

# Field non-destructive determination of nectarine quality under deficit irrigation

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## Abstract

**This study investigated the effects of deficit irrigation (DI) at different fruit growth stages and tree orientation on fruit quality of 'September Bright' nectarines (*Prunus persica* L. Batsch). Control trees were fully irrigated throughout the entire fruit development period, whereas deficit irrigation was supplied at fruit growth stage I (cell division, DI-I), II (pit hardening, DI-II), IIIa (cell expansion, DI-IIIa) and IIIb (fruit maturation, DI-IIIb). Non-destructive measurements of fruit maturity and quality were carried out using portable devices based on near-infrared spectrometry and pigment fluorescence on West- and East-oriented trees of an Open Tatura system. Chlorophyll degradation (index of absorbance difference,  $I_{AD}$ ), soluble solid concentration (SSC), dry matter percentage (DM), anthocyanin (fluorescence excitation ratio anthocyanin relative index, FERARI) and flavonol concentrations (FLAV) were measured in the four weeks prior to harvest. Yield, crop load, fruit weight and diameter, flesh firmness, and skin dark red and light red coverage were measured at harvest. DI-I, DI-IIIa and DI-IIIb caused a significant reduction of fruit size and yield.  $I_{AD}$  and FERARI declined progressively towards fruit maturity but were not significantly influenced by irrigation. FLAV remained stable until harvest despite a short-term response to water deficit at the beginning of stage IIIb before returning to its previous value. Fruit SSC and DM were highest in DI IIIb and DI I, respectively, and similar to control in DI II and DI IIIa. Tree orientation did not affect fruit quality, except for higher  $I_{AD}$  and FLAV in West- compared to East-oriented trees. Overall, all the DI-treatments induced a crop performance loss, even though different crop parameters were affected, with DI-IIIa causing the most detrimental effect on fruit quality. The non-destructive devices proved useful in determining fruit quality in situ.**

**Keywords:** chlorophyll, dry matter, fluorescence, fruit, near-infrared, phenolics, sugars, tree orientation.

## INTRODUCTION

In a global warming, desertification and food security context, sustainable irrigation management is paramount to improve productivity and save water in orchard systems. Deficit irrigation (DI) affects tree and fruit physiology based on the phenological stage it is applied. In stone fruit, DI with no or low water supply in the stage of pit hardening (stage II) has been successfully linked to minor or no influence on final fruit size or yield (Li et al., 1989; Scalisi et al., 2019). Since final fruit size is the main focus for most growers, DI effects on other fruit quality parameters are often disregarded or poorly considered. Understanding fruit quality is a complex task, as its definition differs among markets and consumer preferences.

The availability of water at different fruit developmental stages affects plant primary and secondary metabolism and physiology with compound effects on fruit quality characteristics (Naor et al., 1999 and 2001; Pliakoni et al., 2010; Lopez et al., 2011; Thakur and Singh, 2012). When measured before harvest, fruit quality parameters help forecast harvest time and allow growers to act in a timely manner to improve fruit characteristics.

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Harvest time is driven by maturity indices that vary among crops and traditional quality parameters can be destructively assessed at harvest. In peach and nectarine, flesh firmness, ground colour, soluble solid concentration (SSC) and titratable acidity have been widely linked to fruit maturity (Crisosto, 1994; Infante, 2012). However, since their standard determination needs fruit destructive analyses, non-invasive portable spectrometers have been tested to determine fruit quality traits directly in the field. Subedi et al. (2006) used a handheld short-wave near-infrared device equipped with a silicon diode detector and a tungsten halogen light source to determine SSC and dry matter concentration (DM). Ziosi et al., (2008), studied the relationship of the index of absorbance difference ( $I_{AD}$ ) obtained with a DA-meter to chlorophyll-*a* and ethylene production. During maturation the value of  $I_{AD}$  slowly decreases as chlorophyll is degraded. Furthermore, Cerovic et al. (2008) used a non-contact autofluorescence optical device for the estimation of phenolics by fruit fluorescence.

