



Effect of Salicylic and Ascorbic Acids Foliar Application on Picual Olive Trees Growth under Water Stress Condition

Ahmed A. El Refaey¹, Yahya I. Mohamed², Samy M. El-Shazly³ and Asmaa A. Abd El Salam²

¹ Soil & Water Science Department, Faculty of Desert and Environmental Agriculture, Fuka, Matrouh University, Egypt;

² Pomology Department, Faculty of Desert and Environmental Agriculture, Fuka, Matrouh University, Egypt

³ Pomology Department, Faculty of Agriculture, El-Shatby, Alexandria University, Egypt.

Abstract

The present study was conducted during the 2018 and 2019 seasons on 5 years of Picual olive trees (*Olea europaea L.*), grown in calcareous soil at the farm of the Faculty of Desert and Environmental Agriculture, Fuka, Matrouh governorate, Egypt. The main objective was to investigate the effect of foliar application with salicylic acid (SA) (100, and 150mg/L), and ascorbic acid (AA) (50, and 100mg/L) under water stress conditions (rain-fed, 50 and 100 % of crop evapotranspiration (ET_c) on growth, physiological characteristics, yield, and fruit quality. The results showed that the highest value of leaf area, shoot length, and yield (kg/tree) were recorded with the treatment salicylic acid (150 mg /L) in both seasons compared with the control. The ascorbic acid at 100 mg/L recorded the highest leaf chlorophyll content and maximum fruit quality compared with the control. Also, sprayed with salicylic acid 100 mg/L and Ascorbic acid at 100 mg/L gave the highest response for leaf content of N, P, K, Ca and Mg (%) compared to other treatments. In addition, 150 mg/L salicylic acid treatment showed the highest value for relative water content associated with the lowest value of leaf proline content compared with control. There were no significant differences between 100% ET_c, and 50% ET_c irrigation treatment in the presence of salicylic acid 150 mg L or ascorbic acid 100 mg L for enhancing most of the studied characteristics. Finally, spraying Picual olive trees with salicylic and ascorbic acids had a positive impact on trees growth, yield, and fruit quality under stress conditions compared to rain-fed treatment in both seasons.

Keywords :Salicylic acid; Ascorbic acid; Proline; Olive trees; Picual; Drought stress

Introduction

Olive (*Olea europaea L.*) is considered as the main and best adapted fruit species in many arid and semi-arid regions around the Mediterranean (Villalobos et al., 2000 and Moriana et al., 2002). Oleaceae family consisted of 30 genera of deciduous trees and shrubs such as olive tree and its relatives comprising about 600 species (Atta-ur-Rahman, 1990). The olive tree is very popular in the Mediterranean area, moreover is the major agricultural crop in the Mediterranean Coastal Basin, and it accounts for around 95% of the world's olive cultivated area (FAO, STAT, 2022).

Egypt has been a world leader in growing olives, according to the ministry of agriculture; the Egyptian olive production was about 932,927 tons produced from 100,826 ha in the year 2020 (FAO, STAT, 2022).

The olive tree can tolerate different types of stresses, i.e. drought, and salinity. Moreover, olive tree is cultivated, on the North-Western coast of Egypt, on very little rainfall (220 mm per year) and grown on poor nutrients calcareous soils (Marsal et al., 2002). But, Water stress, especially at the early growing season, could reduce the yield because of its influence on flowering and fruit sets (Moriana et al., 2003; Farooq et al., 2009). Recently, exogenous use of various growth regulators such as salicylic acid (SA) and ascorbic acid (AA) is widely used for enhancing growth versus abiotic stresses.

Salicylic acid plays an essential role in plant metabolisms such as photosynthesis, antioxidant defense system, nitrogen, proline metabolism; and plant-water relation especially under abiotic stresses (Khan et al., 2015). The exogenous application of salicylic acid (SA) could be an adequate short-term

*Corresponding author e-mail: ahmedelrefaey@alexu.edu.eg.;

Received: 17/2/2022; Accepted: 21/2/2022

DOI: 10.21608/ejss.2022.122443.1493

©2022 National Information and Documentation Center (NIDOC)

solution to reduce the unfavorable impacts of abiotic stress. SA is an alert phytohormone with various roles control of plant metabolism and abiotic stress tolerance (Arfan *et al.* 2007; Khan *et al.*, 2015). The influences of these materials on crop quality have less attention than its effect on yield, due to the difficulty of detecting and sometimes are not consensual. Using SA increased the yield of olive (Khalil *et al.*, 2012; Abdel-Razik *et al.*, 2011), peach (El-Shazly *et al.*, 2013), and strawberry (Kazemi, 2013) crops. Also, the quality of strawberry fruits was improved, with a higher accumulation of total phenols, both flavonoids and non-flavonoids, soluble solids, and vitamin C as a result of foliar application with SA (Kazemi, 2013).

An antioxidant such as ascorbic acid is safe for humans, and the environment (Elade, 1992). The application of exogenous ascorbic acid could reduce oxidative stresses (Shalata and Peter, 2001). It is considered an antioxidant and associated with other components of the antioxidant system. It protects plants against oxidative damage and plays various roles in plant growth, such as in cell division, cell wall expansion, and other developmental processes (Lee and Kader, 2000). Moreover, ascorbic acid functions as an enzymatic cofactor, and it plays significant roles in several physiological processes, such as photosynthesis, photo-protection, stress resistance, hormones biosynthesis, and cell wall ingredients (Davey *et al.*, 2000; Conklin and Barth, 2004). Ascorbic acid has auxinic action. It has a synergistic influence on improving the growth, flowering, yield, and fruit quality of fruit crops (Barth *et al.*, 2006).

The main objective was to determine the influence of spraying ascorbic and salicylic acids on growth physiological characteristics, yield, and fruit quality of olive variety "Picual" under water stress conditions.

Material and Method

Experimental design

A field experiment was performed during two seasons (2018 and 2019) at the experimental farm of Faculty of desert and environmental Agriculture (E = 27.912408', N = 31.072289'), Fuka, Matrouh governorate. The study was performed using olive cultivar Picual (dual purpose; table and oil). The experimental olive trees were 5 years old and had the same vigor and homogenous appearance. The applied treatments were implemented during the two growing seasons. The trees were trained according to the cup system and spaced 4m x 5. The drip irrigation system and routine cultural practices (nutrition, pruning, disease control) were set up.

The experiment was designed as a split-plot with three treatments of irrigation: 1) Irrigation by 100% from (ET_C); 2) Irrigation by 50% (ET_C), and 3) control rain-fed irrigation. The subplots were occupied by the seven spray treatments were as follows: 1) spray with ascorbic acid 50 µg/L (T1); 2) spray with ascorbic acid 100 µg/L (T2); 3) spray with salicylic acid 100 µg/L (T3); 4) spray with salicylic acid 150 µg/L (T4); 5) spray with ascorbic acid 50 µg/L+ salicylic acid 100 µg/L (T5); 6) spray with ascorbic acid 100 µg/L+ salicylic acid 150 µg/L (T6), and 7) control: using spray distilled water (T7). These treatments were applied at two different times (1st May and 1st June at the two seasons, respectively), and each tree was sprayed with 5 L of each solution.

