Food Chemistry 263 (2018) 67–73

Contents lists available at ScienceDirect

Food Chemistry

journal homepage: www.elsevier.com/locate/foodchem

Health and taste related compounds in strawberries under various irrigation regimes and bio-stimulant application

Burcak Kapur[a], Mehmet Ali Sarıdaş[b,*], Eser Çeliktopuz[a], Ebru Kafkas[b], Sevgi Paydaş Kargı[b]

[a] Department of Agricultural Structures and Irrigation Engineering, University of Çukurova, Adana 01330, Turkey
[b] Department of Horticulture, University of Çukurova, Adana 01330, Turkey

ARTICLE INFO

Keywords:
Antioxidant
Eating quality
Phenol
Seaweed extract
Deficit irrigation

ABSTRACT

Strawberry has a unique status within the fruit species in terms of health and taste related compounds. This experimental study concerned the application of a bio-stimulant at various drip irrigation levels (IR125, IR100, IR75 and IR50). The effects of the bio-stimulant (seaweed extract) on the eating quality, i.e., the taste-related (TSS, fructose, glucose, sucrose and citric, malic, L-ascorbic acid), and health-related (antioxidant activity, total phenol, myricetin and quercetin) compounds were studied in two strawberry cultivars. The 'Rubygem' with its higher sugar and lower acid content has been more preferable than the 'Kabarla' cultivar. The bio-stimulant contributes to taste by improving the TSS, fructose, sucrose and also to health by increasing the quercetin content of the fruit which is associated to the cardiovascular properties and cancer reducing agents. The experiment conducted revealed significant increases only in the TSS contents and antioxidant activity under the IR50 and IR75 deficit irrigation treatments.

1. Introduction

Berries are highly consumed for their attractive colour, delicate texture, and unique flavor and known as rich sources of natural antioxidants (Manganaris, Goulas, Vicente, & Terry, 2014). Their consumption has been associated to various cardiovascular, anti-proliferative and neurologic benefits. Berry fruits have vitamins, minerals, folate and fiber which are the main dietary components and also contain phytochemical compounds mostly represented by polyphenols (Giampieri et al., 2012). Enhancement of strawberry production needs the availability of high water sources due to their large leaf area, shallow root system, and fruits with high water contents (Chandler & Ferre, 1990). According to a UNESCO report, one-third of the world's population lives under the water scarcity condition, and it is predicted that this will reach over two-thirds in 2025 (UNESCO, 2009). Increasing world population demands increased land and water use. In this context, although farmers of the old world are knowledgeable and experienced in traditional crop production, they are in need of the contemporary clues obtained via the field experiments conducted for Sustainable land and water management. Farmers generally use their past experience to determine the optimal amount of irrigation water by observing the weather conditions and the visual plant indicators of stress. However, they tend to use excessively or inadequately supplied water, which can have a negative impact on the vegetative part of the crop and its fruit size (Ghaderi, Normohammadi, & Javadi, 2015; Liu et al., 2007). The amount of water applied not only affects yield and the vegetative part of the plant (Adak, Gubbuk, & Tetik, 2018; Yuan, Sun, & Nishiyama, 2004) but also influences the fruit quality parameters of taste and health related compounds (Giné-Bordonaba & Terry, 2010; Giné-Bordonaba and Terry, 2016; Weber et al., 2017). Deficit irrigation is applied not only to decrease water consumption but also to improve the quality parameters of taste and health via related compounds (Adak, et al., 2018; Giné-Bordonaba & Terry, 2010; Weber et al., 2017). Quality parameters of fruit such as color, firmness and chemical composition influence the consumer preference (Ornelas-Paz et al., 2013). Fruit flavor is mainly determined by the contents of total soluble solids, total acids and their ratios (Krüger et al., 2012). The content of taste and health-related compounds in strawberry could vary depending on the genotype (Kosar, Kafkas, Paydaş, & Baser, 2004), agricultural practice, environment, and maturity (Tulipani et al., 2011). However, the genotypic properties of strawberries were found to be more influential than cultivation practices for quality parameters (Capocasa, Scalzo, Mezzetti, & Battino, 2008). Some pre-harvest practices were applied together with irrigation levels for increasing health-related compounds (Giné-Bordonaba & Terry, 2016). Bio-stimulants have been gaining attention especially in sustainable agriculture, because their application enhances several physiological processes, that increase nutrient use efficiency, encouraging plant development and reducing

* Corresponding author.
E-mail address: masaridas@cu.edu.tr (M.A. Sarıdaş).

https://doi.org/10.1016/j.foodchem.2018.04.108
Received 16 February 2018; Received in revised form 23 April 2018; Accepted 24 April 2018
Available online 30 April 2018
0308-8146/ © 2018 Elsevier Ltd. All rights reserved.

fertilizer consumption (Bulgari, Cocetta, Trivellini, Vernieri, & Ferrante, 2015; Kunicki, Grabowska, Sekara, & Wojciechowska, 2010).

To the best of our knowledge, there has been no research conducted to investigate the combined effects of irrigation regimes with bio-stimulant application on health and taste-related compounds under farmers field conditions in the Mediterranean region of Turkey. Accordingly, the aim of this study is to determine the influence of the different irrigation levels (IR125, IR100, IR75, and IR50), bio-stimulant applications and their combined effects on health (total antioxidant, total phenol, quercetin and myrcetin) and taste-related (TSS, individual sugars and acids) compounds for two strawberry cultivars (Rubygem cv. and Kabarla cv.).

2. Materials and methods

The experiment was conducted in a high tunnel at the experimental farm of the Horticulture Department of the University of Çukurova (latitude: 36°59′9N, longitude 35°27′7E and altitude 20 m above sea level). Strawberries (*Fragaria* x *ananassa* Duch. Rubygem, Kabarla cv.) were planted on November 10 in 2015 and harvesting continued until June 8 in 2016.

The berries were planted in trapezoidal raised beds measuring 0.70 m from the base, 0.50 m at the top, with a height of 0.30 m. The distance between the beds was 0.4 m and the beds were covered by 0.05 mm-thick two-sided polyethylene mulch cover with a grey upper side and black under side suitable to the conventional cultural practices in the area. After planting, equal amounts of water was applied to all treatments until the plants were 3 trifoliate (28 January). Fertilizer was applied uniformly to all treatments during the trial through drip irrigation and foliar spraying. Plant protection was carried out by the timely spraying of the agricultural pesticides. This production system was described previously by Saridas, Paydas Kargı, Kapur, and Celiktopuz (2017).

