

## PHENOTYPIC AND BIOCHEMICAL PARAMETERS OF FOUR SWEET CHERRIES (*PRUNUS AVIUM L.*) CULTIVARS GROWN IN AGRO-ECOLOGICAL CONDITIONS OF MIDDLE ATLAS OF MOROCCO

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### ABSTRACT

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*The objective of this study is to investigate the phenotypic and biochemical parameters of selected sweet cherry (*Prunus avium*) from middle region of Morocco. 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 "Toufselt") in the Middle Atlas. The free radical scavenging activity was evaluated spectrophotometrically using 1,1-diphenyl-2-picrylhydrazine (DPPH) and ABTS assay. The fruit weight, and pulp percentage were studied and found to range between 237–329 g, 38.14–42.22%, 39.21–44.36 g, and 52.27–57.48%, respectively.*

*The total phenolic and total anthocyanin content ranged from 305.99 and 306.67 mg EqGal/100g D.W, total flavonoid contents were within the range of 481.73-517.67 mgeqRE/100g D.W, and total anthocyanin contents were between 1.09 and 2.89 mg Eqcyanidin 3-glucoside/100g D.W. Antioxidant activity ranged from 17.18 to 18.11 mg EqTrolox/100g D.W for DPPH assay and from 27.97 to 29.60 mg EqTrolox/100f D.W for ABTS 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 "Toufselt" 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 ( $r^2=0.73$ ) show that antioxidant activity of cherry fruit depends on total polyphenols.*

**Keywords:** pomological properties, total phenolic, anthocyanins, sweet cherry, DPPH, diphenylpicrylhydrazyl, free radical, antioxidant activity ABTS, flavonoids.

## 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 (Vavilov, 1951). In Morocco, the sweet cherry culture occupies an area of 2000 hectares, with an annual production of 14.100 tones (DRA, 2014). The most popular sweet cherry varieties cultivated in Morocco are 'Bigarreau Van' and 'Bigarreau Burlat' (Oukabli, 2004; Kodad et al., 2010) and the cultivars 'Napoleon' used as pollinizer. However, there are other varieties cultivated at small scale such as 'Cerisette' and 'Coeur de pigeon' (Oukabli, 2004). 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 (Pérez-Sánchez et al. 2010).

This species presents a great economic importance due to the nutritional, technological and commercial value of the fruits (Pérez-Sánchez et al., 2010). The nutritional importance especially depends on the biochemical composition, which represents a major source of antioxidant compounds (Usenik et al., 2008). It is widely accepted for quality characteristics of the fruits like skin color, texture, sugar content, sourness, and volatile composition (Diaz-Mula et al., 2009). Anthocyanin contents and the ratio of total solids/total acidity (known maturity) are other factors in consumers' acceptance (Martinez-Romero et al., 2006). Anthocyanins are plant pigments that are responsible for the color of many fruits, including sour cherry (Pedisic et al., 2009) and pomegranate (Varasteh et al. 2012). A recent increase in the interest for nutraceuticals has led to select for higher phenolic contents in fruits (Kim et al., 2005). 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 (Naderiboldaji et al. 2008). Many studies have been conducted to evaluate their properties in terms of quality and bioactivity. (Kim et al. 2005; Hegedús et al. 2013; Papp et al. 2015).

The antioxidant capacity of cherries is due to the presence of phenolics such as anthocyanin and melatonin (Seeram et al., 2001) Sweet cherries are rich in these types of phenolic compounds (Kim et al. 2005). Because of these phenolics, cherries rank first followed by other 19 fruits when comparing their antioxidant capacity (Vinson et al., 2001). Fresh cherries are rich in anthocyanins, they are responsible of skin color of cherries (Seeram et al., 2001) which is considered the most important indicator of

quality and maturity of fresh cherry (Esti et al., 2002). Phenolic antioxidants have many positive effects on the human health like anti-carcinogenic and anti-inflammatory effects which makes them important in nutrition (Usenik et al., 2008). Polyphenolics have been also demonstrated to have antiviral, anti-allergenic, anticarcinogenic activities as well as beneficial effects on gut microbiome and epigenetic effects (Martin et al. 2013).

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 variability of fruit weight, percentage of pulp, titratable acidity, maturity index, total phenolics, 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 phenolics and antioxidant activity.

