Scientia Horticulturae 199 (2016) 63–70

Contents lists available at ScienceDirect

Scientia Horticulturae

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

Effect of deficit irrigation and methyl jasmonate application on the composition of strawberry (*Fragaria* x *ananassa*) fruit and leaves

Jordi Giné-Bordonaba[1], Leon A. Terry*

Plant Science Laboratory, Cranfield University, Bedfordshire MK43 0AL, UK

ARTICLE INFO

Article history:
Received 5 October 2015
Received in revised form 9 December 2015
Accepted 13 December 2015
Available online 31 December 2015

Keywords:
Anthocyanins
Berry size
Drought
Organic acids
Sugars

ABSTRACT

Drought stress is among the most severe environmental risks threatening strawberry production. In the present study, the effect of deficit irrigation (DI; 50 mL/day) and/or elicitation with methyl jasmonate (MeJA; 0.1 mM) on the composition of secondary fruit and leaves from three strawberry pre-commercial cultivars (253/29, 279/4 and 279/5) was investigated and compared to plants kept at or near field capacity (200 mL/day). For certain cultivars (253/29), DI applied at green stage of fruit development resulted in a considerable reduction in berry size (1.7-fold). In other cultivars (279/4 and 279/5), fruit size was comparable in DI-treated and fully irrigated plants. Changes in the major sugars and organic acids of strawberry leaves and fruit were cultivar and organ dependent and were associated to an osmotic adjustment strategy within the plant to counteract the effects of drought. Overall, elicitation with MeJA had a minimal effect on plant growth and morphological traits. Nevertheless, MeJA increased fructose content of DI-treated leaves and palliated the differences in glucose content of fruit from different water treatments. The most pronounced effect of MeJA was related to an enchance synthesis and accumulation of pelargonidin-3-glucoside (nearly 2-fold) in red-ripe fruit from cultivar 279/5.

1. Introduction

Berries have long been recognized to play an important role in human nutrition, providing health-benefits against a wide range of diseases, mainly due to their elevated content in certain bioactives including ascorbate, anthocyanins, phenolic acids, carotenoids, etc. (Giné Bordonaba and Terry, 2011a; Manganaris et al., 2013). Most bioactive compounds within the plants are secondary metabolites whose synthesis can be triggered in response to biotic and abiotic stresses, such as UV radiation, drought, wounding as well as infections (Terry and Joyce, 2004; Terry et al., 2007a; Jahangir et al., 2009). In the particular case of strawberries, several studies have demonstrated the effect that certain preharvest treatments or cultivation practices have on strawberry fruit biochemistry (Terry et al., 2007a; Keutgen and Pawelzik, 2008; Crespo et al., 2010; Giné Bordonaba and Terry, 2010), including the effect on the concentration of certain taste- and health-related compounds. For instance, earlier works demonstrated that besides the positive environmental effects (i.e. water savings), deficit irrigation (DI) in strawberry plants resulted in berries with higher concentrations of anthocyanins and antioxidant capacity (Terry et al., 2007a) as well as other markers of strawberry fruit quality. Later studies revealed that such effects were cultivar dependent (Giné Bordonaba and Terry, 2010). Nonetheless, DI applied to strawberry plants has been linked with a significant reduction in fruit size and yield (Blatt, 1984; Serrano et al., 1992; Krüger et al., 1999; Liu et al., 2007; Terry et al., 2007a) which also seems to be cultivar dependent (Giné Bordonaba and Terry, 2010).

