

Application of Superabsorbent and Mulch on Some Traits of African Marigold (*Tagetes erecta*) under Irrigation Intervals

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Water scarcity is an important obstacle to the development and expansion of the urban landscape in the arid and semi-arid regions of the world. Nowadays, organic and artificial materials can be used to reduce water consumption without impairing the quality and quantity of urban landscape. In order to study the effect of mulch type on reducing irrigation interval for a popular seasonal plant in the Middle East, African marigold, an experiment was carried out in Mashhad, Iran during 2016-2017 in which two treatments were applied including mulch type (typical soil as control, palm fiber mulch, plastic mulch, and superabsorbent) and irrigation intervals (3, 6 and 9 days). Some morphological, biological, physiological and biochemical traits were evaluated. Results indicated that the superabsorbent could significantly increase most recorded traits. The palm fiber mulch also improved plant height, flower number, and dry shoot weight. In most traits, the 3-day irrigation interval indicated a significant increase versus the 6- and 9-day intervals, but plant height, flower number, and photosynthetic rate did not differ significantly between the 3-day and 9-day irrigation intervals. The findings show that superabsorbent and at the next level, palm fiber mulch with 9-day irrigation intervals can be suggested for African marigold in the arid and semi-arid climates.

Abstract

Keywords: Arid and semi-arid regions, Irrigation interval, Palm fiber mulch, Seasonal plants, Superabsorbent.

INTRODUCTION

Climate change is the biggest challenge for mankind in the 21st century. In the recent decades, vast areas of the world, especially Middle Eastern countries, have been struggling with severe water shortages due to temperature rise, precipitation decline, and the increased rate of evaporation (Saleh, 2010). In addition to the climate-related problems, other factors, such as population growth, urban development, water pollution, and excessive use of energy resources have contributed to water scarcity. Some studies show that in the present more than 50 percent of the world's population lives in urban areas, which is expected to increase by 66% by 2050.

Although, living in a green, beautiful and healthy city is an important citizenship right, the development of urban landscapes such as parks and public gardens faces many problems due to the lack of water resources in many urban areas. Evidence suggests that in arid and semi-arid regions of the world, a significant proportion of drinkable water is used to irrigate the urban landscapes. Therefore, to provide long-term water conservation, it is crucially important to design landscapes that consume less water than traditional landscapes (Zollinger *et al.*, 2006). However, the water requirements and the response of many ornamental plants to drought stress are still not well defined in landscapes (Mee *et al.*, 2003).

One of the most effective methods of water saving is the use of mulch in agriculture and urban landscapes. Mulch refers to a layer of material applied to the surface of soil to conserve its moisture, improve its fertility and health, reduce the growth of weeds, and enhance the visual attractiveness of the area. Mulch is used to produce both commercial and horticultural products, and when applied properly, they can significantly improve soil fertility (Brickell, 2003). Studies have indicated that organic and inorganic mulches can control weeds and regulate soil temperature (Skroch *et al.*, 1992). Sawdust mulch improved the growth of two marigold cultivars (Pakdel *et al.*, 2015), and polyethylene mulch contributed to maintaining soil moisture and increasing water use efficiency in black saxaul seedlings (Tahan *et al.*, 2015).

Another effective method to conserve water in agriculture and urban landscapes is to apply the new chemical polymers such as superabsorbent polymers (SAPs). SAPs have special properties owing to their three-dimensional structure. These polymers contain hydrophilic groups that can absorb liquids (may absorb probably 300 times more) and improve water conservation in soils (Omidian *et al.*, 2005). Since SAPs can increase water use efficiency and alleviate the effects of drought stress on plants, proper use of them in dry and semi-dry regions is beneficial (Bakass *et al.*, 2002). Positive effects of SAPs on drought stress reduction in marigold (Dehbashi *et al.*, 2014) and sports turf (Sheikmoradi *et al.*, 2009) have been investigated.

