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Research Article

THE EFFECTS OF IRRIGATION ON CHLOROPHYLL CONTENT OF POMEGRANATE (*Punica granatum* L.) TREES

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ABSTRACT. Pomegranate (*Punica granatum* L.) is a characteristic species of the Mediterranean area. It is well adapted to the growth conditions in Turkey and is frequently found growing in wild or semi wild conditions. Pomegranates can tolerate long duration of drought once the plant is established but regular irrigation is mandatory in commercial production. In this research the effect of different irrigation on content of leaf pigments of four different pomegranate cultivars was determined. Three different water amounts were applied by drip irrigation system. First leaf samples were taken a week before starting the irrigation treatments and continued until the end of the harvest season with four weeks interval and they were taken five times. Results showed that all chlorophyll components (a, b and total) were affected by the water and they were higher in the irrigated trees than non irrigated trees.

Keywords: *Pomegranate, drought stress, irrigation, chlorophyll a, chlorophyll b*

INTRODUCTION

The pomegranate (*Punica granatum* L.) is one of the oldest known edible fruits. Its history dates to very ancient times [5, 13]. This fruit tree is one of the species mentioned in the holy books and is often associated to fertility [16].

In Turkey, the number of pomegranate orchards started to increase rapidly since the last decade of the 1900s. Pomegranate grows mainly in tropics and subtropics climates; also in warm temperature zone. It adapts to all kinds of soils and climate and tolerant to salinity and drought conditions. Besides, it is tolerant to iron chlorosis and limestone in the soil [8].

Regular irrigation and nutrition are required for obtaining high quality and quantity. Fruit cracking and splitting is an important problem in the pomegranate cultivation. Fruit splitting occur because of irregular irrigation practices or excessive rain during the maturation period and it is accepted a major fruit detect [6, 7, 19]. Also, it is due to extreme fluctuations in day and night temperatures, soil moisture and relative humidity, dry wind, heavy rain or irrigation following a dry condition in developed fruits [8]. Especially excessive nitrogenous fertilization increases fruit cracking [10]. Trees take up the plant nutrients from the soil by their roots as dissolved in water. For this reason, even if there are enough nutrients around its root, because of inadequate water supply it is not possible to take up the nutrient by the plants. It is necessary to provide irrigation without causing hydric stress in the plants for optimum growth and quality fruit production. Optimal irrigation duration, frequency and amount depend on some factors. These factors are soil type, temperature and relative humidity, tree size, age, rootstock, specie, variety, growing phase of tree and evaporation [10]. The applied water affects vegetative and generative growth, yield and fruit quality of trees. The harmful effect of water stress on plant growth is attributed to the decreased osmotic potential of the growing medium and nutrient ions shortage.

The chlorophylls are virtually essential pigments for the photosynthesis. Solar radiation absorbed by a leaf is largely a function of the foliar concentrations of photosynthetic pigments. Therefore low chlorophyll concentrations can directly limit photosynthetic potential and hereby primary production [9]. Chlorophyll amount gives an indirect estimation of the nutrient status because much of leaf nitrogen is incorporated in chlorophyll [18]. In addition, leaf chlorophyll amount is closely related to plant stress [17].

The goal of this study is to examine effects of different irrigation amounts on leaf chlorophyll contents of pomegranate trees.

MATERIALS AND METHODS

This study was done with 'İzmir 1', 'İzmir 2', 'İzmir 1499' and 'İzmir 1513' pomegranate varieties in Bornova County of İzmir province, Turkey.

Three different irrigation water amounts (S_0 = no irrigation, S_1 = 50% and S_2 = 100% of the water quantity evaporating from class A pan) were applied by drip irrigation system. Evaporation was measured with class A-pan. The crop pan coefficients (Kcp) for I_1 and I_2 irrigation treatment was taken 0.50 and 1.00, respectively. Irrigations were applied with 7-day interval. The applied quantity of irrigation water was calculated as given below formula.

$$I = Epan \times Kcp \times A \times P$$

Where:
I: The quantity of irrigation water (liters);
Epan: The quantity of evaporation in class A pan (mm);
Kcp: Crop pan coefficient;
A: Tree area (18 m^2 per tree);
P: Wetted area percentage (30%).

