Effects of Regulated Deficit Irrigation and Superabsorbent Polymer on Fruit Yield and Quality of 'Granny Smith' Apple

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Summary

Water resource limitation has caused increasing interest in changing irrigation strategy to improve water use efficiency. In this study, effect of regulated deficit irrigation (RDI) (75 and 50% ET_c) in combination with different concentration of superabsorbent polymer (SAP) (0, 100, 200 and 300 g/tree) on fruit yield and quality of 'Granny Smith' apple was investigated during two consecutive growing seasons. The tree irrigated with 100 ETC was used as control. No significant difference was found between control and RDI and SAP treatments for apple fruit yield. But in the second year of the experiment, apple trees treated with 200 g/tree superabsorbent showed by 15% greater fruit weight. The fruit soluble solid content (SSC) was significantly affected by irrigation regimes and superabsorbent polymer. The highest SSC was found in RDI₅₀ in two years and 200 g/tree superabsorbent in the first year. In the first season, titratable acidity (TA) was significantly affected by SAP levels. The apple trees that received 300 g/tree of SAP, had the highest content of TA in the first season. No significant difference was observed among treatments regarding dry matter percentage and firmness. Total phenolic content of apple fruits was significantly affected by both RDI and SAP treatments in both seasons. In each level of SAP application, by reducing RDI percent total phenolic content increased. The highest phenolic content was found when the trees were treated with 50% RDI with and without SAP treatment.

Key words

dry matter, deficit irrigation, hydrogel, fruit quality

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Received: November 12, 2018 | Accepted: September 9, 2019

Introduction

Water deficiency is the main limiting factor for producing horticulture crops in arid and semi-arid areas in the world. Therefore, it is necessary to develop new strategies to improve water use efficiency without undesirable effect on crop yield and quality (Fereres & Soriano, 2007). Regulated deficit irrigation (RDI) could be one of the most effective strategies. In fact, RDI is the practice of reducing applied water at selected phenological stages to plants less sensitive to water deficit, and can be a practicable water saving practice for arid and semi-arid climates with the minimum impact on yield and fruit quality (Chalmers et al., 1981). This strategy is an important water-saving technique developed by Chalmers et al. (1981). The aims of RDI are to save water, control excessive vegetative growth, and improve or maintain yield and fruit quality. Girona et al. (2005) show that a single RDI regime reduced irrigation by 13-24%, while combined regimes reduced it by 23-35%. Dos Santos et al. (2007) reported that RDI saved irrigation water by 50% in grapevines. Furthermore, the study also showed that RDI improved grape berry quality (Munitz et al., 2017). They found that full irrigation from flowering to bunch closure and lower irrigation from bunch closure to harvest had the potential to generate the best balance between vegetative growth, high yield and berry quality with enhanced color and aroma compounds. Withholding irrigation from 120 to 150 days after flower bloom not only increased in fruit SSC, soluble sugars and dry matter content but also resulted in an increase in water use efficiency (Cheng et al., 2012). Recently, Galindo et al. (2017) have shown that a short period of irrigation restriction at the end of ripening period brings the harvest time forward, saves water, enhances fruit bioactive compounds (anthocyanins, phenolic compounds, punicalagin and ellagic acid) and increases the price of the fruit without affecting marketable quality.

The application of superabsorbent polymers (SAPs) or hydrogel could be another strategy to save water in arid and semi-arid climate (Zangooinasab et al., 2012). Superabsorbent polymers that are chemical compounds derived from petrol oil are used in order to increase water and fertilizer use efficiency in dry conditions (Hashembeig Mahalati, 2008). They found that hydrophilic gel material having a three dimensional structure of the polymer chain had high capacity to absorb and maintain water. They can take up to hundreds times more than their weight, and release it gradually to plants under water deficiency condition. Furthermore, incorporation of SAPs to soil improves physical property and water-availability capacity that results in increasing plant growth and yield and reduces crop irrigation demand (Yazdani et al, 2007). Zhang et al (2012) reported that SAP could fully conserve water and increase crop yield and quality when in combination with proper irrigation. In "Grand Nain" Banana plants showed that application of SAPs increased bunch weight and fruit yield under the water regime of (7000 m³/ha per year) 87.5% of recommended amount of irrigation (Kassim et al., 2017). Liu et al. (2016) reported that superabsorbent polymers increased total dry weight, chlorophyll and soluble sugar in leaves of coffee tree. Overall, the objectives of current study are to evaluate the effect of RDI and SAPs alone or in combination on fruit yield and quality of 'Granny Smith' apple.