The use of irrigation intervals with mulches and superabsorbent has been investigated by some practitioners. One study indicated that the application of superabsorbent with increased irrigation interval in sweet peppers reduced the growth, yield, chlorophyll content, and relative water content while total soluble solid (TSS) and proline content were increased (Sayyari and Ghanbari, 2012). An irrigation interval of 10 days with black and white PVC mulch could improve the quantity and quality of *Gladiolus* production (Fakhraei Lahiji *et al.*, 2012). A study on rose plants also showed that when superabsorbent was applied, irrigation interval could be increased without any adverse impacts on the growth of roses (Jalili *et al.*, 2013).

In the present study, an attempt is made to investigate the negative effects of water stress on marigold, along with an increase in irrigation intervals in order to reduce water use by using different types of mulch and superabsorbent.

MATERIALS AND METHODS

The study was conducted at the Research Center for Plant Science, Ferdowsi University of Mashhad, Iran from the winter of 2016 to the summer of 2017. To evaluate different types of mulch and irrigation intervals on some traits of marigold (*Tagetes erecta*) as an ornamental plant in the landscape, a cultivar of marigold namely 'Taishan' was prepared. The experiment was factorial

based on a completely randomized design with four replications. The factors included mulch at four levels (typical soil as control, palm fiber mulch, plastic mulch, and superabsorbent) and irrigation interval at the levels of 3, 6 and 9 days. The physical and chemical characteristics of the superabsorbent are presented in Table 1.

Table 1. The physical and chemical characteristics of superabsorbent applied in the study.

Characteristics	Value
Appearance	Powder/Brown
Humidity	Less than 5%
Toxicity and smell	No
Density (g/cm ³)	0.8
pH (solution)	6-7
Particle size (µm)	500-800
Durability in the soil (year)	7-5
Water absorbent capacity (g/g)	380

In the end of experiment some morphological traits (plant height, number of flowers per plant, flower diameter, shoot fresh weight, and shoot dry weight), biological traits (flowering date and flowering period), physiological traits (photosynthesis rate and relative water content) and biochemical traits (proline and chlorophyll content) were recorded.

Morphological traits

Plant height (PH) was measured from the soil level to the tip of the plant by a ruler in cm. Since cv. 'Taishan' consists of several flowers per plant, after flowering, the number of flowers (NF) was counted. At the end of the complete growth of flowers, flower diameter (FD) was measured using a caliper in cm. After the end of the experiments and plant growth period, the bushes were removed from the soil. Then, their shoots and roots were separated. Next, fresh shoots and roots were placed inside paper envelopes and oven-dried at 70°C for 48 hours. The dried shoots (DS) and dried roots (DR) were weighed with digital balance with an accuracy of 0.01 g in grams.

Biological traits

Flowering period was calculated from the first day of flower opening until the emergence of flower wilting symptoms. The flowering date was recorded from the opening of the first flower per plant.

Physiological traits

To calculate relative water content (RWC) and leaf fresh weight (FW) of samples were determined. Then, they were placed in distilled water at 4°C in the dark for 24 h to rehydrate, and the turgid weight (TW) was determined. The leaves were oven-dried at 75°C for 24 h, and the dry weight (DW) was determined. RWC was calculated using the following formula (Silveira *et al.*, 2003):

$$RWC = (FW - DW) / (TW - DW) \times 100.$$

The photosynthesis rate was measured by a portable photosynthesis system (LCA- 4) using the expanded and young leaves during the growth period of the marigold.