The trail was implemented as a 2 × 4 × 2 factorial scheme in split split plot design at 3 replicates at totally 48 plots. The strawberry plant was subjected to four irrigation water levels as IR50, IR75, IR100, IR125 in which the water quantities applied were 0.5, 0.75, 1.00 and 1.25 times the pan evaporation measured by the US Weather Service Class A pan with a standard 120.7 cm diameter and 25 cm depth placed over the crop canopy in the center of the high tunnel.

The commercial bio-stimulant applied in this experimental study is natural, non-toxic and a new generation product obtained from seaweed extraction with organic certification. Bio-stimulant applications started approximately 2 months after planting with foliar application (Table 1).

The content of the bio-stimulant was certified by the BCS Öko-Garantie GMBH, Nurnberg, Germany as seaweed extract containing 67% organic matter, 1.5% K2O , 18% alginic acid and 250 ppm gibberellic acid.

Irrigation water (salinity 0.18 dS/m) was applied using drip tubes with emitters spaced every 0.3 m, with a flow rate of 2.7 L h^{-1} and the amount of irrigation water was calculated by using Eq (1).

$$t = \frac{A \times Ep \times pc \times kcp}{q \times n} \quad (1)$$

where, t is the irrigation time (hour), A is the area of the plot (m^2), Ep is the cumulative free surface water evaporation at irrigation interval (mm), pc is the plant cover (%), kcp is the crop-pan coefficients of 0.5, 0.75, 1.00, 1.25 for various irrigation regimes throughout the trial, q is the flow rate of emitters and n is the number of emitters in the plot.

Table 1
Application times and dose of bio-stimulant.

Application No	Application time	Dose of application
1	12.01.2016	20 g/15 l water
2	02.02.2016	20 g/15 l water
3	22.02.2016	20 g/15 l water
4	14.03.2016	20 g/15 l water

All results given here were obtained from the fruits harvested on 10 May 2016 for which the yield was maximum. The effect of different irrigation regimes can be seen clearly from the results. From the initiation of the treatment to the end of the trial, a total of 417, 345, 274 and 203 mm of water were applied to treatments IR125, IR100, IR75 and IR50 respectively.

As a pomological analysis, total soluble solids in fruit juice of each plot were determined by a hand-type refractometer (ATAGO ATC-1, Tokyo, Japan). The Shimadzu HPLC system, equipped with a pump and a refractive index detector (RID-10A), was used for sugar analysis (fructose, glucose, sucrose). The sugar contents were determined by using an Inertsil NH2 column (4.6 mm × 250 mm) maintained at 40 °C. The organic acid contents (citric, malic, L-ascorbic acid) were analyzed by using an HPLC system (Agilent 1100 series G1322A; Germany) according to the method of Bozan, Tunalıer, Koşar, Altıntaş and Başer (1997). An Aminex HPX-87H column (300 mm × 7.8 mm) was used in the HPLC system, which was controlled by Agilent software run on a personal computer. The organic acid content of the samples was determined qualitatively and quantitatively at a wavelength of 210 nm by comparison of the external standard calibration curve and the retention time of the standard.

Vitamin C analysis of the fruit juice of the strawberries was carried out by using an HPLC system (LC-10A HPLC series; Shimadzu, Kyoto, Japan), a UV detector, and a Prevail organic acid column (150 mm × 4.6 mm, 5 µm), according to the method developed by Bozan, Tunalıer, Koşar, Altıntaş and Başer (1997). Determination of the vitamin C content of the samples was performed qualitatively and quantitatively at a wavelength of 242 nm by comparison of the external standard calibration curve and the retention time of the standard.

The total phenolic content of the fruit juices was determined by using a Folin–Ciocalteu reagent with the modified method of Spanos and Wrolstad (1990). The absorbance of all samples was measured at 760 nm utilizing a Multiskan GO microplate spectrophotometer. The results were expressed as mg gallic acid equivalent/g weight (mg GAE/g FW) (Bayır, 2007).

The FRAP assay was done according to the method of Benzie and Strain (1996), with some modifications. The results were expressed in µmol TE/g fresh mass. Additional dilution was needed if the FRAP value measured was over the linear range of the standard curve. The content of myricetin and quercetin was determined according to the method of Kosar et al. (2004) by using of HPLC/fluorescence detector.

The experiment was conducted as a three factors randomized complete block design with split split plot with three replications. The data obtained were analyzed with the statistical program JMP version 5.0.1 (SAS Institute Inc., Cary, NC). ANOVA was carried out to determine the differences between the cultivars, irrigation regimes and bio-stimulant application in terms of taste-related and health-related compounds. A least significant difference test was conducted to examine the differences among the groups. The comparisons that yielded $P \leq 0.05$ were considered to be statistically significant.

3. Results and discussion

3.1. Taste related fruit quality indicators

Significant differences between the cultivars treated with varying irrigation regimes were determined for TSS and individual sugars (Table 2). The TSS, fructose and glucose levels are higher in 'Rubygem' than 'Kabarla'. However, Kabarla has a higher sucrose level. These results highlight the differences in the sugar accumulation mechanisms between the 'Rubygem' and 'Kabarla' cultivars with different contents of total soluble solids (TSS) and amounts of individual sugars. Our

Table 2
Effects of the applied bio-stimulant under different irrigation level of strawberry varieties on TSS (%) and individual sugars.