## MATERIAL AND METHODS

### Plant material

Fruits were harvested in commercial orchards situated in two locations of the Middle Atlas regions [“Toufselt” (Azrou) and “Laanoceur” (Sefrou)]. “Toufselt” 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 “Laanoceur” 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.

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 (0.5kg) 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.

### Chemicals and reagents

1,1-Diphenyl-2-picrylhydrazyl (DPPH) and ABTS was from the Fluka company (Switzerland). Trolox (6-hydroxy-2, 5, 7, 8-tetra methylchroman-2-carboxylic acid), which is a hydrophilic analogue of vitamin E, Gallic were purchased from Sigma-Aldrich (St. Louis, MO, USA).

### **Phenotypic characteristics**

For each cultivar, twenty fruits were analyzed for physical characteristics. Fruit weight was measured on an electronic balance with a sensitivity weighing of  $\pm 0.01$  g and randomly harvested 20 fruits from different parts of trees with four replications were used for measurement. The percentage of pulp were calculated using the formula  $(\text{Fruit weight} - \text{stone weight}) / \text{Fruit weight} * 100$ .

### **Titration acidity, total soluble solids and maturity index of sweet cherry fruit**

The titration acidity (TA) was determined by titration to pH 8.1 using a digital pH meter (Thermo Orion 3 star) at 21°C with 0.1 M NaOH solution and expressed as g of malic acid per liter of juice (IFU, 1996). The instrument was calibrated to 0°Brix using distilled water (Giusti and Wrolstad, 2001).

The total soluble solids (TSS) were determined with a digital refractometer (Mettler-Toledo GmbH, 30 PX, Switzerland, calibrated using distilled water). Results were reported as Brix at 21°C.

Maturity index was calculated as the ratio between Total soluble solids and titration acidity. The pulp of fruit, was obtained from extraction of juice from the five fruits of each cultivar taken at random and expressed as volume of juice per 100 g of fruits. The maturity index (MI) was calculated by dividing the total soluble solid with titration acidity. The percentage of pulp were calculated using the formula  $(\text{Fruit weight} - \text{stone weight}) / \text{Fruit weight} * 100$ .

### **Preparation of sample and Biochemical parameters**

Ten fruits per sample were frozen at  $-20^{\circ}\text{C}$  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.

### **Total phenolic content (TP)**

The total phenolics were determined according to Slinkard and Singleton (1977). 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 D.W).

### Total anthocyanins content (TA)

The TAC in sweet cherry extract was determined using the pH differential methods, with slight modifications Giusti and Wrolstad (2001). Briefly, 0.4 mL sweet cherry extract samples were taken, and one of them was adjusted to 10 mL with potassium chloride (25mM) buffer, pH 1.0, and the other with sodium acetate (0.4M) buffer, pH 4.5. After equilibrium at 15 min, the absorbance was measured at 510 nm and 700 nm, respectively, using an UV/visible spectrophotometer (spectrophysics Jasco UV 1700, Japan). The TAC was calculated as milligrams of cyanidin-3-glucoside (C3G) equivalents (molar extinction coefficient 26,900).

The Absorbance (A) was expressed as follows:

$A = [(A_{520\text{ nm}} - A_{700\text{ nm}})]_{\text{pH}1.0} - [(A_{520\text{ nm}} - A_{700\text{ nm}})]_{\text{pH}4.5}$  and TAC of fruit was presented as mg cyanidin-3-glucoside/100g (DW) of sweet cherry fruit and was determined using equation below:

$$\text{TAC} = (A * \text{MW} * F * 100) * 1 / \text{MA}$$

With: A: absorbance; MW: molecular weight (449.2 g mol<sup>-1</sup>); F: dilution factor (10); MA: molar absorptivity of cyanidin-3-glucoside (26,900).

### Total flavonoids content (TF)

Total flavonoids were determined according to the method described by Lamaison and Carnat (1990). 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 D.W (mg ER/100g D.W).

### Antioxidant activity

#### Radical scavenging activity assay (DPPH)

Antioxidant activity was also determined using the DPPH (1,1-diphenyl-2-picrylhydrazyl) method reported by Brand-Williams et al. (1995) 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 D.W.