Biochemical traits

To calculate chlorophyll *a* and *b* and carotenoids, 100 mg of the fresh leaf was homogenized

in liquid nitrogen in a microtube by an electrical homogenizer. Then, 1 ml of ethanol was added to the sample as a solvent. The samples were kept in the dark and cold conditions for 24 h and then, they were mixed by vortex for 5 minutes. The extract was centrifuged at 3000 rpm for 5 min. The supernatant was diluted 10 times with methanol. The absorbance was read in a spectrophotometer at 470, 664 and 648 nm (Dere *et al.*, 1998). Chlorophylls and carotenoid contents were calculated using the following equations:

$$(1) \text{Chl a } (\mu\text{g/ml}) = 13.36 \times A_{648} - 5.19 \times A_{664}$$

$$(2) \text{Chl b } (\mu\text{g/ml}) = A_{648} \times 27.43 - 8.12 \times A_{664}$$

$$(3) \text{Chl (total)} = \text{Chl a} + \text{Chl b}$$

$$(4) \text{Chl a / Chl b} = \text{Chl a} / \text{Chl b}$$

To measure proline, samples of fresh leaves were weighed (0.1 g) and homogenized in liquid nitrogen in a microtube by an electrical homogenizer. Afterward, 1 ml of 3% sulphosalicylic acid was added to the sample as a solvent. Then, the extract was centrifuged at 3500 g for 10 min and the supernatant was kept at 4°C to determine the proline content. An aliquot of this supernatant was taken and after adding reactive ninhydrin acid reagent (ninhydrin, phosphoric acid 6 M, glacial acetic acid) and glacial acetic acid at 99%, it was placed in a water bath at 100°C for 30 min. Soon after the removal from the water bath, the microtubes were cooled in an ice bath. The absorbance was read in a spectrophotometer at 520 nm as indicated by Bates *et al.* (1973). The proline content in fresh tissue was calculated by comparing the sample absorbencies with the standard proline curve in a concentration range of 0 to 25 mg/L. The result of proline concentration was expressed in $\mu\text{g/g}$ FW (fresh weight).

The data were subjected to the analysis of variance (ANOVA) and mean values were compared with Duncan's multiple range test ($P < 0.05$) using the Minitab 17 software package.

RESULTS AND DISCUSSION

Morphological traits

As can be seen in Table 2, mulch type, irrigation interval, and their interaction significantly affected plant height, the number of flowers and dry shoot weight (at least $P < 0.05$). Root dry weight was significantly different only among irrigation intervals ($P < 0.01$). The maximum plant height (25.3 cm) was recorded in the superabsorbent that had no significant difference with the palm fiber. The lowest plant height was obtained in the 6-day irrigation interval that indicated a significant decrease versus the 3-day (17.5%) and 9-day (12%) irrigation intervals. The highest plant height was observed in the plants treated with the superabsorbent under all irrigation intervals (Fig. 1).

Table 2. Means squares of the morphological traits of the marigold treated with various mulch types and irrigation intervals.

SoV	df	Plant height	Flower number	Flower diameter	Shoot dry weight	Root dry weight
Mulch	3	26.82**	23.44*	0.21 ^{ns}	80.41**	1.53 ^{ns}
Irrigation	2	80.25**	95.06**	0.90 ^{ns}	179.16**	21.33**
Mulch \times irrigation	6	16.57**	21.50*	0.57 ^{ns}	64.12**	2.49 ^{ns}
Error	36	3.76	6.57	0.45	18.11	1.10
Total	47	-	-	-	-	-
Standard deviation	-	3.18	3.65	0.68	5.90	1.47
CV (%)	-	13.52	60.83	10.98	43.76	62.95

*, ** and ns show significance at the 5% and 1% probability levels and insignificance according to Duncan's test, respectively.

