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Review

Natural antioxidants and antioxidant capacity of Brassica vegetables: A review

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Abstract

Dietary antioxidants, such as water-soluble vitamin C and phenolic compounds, as well as lipid-soluble vitamin E and carotenoids, present in vegetables contribute both to the first and second defense lines against oxidative stress. As a result, they protect cells against oxidative damage, and may therefore prevent chronic diseases, such as cancer, cardiovascular disease, and diabetes. Brassica vegetables, which include different genus of cabbage, broccoli, cauliflower, Brussels sprouts, and kale, are consumed all over the world. This review focuses on the content, composition, and antioxidant capacity both lipid- and water-soluble antioxidants in raw Brassica vegetables. The effects of post-harvest storage, industrial processing, and different cooking methods on stability of bioactive components and antioxidant activity also are discussed.

Keywords: Brassica vegetables; Antioxidant vitamins; Phenolic compounds; Antioxidant activity

Contents

1. Introduction

In recent years, increasing attention has been paid to the role of diet in human health. Several epidemiological studies have indicated that a high intake of plant products is associated with a reduced risk of a number of chronic diseases, such as atherosclerosis and cancer (Gosslau & Chen, 2004; Gundgaard, Nielsen, Olsen, & Sorensen, 2003; Hashimoto, Kawamata, Usui, Tanaka, & Uda, 2002; Kris-Etherton, Etherton, Carlson, & Gardner, 2002; Law & Morris, 1998; Temple, 2000). These beneficial effects have been partly attributed to the compounds which possess antioxidant activity. The major antioxidants of vegetables

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are vitamins C and E, carotenoids, and phenolic compounds, especially flavonoids. These antioxidants scavenge radicals and inhibit the chain initiation or break the chain propagation (the second defense line). Vitamin E and carotenoids also contribute to the first defense line against oxidative stress, because they quench singlet oxygen (Krinsky, 2001; Shi, Noguchi, & Niki, 2001). Flavonoids as well as vitamin C showed a protective activity to α-tocopherol in human LDL, and they can also regenerate vitamin E, from the α-chromanoxy radical (Davey et al., 2000; Zhu, Huang, & Chen, 2000).

Nutrient antioxidants may act together to reduce reactive oxygen spieces level more effectively than single dietary antioxidants, because they can function as synergists (Eberhardt, Lee, & Liu, 2000; Ohr, 2004; Rossetto et al., 2002; Trombino et al., 2004). In addition, a mixture containing both water-soluble and lipid-soluble antioxidants is capable of quenching free radicals in both aqueous and lipid phases (Chen & Tappel, 1996). For example, with the liposome oxidation method, the activity of combination of quercetin or catechins plus α-tocopherol was significantly higher than the sum of the individual activities (Murakami, Yamaguchi, Takamura, & Matoba, 2003). Combinations of α-tocopherol or vitamin C plus phenolic compounds also provided synergistic effects in human erythrocyte membrane ghosts and phosphatidylcholine liposome systems (Liao & Yin, 2000).

Brassica vegetables belong to Cruciferous family, and include different genus of cabbage (white, red, savoy, swamp, chinese), cauliflower, broccoli, Brussels sprouts and kale. These vegetables possess both antioxidant and anticarcinogenic properties (Cohen, Kristal, & Stanford, 2000; Chu, Sun, Wu, & Liu, 2002; Verhoeven, Verhagen, Goldbohm, van den Brandt, & van Poppel, 1997). In addition to antioxidant vitamins, carotenoids, and polyphenols, Brassica vegetables provide a large group of glucosinolates, which according to Plumb et al. (1996) possess rather low antioxidant activity, but the products of their hydrolysis can protect against cancer (Keum, Jeong, & Kong, 2004; Paolini, 1998).

Variation in the antioxidant contents of Brassica vegetables is caused by many factors: variety, maturity at harvest, growing condition, soil state, and condition of post-harvest storage (Jeffery et al., 2003; Kurilich et al., 1999; Lisiewska & Kmiecik, 1996; Vallejo, Tomas-Barberan, & Garcia-Viguera, 2002; van der Berg et al., 2000). In addition, Brassica vegetables can be cooked in many ways, while cabbage, broccoli and cauliflower may be eaten raw as the ingredients of different salads. Kale may be prepared in the same way as spinach and its small amounts are used as an excellent component of salads.

Vegetable industrial processing such as blanching, canning, sterilization and freezing, as well as domestic cooking, is expected to affect the content, composition, antioxidant activity and bioavailability of antioxidants. In addition, operations such as cutting and slicing may induce a rapid enzymatic depletion of several naturally occurring antioxidants as a result of cellular disruption which allows contacts of substrates and enzymes. Generally, the antioxidant concentrations and activities in processed vegetables were lower than those of the corresponding raw samples. This was caused by their degradation, but also by absorption of water during boiling, which diluted the compounds and decreased their content per weight unit. The losses during Brassica vegetable processing need to be taken into account when calculating the dietary intake of dietary antioxidants from processed food.

