosclerosis [68].

Various underutilized plant materials, such as vegetable by-products, have become attractive due to contents of certain compounds with biological

Tab. 1. Studies on functional properties and therapeutic effects of leaves.

Material	Product	Concentration/Content	Functional properties and therapeutic effects	Reference
Smallantus sonchifolius (yacon) leaves	Aqueous, leaf rinse and polar extracts	0.25–1.00 μg·ml^{-1}	In vitro anti-inflammatory activity	[34]
Ficus glumosa (fig) leaves	Aqueous extracts of fresh samples	225.3–375.0 mg·kg^{-1}	Hypolipidemic and anti-atherosclerotic effects, which could explain the use of the plant by traditional healers in the treatment of hypertension, cardiovascular diseases and diabetes	[35]
Ribes nigrum (blackurrant) leaves	Extracts of fresh samples	0.05–0.50 μg·ml^{-1}	Protection of the organism against oxidative stress, preventing the development of dangerous diseases	[36]
Punica granatum (pomegranate) leaves	Extracts of four different solvents	19.50–669.35 μg·ml^{-1}	Antioxidant activities, related to antiparasitic and antimicrobial effects	[37]
Psidium guajava (guava) leaves	Extracts of dried samples	100 μg·ml^{-1}	Antioxidant mechanisms of extracts attributed to their free radical-scavenging ability	[38]
		30 μg·ml^{-1}	Decrease in prostraglandin E_2 production in lipopolysaccharides-induced RAW264.7, providing a crude extract with high anti-inflammatory activity	[39]
Moringa oleifera (moringa) leaves	Extracts of dried samples	600 mg·kg^{-1} daily for 12 weeks	Amelioration of genes expression, paralleled by a reduction in body weight and improvement of the atherogenic and coronary artery index, as well as glucose level and insulin resistance of obese rats	[40, 41]
Chenopodium quinoa (quinoa) leaves	Different aqueous extracts obtained by squeezing, decoction of fresh and dried leaves, maceration of fresh and dried leaves, percolation of dried material and Soxhlet extraction of dried leaves	100 μg·ml^{-1}	Significant reduction of intracellular reactive oxygen species, which means that these leaves present high antioxidant capacity	[42]

activities [69, 70]. Several pharmaceutical studies report that polyphenols from sweet potato leaves present health-promoting biological activities [27] such as anti-carcinogenesis (determined by growth suppression of cancer cells by caffeoylquinic acids), anti-atherogenic and cardiovascular (related to antioxidant mechanisms) and antimicrobial [71, 72].

Leaves of spices have been also related to several functional properties. Uliana et al. [73] described Brazilian rose pepper leaves as a source of antibacterial substances, such as phenolics, according to solvent extraction.

In addition to all related functional properties of leaves, recent studies were conducted in order to evaluate anti-hyperglycemic and anti-hypertensive activity of leaves. These properties are justified by the abundance of phenolic compounds in leaves, in general. [3]. These substances exhibit an expressive association with the inhibition mechanisms of enzymes responsible for increasing of glucose contents and blood pressure by mechanisms still poorly understood [74].

Despite the effectiveness of various substances used for diabetes control, there is a growing demand to promote the use of natural products to the management of this disorder and its complications [75]. Recently, it was shown that polyphenols have a potential to contribute in the management of type 2 diabetes [76] because they are responsible for inhibition of α-amylase and α-glucosidase [70, 77], reducing the postprandial increase of blood glucose and, in this way, their use can be a strategy in the blood glucose control [78]. However, according to Kwon et al. [79], natural inhibitors of these enzymes from plants show lower inhibitory effects in α-amylase activity when compared to α-glucosidase. These authors suggest that this could represent an effective therapy for postprandial hyperglycemia.

Tab. 2. Content of phenolic compounds in vegetable leaves.