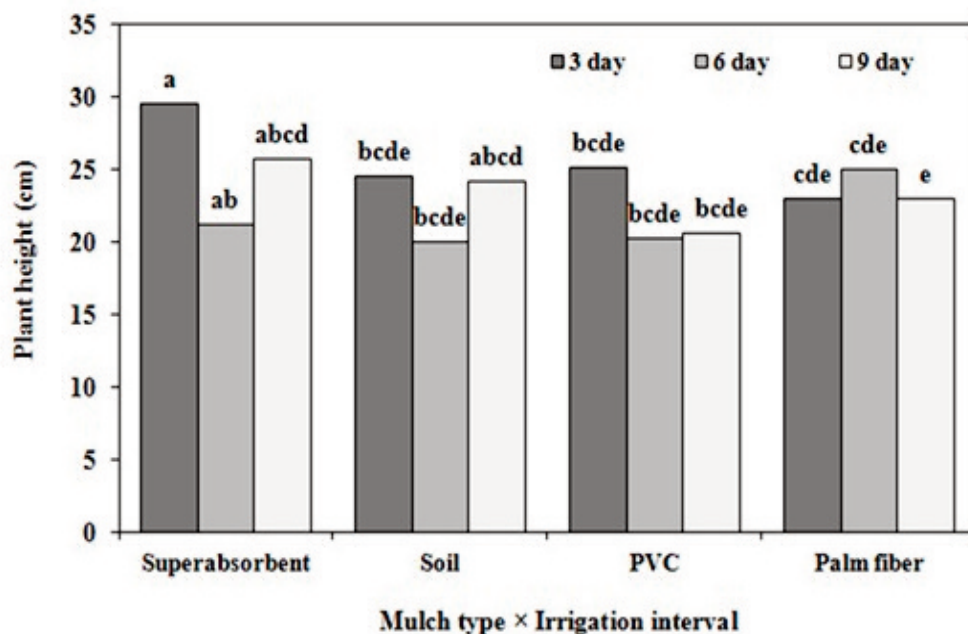


Fig. 1. The interactive effect of mulch type and irrigation interval on the plant height of the marigold.

The most and least number of flowers were recorded in the superabsorbent (7.42) and PVC (4.1) treatments, respectively. Among the mulch type treatments, a significant difference in flower number was only recorded between the superabsorbent and PVC. The highest flower number was obtained from the 3-day irrigation interval in which it was significantly higher than that of the 6-day (49 %) and 9-day (47 %) irrigation intervals. The interaction of the mulch type and the irrigation interval was complex for the flower number (Fig. 2).

The lowest shoot dry weight was recorded in PVC (10.14 g) although this treatment had no significant difference from the soil. The 3-day irrigation interval with the highest shoot dry weight (17.3 g) and the highest root dry weight (3.67 g) indicated a significant difference from the 6-day and 9-day irrigation intervals. In most interactions between the mulch type and the irrigation interval, there was no significant difference (Fig. 3).

Water stress generally affects plant growth and development negatively and it can reduce plant height by reducing cell enlargement and division (Manivannan *et al.*, 2007; Yazdani *et al.*, 2007). Superabsorbents improve the growth of roots by improving the physical conditions of substrates, thereby increasing plant height. Some studies have confirmed the positive effect of superabsorbent on the height of various plants (Esfandiari *et al.*, 2009).

However, it has been documented that different drought stress conditions may have different effects on flowering although drought at flowering commonly results in barrenness (Lichtfouse *et al.*, 2009; Wu *et al.*, 2017). Jaimez *et al.* (2000) and Bissuel-Belaygue *et al.* (2002) reported that drought stress significantly affected flower characteristics (the number of aborted flowers, bulb inflorescence length, and the number of floral buds) in some ornamental plants. The closure of stomata to prevent transpirational water loss is the preliminary plant response to water stress that results in the decline of photosynthesis rate (Mahajan and Tuteja, 2005; Mansfield and Atkinson, 1990). The decline in the photosynthesis rate will subsequently reduce flower formation and number (Salehi and Bahadoran, 2015).

Drought stress can decrease plant dry weight by reducing the available water content, thereby decreasing leaf area and number as well as plant height (Ahmadi Azar *et al.*, 2015). Some studies have indicated that the incorporation of superabsorbent polymer into the soil improves total dry weight in plants (El-Hady *et al.*, 2002; Liu *et al.*, 2013).

