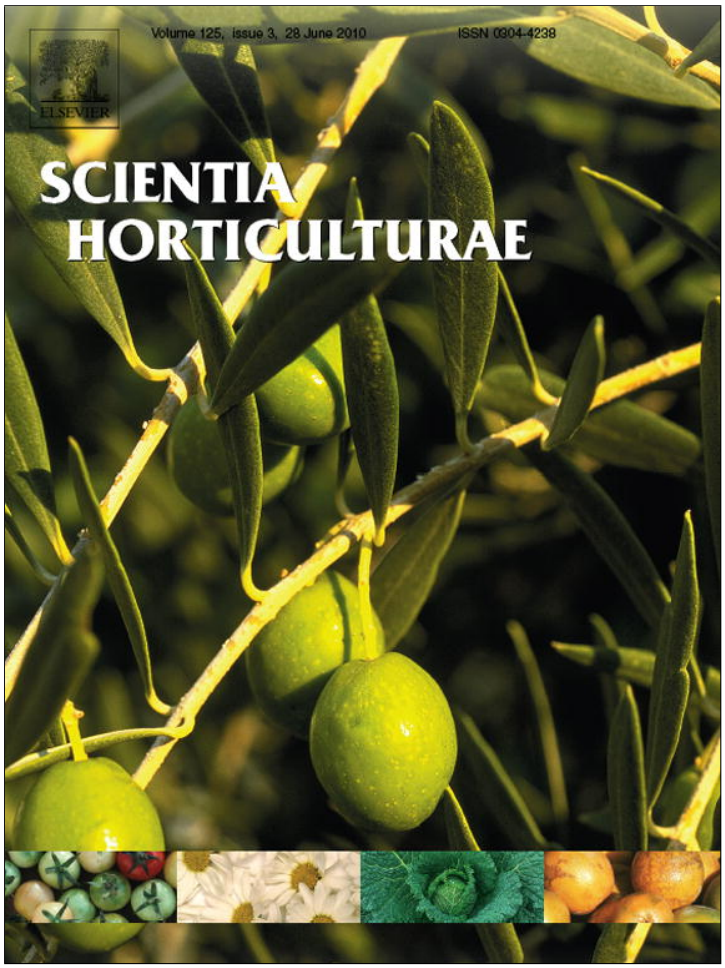

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Effects of cultivar, orchard elevation, and storage on fruit quality characters of sweet cherry (*Prunus avium* L.)

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ABSTRACT

Fruit quality characters were analysed in the sweet cherry cultivars, Burlat, Van, Tragana and Mpakirtzeika, harvested from low (39–59 m), medium (216 m) or high (490–546 m) elevation sites. The effects of storage for 2 or 4 days at 2 °C and 1 day at 20 °C on the fruit antioxidant contents were also evaluated. Tragana and Mpakirtzeika had greater fruit fresh weight (FW) and total soluble solid content compared to Van and Burlat, the latter being the most red colored. Tragana and Burlat had greater total phenolic content and total antioxidant capacity, measured by DPPH extinction, compared to Mpakirtzeika and Van (mean values 204.4 mg vs. 103.7 mg gallic acid equivalent 100 g^{-1} FW, and 176.1 mg vs. 79.3 mg ascorbic acid equivalent 100 g^{-1} FW, respectively). The geographic elevation had a marked influence on the cherry antioxidant content in all studied cultivars, apart from Van, with high elevation orchards producing cherries with greater contents of antioxidant compounds compared to lower elevation orchards. Changes in the antioxidant contents during storage were depended on the cultivar and some times on the orchard elevation. Total antioxidant capacity was significantly correlated with total phenolic content in Tragana, Burlat and Mpakirtzeika, but not in Van; nevertheless this was not the case during storage.