### Antioxidant activity by the ABTS method

Antioxidant activity was measured using an improved ABTS method as described by Re et al. (1999) method according to TEAC (Trolox Equivalent Antioxidant Capacity) The ABTS radical cation solution was prepared through the reaction of 7mM of ABTS•+ (2,2'-azinobis (3-ethylbenzothiazoline-6-sulphonic acid)) and 2.45 mM potassium persulphate, after incubation at 23°C in the dark for 12–16 h. The ABTS solution was then diluted with 100 mM phosphate buffer (pH 7.6) to obtain an absorbance of  $0.700 \pm 0.005$  at 734 nm. ABTS solution (3.9 mL; absorbance of  $0.700 \pm 0.005$ ) was added to 0.1 mL of the extract sample and mixed vigorously. The reaction mixture was allowed to stand at 23°C for 10 min and the absorbance at 734 nm was immediately recorded. A standard curve was obtained using Trolox standard solution at various concentrations (ranging from 0 to 30 µM) in 80% methanol. The absorbance of the reaction samples was compared to that of the Trolox standard and the results were expressed in terms of Trolox equivalents (TE)/100g D.W of sweet cherry.

### Statistical analysis

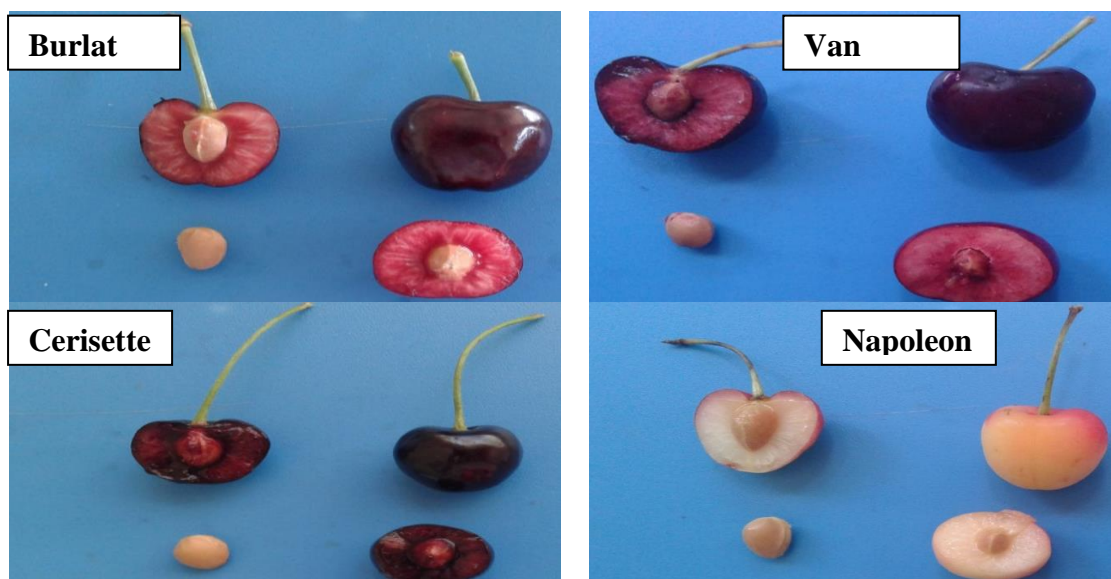
All statistical analyses were performed using the SAS2000 program (SAS Institute, Cary, NC, USA). Analysis of variance used the PROC GLM procedure to distinguish the genotype and location effect. The genotype factor was hierarchical to the factor location because the trees were not repeated between sites. To draw a general conclusion among the four cherry locations, the population was considered as a random effect (Steel and Torrie, 1960) Means were separated by Duncan's multiple range test ( $P < 0.05$ ). Pearson's correlation coefficients were calculated using the PROC CORR procedure.

## RESULTS AND DISCUSSION

### Physical and physicochemical parameters determination

The physical aspect of four sweet cherry fruits investigated is presented in figure 1 and Table 1. The result shows that some physicochemical properties of the four sweet cherry cultivars. Fruit weight of "Van" cultivar was significantly greater than that of the "Burat", "Napoléon" and "Cerisette" fruits. On the other hand, "Burlat" cultivar registered similar fruit weight to that of "Napoleon" cultivar. The weight of four sweet cherry cultivars "Burlat", "Van", "Napoleon", and "Cerisette" was 4.91, 5.79, 4.77 and 4.31g respectively. Fruit weight of eight commercial cherry cultivars and four new selections (*Prunus avium* L.) from the breeding program at Agriculture and Agri-Food Canada was between 8.8 and 14.5 g (Girard and Kopp, 1998). Our results were lower than those of commercial cherry cultivars. The percentage of pulp for "Van" fruits (96.26%) was significantly greater than that of "Burlat" (96.09%), "Napoleon" (4.77%) and "Cerisette" (4.31%) fruits (Table 1). It is important to indicate that consumers generally prefer sweet cherries with large pulp amounts (Pérez-Sánchez et al. 2010).