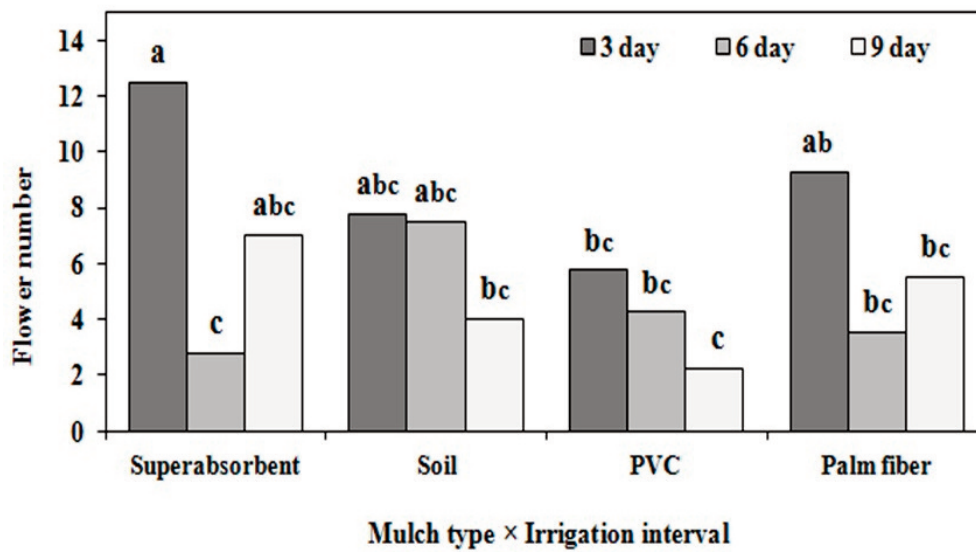


Fig. 2. The interactive effect of the mulch type and irrigation interval on the flower number of the marigold.