Plant source	Compounds	Content [g·kg^{-1}]	Reference
Ipomoea batatas (sweet potatoes)	Total phenolic compounds	49–250	[3]
Smallantus sonchifolius (yacon)	Total phenolic compounds	42	[47]
Eugenia uniflora (pitanga)	Total polyphenols	26.0–233.8	[60]
	Total flavonoids	15–153	
Prunus cerasus (cherries)	Total phenolic compounds	93.97	[49]
	Polymeric procyanidins	41.80	
	Chlorogenic acid	18.96	
Vaccinium ashei (blueberries)	Total phenolic compounds	11.3	[16]
Citrus sinensis and *Citrus aurantium* (oranges)	Total phenolic compounds	12.54–44.41	[51]
Punica granatum (pomegranate)	Total phenolic compounds	19.50–669.35	[37]
Carica papaya (papaya)	Flavonoids	2.31–2.63	[52]
	Phenolic acids	2.81–3.18	
	Flavonoids	59.3–112.0	[55]
Moringa oleifera (moringa)	Phenolics	80.3	[61]
	Flavonoids	170	
	Flavonols	75.4	
Olea europaea (olives)	Polyphenols	2.88	[64]
Brassica oleracea (cabbage)	Polyphenols	0.44–5.71	[65]
Schinus terebinthifolius (spices)	Phenolics	69.6–221.6	[73]
	Flavonoids	69.7–243.1	

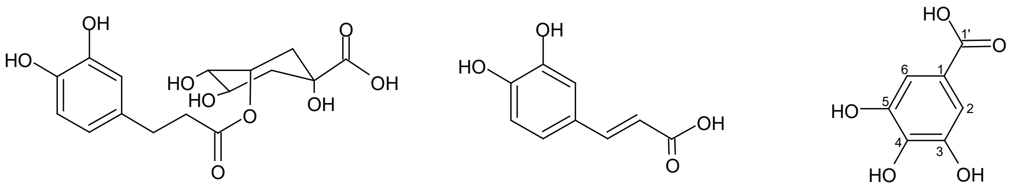

Fig. 4. Chemical structures of some phenolic compounds found in foods [45, 46].

Quercetin and its derivatives can decrease postprandial hyperglycemia by limiting glucose absorption [57].

The doses evaluated in the following studies are shown in Tab. 3. Jaiswal et al. [15] showed that leaf extracts from *M. oleifera* reduced hyperglycemia in severe diabetic rats after 21 days of treatment. Baroni et al. [80] reported that yacon leaf extracts decreased hyperglycemia by improving insulin sensibility through its phenolic compounds in diabetic rats. Ntchapda et al. [35] evaluated aqueous extracts from fig leaves, suggesting that they are effective in the treatment of diabetes. A study conducted by Al-Attar and Zari [81] concluded that crude extracts of *Camellia sinensis* leaves reduced the levels of serum glucose in diabetic mice. Pomegranate leaves have been used in Ayurvedic medicines for the treatment of diabetes [20]. Persimmon leaves showed a significative decrease in insulin resistance index [82].

Several studies dealt with phenolic compounds and their anti-hypertensive effects, since it has been demonstrated that type 2 diabetes and hypertension are metabolic disorders strongly related to each other [83]. The mechanism by which these compounds act consists in their ability to inhibit the action of angiotensin I-converting enzyme (ACE) in vitro and in vivo [79]. ACE converts angiotensin I to angiotensin II, which is a vasoconstrictor, and its inhibition could contribute to the treatment of high blood pressure and can represent an alternative to decrease the consumption of synthetic ACE inhibitors [77].

Leaves from stevia [84], pomegranate [85],

Tab. 3. Effects on hyperglycemia and hypertension of extracts from vegetable leaves with their respective doses associated to contents of phenolic compounds.