1. Introduction

Apart from several essential dietary components, such as vitamins, minerals, protein and carbohydrate, cherries also contain other phytonutrients that may provide benefits beyond the prevention of dietary deficiencies such as compacting multiple disease states. The consumption of sweet or sour cherries reduce the risk of cancer (Kang et al., 2003), pain from arthritis and inflammation (Jacob et al., 2003; Seeram et al., 2002), symptoms of exercise-induced muscle damage (Connolly et al., 2006), oxidative stress in older people (Traustadóttir et al., 2009), and offer protection against neurodegenerative diseases (Kim et al., 2005). The beneficial effects of cherries may be attributed to the presence of phenolics such as anthocyanins, and melatonin that exert potent antioxidant capacity (Seeram et al., 2002; Vinson et al., 2001; Burkhardt et al., 2001).

Clearly, the genetic background is the first parameter with the potential to influence the antioxidant content in a commodity. Cherries ranked first distantly followed by other 19 fruits when comparing their antioxidant capacity (Vinson et al., 2001). Significant inter-cultivar variation in the phenolic content and antioxidant capacity has also been documented in cherries (Usenik and Fabčič, 2008; Gao and Mazza, 1995).

Parameters such as climatic, agronomic (crop load, culture in greenhouses or fields, biological culture, etc.) and degree of ripening may also exert an important role in the phenolic content of fruit tissues (Drogoudi et al., 2009; McGhie et al., 2005; Serrano et al., 2005; Gonçalves et al., 2004a,b). The biosynthesis of phenolic compounds is triggered by exposure to stress conditions, as a natural defence system, generating a significant amount of the above variability (Rivero et al., 2001; Kataoka et al., 1996; Cohen et al., 1994). Little is known on the effects of climatic parameters on fruit quality and antioxidant contents in sweet cherry. Although there is little scientific evidence, it is generally accepted that cherries from higher compared to lower elevations are of better quality. In the only relevant study, the cherry phenolic and anthocyanin contents were greater in a warmer compared to a cooler year (Gonçalves et al., 2004a).

During storage the fruit metabolism continues inducing changes in the phenolic and other antioxidant content (Amarowicz et al., 2008). In sweet cherries storage was found to have variable effects on the total phenolic and anthocyanin contents which were depended on the cultivar and the storage conditions (Mozetič et al., 2006; Gonçalves et al., 2004a,b; Esti et al., 2002).

The objectives of the present work were to study the effects of different cultivars, geographic origin, with orchards located in

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Table 1
Areas, elevation, cultivars, harvest date, tree age and total and maximum yield of trees assayed.

Area/elevation	Cultivar	Harvest date	Tree age (years)	Maximum estimated yield (kg/tree)
Kolindros/39 m	Burlat	19 May	11	60–70
Makrochori/59 m	Tragana	01 June	20	100–120
Giannakochori/216 m	Burlat	16 May	16	70–80
	Van	02 June	16	70–80
	Tragana	02 June	16	90–100
	Mpakirtzeika	21 June	16	70–80
Rodochori/490 m	Van	15 June	11	70–80
	Tragana	09 June	11	70–80
	Mpakirtzeika	23 June	11	70–80
Arnissa/546 m	Tragana	23 June	18	90–100
	Mpakirtzeika	23 June	14	70–80

different elevations, and storage conditions on sweet cherry quality. The hypothesis tested was that cherries harvested from geographic areas with higher altitude, where stress conditions are more pronouncing, would contain more antioxidants compared to those harvested from lower altitudes.

2. Materials and methods

Fruit from the sweet cherry (*Prunus avium* L.) cultivars Burlat, Van, Tragana and Mpakirtzeika (the latter two being of Greek origin) were harvested during commercial maturation from orchards positioned in the prefectures of Pieria, Imathia and Pella, in northern Greece. The orchards were in different elevations, with low elevation sites being Makrochori, Imathia (40°32′46.96″N, 22°13′55.16″E, at 59 m altitude) and Kolindros, Pieria (40°30′28.43″N, 22°30′43.35″E, at 39 m altitude), a medium elevation site being Giannakochori, Imathia (40°40′55.65″N, 22°04′06.50″E, at 216 m altitude) and high elevation sites being Rodochori, Imathia (40°41′30.10″N, 22°01′20.59″E, at 490 m altitude) and Arnissa, Pella (40°48′46.90″N, 21°48′51.65″E, at 546 m altitude). All cultivars were grafted on Mazzard rootstock and the harvest date, tree age and maximum yield of trees assayed are presented in Table 1.