2. Water-soluble antioxidants

2.1. Vitamin C

Vitamin C, which includes ascorbic acid and its oxidation product—dehydroascorbic acid, has many biological activities in human body. Block et al. (2004) have found that vitamin C can reduce levels of C-reactive protein (CRP), a marker of inflammation and possibly a predictor of heart disease. More than 85% of vitamin C in human diets is supplied by fruits and vegetables (Davey et al., 2000; Lee & Kader, 2000). Biological function of L-ascorbic acid can be defined as an enzyme cofactor, a radical scavenger, and as a donor/acceptor in electron transport at the plasma membrane. Ascorbic acid is able to scavenge the superoxide and hydroxyl radicals, as well as regenerate α-tocopherol (Davey et al., 2000).

The content of vitamin C among Brassica vegetables varies significantly between and within their subspecies (Table 1). Vitamin C levels varied over a 4-fold in broccoli and cauliflower, 2.5-fold in Brussels sprouts and white cabbage, and twice in kale. The cause of reported variations in vitamin C content might be related to the differences in genotype (Kurilich et al., 1999; Vallejo et al., 2002). Climatic conditions also might alter vitamin C level (Howard, Wong, Perry, & Klein, 1999). Lisiewska and Kmiecik (1996) reported that nitrogen fertilization did not affect the content of vitamin C in broccoli, but increasing amount of nitrogen fertilizer from 80 to 120 kg/ha decreased the vitamin C content by 7% in cauliflower. Generally, among Brassica vegetables, white cabbage is the poorest source of vitamin C. However, for example in Poland, white cabbage is the most popular species of Brassica vegetables.

Dehydroascorbic acid (DHA)—oxidation product of ascorbic acid is unstable at physiological pH and it is spontaneously and enzymatically converted to 2,3-diketogulonic acid (Davey et al., 2000). According to Gokmen, Kahraman, Demir, and Acar (2000), DHA was the dominant form of vitamin C in cabbage, with 4-fold higher level than ascorbic acid. In contrast to this report, Vanderslice, Higgs, Hayes, and Block (1990) observed that the contribution of DHA to the total vitamin C contents was 14% or 8% in cauliflower and broccoli, respectively. These authors did not find DHA in fresh cabbage. Those values were in agreement with that reported for broccoli by Vallejo, Tomas-Barberan, and Garcia-Viguera (2003),

Table 1
Ascorbic acid (AA) content of Brassica vegetables (mg/100 g edible portion)

Vegetable	AA content	References
Broccoli	34–93	Favell (1998)
	41–64	Franke et al. (2004)
	74.8	Bahorun et al. (2004)
	75	Hussein et al. (2000)
	84	Hrncirik et al. (2001)
	93	Chu et al. (2002)
	103	Zhang & Hamauzu (2004)
	112	Murcia et al. (2000)
	113	Davey et al. (2000)
	54–120	Kurilich et al. (1999)
	43–146	Vallejo et al. (2002)
Brussels sprouts	76	Pfendt, Vukasinovic, Blagojevic, and Radojevic (2003)
	87–109	Davey et al. (2000)
	192	Czarniecka-Skubina (2002)
White cabbage	18.8	Bahorun et al. (2004)
	25.6	Gokmen et al. (2000)
	28.2	Pfendt et al. (2003)
	23–33	Kurilich et al. (1999)
	32	Chu et al. (2002)
	43	Puupponen-Pimia et al. (2003)
	44	Hrncirik et al. (2001)
	46–47	Davey et al. (2000)
Kale	92.6	Pfendt et al. (2003)
	186	Davey et al. (2000)
Cauliflower	17.2	Pfendt et al. (2003)
	40–44	Kurilich et al. (1999)
	49.9	Bahorun et al. (2004)
	64	Hrncirik et al. (2001)
	64–78	Davey et al. (2000)
	81	Puupponen-Pimia et al. (2003)

because the contribution of DHA to the total vitamin C contents was 11.3%.

In addition to ascorbic and dehydroascorbic acid, *Brassica* vegetables include ascorbigens, which are formed as the result of the reaction between ascorbic acid and degradation products of indol-3-ylmethylglucosinolates produced in the myrosinase-catalysed degradation (Buskov et al., 2000; Hrncirik, Valusek, & Velisek, 2001). According to Buskov et al. (2000), generally, 30–60% of the indol-3-ylmethylglucosinolates in the Brassica plants were transformed into ascorbigens. With regard to ascorbic acid, Hrncirik et al. (2001) suggested that the decrease of ascorbic acid content, as a result of its transformation into ascorbigen, will probably not reach more than 10% during processing of Brassica vegetables.