	Application	Cultivar	Irrigation Regime				Cul X App	App. Av.
		Rubygem	IR50	IR75	IR100	IR125		
TSS (%)	Control		9.00 a	8,50 b	9.05 a	8.15 de	8.68	8.33 B
		Kabarla	8.00 e	8.00 e	8.00 e	8.00 e	8.00	
	App X Irr		*8.50 B*	*8.25 C*	*8.53 B*	*8.08 D*		
	Bio stimulant	Rubygem	8.55 b	9.15 a	8.50 b	9.00 a	8.80	8.45 A
		Kabarla	8.25 cd	8.40 bc	7.75 f	8.00 e	8.10	
	App X Irr		*8.40 B*	*8.78 A*	*8.13 CD*	*8.50 B*		
	Irrigation Av.		8.45 A	8.51 A	8.33 B	8.29 B		
LSD_{app}*** = 0.063 LSD_{irr}*** = 0.089 $LSD_{appxirr}$*** = 0.13 $LSD_{appxirrxcul}$*** = 0.18								
Fructose (g/100 g FW)	Application	Cultivar	Irrigation Regime				Cul X App	App. Av.
	Control	Rubygem	IR50	IR75	IR100	IR125		
			3.66	3.56	3.82	3.63	3.67	3.23 B
		Kabarla	2.90	3.86	2.71	2.68	2.79	
	App X Irr		*3.28 BC*	*3.21 BC*	*3.27 BC*	*3.15 C*		
	Bio stimulant	Rubygem	3.63	3.64	3.67	3.83	3.70	3.30 A
		Kabarla	3.04	2.76	2.76	3.08	2.91	
	App X Irr		*3.33 AB*	*3.20 BC*	*3.21 BC*	*3.45 A*		
	Irrigation Av.		3.31	3.20	3.24	3.30		
LSD_{app}* = 0.071 $LSD_{appxirr}$** = 0.14								
Glucose (g/100 g FW)	Application	Cultivar	Irrigation Regime				Cul X App	App. Av.
	Control	Rubygem	IR50	IR75	IR100	IR125		
			3.44	3.37	3.62	3.44	3.47	3.00
		Kabarla	2.62	2.65	2.47	2.45	2.55	
	App X Irr		*3.03 B*	*3.0 B*	*3.05 AB*	*2.95 B*		
	Bio stimulant	Rubygem	3.39	3.48	3.44	3.61	3.48	3.05
		Kabarla	2.62	2.54	2.49	2.79	2.61	
	App X Irr		*3.0 B*	*3.01 B*	*2.97 B*	*3 .20 A*		
	Irrigation Av.		3.02	3.01	3.01	3.07		
$LSD_{appxirr}$* = 0.029								
Sucrose (g/100 g FW)	Application	Cultivar	Irrigation Regime				Cul X App	App. Av.
	Control	Rubygem	IR50	IR75	IR100	IR125		
			0.11 def	0.12 de	0.06 f	0.14 d	0.11 C	0.21 B
		Kabarla	0.37 a	0.28 c	0.31 bc	0.29 c	0.31 B	
	App X Irr		0.24	0.20	0.18	0.21		
	Bio stimulant	Rubygem	0.12 de	0.07 ef	0.12 de	0.09 def	0.10 C	0.23 A
		Kabarla	0.37 a	0.35 ab	0.34 abc	0.38 a	0.36 A	
	App X Irr		0.25	0.21	0.23	0.24		
	Irrigation Av.		0.25 A	0.21 B	0.21 B	0.23 AB		
LSD_{app}* = 0.02 LSD_{irr}* = 0.029 $LSD_{culxapp}$* = 0.029 $LSD_{appxirrxcul}$** = 0.057								

(1): Differences between the means were showed with different letters.
(2): N. S.: Not Significant, ***: $p < 0.001$; **: $p < 0.01$; *: $p < 0.05$.

results show that the fructose content is the highest (from 2.68 to 3.83 g/100 g FW), followed by glucose (2.45 to 3.62 g/100 g FW) and sucrose (0.06 to 0.38 g/100 g FW). One of the most important factors for the consumer preference is the sweetness of the strawberry, which is determined by the quality and quantity of these sugars. Contributions of these sugars to the sweetness are also different, fructose is considered to be 1.8 times sweeter than sucrose, and the sweetness of glucose is 60% of sucrose. Significant differences to be observed between the cultivars in our results are very important to emphasize influences of the irrigation regimes and the bio-stimulant application on the taste-related compounds. The bio-stimulant applications also significantly affected all the taste-related parameters positively except the glucose content, which exhibited only a small increase. The highest TSS content of 8,78% was found in IR75. However, a consistent difference was not observed among the irrigation regimes tested in terms of TSS and individual sugars.

The highest fructose was determined for the IR125 plot with 3.20 g/100 g. Interaction between the bio-stimulant application and the irrigation regimes significantly affected TSS and individual sugar contents except sucrose. Neither irrigation nor the bio-stimulant application had a significant effect on the glucose content of the fruits (Table 2), but interestingly their interaction did. This outcome may be utilized as a tool to increase the glucose content of the fruits. A significant triple interaction was found only in the TSS content that may provide us different options; if the bio-simulant was used at 'Rubygem' with IR75 (9.15%) or IR125 (9.00%). These could be the best options to obtain the highest TSS. Nevertheless, IR50 (9.00%) or IR100 (9.05%) could be the best option for the same cultivar and the control treatments. Our results agreed with those of Olsson et al. (2004), stating that glucose and fructose were the dominant sugars obtained together with considerable amounts of sucrose and different strawberry varieties had different contents of sugar. Similar results were also determined by Winardiantika, Lee, Park, and Yeoung (2015) fluctuating at harvesting periods. However, it was found by Giné-Bordonaba and Terry (2010) that the dominant sugar is sucrose in different strawberry cultivars both at the dry and fresh weight basis. Many researchers reported some qualitative and quantitative variations in the composition of individual sugars and TSS depending on the cultivar, genotypes, ripening, position of fruit at the cluster and harvesting time thus influencing the eating quality and consumer's preference of the fruits (Giné-Bordonaba & Terry, 2010; Olsson et al., 2004; Winardiantika, et al., 2015). There is no previously published study on the effects of bio-stimulants on individual sugars, which greatly influence consumer preference. A bio-stimulant, with an important role in sustainable agriculture, provides reduction in fertilization and other chemical compound applications (Russo & Berlyn, 1991). Two recent studies investigated the effects of