Fruit with the pedicel (about 1.5 kg) was harvested from four trees, transferred to the lab and separated in three parts of 0.5 kg. Fruit fresh weight (FW), color, total soluble solid and total acid contents were measured in 20 fruits from the first part. The fruit color L^* (brightness or lightness; 0 = black, 100 = white), a^* ($-a^*$ = greenness, $+a^*$ = redness) and b^* ($-b^*$ = blueness, $+b^*$ = yellowness) variables were measured using the chromatometer Minolta (Model CR-200; Minolta, Ramsey, NJ, USA), total soluble solids using a refractometer (Model PR-1; Atago, Tokyo, Japan) and expressed as %, and total acidity (TA) were analysed in juice by titration with 0.1 N NaOH and expressed as malic acid content (g L^{-1}).

In the second part the pedicel and stones were removed and the remaining fruit was stored at −80 °C for analyses of total antioxidant capacity and total phenolic content.

The remaining third part of fruit (0.5 kg) was stored in polyethylene bags at 1–2 °C for 2 or 4 days and in the shelf at 20 °C for 1 day. Measurements of fruit antioxidant contents were made on day 3 and day 5.

2.1. Antioxidant capacity and total phenolics assay

Frozen samples (about 1 g) from the flesh of three replicate fruits were homogenized in 8 mL of 80% MeOH/H_2O (v/v) in mortar and pestle. The extract was centrifuged at 10,000 × *g* for 10 min at 4 °C and the supernatant recovered.

Antioxidant capacity was measured using the stable 1,1-diphenyl-2-picryl hydrazyl (DPPH) free radical (Blois, 1958), which has an intense violet color, but turns colorless as unpaired electrons are sequestered by antioxidants. Reaction mixtures containing 0 or 20 μL extract, 2300 μL of 106.5 μM DPPH in MeOH and 680 μL H_2O were vortexed, and then held at room temperature in darkness for 4 h. The absorbance of the reaction mixtures was measured at 517 nm and AEAC (mM) was extrapolated from a standard curve prepared using 0–2.7 mM ascorbate.

Total soluble phenolics contents were determined according to the Folin-Ciocalteu's procedure (Singleton and Rossi, 1965) and the results were expressed as mg gallic acid equivalent 100 g^{-1} FW. All analyses were made in triplicate.

2.2. Statistical analyses

Analysis of variance (ANOVA) and correlation analysis were performed using SPSS (SPSS Inc., Chicago, IL, USA). Treatment means were separated using the Duncan's multiple range test where ANOVA *F*-tests were significant, using a *P*-value of 0.05.

3. Results and discussion

A comparison among the studied cultivars on the fruit quality characters was made for fruit harvested from Giannakochori, where all cultivars received the same cultivation practices (Table 2). Mpakirtzeika had the greatest fruit FW (11.4 g), followed by Tragana (10.5 g) and the lowest values were found in Burlat and Van (mean 9.5 g). Tragana contained the greatest total soluble solid content (18.0%), followed by Mpakirtzeika and Van (mean 15.6%), and the lowest value was found in Burlat (13.5%). Total acid content followed the descending order, Mpakirtzeika (12.2 g L^{-1}) > Van (10.3 g L^{-1}) > Tragana (9.1 g L^{-1}) > Burlat (3.3 g L^{-1}). Burlat cherries were the most red colored (a^* = 28.7) followed by Van (a^* = 24.1) and the rest cultivars (mean a^* = 19.7). Mean values of L^* and b^* parameters were also higher in Burlat compared to the rest cultivars. Fruit FW, sweetness and skin color are parameters that greatly affect the consumer acceptance of sweet cherry (Crisosto et al., 2003). Therefore, when considering fruit FW and total soluble solid content the Greek cultivars Tragana and Mpakirtzeika are of superior quality compared to Van and Burlat, although in respect to coloration Burlat would be more favored to consumers.

