

# Fruit quality attributes of blackberry grown under limited environmental conditions

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## ABSTRACT

Fruit quality attributes were studied for two consecutive years in seven blackberry cultivars grown in a Serbian climate and on acidic soil. Physical parameters [berry weight (BW), size and shape] and chemical parameters [soluble solids content (SSC), acidity, total phenolic (TPH) and flavonoid content (TFC) and total antioxidant capacity (TAC)] were evaluated. A high variability was found in the set of the evaluated blackberry cultivars and significant differences were found among them in all studied quality attributes. Year-by-year variations were observed for all quality traits, except berry length (L) and berry shape index (BSI). A high correlation was found among TPH and TAC. In addition, most of cultivars had good adaptation capability and respectable fruit quality attributes, and also had good potential as a commercial crop for fresh and processing markets and future breeding programs.

**Keywords:** acidic soil; antioxidant capacity; berry size; *Rubus fruticosus* L.; soluble solids content

Blackberries are popular fruits widely distributed throughout nature, except the polar regions, and grown worldwide. World consumption of blackberries increased in the past few decades, and they are consumed fresh, frozen, or commercially processed into a variety of foods and other products such as jam, wine, tea, ink, dyes, fruit leather, ice cream, cake pastry, and medicine (Eyduran et al. 2008). Berries of this species had pleasant flavor and high nutritive and health value. Blackberries are rich sources of carbohydrates, dietary fibers, vitamins, minerals and other bioactive compounds (Wang 2007). Additionally, berry fruits are rich in phenolic compounds such as phenolic acids and anthocyanins (Pantelidis et al. 2007), flavonoids (Siriwoharn and Wrolstad 2004, Cho et al. 2005), flavonols, ellagitannins, gallotannins and proanthocyanidins (Reyes-Carmona et al. 2005), which demonstrated considerable antioxidant properties. Flavonoids and phenolic compounds in the berries are anti-carcinogens and have anti-neurodegenerative and anti-inflammatory effect,

therefore, blackberry berries are used in medicine (Wang 2007). Also, the phenolic contents of berries are therefore an important parameter for the evaluation of their antioxidant properties and quality. On the other hand, in view of the increasing consumer attention to the nutritional value of their diets, the antioxidant content of blackberries and in particular their phenolic content ought to be considered as an important trait for breeding programmes (Clark and Finn 2011).

The exact composition of nutraceuticals in blackberries is highly dependent on the cultivars, cultural practices and numerous pre-harvest factors, especially climatic and soil conditions (Prange and De Ell 1997). Additionally, blackberry is a crop of mild climate and can easily adapt to different ecological conditions (Eyduran et al. 2008).

Serbia accounted for 69% of the blackberry area in Europe and produced 27 557 t, the fourth highest production in the world after USA, Mexico and China (Strik et al. 2007). In this country, only

semi-erect blackberry types were grown with the predominant cultivar being Thornfree, somewhat Dirksen Thornless and Black Satin. In past few years, Čačanska Bestrna, a new Serbian cultivar that produces high yield and large fruits is being widely planted. However, the physical and chemical properties of the berry of these and other cultivars are slightly tested, while the total phenolic and total flavonoid content and antioxidant capacity were not investigated. From these purposes, the aim of this work was to evaluate and quantify main physical and chemical attributes of seven blackberries grown under limited climatic and soil conditions in Cacak, Western Serbia.

## MATERIAL AND METHODS

### Plant material and experimental procedure.

Blackberry (*Rubus fruticosus* L.) fruits of seven cultivars [Dirksen Thornless (DT), Thornfree (TF), Čačanska Bestrna (ČB), Black Satin (BS), Loch Ness (LN), Chester Thornless (CT), Navaho (NV)] were evaluated in 2010 and 2011. The experimental orchard was established in 2000 and was located near Cacak, Western Serbia. The blackberries were planted in rows spaced 3.0 m apart with plants set at 1.4 m apart in the row, and trained as a four-wire trellis, in the randomized block design with four replicates for each cultivar. Standard cultural practices were applied, except irrigation.

