Full Length Research Paper

Selected cultivars of cornelian cherry (*Cornus mas* L.) as a new food source for human nutrition

Otakar Rop^{1*}, Jiri Mlcek¹, Daniela Kramarova² and Tunde Jurikova³

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The aim of this work was to determine antioxidant activity in 12 cultivars of cornelian cherry (*Cornus mas* L.). Two assays based on ion reduction of ABTS (2,2´-azinobis(3-ethylbenzothiazoline-6-sulphonate) and DPPH (2,2-diphenyl-1-picrylhydrazyl) radicals were used for antioxidant activity. Total phenolic content of the fruit was analysed by Folin-Ciocelteau colorimetric method and ascorbic acid content was analysed using column chromatography - electrochemical detector (Coulochem III). The highest amounts of total phenolic content were found in cultivars 'Vydubeckij' and 'Titus' as 7.96 and 8.11 g gallic acid kg⁻¹ of fresh mass (FM). High correlation between polyphenols and antioxidant activity in fruits of the cultivars was observed ($r^2 = 0.970$ for DPPH test and $r^2 = 0.978$ for ABTS test). The highest total content of ascorbic acid was determined in cultivar 'Olomoucky', with the value of 3.11 g kg⁻¹ FM. This study attempts to contribute to the knowledge of human nutritional properties of these cornelian cherry cultivars and may be useful for the evaluation of dietary information and further propagation of cultivation and utilization of this fruit in the world.

Key words: Cornelian cherry, phenolics, antioxidant activity, ascorbic acid.

INTRODUCTION

Cornelian cherry (*Cornus mas* L.) belongs to the family Cornaceae. It is a tall deciduous shrub or small tree from 5 to 8 m high. This plant is popular in southern Europe with the northern limit being southern Belgium and central Germany (Mamedov and Craker, 2004). In the Czech and Slovak Republic, it is spread in the area of the White Carpathian Mountains (Tetera, 2006).

Cornelian cherry is a widely distributed species in Europe and it grows up to 1400 m. The species is highly

Abbreviations: ABTS, 2,2´-Azinobis(3-ethylbenzothiazoline-6-sulphonate); **DPPH**, 2,2-diphenyl-1-picrylhydrazyl; **FM**, fresh mass.

tolerant to diverse abiotic and biotic conditions. The bloom time in Central European conditions begins early in the spring. The fruits are very valuable for fresh consumption and for processing to produce syrups, juices, jams (Brindza et al., 2007), spirits and other traditional products (Tesevic et al., 2009). The stone fruits are in full maturity continuously during early autumn (Kutina, 1991).

The cultivation of cornelian cherry in the Caucasus and Central Asia has occurred for centuries, mainly for food and medicine, but also as an ornamental plant (Mamedov and Craker, 2004). Nowadays, Turkey is an important centre of cornelian cherries (Ercisli et al., 2008), especially in northern Anatolia (Ercisli, 2004). Generally, the historical areas of cornelian cherry occurrence are important for an adaptation of some genotypes to different local conditions in different regions of several countries (Yilmaz et al., 2009a).

¹Department of Food Technology and Microbiology, Faculty of Technology, Tomas Bata University in Zlin, Namesti T. G.

Masaryka 275, 762 72 Zlin, Czech Republic.

²Department of Food Biochemistry and Analysis, Faculty of Technology, Tomas Bata University in Zlin, Namesti T. G. Masaryka 275, 762 72 Zlin, Czech Republic.

³Department of Natural and Informatics Sciences, Faculty of Central European Studies, Constantine the Philosopher University in Nitra, Drazovska 4, 949 74 Nitra, Slovak Republic.

^{*}Corresponding author. E-mail: rop@ft.utb.cz. Tel: +420 576 031 129. Fax: +420 577 210 172.

The aim of our work was to monitor antioxidant activity. total phenolic content and the content of ascorbic acid in cultivars of cornelian cherry. The consumption of foodstuffs with a high amount of antioxidant compounds has a positive impact on human health, particularly the prevention of cancer and other inflammatory diseases. Only little information about antioxidant activity and main bioactive components in particular cultivars of cornelian cherries is available in scientific works. 12 cultivars of cornelian cherries were investigated. The cultivars used were ´Elegantnyj´, Jalt´, 'Kijevskij', 'Lukianovskij', 'Vydubeckij' which are Russian in origin, 'Devin', 'Olomoucky, 'Ruzynsky', 'Sokolnicky', 'Titus' which are Czech and Slovak in origin and 'Joliko' and 'Fruchtal' which are Austrian in origin (Tetera, 2006). The main aim of our work was to popularize this fruit species for propagation in other continents in the world and draw attention to the potential of European cultivars as a new promising fruit species.