The titratable acidity for “Napoleon” cultivar (8.6 eqmalic acid g/l of juice) was significantly greater than that of the “Burlat” (7.38 geq malic acid /l, “Van” (6.81 geq malic acid /l) and “Cerisette” (6.71g/l malic acid) fruits. The titratable acidity values for “Burlat”, “Van” and “Cerisette” cultivars were similar. “Cerisette” cultivar had significantly higher maturity index (2.26) compared to that of “Burlat” (1.86), “Van” (1.88) and “Napoleon” (1.44). Such differences in maturity index may be related to differences in maturity during fruit collection (Vavoura et al. 2015).



**Figure 1. Morphological characteristic of Sweet cherry cultivars.**

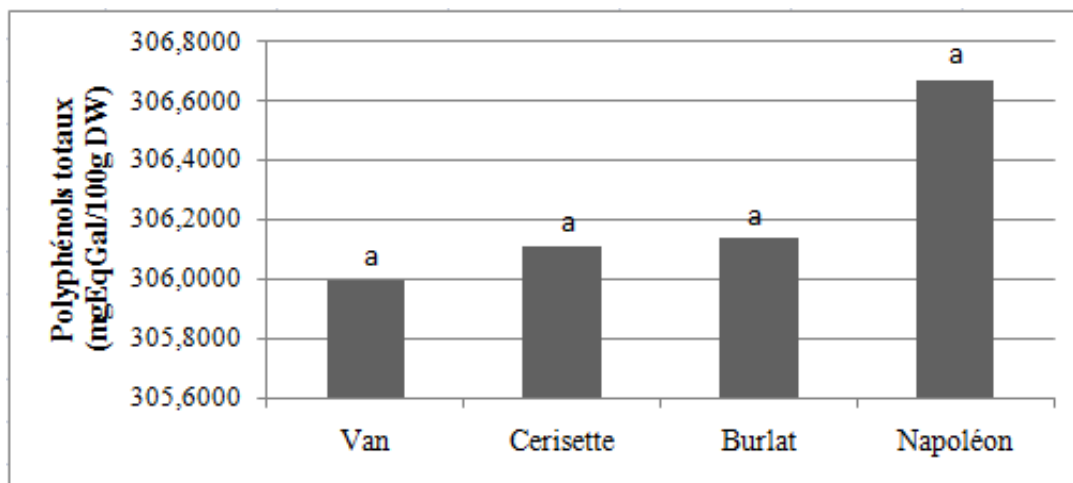
**Table 1. Some physicochemical properties of the four sweet cherry cultivars.**

Cultivar	Fruit weight(g)	Percentage of pulp %	Acidity( e g Eqmalicacid per liter of juice of fruit	Maturity index
Burlat	4.91b	96.09ab	7.38b	1.86b
Van	5.79a	96.26a	6.81b	1.88b
Napoleon	4.77b	95.95b	8.60a	1.74b
Cerisette	4.31c	95.08c	6.71b	2.26a

**Biochemical parameters**

The total phenolics ranged from 305.99 to 306.67mg GAE/100g DW (Figure 2). The highest total phenolics content was recorded in the fruits of ‘Napoléon’ cultivar (306.67mgGAE/100gDW) and the lowest value was recorded for ‘Van’ (305.99mg GAE/100gDW). The values of total phenolics content in this study were lower compared to those reported in the literature for others varieties. (Kim et al. 2005;Serra et al. 2011; Petković et al. 2014).In addition, the value of total phenolic content of sweet cherries varied from 78 to 500 mg GAE /100 g DW.(Kim et al. 2005; Dragović-Uzelac