Weather conditions of Cacak are characterized by the mean growing season temperature and total rainfall of 17.0°C and 408.6 mm for the long-term averages, respectively. However, substantial rainfall deficiency and high air temperatures were observed in 2011, especially during ripening (Table 1).

Soil texture in blackberry orchard is sandy-loam, moderate in organic matter (1.62%) and poor of N (0.14%); soil pH in KCl 0.01 mol/L was 4.86. The contents of available soil P, K, Ca and Mg were 178 mg/kg, 220 mg/kg, 0.39% and 6.2 mg/kg, respectively. For blackberries grown under Serbian conditions, optimal amount of rainfall during growing season is 450 mm, and soil pH between 6 and 7 (Milosevic 1997). However, rainfall deficiency, high temperatures and acidic soil may have affected the results of this study, as previously reported (Milošević and Milošević 2011).

Mature berries (30 per replicates for each cultivar) assessed by full colour development were harvested in both years. The berries were transported to the laboratory in the same day for sample preparation and analysis.

Table 1. Mean monthly and growing season temperature and rainfall in Cacak in 2010 and 2011

Month	Air temperature (°C)			Rainfall (mm)		
	2010	2011	normal*	2010	2011	normal*
April	13.3	12.2	11.5	52.0	23.5	33.3
May	17.9	15.5	16.8	98.8	83.2	59.3
June	21.3	20.7	20.0	81.0	64.8	86.1
July	23.5	22.3	21.5	90.0	36.0	75.5
August	23.7	23.4	21.2	78.5	0.0	50.0
September	17.3	21.3	16.7	25.0	32.4	42.7
October	10.0	16.3	11.4	63.0	57.2	61.7
Mean or total	16.5	18.8	17.0	488.3	297.1	408.6

\*normal refers to the long-term average (45-year average, i.e. 1965–2010 period)

**Fruit physical properties.** BW (g) was measured by a Tehnica ET-1111 technical scale (Iskra, Horjul, Slovenia) with a sensitivity of  $\pm 0.01$  g. For each berry, two linear dimensions, length (L) and width (W) in mm were measured using a digital caliper gauge Starrett 727 Series (Athol, MA, USA). On the basis of the measured data, BSI was calculated as L/W ratio.

**Determination of soluble solids content and titratable acidity.** SSC and titratable acidity (TA) were determined in juice extracted using a food processor in three replicates of 10 berries. The SSC (°Brix) was determined using a hand refractometer Milwaukee MR 200 (ATC, Rocky Mount, USA) at 20°C. For TA (% of malic acid), prepared juice was titrated with 0.1 mol/L NaOH, up to pH 8.1 using a titrimeter Metrohm 719S (Titrino, Herisau, Switzerland). On the basis of the measured data, SSC/TA ratio or ripening index (RI) was calculated.

**Determination of total phenolic and flavonoid content and total antioxidant capacity.** The TPH [mg GA/g dry extract (d.e.)], TFC (mg RU/g d.e.) and TAC (AA/g d.e.) were determined spectrophotometrically using UV-VIS spectrophotometer MA9523-SPEKOL 211 (Iskra, Horjul, Slovenia), according to the methods described by Gutfinger (1981), Prieto et al. (1999) and Brighente et al. (2007), respectively.

**Statistical analysis.** Data were subjected by analysis of variance, and treatments were compared using the *LSD* test at  $P \leq 0.05$  using SPSS (SPSS Inc., version 8.0, Chicago, USA). Correlations among TPH, TFC, and TAC for all berries was calculated according to the Pearson's test at  $P = 0.05$ .

## RESULTS AND DISCUSSION

**Evaluation of fruit size and fruit shape.** There were large variations between the tested blackberry cultivars in both seasons (Table 2). The ČB and LN had the largest BW for two years; TF and DT in 2010, and TF in 2011 had the smallest BW. Presented results were much higher than the findings of Eyduran et al. (2008) for BS, NV, LN, and CT, similar to the observations by Miletić et al. (2006) for ČB and BS, and lower than those of Vrhovsek et al. (2008) for BS, ČB, CT, and LN.

