# Combined effects of deficit irrigation and mulching on fruit quality and bioactive compounds of VNR Bihi guava (*Psidium guajava*)

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#### **ABSTRACT**

The present study was carried out during 2019-20 and 2020-21 at Govind Ballabh Pant University of Agriculture and Technology, Pantnagar, Uttarakhand to assess the combined effects of deficit irrigation and mulching on a 5-yearold VNR Bihi guava (Psidium guajava L.) orchard. The experiment consisted of three irrigation regimes, viz. deficit irrigation at 50% ET<sub>c</sub> (DI<sub>50</sub>); deficit irrigation at 75% ET<sub>c</sub> (DI<sub>75</sub>); and full irrigation at 100% ET<sub>c</sub> (FI<sub>100</sub>); and four mulching treatments, viz. silver-black mulch (MSB); black mulch (MB); organic mulch (MOM); and without mulch (M<sub>WM</sub>) laid out in a factorial randomised block design (F-RBD) with three replications. Results showed that fruit number and yield/tree were recorded highest in trees irrigated with  $\mathrm{DI}_{75}$  and mulched with silver-black mulch. Under treatment DI<sub>50</sub> fruit had the highest concentration of soluble solids, with Brix levels of 12.35°B and 12.78 °B during the years 2019-20 and 2020-21, respectively. Total sugars content was significantly increased under  $DI_{50}$  by 7.73 and 7.36%, respectively as compared to FI<sub>100</sub> during both the years of experimentation. The biochemical properties such as total phenolics, total flavonoids and antioxidant (DPPH) activity also increased as irrigation regimes decreased from FI<sub>100</sub> to DI<sub>50</sub>. The total phenolic and flavonoids content were improved by 19.19 and 33.44%, respectively under DI<sub>50</sub> when compared to FI<sub>100</sub>. Significant differences among different mulch treatments with respect to quality parameters were also observed except DPPH activity. Silver-black mulch followed by black mulch resulted in higher soluble solids content, total sugars, ascorbic acid, total phenolic content, total flavonoids and DPPH activity. These findings demonstrate the positive effect of water deficit practice on bioactive compounds and the appropriateness of deficit irrigation and mulching as irrigation management strategies in the fruit orchards that improve fruit commercial value and conserve water.

**Keywords**: Deficit irrigation, DPPH activity, Mulching, Sugars, Total phenolic

Guava (*Psidium guajava* L.) is a significant fruit crop grown in tropical and subtropical areas. It is widely recognised as the "Apple of the Tropics" (Chopda and Barrett 2001) and is referred as the "Poor Man's Apple" since it is consistently accessible and affordable for the typical individual. In India, the cultivation of this crop spans across 2.92 million hectares, resulting in an output of 43.61 million metric tonnes and productivity of 14.93 tonnes/ha (MoA and FW 2022). Guava is renowned for its great nutritional value as it contains ample amount of vitamin C, vitamin A, carbohydrates, and other minerals. Guava contain powerful antioxidant and free radical-scavenging capabilities (Naseer *et al.* 2018, Mishra *et al.* 2022, Bhardwaj and Naruka 2023).

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Deficit irrigation (DI) is an efficient agricultural technique that aims to save water. It involves subjecting crops to mild water stress, which helps boost water productivity and does not reduce crop production significantly. Additionally, DI can improve the quality of horticultural commodities (Edwards and Clingeleffer 2013, Zarrouk et al. 2016, Jat et al. 2022). Deficit irrigation during fruit growth phase improved the quality of fruit by boosting flavour of fruit, which was associated with an increment in soluble solids content (SSC), sugars (Galindo et al. 2018) and bioactive substances such as anthocyanins, phenolics, and antioxidant activity (Thakur and Singh 2013, Martinez-Nicolas et al. 2019) in numerous fruit crops. Mulching is also commonly employed with deficit irrigation as it enhanced the nutrient absorption and assimilation, resulting in a greater proportion of reducing sugars and non-reducing sugars, which ultimately enhanced the quality of fruit (Joshi et al. 2012, Meena et al. 2024).

