Red Beets as a Source of Bioactive Compounds

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Beetroot (Beta vulgaris L.) is a vegetable that is consumed worldwide in the form of juices, soups, or salads. It is also known for its high content of biologically active substances such as betalains, polyphenolic compounds, vitamins, carotenoids, and other nutrients including, sodium, potassium, and magnesium. The distribution of these compounds in the plant is diverse, some occur in greater amounts in the leaves (e.g., vitamin A, B6) and others are in the tubers (e.g., folate, lycopene). The concentration of bioactive compounds in beetroot also depends on its variety and growing conditions.

red beets bioactive compounds

betalains phenolic compounds

1. Introduction

Beta vulgaris (beet) is an edible plant in the subfamily Betoideae of the family Amaranthaceae. It has several cultivar groups such as sweet beet, garden beet, chard, and mangoldwurzel. Red beetroots (Beta vulgaris) have been cultivated for many hundreds of years in all temperate climates. Surprisingly, the green parts of the beetroot were consumed initially in the form of chard ^[1]. The dish was so popular that the ancient Greeks and Romans developed a method of growing beetroot also during the hot summer months. It should be emphasized that this variety had only thin and fibrous roots, which were occasionally used in medicine. Consumption of underground beet parts was first reported in Germany and Italy in 1542^[2]. However, that beet variety was significantly different from the one we know today and rather closely resembled a parsnip. The beetroot variety that is still cultivated today appeared in the late 16th century, and is believed to have originated from a prehistoric North African root vegetable. Currently, most beetroots are consumed as a vegetable and also as a juice. Beetroots can also be processed into a powder for use as a food colorant E162 and additive to cosmetics [3][4][5][6].

Although beets have been known for centuries, understanding of their value is still limited. New reports about bioactive compounds in beets suggest that they fit the definition of a functional food, as they offer health benefits that extend beyond their nutritional value [7][8][9]. In addition to proteins, carbohydrates, fat, amino acids, fatty acids, phytosterols, minerals, and fibres, beetroot contains also vitamins, nitrate, polyphenolic compounds, and betalains, a class of red and yellow natural pigments found in plants. Betalains are considered to be strong antioxidant agents as they can decrease oxidative stress by efficiently removing the reactive oxygen species [9][10][11]. Other health benefits of betalains include antimicrobial, antiviral, and anti-inflammatory activities [12][13][14]. Several studies showed the tumour-chemopreventive effects of beetroot extracts in laboratory animals [15][16][17]. This is probably a

synergistic effect of both betalains and other compounds present in beetroot, e.g., polyphenolic compounds. The scheme of the main bioactive compounds present in beetroot is depicted in **Figure 1**.

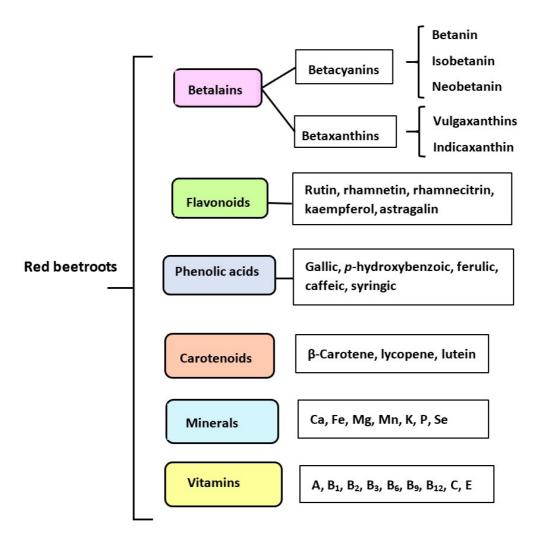


Figure 1. Main bioactive compounds present in red beetroots.