The BW has a direct effect on the marketability and acceptance of blackberries in both fresh market and processing. Traditionally, large berries are preferred by consumers, but excessive berry weight (possibly > 15.0 g) is usually not desired for processed or fresh market use (Clark and Finn 2011). According to the above authors, the ideal BW for fresh market use ranges from 8 to 10 g. In our study, only ČB and LN had values close to 8.0 g. It could be said that under irrigation this trait can be better.

The L and W were the highest in LN in 2010, whereas the highest L in 2011 was found in DT and W in ČB and BS, also in that year. Similar data were reported by Miletić et al. (2006). The highest berry shape index (BSI) values were observed in DT and the lowest in NV in both seasons. All cultivars had values beyond 1, and supposing an elongated shape of the berries (Gerçekcioglu and Esmek 2005). The BW and W significantly differed between years, being higher in 2011 than in 2010 (Table 2), which could be due to the impact of

environmental conditions (Eyduran et al. 2008). L and BSI were similar in both years.

**Evaluation of soluble solids content and titratable acidity.** The SSC is a better indicator of blackberry maturity and also very important in the food industry and critical in comparative studies where variations by cultivar and environment are high (Clark and Finn 2011). In this study, significant differences among cultivars for SSC were found (Table 3). The highest SSC was observed in NV for two years, and in DT in 2011. The lowest SSC was recorded in ČB in 2010 and in BS in 2011. Great variability for SSC among cultivars was previously reported (Pantelidis et al. 2007). Generally, SSC of at least 10% provides for a 'sweet' eating experience for fresh blackberries (Clark and Finn 2011). In our study, LN, CT, and especially NV, provide SSC close to 10°Brix, which is in agreement with observation of the above authors. Significant year-by-year differences for SSC were found, being higher in 2011 than in 2010 (Table 3), due to the higher mean monthly temperatures and rainfall absence during maturity (Naumann and Wittenburg 1980).

The TA is a very important quality attribute, influencing notably berry taste (Vrhovsek et al. 2008). Data presented in Table 3 indicated that TA significantly varied among cultivars in both years. The highest value was found in ČB, and the lowest in NV. Great variability considering TA was previously found (Gerçekcioglu and Esmek 2005). Year-by-year variations of TA were significant (Table 3). For example, except other factors, the