This work aimed to investigate the effects of deficit irrigation at different stages of fruit development and tree orientation on nectarine fruit quality traits using portable field devices prior to harvest.

## **MATERIALS AND METHODS**

### **Experimental design**

The experiment was conducted in 2018 at the research station of Agriculture Victoria, Tatura, Australia (36°26'7.2" S and 145°16'8.4" E, 113 m a.s.l.). Four-year old 'September Bright' nectarine (*Prunus persica* L. Batsch) trees trained to an Open Tatura system (2,222 trees/ha) along North–South orientated rows were subjected to full irrigation (control) and deficit irrigation (DI) at fruit developmental stages I (cell division, DI-I), II (pit hardening, DI-II), IIIa (cell expansion, DI-IIIa) and IIIb (fruit maturation, DI-IIIb). Control trees were irrigated to 100 % of crop evapotranspiration, whereas DI trees were supplied a volume of water ranging from 0 to 40 % of the control trees at each stage. Within a randomised-block designed experiment, three blocks were selected for measurements of crop parameters. The experimental orchard had a clay loam soil texture, trees were fertigated and thinned at the start of fruit developmental stage I and summer-pruned in stage II.

### **Determination of crop parameters**

September Bright is a late-maturing yellow-fleshed/red skin nectarine cultivar that reaches its ripening stage in early March in the Southern Hemisphere. The fruit of this cultivar have a predominantly dark-red cover colour, with occasional light-red areas, and a ground yellow colour is only evident in small portions of the fruit. Fruit quality parameters were non-destructively measured in situ on the West- and East-oriented trees of the Open Tatura training system, on a total of 72 trees, on three fruit per tree. Fruit were measured on the inside of the Open Tatura canopies, hence measured fruit in West-oriented trees received more sunlight in the morning, whereas measured fruit in East-oriented trees received more sunlight in the afternoon. Measurements were carried out at weekly intervals starting at 26 days before harvest (DBH) and ending with harvest.

Soluble solid concentration (SSC) and dry matter percentage (DM) were estimated with a Felix F-750 NIR portable spectrometer (Felix Instruments, Camas, WA, USA) using models based on the association between the second derivative of the absorbance in the 729 – 975 nm spectrum and destructively determined SSC and DM (fruit N = 85).

Chlorophyll degradation was measured using a DA-meter (TR Turoni, Forlì, Italy) on two opposite sides of each fruit and expressed as index of absorbance difference ( $I_{AD}$ ) calculated from absorbance measurements at 670 and 720 nm wavelengths. In 'September

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Bright' nectarine, fruit are considered ready to harvest at  $I_{AD}$  between 1.20 and 0.50 (Horticulture Industry Network, 2019).

Anthocyanin and flavonol concentrations were estimated using a Multiplex® 3 handheld device (Force A, Orsay, France). This device is an improved version of the one used by Cerovic et al. (2008) equipped with Red-Blue-Green (RGB) matrices light sources at 470 nm (blue), 516 nm (green) and 635 nm (red), and three fluorescence detectors for yellow (YF), red (RF) and far-red (FRF) (Baluja et al., 2012). Anthocyanins and flavonols were expressed as fluorescence excitation ratio anthocyanin relative index (FERARI) and flavonol index (FLAV), respectively, calculated as reported by Baluja et al. (2012) and Bahar et al. (2012).

Fruit were harvested on 3 March 2018 and brought to the laboratory to determine yield, crop load, fruit weight, fruit diameter, flesh firmness, and dark and light red coverage using a commercial fruit grader equipped with optical sensors (Compac InVision 9000, Compac Sorting Equipment Ltd, Australia) and a near infra-red (NIR) reflectance spectrometer (Taste Technologies Ltd, New Zealand).