In the context of guava, research on the influence of deficit irrigation and mulching on fruit quality and bioactive

compounds is limited. Therefore, an experiment was planned to assess the cumulative effects of two deficit irrigation strategies ( $\mathrm{DI}_{50}$  and  $\mathrm{DI}_{75}$ ) along with mulches compared to full irrigation on the fruit quality characteristics and bioactive compounds of VNR Bihi guava.

## MATERIALS AND METHODS

The present study was carried out during 2019–20 and 2020–21 at Govind Ballabh Pant University of Agriculture and Technology, Pantnagar, Uttarakhand. The experiment was performed on 5-year-old VNR Bihi guava trees and a semi-high density planting technique included trees spaced at 5 m × 3 m was used. The region has a sub-humid, sub-tropical climate with an annual precipitation of 1450 mm, of which 70% falls between July–September. The experimental land soil has been classified as a mollisol, and it has a silty loam texture, with neutral pH of 7.1, an EC of 0.36 ds/m, organic carbon (0.67%), low levels of nitrogen (183.76 kg/ha), medium levels of phosphorus and potassium (28.92 and 220.34 kg/ha, respectively).

A factorial randomized block design (F-RBD) with three replications and two variables was applied to conduct the experiment (deficit irrigation and mulching). Drip irrigation was used with 16 mm lateral pipes and four on-line 6 litre/h pressure compensated drip emitters/tree during the fruit-growing season. Three different irrigation treatments, viz. deficit irrigation at 50% ET<sub>c</sub> (DI<sub>50</sub>); deficit irrigation at 75% ET<sub>c</sub> (DI<sub>75</sub>); and full irrigation at 100% ET<sub>c</sub> (FI<sub>100</sub>) were used. Silver-black mulch (M<sub>SB</sub>); black mulch (M<sub>B</sub>); organic mulch (M<sub>OM</sub>); and without mulch (M<sub>WM</sub>) were used as four mulching treatments. Mulches made of 100  $\mu$  thickness by 1.2 m wide polyethylene in the colours silverblack and black were utilised. The following calculation, which suggests utilizing 100% class-A pan evaporation rate for irrigation scheduling, was used:

$$ET_c = K_p \times K_c \times E_p$$

where  $\rm ET_c$ , Evapotranspiration by crop (mm/day);  $\rm K_p$ , Pan coefficient (0.7);  $\rm K_c$ , Crop coefficient (0.8);  $\rm E_p$ , Open pan evaporation (mm)

The relationship provided in INCID was used to estimate the amount of water supplied by a drip irrigation method (Rao 1994). The amount of precipitation and irrigation water used by each plant under various irrigation level during the fruit-growing phase is shown in Table 1.

The fruit number/plant was manually counted. The average fruit weight (g) was multiplied by the fruit number per tree to determine the yield/tree. The average yield/tree was calculated and expressed as kg/tree.

Data were collected for various parameters by using standard protocols. The Erma Hand Refractometer was used to measure the soluble solids content (SSC) in fruit juice, with the reading indicated as °brix. The concentration of ascorbic acid in fruit samples was determined using a 2,6 dichlorophenol-indophenol (DCPIP) titration method (Ranganna 1997). The fruits total sugar content, as well as their reducing and non-reducing sugar content were

Table 1 Precipitation and irrigation water applied per plant during fruit growth period under different irrigation regimes

Treatment	E <sub>pan</sub> (mm)	Rainfall (mm)	Irrigation water applied (mm)					
2019–20								
$DI_{50}$	455.2	541	401.47					
DI <sub>75</sub>	455.2	541	602.21					
$FI_{100}$	455.2	541	802.94					
2020–21								
$DI_{50}$	552.9	82	487.66					
DI <sub>75</sub>	552.9	82	731.48					
FI <sub>100</sub>	552.9	82	975.31					

 $\mathrm{DI}_{50}$ , Deficit irrigation at 50%  $\mathrm{ET_c}$ ;  $\mathrm{DI}_{75}$ , Deficit irrigation at 75%  $\mathrm{ET_c}$ ;  $\mathrm{FI}_{100}$ , Full irrigation at 100%  $\mathrm{ET_c}$ .

analysed using the Lane and Eynon method (Ranganna 1997).