Table 2. Berry weight, berry size and berry shape index of blackberry cultivars

Treatment	Berry weight (g)		Length (mm)		Width (mm)		Berry shape index	
	2010	2011	2010	2011	2010	2011	2010	2011
Dirksen Thornless	4.54 ± 0.02 <sup>e</sup>	6.91 ± 0.23 <sup>c</sup>	27.28 ± 0.58 <sup>b</sup>	28.10 ± 0.63 <sup>a</sup>	19.31 ± 0.27 <sup>c</sup>	20.33 ± 0.32 <sup>b</sup>	1.41 ± 0.03 <sup>a</sup>	1.38 ± 0.04 <sup>a</sup>
Thornfree	4.65 ± 0.01 <sup>e</sup>	5.32 ± 0.26 <sup>f</sup>	21.52 ± 0.67 <sup>b</sup>	23.69 ± 0.69 <sup>d</sup>	17.52 ± 0.30 <sup>d</sup>	19.31 ± 0.21 <sup>c</sup>	1.23 ± 0.03 <sup>d</sup>	1.22 ± 0.03 <sup>e</sup>
Čačanska Bestrna	7.57 ± 0.00 <sup>a</sup>	7.61 ± 0.22 <sup>a</sup>	26.62 ± 0.53 <sup>c</sup>	27.54 ± 0.52 <sup>b</sup>	20.31 ± 0.20 <sup>b</sup>	21.35 ± 0.20 <sup>a</sup>	1.31 ± 0.03 <sup>b</sup>	1.29 ± 0.03 <sup>c</sup>
Black Satin	6.45 ± 0.04 <sup>b</sup>	7.24 ± 0.50 <sup>b</sup>	25.96 ± 0.82 <sup>e</sup>	27.08 ± 0.84 <sup>b</sup>	20.40 ± 0.51 <sup>b</sup>	21.28 ± 0.58 <sup>a</sup>	1.27 ± 0.02 <sup>c</sup>	1.27 ± 0.02 <sup>d</sup>
Loch Ness	7.76 ± 0.01 <sup>a</sup>	7.61 ± 0.34 <sup>a</sup>	28.13 ± 0.80 <sup>a</sup>	27.15 ± 0.81 <sup>b</sup>	21.78 ± 0.56 <sup>a</sup>	20.69 ± 0.53 <sup>b</sup>	1.30 ± 0.05 <sup>b</sup>	1.32 ± 0.06 <sup>b</sup>
Chester Thornless	5.31 ± 0.00 <sup>c</sup>	6.11 ± 0.23 <sup>d</sup>	24.13 ± 0.27 <sup>d</sup>	25.01 ± 0.29 <sup>c</sup>	19.72 ± 0.34 <sup>c</sup>	20.82 ± 0.33 <sup>b</sup>	1.23 ± 0.02 <sup>d</sup>	1.20 ± 0.02 <sup>f</sup>
Navaho	5.39 ± 0.20 <sup>c</sup>	5.90 ± 0.38 <sup>e</sup>	22.65 ± 0.49 <sup>f</sup>	23.12 ± 0.48 <sup>d</sup>	19.46 ± 0.24 <sup>c</sup>	19.80 ± 0.22 <sup>c</sup>	1.16 ± 0.03 <sup>e</sup>	1.17 ± 0.03 <sup>g</sup>
Mean over years	5.95 ± 0.50 <sup>B</sup>	6.67 ± 0.34 <sup>A</sup>	25.18 ± 0.94 <sup>A</sup>	25.96 ± 0.75 <sup>A</sup>	19.78 ± 0.49 <sup>B</sup>	20.51 ± 0.28 <sup>A</sup>	1.27 ± 0.03 <sup>A</sup>	1.26 ± 0.03 <sup>A</sup>

Means followed by the same small letters, within the same column, are not significantly different (*LSD* at  $P \leq 0.05$ ); means followed by the same capital letters in latest row, are not significantly different (*LSD* at  $P \leq 0.05$ )

Table 3. Soluble solids, titratable acidity and ripening index of blackberry cultivars

Cultivar	Soluble solids (°Brix)		Titratable acidity (%)		Ripening index	
	2010	2011	2010	2011	2010	2011
Dirksen Thornless	6.80 ± 0.26 <sup>e</sup>	9.76 ± 0.13 <sup>a</sup>	1.51 ± 0.27 <sup>d</sup>	1.24 ± 0.15 <sup>c</sup>	4.50 ± 0.33 <sup>d</sup>	7.87 ± 0.81 <sup>b</sup>
Thornfree	7.70 ± 0.18 <sup>d</sup>	8.66 ± 0.13 <sup>c</sup>	1.72 ± 0.03 <sup>b</sup>	1.60 ± 0.41 <sup>b</sup>	4.48 ± 0.71 <sup>d</sup>	5.41 ± 0.48 <sup>e</sup>
Čačanska Bestrna	6.40 ± 0.14 <sup>g</sup>	7.82 ± 0.18 <sup>d</sup>	1.89 ± 0.66 <sup>a</sup>	1.64 ± 0.41 <sup>a</sup>	3.39 ± 0.02 <sup>e</sup>	4.77 ± 0.69 <sup>g</sup>
Black Satin	6.70 ± 0.39 <sup>f</sup>	6.89 ± 0.14 <sup>e</sup>	1.57 ± 0.19 <sup>c</sup>	1.42 ± 0.06 <sup>c</sup>	4.27 ± 0.67 <sup>d</sup>	4.85 ± 0.43 <sup>f</sup>
Loch Ness	9.25 ± 0.29 <sup>b</sup>	9.35 ± 0.12 <sup>b</sup>	1.56 ± 0.33 <sup>c</sup>	1.42 ± 0.20 <sup>c</sup>	5.93 ± 0.09 <sup>c</sup>	6.58 ± 0.91 <sup>d</sup>
Chester Thornless	9.20 ± 0.20 <sup>c</sup>	9.27 ± 0.19 <sup>b</sup>	1.44 ± 0.13 <sup>e</sup>	1.27 ± 0.22 <sup>d</sup>	6.39 ± 0.03 <sup>b</sup>	7.30 ± 0.55 <sup>c</sup>
Navaho	9.35 ± 0.20 <sup>a</sup>	9.67 ± 0.22 <sup>a</sup>	1.33 ± 0.03 <sup>f</sup>	1.08 ± 0.11 <sup>f</sup>	7.03 ± 0.19 <sup>a</sup>	8.95 ± 0.79 <sup>a</sup>
Mean over years	7.91 ± 0.50 <sup>B</sup>	8.77 ± 0.40 <sup>A</sup>	1.57 ± 0.07 <sup>A</sup>	1.38 ± 0.08 <sup>B</sup>	5.14 ± 0.50 <sup>B</sup>	6.53 ± 0.61 <sup>A</sup>