### **Crop performance**

The overall crop performance was assessed for DI fruit in relation to control. A performance index of DI-I, DI-II, DI-IIIa and DI-IIIb fruit was calculated for each measured crop parameter as the ratio of its measured value to the control (i.e. DI crop parameter / control crop parameter). The overall crop performance index (CPI) was estimated by calculating the weighted average of all the performance indices for each parameter, using weights (W) based on the subjective assessment of the importance of each parameter for crop profitability in the regional context of the Goulburn Valley (i.e. ranging from 1, slightly important, to 5, very important). The following W values were used for each parameter: yield [W=5], crop load [W=3], fruit diameter [W=5], flesh firmness [W=3], dark red coverage [W=3], light red coverage [W=1], SSC [W=4], dry matter [W=1],  $I_{AD}$  [W=3], FLAV [W=1] and FERARI [W=1]. Fruit weight was not considered in this analysis as it is highly correlated to fruit diameter. Crop performance loss (CPL) was calculated for each DI treatment as the difference between 1 (i.e. CPI of control fruit) and the CPI of each DI treatment.

### **Statistical analysis**

Data were analysed by analysis of variance (ANOVA) using SYSTAT (Systat software Inc., Chicago, Illinois, USA) procedures. The F-750 Model Builder software (Felix Instruments, v1.3.0.177) was used to conduct a partial least squares (PLS) regression analysis for the SSC and DM models. SigmaPlot procedures (Systat software Inc., Chicago, Illinois, USA) were used for the cross-validation of SSC and DM regression models.

## **RESULTS AND DISCUSSION**

### **Fruit quality and crop parameters**

Deficit irrigation at different fruit developmental stages significantly affected yield, fruit weight and diameter, flesh firmness, and dark red and light red coverage (Table 1). The same parameters were not significantly different in West- and East-oriented trees. DI-IIIb caused over 50% of yield loss compared to control trees. This result was driven by a reduced fruit weight (and diameter), as crop load was not significantly affected by deficit irrigation at any of the stages (Table 1). DI-I and DI-IIIa also led to a decrease of yield, fruit diameter and weight, although not as pronounced as for DI-IIIb. The only DI treatment that did not significantly affect yield, fruit weight and diameter was DI-II, in line with literature on nectarine (Naor et al., 1999; 2001). Flesh firmness was slightly reduced when deficit

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irrigation was applied at stage II and IIIa compared to the control, meaning that these two deficit irrigation strategies might lead to an earlier fruit maturity. In contrast, DI-IIIb induced an increase in flesh firmness that might reflect delayed ripening, as previously observed by Lopez et al. (2011) when no irrigation was applied in stage III in the late peach 'Ryan Sun'. Fruit skin dark red coverage was increased by DI-II, DI-IIIa and DI-IIIb, with the latter inducing the strongest effect (i.e. highest %). Conversely, the light red coverage was not affected from deficit irrigation in stage IIIb, but instead dropped in DI-II and DI-IIIa fruit.

In line with previous studies on nectarine (Pliakoni and Nanos, 2010; Thakur and Singh, 2012), deficit irrigation produced an overall significant effect on SSC and DM, with DI-IIIb trees yielding fruit with the highest SSC and DM at harvest, followed by DI-I (Figure 1A and B). DI-II and DI-IIIa did not significantly affect SSC and DM, despite the latter yielding smaller fruit (Table 1). Therefore, DI-IIIa fruit had a similar water content (i.e. as dry matter is not significantly different) but a likely lower import of non-structural carbohydrates when compared to control fruit. This confirms that fruit size only partly explains the variation in sugars by dilution/concentration, in line with the findings from Lopresti et al. (2015) who found that fruit mass has little effect on sucrose (i.e. the most relevant sugar in nectarine) concentration. In DI-IIIb fruit, SSC and DM started to be significantly higher than control trees at 12 DBH, whereas in DI-I only at 5 DBH (Figure 1A and B), with the former being likely subjected to a sugar concentration effect due to low cell number. Tree orientation did not influence SSC (i.e. SSC in West trees =  $16.2 \pm 0.19$  °Brix cf. East trees =  $16.0 \pm 0.18$  °Brix,  $P = 0.483$ ), but generated a significant effect on DM, with fruit from West-oriented trees having the highest DM, thus the lowest water content at harvest (i.e. DM in West trees =  $18.3 \pm 0.22$  % cf. East trees =  $17.7 \pm 0.24$  %,  $P = 0.003$ ). This may have been due to differences in fruit temperature in response to light exposure in the morning and afternoon.