Plant source	Doses evaluated	Effects	Reference
Moringa oleifera (moringa)	200 mg·kg^{-1}	Hyperglycemia control	[15]
Smallantus sonchifolius (yacon)	400 mg·kg^{-1}, a day	Hyperglycemia decreasing	[80]
Ficus glumosa (fig)	225.4, 300.0 and 375.0 mg·kg^{-1}	Effective in the diabetes control	[35]
Camellia sinensis (tea)	0.5 ml, a day	Serum glucose decrease	[81]
Stevia rebaudiana (stevia)	50 μg·ml^{-1}	Anti-hypertensive activity	[84]
Punica granatum (pomegranate)	200 mg·kg^{-1} and 400 mg·kg^{-1}	Anti-hypertensive activity	[85]
Hancornias peciosa (mangaba)	0.03, 0.10 and 1.00 mg·kg^{-1}	Anti-hypertensive activity	[86]
Olea europaea (olives)	12.5–25.0 mg·kg^{-1}, twice a day	Anti-hypertensive activity	[87]
Hibiscus sabdariffa (hibiscus)	< 5 g·kg^{-1}	Anti-hypertensive activity	[88]
Morus alba (mulberry)	50 and 100 mg·kg^{-1}	Anti-hypertensive activity	[89]

Doses are expressed per kilogram of body weight.

mangaba [86], olive [87], hibiscus [88–90], mulberry [91], *M. oleifera* [92–94] and sweet potatoes [95] were also reported by the scientific literature as sources of phenolics (Tab. 3), antioxidant capacity of which was related to their anti-hypertensive activity.

Potential of vegetable leaves in food technology

By-products of many vegetables can be introduced in the daily diet and/or can be used in the food industries due to their high content of phenolic compounds and other nutrients [51]. There are several ways of using vegetable crops leaves; dried and used as teas [8]; dried and powdered as flours [9, 10, 18, 94]; or in natura.

Recent studies reported the use of dried leaves as ingredients in the preparation of several processed food products (Tab. 4). These applications include the production of ingredients for bakery products such as quinoa leaves [10, 14], cassava leaves [18], carrot leaves [96, 97] and beet leaves [94]. These studies demonstrated that the use of by-products provided increase in the antioxidant capacity and the levels of several nutrients such as fibre, proteins, minerals, carotenoids and phenolic compounds of processed products. Moreover, the addition of flour from carrots and beet leaves (6 %), as carrot and beet leaves in the formulation of cookies, provided good acceptability and favourable technological properties, as potential to be used in extrusion and bakery [94]. In all of these applications, the leaves were used in the form of a powder, i.e. dried and powdered.

The addition of flour (1, 2, 3, 4 and 5%) from quinoa leaves to bread could decrease loaf volume and increase hardness, cohesiveness and gumminess of this food product [14]. The incorporation of green tea extracts in breads (1.5 g·kg^{-1} and 5.0 g·kg^{-1}) showed changes in crumb appearance, texture properties and taste profiles of them [99].

Phenolics and proteins may raise two types of interactions, covalent or non-covalent, which might lead to precipitation of proteins through multisite interactions and multidentate interactions [100]. The non-covalent binding of phenolic compounds did not present effects on the secondary structure of proteins, but in the tertiary structure [101], while covalent binding affected both secondary and tertiary structures [102]. These characteristics could be responsible for changes in solubility, thermal stability and digestibility of food proteins [101, 103].

The use of phenolics sourced in natural additives of food was also studied. Researches on polyphenols showed that they can have a positive effect on lipid oxidation, colour stability and antioxidant activity in meat products [13, 104]. Nowak et al. [13] found that the addition of leaf extracts of cherry and blackurrant leaves (5 mg·kg^{-1} and 10 mg·kg^{-1}, respectively) increased the shelf life of vacum-packed sausages.

Main technologies in the reuse of leaves by the food industry

Crude extracts of polyphenols from leaves contain chlorophyll, proteins, polysaccharides and other impurities, which limit the application of leaf polyphenols. For this reason, an efficient purification method is required to obtain high purity polyphenols from leaves [105].

The extraction of polyphenolic compounds depends on the polarity of the solvent, method and duration of extraction, which determine both the qualitative and the quantitative composition of these extracts. The polarities of phenolics vary significantly and it is difficult to develop a universal method for optimal extraction of all phenolic compounds [106].

The methods of purification of plant polyphenols are based on extraction with organic solvents, membrane separation or supercritical extraction [107, 108]. However, these methods present some drawbacks, such as long extraction cycles or high cost, which make them unviable for use on industrial scale. Solid-liquid or liquid-liquid extraction can also be used, followed by chromatography. This method is not very effective due to the extensive use of reagents, energy consumption and because it is laborious [109].

