




Determination of the antioxidant and mineral contents of raspberry varieties

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Abstract

Humans maintain their health by consuming a variety of vegetables and fruits that contain antioxidants, both enzymatic and no enzymatic. Raspberry is one of the most diverse genus of true dicotyledonous plants, which includes 12 subspecies and about 429 species. Raspberry fruit is rich in antioxidant compounds, especially polyphenols. Two species of raspberry were studied to determine the amount of antioxidants and phenolic and flavonoid compounds in their fruits at three different stages of fruit ripening immature, semi-ripe and mature. Natural samples of *Rubus idaeus* and *Rubus strigosus* were collected. In this study, the fruit extracts of two species were stored at -23 °C for about six months. Free radical cleansing and Ferric reducing antioxidant power methods were used to determine the antioxidant activities of the extracts. The antioxidant activity of both methods revealed a higher mean value in extracts from fully matured fruits compared with immature and semi-ripe fruits. The results showed that the antioxidant activity of *Rubus strigosus* is 9%, 10%, and 8% higher than *Rubus idaeus* in the stages of immature, semi-ripe, and full maturity, respectively.

Keywords: raspberry; free radical; matured stage; *Rubus strigosus*; *Rubus idaeus*.

Practical Application: In the current study, it was tried to determine the antioxidant contents of raspberry varieties.

1 Introduction

Raspberry is one of the most diverse genus of true dicotyledonous plants, which includes 12 subspecies and about 429 species (Beekwilder et al., 2005; Chen et al., 2020). Its four species have the highest economic value as fruit species (Wang & Lin, 2000). This diversity is due to the wide types of fruits and pigments, such as anthocyanins in this genus, which cause red, blue, and purple colors in this genus (Sitpayeva et al., 2021). Raspberry fruit is a complex type and after pollination in the form of a small fruit called a buck to the national existence (Yang et al., 2019). The fruit is conical and produces fewer seeds, and sometimes there are secretory spheres on the shafts. Environmental conditions are often among the most important factors affecting a cultivar's fruit chemical composition (Urbina-Suarez et al., 2021). Raspberry species are widely distributed worldwide and have wild, cultivated species and different genotypes that are distributed from the North Pole to Australia (Vukoja et al., 2021).

The use of natural extracts or plant fragrances is one of the safest and safest ways to control diseases (Diaconeasa et al., 2019; Contreras et al., 2021). Raspberry fruit is rich in antioxidant compounds, especially polyphenols. Raspberries contain many compounds that include vitamins such as E, C, A, and acetic acid, mineral chloride, and Fe, Co, Mn, Al, Cu, Zn. The importance of natural antioxidants in human health and reducing the risks of heart disease, cancer, and diabetes has led to much attention being paid to the use of these compounds (Ponder & Hallmann, 2019; Tan et al., 2021). Phenolic compounds in plant samples are one of the best sources of natural antioxidants, and raspberry fruit is one of the most important sources of polyphenolic compounds that have received less attention so far (Yao et al., 2020). Raspberry phenolic compounds prevent oxidation and liposomes in the body and significantly eliminate free radicals (Elias et al., 2020; Kashchenko et al., 2021). The total antioxidant activity

Received 13 Oct., 2021

Accepted 21 Dec., 2021

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of raspberries, total phenols, and flavonoids was significantly higher than strawberries so that the antioxidant properties of raspberries were about twice that of strawberries (García et al., 2020; Vázquez-González et al., 2020). The amount of phenol and anthocyanin in raspberries is significantly higher than in cherries and plums. Total phenol content in the ripe berries is more than unripe fruits to handle the increased amount of phenolic compounds (Maro et al., 2013; Ishkeh et al., 2021).

Free radicals are electrophiles that damage cellular components and destroy proteins, lipids, and, most importantly, nucleic acids, and cause oxidative stress (Jiang et al., 2020; Kornobis, 2021). Increased oxidative stress in people with diabetes leads to a wide range of diseases such as retinopathy, nephropathy, neuropathy, cardiovascular disease, and sexual and hormonal disorders. To counteract oxidative stress, antioxidants are molecules in which single electrons or hydrogen atoms react to free radicals in oxidized cells and tissues (Maksimović et al., 2013; Tarasevičienė et al., 2020). As mentioned, raspberries are a rich source of natural antioxidants because it contains large amounts of phenols, flavonols, and anthocyanins and is, therefore, a free radical scavenger (Nardini & Garaguso, 2020; Szymanowska et al., 2021).

Due to the fact that sufficient studies on antioxidant activity have not been performed at present, the aim of this study was to investigate the protective effects of raspberry fruit extract on antioxidant activity and mineral performance. Since fruit variety and genotype affect the biologically active compounds of raspberries, so in this study, the antioxidant activity of two types (*R. idaeus* and *R. strigosus*) of raspberries has been investigated.