Jasmonic acid (JA) and its methyl ester methyl jasmonate (MeJA) are naturally occurring plant hormones which have been shown to regulate a wide range of physiological and biological processes (Cheong and Choi, 2003; Rohwer and Erwin, 2008), including responses to drought stress. Given the capacity of MeJA to act as an elicitor and considering that this compound is already classified as Generally Recognise As Safe (GRAS) substance by the U.S. Food and Drug Administration (Wang et al., 2009) it may have a potential for enhancing the synthesis of bioactive compounds (Pérez-Balibrea et al., 2011) and increase fruit quality whilst palliating the negative effects of DI. In this context, Wang (1999) reported that preharvest application of MeJA (0.01–0.1 mM) resulted in changes in plant metabolism that rendered strawberry leaves to better withstand in vitro water stress. Preharvest

* Corresponding author. Fax: +44 1234 758380.
E-mail address: l.a.terry@cranfield.ac.uk (L.A. Terry).
[1] Present address : Institute for Food and Agricultural Research and Technology (IRTA), Parc Científic i Tecnologic Agroalimentari de Lleida, Postharvest Programme, Lleida, Parc de Gardeny 25003, Spain.

http://dx.doi.org/10.1016/j.scienta.2015.12.026
0304-4238/

application of MeJA seems to alter stomatal opening in strawberry (Wang, 1999) and other crops (Horton, 1991) resulting in better transpiration control and hence potentially improving water stress tolerance. In soybean, MeJA has been shown to ameliorate the damaging effects of drought stress by modifying endogenous phytohormones and polyamines (Hassanein et al., 2009). Moreover, both preharvest and postharvest application of MeJA has been associated with greater antioxidant capacity in Chinese red bayberry (Wang et al., 2009) and strawberry (Ayala-Zavala et al., 2005) as well as enhanced anthocyanin synthesis in apples (Rudell and Mattheis, 2008) or other berries (Wang, 2003; Wang et al., 2008).

This study was conducted to further understand the response mechanisms of strawberry plants to water stress conditions and to elucidate whether or not elicitation with MeJA may be a suitable alternative to minimise the negative effects of DI on berry weight of three different pre-commercial strawberry cultivars whilst maintaining/enhancing the taste- and health-related composition of the fruit. Special attention was given to quantifying sugar and organic acids in both fruit and leaves as major respiratory substrates, and anthocyanins and ascorbic acid concentrations in fruit.

2. Materials and methods

2.1. Plant materials and experimental design

Three different maiden year cold-stored strawberry pre-commercial cultivars (*viz.* 253/29, 279/4 and 279/5) were supplied by Redeva (Surrey, UK) and grown in a glasshouse (April–July) in 1 L capacity pots containing commercial standard compost. Cultivars were selected by Redeva breeders to assess their potential adaptation to dry climates and drought. A completely randomised design was adopted considering cultivar, water treatments (50 or 200 mL day^{-1}) and MeJA treatments (none or 0.1 mM) as the principal sources of variation. Prior to commencing water treatments plants were kept at or near field capacity (*ca.* 0.7 m^3 of water per m^3 of soil; conductivity ca. 850 mV) for approximately three weeks following the methodology described in earlier works (Terry et al., 2007a; Giné Bordonaba and Terry, 2010). Water treatments started once the majority of primary fruit from the primary truss were at green I stage of development (prior to the second fruit expansion growth phase; Terry et al., 2004). Then, plants were irrigated daily (*ca.* 09:00 h) with either 50 or 200 ml day^{-1} over an eight-week period. Methyl jasmonate (Sigma, Dorset, UK) treatments at 0.1 mM + 0.05% Tween-20 (Wang, 1999) were applied as a foliar spray to incipient runoff every 72 h. MeJA treatments started when the majority of the primary fruit from the primary truss were at white stage of development. Similarly, control plants were sprayed with a 0.05% Tween-20 solution.

2.2. Soil moisture content and environmental monitoring

Soil moisture content, recorded as the conductivity from the growing media (mV), was measured daily (*ca.* 16.00 h) by time-domain-reflectometry (TDR) using a Thetaprobe (ThetaKit type TK3, Delta-T devices, Cambs., UK). Hourly temperatures within the glasshouse were recorded by means of a Tiny Tag Ultra 2 data logger (Gemini Data Logger, Sussex, UK), shielded from solar radiation. Mean temperature inside the glasshouse through the growing period was 21 °C.