Soil and Water analysis

Soil samples were collected from the surface to 85 cm depth. Samples were air-dried, ground, crushed, and passed through a 2mm sieve. The chemical and physical properties of experimental soil, soil extract are presented in Table (1) and the water irrigation analysis is presented in Table (2) according to the methods described by Page *et al.*, (1982). K and Na were measured by flame photometer (Black, 1965) and CaCO₃ was measured by calcimeter (Alison and Moodle, 1965). The particles size distribution was determined by the hydrometer method (FAO, 1970), and the organic matter was measured according to the method of Walkley- Black (Black, 1965).

Plant analysis and growth measurements

Growth measurements were taken every month from March to October on each tree, at the beginning of the two growing seasons (2018 and 2019), where five shoots aged one year, were selected around the canopy of each tree. The final shoot lengths and basal diameters of each tree were measured in April, June, August, and October for the two growing seasons.

In order to measure the leaf area, of each sampling, 20 leaves; were chosen, weighed, and scanned every year using an image analysis software (APS Assess, Winipeg, Canada). After harvesting, the entire production of each tree was weighted to calculate the yield, and then ten fruits per tree were used to measure fruit weight.

For the chlorophyll content, three fresh leaf samples were selected from each plant at the end of both experimental seasons. Total leaf chlorophyll was determined as maintained by Yadava (1986), using a Minolta SPAD 502 plus chlorophyll-meter model. Leaf relative water content (RWC) was determined according to Barrs and Weatherley (1962).

TABLE 1. Some physical and chemical characteristics of experimental soil

Parameters	Depth (cm)			
	0-20	20-40	40-60	60-85
Particle size distribution (%)				
Sand	85	85	82	72
Silt	5	5	5	10
Clay	10	10	13	15
Texture	Loamy	Loamy	Loamy	Loamy
	Sand	Sand	Sand	Sand
pH	7.73	8.18	8.15	7.97
EC _e (dS/m)	1.05	1.38	1.14	3.12
Soluble cations (meq/L)				
Ca ⁺⁺	5.00	5.00	5.00	6.50
Mg ⁺⁺	4.50	3.50	4.50	2.00
Na ⁺	0.50	4.50	1.50	5.50
K ⁺	0.17	0.01	0.17	0.22
Soluble anions (meq/L)				
CO ₃ ⁻	--	--	--	--
HCO ₃ ⁻	4.50	6.50	10.00	8.00
Cl ⁻	5.50	3.00	3.25	17.00
SO ₄ ⁻	0.50	0.88	0.01	6.20
Organic matter (%)	2.41	2.06	1.39	1.86
CaCO ₃ (%)	20.78	18.44	17.07	18.83

TABLE 2. Chemical properties of the applied irrigation water

pH	EC ds/m	Na ⁺	K ⁺	Ca ⁺⁺	Mg ⁺⁺	CO ₃ ⁻	HCO ₃ ⁻	Cl ⁻	SO ₄ ⁻	SAR
		meq / L								
7.84	1.08	6.40	0.50	4.40	1.10	--	2.40	6.00	3.30	3.85

For chemical leaf analysis, fully developed leaves (30–50) were collected from the middle portion of non-bearing branches in July for the two seasons. Leaf samples were rinsed in deionized water, dried at 60 °C for 72 h, and pulverized. Total N, P, K, Ca, and Mg contents of the leaves were determined after digestion with sulfuric acid and hydrogenperoxide. The concentrations of N were determined by MicroKjeldehl method (Jackson, 1973), Phosphorus was determined using the vanadomolybdic acid method (Chapmann and Pratt, 1961), Calcium, and Magnesium were determined by titration with Na₂-EDTA solution (Page et al, 1982). Proline determination was carried out according to the method of Bates (1973).

Statistical analysis

Statistical analyses were performed using statistix.8 Statistical (Analytical Software,

TallahsseeFL, USA). Means of each treatment were compared using the Least Significant Difference (LSD) at 5% according to (Steel and Torrie, 1985).

Results and Discussion

Vegetative growth measurements

The influence of different irrigation levels and salicylic acid (SA) and ascorbic acid (AA) foliar application on Picual olive growth (shoot length, cm and leaf area, cm²) for two seasons (2018 and 2019) wereshownin Table(3) and figure (1), respectively. Regarding the effect of the different irrigation levels on shoot length, the level of applied irrigation did not present a constant trend. Moreover, in the first season, it was observed that trees irrigated by 100% ET_c were significantly higher in shoot length than the rest treatments. On other hand, the Rain-fed treatment gave

the lowest values compared to other treatments. This may be due to result of deficit water stress at any growth stage of the plant causes negative influences on growth and progress according to the stress severity (Dichio *et al.* 2002). Significant differences in the shoot lengths were observed on trees treated with 100 % ET_c compared to the treatment of rain-fed, the highest value of shoot length of 13.2 cm was noticed with 100 % ET_c and the lowest value is 11.7 cm was observed in trees treated with rain-fed, respectively, during the two studied seasons. However, in the second season, there were no significant differences between the levels of irrigation. For the two seasons, it seems that shoot length is increased with an increasing rate of ascorbic or salicylic acids.

For leaf area results, it was observed a decrease of leaf area in the second season than the first one (Fig. 1). The effect of applying and increasing the rate of ascorbic or salicylic acids (from 50 to 100 mg AA /L and 100 to 150 mg SA/L) had a remarkable effect compared to the control. The highest value was presented by 100% ET_c or 50%ET_c plus 150 mg/L SA in both seasons. Moreover, the highest values of leaf area (11.9 and 7.5 cm²) were noticed with the 150 mg/L salicylic acid treatment (T5) in both seasons (Fig. 1).

The obtained results are in agreement with previous findings by Moriana *et al.* (2002 and 2003), El-Hosieny (2015), Randa (2015), Abd-El-Rhman and Attia (2016), and Mohamed *et al.* (2018). The positive effect of ascorbic acid treatments in enhancing cell division and cell enlargement could explain the positive effect of ascorbic acid in increasing of leaf area values and that was previously observed by Omar (1999). Similarly, the effects of salicylic acid in enhancing tolerance of fruit crop species to stresses and promoting effect of salicylic acid on cell division can explain the present results (Joseph *et al.*, 2010).

Yield and fruit weight

The obtained yield and fruit weight for the two experimental years are presented in Table (4). The "on" year of 2018 returned significantly different yield between 100%ET_c and the two other treatments, while the 50%ET_c yield was not significantly different from rain-fed treatment in the first season where the highest value yield was (4.16 Kg) with treatment 100% ET_c. In the second season the "off", each of 100% ET_c or 50% ET_c had a significant effect on the yield greater than rain-fed and there was no difference between

100%ET_c and 50%ET_c. That could indicate the fact that reducing water supply could reduce the roots, and biomass yields (Yassin *et al.*, 2021).

Results in Table (4) represented significant differences among treatments on fruit weight. The irrigation quantity (100%ET_c) had the advantage in fruit weight in both seasons consequently. Decreasing irrigation quantity to 50% significantly decreased fruit weight, and more decrease was noticed with rain-fed treatment. The results showed that the water stress decrease the fruit weight and the application of irrigation water for olive trees in both "on" or "off" years was more affected to increase it compared with the control (Rain-fed) and this might be due to the trees take its time to build up more carbohydrates and then raising the weight of the fruit. These results were in agreement with d'Andria *et al.* (2004) and Lavee *et al.* (2007) on supplemental irrigation effect. Also, results in Table (4) indicated that the fruit weight of Picual olive trees, increased with increasing the applied rates of foliar ascorbic acid, meanwhile it decreased with increasing salicylic acid rate. In this respect, the T3 (ascorbic acid 100mg/L) treatment induced higher fruit weight than the salicylic acid (150 mg/L), while the T2 induced the lowest effect.