2. Betalains

Betalains are water-soluble secondary metabolites that occur in many plants of the order *Caryphyllales*. They have a higher solubility in water compared to anthocyanins, and are stable in the pH range of 3–7, thus, can be used in low-acid and neutral foods ^{[18][19]}. Betalamic acid is a component of all betalains and its substituent type defines the class of betalain ^[20]. Betalamic acid can spontaneously condense with cyclo-DOPA (3,4-dihydroxyphenylalanine, a precursor of betalain pigment in plants) yielding the violet betacyanins, or with amines or amino acids to produce yellow betaxanthins ^[21]. Their general chemical structures are presented in **Figure 2** ^[18]. Red beets are the main edible source of these compounds. According to Delgado-Vargas et al., betacyanins content in beetroot accounts for 75–95% of the total pigments, the remaining 5–25% corresponds to betaxanthins ^[22]. The violet betacyanin is the most studied of all the betalain groups. Beetroot contains also its epimer, isobetanin, as well as other pigments like betaxanthins vulgaxanthin I and miraxanthin V ^[23]. Four betacyanins (betanin, prebetanin, isobetanin,

neobetanin) and three betaxanthins (vulgaxanthin I, vulgaxanthin II, indicaxanthin) were identified in the water fractions of crude extracts of beetroot peel ^[24]. In beetroot juice samples, the presence of red pigments was reported in ranges of 797.5–421.7 mg·L⁻¹ for betanin and 321.7–423.1 mg·L⁻¹ for vulgaxanthin ^[25]. Concentrations of both dyes in purchased organic beetroot juice were higher than those in freshly squeezed juice ^[26]. However, the characteristic color quickly lost its intensity after storage at 20 °C, and after some time it was possible to notice the presence of a flocculent precipitate. Degradation products of betanin are isobetanin, decarboxylated betanin, betalamic acid, and cyclodopa glucoside ^[27]. Kazimierczak et al. determined higher concentrations of betalains in pure beetroot juices in comparison to those combined with other components (e.g., apple, lemon) ^[28].

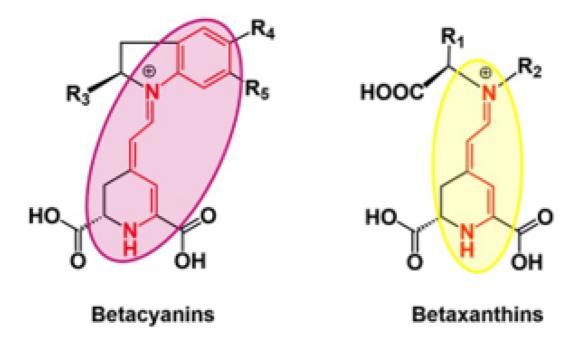


Figure 2. General chemical structures for betacyanins and betaxanthins. Reprinted with permission from [18].

3. Flavonoids and Polyphenolic Acids

The high antioxidant capacity of beets is due not only to the high content of betalains, but also other phenolic compounds—flavonoids and polyphenolic acids. Significant amounts of catechins and polyphenolic acid (ferulic, protocatechuic, vanillic, p-coumaric, p-HBA, syringic, and caffeic) have been detected in beets ^{[29][30][31][32]}. However, the composition of plant extract depends on many factors, including the original plant (variety, maturation state), conditions of its cultivation (climate, harvesting), as well as its storage condition ^[33].

Kavalcowa et al. showed how much the total content of polyphenolic compounds depends on the beet variety and the place of its cultivation ^[32]. The highest content of total polyphenols was found in the variety of Renova from Sliač, in Slovakia (1280.56 μ g·g⁻¹), followed by red beetroot from Zohor. In addition, the variety of Zohor, obtained from Sihelné, had the lowest concentration of polyphenols (820 μ g·g⁻¹). In the case of the variety Monorubrawe, the highest value of total polyphenols was determined in samples from the area of Sihelné (1201.6 μ g·g⁻¹), followed by red beetroot from Zohor (988.66 μ g·g⁻¹). The variety-specific differences