Materials and Methods

Experimental Conditions and Plant Materials

The experiment was performed during 2015 and 2016 at a commercial apple orchard located in Alborz province, Iran (36°N, 50.31 °Wand 1200M). The apple trees (Malus domestica Borkh) cultivar Granny Smith were 6 years old and grafted on seedling rootstocks which were planted in a 4×4.5 m space. The trees were drip irrigated using two drip irrigation lines for each row and four emitters per tree. Tree irrigation requirement was scheduled weekly according to daily ETo, calculated using the Penman-Monteith equation (Allen et al., 1998). The average thirty- year meteorological data are summarized in Table 1. The experimental design was complete block design with three replications. The standard plot was made up of five trees. The three central trees were used for evaluation and two other trees were used as guard trees. The soil properties are also summarized in Table 2.

Treatments

Two regulated deficit irrigation (RDI) strategies were compared with full irrigation (Control). The control tree received full irrigation over fruit growth and development (ETc 100), and 75% ETc and 50% ETc. RDI treatments were applied over fruit growth and development to harvest.

Meteorological data were obtained from local synoptic weather station near the orchard. Irrigation scheduling was set based on holding capacity of soil for unavailable moisture and defined treatments using following relations (Equation 1 and 2). Therefore, the control received full irrigation over fruit growth and development season (32 L/tree per hour) and 75%, and 50% of full irrigation water for treatments.

$$In = [(F_c - PWP / 100) \times \rho_b \times Dr \times F]$$

$$I_i = In / [K \times (ETc)]$$

In: Net irrigation water content (mm), F_c : Field capacity (%), *PWP*: Permanent wilting point (%), ρb : Soil bulk density (gr·cm⁻³), Dr effective depth of root (120 cm) I: Irrigation interval (day), K: Coefficient of plant, ETc: Plant water requirement.

The superabsorbent polymer (8.5 g·L-1 mass density with pH 7.5) was obtained from Tarava Company, Esfehan, Iran and incorporated (0, 100, 200 and 300 g/tree) with soil just under dripline into 30 cm deep. The superabsorbent used in this study was a potassium base that does not cause salinity in the soil after decomposition. In 100% regulated deficit irrigation, the superabsorbent was not used, but at others 75 and 50%, four different levels of superabsorbent were used.

Fruit Yield and Quality Analysis

In both years of the study the fruits were harvested at the commercial maturity stage, which was almost coincident with the middle of September. The fruit samples were harvested from four branches at each side of tree canopy. The remaining fruits were weighed to determine total yield. Finally, the fruit yield was reported as kg/tree.

Average of monthly temperature (°C)	Monthly rainfall (mm)	Relative humidity (%)	Wind Speed (m/s)	Months
14.2	39.1	55	2.5	April
19.2	19.5	49	3.3	May
24.6	2.7	35	3.0	June
27.1	.1 3		3.5	July
26.8	1.2	19	3.9	August
22.9	1.6	24	3.4	September
17.1	15.1	30	3.2	October
9.9	27.7	42	3.1	November
4.6	33.5	54	3.8	December
1.8	30.8	68	3.6	January
4.1	32.1	61	2.7	February
8.7	45.4	58	2.6	March

Table 2. Some physical and chemical properties of soil

	1 1	
Property	Unit	Value
рН	-	8.1
EC	ds/m	1.31
OM	%	2
N	%	0.2
P	ppm	73.8
K	ppm	1478
Fe	mg/kg	5.01
Zn	mg/kg	1.8
Mn	mg/kg	7
Tissue	-	Clay Loam

At harvest, randomly 10 apple fruits were evaluated for flesh firmness, titratable acidity (TA), and soluble solids concentration (SSC). Flesh firmness was measured at two opposite points on equatorial side using a penetrometer with an 8 mm diameter probe. SSC was determined with a digital refractometer and TA was measured by titration of juice with 0.1 M NaOH and phenolphthalein as an indicator.