The estimation of total phenolics in the fresh fruit sample was conducted using the Folin-Ciocalteau process as described by Singelton (1999). The determination of the total flavonoid concentration was conducted using the methodology outlined by Zhishen *et al.* (1999). The DPPH radical scavenging activity of the guava was evaluated using the protocol recommended by Huang *et al.* (2016). The DPPH activity of the fruit sample was determined as:

DPPH (%) = 
$$\frac{1 - \text{Absorbance of sample}}{\text{Absorbance of control}} \times 100$$

Statistical analysis: The data obtained from the two factorial randomized block design were subjected to statistical analysis using doebioresearch package (Popat and Banakara 2020). For the significant component of ANOVA, the means of different levels of factors and their interactions were compared using a Duncan's Test.

## RESULTS AND DISCUSSION

*Yield attributes*: The fruit number/tree increased as the irrigation level was elevated from  $\mathrm{DI}_{50}$ – $\mathrm{DI}_{75}$  and slightly decreased in  $\mathrm{FI}_{100}$  over  $\mathrm{DI}_{75}$  during both the experimental seasons (Table 2). In 2019–20, the highest reported number of fruits (88.00) was found in  $\mathrm{DI}_{75}$  whereas the lowest (73.50) was noted in  $\mathrm{DI}_{50}$ . Similar pattern was also noted for 2020–21, with the greatest (77.50) in  $\mathrm{DI}_{75}$  and the lowest (64.75) in  $\mathrm{DI}_{50}$ . Furthermore, the fruit number harvested/tree increased in 2019 compared to 2020, owing to improved flowering, greater average rainfall and minimal fruit cracking.

In 2019–20, trees irrigated at DI<sub>75</sub> produced the highest fruit yield of 44.51 kg/tree and lowest (32.18 kg/tree) in DI<sub>50</sub>. In 2020–21, similar pattern was observed with the highest (46.64 kg/tree) fruit yield reported in DI<sub>75</sub>. However, minimum yield (34.08 kg/tree) was obtained in DI<sub>50</sub>.

The maximum fruit yield seen under  $\mathrm{DI}_{75}$  may be attributed to water deficit in the root zone, which inhibited plant vegetative growth while having minimal impact on

Table 2 Effect of deficit irrigation and mulching on yield attributes of VNR Bihi guava

Treatment		No. of fruits/tree			Yield (kg/tree)	
	2019–20	2020–21	Pooled	2019–20	2020–21	Pooled
Irrigation levels						
$DI_{50}$	73.50	64.75	69.13	32.18	34.08	33.13
DI <sub>75</sub>	88.00	77.50	82.75	44.51	46.64	45.57
FI <sub>100</sub>	85.75	75.00	80.38	42.93	44.69	43.81
SEm±	1.209	0.849	0.707	0.474	0.463	0.230
CD (P=0.05)	3.545	2.489	2.074	1.390	1.358	0.674
Mulching						
Silver-black	85.33	74.67	80.00	42.55	44.57	43.56
Black	83.33	73.33	78.33	40.75	42.44	41.59
Organic	81.33	71.67	76.50	38.94	40.91	39.92
Without mulch	79.67	70.00	74.83	37.26	39.29	38.27
SEm±	1.396	0.980	0.817	0.547	0.535	0.265
CD (P=0.05)	4.094	2.874	2.395	1.605	1.568	0.778
Interaction (A × B)	NS	NS	NS	2.780	2.715	1.348