The same small letters in columns show insignificant differences ( $P \leq 0.05$ ) by *LSD* test among cultivars; the same capital letters in the latest row show insignificant differences ( $P \leq 0.05$ ) by *LSD* test between years

loss of TA in blackberry fruits was accelerated with increasing pre-harvest temperatures (Naumann and Wittenburg 1980). Namely, in our trial, mean monthly temperatures during all maturity period in 2010 were notably lower than in 2011 (Table 1).

The RI plays an important role for evaluating the eating quality, consumer acceptance (Perkins-Veazie and Collins 2001) and optimum time for harvesting (Kafkas et al. 2006). The RI of berries was significantly dependent on the cultivars in two seasons (Table 3). Higher value was observed in NV, and lower in ČB. Türemiş et al. (2003) also found that NV had the best RI, whereas LN had the

poorest. The results show significant year-by-year differences (Table 3), suggested environmental factors and growing seasons influence on RI levels (Reyes-Carmona et al. 2005).

All the above results indicated a very complex nature of the accumulation of SSC and TA in fruits of blackberries and importance of the cultivar's choice in order to maximize the potential performance of a genotype under the cultural management in some years (Siriwoharn et al. 2004).

**Evaluation of total phenolic and flavonoid content and antioxidant capacity.** There were large variations in TPH among cultivars (Table 4).

Table 4. Total phenolic and flavonoid content, and total antioxidant capacity of blackberry cultivars