Recently, macroporous adsorption resins were used in the purification of bioactive constituents of natural extracts due to their high efficiency [110–112]. They were used in the separation and purification of biologically active substances due to their physico-chemical stability, high adsorption and easy utilization [113]. In addition, adsorption is an environmentally friendly technique, allowing the separation of selected compounds from diluted solutions, and is used to recover phenolic compounds from plants [68, 114]. Macroporous resins exhibit high adsorption capacity not only due to polarity similar to the target compounds, but also because of their large contact surface and average pore diameter [110–112]. They can selectively adsorb aqueous and non-aqueous system constituents through electrostatic force, hydrogen bonding and complexation interactions [115].

On the other hand, acccording to Nantitanon et al. [116], the use of ultrasound was the best method to extract phenolics from guava leaves, followed by Soxhlet extraction and maceration, based on extraction efficiency. These authors also concluded that the maturation stage of the leaves is important and should be considered.

A study conducted by Xi et al. [105] concluded that purification of polyphenols from sweet potato leaves with AB-8 macroporous resin was highly economically and environmentally efficient, with a great potential for industrial production. In addition, the purified products had high antioxidant capacity (62 μg·ml^{-1} of ascorbic acid equivalent).

Challenges for the use of leaves by the food industry

The use of leaves in human food means a viable alternative to valorize wastes [51]. However, there are relevant aspects that have to be considered in the study of these food matrices. Some leaves may contain potentially toxic compounds. The toxicity of cassava leaves, for example, restricts their use. This toxicity is related to the presence of cyanide and the technique of kneading and tearing the

Tab. 4. Studies on leaf flours, obtained by drying and powdering, and their application in food technology.

Plant source	Study	Dose [%]	Technological and/or sensory effects	Reference
Chenopodium quinoa (quinoa)	Bread formulation	3	Valuable supplements to the development of bread with improved functional properties; the supplementation increased antioxidant potential of the product without compromising sensory quality	[10]
	Bread fortification	1	Fortification affected positively the antioxidant and phenolic contents, the quality and biological effects; interactions were identified between phenolics, proteins and starch, which affected the antioxidant capacity, starch digestibility and functional properties of breads	[14]
Moringa oleifera (moringa)	Ice cream formulation.	7–13	The product presented high nutritional value and sensory viability	[9]
Manihot esculent (cassava)	Flour preparation	–	Source of proteins, vitamins C, β-carotene and minerals; the processing reduced cyanide levels	[98]
	Preparation of snacks	10	High levels of proteins and fibres; can be used as materials for extrusion process	[18]
Daucus carota (carrot)	Preparation of cookies	10	Excellent sources of fibres and minerals, good acceptability	[96]
	Flour preparation for food products	30	Alternative method for producing products in general, such as pasta, cakes and breads	[97]
Beta vulgaris (beet)	Cookies preparation	6	Source of proteins and fibre, satisfactory sensory properties	[95]

Doses of the product used are presented.

cassava leaves before drying them can decrease the hydrocyanic acid contents. It was observed that the drying process of cassava leaves is efficient to promote the reduction of hydrocyanic acid [98]. Antia et al. [117] found low levels of toxic compounds in sweet potato leaves, except for oxalate, content of which can be reduced through cooking.

The extraction and purification of phytochemicals present in leaves are also challenges for the food industry, since several extraction techniques can lead to a decrease in these compounds or even generate negative impacts to the environment and/or the consumers' health.

CONCLUSIONS

Several studies reported that phenolic compounds found in vegetable leaves can be related to health-promoting properties, such as anti-inflammatory, antioxidant, antibacterial, anti-hypertensive and anti-hyperglycemic activities. These materials of high nutritional value can be incorporated in the human diet or used as ingredients for food products. Furthermore, the practice of full use of food is a strategy to promote sustainability, reducing wastes and improving the economy. Obtaining phenolic compounds from leaves is an alternative to add value to these by-products, with the purpose to provide new opportunities for industries to develop functional foods, cosmetics and pharmaceuticals products.

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