between the highest and lowest polyphenol contents were 460.56 μ g·g⁻¹ in the Renova variety and 212.94 μ g·g⁻¹ in the Monorubrawe variety. The authors concluded that the observed differences may be due to the high humus (4.0%) and potassium (520 μ g·g⁻¹) content in the soil. Studies conducted by Kujala et al. clearly showed that the total phenolics content decreases in order: peel (50%), crown (37%), and flesh (13%) [34]. The comparison of the phenolic composition in betalain extracts from intact B. vulgaris cv. Detroit Dark Red plants and hairy root cultures was described by Georgiev et al. [11]. The authors proved that the concentrations of selected polyphenolic compounds were even more than 100 times higher in extracts from hairy root cultures than in extracts obtained from intact plants. Moreover, rutin was present only in hairy root cultures, while chlorogenic acid was found only in intact plants. While quercetin was not reported in this study, Pratimasari and Puspitasari proved its presence in the purified extract of beetroot leaf ^[35]. In the study performed by Platosz et al., guercetin was detected in fresh red beet, fermented red beet, and commercial fermented beet juice, in concentrations of 0.023 $\mu g \cdot g^{-1}$, 0.002 $\mu g \cdot g^{-1}$. and 0.009 μ g·mL⁻¹, respectively ^[29]. The results obtained by Georgiev indicated that the extract of the hairy roots had substantially higher concentrations of the identified phenolic compounds than did the intact plants, as noted above [11]. Moreover, there were differences in the phenolic profiles of the studied extracts. Chlorogenic acid was present only in the extract from intact plants, while rutin was found only in the extract from hairy root cultures. As the authors emphasize, it is a valuable discovery as rutin interacts synergistically with many other biologically active compounds present in the plant material enhancing its antioxidant properties ^[36]. The fermentation process caused an increase in the content of phenolic acids and a reduction in the content of their conjugated forms, while the same process caused a decrease in the content of free flavonoids ^[29]. In the case of the studies described by Platosz, significant differences in the concentrations of polyphenolic acids were observed between the extracts from fresh and fermented beetroot ^[29]. Such large differences were not observed in the case of flavonoids, their concentrations in most cases were rather similar.

4. Carotenoids

Carotenoids are another group of bioactive compounds present in beetroots; however, they are present rather in small amounts ^[37]. The content of these compounds strictly depends on the part of the plant that is being tested; the highest content of carotenoids is found in beetroots peel, followed by pulp, leaves, and stalks ^[8]. This is because these compounds are accumulated in the green parts of the plants, namely in chloroplasts, as a mixture of α - and β -carotene, β -cryptoxanthin, lutein, zeaxantin, violaxanthin, and neoxanthin ^[38]. The main carotenoids in red beets are β -carotene and lutein, which pose strong anticancer properties ^[16]. Red beets are not a very good source of red carotenoid lycopene in comparison to tomatoes, watermelon, or papaya ^[8]. The lycopene content was 30.0 ± 0.3 µg per 100 g of beet tubers and was not detected in the leaves. α -Carotene was detected in the leaves and tubers of the beetroot, and its concentration in tubers was almost seven times higher than in leaves. However, the leaves are a good source of β -carotene (11.64 µg per 100 g) and lutein (1.503 µg together with zeaxanthin) ^[39]. The presence of these compounds was not confirmed in beet tubers ^[8].

A higher concentration of carotenoids was found in organic stalks and cooked pulp in comparison to the conventionally grown plants ^[40]. This is in line with reports in the literature showing the content of β -carotene is

higher in organically cultivated plants in comparison to conventionally grown plants ^[41]. However, significant differences were only observed between these two parts. After cooking the beet peel and pulp, a decrease in the carotene content was observed; however, its content was still higher in organic beets after this process ^[40].

5. Minerals

Beetroot contains natural minerals that are essential for the proper functioning of the human body. Wruss et al. analyzed the mineral composition of seven different popular beetroot varieties grown in Upper Austria ^[31]. Obtained values were in a range similar to those reported in previous studies ^[42]. However, as the authors highlighted, data regarding the concentration of trace elements and minerals in beet juices are still limited. It is also crucial to note which part of the plant is tested, as the content of individual elements varies among leaves and tubers ^[8]. The concentration of copper ions was more than two times higher in leaves, in comparison to tubers (0.191 and 0.075 mg per 100 g of beets, respectively) ^[8]. A greater difference was observed for iron, for which 2.57 mg was found in leaves and 0.80 mg in tubers (values per 100 g of beets). Also, calcium, sodium, potassium, and magnesium were detected in much higher concentrations in leaves. The concentrations of zinc, phosphorus, and manganese did not significantly differ from each other.

