Rubi idaei fructus as a Source of Antioxidants

Subjects: Plant Sciences

Contributor: Mirosława Chwil , Renata Matraszek-Gawron , Mikołaj Kostryco , Monika Różańska-Boczula

Given the increased incidence of lifestyle diseases, scientists are searching for natural bioactive compounds with a broad spectrum of activity but no side effects to be used in the production of pharmaceutical and cosmetic formulations. *Rubi idaei fructus* is a promising source of antioxidants that can serve as substitutes for synthetic agents in prophylaxis and adjuvant therapies.

raspberry Rubi idaei fructus bioactive compounds fatty acids amino acid flavonoids

1. Oxidative Stress and Its Effects and Defense Mechanisms against ROS

Oxidative stress plays an important role in the etiology and course of lifestyle diseases. It is associated with a disturbed balance between oxidants and antioxidants having an ability to detoxify the highly reactive by-products of metabolic transformations [1][2]. Reactive oxygen (ROS) and reactive nitrogen (RNS) species include oxygen free radicals and non-radical derivatives [3][4]. The most important sites of ROS generation in a plant cell include the chloroplasts, peroxisomes, and mitochondria. Due to the large amounts of ROS formed in the chloroplasts, proteins involved in photosynthetic electron transport are often oxidized [4][5]. At moderate concentrations, ROS and RNS mediate the regulation of cell signaling transduction processes, and their excessive production activates nucleases, damages (deoxyribonucleic acid) DNA, and results in cytotoxic activity [6][7][8]. Oxidative stress damages lipids and oxidizes cell membranes, changes the structure and modifies the functions of proteins due to the oxidation of amino acid residues, and damages carbohydrates by breaking the glycosidic bonds of polysaccharides [9][10]. It reduces adenosine triphosphate (ATP) production via the inhibition of oxidative phosphorylation, leading to the oxidation of low-molecular compounds [11][12], and induces the formation of free radical oxidation products, resulting in apoptotic cell death [13][14].

The enzymatic system protecting cells from ROS includes superoxide dismutase, which catalyzes $O_2^{\bullet-}$ dismutation, and catalase participating in H_2O_2 disproportionation [15]. Enzymes associated with the glutathione–ascorbic pathway, i.e., ascorbate peroxidase, dehydroascorbate reductase, and glutathione reductase, are involved in ROS detoxification as well [16][17]. In the non-enzymatic system, the small-molecular antioxidant compounds acting as scavengers are represented by membrane-active lipophilic carotenoids, flavonoids, and vitamins C and B_6 [18][19]. Vitamin C scavenges $^1O_2$, $OH^{\bullet}$, $O_2^{\bullet-}$ superoxide radicals, and ONOOH; acts a cofactor for ROS detoxifying enzymes; and regulates cell metabolism systems [17][20].

2. Rubi idaei fructus as a Source of Antioxidants

Recently, interest in berries has significantly increased together with the increased consumption associated with their high nutritional value and phytotherapeutic properties. *Rubi idaei fructus* is a promising source of antioxidants that can serve as substitutes for synthetic agents in prophylaxis and adjuvant therapies [21][22]. *R. idaeus* is an important species for consumption, processing, medicinal, and cosmetic purposes. The majority of commercial cultivars derive from red raspberry [23]. On a global scale, the cultivation area of this species is expanding [24][25][26]. New fruit storage technologies contribute to the maintenance of an unchanged chemical profile, i.e., the content of anthocyanins, vitamins, and tannins. These phytochemicals exhibit antioxidant [27], anti-inflammatory [28], antibacterial [29], immunomodulatory [30], and anticancer activity [31].