the bio-stimulant applications on plant growth, rooting, biomass of newly formed roots, early flowering and fruiting (Marfá, Cáceres, Polo, & Ródenas, 2008), frost resistance, yield and pomological characteristics (Bogunovic, Duralija, Gadze, & Kisic, 2015). According to the current results, the bio-stimulant could be a useful tool for improving TSS and individual sugars. Additionally, strawberry production could be more efficient by using evapotranspiration (ET)-based irrigation management (Yuan, et al., 2004). The accumulation of TSS in the strawberry fruit is induced by deficit irrigation (IR50, IR75), but the levels of soluble sugars do not increase with the severity of the drought stress. An additional interesting outcome of this experimental study is that fructose and glucose are not affected by irrigation regimes, whereas the plot with the lowest (IR50) and the highest (IR125) irrigation levels promoted sucrose accumulation in the fruit. The study conducted by Weber et al (2017), reveals the similar positive effects of deficit irrigation on the TSS contents of different cultivars. . However, in our experiment the irrigation regimes do not influence the fructose and glucose contents of the fruits. Response of the water stress is cultivar dependent due to different improved osmotic adjustment strategies (Adak, et al., 2018; Giné-Bordonaba & Terry, 2016). Many studies reported conflicting results of inconsistent impacts of deficit irrigation on sugar accumulation for different strawberry cultivars. This conflict may arise via the cultivation conditions of the experiment. The difference between the current study and the study conducted on soilless cultures by Giné-Bordonaba and Terry (2016) and Adak et al. (2018), , may be due to the lower water holding capacity of the latter that leads to the increasing effect of deficit irrigation. The effects of deficit irrigation on plant growth are generally quick and severe in soilless cultures compared to field conditions. In this respect, These are rooted from limited growing medium volume and exhaust of water source quickly compared with field conditions. Taste-related compounds such as TSS, fructose, glucose were significantly affected by the interaction between the irrigation regime and application ($P \leq 0.05$). Consequently, this revealed that the effect of the bio-stimulant application was different for the irrigation regimes tested. In this context, the highest fructose content (3.45 g/100 g) was obtained from the IR125 x bio-stimulant interaction, whereas IR50 produced the best value (3.28 g/100 g) among the control plots of the different irrigation regimes. The triple interaction was found statistically important in terms of TSS and sucrose contents. The influence of sucrose on the taste could be neglected due to its low contents. However, TSS could be improved with a good strategy by choosing the right cultivar, application and water saving irrigation regime.

Higher individual acid (taste and health-related components) contents were found in the 'Kabarla' cultivar compared to the 'Rubygem' (Table 3). The bio-stimulant application was found to have no effect on the individual acid contents, which were also inconsistent with the irrigation regimes. The results indicate that the cultivars exhibited more significant effects on strawberry individual acid contents than the bio-stimulant application. The lowest (IR50) and the highest (IR125) water supplies significantly improved the citric and L-ascorbic acid contents of fruits, whereas only the lowest irrigation regime (IR50) significantly contributed to the malic acid content of the fruits. The highest citric and L-ascorbic acid contents were found in the IR125 plot with a bio-stimulant application of 0.81 g/100 g and 53.5 mg/100 g values, respectively. Contrary to these outcomes, the highest malic acid content (0.18 g/100 g) was determined in the IR50 plot with the control plants. The triple interaction of the factors examined was found statistically significant in terms of individual acids. Citric, malic and L-ascorbic acid contents varied from 0.60 to 0.84 g/100 g, from 0.13 to 0.18 g/100 g and from 34.3 to 60.5 mg/100 g among the interactions, respectively. Parallel to the current results, Terry, Chope, and Giné-Bordonaba, (2007); Giné-Bordonaba and Terry, (2010) also determined three major acids, namely the citric, malic and ascorbic acids within the cultivars, respectively. Although the antioxidant activity of strawberry is mainly determined by the phenolic compounds, the L-ascorbic acid content contributed up to 10% to the antioxidant activity (Kalt, Forney, Martin, & Prior, 1999). Numerous studies (Giné-Bordonaba & Terry, 2016; Winardiantika, et al., 2015; Yıldız et al., 2014) manifested that individual acid contents were influenced by genotypes, the ripening stage, the pre-harvest application, irrigation and the growing medium. The citric acid contents of strawberry fruits grown in different growing substrates were reported to vary from 0.36 to 0.52 mg/g per FW (Palencia, Giné-Bordonaba, Martínez, & Terry, 2016). In another study conducted by Yildiz et al. (2014) the genotypes were found effective on vitamin C contents and especially more effective in wild genotypes. Similar to the current results, Winardiantika, et al. (2015) determined that citric acid was the dominant acid in the strawberry fruit followed by malic acid. They also documented that the amounts of both acids differed in different cultivars and harvest times. The citric acid content of Winardiantika, et al. (2015) was lower than ours, whereas the malic acid content was found to be higher due to the genetic variability of the cultivars and cultivation conditions. The influence of the genotype on the acid contents was also reported by Zheng, Kallio, & Yang (2009), where they found differences not only between species but also between cultivars due to regulated organic acid accumulation within the fruit. Moreover, the effect of the soil water level was found to be significant but inconsistent in terms of individual acids. Similar to this inconsistency determined by Zheng et al (2009), the highest citric acid content (0.75 g/100 g) was determined in the fruits obtained from the IR50 or IR125 plots of our study. A similar trend was also observed in the L-ascorbic acid content from the IR125 (53.5 mg/100 g) and IR50 (43.3 mg/100 g) plots. Contrary to these two acids, the malic acid content was increased only in the lowest irrigation plot (IR50) by 0.18%. Weber et al. (2017) found different responses of the cultivars grown in various soil moisture contents in terms of total acid contents. Similar outcomes were documented by Giné-Bordonaba and Terry (2010), who detected increases in organic acid contents under deficit irrigation in some of the strawberry cultivars tested. However, Adak, et al., (2018), reported a clear decrease in the citric acid content for all the cultivars examined under water stress. Regarding acid contents, a cultivar dependent response was obviously observed. However, another reason for the differences could be rooted from growing environments, cultivation applications, growth medium (soil or soilless), and harvesting time of fruit (Adak, et al., 2018; Giné Bordonaba & Terry, 2010; Weber et al., 2017).