Extraction of phenolic compounds was performed according to the method described by D' Angelo et al. (2007). Briefly, 0.5 grams of fruit sample were powdered in mortar and macerated overnight in methanol: acetic acid (85:15 v/v) solution at 4°C.

Then, samples were centrifuged for 10 minutes at 10,000 rpm. It was evaluated using a colorimetric method described previously by D' Angelo et al. (2007). The method involves the reduction of Folin-Ciocalteau reagent by phenolic compounds, with a concomitant formation of a blue complex. In this study, 70 μL of the extract was mixed with 130 μL of distilled water and 1 mL of Folin-Ciocalteau reagent diluted 10 times with distilled water. After 5 minutes, 800 μl of sodium carbonate (7.5 %, w/v) was added. Then, the mixture was incubated for 1.5 hours at room temperature, and the absorbance was read at 760 nm using an UV-Vis spectrophotometer (Unicam He Cambridge, UK). The measurement was compared to a standard curve prepared with gallic acid solution. The total phenolic content was expressed as mg of gallic acid equivalents per gram of fresh weight (mg GAE g-¹ FW).

The percentage of antioxidant activity of each substance was assessed by DPPH free radical assay (D' Angelo et al., 2007).

Statistical Analysis

The analysis of variance was performed on all fruit data as a factorial design. All statistical analyses were undertaken using the general linear model (GLM) procedure of the SAS version 9.0. The Duncan multiple range test ($P \le 0.01$) was used to evaluate differences between treatments.

Results and Discussion

Fruit Yield

The results showed that fruit yield of apple trees was not significantly affected by RDI and SAP treatments during two growing seasons (Table 3).

Table 3. The analysis variance of regulated deficit irrigation (RDI) and superabsorbent polymer (SAP) on fruit yield and quality of Granny Smith apple during two growing years

Source DI	DE	Mean Square							
	DF	Fruit Yield	Fruits Weight	Dry Matter	SSC	TA	Firmness	Total Phenolics	Antioxidant Activity
2015									
Block	2	44.69 ^{ns}	1903.1*	$0.05^{\rm ns}$	4.93 ^{ns}	1.52**	0.78*	$0.004^{ m ns}$	5.51 ^{ns}
SAP	3	$3.29^{\rm ns}$	328.1 ^{ns}	$0.15^{\rm ns}$	4.03*	0.017*	0.08^{ns}	0.001^{ns}	155.14 ^{ns}
RDI	2	1.86 ^{ns}	144.5 ^{ns}	2.57 ^{ns}	5.18*	$0.005^{\rm ns}$	0.05^{ns}	$0.0004^{ m ns}$	55.14 ^{ns}
SAP×RDI	6	$10.82^{\rm ns}$	917.7 ^{ns}	1.03 ^{ns}	1.19 ^{ns}	0.007^{ns}	$0.11^{\rm ns}$	0.013*	86.95 ^{ns}
CV		17.77	16.04	7.61	8.38	13.71	7.73	18.14	23.85
2016									
Block	2	450.8 ^{ns}	1709.6*	5.95 ^{ns}	5.97*	0.20**	$0.39^{\rm ns}$	0.01 ^{ns}	426.09 ^{ns}
SAP	3	29.7 ^{ns}	1023.7*	1.89 ^{ns}	1.03 ^{ns}	$0.007^{\rm ns}$	$0.11^{\rm ns}$	0.28 ^{ns}	298.09 ^{ns}
RDI	2	16.86 ^{ns}	916.7 ^{ns}	3.3 ^{ns}	4.82*	$0.007^{\rm ns}$	$0.49^{\rm ns}$	0.001^{ns}	422.56 ^{ns}
SAP×RDI	6	$30.78^{\rm ns}$	170.5 ^{ns}	12.4 ^{ns}	$0.54^{\rm ns}$	0.015^{ns}	$0.36^{\rm ns}$	1.27*	67.54 ^{ns}
CV		23.53	15.1	21.7	5.52	9.29	8.59	19.24	24.52