Table 1. Fruit quality parameters obtained at harvest in 'September Bright' nectarine trees under full irrigation (control) and deficit irrigation at stages I (DI-I), II (DI-II), IIIa (DI-IIIa) and IIIb (DI-IIIb). Different letters indicate significant differences within each column at  $P < 0.05$  (Tukey's test).

Irrigation treatment	Yield (kg/tree)	Crop load (No fruit)	Fruit weight (g)	Fruit diameter (mm)	Flesh firmness (kgf)	Dark red coverage (%)	Light red coverage (%)
Control	19.6 a	183	109 a	57.7 a	5.8 b	56.3 cd	11.2 ab
DI-I	13.8 b	156	88 b	53.2 b	5.9 b	50.0 d	12.1 a
DI-II	17.9 a	162	111 a	58.2 a	5.4 c	64.9 ab	9.7 b
DI-IIIa	13.7 b	148	93 b	54.4 b	5.4 c	62.6 bc	9.7 b
DI-IIIb	9.6 c	156	62 c	46.7 c	6.7 a	70.0 a	12.6 a

Overall,  $I_{AD}$  steadily decreased starting from 19 DBH until harvest (Figure 1C) as chlorophyll slowly degraded in the fruit as per studies on other nectarine cultivars (Reig et al., 2012; Bonora et al., 2013). None of the deficit irrigation treatments showed significant differences compared to control fruit; however, DI-I always maintained the highest  $I_{AD}$ , suggesting a slight delay in maturation compared to other DI treatments. At harvest, DI-IIIb fruit reached similar  $I_{AD}$  levels to DI-I, as the ongoing deficit in stage IIIb eventually slowed down maturation. Delayed maturity was previously observed in peach when no irrigation was supplied in stage III (Lopez et al., 2011). In DI-II and DI-IIIa,  $I_{AD}$  was always the lowest, showing a tendency to advance maturation (Figure 1C). At harvest, fruit from East trees had a significantly lower  $I_{AD}$  than fruit from West trees (i.e.  $I_{AD}$  in West trees =  $0.78 \pm 0.03$  cf. East

trees =  $0.64 \pm 0.03$ ,  $P = 0.002$ ) suggesting that morning or afternoon sunlight exposure might influence maturity, with the latter causing high fruit chlorophyll degradation.

Deficit irrigation showed almost no effect on FLAV prior to harvest, even though a week after the beginning of DI-IIIb (19 DBH) a significant rise of FLAV was detected, to then return to a level near that of other irrigation treatments (Figure 1D). Thus, there seems to be a quick and temporary response of flavonols to water deficit. The overall pattern of FERARI followed a decreasing trend (Figure 1E), meaning that in this red-skin cultivar anthocyanins are slowly degraded and likely transformed into other pigments or phenolics. Also, FERARI did not seem to have a clear significant response to deficit irrigation at different fruit developmental stages, despite DI-IIIb fruit decreasing their anthocyanin levels two weeks after the beginning of stage IIIb (12 DBH) to then rise to the highest level observed among treatments at harvest, which was equivalent to FERARI in DI-I (Figure 1E). Tree orientation significantly affected FLAV (i.e. FLAV in West trees =  $7.96 \pm 0.28$  cf. East trees =  $6.36 \pm 2.40$ ,  $P < 0.001$ ) but not FERARI (i.e. FERARI in West trees =  $0.83 \pm 0.01$  cf. East trees =  $0.80 \pm 0.01$ ,  $P = 0.143$ ) at harvest. Fruit receiving more sunlight in the morning (i.e. in West trees), expressed a higher concentration of flavonols when compared to those hit by more light in the afternoon. This could be explained by the combined effect of light exposure and air temperature, but further studies need to be carried out to confirm these assumptions. The lack of an effect of deficit irrigation on flavonols and anthocyanins at harvest is in line with the findings of Pliakoni et al. (2010) who observed that the two phenolic compounds are not affected by regulated deficit irrigation (i.e. DI at stage II) despite total phenolic concentration being higher than in full-irrigated trees.