There is an obvious lack of research concerning the effects of bio-stimulant applications on individual acids for strawberry. Despite this drawback, the applications of MeJa (methyl jasmonate) conducted by Giné-Bordonaba and Terry (2016) revealed significant increases at the fruit citric acid contents, but cultivar dependent contents of malic acid under pre harvest applications. This is similar to our results pointing to significant increases in the malic acid contents of 'Kabarla' fruits under bio-stimulant applications. In this context, as it is in our case, regarding the individual acids, the significant interactions of all the factors examined provides various options to obtain higher individual acid contents. We determined the highest citric and L-ascorbic acid contents by bio-stimulant applications in the 'Rubygem' fruits of the IR125 plot and the highest malic acid contents in the fruits of the 'Kabarla' of the IR50 plot. Results emphasize that the effect of the cultivar on individual acid contents of the strawberry fruit was higher and clearer than the effect of the irrigation regimes and bio-stimulant applications.

3.2. Health related fruit quality indicators

The antioxidant and phenol contents of fruits and vegetables are well-known to be responsible for decreasing the risks of oxidative stress mediated cardiovascular and neurodegerative diseases as well as cancer (Giampieri et al., 2012). Among the edible foods, berries are the best sources of antioxidant and phenol contents which are 4, 10 and 40 fold higher than the contents of other fruits, vegetables and cereals respectively (Halvorsen et al., 2002). Strawberries have higher

Table 3
Effects of the applied bio-stimulant under different irrigation level of strawberry varieties on individual acids.

	Application	Cultivar	Irrigation Regime				CulXApp	App. Av.
			IR50	IR75	IR100	IR125		
Citric (g/100 g FW)	Control	Rubygem	0.80 ab	0.57 e	0.74 cd	0.60 e	0.68	0.72
		Kabarla	0.78 a-d	0.76 bcd	0.73 d	0.79 abc	0.77	
	App X Irr		*0.79 A*	*0.66 D*	*0.73 B*	*0.70 BCD*		
	Bio stimulant	Rubygem	0.62 e	0.62 e	0.61 e	0.84 a	0.67	0.73
		Kabarla	0.78 a-d	0.82 ab	0.76 bcd	0.78 a-d	0.78	
	App X Irr		*0.70 BCD*	*0.72 BC*	*0.69 CD*	*0.81 A*		
	Irrigation Av.		0.75 A	0.69 B	0.71 B	0.75 A		
LSD_{irr}** = 0.032 $LSD_{appxirr}$*** = 0.045 $LSD_{appxirrxcul}$*** = 0.063								
Malic (g/100 g FW)	Application	Cultivar	Irrigation Regime				CulXApp	App. Av.
	Control	Rubygem	IR50	IR75	IR100	IR125		
			0.18 ab	0.13 e	0.16 cd	0.13 e	0.152 C	0.16
		Kabarla	0.18 ab	0.17 bc	0.17 bc	0.16 cd	0.168 B	
	App X Irr		0.18 A	0.15 CD	0.16 B	0.15 D		
	Bio stimulant	Rubygem	0.15 de	0.13 e	0.13 e	0.17 bc	0.144 C	0.16
		Kabarla	0.19 a	0.19 a	0.17 bc	0.18 abc	0.182 A	
	App X Irr		0.17 AB	0.16 BC	0.15 D	0.17 AB		
	Irrigation Av.		0.18 A	0.16 B	0.16 B	0.16 B		
LSD_{irr}*** = 0.0083 $LSD_{culxapp}$*** = 0.0083 $LSD_{appxirr}$*** = 0.012 $LSD_{appxirrxcul}$*** = 0.017								
L-ascorbic (mg/100 g FW)	Application	Cultivar	Irrigation Regime				CulX App	App. Av.
	Control	Rubygem	IR50	IR75	IR100	IR125		
			46.6 bc	35.5 e	47.4 b	39.6 cde	42.3	43.5
		Kabarla	44.5 bcd	39.9 cde	48.5 b	45.6 bc	44.6	
	App X Irr		45.5 BC	37.7 DE	47.9 B	42.6 CD		
	Bio stimulant	Rubygem	38.2 de	36.6 e	34.3 e	60.5 a	42.3	43.7
		Kabarla	48.5 b	45.4 bcd	39.8 cde	46.5 bc	45.0	
	App X Irr		43.3 BC	41.0 CDE	37.1 E	53.5 A		
	Irrigation Av.		44.4 AB	39.4 C	42.5 BC	48.1 A		
LSD_{irr}*** = 3.73 $LSD_{appx\ irr}$*** = 5.27 $LSD_{appxirrxcul}$*** = 7.46								

(1): Differences between the means were showed with different letters
(2): N. S.: Not Significant, ***: $p < 0.001$; **: $p < 0.01$; *: $p < 0.05$

antioxidant contents varying from 2 to 11 fold than apples, peaches, grapes, tomatoes, oranges, and kiwifruit (Scalzo, Politi, Pellegrini, Mezzetti, & Battino, 2005; Wang, Zheng, & Galletta, 1996). Table 4 reflects the results obtained from our studies concerning the health-related compounds, antioxidant activity, total phenol, quercetin and myricetin contents versus the growth variants (cultivar, irrigation and application) and their interactions. Deficit irrigation (IR50 and IR75) was determined to increase antioxidant activity, whereas there were no quality variations concerning genotypes and application of bio-stimulants. The antioxidant activity varied from 79.7 (IR100) to 82.0 (IR50) μmol TE/g for the different irrigation regimes of our experiment. The bio-stimulant application also decreased the total phenol content of the fruits, whereas conversely increased the quercetin contents. Moreover, the bio-stimulant application did not affect the antioxidant activity of the fruit, but the total phenol content decreased significantly (Table 4). The highly reactive polyphenols play an important role in plants as antioxidants, thus, provide protection against some sources of damage (UV, pathogens, etc.) as stated by Schijlen et al. (2006). Some studies reported an increase in the total phenol contents under the stress conditions of deficit irrigation, and thermal and light stress conditions for various fruit species (Adak, et al., 2018; Yuri et al., 2014). The decrease documented in our study could be explained by the alleviation of the unfavorable cultivation conditions, which encouraged phenol accumulation in the fruit. The significant interaction between the application and the irrigation regimes confirms this hypothesis with the highest accumulation of phenol for the IR75 (216.3 mgGAE/gext) and IR50 (215.5 mgGAE/gext) plots in the control plants while the lowest total phenol contents were obtained for the same irrigation regimes of IR75 (184. 5 mgGAE/gext) and IR50 (185.9 mgGAE/gext) plots with bio-stimulant application. In contrast to the well-known fact of polyphenol contribution to the antioxidant activity no decrease was observed in the antioxidant activity of our study, while the total phenol content decreased with the bio-stimulant application. Other secondary metabolites such as vitamin C, E or the carotenoid contents of the fruit and in turn the antioxidant activity may be improved by bio-stimulant activity. A significant triple interaction was evident in our study for all the factors that could provide different ways for obtaining the highest antioxidant activity and the total phenol content.

