

PHYTOCHEMICAL CHARACTERISTICS AND ANTIOXIDANT CAPACITY OF FRUIT EXTRACTS OF DIFFERENT *PRUNUS* SPECIES

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Abstract

Fruits are one of the major sources of polyphenol compounds in human diet. These compounds are known to have many health-promoting activities, especially anticancer, antiradical and antioxidant effects. In this work, seven different *Prunus* species traditionally grown in south Bačka region of Vojvodina were investigated: blackthorn (*P. spinosa*), plum (*P. domestica*), apricot (*P. armeniaca*), cherry plum (*P. cerasifera*), sweet cherry (*P. avium*), sour cherry (*P. cerasus*) and mahaleb cherry (*P. mahaleb*). Freeze-dried fruits were evaluated in terms of their phytochemical characteristics and bioactivity, determining total content of phenolics, flavonoids and anthocyanins, total antioxidant capacity and antiproliferative effect on human colon cancer cells (HT29). Blackthorn fruits are the richest in phenolic and flavonoid contents, while mahaleb cherry and sweet cherry had much higher content of total anthocyanins than other examined species. Apricot and cherry plum fruits had the lowest levels of polyphenol compounds, but very high antiproliferative effect, almost the same as blackthorn. This indicates that not only polyphenol compounds contribute to antiproliferative effects. Concerning total antioxidant activity, blackthorn, sweet cherry, sour cherry and mahaleb cherry showed the highest capacity of scavenging DPPH radical and ferric reducing activity power.

Introduction

Regular consumption of fruits has been associated with reduced risk of developing cancer, neurodegenerative diseases, cardiovascular diseases, diabetes and other chronic diseases. These benefits are often attributed to their various phytochemical content and strong antioxidant activity. Fruits are abundant in phenolic compounds, plant secondary metabolites, which greatly contribute to their health promoting effects.^{1,2} Health effects of polyphenols are often attributed to their antioxidant activity which is mediated by a variety of mechanisms, including reduction or scavenging of ROS, chelation of transition metal ions and inhibition of enzymes involved in oxidative stress.³

Prunus L. genus belongs to Amygdaloideae (or Prunoideae) subfamily of Rosaceae family. The subfamily Amygdaloideae differs from other rosaceous subfamilies by having a drupe, a fleshy fruit with a stony endocarp or stone. This genus includes the plums, cherries, peaches, apricots and many other stone fruits which are widely consumed and present good sources of phytochemicals in human diet.⁴

There are earlier reported many bioactivities and phytochemicals of *Prunus* species.⁵⁻⁸ Within this context, the aim of this work was to evaluate and compare the antioxidant and antiproliferative effects of seven *Prunus* species traditionally grown in Serbia, in south Bačka region of Vojvodina, in an effort to distinguish promising functional fruits.

Experimental

It were assessed freeze-dried fruit extracts of seven different *Prunus* species: blackthorn (*P. spinosa*, genotype B1), plum (*P. domestica*, cultivar Čačanska rodna), apricot (*P. armeniaca*, cultivar DM), cherry plum (*P. cerasifera*, wild type), sweet cherry (*P. avium*, wild type), sour cherry (*P. cerasus*, cultivar Oblačinska) and mahaleb cherry (*P. mahaleb*, wild type). For spectrophotometric assays 50% acidic methanol (1% HCOOH) fruit extracts were prepared and water extracts were used for determination of antiproliferative activity.

Total phenolic content (TPC) in fruit extracts was determined spectrophotometrically according to the Folin–Ciocalteu method.⁹ The results were expressed as milligram of gallic acid equivalents per gram of dry weight (mg GAE/g DW).

Total flavonoid content (TFC) was measured using aluminium chloride assay and results were expressed as milligram of quercetin equivalents per gram of dry weight (mg QE/g DW).¹⁰

Total anthocyanin content (TAC) was determined by pH differential method and expressed as cyaniding-3-glucoside equivalents per gram of dry weight (mg CGE/g DW).¹¹

Total antioxidant activity was determined using scavenging effect on 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical¹² and measuring the ferric reducing activity power (FRAP).¹³

Results of DPPH assay were given as reciprocal value of IC₅₀ (the concentration of extract required to scavenge 50% of radical) and for FRAP test in milligram of ascorbic acid equivalents per gram of dry weight (mg AAE/g DW).

Antiproliferative activity (APA) assay was performed using human colon cancer cells HT29.¹⁴ Cell proliferation was determined using the colorimetric MTT assay. Results were expressed as reciprocal value of ED₅₀ (effective dose), the amount of sample necessary to decrease 50% of the cellular viability.

Results and discussion

In this present work, fruits of seven *Prunus* species, were compared by the content of phenolic compounds and antioxidant activities, as well as antiproliferative potential against human cancer cells from colon (HT29).

The content of total phenolic, flavonoid and anthocyanin compounds are presented in Table 1. Blackthorn fruits were the richest in phenolic and flavonoid contents (30.32 mg GAE/g DW and 3.24 mg QE/g DW), while mahaleb cherry and sweet cherry had much higher content of total anthocyanins (11.11 and 9.76 mg CGE/g DW) than other examined species.