Irrigation was started 16th June and finished 8th September. Applied water quantities are given in Table 1.

First leaf samples were taken a week before beginning irrigation and continued until harvest season with four weeks interval.

Analysis of Chlorophyll a (Chl- a), Chlorophyll b (Chl- b), and total chlorophyll (Total chl) in leaf was performed by Arnon [2]. Calculation was done following formulas.

$$\text{Chl a} = [(0.0127 * A_{663.0}) - (0.00269 * A_{645})] * 100$$
$$\text{Chl b} = [(0.0229 * A_{645.0}) - (0.00468 * A_{663})] * 100$$
$$\text{Total chl} = [(0.0202 * A_{645.0}) - (0.00802 * A_{663})] * 100$$

The experiment was designed in a randomized complete block design with four replications. In each replication one tree was used. Statistical analyses of all data were performed with SPSS Version 16.0 (SPSS Inc., Chicago, IL, USA). Differences between the means were compared by Duncan test at a significance level of $P<0.05$.

Table 1. *Applied amount of irrigation water (L/tree)*

Irrigation	Water quantity[1]		Irrigation	Water quantity	
No	S_1	S_2	No	S_1	S_2
1	140	281	8	173	346
2	105	210	9	174	348
3	165	329	10	158	317
4	140	280	11.	188	377
5	153	306	12	127	254
6	169	337	13	115	230
7	176	352	Total	1984	3968

[1]S_1 and S_2:= 50 and 100% of the water quantity evaporating from class A pan, respectively.

RESULTS

Leaf average chl-a, chl-b and total chlorophyll were affected depend on irrigation water amount and varieties. These differences were statistically significant. Average leaf chl-a, chl-b and total chlorophyll of irrigated (50% or 100%) trees were higher than non-irrigated trees. All average leaf chlorophyll parameters of İzmir 2 pomegranate variety's trees were higher than other trees of varieties. All the other varieties were statistically similar and formed in the second group. The leaf chlorophyll content was the lowest at the beginning of growing season. The highest values were found in the end of season.

Under non-irrigation treatment, Chl-a content of İzmir 1, İzmir 2, İzmir 1499 and İzmir 1513 varieties were 5.350, 3.882, 3.610 and 3.731 mg/L at the early of season, respectively. It was increased during the summer period and reached 6.701, 7,925, 6.466 and 8.092 mg/L in the October. Especially, it increased too much in the leaf of İzmir 1513. In I_1 treatments, Chl-a content of İzmir 1, 2, 1499 and 1513 varieties were 4.629, 3.393, 3.537 and 4.310 mg/L at the beginning of season, respectively and these values increased during the season and reached 9.172, 8.659, 8.987 and 7.403 mg/L in the October. Similar changes occurred at full (I_2) irrigation treatment.

Chl-b content showed similar trend to chl-a. It was lower in the beginning of the irrigation season than end of season. The highest chl-b contents were obtained from 100% (I_2) treatments. But there were not differences between I_1 and I_2 treatments statistically. Chl-b content of İzmir 1, 2, 1499 and 1513 varieties were found as 1.526, 1.105, 1.018 and 0.977 mg/L at the beginning of season, respectively in the non-irrigation treatment. It was increased during the summer period (except İzmir 1) and reached 1.175, 1.557, 1.481 and 1.375 mg/L in the October.

In I_1 treatments, leaf chl-b contents were found as 1.309, 0.942, 1.104 and 1.223 mg/L of İzmir 1, İzmir 2, İzmir 1499 and İzmir 1513 varieties at the beginning of season, and these values increased during the season and reached 2.623, 2.474, 2.753 and 2.131 mg/L in the October. It increased too much in the leaf of İzmir 2 variety. Similar changes found at 100% (S_2) irrigation.