2 Material and methods

In this study, the fruits of two species of raspberries, *R. idaeus* and *R. strigosus* have been evaluated at different stages of fruit ripening. For this purpose, 1 g of harvested fruits was soaked in 23 mL of methanol solvent and crushed. The slaps were filtered, and for this purpose, a metabolic extract of raspberry fruits was prepared for experiments related to the measurement of antioxidants and the content of total phenol and total flavonoids. The extracts were stored in 2 mL microtubes at -23 °C in the laboratory until frozen for about six months until the beginning of the experiments. In this study, two methods were employed to assess the antioxidant activity. First, based on radical cleansing method and ferric reducing antioxidant power.

2.1 Radical cleansing method

This method placed 3.9 mL 2,2-diphenyl-1-picrylhydrazyl 0.1 mL of Heller extract in test tubes and kept in the dark for 33 minutes. After that, the absorbance of the solution was read at 517 nm. The percentage was released using (Equation 1) to calculate the radical inhibition (Brand-Williams et al., 1995).

$$SA (\%) = 100 \times \left(\frac{A_{blank} - A_{sample}}{A_{blank}} \right) \quad (1)$$

Where, A_{sample} and A_{blank} , SA(%), indicates sample adsorption, control adsorption, and the percentage of free radical scavenging, respectively.

2.2 Ferric reducing antioxidant power

According to this method, 2,4,6-Tripyridyl-s-triazine in the presence of antioxidants, it reduces the Fe^{2+} to Fe^{3+} solution turns purple. Depending on the amount of regenerative power, the absorption increases depending on the concentration of antioxidants. Using this change in absorption (increase in absorption at 593 nm), the antioxidant properties of various compounds can be evaluated (Lahlou, 2004; Rasheed et al., 1995). For this purpose, 1.5 mL of ferric reducing antioxidant power, 25 mL of acetate buffer, 2.5 mL of iron chloride III solution were mixed together. Raspberries were added to the test tubes containing the studied extracts and kept at 32 °C for 1 minute. Then 50 μ L of ferrous sulfate was added to the respective tubes and kept again for 13 minutes at 32 °C. The intensity of the blue color (bluish-purple) was read by measuring the absorbance at a wavelength of 593 nm in front of the control.

2.3 Assessment of total phenol content

The phenol content of the whole sample was measured using Folin Ciocalteu's phenol reagent. 0.1 mL of the diluted sample with 2 mL of Na_2CO_3 was poured into a test tube and kept at room temperature for two minutes. After that, 0.1 mL reagent was added to it. The reaction mixture was kept at room temperature for 33 minutes at room temperature and then readily absorbed in the visible region at a wavelength of 224 nm.

2.4 Measurement of total flavonoids

Flavonoid content was determined by the aluminum chloride colorimetric method. 213 μ L of the extract, 75 μ L of $NaNO_2$, 150 μ L of $AlCl_3$ and 500 of 3 μ L of $NaOH$ were added. The final volume was increased to 2.6 mL by adding distilled water. The adsorption of the solution was read after 1 minute at a wavelength of 2 nm by a spectrophotometer.

3 Results and discussion

In this section, the results related to the Mineral, Antioxidant, And Bioactive contents of Raspberry Varieties are investigated for the determination of the Antioxidant and Mineral contents.

3.1 Antioxidant activity of extracts based on the radical cleansing method

Analysis of variance of different traits related to antioxidant activity in two raspberry cultivars showed that the cultivar significantly affected all traits. The percentage of free radical scavenging in *R. idaeus* 55.2 to 81.4 mg/mL varied from immature to complete maturity, respectively (Figure 1). In *R. strigosus* from immature to complete maturity, this parameter varied from 56.1 to 89.2, respectively. There was a significant difference between different stages of immature, semi-ripe, and full maturity in both species. However, no significant difference was observed between the two studied species in terms of the percentage of free radical scavenging in the mating stage.

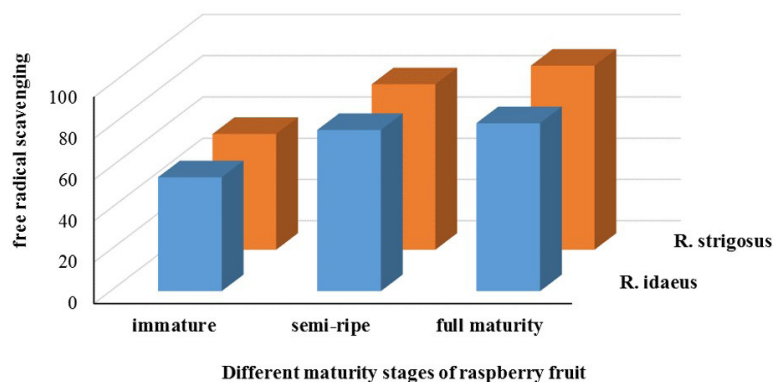


Figure 1. The level of antioxidant activity obtained by free radical scavenging.