DI<sub>50</sub>, Deficit irrigation at 50% ET<sub>c</sub>; DI<sub>75</sub>, Deficit irrigation at 75% ET<sub>c</sub>; FI<sub>100</sub>, Full irrigation at 100% ET<sub>c</sub>. NS, Non-significant.

leaf photosynthetic rate. As a result, the plants directed a larger amount of photosynthates into the process of reproductive growth instead in the direction of vegetative growth (Panigrahi *et al.* 2012). Guava is a resilient tree that can be grown in regions with low rainfall and limited water availability. Consequently, it is possible that this is the reason why trees irrigated at a mild water deficit performed better than those irrigated at full irrigation. Panigrahi *et al.* (2012) in Nagpur mandarin, Singh *et al.* (2015) in guava and Soliman *et al.* (2018) in apple reported similar findings.

The results indicated that plants treated with silver-black mulch (M<sub>SB</sub>) exhibited a significantly greater fruit number/ tree and the maximum fruit yield per tree followed by black mulch and the lowest fruit number/tree and fruit yield were observed in plants without mulch (M<sub>WM</sub>) throughout the two years of experiment (Table 2). The positive influence on yield characteristics resulting from different mulches can be linked to the modification of the microclimate in favour of the guava tree. This involves the regulation of temperature, the maintenance of ideal soil moisture levels, and the reduction of weed competition, while simultaneously enhancing nutrient and moisture availability for the fruit tree (Singh *et al.* 2020).

Soluble solids content (°B): The concentration of soluble solids (SSC) decreased when the irrigation level elevated from  $\mathrm{DI}_{50}$ – $\mathrm{FI}_{100}$  during both the experimental seasons (Fig. 1A). In 2019–20, fruits harvested from trees irrigated with  $\mathrm{DI}_{50}$  had an SSC of 12.35 °B, followed by 12.00 °B in  $\mathrm{DI}_{75}$  and the lowest (11.25 °B) in  $\mathrm{FI}_{100}$ . In addition, a minor increase in SSC was observed during 2020–21 when guava trees were irrigated at  $\mathrm{DI}_{50}$  and the SSC value was recorded 12.78 °B, but the lowest value was obtained in guava trees irrigated at  $\mathrm{FI}_{100}$  (11.48 °B). The rise in concentration of soluble solids in water deficit

treatments may be attributed to the accelerated breakdown of starch and subsequent rise in sugar accumulation in fruits caused by reduced irrigation due to water constraint (Ebel *et al.* 1993). These result aligns with the studies reported by Panigrahi *et al.* (2014) in Kinnow and Selahvarzi *et al.* (2017) in pomegranate.

The guava plants that were mulched with  $\rm M_{SB}$  mulch exhibited a greater SSC of 12.17 °B, while those mulched with  $\rm M_{B}$  mulch had an SSC of 12.00 °B, in contrast to the fruits derived from  $\rm M_{WM}$  which had an SSC of 11.53 °B. During the period of 2020–21, the highest SSC of 12.50 °B was seen in plants treated with  $\rm M_{SB}$  mulch followed by an SSC of 12.30 °B in plants treated with  $\rm M_{B}$  mulch. Both of these treatments resulted in significantly higher SSC values compared to the fruits produced from plants that were not mulched, which had an SSC value of 11.83 °B. Mulching guarantees optimal soil moisture by minimizing evaporation from the soil, which ultimately results in an increase in the soluble solids in guava fruits (Sagar *et al.* 2019).