6. Vitamins

Beetroots and beet products are also great sources of vitamin C, vitamin A, and vitamins from the B group. The Bgroup vitamins are a collection of 8 water-soluble vitamins including thiamine (B1), riboflavin (B2), niacin (B3), pantothenic acid (B5), pyridoxine (B6), biotin (B7), folate (B9), and cyanocobalamin (B12). According to Odoh and Okoro 100 g of this plant contains: vitamin A (2.6 mcg), K, (3.2 mcg), C (4.36 mg), E (0.18 mg) B3 (0.03 mg), B6 (90 mg), B2 (0.034 mg), and B5 (0.151 mg) ^[39]. These values are in agreement with findings published by Neha et al. ^[43]. Similar concentrations of vitamins mentioned above were reported by the US Department of Agriculture in the most recent release of the USDA National Nutrient Database for Standard Reference, a major source of food composition data ^[44]. Analysis of vitamin C in different parts of the beetroot was conducted by Rosseto et al. ^[40]. According to this study, the highest concentration of ascorbic acid was found in the pulp of the beet and the lowest in its leaf. The concentrations of vitamin C, a well-known antioxidant, vary by plant part; in decreasing order of concentration, the parts are: pulp > skin > leaf > stalk for organic beet and pulp > stalk > skin > leaf for conventionally grown beet. This suggests that the method of cultivation influences the concentration of ascorbic acid in a specific part of the plant. Part-specific concentrations were observed in beets from organic and conventional cultivation. However, the highest level of vitamin C was detected in each part of plants fertilized with organic manure ^[40].

7. Other Compounds

Nitrate (NO_3^-) is one of the most important inorganic compounds in red beets, and their ingestion provides a natural means of increasing in vivo nitric oxide (NO) ^[8]. This can prevent several pathologies such as hypertension

and endothelial dysfunction ^{[45][46]}. For a long time, nitrates, which can be converted into nitrosamines, were thought to be harmful substances. These, in turn, lead to endocrinological diseases, defects in human fetuses or cancer ^[47]. Today it is known that red beetroot nitrates are one of the most important nutrients. In general, nitrates provide ergogenic and cardio-protective properties. Nitrates present in beetroots are converted into nitrite and nitric oxide, which are responsible for lowering blood pressure and vasoprotection ^[48]. Jonvik et al. reported that beetroot juice lowers blood pressure to a greater extent than sodium nitrate ^[49]. Many studies have focused on the impact of beetroot juice consumption on the body's efficiency, mainly during intense physical exercise.

Volino-Souza et al. performed a randomized, clinical, crossover, double-blind study which has shown that the consumption of 140 mL of juice from red beets improved macrovascular endothelial function ^[50]. However, no oxygen saturation parameters in muscle tissue of pregnant women were observed. Other studies showed that one week of supplementation with beetroot juice improved submaximal endurance and reduced blood pressure in older patients with heart failure and preserved ejection fraction ^[51]. Subsequent works emphasize the beneficial effect of beetroot supplementation on the body's efficiency during physical exertion, as well as its impact on the speed of regeneration ^{[52][53]}.