Table 1. Total phenolic, flavonoid nad anthocyanin contents of fruit extracts*

Common name	Latin name	TPC (mg GAE/g DW)	TFC (mg QE/g DW)	TAC (mg CGE/g DW)
blackthorn	<i>P. spinosa</i>	30.32 ± 3.70	3.24 ± 0.09	4.65 ± 0.08
plum	<i>P. domestica</i>	8.79 ± 0.70	0.59 ± 0.01	0.25 ± 0.02
apricot	<i>P. armeniaca</i>	3.60 ± 0.06	0.22 ± 0.01	0.01 ± 0.00
cherry plum	<i>P. cerasifera</i>	2.92 ± 0.09	0.20 ± 0.01	0.18 ± 0.01
sweet cherry	<i>P. avium</i>	20.04 ± 0.34	2.68 ± 0.02	9.76 ± 0.00
sour cherry	<i>P. cerasus</i>	17.76 ± 0.03	0.95 ± 0.01	3.94 ± 0.08
mahaleb cherry	<i>P. mahaleb</i>	18.82 ± 0.12	3.64 ± 0.14	11.11 ± 0.28

*Values are expressed as means of three replications ± standard deviation.

Abbreviations: TPC-Total phenolic content; TFC-Total flavonoid content; TPC-Total anthocyanin content; GA-Gallic acid, Q-Quercetin; CG-Cyanidin-3-glucoside; E-equivalents.

Extracts of all fruits showed good antioxidant and antiproliferative activities, but blackthorn was distinguished as much better than the others, especially in scavenging DPPH radical and

ferric reducing antioxidant power as shown in Figure 1. Very high antioxidant activity was also noticed for sweet cherry, sour cherry and mahaleb cherry fruit extracts.

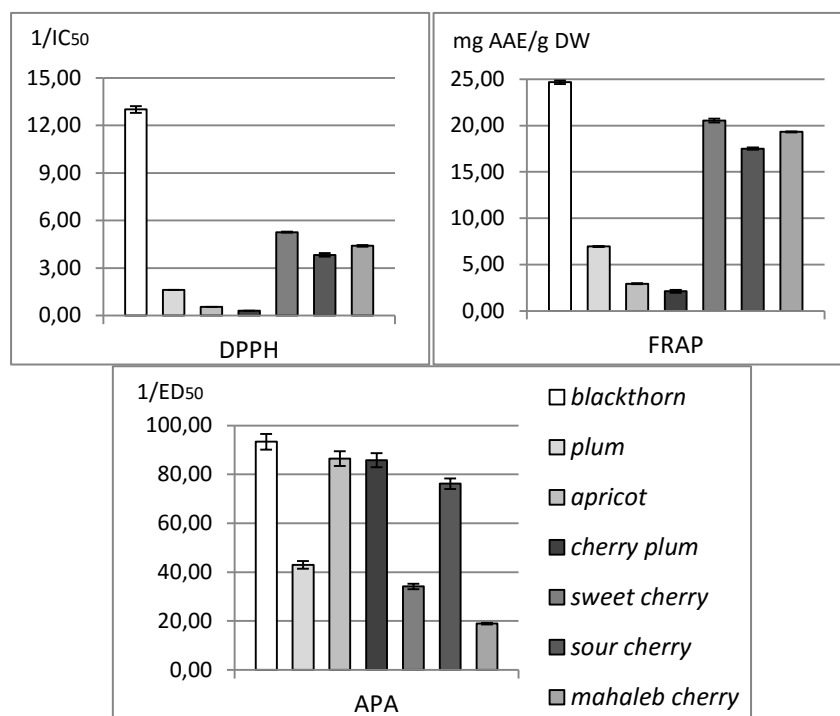


Figure 1. Scavenging activity on DPPH radical, ferric reducing antioxidant power (FRAP), and antiproliferative activity (APA) on human colon cancer cells HT29 of different *Prunus* species.

The lowest values of total phenolics, flavonoids and anthocyanins were assessed in apricot and cherry plum fruit extracts and these two fruit extracts showed much lower antioxidant capacity comparing to other species. On the other hand, apricot and cherry plum fruit extracts very strongly inhibited proliferation of HT29 cells, almost as strong as blackthorn, which was the most effective species (Figure 1). This suggests that not only polyphenol compounds contribute to antiproliferative effect, but some other phytochemicals, as well.

Conclusion

The present investigation was carried out to compare phytochemical characteristics and antioxidant and antiproliferative activities of seven *Prunus* species commonly used in human diet. According to obtained data, all examined *Prunus* species showed remarkable antioxidant and antiproliferative effects. Results obtained for apricot and cherry plum fruits, indicates that not only polyphenol compounds contribute to antiproliferative effect, because these two fruit extracts greatly inhibited proliferation of HT29 human colon cancer cells, but were not so abundant with total phenolic compounds and did not show so high antioxidant capacity as the other ones. Among all species, blackthorn was distinguished by the best potential to be used as a source of functional ingredients, but the potential of other species, should not be neglected, also.

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