2.2. Phenolic compounds

Phenolic compounds are a large group of the secondary metabolites widespread in plant kingdom. They are categorized into classes depending on their structure and subcategorized within each class according to the number and position of hydroxyl group and the presence of other substituents. The most widespread and diverse group of the polyphenols are the flavonoids which are built upon C_6–C_3–C_6 flavone skeleton. In addition, other phenolic compounds such as benzoic acid or cinnamic acid derivatives have been identified in fruits and vegetables (Aherne & O'Brien, 2002; Robards, Prenzler, Tucker, Swatsitang, & Glover, 1999).

Phenolic compounds, especially flavonoids, possess different biological activities, but the most important are antioxidant activity, capillary protective effect, and inhibitory effect elicited in various stages of tumor (Cook & Samman, 1996; Czeczot, 2000; Hollman, Hertog, & Katan, 1996; Kuntz, Wenzel, & Daniel, 1999). Phenolics are able to scavenge reactive oxygen spieces due to their electron donating properties. Their antioxidant effectiveness depends on the stability in different systems, as well as number and location of hydroxyl groups. In many in vitro studies, phenolic compounds demonstrated higher antioxidant activity than antioxidant vitamins and carotenoids (Re et al., 1999; Vinson, Dabbagh, Serry, & Jang, 1995).

The studies on phenolic profiles of Brassica vegetables have been focused mainly on broccoli florets, which are popular in Western Europe countries and USA. Broccoli is a source of flavonol and hydroxycinnamoyl derivatives. Price, Casuscelli, Colquhoun, and Rhodes (1998) identified the main flavonol glycosides present in broccoli florets as quercetin and kaempferol 3-*O*-sophoroside. Three minor glucosides of these aglycones were also detected, namely isoquercitrin, kaempferol 3-*O*-glucoside and kaempferol diglucoside. The predominant hydroxycinnamoyl acids were identified as 1-sinapoyl-2-feruloylgentiobiose, 1,2-diferuloylgentiobiose, 1,2,2′-trisinapoylgentiobiose, and neochlorogenic acid (Vallejo et al., 2003). In addition, 1,2′-disinapoyl-2-feruloylgentiobiose and 1,2-disinapoyl-gentiobiose, 1-sinapoyl-2,2-diferuloyl gentiobiose, isomeric form of 1,2,2′-trisinapoylgentiobiose, and chlorogenic acids were found in broccoli (Price, Casuscelli, Colquhoun, & Rhodes, 1997; Vallejo et al., 2003). Total amounts of feruloylsinapoyl esters of gentiobiose and caffeic acid derivatives in 14 cultivars of broccoli varied from 0 to 8.25 mg/100 g, and from 0 to 3.82 mg/100 g, respectively.

Nielsen, Olsen, and Petersen (1993) showed that cabbage contains a mixture of more than 20 compounds of which seven have been identified as 3-*O*-sophoroside-7-*O*-glucosides of kaempferol and quercetin with and without further acylation with hydroxycinnamic acids. In addition, unmodified kaempferol tetraglucosides or their derivatives acylated with either sinapic, ferulic or caffeic acid were found in cabbage leaves (Nielsen, Norbek, & Olsen, 1998).

Red pigmentation of red cabbage is caused by anthocyanins, which belong to flavonoids. Red cabbage contains more than 15 different anthocyanins which are acylglycosides of cyanidin (Dyrby, Westergaard, & Stapelfeldt, 2001; Mazza & Miniati, 1993). Total anthocyanins content in red cabbage was 25 mg/100 g (Wang, Cao, & Prior, 1997) or

44.4–51.2 mg/100 g for anthocyanidins released after acid hydrolysis (Franke, Custer, Arakaki, & Murphy, 2004). In Japan red cabbage is a source of red food colorants and the preparation of these pigments is described in several patents (Bridle & Timberlake, 1997).

The content of polyphenols in vegetables, like levels of other phytochemicals, can be influenced by various factors such as varieties, climatic conditions and cultural practices, maturity at harvest, and storage conditions. In the case of phenolic compounds, which are highly reactive spieces, sample preparation method is also very important. There have been only a few studies that evaluated the content of polyphenols in Brassica vegetables (Table 2). Phenolic contents ranged from 15.3 mg/100 g fresh weight in white cabbage to 337.0 mg/100 g in broccoli. Phenolic level in broccoli varied from 34.5 to 337.0 mg/100 g. In plants, phenolics occur in soluble forms as well as in combination with cell wall components—bound phenols. According to Chu et al. (2002), participation of bound phenolics in total phenolics varied from 20.5% in broccoli to 32.9% in cabbage. Bound phenols were quantified in the extracts after base hydrolysis of residues following the solvent-soluble extraction.