Total phenolic content was greater in cultivars Burlat and Tragana compared to Mpakirtzeika and Van (212.3, 196.5, 104.2 and 103.1 mg gallic acid equivalent 100 g^{-1} FW) and a similar trend was found for the total antioxidant capacity (184.2, 168.0, 93.5 and 65.0 mg ascorbic acid equivalent 100 g^{-1} FW, respectively) (Table 2). Similar mean values of total phenolics for Burlat and Van cherries were reported by Gonçalves et al. (2007) and Gao and Mazza (1995) while lower values for Burlat was reported by Usenik et al. (2008). Considering that in the study by Usenik et al. (2008) Burlat was found to have the greatest antioxidant capacity among

Table 2
Fruit weight (g), color parameters L^*, a^* and b^*, total soluble solids (TSS) (%), total acids (g L^{-1}), total phenolics (mg of gallic acid equiv. 100 g^{-1} FW) and ascorbate equivalent antioxidant capacity (AEAC) (mg of ascorbic acid equiv. 100 g^{-1} FW) of different cherry cultivars and areas.

Cultivars	Areas/elevation	Fruit weight	Color parameters			TSS	Total acids	Total phenolics	AEAC
			L^*	a^*	b^*				
Burlat	Kolindros/39 m	10.1	32.0	30.4	−0.59	15.5 a	2.9 b	153.8 b	135.7 b
	Giannakochori/216 m	9.7	32.1	28.7	−1.56	13.5 b	3.3 a	212.3 a	184.2 a
Van	Giannakochori/216 m	9.2	29.1	24.1 a	−4.17 a	15.6 b	10.3 a	91.7	65.0
	Rodochori/490 m	8.8	28.7	17.3 b	−6.76 b	19.7 a	8.0 b	103.1	79.3
Tragana	Makrochori/59 m	7.4 b	27.9 b	14.5 bc	−7.64 bc	17.8	10.1 a	143.4 c	120.8 c
	Giannakochori/216 m	10.5 a	29.3 a	18.7 a	−6.23 a	18.0	9.1 ab	196.5 b	168.0 b
	Rodochori/490 m	9.9 a	27.7 b	15.0 b	−7.22 b	17.1	8.2 b	265.4 a	209.3 a
	Arnissa/546 m	7.0 b	28.1 b	11.8 c	−8.27 c	18.3	8.7 b	212.7 b	175.4 b
Mpakirtzeika	Giannakochori/216 m	11.4 a	28.5 b	20.6 b	−4.95	15.5	12.2	104.2 b	93.5 b
	Rodochori/490 m	10.2 b	31.2 a	25.8 a	−3.53	15.7	11.1	197.2 a	138.6 a
	Arnissa/546 m	7.9 c	30.2 a	20.9 b	−5.47	15.7	11.7	179.7 a	145.5 a

Means separation within elevation sites by Duncan's multiple range test ($P \leq 0.05$).