Ascorbic acid treatment (100mg/L) induced the highest increase in fruit weight in comparison to the other treatments and recorded 4.0, 3.6 g for fruit weight in the first and second seasons, respectively. The treatment T3 in 100% ET_c induced higher increases in fruits weight than the control treatment. The obtained data gave a maximum value (6.0 and 5.5 g) for fruit weight parameters. Ascorbic acid acts as antioxidants, therefore expected increments of carbohydrates supplied to fruits can explain this improvement in yield fruit weight and flesh oil content obtained in this experiment (Johnson *et al.*, 1999; Maksoud *et al.*, 2009). Increasing yield under foliar application of salicylic acid could be ascribed to the well-known roles of salicylic acid on photosynthetic parameters and plant water relations. Fariduddin *et al.* (2003) referred that exogenous application of salicylic acid could improve the photosynthetic rate, internal CO₂ concentration, and water use efficiency in *Brassica juncea*. According to Shakirova *et al.* (2003), the favorable influences of salicylic acid on yield can be due to its influence on other plant hormones. Salicylic acid altered the auxin, cytokine in, and ABA balances in wheat and increased the growth and yield under both normal and saline conditions.

TABLE 3. Effect of water level, salicylic acid (SA) and ascorbic acid (AA) and their interaction on vegetative growth parameters shoot length of Picual olives in 2018 and 2019 seasons

Treatments		Shoot length (cm)	
		2018	2019
100%ET_C	T1= Control	9.7 fg	8.4 d
	T2= AA 50 mg/L	12.3 de	11.4 bcd
	T3= AA 100 mg/L	12.2 de	16.2 a
	T4= SA 100 mg/L	14.7 bc	12.2 ab
	T5= SA 150 mg/L	16.4 a	14.6 ab
50% ET_C	T1= Control	9.1 g	10.8 ab
	T2= AA 50 mg/L	11.2 ef	11.2 ab
	T3= AA 100 mg/L	13.1 d	12.8 bcd
	T4= SA 100 mg/L	13.3 cd	10.2 d
	T5= SA 150 mg/L	15.1 ab	15.3 bcd
Rain-fed	T1= Control	8.4 g	12.4 abcd
	T2= AA 50 mg/L	9.8 fg	14.0 abc
	T3= AA 100 mg/L	12.1 de	14.6 ab
	T4= SA 100 mg/L	13.2 d	10.4 cd
	T5= SA 150 mg/L	14.8 b	12.6 abcd

Means within a column followed by different letter (s) are statistically different at 0.05% level.

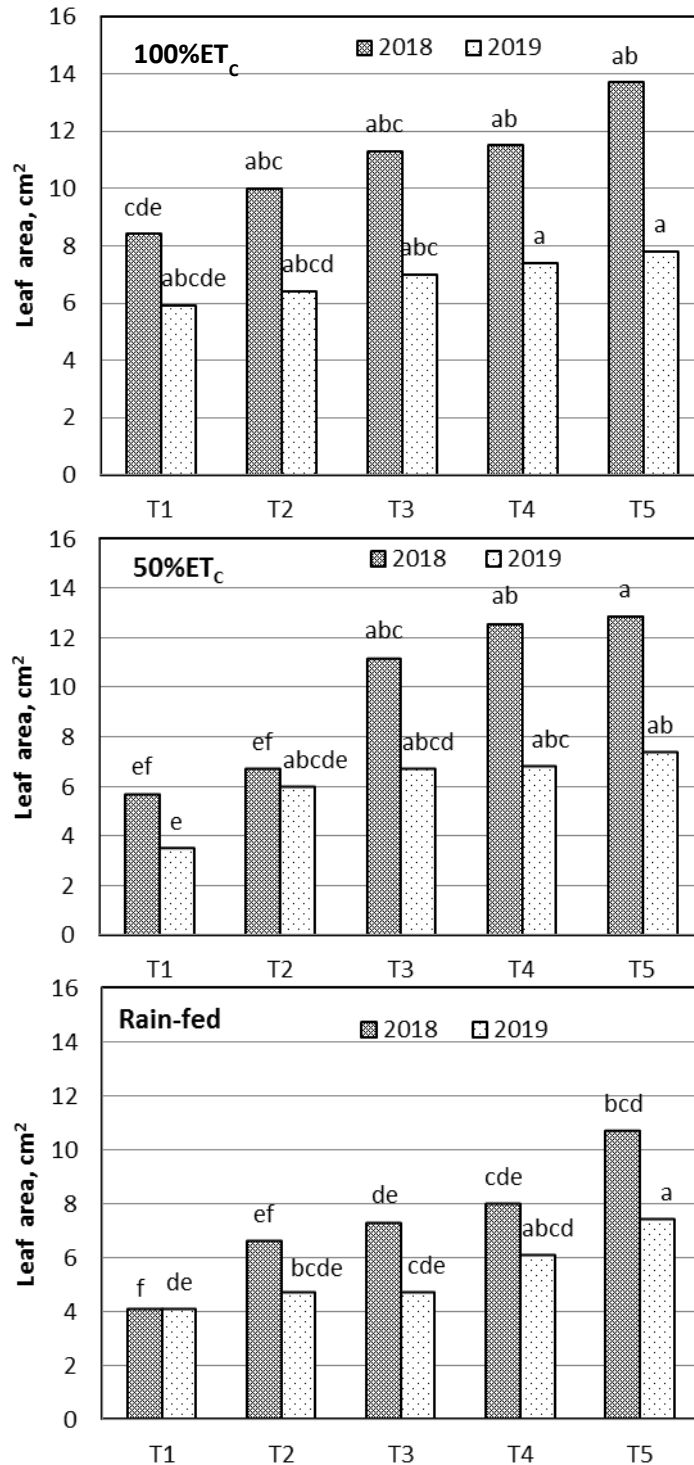


Fig. 1. Effect of water level (rain-fed, 50% ET_c, and 100% ET_c) and foliar applications with ascorbic acid (T2= 50 mg/L, and T3= 100 mg/L) and salicylic acid (T4= 100 mg/L, and T5= 150 mg/L) compared to control treatment without applications (T1) on leaf area (cm²) of Picual olives in 2018 and 2019 seasons (Each column labeled by different letter (s) is statistically different at 0.05% level)

TABLE 4. Effect of water regime, salicylic acid (SA), ascorbic acid (AA) or their interaction on yield (kg/tree) and Fruit weight (g) of Picual olives in 2018 and 2019 seasons

Treatments		Yield (kg/tree)		Fruit weight (g)	
		2018	2019	2018	2019
100% ET_C	T1= Control	0.1 f	0.072 f	1.1 g	0.8 e
	T2= AA 50 mg/L	2.6 c d	1.78cd	3.6 cde	3.5cd
	T3= AA 100 mg/L	5.7 b	3.85 b	6.0 a	5.5 b
	T4= SA 100 mg/L	5.8 ab	3.92 a b	4.9 b	4.4bc
	T5= SA 150 mg/L	6.6 a	4.43 a	4.7 b	4.3 a
50% ET_C	T1= Control	0.1 f	0.075 f	0.8 g	0.8 e
	T2= AA 50 mg/L	1.3 d e f	0.86 def	3.2 def	3.3 d
	T3= AA 100 mg/L	3.5 c d	1.68 c d e	4.0 c	3.3 d
	T4= SA 100 mg/L	4.9c	2.51 c	3.8 cd	3.4 cd
	T5= SA 150 mg/L	6.5 a b	3.46 a b	3.7 cd	3.49 b
Rain-fed	T1= Control	0.1 f	0.35 f	0.7 g	0.7 e
	T2= AA 50 mg/L	1.2 e f	0.30 d e f	1.0 g	1.0 e
	T3= AA 100 mg/L	3.5 c d	0.65 c d e	2.8 ef	2.9 d
	T4= SA 100 mg/L	3.7 c d	0.69 c d	2.8 f	2.9 d
	T5= SA 150 mg/L	4.3 c	1.26 d	1.2 g	1.2 e

Means within a column followed by different letter (s) are statistically different at 0.05% level.