Phenolics in vegetables also exist in both free and conjugated forms. Generally, in fresh vegetables only conjugated flavonoids are present but aglycones may be found as a result of food processing. Most studies on vegetable flavonoid levels determined aglycones after hydrolysis of food extracts by heat and acid, because determination of individual flavonoid glycosides is difficult, because of lack of reference compounds. After hydrolysis, HPLC analysis showed that quercetin was the predominant flavonol aglycone in Brassica vegetables. Its level in Mauritian Brassica vegetables varied from 3.9 mg/100 g in cauliflower to 39.0 mg/100 g in Chinese cabbage (Bahorun, Luximon-Ramma, Crozier, & Aruoma, 2004). However, Chu, Chang, and Hsu (2000) reported much lower contents of quercetin for Brassica vegetables cultivated in Taiwan: 0.004 mg/100 g for white cabbage and 0.024 mg/100 g for Chinese cabbage. Kaempferol and myricetin derivatives were also present in Brassica vegetables, but myricetin was not present in broccoli, white cabbage, purple cabbage, and cauliflower. According to Bahorun et al. (2004), apigenin and luteolin were flavones detected in the hydrolysed extracts of different Brassica vegetables, except for broccoli. Among four Taiwan Brassica vegetables studied by Chu et al. (2000), the levels of flavone were higher than those of flavonol in all tested vegetables. Apigenin was the predominant flavone aglycone in these vegetables except Chinese cabbage, where luteolin content was nearly 4-fold higher than apigenin content.

3. Lipid-soluble antioxidants

3.1. Carotenoids

Carotenoids (carotens and xanthophylls) are yellow, orange, and red pigments present in many fruits and vegetables. Several of them are precursors of vitamin A (i.e. β-carotene, γ-carotene, and β-cryptoxanthin), and due to conjugated double bonds they are both radical scavengers and quenchers of singlet oxygen. Lower serum β-carotene levels have been linked to higher rates of cancer and cardiovascular diseases, as well as to increased risk of myocardial infarction among smokers (Rice-Evans, Sampson, Bramley, & Holloway, 1997).

Among the 22 species of vegetables investigated by Muller (1997), kale, red paprika, leaf of parsley, spinach, Lamb's lettuce, carrot, and tomato were very rich in carotenoids (over 10 mg/100 g edible portion). In this study, Brussels sprouts were 11th (6.1 mg/100 g), broccoli 16th (1.6 mg/100 g), red cabbage 20th (0.43 mg/100 g), and white cabbage 21st (0.26 mg/100 g) in the rank order based on total carotenoid content. Table 3 shows the range of concentrations of important dietary carotenoids from Brassica vegetables. Lutein and β-carotene are the dominant carotenoids in cruciferous vegetables. The highest lutein + zeaxanthin values were obtained for kale (3.04–39.55 mg/100 g). The amount of these xanthophylls was moderately high (0.78–3.50 mg/100 g) in broccoli and

Table 2
Phenol content of Brassica vegetables (mg of gallic acid/100 g edible portion)

Origin	Extraction solvent	Vegetable	Total phenols	References
Mauritius	Acetone/water (70:30 v/v)	Broccoli	82.2 ± 8.9	Bahorun et al. (2004)
		Cauliflower	27.8 ± 1.5	
		Chinese cabbage	118.9 ± 12.5	
		White cabbage	15.3 ± 2.1	
United States	Acetone/water (80:20 v/v)	Broccoli	80.8 ± 1.2	Chu et al. (2002)
		Cabbage	36.7 ± 6.9	
United States	Acetone/water/acetic acid (70:29,5:0.5 v/v/v)	Broccoli	337 ± 62	Wu et al. (2004)
		Cabbage common	203 ± 31	
		Cabbage red	254 ± 18	
		Cauliflower	274	
Japan	Methanol/water (80/20 v/v)	Broccoli	34.5 ± 1.0	Zhang and Hamauzu (2004)

Table 3
Carotenoids content of Brassica vegetables (mg/100 g edible portion)