The content of nitrates in beetroot depends on its variety. Wruss et al. analyzed nitrates content in seven beetroot varieties grown in Upper Austria ^[31]. The highest content was detected in the Mona Lisa variety (4626 ± 568 mg·L⁻¹), while the lowest was found in Robuschka ($564 \pm 129 \text{ mg·L}^{-1}$). The mean value across all analyzed cultivars was 1970 mg·L⁻¹, but the established standard deviation was 1395 mg·L⁻¹, which demonstrates the variability of nitrate content among beet varieties. Gallardo and Coggan studied the nitrate and nitrite content of beet juice products marketed to athletes ^[54]. The NO₃⁻ concentration of powders was significantly higher than that of concentrates ($174 \pm 63 \mu$ mol·g⁻¹ and $70 \pm 39 \mu$ mol·mL⁻¹, respectively). A lower concentration of nitrates was found in mixed drinks ($13 \pm 5 \mu$ mol·mL⁻¹) and bulk juices ($18 \pm 11 \mu$ mol·mL⁻¹). Authors highlighted that regardless of the type of product, there was considerable variability in NO₃⁻ concentration/content between products and, often, between samples of the same product ^[53]. The evaluation of nitrate and nitrite contents of beetroot from different regions of Brazil and the USA showed the highest nitrate ($31.2 \pm 0.010 \text{ mmol·L}^{-1}$) and nitrite ($0.45 \pm 0.005 \text{ mmol·L}^{-1}$) contents in US beets when compared to beetroots from Brazil ^[55]. In Brazil, Rio de Janeiro was the region that showed the highest nitrate content ($17.1 \pm 0.020 \text{ mmol·L}^{-1}$), while Rio Grande do Norte presented the highest nitrite content ($0.13 \pm 0.010 \text{ mmol·L}^{-1}$).

Red beetroots have more sugar than many other vegetables. According to Wruss et al., its average total content was 77.5 \pm 10.2 g·L ⁻¹ (7.8%) ^[31]. In the roots, sucrose was the most commonly identified sugar (94.8%), followed by glucose (3.3%) and fructose (1.9%). However, glucose was the major sugar in the shoots ^[56]. Red beetroot can be considered as a part of a healthy diet for diabetic patients. It was found that raw, red beetroot consumption by diabetes mellitus type 2 patients for 8 weeks, has beneficial impacts on cognitive function, glucose metabolism, and other metabolic markers ^[57]. The list of biologically active compounds in beets and beet products is presented in **Table 1**.

Unfortunately, in contrast to its health benefits, beetroots contain also oxalic acid; high levels of which lead to the production of kidney stones and their related negative health outcomes ^[58]. Moreover, oxalate, by chelating metal ions (such as magnesium and calcium), may inhibit their absorption. Thus, the consumption of red beetroots in large amounts may lead to negative health effects.

Compound	Concentration *	Sample	References
Betalains			
	128.7 ± 22.0	beet	[<u>8]</u>
	797 - 421.7	beet juices	[25]
Betanin	797 ± 24.0	Organic beet juice	[<u>59</u>]
	406 ± 17.0	Conventional beet juice	[<u>59</u>]
	705 ± 156	Beet extract	[<u>31</u>]
	321 - 432.1	Beet juices	[25]
	424 ± 16.0	Organic beet juice	[<u>59</u>]
Vulgahantin I	311 ± 13.0	Conventional beet juice	[<u>59</u>]
	397 ± 100	Beet extract	[<u>31]</u>
Flavonoids			
	0.27 ± 0.091	Organic beets	[<u>28]</u>
Myricetin	0.30 ± 0.109	Conventional beets	[<u>28]</u>
	0.14 ± 0.004	Organic beets [28]	
Luteolin	0.13 ± 0.003	Conventional beets	[28]
	0.13 ± 0.017	Organic beets	[28]
	0.010 ± 0.009	Conventional beets	[<u>28</u>]
Quercetin	0.0023	Fresh red beets	[29]
	0.009	Commercial juice	[29]
Epicatechin	3.20	Intact beet	[<u>11</u>]
	2.1 ± 0.100	Commercial juice	[25]

Table 1. The content of bioactive compounds in red beets and beet juices.