Table 2. *Leaf chlorophyll –a content of pomegranate varieties (mg/L)*

Irrigation	Variety	Sampling Date

		05/06	03/07	31/07	28/08	05/10	Average
	İzmir 1	5,350	5,718	6,821	5,229	6,701	**5,964**
	İzmir 2	3,882	7,700	6,563	7,366	7,925	**6,687**
0 %	İzmir 1499	3,61	3,487	3,580	5,629	6,466	**4,554**
	İzmir 1513	3,731	5,817	5,322	5,927	8,092	**5,778**
	Mean	**4,143**	**5,681**	**5,5715**	**6,038**	**7,296**	**5,746**
	İzmir 1	4,629	6,657	5,887	5,546	9,172	**6,378**
	İzmir 2	3,393	8,773	6,149	7,77	8,659	**6,949**
50 %	İzmir 1499	3,537	7,361	6,049	6,985	8,987	**6,584**
	İzmir 1513	4,31	7,531	6,453	7,514	7,403	**6,642**
	Mean	**3,967**	**7,581**	**6,135**	**6,954**	**8,555**	**6,638**
	İzmir 1	3,793	8,7	5,497	7,607	-	**6,399**
	İzmir 2	5,049	9,695	7,668	7,913	-	**7,581**
100 %	İzmir 1499	4,433	7,465	5,192	7,549	-	**6,160**
	İzmir 1513	4,892	6,881	5,62	7,616	-	**6,252**
	Mean	**4,542**	**8,185**	**5,994**	**7,671**	-	**6,598**
LSD $_{0,05\ irr}$		0,376**		LSD $_{0,05\ irr}$		0,586**	
LSD $_{0,05\ vart}$		0,530**		LSD $_{0,05\ vart}$		1,051**	
LSD $_{0,05\ irr*var}$		0,903*		LSD $_{0,05\ irr*var}$		1,186*	
LSD $_{0,05\ irr*var\ *date}$		ns					

*p<0,05 **p<0,01 ns non-significant

Table 3. *Leaf chlorophyll –b content of pomegranate varieties (mg/L)*

Irrigation	Variety	Sampling Date					
		05/06	**03/07**	**31/07**	**28/08**	**05/10**	**Average**
	İzmir 1	1,526	1,664	2,223	1,463	1,863	**1,748**
	İzmir 2	1,105	2,254	1,927	2,173	2,367	**1,965**
0 %	İzmir 1499	1,018	1,911	1,118	1,626	2,062	**1,547**
	İzmir 1513	0,977	1,762	1,534	1,733	2,286	**1,658**
	Mean	**1,157**	**1,898**	**1,7005**	**1,745**	**2,1445**	**1,730**
	İzmir 1	1,309	2,028	1,708	1,608	2,623	**1,856**
	İzmir 2	0,942	2,709	1,796	2,329	2,474	**2,050**
50 %	İzmir 1499	1,104	2,289	1,886	2,061	2,753	**2,019**
	İzmir 1513	1,223	2,272	1,876	2,169	2,131	**1,934**
	Mean	**1,1445**	**2,3245**	**1,8165**	**2,04175**	**2,495**	**1,965**
	İzmir 1	1,175	2,624	1,649	2,142	-	**1,898**
	İzmir 2	1,557	2,970	2,256	2,365	-	**2,287**
100 %	İzmir 1499	1,481	2,390	1,555	2,370	-	**1,949**
	İzmir 1513	1,375	2,144	1,592	2,327	-	**1,860**
	Mean	**1,397**	**2,532**	**1,763**	**2,301**	-	**1,998**
LSD $_{0,05\ irr}$		0,153*		LSD $_{0,05\ irr}$		0,100**	
LSD $_{0,05\ vart}$		0,171**		LSD $_{0,05\ vart}$		0,338**	
LSD $_{0,05\ irr*var}$		ns		LSD $_{0,05\ irr*var}$		ns	
LSD $_{0,05\ irr*var\ *date}$		ns					