The phenolic compounds are categorized into four groups, which include phenolic acids, flavonoids, tannin and stilbenes in the berry fruit (Paredes-Lopez, Cervants-Ceja, Vigna-Perez, & Hernandez-Perez, 2010). The major flavonols are kaemferol and quercetin, whereas myricetin is found rarely in strawberry (Skupień & Oszmiański 2004). The results of our study for quercetin and myricetin are presented in Table 4. The previous researchers (Kosar, et al., 2004; Olsson et al., 2004; Skupień & Oszmiański 2004) indicated that myricetin and quercetin would be influenced by the cultivar, ripening, storage and cultivation environment. There is evidently a lack of research regarding the effect of myricetin and quercetin contents of the fruit as health related compounds with bio-stimulant applications combined with irrigation regimes. Skupień & Oszmiański (2004) found higher quercetin than myricetin in all the cultivars examined, which ranged from 63.2 to 124.0 mg/100 g and 0.74 to 3.88 mg/100 g, respectively. However, our results indicated a higher content of myricetin than quercetin. This may be explained over the use of different genetic stocks of strawberry and cultivation conditions. A significant impact of the cultivars on the quercetin content of the fruits was not observed in our experiment. However, the application significantly promoted the quercetine content of the fruit, whereas the myricetine content of the 'Kabarla' cultivar (10.1 mg/100 g) was found to be higher than that of the 'Rubygem' (8.5 mg/100 g). Although the effect of irrigation regimes on the myricetine and quercetine contents were not consistent, the amount of water significantly influenced these compounds ($p \leq 0.001$). The highest myricetine content was found in the IR75 plot (10.3 mg/100 g),

Table 4
Effects of the Applied Bio-stimulant Under Different Irrigation levels of Strawberry Varieties on Health-Related Compounds.

	Application	Cultivar	Irrigation Regime				Cul XApp	App. Av.
			IR50	IR75	IR100	IR125		
Antioxidant (μmol TE/g FW)	Control	Rubygem	82.0 ab	82.8 a	79.5 bc	82.1 ab	81.6	80.98
		Kabarla	82.4 ab	80.3 ab	76.9 c	81.7	80.4	
	App X Irr		*82.2 A*	*81.6 A*	*78.2 B*	*81.9 A*		
	Bio stimulant	Rubygem	81.6 ab	82.8 a	79.7 abc	82.1 ab	81.5	80.79
		Kabarla	81.9 ab	82.0 ab	82.5 ab	73.7 d	80.0	
	App X Irr		*81.8 A*	*82.4 A*	*81.1 A*	*77.9 B*		
	Irrigation Av.		82.0 A	81.9 A	79.7 B	79.9 B		
LSD_{irr}*** = 1.61 $LSD_{appxirr}$** = 1.61 $LSD_{appxirrxcul}$*** = 3.22								
Total Phenol (mgGAE/gext/100 g FW)	Application	Cultivar	Irrigation Regime				Cul X App	App. Av.
	Control	Rubygem	IR50	IR75	IR100	IR125		
			191.6 bcd	175.5 cde	188.9 bcd	204.3 b	190.1 B	203.0 A
		Kabarla	239.4 a	257.2 a	172.4 de	195.1 bc	216.0 A	
	App X Irr		*215.5 A*	*216.3 A*	*180.6 C*	*199.7 B*		
	Bio stimulant	Rubygem	190.5 bcd	175.5 cde	175.3 cde	164.7 e	176.5 C	171.2 B
		Kabarla	122.1 f	129.9 f	203.8 b	208.3 b	166.0 C	
	App X Irr		*156.3 D*	*152.7 D*	*189.5 BC*	*186.5 BC*		
	Irrigation Av.		185.9	184.5	185.1	193.1		
LSD_{app}*** = 7.66 $LSD_{culxapp}$*** = 10.8 $LSD_{appxirr}$*** = 15.3 $LSD_{appxirrxcul}$*** = 21.7								
Myrcetin (mg/100 g FW)	Application	Cultivar	Irrigation Regime				CulXApp	App. Av.
	Control	Rubygem	IR50	IR75	IR100	IR125		
			10.0 cd	6.79 fg	8.70 de	6.41 g	7.98 C	9.48
		Kabarla	11.34 bc	11.29 bc	7.87 efg	11.10 bc	10.4 A	
	App X Irr		*10.67 AB*	*9.04 CD*	*8.29 DE*	*8.76 D*		
	Bio stimulant	Rubygem	6.37 g	14.98 a	7.70 efg	7.34 efg	9.09 B	9.19
		Kabarla	8.21 ef	8.01 ef	12.42 b	10.83 c	9.87 A	
	App X Irr		*7.29 E*	*11.49 A*	*10.06 BC*	*9.09 CD*		
	Irrigation Av.		8.98 B	10.27 A	9.17 B	8.92 B		
LSD_{irr}*** = 0.76 $LSD_{culxapp}$*** = 0.76 $LSD_{appxirr}$*** = 1.08 $LSD_{appxirrxcul}$*** = 1.53								
Quercetin (mg/100 g FW)	Application	Cultivar	Irrigation Regime				CulXApp	App. Av.
	Control	Rubygem	IR50	IR75	IR100	IR125		
			4.53 b	1.09 f	5.51 a	1.31 ef	3.11 A	2.52 B
		Kabarla	2.13 d	1.96 de	1.91 de	1.72 def	1.93 B	
	App X Irr		*3.33 BC*	*1.52 D*	*3.71 BC*	*1.52 D*		
	Bio stimulant	Rubygem	1.64 def	6.16 a	4.59 b	1.59 def	3.49 A	3.34 A
		Kabarla	5.81 a	3.26 c	1.70 def	2.00 de	3.11 A	
	App X Irr		*3.72 B*	*4.71 A*	*3.14 C*	*1.79 D*		
	Irrigation Av.		3.53 A	3.12 B	3.43 AB	1.65 C		
LSD_{app}*** = 0.284 LSD_{irr}*** = 0.40 $LSD_{appxirr}$*** = 0.59 $LSD_{appxirrxcul}$*** = 0.80								