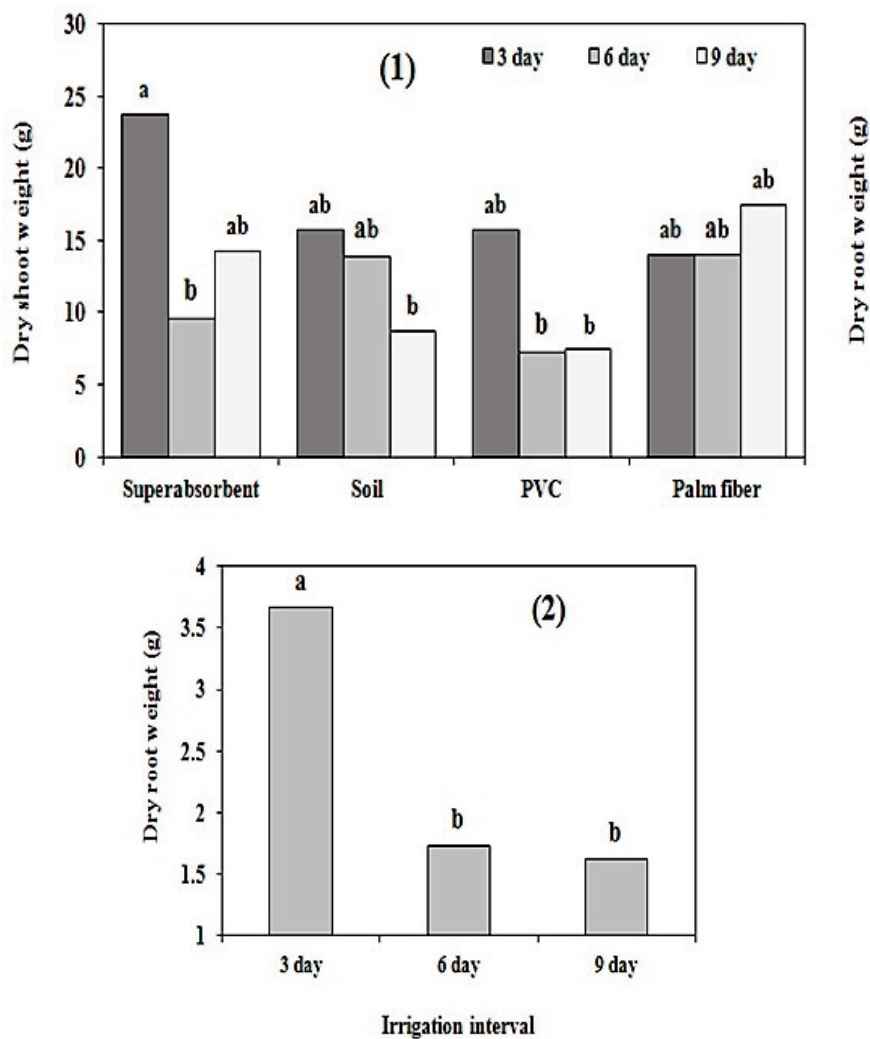


Fig. 3. The interactive effect of the mulch type and irrigation interval on shoot dry weight (1) and the effect of irrigation interval on root dry weight of the marigold.

Biological traits

The results indicated that out of the two recorded biological traits, the flowering period was significantly affected by only the irrigation interval ($P < 0.01$) (Table 3). As depicted in Fig. 4, the longest flower period was obtained from the 3-day irrigation interval (13.1 days) although this treatment was not significantly different from that of the 9-day irrigation interval (12.4 days).

It seems that water stress can shorten the flowering period due to the reduction of photosynthesis and consequently the decline of leaf area and the transfer of assimilated compounds to flowers (Jafarzadeh *et al.*, 2013). However, a significant difference was only observed between the 3-day and 6-day irrigation intervals in the present study.

Table 3. Means squares of biological traits of the marigold plants treated with various mulch types and irrigation intervals.

SoV	df	Flowering period	Flowering date
Mulch	3	1.41 ^{ns}	18.05 ^{ns}
Irrigation	2	5.65 ^{**}	1.02 ^{ns}
Mulch × irrigation	6	1.32 ^{ns}	14.66 ^{ns}
Error	36	1.01	11.76
Total	47	-	-
Standard deviation	-	1.13	2.17
CV (%)	-	9.07	23.18

*, ** and ns show significance at the 5% and 1% probability levels and insignificance according to Duncan's test, respectively.

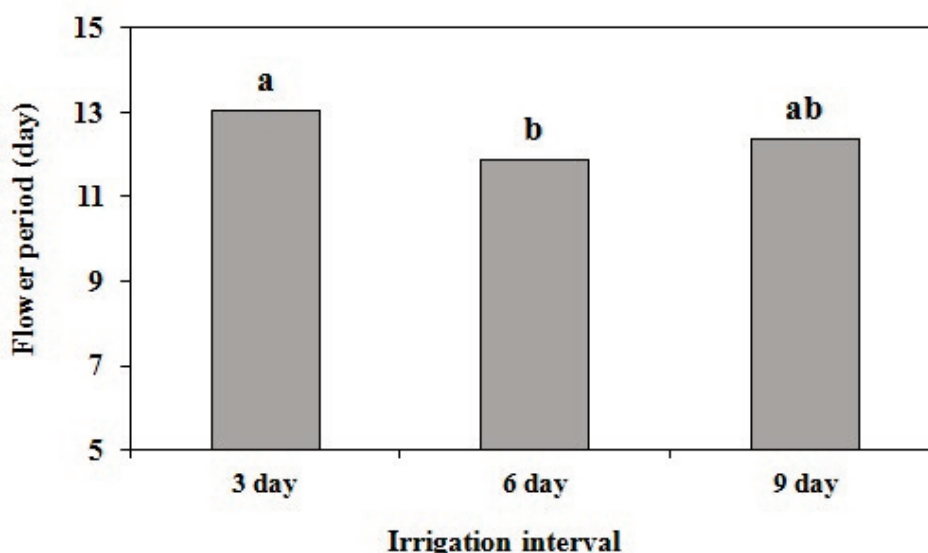


Fig. 4. The effect of the mulch type, irrigation interval and their interaction on the flowering period of marigold.