*p<0,05 **p<0,01 ns non-significant

Table 4. *Leaf total chlorophyll content of pomegranate varieties (mg/L)*

Irrigation	Variety	Sampling Date					
		05/06	**03/07**	**31/07**	**28/08**	**05/10**	**Average**
0 %	İzmir 1	6,877	7,382	9,044	6,693	8,564	**7,712**
	İzmir 2	4,987	9,954	8,490	9,539	10,293	**8,653**
	İzmir 1499	4,628	5,398	4,698	7,255	8,528	**6,101**
	İzmir 1513	4,708	7,580	6,856	7,660	10,378	**7,436**
	Mean	**5,300**	**7,579**	**7,272**	**7,78675**	**9,441**	**7,476**
50 %	İzmir 1	5,938	8,685	7,594	7,154	11,795	**8,233**
	İzmir 2	4,335	11,483	7,945	10,100	11,133	**8,999**
	İzmir 1499	4,641	9,651	7,935	9,046	11,739	**8,602**
	İzmir 1513	5,533	9,803	8,329	9,683	9,534	**8,576**
	Mean	**5,112**	**9,905**	**7,951**	**8,996**	**11,050**	**8,603**
100 %	İzmir 1	4,968	11,324	7,146	9,749	-	**8,297**
	İzmir 2	6,607	12,664	9,924	10,278	-	**9,868**
	İzmir 1499	5,914	9,856	6,747	9,919	-	**8,109**
	İzmir 1513	6,267	9,026	7,212	9,942	-	**8,112**
	Mean	**5,939**	**10,718**	**7,757**	**9,972**	-	**8,596**
$LSD_{0,05\ irr}$		0,517**		$LSD_{0,05\ irr}$		0,762**	
$LSD_{0,05\ vart}$		0,690**		$LSD_{0,05\ vart}$		1,367**	
$LSD_{0,05\ irr*var}$		ns		$LSD_{0,05\ irr*var}$		ns	
$LSD_{0,05\ irr*var\ *date}$		ns					

*p<0,05 **p<0,01 ns non-significant

DISCUSSION AND CONCLUSION

Under drough conditions, plants reduce water loss by stomatal closure which results both in a reduction of transpiration and the inhibition of photosynthesis coupled with reducing CO_2 uptake. The amount of available metabolites required for the development of plants decreases because of decreasing photosynthesis. The photosynthetic activity of the crops is one of the important factors influencing the yield that can be observed by the measurement of physiological traits such as leaf chlorophyll content, net photosynthetic rate, and stomatal conductance. In this study, the effect of irrigation on leaf chlorophyll content was investigated. The chlorophyll content of a leaf is the indicator of a plant's physiological condition A large part of the total chlorophyll was determined to be Chl a. C*hlorophyll a*, is the primary photosynthetic pigment in green plants for the transfer of light energy to a chemical acceptor [11] and chlorophyll a was more resistant to dehydration than other chlorophyll components [14]. In higher plant leaves, chlorophyll content changes along the different stages of plant development. Thus, all chlorophyll parameters changed during the season. Chl-a, chl-b and total chlorophyll of all varieties' were low at early June. They started to increase throughout development periods and reached the highest level at the harvest period. Chlorophyll is the basic unit of plant energy systems during the photosynthesis event [4]. Besides light quality, chlorophyll production and activity are influenced by nutrition and chemical metabolites produced in the plant system. Therefore, chlorophyll amounts increased as the leaves got their full size [9]. Water defiency has an indirect effect on photosynthesis. Total chlorophyll content was higher in the irrigated trees than non-irrigated trees, in this study.

As the nutrients uptake from roots increased by watered [10], the amount of leaf chlorophyll of the irrigated trees increased. Increased uptake of Fe in these treatments also plays important role in chlorophyll formation [15]. Basiouny reported that chlorophyll content of peach leaves from irrigated trees was higher than that of tree

leaves from nonirrigated trees [3]. It was found that chlorophyll a, b and total reduced under drought stress conditions in the pear [12] and fig [1]. Furthermore, leaf chlorophyll content is closely related to plant stress and senescence. The results of this experiment showed that irrigation had effect on leaf chlorophyll content of pomegranate trees.

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