2.3. Fruit, leaf and runner sampling

From each plant, all fruit from the primary truss were harvested at red stage. Four fully expanded leaves of similar size and age per plant were excised towards the end of the trial (30 days after initiation of water treatments and when all experimental fruit had been harvested) and the length and surface area of the leave recorded. On the last day of the experiment, following leaf sampling, the length as well as the total runner density (g) was determined for each plant. After harvest or excision, objective color of fruit and leaves was measured using a Minolta CR-400 colorimeter and a DP-400 data processor (Minolta Co., Ltd., Japan) with an 8 mm light-path aperture, respectively (Terry et al., 2007b). Berry and leave weight was measured and recorded and thereafter immediately snap-frozen in liquid nitrogen and stored briefly at −40 °C before being freeze–dried in an Edwards Modulyo freeze drier (W. Sussex, UK) for 6 and 4 days at 0.015 kPa, respectively. Lyophilized samples were subsequently ground in a pestle and mortar, weighed and returned to the freezer until use. All reagents were purchased from Sigma (Dorset, UK) unless otherwise stated.

2.4. Extraction and quantification of sugars and organic acids

Sugars from both freeze–dried berries and leaves were extracted using 62.5% (v/v) aqueous methanol as described elsewhere (Terry et al., 2007a). Sugar content was determined using an Agilent 1200 series HPLC binary pump system (Agilent, Berks., UK), equipped with an Agilent refractive index detector (RID) G1362A. Strawberry extracts (20 μL) were diluted (1:10), and injected into a Rezex RCM monosaccharide Ca+ (8%) column of 300 mm × 7.8 mm diameter (Phenomenex, CA, USA; Part no. 00H-0130-K0) with a Carbo-Ca^{2+} guard column of 4 mm × 3 mm diameter (Phenomenex,; Part no. AJ0-4493). Column and oven temperature as well as the mobile phase conditions were those reported earlier (Giné Bordonaba and Terry, 2010). Extracts for organic acids determination were prepared as described elsewhere from both berry or leave freeze–dried samples (Giné Bordonaba and Terry, 2010). L-ascorbic, citric, and malic acid contents in extracts were detected at 210 nm using the same HPLC system as described above equipped with an Agilent DAD G1315B/G1365B photodiode array with multiple wavelength detector. The mobile phase (1.0 mL min^{-1}) was analytical grade degassed 0.2% (w/v) metaphosphoric acid in H_2O (Giné Bordonaba and Terry, 2009). The presence and abundance of individual sugars or organic acids were automatically calculated by comparing sample peak area to standards (0.025-2.5 mg mL^{-1}) using ChemStation Rev. B.02.01.

2.5. Antioxidant capacity of strawberry leaves

Antioxidant capacity from strawberry leaves was measured using the FRAP assay as described in earlier works (Terry et al., 2007a; Giné Bordonaba and Terry, 2012) with some modifications. A 50 μL aliquot of diluted sample extract (1:9; v/v) or Fe^{2+} ($FeSO_4 \cdot 7H_20$) standards (0–5.0 mM) was added to 3.6 mL of freshly prepared FRAP working solution (*viz.* 5 mL of 10 mM TPTZ (2,4,6-tripyridyl-2-triazine) in 40 mM HCl + 5 mL of 10 mM $FeCl_3$ in 50 mL of 300 mM acetate buffer). The reaction mixture was incubated at 37 °C for 10 min and absorbance measured spectrophotometrically at 593 nm using a Camspec M501 UV–vis spectrophotometer. Antioxidant capacity was expressed as the concentration of antioxidants having a ferric reducing ability (mmols Fe^{2+} g^{-1} DW).

2.6. Analysis of individual anthocyanins

Individual anthocyanins were extracted using the methodology described in earlier works (Giné Bordonaba and Terry, 2011b) by mixing 150 mg of freeze–dried fruit sample with 3 mL of 70% (v/v) methanol and 0.5% (v/v) HCl in HPLC-grade water. The slurry obtained was held at 35 °C in a water bath with constant shaking for 1.5 h; mixing the samples every 15 min. Finally, the flocculate obtained was filtered through a 0.2 μm Millex-GV syringe driven filter unit (Millipore Corporation, MA) and the clear extract