(1): Differences between the means were showed with different letters.
(2): N. S.: Not Significant, ***: $p < 0.001$; **: $p < 0.01$; *: $p < 0.05$.

whereas the fruit grown in IR50 plot produced the highest quercetine content (3.53 mg/100 g). The interaction between the application and the irrigation was found to be significant for both flavonols. Interestingly, the highest myricetine and quercetine were determined in the IR75 plot applied bio-stimulant, with 11.5 and 4.7 mg/100 g values, respectively. The triple interaction for both flavonols was found to be significant and the highest value was observed for the 'Rubygem' cultivar in the same plot (IR75 with bio-stimulant).

4. Conclusions

This study clearly revealed the differences between the cultivars in terms of taste-related compounds. However, no significant variation was found among the health-related compounds except that in myricetine. The results of this research indicate that the 'Rubygem'cultivar can be more acceptable by the consumer with its higher sugar and lower acid content than the 'Kabarla' cultivar. The bio-stimulant, which is widely used in sustainable agriculture without any negative effect on human health, contributed to taste by improving TSS, fructose, sucrose and health-related compounds and increasing the quercetine content of the fruit. The deficit irrigations (IR50 and IR75) significantly increased TSS and antioxidant activity, whereas the other irrigation applications had inconsistent effects on the taste and health-related compounds. These results are very important for water saving in terms of contributing to sustainable agriculture suffering from intensive production. Current results may have a widespread influence on strawberry production for the Mediterranean region, in which a very big portion (approximately 70%) is acquired of the strawberry production in Turkey.

Conflict of interest

The authors have declared no conflict of interest.

Acknowledgements

This work was financially supported by the Coordination Unit of the Scientific Research Projects of the Çukurova University via the project FDK-2016-5886.

S. Kapur (Ph. D, Aberdeen University, UK), retired faculty of Çukurova and Maryland Universities (European Division) is acknowledged for the language editing of the text.

References

Adak, N., Gubbuk, H., & Tetik, N. (2018). Yield, quality and biochemical properties of strawberry cultivars under water stress. *Journal of the Science of Food and Agriculture,*

