PHENOLIC COMPOUNDS IN SWEET CHERRY (*PRUNUS AVIUM* L.) PETIOLES AND THEIR ANTIOXIDANT PROPERTIES

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Abstract: *Sweet cherries (Prunus avium L.) contain various phenolic compounds which contribute to total antioxidant activity. Besides fruits, petioles also contains siginificant amount of phytonutrients and could be used for health-promoting herbal teas or infusions. Many dietary phenolics are known to provide health-improving benefits due to their various biological activities. The possible health beneficial effects included antioxidant, antialergic, anticarcinogenic, antimicrobial, antimutagenic, and antiinflamatory properties. Content of total polyphenols, tannins, and flavonoids and antioxidant capacity in a dry petioles of a number of selected sweet cherry genotypes were investigated spectrophotometrically. The differences in total polyphenolic contents, total tannins, antioxidant activity, and total flavonoid in petioles among sweet cherry genotypes were statistically significant. Total polyphenols content ranged from 12.96 to 31.85 mg gallic acid equivantents/g dry petioles weight and total tannins content ranged from 6.31 to 9.77 mg gallic acid equivalent/g dry petioles weight. Flavonoids have been knows to reduce oxidative stress in biological systems due to their antioxidant capacities. Total flavonoids were within the range 0.44-1.94 mg of rutin equivalents/g dry petioles weight. Cherries from the cultivars that are abundant in total phenolic content contained also more flavonoids. Antioxidant activity of sweet cherry petioles is related with the total polyphenolics, and flavonoids but not with tannins content. The highest total polyphenol and flavonoid content and antioxidant activity was observed in the petioles of Hedelfinger sweet cherry cultivar while the lowest antioxidant activity and total polyphenol and flavonoid content was recorded in the petioles of Rita cultivar. Genotypes with highrt poliphenolic contents also showed the higher DPPH-radicals scavenging activities. Petioles of sweet cherry fruits are a significant source of different phenolic compounds, and could be considered a good source of natural antioxidants. The difference in the sweet cherry genotypes in terms of total polyphenolics is due to genetic variations, as all genotypes were the same age and grown under the same ecological conditions.*

Key words: *Sweet cherry petioles, phenolics, tannins, flavonoids, antioxidant capacity*

INTRODUCTION

There is great interest in determining the role of compounds of plant origin, phytonutrients, in promoting improved health. Natural antioxidants found in foods may play an important role in preventing the formation of free radicals and the subsequent formation of lipid peroxides. Phenolic compounds have antioxidant properties and can protect against degenerative diseases (i.e., heart disease and cancer) in which reactive oxygen species (superoxide anion, hydroxyl radicals, and peroxy radicals) are involved (CAPECKA et al., 2005).

The general definition of phenolic compounds is any compound containing a benzene ring with one or more hydroxyl groups. Phenolic acids, flavonoids, condensed tannins, and coumarins are examples. Phenolics range from a simple, low molecular-weight, single aromatic ringed compounds to large and complex tannins and derived polyphenols. They can be classified based on the number and arrangement of their carbon atoms in flavonoids and non-flavonoids (DYKES AND ROONEY, 2007).

Most of the literature on plant phenolics focuses mainly on those in fruits, vegetables,

wines, and teas. In recent years, there been an increasing effort to find foods and beverages with high antioxidant contents and health-promoting properties. Herbs traditionally used in folk medicine have attracted consumer interest because of their long historical consumption and ready acceptability. Herbal teas have gained popularity all over the world due to their antioxidant activity. Over 80% of the world's population relies on largely plant-based traditional medicine for primary healthcare needs (KARAKAYA AND EL, 2006; HEONG et al. 2011). Herbal teas can be easily prepared from any part of a plant, including the roots, flowers, seeds, berries and bark. There are many proposals for the evaluation and exploitation of agricultural byprodacts as well (DIMITRIOUS, 2006). Sweet and sour cherry steam make an enjoyable infusion. In Serbia, this infusion is often taken for its detoxifying and diuretic properties.

The aim of this study was to determine the content of different phenolic compounds and antioxidant activity of dried petioles of 17 different sweet cherry cultivars.

MATERIAL AND METHODS

Fruits of sweet cherry cultivars were collected in 2010 from the productive orchard "Sloga" Kać in vicinity of Novi Sad, Serbia. Petioles of 17 red-coloured cultivars (Sándor, Katalin, Kavics, Rita, Margit, Peter, Linda, Aida, Alex, Carmen, Sunburst, Summit, New Star, Burlat (Bigarreau Burlat), Germerdorf 3, Hedelfinger and Majeva rana), were included in this study. Cherry fruits were picked at commercial maturity on the basis of fruit colour. Petioles were air-dried at ambiental temperature to the constant weight.