Vegetable	α-carotene	β-carotene	Lutein + zeaxanthin	References
Broccoli	0.001	0.78	2.45	Holden et al. (1999)
	0	0.28	0.80	Muller (1997)
	tr	1.00	1.80	Heinonen et al. (1989)
	0–0.07	0.37–2.42	NA	Kurilich et al. (1999)
	NA	1.24–1.92	2.42–3.50	de Sa et al. (2004)
	0	0.55	0.78	Mukovic, Gams, Draxl and Pfannhauser (2000)
	NA	0.63	1.05	Zhang and Hamauzu (2004)
Brussels sprouts	0.006	0.45	1.59	Holden et al. (1999)
	0.05	0.63	2.71	Muller (1997)
	—	0.43	0.92	Heinonen et al. (1989)
	0–0.01	0.77–1.02	NA	Kurilich et al. (1999)
White cabbage	0	0.07	0.31	Holden et al. (1999)
	0	0.02	0.08	Muller (1997)
	tr	0.07	0.15	Heinonen et al. (1989)
	<0.01	0.01–0.13	NA	Kurilich et al. (1999)
	0	0.41	0.45	Murkovic et al. (2000)
Red cabbage	0	0.05	0.15	Muller (1997)
	tr	0.02	0.03	Heinonen et al. (1989)
Kale	0	9.23	39.55	Holden et al. (1999)
	0.15	7.28	18.63	Muller (1997)
	0.05–0.07	3.65–6.08	NA	Kurilich et al. (1999)
	NA	2.84–4.38	3.04–5.26	de Sa et al. (2004)
Cauliflower	0	<0.01	0.02	Muller (1997)
	—	0.01	0.03	Heinonen et al. (1989)
	ND	0.07–0.08	NA	Kurilich et al. (1999)

ND—below the level of detection; NA— not analysed

Brussels sprouts. In addition to lutein and *trans*-β-carotene, *cis*-β-carotene was reported in Brussels sprouts, broccoli and green cabbage (Hart & Scott, 1995; Muller, 1997). According to Muller (1997), Brassica vegetables also contain cryptoxanthin, neoxanthin and violaxanthin, but Heinonen, Ollilainen, Linkola, Varo, and Koivistoinen (1989) detected cryptoxanthin only in broccoli (0.024 mg/100 g).

3.2. Vitamin E

In addition to carotenoids, vitamin E also belongs to a group of lipid-soluble antioxidants. The biological activity of vitamin E exhibit tocopherols and tocotrienols, especially α-tocopherol. The predominant reaction responsible for tocopherol antioxidant activity is hydrogen atom donation, where a tocopheroxyl radical is formed (Lampi, Kamal-Eldin, & Piironen, 2002). Vitamin E shows protective effects against the coronary heart disease due to inhibition of LDL oxidation (Stampfer & Rimm, 1995).

Although vegetables in addition to fats, oils and cereal grains, constitute the major source of vitamin E in our diet, there are only few data of tocopherol content in vegetables. The descending order of total tocopherols and tocotrienols in Brassica vegetables is as follows: broccoli (0.82 mg/100 g) > Brussels sprouts (0.40 mg/100 g) > cauliflower (0.35 mg/100 g) > chinese cabbage (0.24 mg/100 g) > red cabbage (0.05 mg/100 g) > white cabbage (0.04 mg/100 g) (Piironen, Syvaoja, Varo, Salminen, & Koivistoinen, 1986). Kurilich et al. (1999) have also reported similar rank on the basis of concentration, but in their study total tocopherol values were about 2-fold higher. These differences are probably caused by the differing varieties and growing conditions. According to these authors, kale was the best source of α-tocopherol and γ-tocopherol (2.15 mg/100 g). Piironen et al. (1986) reported that α-tocopherol was predominant tocopherol in all Brassica vegetables, except in cauliflower, containing predominantly γ-tocopherol. In contrast, Kurilich et al. (1999) reported lower concentration of γ-tocopherol than α-tocopherol in cauliflower.

In general, the best sources of lipid-soluble antioxidants are kale and broccoli. Brussels sprouts have moderate levels of the above-mentioned compounds, while cauliflower and cabbage are characterized by their relatively low amounts.

4. Stability of dietary antioxidants during processing and storage of Brassica vegetables

Among natural antioxidants, vitamin C is considered as an indicator of the quality of food processing, due to its

high degree of water solubility and low stability during heat treatment. Brassica vegetables after harvest have been stored until sale and use by consumer at ambient or chill temperatures. Generally vegetables showed a rapid loss of ascorbic acid (AA) at ambient temperature. Favell (1998) noticed a steady loss of this compound during storage of broccoli, after 7 and 14 days storage at 20 °C AA content decreased to 44% and to 28% of the original amount, respectively. However, when broccoli was stored at 4 °C, the retention of AA was very good with no loss after 7 days and only 20% loss after 21 days. According to Leja, Mareczek, Starzyńska, and Rożek (2001) AA content in non-packed broccoli was reduced by 26% after 3 days of storage at room temperature, but in case of an application of commercial polymeric film, retention of AA was 100%. On the contrary, the reducing of the storage temperature to 5 °C resulted in an increase (25%) of AA content in non-packaged samples and insignificant decrease (3%) in packaged broccoli. The levels of vitamin C also slightly decreased (2.4% loss) when broccoli was wrapped in low-density polyethylene and stored for 7 days in a cold room at 1 °C (Vallejo et al., 2003). Furthermore, the contribution of dehydroascorbic acid to the total vitamin C contents under above conditions increased from 11.3% to 31.2%.