98(1), 304–311.
Bayır, A. (2007). *Bazı Şaraplık Üzüm Çeşitlerine Ait Çekirdeklerin Toplam Fenolik Madde İçerikleri ve Antiradikal Aktivitelerinin Belirlenmesi.* Antalya, Türkiye: Yüksek Lisans Tezi, Akdeniz Üniversitesi.
Benzie, I. F. F., & Strain, J. J. (1996). The ferric reducing ability of plasma (FRAP) as a measure of "antioxidant power": The FRAP assay. *Analytical Biochemistry, 239*, 70–76.
Bogunovic, I., Duralija, B., Gadze, J., & Kisic, I. (2015). Biostimulant usage for preserving strawberries to climate damages. *Horticultural Science (Prague), 42*(3), 132–140.
Bozan, B., Tunalıer, Z., Koşar, M., Altıntaş, A., & Başer, K.H.C. (1997). Comparison of ascorbic and citric acid contents in 'Emphasis Type.' In: Proc. 11. Symp Plant Origin Crude Drugs, Ankara, 258 s.
Bulgari, R., Cocetta, G., Trivellini, A., Vernieri, P., & Ferrante, A. (2015). Biostimulants and crop responses: A review. *Biological Agriculture & Horticulture, 31*(1), 1–17.
Capocasa, F., Scalzo, J., Mezzetti, B., & Battino, M. (2008). Combining quality and antioxidant attributes in the strawberry: The role of genotype. *Food Chemistry, 111*, 872–878.
Chandler, C. K., & Ferree, D. C. (1990). Response of 'Raritan' and 'Surecrop' strawberry plants to drought stress. *Fruit Varieties Journal, 44*, 183–185.
Ghaderi, N., Normohammadi, S., & Javadi, T. (2015). Morphophysiological responses of strawberry (Fragariaxananassa) to exogenous salicylic acid application under drought stress. *Journal of Agricultural Science and Technology, 17*, 167–178.
Giampieri, F., Tulipani, S., Alvarez-Suarez, J. M., Quiles, J. L., Mezzetti, B., & Battino, M. (2012). The strawberry: Composition, nutritional quality, and impact on human health. *Nutrition, 28*, 9–19.
Giné-Bordonaba, J., & Terry, L. A. (2010). Manipulating the taste related composition of strawberry fruits (Fragaria x ananassa) from different cultivars using deficit irrigation. *Food Chemistry, 122*, 1020–1026.
Giné-Bordonaba, J., & Terry, L. A. (2016). Effect of deficit irrigation and methyl jasmonate application on the composition of strawberry (Fragaria x ananassa) fruit and leaves. *Scientia Horticulturae, 199*, 63–70.
Halvorsen, B. L., Holte, K., Myhrstad, M. C. W., Barikmo, I., Hvattum, E., Remberg, S. F., ... Blomhoff, R. (2002). A systemic screening of total antioxidants in dietary plants. *Journal of Nutrition, 132*, 461–471.
Kalt, W., Forney, C. F., Martin, A., & Prior, R. L. (1999). Antioxidant capacity, vitamin C, phenolics, and anthocyanins after fresh storage of small fruits. *Journal of Agriculture and Food Chemistry, 47*, 4638–4644.
Kosar, M., Kafkas, E., Paydaş, S., & Baser, K. H. C. (2004). Phenolic composition of strawberry genotypes at different maturation stages. *Journal of Agricultural and Food Chemistry,* 1586–1589.
Krüger, E., Josuttis, M., Nestby, R., Toldam-Andersen, T. B., Carlen, C., & Mezzetti, B. (2012). Influence of growing conditions at different latitudes of Europe on strawberry growth performance, yield and quality. *Journal of Berry Research, 2*, 143–157.
Kunicki, E., Grabowska, A., Sekara, A., & Wojciechowska, R. (2010). The effect of cultivar type, time of cultivation, and biostimulant treatment on the yield of spinach (*Spinacia oleracea* L.). *Folia Horticulturae, 22*, 9–13.
Liu, F., Savić, S., Jensen, C. R., Shahnazari, A., Jacobsen, S. E., Stikić, R., & Andersen, M. N. (2007). Water relations and yield of lysimeter-grown strawberries under limited irrigation. *Scientia Horticulturae, 111*, 128–132.
Manganaris, G. A., Goulas, V., Vicente, A. R., & Terry, L. A. (2014). Berry antioxidants: Small fruits providing large benefits. *Journal of the Science of Food and Agriculture, 94*, 825–833.
Marfá, O., Cáceres, R., Polo, J., & Ródenas, J. (2008). Animal protein hydrolysate as a biostimulant for transplanted strawberry plants subjected to cold stress. *Acta Hortic (ISHS), 842*, 315–318.
Olsson, M. E., Ekvall, J., Gustavsson, K. E., Nilsson, J., Pillai, D., Sjöholm, I., ... Nyman, M. G. L. (2004). Antioxidants, low molecular weight carbohydrates, and total antioxidant capacity in strawberries (Fragaria x ananassa): Effects of cultivar, ripening, and storage. *Journal of Agriculture and Food Chemistry, 52*, 2490–2498.
Ornelas-Paz, J. J., Yahia, E. M., Ramírez-Bustamante, N., Pérez-Martínez, J. D., Escalante-Minakata, M. P., Ibarra-Junquera, V., & Ochoa-Reyes, E. (2013). Physical attributes and chemical composition of organic strawberry fruit (Fragaria x ananassa Duch, Cv. Albion) at six stages of ripening. *Food Chemistry, 138*, 372–381.
Palencia, P., Giné-Bordonaba, J., Martínez, F., & Terry, L. A. (2016). Investigating the effect of different soilless substrates on strawberry productivity and fruit composition. *Scientia Horticulturae, 203*, 12–19.
Paredes-Lopez, O., Cervants-Ceja, M. L., Vigna-Perez, M., & Hernandez-Perez, M. (2010). Berries: Improving human health and healthy aging, and promoting quality life-a review. *Plant Foods for Human Nutrition, 65*, 299–1203.
Russo, R. O., & Berlyn, G. P. (1991). The use of organic biostimulants to help low input sustainable agriculture. *Journal of Sustainable Agriculture, 1*, 19–42.
Saridas, M. A., Paydas Kargı, S., Kapur, B., & Celiktopuz, E. (2017). Effect of the bioactivator application on fruit quality under different irrigation regime in "Kabarla" strawberry variety. *American Journal of Plant Biology, 2*(5), 114–119.
Scalzo, J., Politi, A., Pellegrini, N., Mezzetti, B., & Battino, M. (2005). Plant genotype affects total antioxidant capacity and phenolic contents in fruit. *Nutrition, 21*, 207–213.
Schijlen, E., Ric de Vos, C. H., Jonker, H., van den Broeck, H., Molthoff, J., van Tunen, A., ... Bovy, A. (2006). Pathway engineering for healthy phytochemicals leading to the production of novel flavonoids in tomato fruit. *Plant Biotechnol, 4*, 433–444.
Skupień, K., & Oszmiański, J. (2004). Comparison of six cultivars of strawberry (Fragaria x ananasssa Duch.) grown in northwest Poland. *European Food Research and Technology, 219*, 66–70.
Spanos, G. A., & Wrolstad, R. E. (1990). Influence of processing and storage on the phenolic composition of Thompson seedless grape juice. *Journal of Agriculture and Food Chemistry, 38*, 1565–1571.
Terry, L. A., Chope, G. A., & Giné-Bordonaba, J. (2007). Effect of water deficit irrigation and inoculation with Botrytis cinerea on strawberry (Fragaria x ananassa) fruit quality. *Journal of Agriculture and Food Chemistry, 55*(26), 10812–10819.
Tulipani, S., Marzban, G., Herndl, A., Laimer, M., Mezzetti, B., & Battino, M. (2011). Influence of environmental and genetic factors on health-related compounds in strawberry. *Food Chemistry, 124*(3), 906–913.
UNESCO (2009). *The United Nations World Water Development Report 3-Water in a Changing World.* Earthscan, Paris, France: United Nations Educational Scientific and Cultural Organization429.
Wang, H., Cao, G., & Prior, R. L. (1996). Total antioxidant capacity of fruits. *Journal of Agricultural and Food Chemistry, 44*, 701–705.
Weber, N., Zupanc, V., Jakopic, J., Veberic, R., Mikulic-Petkovsek, M., & Stampar, F. (2017). Influence of deficit irrigation on strawberry (Fragaria x ananassa Duch.) fruit quality. *Journal of the Science of Food and Agriculture, 97*, 849–857.
Winardiantika, V., Lee, Y. H., Park, N. I. I., & Yeoung, Y. R. (2015). Effects of cultivar and harvest time on the contents of antioxidant phytochemicals in strawberry fruits. *Horticulture, Environment and Biotechnology, 56*(6), 732–739..
Yildiz, H., Ercişli, S., Hegedus, A., Akbulut, M., Topdas, E. F., & Aliman, J. (2014). Bioactive content and antioxidant characteristics of wild (*Fragaria vesca* L.) and cultivated strawberry (Fragaria x ananassa Duch.) fruits from Turkey. *Journal of Applied Botany and Food Quality, 87*, 274–278.
Yuan, B. Z., Sun, J., & Nishiyama, S. (2004). Effect of irrigation on strawberry growth and yield inside a plastic greenhouse. *Biosystem Engineering, 87*(2), 237–245.
Yuri, J. A., Neira, A., Maldonado, F., Quilodrán, Á., Simeone, D., Razmilic, I., & Palomo, I. (2014). Total phenol and quercetin content and antioxidant activity in apples in response to thermal, light stress and to organic management. *Journal of Applied Botany and Food Quality, 87*, 131–138.
Zheng, J., Kallio, H., & Yang, B. (2009). Effects of latitude and weather conditions on sugars: Fruit acids and ascorbic acid in currant (Ribes spp.) cultivars. *Journal of the Science of Food and Agriculture, 89*, 2011–2023.