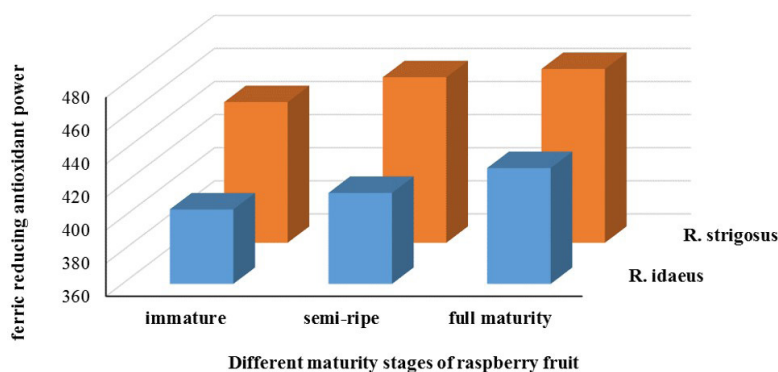


Figure 2. The level of antioxidant activity obtained by ferric reducing antioxidant power.

3.2 Antioxidant activity of extracts based on the ability to ferric reducing antioxidant power

Antioxidant activity was measured based on the ability to ferric reducing antioxidant power between different stages of immature, semi-ripe, and full maturity for *R. idaeus* 405, 415, and 430 mg/mL, respectively. While for *R. strigosus* ferric reducing antioxidant power between different stages of immature, semi-ripe, and full maturity was 445, 460, and 465 mg/mL, respectively (Figure 2). Comparison of the two species results shows a significant difference between the two at different stages. So that the antioxidant activity of *R. idaeus* was higher than *R. strigosus*, this issue can be clearly seen in all three stages of immature, semi-ripe, and full maturity.

3.3 Total phenol content

The total phenol content is completely different between different stages of immature, semi-ripe, and full maturity in both studied species. For *R. idaeus* in three immature, semi-ripe, and fully maturity states are 85, 93, and 96 mg per 100 g, respectively, and for *R. strigosus* in three fruit states are 125, 142, and 166 mg per 100 g, respectively (Table 1).

3.4 Total flavonoid content

The total phenol content is completely different between different stages of immature, semi-ripe, and full maturity in

Table 1. The total phenolic content.

	<i>R. idaeus</i> (mg per 100 g)	<i>R. strigosus</i> (mg per 100 g)
immature	85	125
semi-ripe	93	142
full maturity	96	166

Table 2. The total flavonoids content.

	<i>R. idaeus</i> (mg per 100 g)	<i>R. strigosus</i> (mg per 100 g)
immature	6	7
semi-ripe	7	8
full maturity	9	9

both studied species. For *R. idaeus* in three immature, semi-ripe, and fully maturity states are 6, 7, and 9 mg per 100 g, respectively, and for *R. strigosus* in three fruit states are 7, 8, and 9 mg per 100 g, respectively (Table 2). While in both species, there was no significant difference between full and semi-mature stages.

4 Conclusion

Temperature, humidity, respiration rate after fruit ripening, and susceptibility to fungal infections effectively reduce the shelf life of fruits, including raspberries. However, due to the high nutritional value of raspberries, the use of raspberry extract can be kept the high quality and healthy without compromising its nutritional properties. In this study, the fruit extracts of two species (*R. idaeus* and *R. strigosus*) were stored at -23 °C for about six months. Raspberry fruit extract was evaluated for its antioxidant properties at different developmental stages according to different tests. The results showed that the antioxidant activity in the full ripening stage was higher than other fruit ripening stages in both species. Meanwhile, the content of total phenol and total flavonoids was also higher in the full ripening stage than in other stages. In addition, antioxidant activity, total phenol content, and total flavonoid content between the two species at different stages of ripening show a significant difference.

The results obtained to evaluate the radical cleansing method's antioxidant properties and Ferric Reducing Antioxidant Power were similar. In both methods, in the two studied species, from an immature state to the stage of full maturity of the excretory axis of the anterior axillary effusion. Polyphenols are composed of hydroxy groups as electron donors. Polyphenols, as electron donors, make free radicals suspect stable radicals. In this study, the amount of phenolic compounds gradually increased from immature to full maturity in both studied species. Flavonoids have hydroxyl groups that counteract the oxidative stress created by free radicals through the process of scavenging or inactivating free radicals. These compounds are widely found in fruits and vegetables. In the present study, the highest levels of flavonoids were related to the semi-ripe and fully mature states in *R. strigosus*. The lowest ones were related to the immature states in *R. idaeus*. In both stages of full fruit ripening, raspberry fruit showed more antioxidant properties than others. In addition, both studied species showed more complete maturity than immature flavonoid heterocysts. Therefore, the consumption of fruit in the full ripening stage is recommended to fight against various cancers and diseases.

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