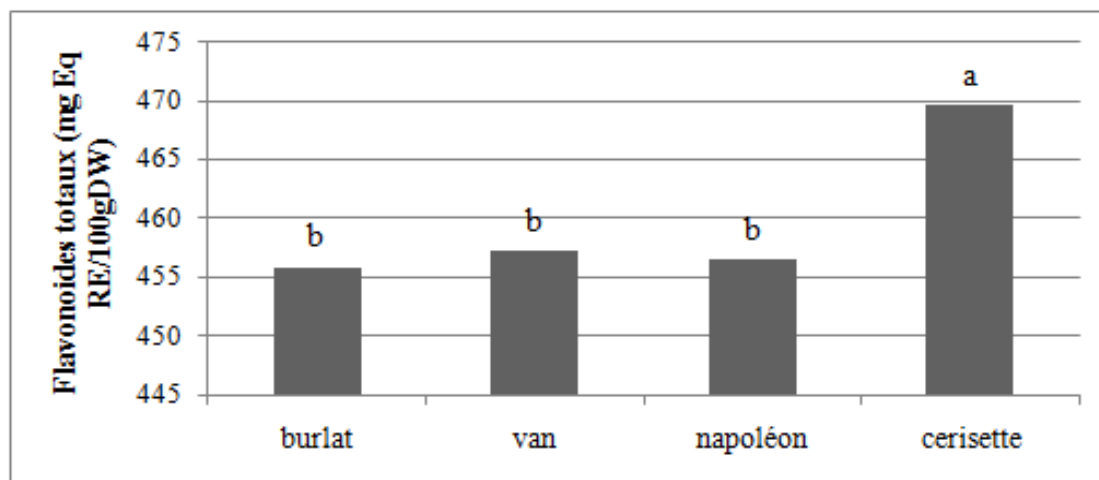
et al. 2010). These differences may be due to the extraction method (Melicháčová et al. 2010). Other factors may explain these results such as genetic factors, environmental conditions and degree of maturity (Hegedúset al. 2013; Bravo, 1988; Goncalves et al. 2004).



**Figure 2. Total phenolics (TPC) 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).**

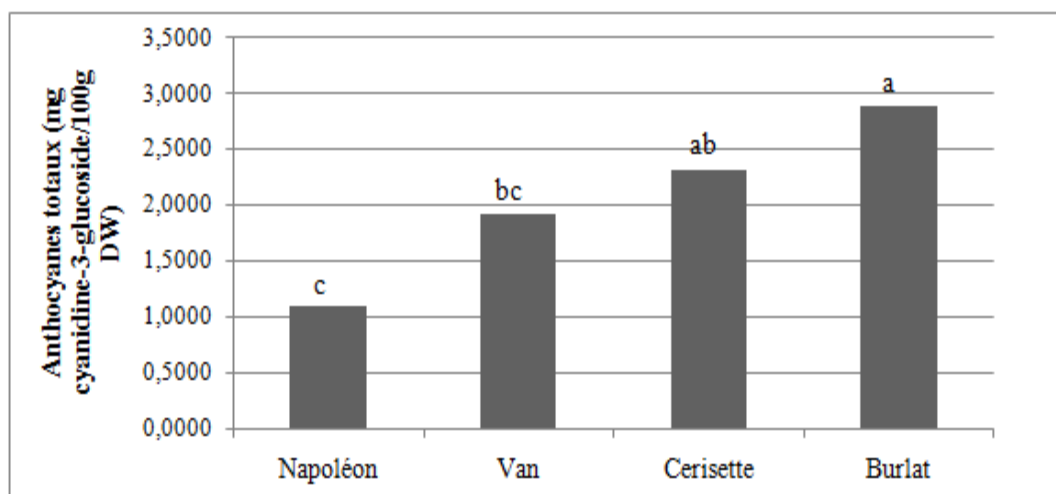
Total flavonoids varied from 481.73–517.67 mg RE/100g D.W (Figure 3). These values are highest than those reported by Prvulović et al. (2011) (42 to 154mg RE/100g D.W). The results showed significant differences for the total flavonoids content among ‘Cerisette’ and the remaining cultivars (Figure 3). The total flavonoids content was reported to be closely correlated with genotypes (Petković et al. 2014). The ‘Cerisette’ has the highest value of total flavonoids (517.67 RE mg/100g D.W), while the lowest value was recorded in ‘Burlat’ (481.73 RE mg/ 100g DW) (Figure 3). Flavonoids were found to be an important part of human diet and are considered as active agent in many medical plants (Martin et al. 2013). Flavonoids have been known to reduce oxidative stress in biological systems due to their antioxidant capacities (Kim et al. 2005).