^{**:} Significant differences at 1% level, *: Significant differences at 5% level, ns: Not significant

These results are in agreement with findings from a previuos study which showed that fruit yield of apple and peach trees wasn't significantly affected by RDI while it saved 17-20% of water (Huang et al., 2001). Another study on pear fruits showed that controlled soil drying reduced shoot growth by 15-25%, and the fruit diameter and fresh weight were not affected by deficit applied during the fruit growth stage. In agreement with this study, Cheng et al (2003) also found that RDI did not effect fruit size and yield.

Unlike the finding of this experiment, Sultana et al (2016) found that when SAP mixed with soil, fruit yield of tomato plants increased. El-Hady and Wanas (2006) reported that SAP reduced watering frequency of container or field grown crops. It also reduced irrigation content from 100 to 85% of the crop water requirements and increased crop yield. But in this study, application of SAP had no significant effect on fruit yield of apple trees (Table 3).

Fruits Weight

No significant difference was found between RDI treatments and control regarding fruit weight in two growing season, 2015 and 2016 (Table 3). In agreement with these results, Cheng et al. (2012) also reported that different water stress regimes had no significant effect on trees' yield and individual fruit weight, but the application of superabsorbent increased fruit weight compared to control samples in the second year (Table 3). Application of 200 g/tree SAP significantly increased fruit weight by 15% compared to control (Fig1).

The previous study confirmed that SAP application improved soil physical properties, water penetration rate, water-holding

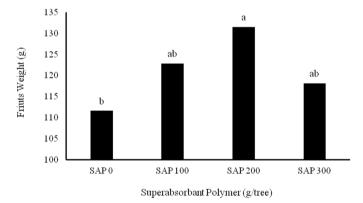


Figure 1. Effect of superabsorbent polymer (SAP) on apple fruits weight in the second year. Values with the different letters are significantly different according to Duncan's Multiple Range Test at P<0.05

capacity and available water (Abedi- Koupai and Sohrab, 2004). However, it released nutrients slowly, which increased nutrient absorption, osmotic moisture of soil and caused a decrease in transplanting stresses that caused an improvement in plant growth reaction and increase in yield.

Fruit Dry Matter

The results showed that RDI and SAP alone or in combination with each other didn't significantly effect fruit dry matter in 2015 and 5016 (Table 3). These results are in agreement with Teng et al (1999) that showed water stress could reduce fruit size and fresh weight but had no effect on the dry weight of pear fruits.

However, Marsal et al (2008) reported that irrigation regimes had significant effects on dry matter of pear fruits. Berman and Dejong (1996) showed that water stress could potentially inhibit fruit dry weight growth as a result of both sink and source limitations. Sink limitations to fruit growth could occur if fruit cell growth is expansive and carbohydrate accumulation processes are sensitive to water status, while source limitations could occur when water stress reduces photosynthesis and restricts the supply of assimilates. Therefore, it seems the RDI treatments in this study were not great enough to affect the sink strength of fruits, thus no significant reduction in dry weight was observed relative to wellwatered controls.

Soluble Solids Content (SSC)

The results of this study showed a significant difference between SAP treatments on fruit SSC in 2015. However, in the second year of experiment, no significant difference was found between SAP treatments. In contrast, RDI treatment significantly affected SSC in both consecutive years (Table 3).

The results of the first year of the study showed that increasing SAP level to 200 g/tree significantly increased SSC but thereafter it decreased (Fig 2). The Granny Smith apple tree treated to SAP_{200} showed the highest fruit SSC (Fig 2).