Ascorbic acid (mg/g): Ascorbic acid content was significantly increased under  $\mathrm{DI}_{50}$  by 24.44% and  $\mathrm{DI}_{75}$  by 13.33% as compared to  $\mathrm{FI}_{100}$  during 2019–20 (Fig. 1B). Similarly, in 2020–21, ascorbic acid increased under  $\mathrm{DI}_{50}$  and  $\mathrm{DI}_{75}$  by 23.65 and 12.90% respectively, when compared with  $\mathrm{FI}_{100}$ . Abiotic stress conditions may have contributed to the rise in concentration of ascorbic acid in the custard apple fruit obtained from the trees that were experiencing a water deficit (Kowitcharoen *et al.* 2018). Although hydrogen peroxide is recognized as a form of stress signaling, stress situations frequently activate ABA production, which can enhance hydrogen peroxide formation during times of stress. This may result in the antioxidant system in plants maintaining or increasing the concentration of ascorbic acid (Kowitcharoen *et al.* 2018). The ascorbic acid content was

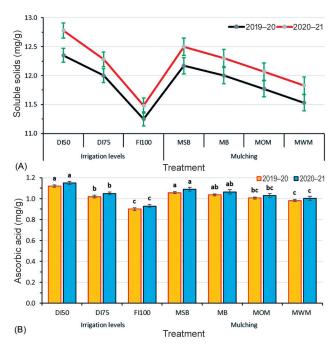


Fig. 1 Effect of deficit irrigation and mulching on (A) soluble solids content (SSC) and; (B) ascorbic acid content. DI<sub>50</sub>, Deficit irrigation at 50% ET<sub>c</sub>; DI<sub>75</sub>, Deficit irrigation at 75% ET<sub>c</sub>; FI<sub>100</sub>, Full irrigation at 100% ET<sub>c</sub>; M<sub>SB</sub>, Silverblack mulch; M<sub>B</sub>, Black mulch; M<sub>OM</sub>, Organic mulch; M<sub>WM</sub>, Without mulch. As per Duncan's multiple range test, different letters (A–D) represent substantially different values at P=0.05. Each data point's vertical bar represents the standard error of the mean.

affected by several types of mulches. Plants treated with  $\rm M_{SB}$  had significantly greater ascorbic acid content, followed by  $\rm M_B$  and the lowest value was seen in plants mulched with  $\rm M_{WM}$  throughout the study period. Ascorbic acid content in guava fruit samples increased under  $\rm M_{SB}$  when compared to  $\rm M_{WM}$  (by 8.16% and 9.00% during 2019–20 and 2020–21, respectively).

Total sugars (%): The total sugar concentration of guava fruits when subjected to various irrigation treatments exhibited a similar pattern to that observed in the soluble solids content during both the years (Table 3). Total sugars content was significantly increased under DI<sub>50</sub> by 7.73% and DI<sub>75</sub> by 4.31% when compared to FI<sub>100</sub> during 2019–20. During 2020-21, total sugars content was significantly increased under DI<sub>50</sub> and DI<sub>75</sub> by 7.36% and 4.72% respectively, as compared to FI<sub>100</sub>. The increased sucrose hydrolysis into glucose and fructose by the invertase enzyme may be the cause of the rise in total sugar content with severe deficit irrigation (Thakur and Singh 2012). Sugar plays the role of a compatible solute that builds up in fruit cells and aids in osmotic adjustment, reducing the loss of turgor in fruit tissues (Navarro et al. 2010). Similar findings of higher total sugars with respect to deficit irrigation were also reported by Thakur and Singh (2013) in nectarines.

The total sugars content of guava fruits improved under tree mulched with  $\rm M_{SB}$  (3.87%) and  $\rm M_{B}$  (2.50%) as compared to un-mulched trees during 2019–20. Similarly, in 2020–21, total sugars content was increased by 3.79% in  $\rm M_{SB}$  and 2.63% in  $\rm M_{B}$  when compared to un-mulched trees. Moreover, silver-black, black and organic mulches were statistically equivalent with each other during the two experimental years. The higher total sugars content of guava fruit in silver-black mulch might be attributed to greater assimilation of carbohydrates favoured under optimum soil hydrothermal regime coupled with higher nutrient absorption and suppressed growth of weeds (Sahu and Sahu 2020). Similar results were also reported by Singh et al. (2015) in guava.