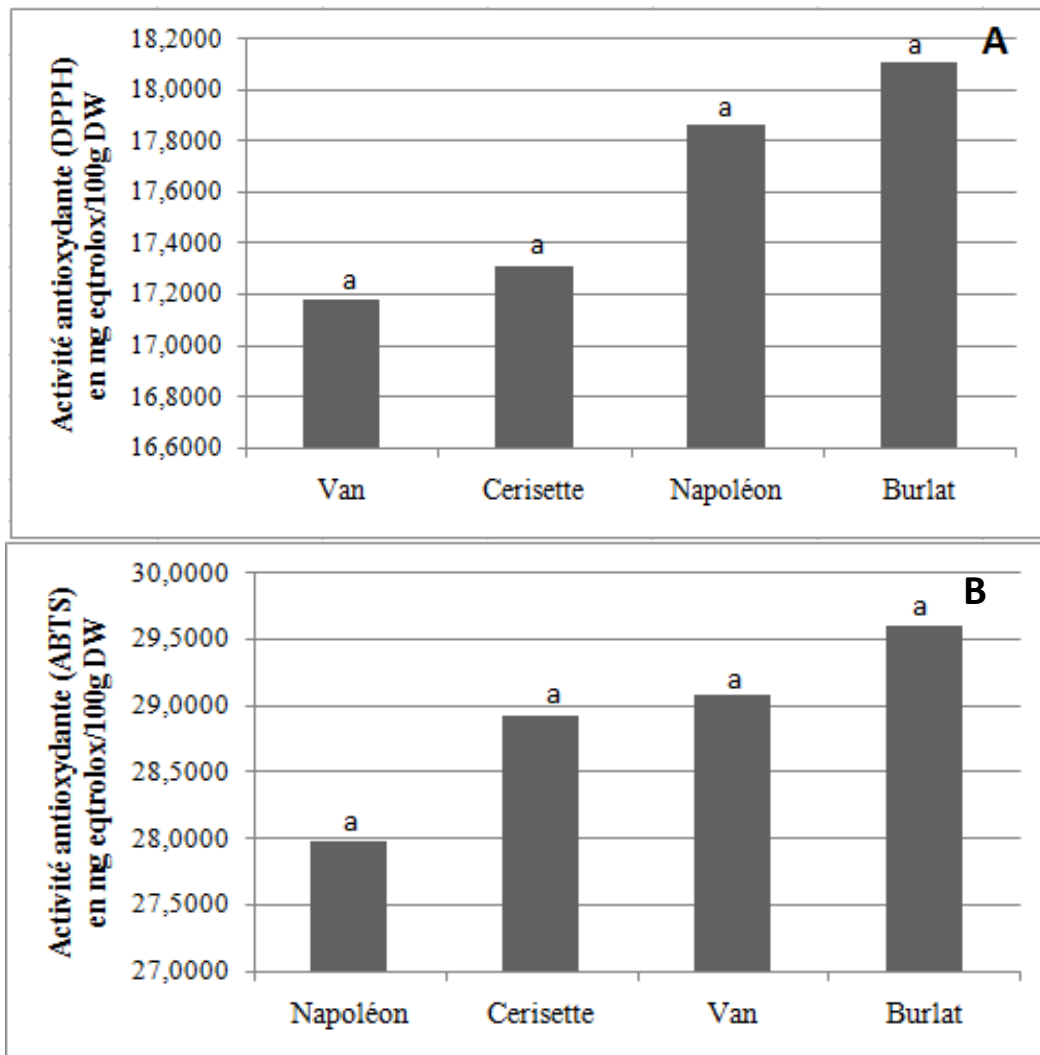
**Figure 3. Total flavonoids of four sweet cherry cultivars. The statistically significant differences among means were tested by the Duncan test at  $p < 0.05$ . (\*Values followed by the same letter are not significantly different at the 5% level)**

The differences in total anthocyanins among the four sweet cherry cultivars were statistically significant (Figure 4). Total anthocyanin contents ranged from 1.09 and 2.89mg cyanidin 3–glucoside/100g D.W (Figure 4). The total anthocyanin concentration was reported to be ranged between 350 and 690mg cyaniding 3–glucoside/100g D.W in some sweet cherry cultivars (Prvulović et al. 2011) which are higher than our results. These differences could be due to differences in methods of extraction (Vangdaland Slimestad, 2006). 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 ‘Napoléon’ (yellow colored fruit). These results are in agreement with those reported by some authors (Vangdaland Slimestad, 2006; Karlidaget al. 2009) 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 (Useniket al. 2008; Prvulovićet al. 2011). However, some exceptional cultivars with high antioxidant capacity and relatively low anthocyanin content were also described (Hegedüset al. 2013). 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 (Karlidag et al. 2009).