Biochemical Constituents

Chlorophyll content

Generally, the results indicated high significant differences between the three irrigation treatments; severe water deficit (rainfall treatment) significantly decreased total chlorophyll per tree (Table 5). The highest value of chlorophyll per tree was recorded for trees irrigated with 100% ET_c (50.958 and 60.529SPAD for 2018 and 2019, respectively). The lowest values in chlorophyll were observed with rainfall treatment (25.992 and 24.992SPAD for 2018 and 2019, respectively). The obtained results regarding the effect of sustained deficit irrigation on leaf chlorophyll agree with the findings of Abo-Taleb *et al.* (1998), and Sary and Elsokkary (2019) that chlorophyll (a & b) of pomegranate trees decreased under severe water stress.

On the other side used foliar application of ascorbic or salicylic acids caused a significant increase in chlorophyll content especially with the highest concentrations compared with untreated trees in the two seasons. Whereas, the highest values were recorded in the treatments T3, Ascorbic acid 100 mg/L, which recorded 69.439 and 70.439SPAD, respectively, for both seasons (Table 5). Zulaikha (2013) reported that the foliar applications of ascorbic acid caused a significant increase in leaf chlorophyll content of olive trees. Similarly, investigation of Blokhina (2003) revealed that ascorbic acid as an important antioxidant increased the chlorophyll in plants. In this respect, (Hayat *et al.*, 2010) reported that SA treatment increased leaf area and enhanced the rate of photosynthesis, total carbohydrates, and photosynthetic pigments thereby, AA application could affect the plant growth regulators (Abbaspour and Babaee, 2017). As for interaction effect, three levels of irrigation and spraying with salicylic and ascorbic acid gave a maximum value of leaf chlorophyll content especially, with T3 in 100% ET_c but there were no significant differences between treatment T3 in 100% ET_c and 50% Etc (Table 5).

Relative water content

The increase in values of the relative water content (%) under water deficit conditions indicates that the plant has got its need from water to accomplish the different plant physiological functions. Relative leaf water content (RWC) is an integrative index of plant water status that is used to estimate the water stress tolerance. Reduction in RWC under drought stress

leads to stomatal closure, which further results in decreased CO₂ assimilation (Gindaba *et al.*, 2004).

Results in figure (2) showed that relative water content (%) of Picual olive leaves was increased as the amount of added irrigation water increased. Therefore, the treatment 100 % ET_c showed the maximum significant RWC values compared with that was getting at 50% and Rain-fall treatments. Abdel-Razik (2011) and Khalifa (2013) reported that mango plants produced low values of relative water content as a result of high water deficit. Loss of turgor and cell elongation limitation in the apex and in the developing leaves of the olive cultivars could be expected due to the decrease of leaf RWC under drought stress (Ings *et al.*, 2013). In comparison with normal irrigation conditions, the shoot growth of drought-stressed 'Dezful', 'Conservolia', and 'Amigdalolia' had fewer influence by drought stress. Since Specific leaf weight (SLW) is recognized as positively correlated with plant growth rate, the highest SLW of 'Conservolia' and 'Amigdalolia' indicated their higher vigor under drought stress (Karimi *et al.*, 2018).

The relative water content (%) in leaves was affected significantly by treatments of ascorbic and salicylic acids (Fig. 2). All treatments significantly increased RWC in comparison with control. The highest content of RWC in leaf was recorded with treatment T5 (81.966 and 88.966%) in the two seasons respectively. This is logical since it is known that both AA and SA are antioxidants and anti-stress agents. Moreover, the lowest significant leaf RWC content was observed in control trees in comparison with the other treatments, which recorded the lowest values (49.640 and 40.640%) during two seasons of study, respectively. These data are in harmony with those reported by Dolatabadian *et al.* (2008). In this, respect, the increase in such character was obtained in plants sprayed with salicylic acid 150 mg/L. Generally, the interaction between irrigation treatments and spray with salicylic and ascorbic acid treatments, it can be noticed that 100% ET_c plus treatment with salicylic acid 150 mg /L gave the highest values of leaf relative water content (79.533 and 85.533%) in the two seasons respectively. No significant differences were noticed between treatment T5 in 100% ET_c and 50% ET_c.

Table 5. Effect of water level, salicylic acid (SA), ascorbic acid (AA) and their interaction on leaf chlorophyll content of Picual olives in 2018 and 2019 seasons

		Chlorophyll (SPAD)	
Treatments		2018	2019
100% ETC	T1= Control	73.951 d	76.99 abcd
	T2= AA 50 mg/L	74.11bcd	79.11bcd
	T3= AA 100 mg/L	83.60 a	88.60 a
	T4= SA 100 mg/L	75.11bc	80.11ab
	T5= SA 150 mg/L	76.08 bc	85.08 bc
50% ETC	T1= Control	49.52 fg	51.52ef
	T2= AA 50 mg/L	51.23 ef	54.23 d ef
	T3= AA 100 mg/L	80.12 ab	86.12 b
	T4= SA 100 mg/L	62.42 d ef	64.42 bcde
	T5= SA 150 mg/L	63.00 def	65.97 abcde
Rain-fed	T1= Control	20.06 cdef	15.06 h
	T2= AA 50 mg/L	25.53 h	27.53 g
	T3= AA 100 mg/L	45.80 gh	31.80 h
	T4= SA 100 mg/L	29.50 def	32.50 hi
	T5= SA 150 mg/L	30.52def	50.52 f

Different letter (s) represents the statistically different at 0.05% level.