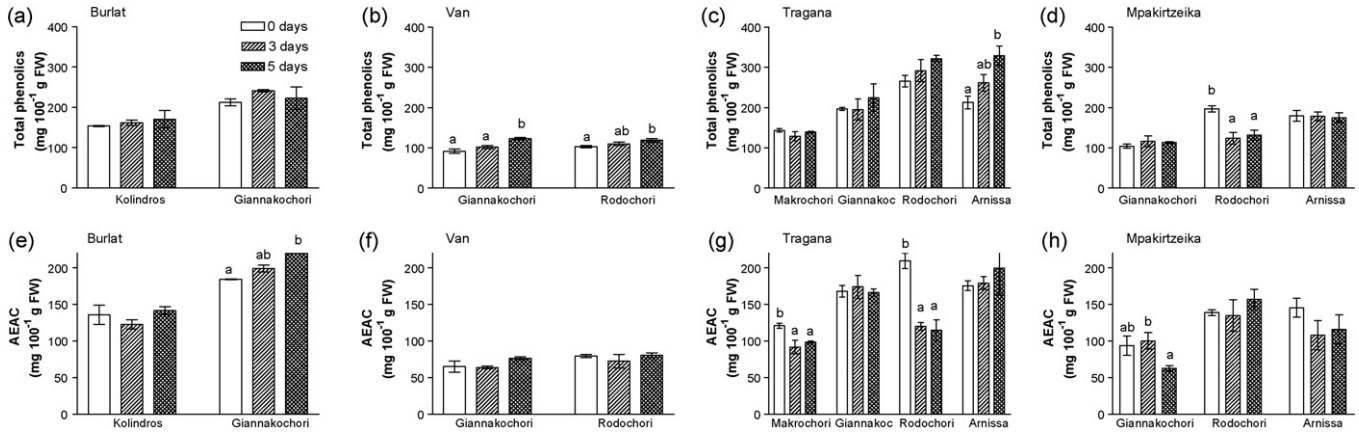

Fig. 1. Changes in the total phenolics (a–d) and ascorbate equivalent antioxidant capacity (AEAC) and (f–h) during storage at 1–2 °C for 2 or 4 days and in the shelf at 20 °C for 1 day in the sweet cherry cultivars Burlat (a and e), Van (b and f), Tragana (c and g) and Mpakirtzeika (d and h) harvested from Makrochori, Kolindros, Giannakochori, Rodochori and Arnissa. Means with different letters are significantly different at $P \leq 0.05$.

other 12 sweet cherry cultivars, it is suggested that both Burlat and Tragana should be considered as fruit with high antioxidant content.

When comparing cherries from different elevation orchards, differences were found in the fruit FW, color, total soluble solid and total acid contents, but these were not related to the elevation site (Table 2). The total phenolic content and antioxidant capacity were generally greater in cherries harvested from higher, compared to lower, elevation orchards (Table 2, Fig. 1). For example, the total phenolic content was 85% greater in the high (490 m) compared to the low (59 m) elevation site in Tragana; 35% and 89% greater in the high (490 m) compared to the medium (216 m) elevation sites in Tragana and Mpakirtzeika, respectively; and 38% greater in the medium (216 m) compared to the lower (39 m) elevation site in Burlat. Nevertheless, similar values of the phenolic content and antioxidant capacity were found between the low (39 m) and medium (216 m) elevation sites in Van and this may be a cultivar characteristic. As it was hypothesised it is possible that the low temperatures in high elevation sites trigger the mechanism for producing phenolics. Similarly, cold stress induced the accumulation of phenolic compounds in apples (Thomai et al., 1998), tomato and watermelon plants (Rivero et al., 2001) and olive tress (Ortega-García and Peragón, 2009).

Effects of storage for 2 or 4 days at 2 °C and 1 day at 20 °C on the total phenolic content and antioxidant capacity were cultivar depended, and some times differed among locations sites. The total phenolic content increased during storage in cultivars Van and Tragana in most location sites, while it was not altered in Burlat or decreased in one site in Mpakirtzeika (Fig. 1). Variable was also the response of the antioxidant capacity during storage in the studied cultivars. Similarly, Gonçalves et al. (2004a) and Esti et al. (2002) found cultivar depended changes in the total phenolics and anthocyanins contents during cold or warm storage.

Total antioxidant capacity measured by DPPH extinction was significantly correlated with total phenolic content in cultivars Tragana ($r^2 = 0.891$), Burlat ($r^2 = 0.680$) and Mpakirtzeika ($r^2 = 0.767$), but not in Van. Usenik et al. (2008) similarly found that antioxidant capacity was significantly correlated with phenolics or anthocyanins or none of them in different sweet cherry cultivar, suggesting that antioxidant capacity is depended on different compounds and is specific for cultivar. In the present study there was no significant correlation between antioxidant capacity and total phenolics during storage in any cultivar. It is suggested that although phenolics may have a significant contribution to antioxidant capacity in some cherry cultivars (Tragana, Burlat and Mpakirtzeika), during storage the presence of other compounds may participate in the antioxidant action that requires further investigation.

4. Conclusions

Cultivar Tragana is of superior quality in respect to physical and antioxidant characteristics, while Mpakirtzeika would be appreciated by consumers interested in cherries with relatively higher acidity. Cherries harvested from higher altitudes had usu-

ally greater phenolic and antioxidant capacity in the three out of four studied cultivars, although the fruit physical characters were not affected by the elevation site. A relatively short storage period for 2 or 4 days at 2 °C and 1 day at 20 °C, induced changes in the antioxidant contents which were depended on the cultivar and the harvesting location.

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