Physiological and biochemical traits

According to Table 4, total chlorophyll was affected by the irrigation interval ($P < 0.05$), RWC by the mulch type ($P < 0.01$) and photosynthesis by the irrigation interval and their interaction ($P < 0.01$) significantly. The highest chlorophyll content (4.156 mg/g) was recorded in the 6-day ir-

rigation interval, but it did not differ significantly from that of the 3-day irrigation interval. The highest photosynthesis rate was obtained from the 3-day and 9-day irrigation intervals in that they were significantly higher than that of the 6-day irrigation interval (47 and 45%, respectively). In the interaction of mulch type and irrigation interval, the heights photosynthesis rate was recorded in the superabsorbent and 9-day irrigation interval ($38.12 \mu\text{mol m}^{-2} \text{s}^{-1}$) although it did not show any significant difference from that of some interactions. The highest RWC was observed in the superabsorbent (74.1%) that which significantly different from the other mulch types (Fig. 5).

The chlorophyll level in living plants is one of the important factors in maintaining photosynthetic capacity (Jiang and Hung, 2001). The results of this study revealed that the chlorophyll content was increased in mild water stress (6-day irrigation interval). Taiz and Zeiger (1991) reported that in the mild water stress conditions, with a decrease in leaf area, the concentration of chlorophyll increased while the severe water stress impeded chlorophyll synthesis. In general, the effect of water stress on the chlorophyll content is very different and depends on environmental conditions, plant genotype, stress duration, and intensity (Antolin *et al.*, 1995).

Some studies have reported that water stress can reduce the photosynthesis rate by stomatal closure and metabolic impairment (Ennahli and Earl, 2005; Flexas *et al.*, 2006). In the present study, the decrease in photosynthesis rate was only observed in mild water stress while with the increase in the irrigation interval (9 days), the photosynthesis rate was at a normal level. Flexas *et al.* (2006) expressed that under natural conditions, water stress normally develops much more gradually over time and it is possible that some acclimation occurs, in addition to day to day variations in response to the variable environmental conditions. Among mulch types, the superabsorbent indicated an appropriate interaction with the 9-day irrigation interval. The increased rate of photosynthesis by applying superabsorbent has been reported by Islam *et al.* (2011) in oat, Mehri *et al.* (2013) in corn, and Najafinezhad *et al.* (2015) in lawn.

The highest RWC in the present study was obtained from the superabsorbent. RWC is an important parameter that is most commonly used to assess plant water status. The application of superabsorbent to the culture media could conserve different amounts of water and thereby improve the medium's water retention capacity. Therefore, in the plant growth and yield traits, RWC increased under water stress condition (Tohidi-Moghadam *et al.*, 2009). In this regard, it has been established that the application of superabsorbent to various plants (Khadem *et al.*, 2010; Najafinezhad *et al.*, 2015; Nazarli *et al.*, 2010) could increase RWC.

Table 4. Means squares of physiological and chemical traits of the marigold plants treated with various mulch types and irrigation intervals.

SoV	df	Total chlorophyll	Chl. a/b	Photosynthesis rate	RWC	Proline
Mulch	3	2.05 ^{ns}	0.16 ^{ns}	45.31 ^{ns}	1038.26 ^{**}	0.02 ^{ns}
Irrigation	2	2.69 [*]	0.29 ^{ns}	665.76 ^{**}	97.92 ^{ns}	0.06 ^{ns}
Mulch × irrigation	6	0.19 ^{ns}	0.10 ^{ns}	390.53 ^{**}	94.96 ^{ns}	0.02 ^{ns}
Error	36	0.77	0.10	42.97	127.01	0.04
Total	47	-	-	-	-	-
Standard deviation	-	0.93	0.34	10.68	13.41	0.14
CV (%)	-	24.81	17.79	51.84	22.04	0.65

*, ** and ns show significance at the 5% and 1% probability levels and insignificance according to Duncan's test, respectively.