Broccoli, cauliflower and Brussels sprouts are available throughout the year as deep-frozen foods. Prior to freezing, vegetables are washed, sometimes cut, and steam or water blanched in order to inactivate enzyme systems, especially oxidative enzymes (e.g. polyphenoloxidase, ascorbic oxidase, peroxidase). Vitamin C losses during blanching were plant species-dependent. Retention level of vitamin C after blanching (3 min, 96 °C) was 84% for cauliflower, but around 70% for cabbage, probably due to cutting it into slices (Puupponen-Pimia et al., 2003). Lower stability of vitamin C at the same temperature was observed by Lisiewska and Kmiecik (1996). In the case of cauliflower there was a 28–32% loss of vitamin C content brought by a 4 min blanching, while in the case of a 3 min blanching of broccoli—41–42%. Similar results have been obtained for Brussels sprouts by Czarniecka-Skubina (2002). After blanching (4.5 min at 93–95 °C) and freezing, loss of vitamin C was 34%. Steam blanching of broccoli decreased AA concentration about 30% (Howard et al., 1999). Murcia, Lopez-Ayerra, Martinez-Tome, Vera, and Garcia-Carmona (2000) observed that losses of vitamin C content in broccoli were 50–51% in florets and 54–55% in steams independently of prolongation of blanching time from 1 to 2.5 min. The freezing operation did not change the level of vitamin C, which is also stable in frozen broccoli and cauliflower during a 12 month storage at –20 °C. Under these conditions, vitamin C contents decreased by 3–18% for broccoli and 6–13% for cauliflower (Favell, 1998; Lisiewska & Kmiecik, 1996). Higher losses were observed for cabbage, which lost 30% of vitamin C after storage under similar conditions (Puupponen-Pimia et al., 2003).

Czarniecka-Skubina (2002) reported that retention of vitamin C in Brussels sprouts strongly depends on the cooking method. High retention of this vitamin was found for cooking in a microwave oven, pressure cooker in steam and acuthermal pot, losses of vitamin C were from 3.7% to 10.6%. On the contrary, low retention of vitamin C was noted for traditional cooking in a pot starting with cold water (loss 38.6%), in pressure cooker starting with boiling water (31.3%), and in a pot starting with boiling water (27.6%). The result obtained by Zhang and Hamauzu (2004) showed that the content of ascorbic acid in broccoli declined dramatically during both conventional and microwave cooking. The authors observed that the time of cooking had a higher influence on ascorbic acid level than the cooking methods. The florets cooked conventionally for 0.5, 1.5 and 5 min lost 19.2%, 47.5%, and 65.9% of ascorbic acid present in fresh florets, respectively. For comparison, in the microwaving cooking florets lost 17.4%, 48.0%, and 65.5%, respectively. With regard to vitamin C content, the canning of Brassica vegetables was the worst preservation method. After blanching and canning of Brussels sprouts, a decrease of vitamin C by 66% was observed, that was about 2-fold higher than in case of blanching and freezing (Czarniecka-Skubina, 2002), and in the case of canned broccoli only 16% of original vitamin C was retained (Murcia et al., 2000).

Phenolic compounds in broccoli, which are stored 7 days at 1 °C, showed 2–3 dozen lower stability in comparison with ascorbic acid (Vallejo et al., 2003). Total flavonoid, sinapic and caffeoyl-quinic derivatives contents decreased up to 61%, 51% and 73% of the initial value, respectively. On the contrary, Leja et al. (2001) observed very good stability of total polyphenols in broccoli during a 7 day storage at 5 °C. The level of those antioxidants in samples packed in polymeric film was the same as in the fresh harvested vegetables. In case of non-packed samples authors noticed a significant increase (26.7%) of phenolic content, probably due to the weight loss that led to concentration of these compounds in the cells. Higher increases of polyphenolic concentration at ambient temperature (20 °C) in comparison to changes during cold storage seem to confirm this fact. The effect of cooking on phenolic compounds in broccoli are shown in Table 4. According to Vallejo et al. (2003), among four cooking methods, steaming led to the retention of the highest levels of flavonoids, caffeoyl-quinic derivatives, sinapic and feruoyl derivatives in edible part of broccoli. On the contrary, microwave treatment caused the highest losses of these phenolic compounds, because there were 32-, 7- and 4-fold higher than for steam cooking, respectively. Generally, sinapic and feruoyl derivatives showed the highest stability, followed by caffeoyl-quinic derivatives, and flavonoids. Price et al. (1998) found a similar retention of flavonoid glycosides in broccoli, which was conventionally cooked, starting with boiling water. The highest retention (>20%) after cooking for 5 min was noticed for quercetin-3-*O*-sophoroside and kaempferol-3-*O*-glucoside. Stability