### **Crop performance**

Deficit irrigation in different fruit developmental stages induced various effects on the crop performance. None of the DI treatments guaranteed a crop performance as high as the one observed in control fruit, although in some stages, reducing irrigation improved some fruit characteristics (Table 2). CPL was observed in all the DI treatments since the most important fruit traits for fruit marketability were yield and fruit size, both significantly affected by water deficit, except for DI-II (Table 2). Water deficit at stage II induced a loss in other quality indices ( $I_{AD}$ , flesh firmness, FLAV and FERARI) that resulted in an overall CPL of approximately 5%. The highest CPL was observed in DI-IIIa fruit (Table 2) where the only improved quality parameter was dark red coverage, whilst DI-IIIb trees produced the smallest CPL despite small fruit (Table 1) since SSC, DM,  $I_{AD}$ , dark red coverage and flesh firmness were significantly improved at harvest.

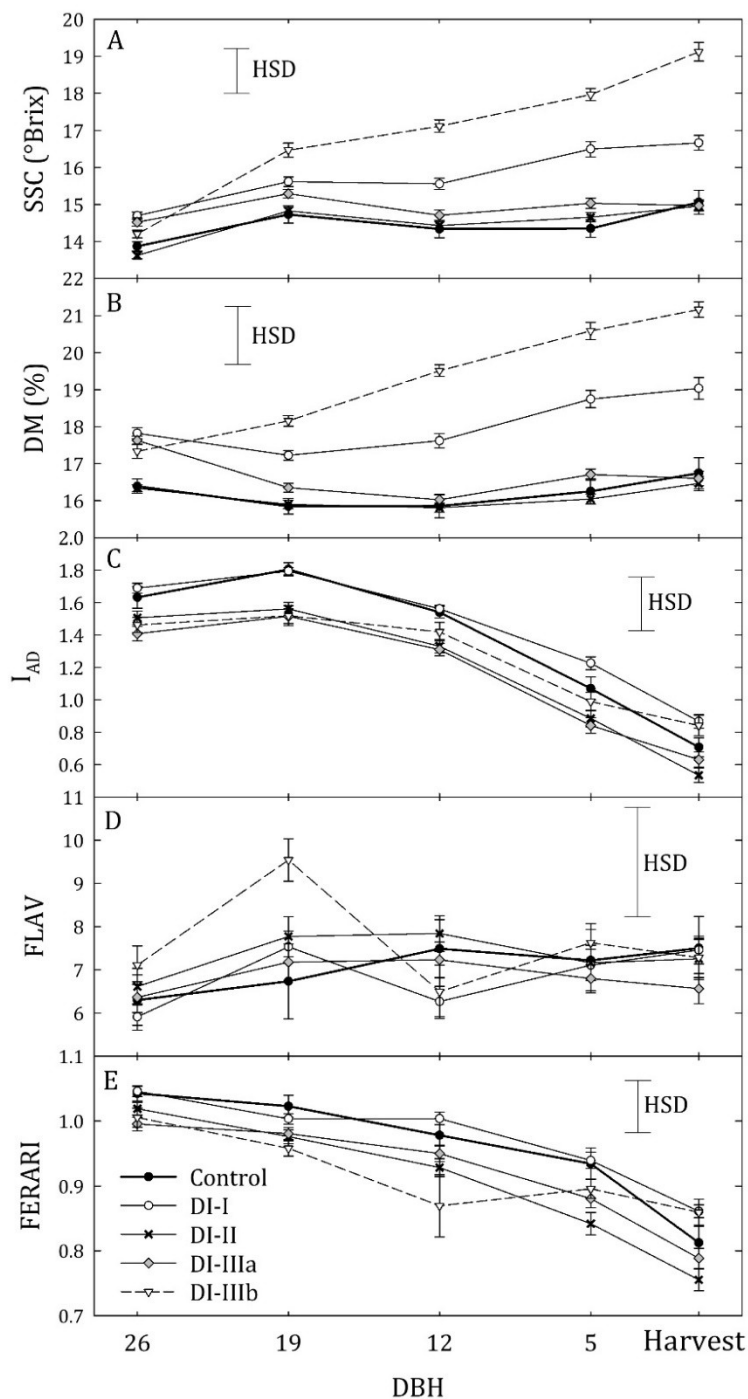


Figure 1. Soluble solid concentration (SSC, A), dry matter percentage (DM, B), index of absorbance difference ( $I_{AD}$ , C), flavonol concentration index (FLAV, D) and fluorescence excitation ratio anthocyanin relative index (FERARI, E) in fruit of 'September Bright' nectarine trees under full irrigation (control) and deficit irrigation at stages I (DI-I), II (DI-II), IIIa (DI-IIIa) and IIIb (DI-IIIb) of fruit development. Data represent average of West- and East-oriented trees and are reported in days before harvest (DBH); HSD bar: Tukey's honestly significant difference at  $P < 0.05$ .

Table 2. Crop performance index (CPI) and loss (CPL) at harvest in 'September Bright' nectarines trees under deficit irrigation at stages I (DI-I), II (DI-II), IIIa (DI-IIIa) and IIIb (DI-IIIb) of fruit growth. Performance indices expressed relative to control fruit.

Crop parameter	DI-I	DI-II	DI-IIIa	DI-IIIb
Yield [W <sup>u</sup> =5]	0.70	0.91	0.70	0.49
Crop load [W=3]	0.85	0.89	0.81	0.85
Fruit diameter [W=5]	0.92	1.01	0.94	0.81
Flesh firmness [W=3]	1.02	0.93	0.93	1.16
