Biochemical composition and antioxidant activity of some sweet cherry (Prunus avium L.) cultivars grown in the Middle Atlas of Morocco

Abstract

The main biochemical composition, contents of total polyphenols, total flavonoids, total anthocyanins and antioxidant activity, were measured in the fruits of four sweet cherry cultivars (‘Burlat’, ‘Van’, ‘Napoleon’ and ‘Cerisette’) grown in two locations (Laanoceur and Toufsett) in the Middle Atlas. The free radical scavenging activity was evaluated spectrophotometrically using 1,1-diphenyl-2-picrylhydrazine (DPPH). Total phenolic contents ranged between 305.99 and 306.67 mg EqGal/100g DW, total flavonoid contents were within the range of 481.73-517.67 eqRE mg/100g DW, and total anthocyanin contents were between 1.09 and 2.89 mg cyanidin 3-glucoside/100g DW. Antioxidant activity ranged from 17.18 to 18.11 mg eqtrolox/100g DW for DPPH method. The highest values of total anthocyanin content and antioxidant activity (DPPH) were recorded in ‘Burlat’. The highest value of total flavonoid content was found in ‘Cerisette’. Cherries from Laanoceur and Toufsett locations are characterized by similar biochemical composition and antioxidant activity, except for total anthocyanin content that shows slightly elevated values in Laanoceur. The close correlation between total phenolic contents and antioxidant activities (r=0.73) show that antioxidant activity of cherry fruit depends on total polyphenols.

Keywords: sweet cherry, biochemical component, antioxidant activity, variability, location

Abbreviations: TA, total anthocyanins; TP, total phenolic; TF, total flavonoids; DPPH, 1,1-diphenyl-2-picrylhydrazyl; MCRDV, mécanisme compétitif de recherche développement et vulgarisation au Maroc

Introduction

Sweet cherry (Prunus avium L.), a fleshy non-climacteric stone fruit belongs to the genus Prunus and is mainly grown in countries with temperate climate. The species is reported to have originated from an area that includes Asia Minor, Iran, Iraq and Syria.1 In Morocco, the sweet cherry culture occupies an area of 2000 hectares, with an annual production of 14.100 tones.2 The most popular sweet cherry varieties cultivated in Morocco are ‘Bigarreau Van’ and ‘Bigarreau Burlat’,3,4 and the cultivars ‘Napoleon’ used as pollinator.3,5 However, there are other varieties cultivated at small scale such as ‘Cerisette’ and ‘Coeur de pigeon’.4 We hypothesized that these varieties differ from each other in some physical and biochemical features. Studies for characterization of sweet cherry fruit may have crucial importance for the producers in designing the necessary harvesting and postharvest technology of sweet cherry production in the world.5

This species presents a great economic importance due to the nutritional, technological and commercial value of the fruits.5 The nutritional importance especially depends on the biochemical composition, which represents a major source of antioxidant compounds.6 It is widely accepted for quality characteristics of the fruits like skin color, texture, sugar content, sourness, and volatile composition.7 Anthocyanin contents and the ratio of total solids/total acidity (known maturity) are other factors in consumers’ acceptance.8 Anthocyanins are plant pigments that are responsible for the color of many fruits, including sour cherry,9 and pomegranate.10 A recent increase in the interest for nutraceuticals has led to select for higher phenolic contents in fruits.11 Cherries, in particular, have been found to offer a good source of antioxidants and contain compounds believed to aid in pain relief of arthritis, gout and headaches.12 Many studies have been conducted to evaluate their properties in terms of quality and bioactivity.13,14

The antioxidant capacity of cherries is due to the presence of phenolics such as anthocyanin and melanin.15 Sweet cherries are rich in these types of phenolic compounds.13 Because of these phenolics, cherries rank first followed by other 19 fruits when comparing their antioxidant capacity.16 Fresh cherries are rich in anthocyanins, they are responsible of skin color of cherries,15 which is considered the most important indicator of quality and maturity of fresh cherry.17 Phenolic antioxidants have many positive effects on the human health like anti-carcinogenic and anti-inflammatory effects which makes them important in nutrition.6 Polyphenolics have been also demonstrated to have antiviral, antiallergic, anticarcinogenic activities as well as beneficial effects on gut microbiome and epigenetic effects.18

As far as we know, this is the first report of the antioxidant capacity and biochemical composition of sweet cherries grown under Moroccan climatic conditions. Thus, the main objective of this work was the determination of the total polyphenols, flavonoids and anthocyanins contents of fruit, as well as the antioxidant activity of fruits of four sweet cherry cultivars and to estimate the correlation between total polyphenols and antioxidant activity.