Table 4
Effect of cooking on phenolic compounds in broccoli

Cooking process	Broccoli parts	Ratio broccoli/water	Time min	Determined value	Retention of compounds %	References
Conventional	Floret	0.5 g/10 ml	0.5	Total phenolics	68.4	Zhang and Hamauzu (2004)
			1.5		44.0	
			5.0		28.1	
	Steam	0.5 g/10 ml	0.5	Total phenolics	86.7	Zhang and Hamauzu (2004)
			1.5		73.3	
			5.0		57.8	
Conventional	Floret	3 g/10 ml	15.0	Total flavonol glycosides	18.0	Price et al. (1998)
				Quercetin 3-*O*-sophoroside	28.0	
				Kaempferol-3-*O*-sophoroside	14.0	
				Isoquercitrin	15.0	
				Kaempferol-3-*O*-glucoside	24.0	
				Kaempferol-diglucoside	19.0	
Conventional	Edible part	10 g/10 ml	5.0	Flavonoids	19.1	Vallejo et al. (2003)
				Caffeoyl-quinic derivatives	39.9	
				Sinapic and feruoyl derivatives	49.3	
Microwaving	Edible part	10 g/10 ml	5.0	Flavonoids	2.8	Vallejo et al. (2003)
				Caffeoyl-quinic derivatives	12.6	
				Sinapic and feruoyl derivatives	23.8	
Steaming	Edible part	10 g/10 ml	3.5	Flavonoids	88.9	Vallejo et al. (2003)
				Caffeoyl-quinic derivatives	91.9	
				Sinapic and feruoyl derivatives	99.7	
High pressure	Edible part	10 g/10 ml	3.0	Flavonoids	46.6	Vallejo et al. (2003)
				Caffeoyl-quinic derivatives	50.7	
				Sinapic and feruoyl derivatives	58.1	

of broccoli phenolics strongly depended on cooking time. A 10-fold (from 0.5 to 5 min) prolongation of the conventional cooking time caused 2-fold higher total phenolic losses in term of florets and 1.5-fold in steams (Zhang & Hamauzu, 2004). Total phenolic losses during blanching/freezing varied from 12% in kale and cauliflower to 58% in broccoli, and phenolic contents continued to decrease slighty during storage (Ismail, Marjam, & Foong, 2004; Ninfali & Bacchiocca, 2003; Puupponen-Pimia et al., 2003).

In terms of carotenoids' stability during food processing, there are opposite reports. Some workers have reported losses of total carotenoids from broccoli, during conventional and microwave cooking (Zhang & Hamauzu, 2004). The florets and stems cooked for 5 min by these both methods lost about 23% of total carotenoids. Among analysed carotenoids, the levels of β-carotene and violaxanthin declined during the conventional and microwave cooking, but the level of lutein increased gradually in both cooking methods. This could be due to transformation of the *cis* isomer of lutein to *trans* form, which has been noted during microwave cooking of broccoli and in case of canned kale (Updike & Schwartz, 2003). De Sa and Rodriguez-Amaya (2004) also noted a reduction of carotenoid concentration both in boiled and stir-fried samples, the loss was as high as 22% and 4%, respectively. On the contrary, some authors reported that thermal processing increased carotenoids concentration, presumably due to greater chemical extractability and enzymatic degradation. According to Hart and Scott (1995), boiled broccoli and cabbage green showed an increase of 31% of total carotenoids. Furthermore, in case of cooked broccoli, a higher increase of β-carotene than lutein has been observed, however for green cabbage the opposite trend was noted. The cooking of frozen Brussels sprouts in water for 15 min has not affected the content of total carotenoids, but resulted in 28% and 2% increases in *cis* β-carotene and lutein contents, respectively, and 7% decrease of β-carotene level.

To sum up, during vegetable processing, the antioxidants thermal breakdown, qualitative changes and leaching into the surrounding water influence the antioxidant capacity of Brassica vegetables.