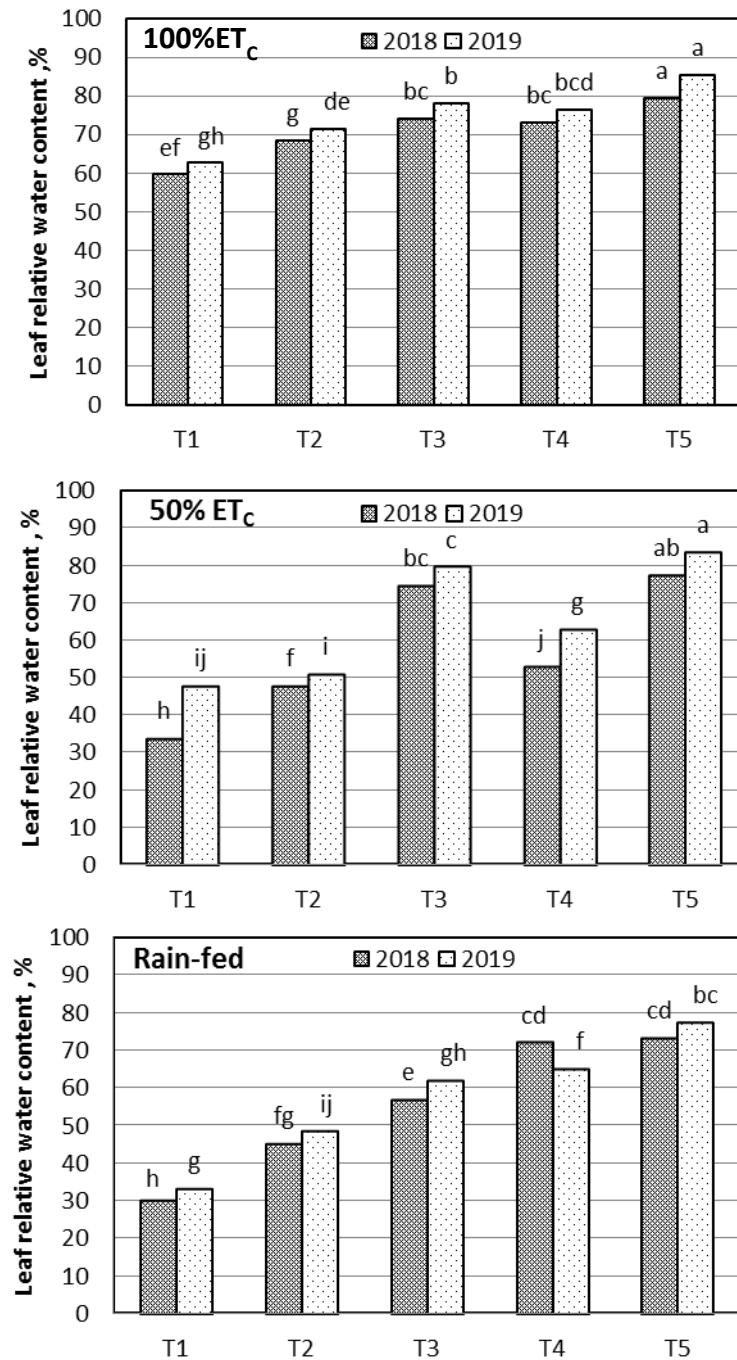


Fig. 2. Effect of water level (rain-fed, 50% ET_c, and 100% ET_c) and foliar applications with ascorbic acid (T2= 50 mg/L, and T3= 100 mg/L) and salicylic acid (T4= 100 mg/L, and T5= 150 mg/L) compared to control treatment without applications (T1) on relative water content (%) of Picual olives in 2018 and 2019 seasons.

(Each column labeled by different letter (s) is statistically different at 0.05% level)

Leaf nutrients

Results in Table (6) showed that the leaf nutrients concentrations were affected by irrigation treatments and increasing the amounts of applied water affect these values significantly in comparison with rain-fed treatment (for season 2018). Leaf N concentrations reached a high value of 1.6% with 100% ETc treatment and this may be due to the fact that giving nitrogen had the chance to move with water flow into root tissues (Smith and Kenuworthy, 1979; Abou-Amer, 2013). The leaf-P concentration increased linearly ($P < 0.05$) as a function of increasing the application of irrigation water, this might be due to the diffuse of it from the soil to the root tissues (Smith and Kenuworthy, 1979; Zahran *et al.*, 1987).

Also, the leaf- K gives a similar trend as a result of irrigation level and the concentration ranged between 0.99 % and 1.21% Table (6). The values measured in the rain-fed treatment indicated the lowest value (0.99 %). Indeed, the low potassium assimilation, make its concentration high in the fruit and this was correlated with oil accumulation (Celano, 1999). Leaf- Ca concentrations were affected by irrigation level where the lowest concentration was noticed with low-irrigation treatments and the highest values were observed with the high-irrigation treatments. Leaf-Mg concentration -response to irrigation level was similar to Ca response. Induced by the water stress, the total amount of mineral elements increased in the irrigated plants with respect to the control (Rain-fed) on olive trees. Moreover, Alegreet *al.*, (2002) found that water stress reduced net assimilation of leaves for nutrients. The effect of different spraying treatments on leaf nitrogen, phosphorus, potassium, calcium and magnesium contents of Picual olives was represented

in Table (6). Nitrogen concentrations in the leaves were affected by various spraying concentrations. Trees sprayed with ascorbic 100 mg/L (T4) gave the highest leaf content of N, P, K, Ca, and Mg (1.51, 0.35, 1.17 and 1.25, 1.51%) respectively, while control treatment gave the lowest leaf contents of N, P, K, Ca, and Mg (1.28, 0.30, 1.02, 1.10 and 1.28%), respectively. Regarding to the interaction effect, irrigation with high water levels and spraying with salicylic and ascorbic acids (T3 in 100% ETc or 50% ETc) recorded the highest values of all determined minerals (N, P, K, Ca and Mg) Table (6). The previous results are in harmony with the findings of El-Badawy (2013) who illustrated that the highest leaf N, P, and K contents in Canino apricot trees were noticed with treating the trees with ascorbic acid at 1000 ppm. Moreover, in mango trees cv. Fagriclan, El-Hosieny (2015) illustrated that spraying 1 Mm ascorbic acid increased leaf N, P and K contents. Also, Randa

(2015) cleared that spraying salicylic acid at 400 ppm once or twice considerably increased N, K and P percentages of orange leaves. In addition, Abd-El-Rahman and Atia (2016) observed that spraying salicylic acid at 1000 ppm gave the maximum leaf mineral contents of N, K, P, Zn, Fe and Mn on olive trees. In this respect, on Manfaloty pomegranate, Abd-El-Rahman *et al.*, (2017) revealed that spraying ascorbic acid at 2000 ppm improved leaf mineral contents (N, K, P, Mg and Ca). Finally, Abdel Aziz *et al.*, (2017) found that spraying salicylic acid at 100 or 200 ppm at three periods (growth start, after fruit set and one month later) increased leaf contents of Zn and Fe on leaves of pomegranate trees.

Generally, the results of the present study showed the capability of olive trees to control the enzymatic antioxidant system and this could be a serious assignment related to drought tolerance that could end cellular harm by active oxygen species through water deficit. It is worthy to note that, ascorbic acid as an antioxidant is very effective in improving the plant biomass (Ejzet *al.*, 2012). Also, the previous results agreed with those obtained by El-Sayed *et al.* (2014).