MATERIALS AND METHODS

Locality description and collection of samples

Fruits were harvested in experimental orchards of Tomas Bata University Zlin within the period of 2007 – 2009. These orchards are situated in the south-western part of the White Carpathians near Zlin, the Czech Republic. The average altitude is 340 m above sea level, and the mean annual temperature and precipitation are 7.9°C and 760 mm, respectively. The soil type was classified as the Mesotrophic Cambisol.

Fruit were harvested in consume ripeness from five trees of each cultivar under study (thus each year in 5 replications) in the course of September. 20 randomly chosen fruits from each tree were used for analyses (that is, altogether 100 per each cultivar).

Sample processing

The fruit of individual trees were processed immediately after the harvest (not later than within two days). Harvested fruits were poured in a mixer and the average sample was obtained by means of quantitation. Each parameter was measured in five replications from the fruit taken from each tree of particular cultivars (n = 25). The results were expressed as average of a three-year experiment.

Total phenolic content (TPC) and free radical scavenging assay

The extraction was performed according to the method described by Kim et al. (2003a), using 10 g of fresh sample which were homogenized for 10 s in an extraction mixture of hydrochloric acid: methanol: water in the ratio 2:80:18. For measurement of TPC, Folin-Ciocalteau reagent was used. The resulting absorbance was measured in the spectrophotometer LIBRA S6 at the wavelength of 765 nm against a blind sample, which was used as reference. The results were expressed as grams of gallic acid (GAE) kg-1 of fresh mass (FM).

Antioxidant activity was measured using the ABTS radical scavenging method described by Sulc et al. (2007). This test is

based on monitoring of the course of inactivation of the cation ABTS⁺, which is produced during the oxidation of 2,2'-azinobis(3-ethylbenzothiazoline-6-sulphonate). ABTS⁺ shows a strong absorbance in the visible region of the electromagnetic spectrum (600-750 nm); this solution is green and its antioxidant activity can be easily measured by means of spectrophotometry. DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging assay was carried out according to the method of Brand-Williams et al. (1995). This test is based on reduction of DPPH⁺ radical. In its radical form, DPPH⁺ absorbs light at 515 nm, but upon reduction by an antioxidant or a radical species, the absorption disappears (Thaipong et al., 2006). In both methods the calculated activity was converted using a calibration curve of the standard and expressed in ascorbic acid equivalents (AAE) (Rupasinghe et al., 2006).

Ascorbic acid content assay

The determination of ascorbic acid content was carried out by a modified method of Miki (1981). 5 g of the sample were extracted in an extraction mixture (methanol:H₂O:H₃PO₄ in the ratio 99:0.5:0.5). The instrument used for ascorbic acid analysis consisted of a solvent delivery pump (ESA Inc., Model 582), guard cell (ESA Inc., Model 5010A, working electrode potential K1 = 600 mV, K2 = 650 mV), chromatographic column - Model Supelcosil LC8 (150.0 x 4.6 mm), 5 μ m particle size and an electrochemical detector (Coulochem III). Chromatographic conditions were constant: 30°C, as a mobile phase methanol was used: H₂O:H₃PO₄ = 99:0.5:0.5, (filtrated through a filter Nylon, 0.2 μ m), type of elution was isocratic, the flow rate of the mobile phase was 1.1 ml min⁻¹, and retention time 1.9 - 2.0 min. The content of ascorbic acid was calculated as g kg⁻¹ of FM.

Statistical analysis

The data obtained were analysed statistically by an analysis of variance (ANOVA) and Tukey's multiple range test for comparison of means (Snedecor and Cochran, 1968) using the statistical package Unistat, v. 5.1.

RESULTS AND DISCUSSION

The results of chemical analyses of samples of cornelian cherry cultivars are shown in Tables 1 and 2.

The total phenolic content ranged from 2.61 to 8.11 g of gallic acid kg-1 of fresh mass. In connection with the decrease of total phenolic content, the free radical scavenging activity of the fruits extracts (a measure of the antioxidant activity) also reduced (see Table 1). The correlation coefficient between the total phenolic content and free radical scavenging activity of DPPH and ABTS were $r^2 = 0.970$ and 0.978, respectively (see Figures 1 and 2). Quite a number of authors (Rupasinghe et al., 2006; Moyer et al., 2002; Jurikova and Matuskovic, 2007) refers to high correlation dependence of polyphenols and antioxidant activity in different fruit species. What is more interesting, the highest content of phenolic substances and antioxidant activity was measured in the cultivars Titus' and 'Vydubeckij', which are Russian in origin. Minor differences in the ascorbic acid contents between

Table 1. Free radical scavenging activity (grams of ascorbic acid equivalent kg^{-1} FM) of extracts of fruits of different cultivars of cornelian cherry (*Cornus mas* L.), n = 25.