Dry plant material (1 g per sample) was ground to a fine powder and extracted with 70% aqueous acetone solution (50 mL) by sonication for 20 minutes in an ultrasonic bath at ambient temperature. The extracts were rapidly vacuum-filtered through a sintered glass funnel and kept refrigerated until assayed.

Total polyphenols were determined colorimetrically (Jenway 6505, UK) by Folin-Ciocalteu procedure (KROYER, 2004). Gallic acid (GAE) was used as a standard (covering the concentration range between 0.1 and 1.0 mg/mL) and results were expressed as miligrams of GAE per gram of dry plant material (DW).

Total tannins content was determined by the Folin-Ciocalteu procedure as above, after removal of tannins by their adsorption on insoluble matrix (polyvinylpolypyrrolidone) (HAGERMANN et al., 2000). Calculated values were subtracted from total polyphenol contents, and total tannin contents were expressed as milligrams of GAE per gram of DW.

Total flavonoids were determined according to the procedure of MARCKAM (1989). The amount of flavonoids was calculated as a rutin equivalent from the calibration curve of rutin standard solutions and expressed as milligrams of rutin per gram of DW.

The potential antioxidant activity of the test samples have been assessed based on scavenging activity of the 70% aqueous acetone sweet cherry extracts of the stable 1,1`-diphenyl-2-picrylhydrazyl (DPPH) free radicals (ABE et al., 1998). DPPH-radical scavenging activity was expressed as % of neutralized free radicals, assuming that the sample with the higher percentage has higher scavenging capacity. All measurements were done in triplicate.

Results were expressed as mean of determinations of 3 independent samples made in triplicates. Statistical significance was tested by analysis of variance followed by comparison of means by Duncan's multiple range test ($P<0.05$) calculated using STATISTICA for Windows version 9.0 (StatSoft, Tulsa, OK, USA). Stepwise multiple regression analyses were used to determine correlation among variables.

RESULTS AND DISSCUSION

The amount of total polyphenolics in the studied sweet cherry petioles are shown in

Table 1. The total polyphenolic contents of sweet cherry petioles were in the range of 12.96-31.85 mg gallic acid per g DW basis. The highest total polyphenolic content was in the petioles of Hedelfinger sweet cherry cultivar (31.85 mg/g), followed by the Katalin (28.31 mg/g) and Majeva rana (27.63 mg/g). The lowest content of total polyphenolic compounds was recorded in the petioles of Aida (12.96 mg/g) and Rita (13.13 mg/g) cultivars. Different medicinal plants and herbal teas contains from 7 mg/g up to 200 mg/g of polyphenolics (PRVULOVIĆ et al, 2005; NAITHANI et al, 2006; KUMARI AND KAKKAR, 2008; HEONG et al, 2011). Genotypes with highest polyphenol content in petioles has also the highest polyphenol content in ripe fruits as well (PRVULOVIĆ et al., 2011). The difference in the sweet cherry genotypes used in this study in terms of total polyphenolics in petioles is due to genetic variations, as all genotypes were the same age and grown under the same ecological conditions. Phenolic compounds serve in plant defense mechanisms, to counteract reactive oxygen species, in order to survive and prevent molecular damage, and damaging by microorganisms, insects and herbivores. Phenolic compounds, at low concentration, may act as an antioxidant and protect plant and plant-based food from oxidative deterioration. Phenolic compounds are important by their multiple biological effects such as antioxidant activity, antimutagenic and/or anticarcinogenic activities, and antiinflamatory action (GRASSMANN et al., 2002; KARAKAYA AND EL, 2006; BOECKLER et al., 2011).

Table 1.

Total polyphenols, tannins and flavonoids content, and antioxidant activity of petioles of different sweet cherry cultivars