Cultivar	Total phenolic content (mg GA/g d.e.)		Total flavonoid content (mg RU/g d.e.)		Total antioxidant capacity (mg AA/g d.e.)	
	2010	2011	2010	2011	2010	2011
Dirksen Thornless	60.08 ± 2.02 <sup>c</sup>	69.46 ± 1.65 <sup>c</sup>	29.17 ± 3.19 <sup>c</sup>	33.53 ± 2.51 <sup>e</sup>	103.99 ± 6.32 <sup>cd</sup>	123.80 ± 8.03 <sup>e</sup>
Thornfree	55.01 ± 1.34 <sup>c</sup>	63.61 ± 2.16 <sup>d</sup>	91.44 ± 4.87 <sup>a</sup>	105.10 ± 3.27 <sup>a</sup>	121.90 ± 9.11 <sup>c</sup>	145.12 ± 14.76 <sup>c</sup>
Čačanska Bestrna	96.01 ± 4.75 <sup>b</sup>	111.00 ± 5.01 <sup>b</sup>	47.53 ± 2.76 <sup>b</sup>	54.63 ± 3.07 <sup>c</sup>	194.06 ± 5.07 <sup>b</sup>	231.03 ± 7.51 <sup>b</sup>
Black Satin	357.42 ± 7.99 <sup>a</sup>	413.20 ± 9.56 <sup>a</sup>	85.01 ± 3.12 <sup>a</sup>	97.71 ± 1.99 <sup>b</sup>	262.68 ± 5.67 <sup>a</sup>	312.72 ± 3.04 <sup>a</sup>
Loch Ness	39.26 ± 1.69 <sup>d</sup>	45.39 ± 2.52 <sup>e</sup>	8.91 ± 1.01 <sup>d</sup>	10.24 ± 2.84 <sup>f</sup>	110.35 ± 4.49 <sup>cd</sup>	131.37 ± 3.18 <sup>d</sup>
Chester Thornless	29.69 ± 1.37 <sup>e</sup>	34.33 ± 2.22 <sup>f</sup>	45.22 ± 4.65 <sup>b</sup>	51.98 ± 3.55 <sup>d</sup>	90.77 ± 3.91 <sup>d</sup>	65.56 ± 2.67 <sup>g</sup>
Navaho	26.99 ± 1.39 <sup>e</sup>	33.52 ± 2.13 <sup>f</sup>	6.02 ± 1.97 <sup>d</sup>	7.69 ± 1.36 <sup>g</sup>	94.39 ± 5.78 <sup>d</sup>	111.06 ± 4.31 <sup>f</sup>
Mean over years	99.99 ± 51.35 <sup>B</sup>	110.07 ± 51.51 <sup>A</sup>	44.75 ± 12.76 <sup>B</sup>	51.55 ± 14.61 <sup>A</sup>	139.73 ± 24.39 <sup>B</sup>	161.88 ± 30.78 <sup>A</sup>

The values for each cultivar are presented as means of triplicate analyses for each year; the same small letters in columns show insignificant differences among cultivars at  $P \leq 0.05$  by *LSD* test; the different capital letters in the latest row show significant differences between years at  $P \leq 0.05$  by *LSD* test

The highest TPH was found in BS and the lowest in CT and NV. Clark et al. (2002) and Cho et al. (2005) found that LN had higher amount of total phenolics than NV, which confirmed our results. Very high variations and discrepancy among cultivars and between years were observed by other authors (Sellappan et al. 2002, Reyes-Carmona et al. 2005). This could be connected with genetic differences, maturity at harvest, cultural practices, different extraction and laboratory methods employed (Clark et al. 2002).

The wide variations were observed among cultivars regarding TFC (Table 4), being the highest in TF, and the lowest in NV. An important lower or higher variability was reported previously (Sellappan et al. 2002, Siriwoharn et al. 2004). The different TFC reported when compared to previous studies may be due to maturity at harvest and cultivar difference (Cho et al. 2005, Reyes-Carmona et al. 2005).

Data in Table 4 showed that TAC values significantly varied among cultivars and between years. The BS and ČB had the highest antioxidant capacity and CT had the lowest. Based on the results of our study, BS, ČB, somewhat TF, ranks highest in terms of antioxidant capacities due to their high acidity content (Wang 2007). Reyes-Carmona et al. (2005) reported a high variability among cultivars and concluded that genotypes were a major factor affecting antioxidant capacity in blackberries.

Differences between years for TPH, TFC, and TAC were observed, being higher in 2011 than in 2010 (Table 4), suggested growing season, climate and region influence on antioxidant power of blackberries (Sellapan et al. 2002). Namely, plants grown in cool day and night temperatures generally had the lowest antioxidant capacity (Wang 2007).

It is well established that a strong and positive relationship exists between TPH and TFC or TAC, although correlation between TPH and TFC was not significant (Table 5). Similar findings were observed by Reyes-Carmona et al. (2005).

Finally, all cultivars had good adaptation capability to acidic soil, high temperatures and insufficient water and can be recommended to growers as a commercial crop in similar conditions.

Table 5. Correlation matrix among total phenolic content, total flavonoid content and antioxidant capacity

Variables	TPH	TFC	TAC
Total phenolic (TPH)	1	0.577	0.906*
Flavonoid content (TFC)	–	1	0.552
Total antioxidant capacity (TAC)	–	–	1

\*significant correlation coefficient at  $P = 0.05$

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