Cultivar name	DPPH radical scavenging activity	ABTS radical scavenging activity
Devin	3.30 ± 0.20^{a}	3.65 ± 0.28 ^a
Elegantnyj	4.11 ± 0.28 ^b	4.62 ± 0.32 ^b
Fruchtal	5.02 ± 0.25°	5.75 ± 0.24 ^c
Jalt	4.61 ± 0.31 ^{bc}	5.02 ± 0.27 ^b
Joliko	5.27 ± 0.24 ^c	5.95 ± 0.25 ^c
Kijevskij	6.83 ± 0.29 ^d	7.16 ± 0.32 ^d
Lukjanovskij	4.58 ± 0.24 ^b	5.04 ± 0.30 ^b
Olomoucky	6.94 ± 0.28 ^d	7.51 ± 0.25 ^d
Ruzynsky	6.85 ± 0.27 ^d	7.41 ± 0.38 ^d
Sokolnicky	4.61 ± 0.30 ^{bc}	4.94 ± 0.26 ^b
Titus	8.90 ± 0.25 ^e	9.64 ± 0.31 ^e
Vydubeckij	9.54 ± 0.32 ^e	10.28 ± 0.34 ^e

Different superscripts in each column indicate the significant differences in the mean at P < 0.05.

Table 2. Total phenolic content (grams of gallic acid.kg $^{-1}$ FM) and ascorbic acid content (grams of ascorbic acid.kg $^{-1}$ FM) of extracts of fruits of different cultivars of cornelian cherry (*Cornus mas* L.), n = 25.

Cultivar name	Total phenolic content	Ascorbic acid
Devin	2.61 ± 0.21 ^a	1.75 ± 0.22 ^a
Elegantnyj	3.41 ± 0.34 ^b	2.13 ± 0.31 ^a
Fruchtal	4.45 ± 0.32 ^c	1.48 ± 0.29 ^a
Jalt	4.00 ± 0.41 bc	2.52 ± 0.36 ^{ab}
Joliko	4.80 ± 0.30^{d}	3.01 ± 0.40^{b}
Kijevskij	5.37 ± 0.21 ^d	2.15 ± 0.28 ^a
Lukjanovskij	3.95 ± 0.30 ^{bc}	1.75 ± 0.27 ^a
Olomoucky	5.55 ± 0.32 ^d	3.11 ± 0.31 ^b
Ruzynsky	5.34 ± 0.34 ^d	3.05 ± 0.25 ^b
Sokolnicky	4.08 ± 0.42 ^{bc}	2.70 ± 0.38^{ab}
Titus	7.96 ± 0.42^{e}	2.91 ± 0.37 ^{ab}
Vydubeckij	8.11 ± 0.40 ^e	2.77 ± 0.33 ^{ab}

Different superscripts in each column indicate the significant differences in the mean at P < 0.05.

cultivars were observed (see Table 2).

The results of measurements performed in 12 cultivars showed a variability of the total phenolic content which ranged from 2.61 to 8.11 g GAE kg⁻¹ FM. It is also of interest that the variability of total antioxidant activity was likewise high (3.30 to 9.54 g AAE kg⁻¹ FM). Regarding the fact that all cultivars were grown under identical conditions and in the same locality, it is possible to conclude that one can clearly see the varietal variability, which is quite typical of fruits (Kim et al., 2003b). High antioxidant activity of fruit species is influenced by a number of chemical compounds. Amongst all of such active compounds, flavonols is one of the most important (Chun

et al., 2003). Our results showed high values of total phenolic content and antioxidant activity in comparison with other results conducted on cornelian cherries (Yilmaz et al., 2009b; Gulcin et al., 2005). A high content of ascorbic acid and antioxidant activity (e.g. in comparison with strawberries, orange fruits, kiwi fruits, etc) was demonstrated in other work, too (Tural and Koca, 2008). In our work on cornelian cherries high contents of ascorbic acid were observed with the values ranging from 1.48 (the cultivar 'Fruchtal') to 3.11 g kg⁻¹ (the cultivar 'Olomoucky').

Vitamin C can also be held for a substance which shows antioxidant activity. Nevertheless, in this case the

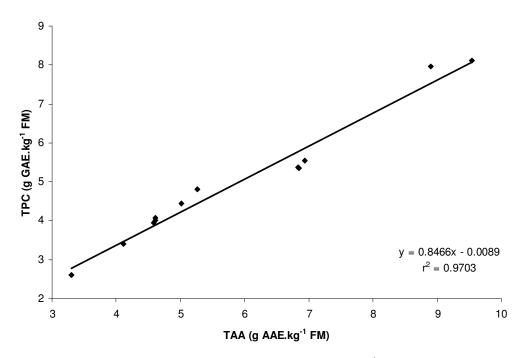


Figure 1. Relationship between total phenolic content (g GAE.kg⁻¹ FM) and DPPH radical scavenging activity (g AAE kg⁻¹ FM) in 12 cultivars of cornelian cherry (*Cornus mas* L.).