Cultivar	Total polyphenols[a]	Tannins[a]	Flavonoids[b]	DPPH values[c]
Hedelfinger	31.85 ± 0.831	7.47 ± 0.607	1.94 ± 0.106	86.94 ± 1.045
Germerdorf 3	18.57 ± 0.607	6.58 ± 0.297	1.44 ± 0.075	54.62 ± 2.923
Majeva rana	27.63 ± 0.336	8.37 ± 0.458	1.21 ± 0.048	72.29 ± 1.045
Burlat	17.67 ± 0.573	7.66 ± 0.567	1.01 ± 0.056	56.42 ± 1.442
New Star	17.52 ± 0.719	7.60 ± 0.551	1.34 ± 0.025	51.27 ± 3.038
Sunburst	16.02 ± 0.341	7.68 ± 0.404	0.71 ± 0.129	41.82 ± 3.462
Summit	21.12 ± 0.117	8.36 ± 0.596	1.38 ± 0.117	57.01 ± 5.246
Margit	18.18 ± 0.576	6.98 ± 0.527	0.81 ± 0.099	52.49 ± 4.309
Sándor	17.74 ± 0.341	7.06 ± 0.556	0.81 ± 0.027	52.55 ± 1.618
Katalin	28.31 ± 0.620	9.77 ± 0.595	1.17 ± 0.080	76.54 ± 2.800
Kavics	26.33 ± 1.036	7.56 ± 0.353	1.94 ± 0.071	68.10 ± 2.465
Rita	13.13 ± 0.248	6.87 ± 0.321	0.44 ± 0.101	29.88 ± 3.130
Peter	23.84 ± 0.549	7.65 ± 0.573	1.28 ± 0.045	64.06 ± 2.050
Linda	20.00 ± 0.500	9.00 ± 0.572	0.74 ± 0.040	56.42 ± 3.958
Aida	12.96 ± 0.432	6.18 ± 0.309	0.76 ± 0.076	36.71 ± 1.464
Alex	16.75 ± 0.545	6.31 ± 0.309	0.67 ± 0.050	42.73 ± 2.686
Carmen	20.40 ± 0.831	6.32 ± 0.364	1.43 ± 0.129	59.23 ± 1.688

Data are mean ± SD values
[a] Expressed as mg of gallic acid equivalents/g of dry plant material.
[b] Expressed as mg of rutin/g of dry plant material.
[c] Expressed as % of neutralised DPPH free radicals.

Differences among the examined petioles of sweet cherry cultivars, regarding their content in total tannins, are not signifficant (Table 1). They range from 6.18 mg GAE/g DW for cultivar Aida, and 6.31 mg GAE/g DW for cultivar Alex, up to 9.00 mg GAE/g DW for cultivar Linda and 9.77 g GAE/g DW for cultivar Katalin. Tannins are widely distributed in the plant kingdom. Tannins are feeding deterrentes to many invertebrate and vertebrate herbivores. Feeding deterance is undoubtly an important mechanism by which tannins protect plant from non-adapted animals. For adapted species, tannins can act as stimulants. The biochemical

activities of tannins range from beneficial antioxidants to damaging prooxidants and toxins. The concentration of tannins varies with plant genotype, tissue developmental stage, and environmental conditions (BARBEHENN AND CONSTABEL, 2011).

The genotype influence the extent of total flavonoid accumulation in the sweet cherry fruits. The contents of flavonoids found in sweet cherries are given in Table 1. The genotypes with high flavonoid contents are Kavics, and Hedelfinger with 1.94 mg of rutin equivalents/g DW. The cultivar with lowest total flavonoid content is Rita with 0.44 g of rutin equivalents/g DW. Flavonoids are present in vegetables, fruits, and beverages derived from plants. Flavonoids have been described as health-promoting and disease-preventing dietary supplements. They are safe and associatd with low toxicity (MOON et al., 2006). Flavonoids have been knows to reduce oxidative stress in biological systems due to their antioxidant capacities (KIM et al., 2005). Most flavonoids are found in nature as *O*- or *C*-glycosides. The glycosylation is important to reduce the reactivity and to increase the water solubility of flavonoids, which in turn prevents their cytoplasmic damage and guarantees their storage in the cell vacuole (CUYCKENS AND CLAEYS, 2005). The presence of glycosides attached to flavonoid aglycons, such as flavonol or anthocyanidin, decreases the antioxidant activity of flavonoid. The reason for this is the glycoside moiety, which interferes with the coplanarity of the flavonoid molecule, decreases the ability to delocalise electrons and by that decreases the antioxidant activity of flavonoid (HEIM et al., 2002).

The antioxidant activity using DPPH method in sweet cherry genotypes are shown in Table 1. A statistical significant difference was found among genotypes. The DPPH-values for investigated extracts varied in a wide range between 29.88% and 86.94% (Table 1). The highest antioxidant activity was observed in Hedelfinger genotype at 86.94%, followed by Katalin genotype (76.54%), and Majeva rana genotype (72.29%). Lowest antioxidant activity was measured in Rita genotype (28.99%) and Aida genotype (36.71%). It is worh mentioning that genotypes with highest poliphenolic contents also showed the highest DPPH-radicals scavenging activities. At the same time, genotypes with the lowest content of total phenolics, such as Rita and Aida, expressed less than half activity of phenolic-rich genotypes. The relationship between antioxidant capacity and different phenolic groups varried between cultivars.

CONCLUSIONS

As the conclusion, this investigation show large variability between petioles of different sweet cherry cultivars in measured chemical attribultes. Antioxidant activity of most cultivars strongly depends on phenolics, and flavonoids, but is not in correlation with the content of tannins. Sweet cherry fruit petioles are a significant source of different phenolic compounds, and could be considered a good source of natural antioxidants.

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