Material and methods

Plant material

Fruits were harvested in commercial orchards situated in tow locations of the Middle Atlas regions [(Toufsett (Azrou) and Laanoceur (Sefrou)]. Toufsett valley is characterized by humid and
temperate climate with an annual average temperature of 10.8°C and an average of rainfall higher than 600mm. The ‘Laanocneur’ locality is characterized by a continental climate with cold winter and hot summer, with annual average of rainfall varied between 400 and 600mm and an annual average temperature of 10.6°C.2

Fruits of four sweet cherry cultivars (‘Burlat’, ‘Van’, ‘Napoléon’ and ‘Cerisette’) were randomly harvested at the optimum commercial maturity based on fruit maturity and color development during May and June 2014. Fruit samples (0.5 kg) were transferred to the pomology laboratory in National School of Agriculture of Meknes immediately after harvest for further fruit quality attribute measurements. To determine the total phenolic content, total anthocyanin content and total antioxidant activity, cherries were pitted manually, frozen in liquid nitrogen and then stored at −20°C until the time of analysis.

Preparation of extract

Ten fruits per sample were frozen at −20°C and lyophilized (CHRIST ALPHA 1–4 LD plus) under vacuum, and 2g of lyophilized fruit were introduced into a flask then 20 ml of methanol was added. After 30 minutes of stirring, the mixture was centrifuged (6000 turn/min, 15min) and kept in the dark until analysis.

Determination of total phenolic (TP)

The total phenolics were determined according to Slinkard and Singleton.23 For 0.25ml of the sample extract (1/10), 0.25ml of Folin-Ciocalteu reagent (2 N) and 2ml of distilled water were added and the mixture was stirred by vortex, then 0.25 ml of sodium carbonate (20% w/v) was added. The extracts were mixed, stirred and then allowed to stand in the dark for 30min before measuring the absorbance at 750nm using a spectrophotometer (Safas UV–Visible spectrophotometer). All samples were prepared in triplicate. The results were expressed as mg gallic acid equivalent in 100g dry weight (mg GAE/100g DW).

Determination of total anthocyanins (TA)

Total anthocyanins (TA) were estimated by the pH differential method according to the protocol described by Giusti and Wrolstad.20 The extract (0.4ml) was dissolved in sodium acetate buffer (0.4M, pH 4.5, 3.6ml) and potassium chloride buffer (25mM, pH 1.0, 3.6 ml) and then the absorbance was recorded at 510nm and 700nm and expressed as mg cyanidin–3–glucoside 100 /g DW.

Determination of total flavonoids (TF)

Total flavonoids were determined according to the method described by Lamaison and Carnat.21 For 1ml of diluted sample, 1ml of aluminium chloride methanolic solution (2%) was added and mixed with vortex. Rutin was used to make the calibration curve. 1ml of diluted sample was separately mixed with 1ml of 2% aluminium chloride methanolic solution. After incubation at room temperature for 15 min, the absorbance of the reaction mixture was recorded at 430 nm. Total flavonoids were expressed as mg of rutin equivalent/100g DW.

DPPH radical scavenging ability (DPPH)

The total antioxidant capacity was measured using the DPPH (1.1–diphenyl–2–picrylhydrazyl) method of Brand-Williams et al.,22 with some modifications. This method aims the evaluation of the effect of free radical scavenging antioxidants on DPPH. Briefly, 0.1ml of the extract was mixed with 3.9ml of DPPH (0.1mM). The mixture is incubated in the dark for 60min and then the absorbance was recorded at 515nm. Results are expressed as mg equivalent trolox/100g DW.

Statistical analysis

All statistical analyses were performed using the SAS2000 program (SAS Institute, Cary, NC, USA). Analysis of variance used the PROC GLM procedure to distinguish the genotype and location effect. The genotype factor was hierarchical to the factor location because the trees were not repeated between sites. To draw a general conclusion among the four cherry locations, the population was considered as a random effect.23 Means was separated by Duncan’s multiple range test (P<0.05). Pearson’s correlation coefficients were calculated using the PROC CORR procedure.