Leaf proline content

According to Figure (3) severe water deficit (rain-fall treatment) significantly increased the leaf proline content. The lowest value of proline content per tree was recorded for trees irrigated with 100% ETc (1.574 and 1.121 ppm in 2018 and 2019 seasons respectively). The highest reduction in proline content was observed under rain-fall treatment (6.184 and 8.184 ppm for 2018 and 2019, respectively). In general, significant differences were observed on trees treated with 100 % ETc and 50% ETc compared to the treatment of rain-fall. On the other side, foliar application of ascorbic or salicylic acid, showed a significant decrease in proline content in trees treated with the highest concentrations, compared with untreated trees. The highest values of leaf proline content were recorded with control treatment (7.773 and 9.773 ppm) in the two seasons respectively (Fig. 3). These findings are logical in the light of the fact that AA and SA are both antioxidants and anti-stress agents. Moreover, the lowest significant leaf proline content was observed in “the Picual olive variety that treated with ascorbic or salicylic in comparison with other treatments. It is worthy to note that T5 recorded the lowest values of leaf proline content (1.435 and 1.135 ppm) during two seasons of study respectively. Generally, the treatment 100% ETc plus salicylic acid at 150 mg L gave the lowest value of proline compared to control and the other treatments (Fig. 3). No significant differences were noticed between treatments 100% ETc + T5 and 50% ETc + T5. These data are in harmony with those reported by

(Dolatabadian *et al.*, 2008). In addition, Hassan (2015) reported that under salt stress conditions, these antioxidants (ascorbic and salicylic acids) had the most beneficial effect on the production of fig and olive trees and increasing the ability of olive and fig trees to

increase proline levels and resistance to drought conditions and increased the plant ability to growth under rain-fed areas of Matarouh, this effect was also reported by Khaled and Fawy (2011).

TABLE 6. Effect of water regime, salicylic acid (SA), ascorbic acid (AA) and their interaction on leaf mineral contents (N, P, K, Ca, and Mg) of Picual olives (in 2018growing season)

Treatments		N	P	K	Ca	Mg
		%				
100% ET_C	T1= Control	1.51 b c d	0.35a b c d	1.18abc	1.31cd	1.51bcd
	T2= AA 50 mg/L	1.56abc	0.35abcd	1.20 ab	1.36 c d	1.56 a b c
	T3= AA 100 mg/L	1.71 a	0.37 a	1.23 a	1.55 a	1.71 a
	T4= SA 100 mg/L	1.65 a b	0.36 a b	1.21 a b	1.49 a b	1.65 ab
	T5= SA 150 mg/L	1.58 a b c	0.36 a b	1.21 a b	1.43 b c	1.58 a b c
50% ETC	T1= Control	1.30fg	0.31 ef	1.04 ef	1.10 fgh	1.30 fg
	T2= AA 50 mg/L	1.34ef	0.32 def	1.05 d e f	1.12 f gh	1.34 ef
	T3= AA 100 mg/L	1.49 def	0.32 c d e f	1.12 c d e	1.21 f gh	1.39 d e f
	T4= SA 100 mg/L	1.39 def	0.33 b c d e	1.10 b c d e	1.15 fg	1.38 def
	T5= SA 150 mg/L	1.37 c d e f	0.33 abcde	1.08 abcde	1.13 e f	1.37 d e f
Rain-fed	T1= Control	1.03 h	0.33 abcde	0.83 h	0.90 i	1.44 cdef
	T2= AA 50 mg/L	1.18 gh	0.24 h	0.95 fg	1.07 gh	1.03 h
	T3= AA 100 mg/L	1.44 def	0.29 fg	1.10 bcde	1.20 ef	1.18 gh
	T4= SA 100 mg/L	1.38 c d e	0.34 abcde	1.16 abcd	1.28 de	1.49cde
	T5= SA 150 mg/L	1.16 gh	0.26 gh	0.92 gh	1.02 hi	1.16 gh

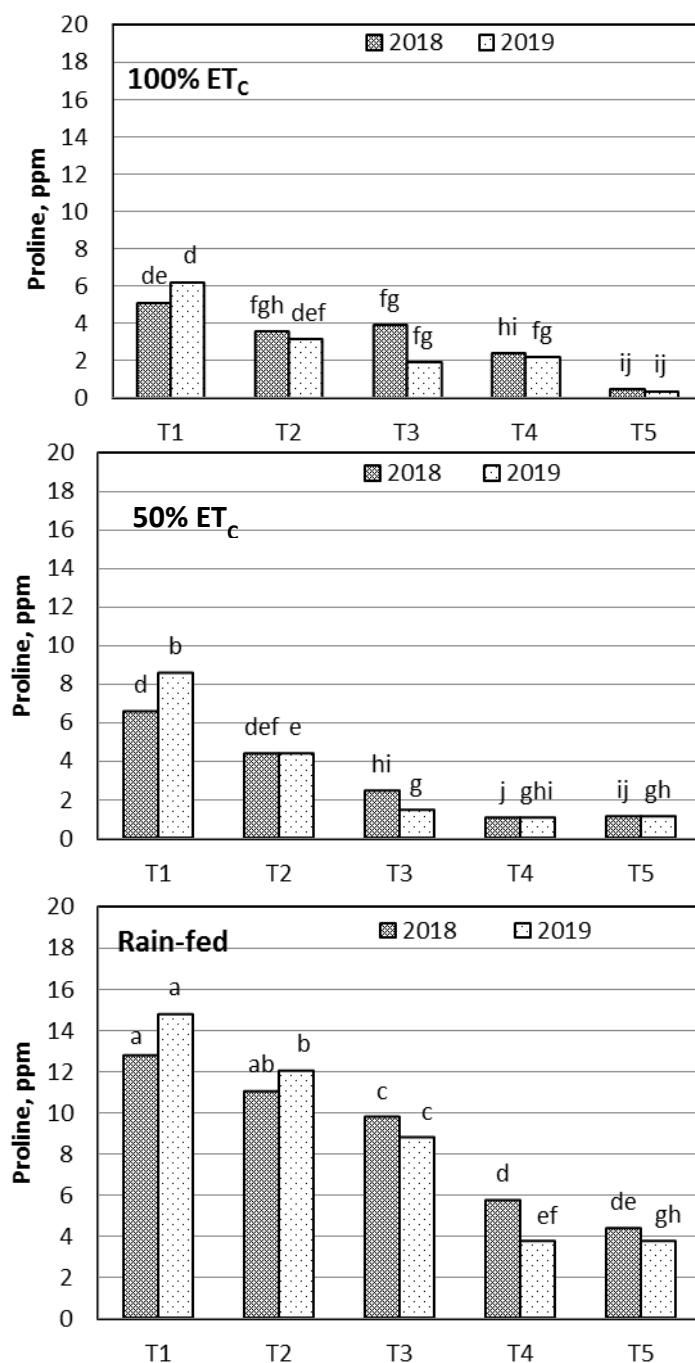


Fig. 3. Effect of water level (rain-fed, 50% ET_c, and 100% ET_c) and foliar applications with ascorbic acid (T2= 50 mg/L, and T3= 100 mg/L) and salicylic acid (T4= 100 mg/L, and T5= 150 mg/L) compared to control treatment without applications (T1) on leaf proline content (ppm) of Picual olives in 2018 and 2019 seasons.

(Each column labeled by different letter (s) is statistically different at 0.05% level)

CONCLUSION

Opposite of 100% ETc, plant growth characteristics of the rain-fed treatment, including shoot length, leaf area per plant, yield, the leaf nutrient concentrations (N, P, K, Ca, and Mg), total chlorophyll, and leaf relative water content, significantly decreased due to water deficit but water shortage significantly increased leaf proline content. The highest values of shoot length, leaf area, relative water content, yield kg/tree, were recorded in the treatments of salicylic acid (150 mg/L). Sprayed with salicylic acid 100 mg/L and ascorbic acid (100 mg/L) treatments gave the highest leaf content of N, P, K, Ca, and Mg (%) compared to other treatments. Ascorbic acid 100 mg/L treatments gave the highest value of leaf chlorophyll content (SPAD) in the two seasons compared to other treatments. Salicylic acid 150 mg/L treatments gave the lowest value of proline content in olive leaves compared with control treatments. In general, there are no significant differences were noticed between irrigation treatment 100% ETc, and irrigation treatment 50% ETc Plus salicylic acid 150 mg/L or ascorbic acid 100 mg/L for characteristics precedent mentioned, so for locality economics, it is recommended to use 50% ETc irrigation plus salicylic acid 150 mg/L or ascorbic acid 100 mg/L.