**Figure 4. Total anthocyanins 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)**

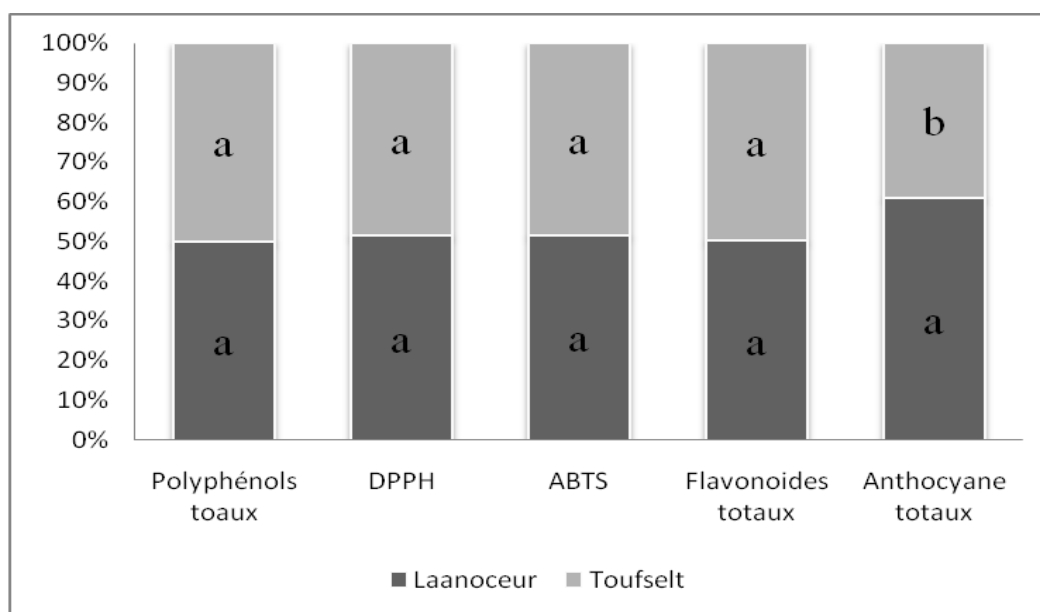
As the antioxidant capacity was measured by two methods namely, ABTS and DPPH assays. The ABTS assay is based on the generation of a blue / green ABTS, which is applicable to both hydrophilic and lipophilic antioxidant systems; whereas DPPH assay uses a radical dissolved in organic media and is, therefore, applicable to hydrophobic systems (Kim et al. 2002) Figure 5, resent the result of the antioxidant activities of sweet cherry cultivars. As can be seen the antioxidant activities (DPPH) (A) produce a less values of AA (DPPH) (17.18 to 18.11mg TRE./100g D.W) than ABTS assay, wich seemed to be a better method for expressing the antioxidant capacity of phenolic compounds in fruits sweet cherry. The values is ranged from 27.97 to 29.60 mg TRE/ D.W ABTS method (B) (Figure 5). Differences among cultivars were non-significant statistically (Figure 5). The highest antioxidant activity was observed in 'Burlat' cultivar (18.11mg TRE/100g DW). The results are in agreement with those reported by Useniket al. (2008) 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). (Karlidag et al. 2009; Vangdal and Slimestad, 2006; Battino et al. 2004). As reported by the litterature, the genetic factor is the first parameter with the potential to influence the antioxidant content in a commodity. Significant inter-cultivar variation in the phenolic content and antioxidant capacity has also been documented in cherries (Prvulović et al. 2012). The antioxidant activity is strongly influenced by the cultivation system, climatic conditions, duration and the technique of preservation of fruits (Sîrbu et al. 2012). 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. (Battino et al. 2004; Serrano et al. 2005; Khanizadeh et al. 2009).