Reducing sugars (%): The different level of deficit irrigation exhibited significant effect on reducing sugars content for two consecutive years in VNR Bihi guava (Table 3). The reducing sugars was significantly higher in  $\mathrm{DI}_{50}$  and  $\mathrm{DI}_{75}$  at 8.41% and 5.62% respectively, than in  $\mathrm{FI}_{100}$  treatment during 2019–20. Likewise, in 2020–21, reducing sugars was increased by 8.78% in  $\mathrm{DI}_{50}$  and 5.71% in  $\mathrm{DI}_{75}$ 

Table 3 Effect of deficit irrigation and mulching on the total sugar, reducing sugar and non-reducing sugar content of VNR Bihi guava

Treatment	Total sugars (%)		Reducing sugars (%)		Non reducing sugars (%)				
	2019–20	2020–21	Pooled	2019–20	2020–21	Pooled	2019–20	2020–21	Pooled
Irrigation levels									
$\mathrm{DI}_{50}$	8.66a	9.37 <sup>a</sup>	9.01 <sup>a</sup>	5.82a	6.15 <sup>a</sup>	5.98 <sup>a</sup>	$2.84^{a}$	3.21a	3.02a
$\mathrm{DI}_{75}$	8.39 <sup>b</sup>	9.11 <sup>b</sup>	8.75 <sup>b</sup>	5.62 <sup>b</sup>	5.95 <sup>b</sup>	5.78 <sup>b</sup>	2.76 <sup>b</sup>	$3.14^{b}$	2.95 <sup>b</sup>
FI <sub>100</sub>	7.99 <sup>c</sup>	8.68c	8.33c	5.33 <sup>c</sup>	5.61 <sup>c</sup>	5.47 <sup>c</sup>	2.65 <sup>c</sup>	3.04 <sup>c</sup>	2.85 <sup>c</sup>
Mulching									
Silver-black	8.51a	9.22a	8.86 <sup>a</sup>	5.71a	6.04 <sup>a</sup>	5.87 <sup>a</sup>	2.81a	3.18 <sup>a</sup>	2.99a
Black	8.39 <sup>ab</sup>	9.11 <sup>ab</sup>	8.75 <sup>ab</sup>	5.62 <sup>ab</sup>	5.95 <sup>ab</sup>	5.78 <sup>ab</sup>	2.76 <sup>ab</sup>	3.15 <sup>ab</sup>	2.96 <sup>ab</sup>
Organic	8.31 <sup>ab</sup>	8.99 <sup>ab</sup>	8.65bc	5.56 <sup>ab</sup>	5.86 <sup>ab</sup>	5.71 <sup>bc</sup>	$2.74^{ab}$	3.12 <sup>ab</sup>	2.93bc
Without mulch	8.18 <sup>b</sup>	8.87 <sup>b</sup>	8.53°	5.47 <sup>b</sup>	5.77 <sup>b</sup>	5.62 <sup>c</sup>	$2.70^{b}$	3.09 <sup>b</sup>	2.89 <sup>c</sup>
Interaction (A × B)	NS	NS	NS	NS	NS	NS	NS	NS	NS

 $DI_{50}$ , Deficit irrigation at 50%  $ET_c$ ;  $DI_{75}$ , Deficit irrigation at 75%  $ET_c$ ;  $FI_{100}$ , Full irrigation at 100%  $ET_c$ . Values marked by a different letter differ significantly according to Duncan's multiple range test ( $P \le 0.05$ ).

as compared to  ${\rm FI}_{100}$ . The increase in concentration of reducing sugars during osmotic adjustment, resulting from the hydrolysis of non-reducing sugars, is likely the reason for the greater proportion of reducing sugars observed under  ${\rm DI}_{50}$  (Navarro *et al.* 2010). The reducing sugars content of guava fruits improved under plants mulched with  ${\rm M}_{\rm SB}$  (4.20%) and  ${\rm M}_{\rm B}$  (2.66%) as compared to un-mulched plants during 2019–20. Similarly, in 2020–21, reducing sugars content was increased by 4.47% in  ${\rm M}_{\rm SB}$  and 3.02% in  ${\rm M}_{\rm B}$  when compared to un-mulched plants. However,  ${\rm M}_{\rm SB}$ ,  ${\rm M}_{\rm B}$  and  ${\rm M}_{\rm OM}$  were statistically equivalent with each other with respect to reducing sugars content during both the years of study.