5. Antioxidant activity of raw and processed Brassica vegetables

Brassica vegetable extracts have been screened for antioxidant activity using different oxidation systems and methods to measure antioxidant capacity (Azuma, Ippoushi, Ito, Higashio & Terao, 1999; Cao, Sofic, & Prior, 1996; Chu et al., 2002; Honer & Cervellati, 2002; Ou,

Huang, Hampsch-Woodill, Flanagan, & Deemer, 2002; Roberts & Gordon, 2003; Wu et al., 2004). For example, the order of the ORAC values of the fresh weight extracts was: kale > Brussels sprouts > broccoli > cauliflower > cabbage (Cao et al., 1996) – Table 5. In another study the linoleic acid emulsion and liposomal phospholipid suspension systems were used to determine the inhibition of lipid peroxidation of vegetables with the following ranking: broccoli > cauliflower > cabbage > Chinese cabbage, and cauliflower > broccoli > cabbage > Chinese cabbage, respectively (Azuma et al., 1999). Generally, among analysed Brassica vegetables Brussels sprouts, broccoli, and red cabbage belong to the group of the highest antioxidant capacity. Common cabbage demonstrated rather low antioxidant activity. Although, in some cases, cabbage revealed the antioxidant activities comparable to the efficiency of broccoli (Ou et al., 2002; Wu et al., 2004). Controversial results had been demonstrated for cauliflower, which showed high activity in liposomal phospholipid suspension system, but low activity in oxygen radical absorption capacity (ORAC) method (Azuma et al., 1999; Wu et al., 2004). The order of antioxidant activities depended on the extraction method, and on the type of the reactive spieces in the reaction mixture (Azuma et al., 1999; Cao et al., 1996). In addition, Kurilich, Jeffery, Juvik, Wallig, and Klein (2002) found significant differences in ORAC values in extracts from the eight broccoli genotypes. The difference between the highest and the lowest total antioxidant capacity was 3-fold. For the above reasons, the data obtained by different researches are sometimes difficult to compare.

To make an overall evaluation of the total antioxidant capacity of vegetables, the activity of both water- and lipid-soluble antioxidants must be considered. In terms of broccoli, Kurilich et al. (2002) reported that the hydrophilic extracts, which include vitamin C and polyphenols, were responsible for 80–95% of the total antioxidant capacity using the ORAC assay. Wu et al. (2004) also found that hydrophilic antioxidants in Brassica vegetables made up >89% of the total antioxidant capacity.

Heat treatments affect the antioxidant activity of vegetables and in many cases has been observed lower antioxidant capacity in processed samples versus raw vegetables. DPPH index of cauliflower decreased by 23%, but in case of cabbage increased by 9% during blanching in water (Puupponen-Pimia et al., 2003). During the conventional and microwave cooking for 5 min, both the florets and stems of broccoli retained about 35% of total antioxidant activity measured by DPPH method (Zhang & Hamauzu, 2004). Similarly, according to Lin and Chang (2005), the extract from broccoli cooked for 10 min at 50 °C showed scavenging activity toward DPPH radicals of 31%, when the extract from fresh broccoli exhibited higher activity by 40%. In cooked broccoli, Wu et al. (2004) observed 21% decrease of total ORAC activity, which was caused more by lipophilic antioxidants than hydrophilic compounds. On the contrary, cooked red cabbage was significantly higher (40%) in ORAC value of hydrophilic extract compared to the raw form.

6. Conclusion

Brassica vegetables are consumed all over the year as the ingredients of different salads or after cooking of raw and frozen vegetables. The contribution of Brassica vegetables to health improvement can be related to their antioxidant capacity. Phenolic compounds with vitamin C are the major antioxidants of Brassica vegetables, due to their high content and high antioxidant activity. On the contrary, lipid-soluble antioxidants (carotenoids and vitamin E) were responsible for up to 20% of the total antioxidant activity of Brassica vegetables. Future research should be focused

Table 5
The antioxidant activities against peroxyl radicals ($ORAC_{ROO\bullet}$—oxygen radical absorbance capacity) of Brassica vegetables

Sample preparation	Vegetable	$ORAC_{ROO\bullet}$ (μmol of Trolox/g)	References
Water extraction + acetone extraction	Kale	17.7	Cao et al. (1996)
	Brussels sprouts	9.8	
	Broccoli	8.9	
	Cauliflower	3.8	
	Cabbage	3.0	
Acetone/water (50:50 v/v) extraction	Broccoli	23–208	Ou et al. (2002)
	Cauliflower	62–152	
	White cabbage	23–146	
Water extraction + hexane extraction	Broccoli	42–137	Kurilich et al. (2002)
Hexane/dichloromethane (1:1 v/v) + acetone/water/acetic acid (70:29.5:0.5 v/v/v)	Red cabbage	22.5	Wu et al. (2004)
	Broccoli	15.9	
	Cabbage common	13.6	
	Cauliflower	6.5	

on relationship between the total antioxidant capacity and the content, as well as composition of antioxidants in Brassica vegetables since: (i) the content and composition of antioxidants vary significantly between and within their subspecies, (ii) antioxidant activity and stability of individual phytochemicals differ significantly, (iii) the vegetable processing (blanching, canning), as well as domestic cooking influence antioxidants content and activity.

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