With increasing water availability in root zone fruit SSC increased. Therefore, the highest SSC was found in RDI₅₀ in the both consecutive seasons (Fig 3 and 4). The RDI₅₀ increased apple fruit SSC by 17.01% and 15.26% in 2015 and 2016 respectively as compared to control.

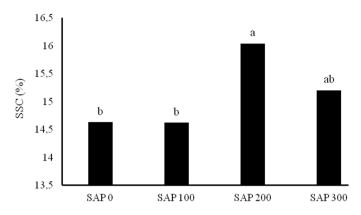
In agreement with this finding, different studies also showed that RDI treatments increased SSC of pear and apple fruits (Teng et al., 1999; Cheng et al., 2012).

Titratable Acidity (TA)

As the analysis of variance showed, the effect of SPA treatment was significant on TA content of Granny Smith apple fruit in the first year. No significant difference was found among RDI on TA in both seasons (Table 3).

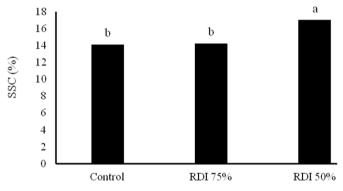
In agreement with this finding, Mpelasoka and Behboudian (2002) found that reduced irrigation on maturity parameters of 'Braeburn' apple expressed that fruit TA was not affected by irrigation. Pérez-Sarmiento et al., (2016) demonstrated that no differences were found in pH and titratable acidity. In contrast, RDI strategies increased significantly TA and SSC in apricot fruits RDI (Pérez-Pastor et al., 2014), whereas Ebel and Proebsting (1993) found that at harvest time, RDI treated fruits were smaller and had a higher SSC and lower TA.

The mean comparison showed that TA in apple fruits was significantly affected by SAP. Similarly, Sendur Kumaran et al. (2001) evaluated the influence of superabsorbent polymers on processing quality of tomato and observed that polymers influenced the quality parameters such as TA of fruits. As the results showed, the highest TA was found when apple trees received 300 g SAP (Fig 5).



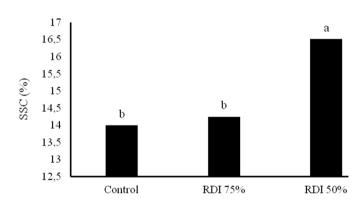
Superabsorbant Polymer (g/tree)

Figure 2. Effect of superabsorbent polymer (SAP) on apple fruits SSC in the first year. Values with the different letters are significantly different according to Duncan's Multiple Range Test at P<0.05



Regulated Deficit Irrigation

Figure 3. Effect of regulated deficit irrigation (RDI) on apple fruits SSC in the first year. Values with the different letters are significantly different according to Duncan's Multiple Range Test at P<0.05



Regulated Deficit Irrigation

Figure 4. Effect of regulated deficit irrigation (RDI) on apple fruits SSC in the second year. Values with the different letters are significantly different according to Duncan's Multiple Range Test at P<0.05

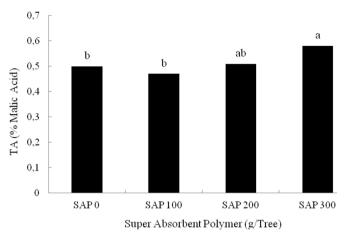


Figure 5. Effect of superabsorbent polymer (SAP) on apple fruits TA in the first year. Values with the different letters are significantly different according to Duncan's Multiple Range Test at P<0.05

Fruit Firmness

Tissue firmness of apple fruits was not significantly affected by RDI and SAP treatments in two consecutive growing seasons (Table 3).

In agreement with these findings, Ebel and Proebsting, (1993) found that apple fruit firmness was not affected by RDI treatments, whereas Mpelasoka and Behboudian (2002) found apple firmness increased in DI (deficit irrigation) fruits. Furthermore, withholding irrigation sometime later in the season could increase total soluble solids and firmness in comparison with fully watered control (Kilili et al., 1996). Mpelasoka et al., (2000) reported that both EDI (early deficit irrigated) and LDI (late deficit irrigated) increased flesh firmness in apple fruits.