Non-reducing sugars (%): The guava trees irrigated at different deficit irrigation regime influenced non reducing sugar content significantly in both the years (Table 3). During 2019–20, fruits harvested from  $\mathrm{DI}_{50}$  level had the maximum (2.84%) non reducing sugar contents followed by  $\mathrm{DI}_{75}$  (2.76%) and minimum (2.65%) in  $\mathrm{FI}_{100}$ . In 2020–21, the highest non-reducing sugar (3.21%) was also achieved in  $\mathrm{DI}_{50}$  and minimum (3.04%) in  $\mathrm{FI}_{100}$ . The impact of various types of mulches on non-reducing sugars was statistically significant during both the years. Pooled data revealed that fruits harvested from the trees mulched with  $\mathrm{M}_{\mathrm{SB}}$  (silverblack) showed maximum (2.99%) non reducing sugars followed by 2.96% in  $\mathrm{M}_{\mathrm{B}}$  (black).

Total phenolics (mg/100 g): The total phenolic content of guava fruit declined as irrigation frequency rise from  $\mathrm{DI}_{50}$  to  $\mathrm{FI}_{100}$  during 2019–20 and 2020–21. However, total phenolics content in guava fruits reported higher in second year of experimentation (Supplementary Fig. 1A). In the first year, the total phenolics content was improved by 19.73% under  $\mathrm{DI}_{50}$  and 9.40% under  $\mathrm{DI}_{75}$  when compared to  $\mathrm{FI}_{100}$ . Similarly, during second year of experimentation, the total phenolics content was increased by 18.69% and 8.51% under DI<sub>50</sub> and DI<sub>75</sub>, respectively. The results align with the findings of Thakur and Singh (2013) in nectarines and Kowitcharoen et al. (2018) in sugar apple. The total phenolics content was improved under  $\rm M_{SB}$  and  $\rm M_{B}$  mulch (8.32% and 6.44% during 2019–20) (7.48% and 5.63% during 2020–21) when compared to un-mulched guava plants. Moreover, silver-black and black mulch were statistically equivalent with each other with respect to total phenolics during both the years. Morra et al. (2021) also detected a significant improvement in fruit quality parameters including total phenolics of tomato under biodegradable mulching.

Total flavonoids (mg/100 g): Pooled data indicates that the total flavonoids were increased under  $\mathrm{DI}_{50}$  and  $\mathrm{DI}_{75}$  (33.44% and 18.67%) when compared to  $\mathrm{FI}_{100}$  (Supplementary Fig. 1B).  $\mathrm{DI}_{50}$  treatment had substantially maximum total flavonoids (22.95 mg/100 g in the year 2019–20 and 23.39 mg/100 g in the year 2020–21) and minimum (15.32 mg/100 g in the year 2019–20 and 15.52 mg/100 g in the year 2020–21) were recorded in  $\mathrm{FI}_{100}$ . Water stress activates the biosynthesis pathway of flavonoids through the upregulation of several regulatory enzymes which involved in flavonoids biosynthesis (Castellarin *et al.* 2007). Navarro

et al. (2010) found similar outcomes, demonstrating that grapefruit plants accumulated a greater quantity of flavonoids when subjected to water stress conditions. During the year 2019–20, significant difference among different mulch treatments with respect to total flavonoids was also observed; being maximum (20.15 mg/100 g) in  $\rm M_{SB}$  (silver-black) and minimum (17.61 mg/100 g) in plant mulched with  $\rm M_{WM}$ . In 2020–21, significantly maximum total flavonoids (20.51 mg/100 g) were recorded in  $\rm M_{SB}$  (silver-black) and minimum (17.87 mg/100 g) in plant mulched with  $\rm M_{WM}$ . However,  $\rm M_{SB}$  (silver-black) and  $\rm M_{B}$  (black) mulch were statistically equivalent with each other during both the experimental years.