Compound	Concentration *	Sample	References
	0.253	Fresh beet	[<u>29]</u>
	0.202	Fermented beet	[<u>29</u>]
	0.034	Commercial juice	[<u>29]</u>
	0.715 ± 0.018	Commercial juice	[25]
Catechin	6.73 ± 0.031	Organic juice	[25]
Polyphenolic acids			
	36.40 ± 23.77	Organic beet	[<u>28]</u>
	65.93 ± 45.38	Conventional beet	[<u>28]</u>
Gallic acid	0.147± 0.008	Commercial juice	[25]
	1.24 ± 0.054	Organic juice	[25]
	1.70 ± 0.55	Beet juice	[<u>60</u>]
	4.67 ± 3.67	Organic beet	[<u>28</u>]
Chlorogenic acid	2.29 ± 2.09	Conventional beet	[<u>28]</u>
	1.80	Intact beet	[<u>11</u>]
	2.22 ± 0.75	Beet juice	[<u>60</u>]
	2.40 ± 0.050	Commercial beet juice	[25]
	0.900 ± 0.008	Organic beet juice	[25]
Caffeic acid	Caffeic acid 0.74 ± 0.40 Organic	Organic beet	[28]
	0.77 ± 0.28	Conventional beet	[28]
	3.70	Intact plant	[<u>11</u>]
	0.120 ± 0.005	Commercial beet juice	[25]
	1.81 ± 0.062	Organic beet juice	[25]
Ferulic acid	0.54 ± 0.37	Organic beet	[<u>28</u>]
	1.71 ± 0.76	Conventional beet	[28]
рНВА	1.2	Intact plant	[<u>11</u>]

Compound	Concentration *	Sample	References
	4.03 ± 0.053	Commercial beet juice	[<u>25</u>]
	6.83 ± 0.095	Organic beet juice	[<u>25</u>]
p-coumarc acid	5.27 ± 0.98	Beet juice	[<u>60</u>]
Sinapic acid	1.99 ± 0.80	Beet juice	[<u>60</u>]
	4.55 ± 2.16	Organic beet	[<u>28</u>]
	5.08 ± 2.10	Conventional beet	[<u>28]</u>
Vitamin C	4.36	Red beet	[<u>39]</u>
	7.20	Red beet	[8]
	4.90	Red beet	[<u>43</u>]
Vitamins B			
	0.034	Red beet	[<u>39</u>]
Riboflavin (B2)	0.040	Red beet	[<u>43</u>]
	2.85 ± 0.064	Commercial beet juice	[<u>25</u>]
	2.43 ± 0.040	Organic beet juice	[<u>25</u>]
Nicotinamide (B3)	0.334	Red beet	[<u>43</u>]
	0.030	Red beet	[<u>39]</u>
	2.49 ± 0.041	Commercial beet juice	[25]
Pantothenic acid (B5)	1.070 ± 0.047	Organic beet juice	[25]
	0.151 Red beets	[<u>39]</u>	
	1.420 ± 0.025	Commercial beet juice	[25]
	1.67 ± 0.038	Organic beet juice	[25]
Pirydoxal (B6)	90	Red beets	[<u>39</u>]
	0.067	Red beet	[<u>43</u>]
Folate (B9)	0.109	Red beet	[<u>8]</u>
Selenium compounds			

Compound	Concentration *	Sample	References
Selenomethionine	0.56 ± 0.020	Organic beet juice	[<u>59</u>]
	0.20 ± 0.01	Conventional beet juice	[<u>59</u>]
Methylselenocysteine	0.08 ± 0.03	Organic beet juice	[<u>59]</u>
	0.20 ± 0.01	Conventional beet juice	[<u>59]</u>
Selenocysteine	0.27 ± 0.02	Conventional beet juice	[<u>59]</u>
Vinerals			
Iron	2.57	Beetroot leaves	[8]
	0.80	Tubers	[8]
Connor	0.191	Leaves	[8]
Copper	0.075	Tuber	[8]
7:	0.365 ± 0.015	Tubers	[8]
Zinc	0.38	Leaves	[8]
Magnesium	23.0	Tubers	[8]
	70	Leaves	[8]
Carotenoids			<,
A-carotene	22.0 ± 2.0	Tubers	^[8] ab
	3.50 ± 0.5	Leaves	[8]
B-carotene	0.012	Leaves	[<u>39]</u> 3C
	0.001	Leaves	[<u>39]</u>
Lycopene	0.030	Tubers	<u>8</u> , 8
Lutein + zeaxanthin	0.001	Leaves	<u>8</u> as

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