References

- Abbaspour, N, Babae, L (2017). Effect of salicylic acid application on oxidative damage and antioxidant activity of Grape (*Vitisvinifera L.*) under drought stress condition. *Int. J. Hort. Sci. Technol*, **4**(1), 29-50.
- Abdel-Razik , A M (2011). Effect of water deficit on growth of some mango (*Mangifera indica L.*) rootstocks. *Nat. Sci.* **11**(10), 136-142.
- Abd-El-Rhman I E, Attia M F (2016). Foliar spray with potassium nitrate and salicylic acid for improving growth, yield and nutrients uptake by olive trees under salinity stress conditions. *Inter. Jour. of Chem Tech Res.*, **9**(12), 230-245.
- Abo-Taleb A S, Noaman F V, Sari El-Deen S (1998). Growth of pomegranate transplants as affected by different water regimes. *Ann. Agric. Sci. Moshtohor.*, **36**(2), 1073- 1091.
- Abou-Amer A I (2013). Effect of rates and methods of N application on growth, fruit yield and mineral content of Manzanillo olive trees. *Egypt. J. Soil Sci.*, **53**(2), 161-173.
- Alegre S, Marsal J, Arbones A, Girona J, Tovar M J (2002). Regulated deficit irrigation in olive trees (*Olea europaea L. cv. Arbequina*) for oil production. *Acta Horticulturae*, **586**, 259-261.
- Alison L E, Moodle C D (1965). Carbonate. In: *Methods of Soil Analysis*, C.A. Black (Ed.). Amer. Soc. Agron. Inc., Madison, Wisc., pp. 1379-1396.
- Arfan M, Athar H R, Ashraf M (2007). Does exogenous application of salicylic acid through the rooting medium modulate growth and photosynthetic capacity in two differently adapted spring wheat cultivars under salt stress? *J. Plant Physiol.* **164**, 685-694
- Atta-ur-Rahman BA (1990). *Studies in natural products chemistry*, 32. Elsevier Science, Amsterdam.
- Barrs H D, Weatherley P E (1962). A re-examination of the relative turgidity technique for estimating water deficits in leaves. *Aust. J. Biol. Sci.*, **24**, 519-570.
- Barth C, Tullio M D, Conklin P L, (2006). The role of ascorbic acid in the control of flowering time and the onset of senescence. *J. Experimental Botany*, **57**, 1657-1665.
- Bates L S, Waldern R P, Teare I D (1973), Rapid determination of free proline for water stress studies. *Plant and Soil*, **39**, 205-207.
- Black C A, Evans D D, White J L, Ensminger L E, Clark F E (1965). *Methods of Soil Analysis*. Parts 1 and 2. Am. Soc. Agron., Inc., Madison, Wisconsin, U.S.A.
- Blokhina O, Virolainen E, Frgerstedt KV (2003). Antioxidants, oxidative damaged and oxygen deprivation stress. A rev. *Ann. of Botany*, **19**, 179-194.
- Celano G B, Dichio G, Montanaro V, Nuzzo A, Palese M, Xiloyannis C (1999). Distribution of dry matter and amount of mineral elements in irrigated and non-irrigated olive trees. *Acta Hort*, **474**:381-384.
- Chapmann H D, Pratt P F (1961). *Methods of Analysis for Soils, Plants and Waters*. Div. Agric. Sci., Univ. of California, Berkeley.
- Conklin P, Barth C (2004). Ascorbic acid, a familiar small molecule intertwined in the response of plants to ozone, pathogens, and the onset of senescence. *Plant Cell Environ*, **27**:959-970.
- d'Andria R, Lavini A, Morelli G, Patumi M, Terenziani S, Calandrelli D, Fragnito, F (2004). Effects of water regimes on five pickling and

- double aptitude olive cultivar (*Olea europaea* L.). *J. Hort. Sci Biotechnol.* **79**(1), 18-25.
- Davey M, Montagu M, Inze D, Sanmartin M, Kanellis A, Smirnoff N, Benzie I, Strain J, Favell D, Fletcher J (2000). Plant L-ascorbic acid: chemistry, function, metabolism, bioavailability and effects of processing. *J Sci Food Agric*, **80**, 825–860.
- Dichio B, Romano M, Nuzzo V, Xiloyannis C (2002). Soil water availability and relationship between canopy and roots in young olive trees (cv Coratina). *Acta Hort.*, **586**, 255–258.
- Dolatabadian A, Sanavy S A M M, Chashmi N A (2008). The effects of foliar application of ascorbic acid (vitamin C) on antioxidant enzymes activities, lipid peroxidation and proline accumulation of canola (*Brassica napus* L.) under conditions of salt stress. *J. Agro. Crop. Sci.* **194**, 206–213.
- Ejaz B, Sajid Z A, Aftab F (2012). Effect of exogenous application of ascorbic acid on antioxidant enzyme activities, proline contents, and growth parameters of *Saccharum* spp. hybrid cv. HSF-240 under salt stress. *Turk J Biol.*, **36**, 630–640.
- Elade Y (1992). The use of antioxidants to control gray mould (*Botryticcibera*) and white mould (*Sclerotinia Aclerotiorum*) in various crops, *Plant Pathol.*, 141, 417-426.
- El-Badawy H E M (2013). Effect of Some Antioxidants and Micronutrients on Growth, Leaf Mineral Content, Yield and Fruit Quality of Canino Apricot Trees. *J. Appl. Sci. Res.*, **9**(2), 1228-1237.
- El-Hosieny , H A M R (2015). Effect of ascorbic and salicylic acid on leaf area, N, P, K content as well as yield and its components of mango (*Mangifera indica*.) Trees. *J. Plant Production*, Mansoura Univ., **6** (10), 1619-1629.
- El-Sayed O M, El-Gammal O H M, Salama A S M (2014). Effect of ascorbic acid, proline and jasmonic acid foliar spraying on fruit set and yield of Manzanillo olive trees under salt stress. *Scientia Horticulturae*, **176** (11), 32–37.
- El-Shazly S M, Eisa A M, Mo'atamed A M H, Kotb H R M (2013). Effect of some agrochemicals preharvest foliar application on yield and fruit quality of "Swelling" peach trees. *Alex. J. Agric. Res*, **58**, 219–229.
- FAO (1970). Physical and Chemical Methods of Soil and Water Analysis. *Soils Bull.* No. 10, FAO, Rome, Italy.
- FAOSTAT (2022) Statistical Division of the UN Food and Agriculture Organization.
<http://www.fao.org/faostat/en/#hom>.
- Fariduddin Q, Hayat S, Ahmed A (2003). Salicylic acid influences net photosynthetic rate, carboxylation efficiency, nitrate reductase activity and seed yield in *Brassica juncea*. *Photosynthetica*, **41**(2), 281-284.
- Farooq M, Wahid A, Kobayashi N, Fujita D, Basra S M A, (2009). Plant drought stress: effects, mechanisms and management. *Agron Sustain Dev.*, **29**(1), 185–212.
- Gindaba J, Rozanov A, Negash L (2004). Response of seedlings of two Eucalyptus and three deciduous tree species from Ethiopia to severe water stress. *Fore. Ecol. Manage.* **201**(1), 119-129.
- Hassan A F (2015) Foliar application of organic acids and antioxidants impact on fruit yield and quality of fig and olive trees in some valleys of Northwestern Coast of Egypt. *Minufiya J. Agric. Res.* **40**(6)1629-1645.
- Hayat Q, Hayat S, Ifran M, Ahmad A (2010). Effect of exogenous salicylic acid under changing environment: a review. *Environmental and Experimental Botany*, **68**, 14–25.
- Ings J., Mur L A J, Robson M P R, Bosch M (2013). Physiological and growth responses to water deficit in the bioenergy crop *Miscanthus × giganteus*. *Front. Plant Sci.*, **4**, 468–475.
- Jackson M L (1973). Soil Chemical Analysis, Prentice Hall, Inc. Englewood Cliffs, N.J.
- Johnson J R, Fahy D, Gish N, Andrews P K (1999). Influence of Ascorbic acid sprays on apple sunburn. *Good Fruit Grower*, **50**(13), 81-83.
- Joseph B, Jini D, Sujatha S (2010). Insight into the role of exogenous salicylic acid on plants grown under salt environment. *Asian J. Crop Sci.*, **2**, 226-235.
- Karimi S, Rahemi M, Rostami A A, Sedaghat S (2018). Drought Effects on Growth, Water Content and Osmoprotectants in Four Olive Cultivars with Different Drought Tolerance, *International Journal of Fruit Science*, **18**:3, 254-267.
- Kazemi M (2013). Foliar application of salicylic acid and calcium on yield, yield component and chemical properties of strawberry. *Bull. Environ. Pharmacol. Life Sci.*, **2**, 19–23.