DPPH radical scavenging activity (%): Pooled data analysis showed that the highest antioxidant activity expressed in terms of 2,2-diphenyl-1-picrylhydrazyl (98.47%) was recorded in DI<sub>50</sub> followed by 96.25% in DI<sub>75</sub> and minimum (90.64%) in FI<sub>100</sub> (Supplementary Fig. 1C). However, the effect of different mulch types on antioxidant activity showed non-significant effect. Antioxidant activity, which is correlated with the presence of bioactive compounds, is indicated by radical scavenging activity. Samieiani and Ansari (2014) discovered that water stress conditions enhanced the DPPH activity in various groundcover plants. It is widely acknowledged that ascorbic acid functions as like antioxidant (Noctor and Foyer 1998). In the present study, the ascorbic acid concentration and the DPPH activity of guava fruits from water-stressed trees were

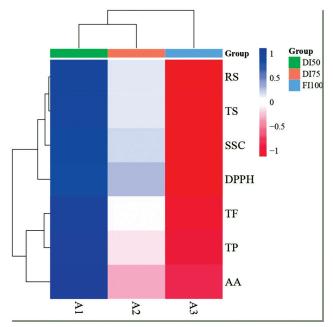


Fig. 2 The heat map depicts the fruit quality traits of VNR Bihi grown under DI<sub>50</sub>, DI<sub>75</sub> and FI<sub>100</sub> irrigation regimes respectively.

RS, Reducing sugars; TS, Total sugars; SSC, Soluble solic content; DPPH, 2,2-diphenyl-1-picrylhydrazyl (antioxident activity); TF, Total flavonoids; TP, Total phenolics; AA, Ascorbic acid.

 $D_{I50}$ , Deficit irrigation at 50%  $ET_c$ ;  $DI_{75}$ , Deficit irrigation at 75%  $ET_c$ ; F1100, Full irrigation at 100%  $ET_c$ .

found to increase. Similarly, Selahvarzi *et al.* (2017) and Kowitcharoen *et al.* (2018) also noted a rise in antioxidant activity in pomegranate and sugar apple, respectively, when subjected to water stress conditions.

Heat map representation of VNR Bihi fruit quality traits: To fully comprehend the relationship among fruit quality parameters and deficit irrigation, a Pearson's correlation heatmap along with clustering analysis was performed (Fig. 2). This test showed that full irrigation (FI) and the majority of the fruit quality parameters were negatively correlated. The heat map results showed that  $\mathrm{DI}_{50}$  treatment was highly positive correlated with ascorbic acid, total phenolics, total flavonoids and sugars respectively, while FI treatment were highly negatively correlated with sugars, soluble solids and antioxidant activity. Clustering analysis revealed that there are mainly 2 mega clusters in which first mega cluster contains 2 sub clusters and second mega cluster contains 3 sub clusters. Cluster analysis also showed that there was a little difference between DI<sub>50</sub> and DI<sub>75</sub> treatments with respect to most of fruit quality parameters.

Our findings confirm the necessity and significance of deficit irrigation strategies as a technique for the effective water management of the VNR Bihi guava orchard in subtropical locations where water shortage is rising as a result of climate change. This research indicates that DI<sub>75</sub> along with silver-black mulch is an effective method for water conservation, improving yield and fruit quality, and increasing the economic profits of farmers. This technique would allow for some degree of water stress during fruit growth period in guava trees.

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