Total Phenolic Content

Total phenolic content of apple fruits was significantly affected by RDI and SAP interaction in both seasons (Table 3). In the first year, the highest phenolic content was found when apple trees were treated with in 100 and 200 gram SAP and irrigated with RDI₅₀ (Fig 6).

In the second year of experiment, the highest fruit phenolic content was found in RDI₅₀ with application of SAP (Fig 7).

In grapevine, it has been shown that periods of water deficit increased phenolic content in the berry skins (Roby et al., 2004). In general, drought stress is known to trigger the plant defense systems resulting in the enhanced production of secondary metabolites (Caliskan et al., 2017). Atkinson et al (2011) demonstrated that content of total phenolics for the cherry fruits under the water stress conditions was considerably higher than control. It has been hypothesized that the activity of phenylalanine ammonia-lyase (PAL), a key enzyme in the biosynthetic pathway of phenolic compounds, is directly involved in the accumulation of polyphenols (Rinaldi et al., 2011). Water stress induces the gene expression and the enzyme activity of PAL, the primary enzyme of phenylpropanoid pathway that is responsible for the biosynthesis of phenolic compounds. Sultana et al., (2016) reported that the total phenol content as the functional antioxidant marker was

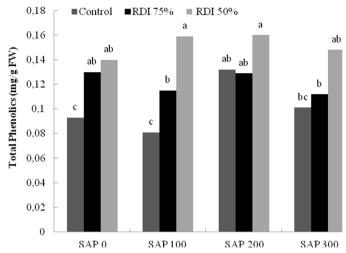


Figure 6. Effect of regulated deficit irrigation (RDI) and super absorbent polymer (SAP) on total phenolic content of apple fruits in the first year. Values with the different letters are significantly different according to Duncan's Multiple Range Test at P<0.05

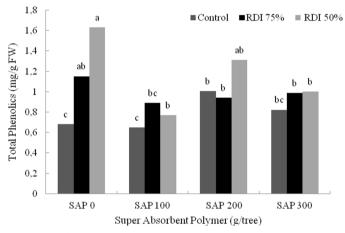


Figure 7. Effect of regulated deficit irrigation (RDI) and super absorbent polymer (SAP) on total phenolics of apple fruits in the second year. Values with the different letters are significantly different according to Duncan's Multiple Range Test at P<0.05

increased in tomato planted with SAP application (3.25 mg/100g) compared to control sample.

Antioxidant Activity

The antioxidant activity of apple fruits was not significantly affected by both RDI and SPA treatments alone or in combination effect (Table 3). In spite of the increase of total phenolic content in RDI and SAP treatments, antioxidant capacity was not significantly affected by these treatments. Previous studies showed that the plants growing under stress conditions react by increasing their antioxidant production from both non-enzymatic systems (e.g., flavonoids, phenolic compounds, vitamins C and E, and carotenoids) and enzymatic systems (e.g., superoxide dismutase, glutathione reductase, catalase, and several peroxidases) (Buendia et al., 2008). They also found that the effect of RDI on different antioxidant compounds was variable.

Conclusion

The results of the current study showed that fruit yield of Granny Smith apple was not significantly affected by RDI and SAP treatments. However, application of superabsorbent polymer with 200 g/tree SAP increased fruit weight by 15% as compared to control. Fruit dry matter and fruit firmness weren't affected by RDI and SAP alone or combination treatment. Furthermore, apple trees treated with RDI significantly showed higher SSC in both consecutive years, but SAP could increase fruit SSC just in the second year of experiment. It seems that SAP increased TA content by improving tree water availability. Total phenolic content of apple fruit was significantly affected by RDI and SAP in both seasons. However, the antioxidant activity of apple fruits was not significantly affected by both RDI and SPA treatments alone or in combination effect.

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