- Khaled H, Fawy H A, (2011). Effect of different levels of humic acids on the nutrient content, plant growth, and soil properties under conditions of salinity. *Soil and Water Res.* **6**(1), 21–29.
- Khalifa S M (2013) Water requirements of mango trees in reclaimed land. *Ph. D. Thesis*, Fac. Agric., Al - Azhar Univ., Cairo, Egypt.
- Khalil F, Qureshi K M, Khan A, Fakharul H, Bibi N (2012). Effect of girdling and plant growth regulators on productivity in olive (*Olea europaea*). *Pak. J. Agric. Res.* **25**, 30–38.
- Khan M I R, Fatma M, Per T S, Anjum N A, Khan N A (2015). Salicylic acid-induced abiotic stress tolerance and underlying mechanisms in plants. *Front. Plant Sci.* **6**, 462.
- Lavee S, Hanoch E, Wodner M, Abramowitch H (2007). The effect of predetermined deficit irrigation on the performance of cv. Muhasan olives (*Olea europaea* L.) in the eastern coastal plain of Israel. *Sci. Hort.*, **112**, 156-163.
- Lee S K, Kader A A (2000). Preharvest and postharvest factors influencing vitamin C content of horticultural crops. *Post Harv. Biol. Technol.*, **20**, 207-220.
- Maksoud M A, Saleh M A, El-Shamma M S, Fouad A A (2009). The beneficial effect of biofertilizers and antioxidants on olive trees under calcareous soil conditions. *World J. Agri. Sci.*, **5** (3), 350-352.
- Marsal J, Mata M, Arbones A, Rufat J, Girona J (2002) Regulated deficit irrigation and rectification of irrigation scheduling in young pear trees: an evaluation based vegetative and reproductive response. *Eur. J. Agron.*, **17**, 111-122.
- Mohamed Y I, Abdel-Sattar M, Moriana A (2018) Effect of several deficit irrigation schedules on fruit set and fruit growth of olive trees in the north coast region of Egypt. *Acta Hort.*, **1199**, 363-368.
- Moriana A, Orgaz F, Fereres E, Pastor M (2003). Yield responses of a mature olive orchard to a water deficits. *J Am Soc. HorticSci*, **128**, 425–431.
- Moriana A, Villalobos F J, Fereres E (2002). Stomatal and photosynthetic responses of olive (*Olea europaea* L.) leaves to water deficits. *Plant Cell Environ*, **25**(3), 395–405.
- Omar A K (1999). Response of Red grapevines (*Vitisvinifera* L.) to some and biofertilizer treatments. *M. Sc. Thesis*. Fac. Agric. Minia Univ. Egypt.
- Page A L, Miller R H, Keeney D R (1982). *Methods of Soil Analysis. Part 2: Chemical and Microbiological Properties*. Second edition, Agronomy J.9, 2, Am. Soc. Agron. Inc., Soil Sci. Soc. Am. Inc. Pub. Madison, Wisconsin, USA.
- Randa E H Y (2015). Effect of spraying salicylic acid on fruiting of Valencia orange trees. *Alex. J. Agric. Res.* **60**(3), 119-126.
- SaryD, Elsokkary I (2019) Effect of irrigation water regime in presence of organic or biological fertilizer on olive trees. *Egypt. J. Soil. Sci.* **59**(1), 67- 84.
- Shakirova F M, Sakhabutdinova A R, Bezrukova M V, Fatkhutdinova R A, Fatkhutdinova D R (2003). Changes in the hormonal status of wheat seedlings induced by salicylic acid and salinity. *Plant Sci.*, **164**, 317-322.
- Shalata A, Peter M N (2001). Exogenous ascorbic acid (vitamin C) increases resistance to salt stress and reduces lipid peroxidation. *J. Exp. Bot.*, **52**, 2207– 2211.
- Smith M W, Kenuworthy A L (1979). The response of fruit trees in Michigan to trickle irrigation. *Communications in Soil Science and Plant Analysis*, **10** (11), 1370-1380.
- Steel R G D, Torrie J H (1985). *Bioestadística: Principios y procedimientos*. Ed. McGraw-Hill, Bogota, 621p. Stewart, V.I. Water in the soil. *Welsh Soils Disc. Group Rpt.*, **4** (1963) 1-10
- Villalobos F J, Orgaz F, Testi L, Fereres E (2000) Measurement and modeling of evapotranspiration of olive (*Olea europaea* L.) orchards. *Eur. J. Agric.*, **13**, 155–163.
- Yadava U L (1986). A rapid and non-destructive method to determine chlorophyll in intact leaves. *Hort.Sci.*, **21**, 1449-1450.
- Yassin O M, Ismail S M, Gameh M A, Khalil F A F, Ahmed E M (2021) Optimizing roots and sugaryields and water use efficiency of different Sugar Beet varieties grown under Upper Egypt conditions using deficit irrigation and harvesting dates. *Egypt. J. Soil Sci.*, **61**(3), 367 – 372.
- Zahran A M, Resk A I, Eissa A M (1987). Effect of irrigation and non-irrigation on leaf mineral composition and yield of olive cultivars. *Ann. Agric. Sci., Fac. Agric., Ain Shams Univ., Cairo, Egypt.*, **32**(1), 591-600.

Zulaikha R (2013). Effect of foliar spray of ascorbic acid, Zn, seaweed extracts force and bio fertilizers vegetative growth and root growth of olive (*OleaEuropea L.*) transplants cv. Hog Blanca. *Int J Pure Appl. Sci. Technol.*, **17**, 79–89.