Dark red coverage [W=3]	0.89	1.15	1.11	1.24
Light red coverage [W=1]	1.08	0.87	0.87	1.13
SSC <sup>v</sup> [W=4]	1.11	0.99	0.99	1.27
DM <sup>w</sup> [W=1]	1.14	0.98	0.99	1.26
I <sub>AD</sub> <sup>x</sup> [W=3]	1.23	0.75	0.89	1.19
FLAV <sup>y</sup> [W=1]	0.99	0.97	1.01	0.97
FERAR <sup>z</sup> [W=1]	1.06	0.93	0.97	1.06
CPI	0.96	0.95	0.91	0.98
CPL	0.04	0.05	0.09	0.02

<sup>u</sup>Weight factor for each crop parameter on a scale from 1 (slightly important) to 5 (very important); <sup>v</sup>soluble solid concentration; <sup>w</sup>dry matter; <sup>x</sup>index of absorbance difference; <sup>y</sup>flavonol index; <sup>z</sup>fluorescence excitation ratio anthocyanin relative index.

## CONCLUSIONS

This study highlighted fruit quality implications of deficit irrigation conducted at different stages of fruit development prior to harvest. None of the DI treatments caused an overall increase in crop performance, despite individual crop parameters being improved with each irrigation treatment. This study only evaluated a few crop parameters, and omitted others (e.g. juice content, acidity, sugar/acid balance, sensory traits) prior to and at harvest, thus crop performance might change based on the inclusion of other crop indices, and in response to post-harvest and storage conditions. Specific irrigation strategies are not applicable in all circumstances but, on the one hand need to rely on consumers' attitude and produce marketability, whereas on the other hand, they should also consider irrigation water price and availability, and impact on management costs such as pruning. The observed effects of tree orientation on I<sub>AD</sub> and FLAV need to be further investigated in relation to light quality and intensity and fruit temperature at different times of the day and season, as they might have the potential to drive changes in the traditional approaches to tree training and orchard design. In conclusion, the portable devices used in this study were considered accurate enough to be used in cause-effect studies in fruit science.

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