Results and discussion

The total phenolics ranged from 305.99 to 306.67mg GAE/100g DW. The highest total phenolics content was recorded in the fruits of ‘Napoléon’ cultivar (306.67mg/g) and the lowest value was recorded for ‘Van’ (305.99mg/g). The values of total phenolics content in this study were low compared to those reported in the literature for others varieties.11,24–26 In addition, the value of total phenolic content of sour cherries varied from 78 to 500 mg GAE /100 g FW.11,25

These differences may be due to the extraction method.28 Other factors may explain these results such as genetic factors, environmental conditions and degree of maturity.12,20,29 The statistical analysis showed no significant effects of the genotype and location on the total phenolics (Table 1). This might be explained by the low number of tested cultivars in this study. If many cultivars with diverse origin are compared, genotype becomes one of the most important factors in determining the antioxidant capacity of fruit.11 Genotype effect is highly significant on total phenolics and total flavonoids (P<0.01), confirming the results reported in the literature.21,31 The total phenolics content is reported to be relative to the environment.32 However, cultivar and location effects were not seen to be significant on the antioxidant activity in our study. The effect of location was significant on total anthocyanins (P<0.05) (Figure 1). The total anthocyanins are determined by environmental factors (light and temperature) and growing conditions (irrigation, planting density and fertilization).31

Figure 1 Total phenolics in fruits of the four sweet cherry cultivars. The statistically significant difference among means was assessed by the Duncan test at P< 0.05.

*Values followed by the same letter are not significantly different at the 5% level.

In the sweet cherry cultivars, antioxidant activities (DPPH) ranged from 17.18 to 18.11 mg trolox eq./100g DW. Differences among cultivars were non-significant statistically (Figure 2). The highest antioxidant activity was observed in 'Burlat' cultivar (18.11 mg eq trolox/100g DW). Our results are in agreement with those of Usenik et al. who analyzed the antioxidant activity of 13 cherry cultivars showing the highest content in 'Burlat' cultivar. Moreover, several authors reported that the antioxidant activity of blackish colored fruit was higher than that in other genotypes, which agrees well with our results ('Burlat' cultivar has blackish colored fruit). The antioxidant activity is strongly influenced by the cultivation system, climatic conditions, duration and the technique of preservation of fruits. Antioxidant capacity is also determined by the biochemical characteristics of each cultivar. The antioxidant capacity of sweet cherries is superior compared with apples or pears but has much lower values than species with small fruits such as the strawberry, raspberry or blueberry.

Total flavonoids varied from 481.73–517.67 mg eqRE/100g DW (Figure 3). These values are highest than those reported by Prvulović et al. (42 to 154 mg eqRE/100g DW). The results showed significant differences for the total flavonoids content among 'Cerisette' and the remaining cultivars (Figure 3). The total flavonoids content was reported to be closely correlated with genotypes. The 'Cerisette' has the highest value of total flavonoids (517.67 eqRE mg/100g DW), while the lowest value was recorded in 'Burlat' (481.73 eqRE mg/100g DW) (Figure 3). Flavonoids were found to be an important part of human diet and are considered as active agent sin many medical plants. Flavonoids have been known to reduce oxidative stress in biological systems due to their antioxidant capacities.

The differences in total anthocyanins among the four sweet cherry cultivars were statistically significant. Total anthocyanin contents ranged from 1.09 and 2.89 mg cyanidin 3–glucoside/100g DW (Figure 4). The total anthocyanin concentration was reported to be ranged between 350 and 690 mg C3G/100g DW in some sweet.
cherry cultivars, which are higher than our results. These differences could be due to differences in methods of extraction. The highest value of total anthocyanin content was recorded in ‘Burlat’ cultivar (blackish colored fruit) and the lowest content of total anthocyanin content was in ‘Napoleon’ (yellow colored fruit). These results are in agreement with those reported by some authors, who reported that anthocyanin content was highest in cultivars with dark red color and lowest in cultivars with pale yellow color. The fruit of ‘Burlat’ was reported to contain very high total anthocyanins content. However, some exceptional cultivars with high antioxidant capacity and relatively low anthocyanin content were also described. The anthocyanin content of fruit species is not stable and is influenced by environmental factors such as light and temperature. The growing conditions such as irrigation, plant density, fertilization, as well as the genotype characteristics also affect anthocyanin content of fruits.

The statistical significance of the differences was examined using a Duncan’s test at p<0.05. *Values followed by the same letter are significantly different at p<0.05.

The biochemical composition of four cultivars studied in Toufselft and Laanoceur is shown in Figure 5. The analysis of variance showed statistically significant differences among means were assessed by the Duncan test at p<0.05. *Values followed by the same letter are not significantly different at p<0.05 level.

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**Conflict of interest**

The author declares no conflict of interest.

**References**