**Figure 5. Antioxidant activity of the four sweet cherry cultivars by DPPH method (A) and ABTS method (B). 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)**

The biochemical composition of four cultivars studied in “Toufselt” and “Laanoceur” is shown in figure 6. The analysis of variance showed no significant effect of the location on biochemical composition and antioxidant activity, except for the total anthocyanins content (Figure 6). The fruits collected from “Laancaeur” location contain more anthocyanins than those collected from Toufselt location. This difference is probably due to growing techniques. Irrigation by drip system at “Laanoceur” has probably positively affected the content of total anthocyanins. Generally, sweet cherries at “Toufselt” and “Laanoceur” regions have similar biochemical composition and antioxidant activity.

A positive correlation was found between total polyphenols and antioxidant activity (DPPH) ( $r^2 = 0.73$ ). Many studies have reported similar correlation between these variables (Chaovanalikit and Wrolstad, 2004; Vangdaland Slimestad, 2006; Karlidaget al. 2009; Damarand Ekşi, 2012). This indicates that the antioxidant activity closely depends on the total polyphenol content of cherry (Melicháčová et al. 2010; Prvulović et al. 2011; Giménez et al. 2014). \*Values followed by the same letter are not significantly different at  $p < 0.05$  level.



**Figure 6. The biochemical composition and antioxidant activity of the four cultivars studied in “Toufselt” and “Laanoceur” locations. 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  $p < 0.05$  level)**

The statistical analysis showed no significant effects of the genotype and location on the total phenolics (Table 2). 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 (Hegedüs et al. 2013). Genotype effect is highly significant on total flavonoids ( $P < 0.01$ ), confirming the results reported in the literature. (Karlidag et al. 2009; Xiao et al. 2015). The total phenolics content is reported to be relative to the environment (Popescu et al. 2014). 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 ( $P < 0.05$ ). Geographical location probably had a strong impact on the anthocyanins content of different sweet cherry cultivars studied (Table 2). Also, the interaction of location with genotype showed significant differences in the total anthocyanin content. The total anthocyanins are determined by environmental factors

(light and temperature) and growing conditions (irrigation, planting density and fertilization) (Karlidag et al. 2009).

**Table 2. Genotype and agroecological effects on the biochemical characteristics of the fruit.**

Source of variation	Ddl	Mean square	F-value	Pr>F
<b>Total phenolics</b>				
Location	1	1.99	3.65	0.0616
Genotype (location)	4	0.94	1.73	0.1576
Error	52	0.54		
<b>Antioxidant activity</b>				
Location	1	2.43	1.07	0.3047
Genotype (location)	4	6.2	2.74	0.0384
Error	52	2.26		
Source of variation	DBL	Mean square	F-value	Pr>F
<b>Total Flavonoids</b>				
Location	1	322.14	0.7	0.4052
Genotype (location)	4	2007.49	4.39	0.0039
Error	52	457.43		
Source of variation	DBL	Mean square	F-value	Pr>F
<b>Total anthocyanins</b>				
Location	1	5.04	4.56	0.0374
Genotype (location)	4	1.98	1.8	0.1436
Error	52	1.1		

## CONCLUSION

The relationship between antioxidant capacity and phenolic contents varied between Cultivars and strongly depends on total polyphenolic contents in fruits of four sweet cherries and could be considered a good source of natural antioxidants. Sweet cherries at “Toufselt” and “Laanoceur” locations have similar biochemical composition and antioxidant activity, except for the total anthocyanins content that shows slightly elevated values at “Laanoceur”. Geographic location in combination with higher precipitations influenced the fortified biosynthesis of anthocyanins in “Laanoceur” more than in “Toufselt” location. Therefore the agro-ecological conditions of the “Toufselt” and “Laanoceur” locations of the Middle Atlas have no effect on total polyphenol, flavonoids and antioxidant activity of sweet cherries produced in the Middle Atlas. If such an alteration is confirmed through several years, the identification of the factors behind elevated anthocyanins levels at “Laanoceur” may help to find growing techniques to increase anthocyanins content in cherry. The relationship between

antioxidant capacity and phenolic contents varied between cultivars and strongly depends on total polyphenolic contents in fruits of sweet cherries and could be considered a good source of natural antioxidants.

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