3. Use of R. idaeus Fruits

Due to their rich chemical composition and pro-health (e.g., antioxidant) activity, *R. idaeus* fruits are an important component of diets [32], functional foods [33], nutraceuticals [34], supplements [35], medicines [36], and natural cosmetics [37]. The bioactive compounds contained in raspberry fruit exert a wide range of beneficial phytotherapeutic effects [38]. The fruits are recommended as part of the prophylaxis of metabolic diseases, e.g., cardiovascular diseases [24], diabetes [39], obesity [40], and Alzheimer's disease [41]. The anti-inflammatory activity of *Rubi idaei fructus* is associated with the inhibitory effect of its compounds on lipoxygenase and cyclooxygenase-2 activity [28]. Additionally, extracts from red raspberries can be used in the prophylaxis and treatment of *Helicobacter pylori* infection [42]. A combination of prebiotics and active chemical compounds from *Rubi idaei fructus* is a natural antimicrobial agent to be used in the production of functional foods and in other branches of the food industry [43]. Raspberry fruit extracts may serve as chemotherapeutic agents, as they have been found to inhibit the migration and invasion of nasopharyngeal cancer cells, the expression of MMP-2, and the ERK1/2 signaling pathway [44]. *Rubi idaei fructus* fruits are characterized by high antioxidant activity [45][46][47]. These raw materials contain enzymatic and non-enzymatic antioxidants inducing the action of enzymes [28][46][48].

Biologically active phenolic compounds are mainly regarded as the most abundant antioxidants in the diet [49]. The total content of phenolic compounds determined with the Folin–Ciocalteau method confirms that raspberry fruits are a good source of antioxidant secondary metabolites [46][50][51]. The methods used, i.e., the ferric reducing antioxidant power (FRAP), 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical, 2,2'-azinobis-(3-ethyl-benzothiazoline-6-sulfonic acid) (ABTS) radical, and $OH^{\bullet}$ scavenging assays, confirm the high free radical scavenging activity of secondary metabolites from *Rubi idaei fructus* raw material [28][29][47][50][51][52][53][54][55]. This indicates that raspberry fruits can be used as a valuable nutritional component in the adjuvant therapy of oxidative stress-related diseases.

4. Selected Methods for the Determination of Oxidative Activity

The comparative analysis of antioxidant properties is based on several oxidation–reduction reactions taking place between an oxidant (a reagent appropriate for a given method, e.g., ABTS cation radical, DPPH, OH radical, FRAP reagent, and Folin) and a reducer, i.e., the sum of antioxidant compounds present in the raw material, e.g., *Rubus idaei fructus*. The content of antioxidants present in a sample is determined based on the decrease or increase in the absorbance of the reaction mixture measured spectrophotometrically at a specific wavelength. The Folin–Ciocalteau procedure is based on the use of Folin's reagent, i.e., a mixture of sodium tungstate (Na_2WO_4), sodium molybdate (Na_2MoO_4), lithium sulphate (Li_2SO_4), bromine water, and concentrated hydrochloric acid and phosphoric acid [56]. In the ABTS and DPPH methods, 2,2'-azinobis(3ethylbenzothiazoline-6-sulfonate) and 2,2-diphenyl-1-picrohydrazyl reagents, respectively, react with antioxidants contained in the raw material and cause discoloration of cation radicals. The degree of discoloration is proportional to the content of the antioxidant compound and is monitored spectrophotometrically at 734 nm and 515 nm wavelengths [57][58]. The FRAP method involves the reaction of the iron-2,4,6-tripyridyl-S-triazine complex obtained from the reaction of 2,4,6-tripyridyl-S-triazine (TPTZ) with iron chloride ($FeCl_3$) in acetic buffer (pH 3.5) with antioxidants present in the raw material. The complex becomes intensely blue, and its concentration is monitored using a spectrophotometer at a wavelength of 593 nm. The principle of the $OH^\bullet$ radical-based method is to produce the radical in the Fenton reaction in a medium with iron sulfate, hydrogen peroxide, and sodium salicylate, measured at a wavelength of 562 nm [59]. These methods facilitate a comparative analysis of the antioxidant capacity of selected raw materials, e.g., *R. idaeus* fruits and juice.

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