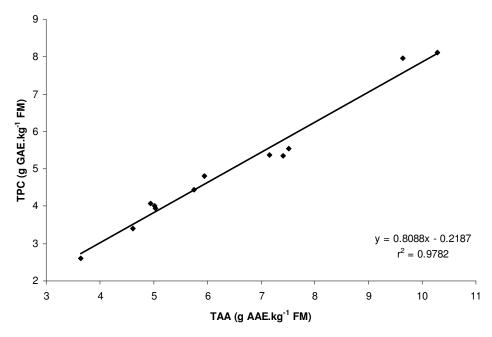


Figure 2. Relationship between total phenolic content (g GAE.kg⁻¹ FM) and t ABTS radical scavenging activity (g AAE kg⁻¹ FM) in 12 cultivars of cornelian cherry (*Cornus mas* L.).

effect on total values of antioxidant activity is problematic (Gil et al., 2002). This can also be associated with a considerably variable content of vitamin C due to the effect of the year, the degree of ripeness, manipulation,

processing, etc (Piga et al., 2003). In addition, it is possible that the levels of other components participating in the formation of antioxidant activity may be rather variable and for that reason the momentary content of

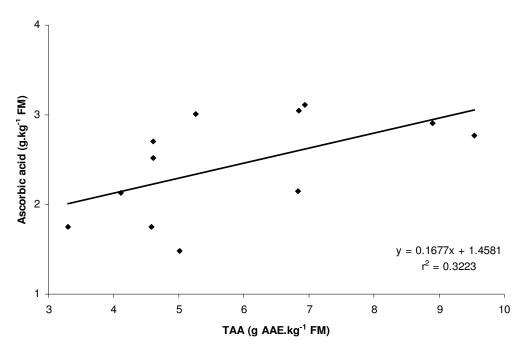


Figure 3. Relationship between total ascorbic acid content (g.kg⁻¹ FM) and DPPH radical scavenging activity (g AAE kg⁻¹ FM) in 12 cultivars of Cornelian Cherry (*Cornus mas* L.).

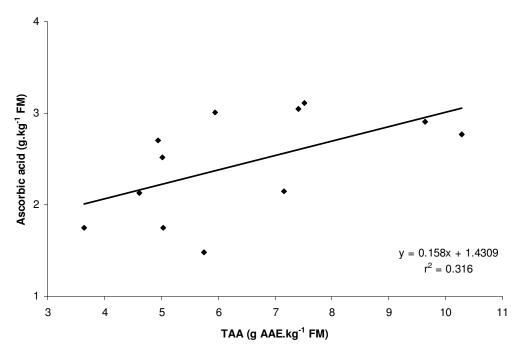


Figure 4. Relationship between total ascorbic acid content (g.kg⁻¹ FM) and ABTS radical scavenging activity (g AAE kg⁻¹ FM) in 12 cultivars of Cornelian Cherry (*Cornus mas* L.).

phenolic substances is considered to be the most important coefficient of correlation with antioxidant activity in fruits (Gil et al., 2002; Chun and Kim, 2004; Vizzotto et al., 2006) and vegetables (Valsikova and Belko, 2004).

Regarding our work, the same is shown in Figures 3 and 4 where we determined the correlation coefficient between the total ascorbic acid content and free radical scavenging activity of DPPH and ABTS to be $r^2 = 0.322$ and

0.316, respectively.

Nevertheless, as it follows from our measurement, within the scope of one fruit species there exist big differences in the content of chemical compounds in the fruit of particular cultivars, which is quite typical of fruit after all (Rop et al., 2009). Central European cultivars of cornelian cherry are generally considered to be a valuable source of substances with high antioxidant activity (Pantelidis et al., 2007). In comparison with other cultivars grown, e.g. in Turkey (Tural and Koca, 2008), they can be a suitable food supplement in relation to a positive impact on human health.

Conclusion

The main contribution of our work was to popularize and draw attention to Central European cultivars of cornelian cherries so that they could become a part of worldwide production and enrich the range of fruit commodities on a global scale. Cornelian cherry cultivars have high biological efficiency-antioxidant activity, total phenolic content and the content of ascorbic acid, which was confirmed in our measurement. This means that in future some cornelian cherry cultivars could be used as a source of new health sources when improving nutritional properties of the world's less traditional fruit species.

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