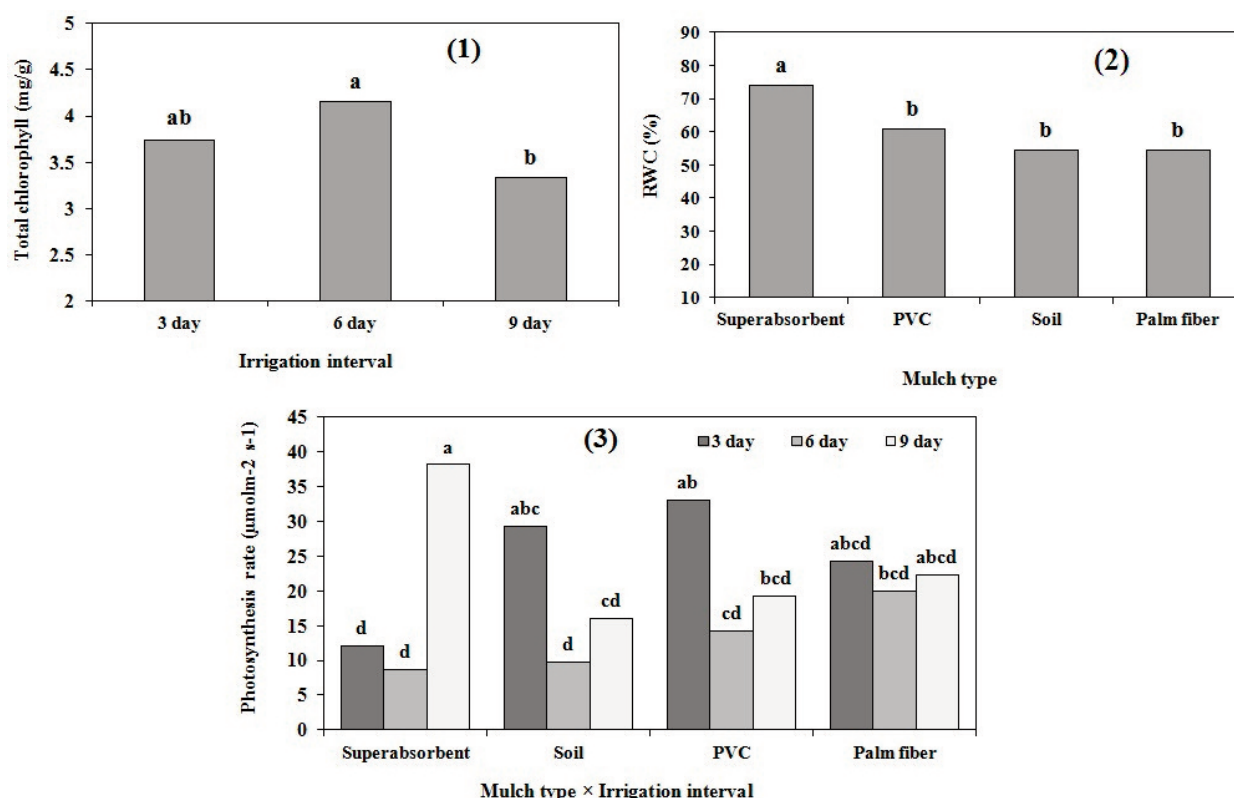


Fig. 5. The effect of the irrigation interval on total chlorophyll (1), mulch type on relative water content (2) and interaction of them in photosynthesis rate (3) in the marigold.

CONCLUSION

The superabsorbent and, at the next level, palm fiber mulch significantly contributed to improving most measured traits. The use of PVC can significantly reduce shoot dry weight and also flower number. Although, in some measured traits, the 3-day irrigation interval was more effective, there was not much difference between the 3-day and 9-day irrigation intervals. In conclusion, the application of superabsorbent, especially palm fiber mulch (since it is cheap and affordable as well as natural), with 9-day irrigation interval to African marigold in urban areas with climates similar to Mashhad (arid and semi-arid) is advisable. In order to reduce water consumption in the urban landscape of arid and semi arid regions, we suggest investigating the effect of using local and natural mulches and new and safe water-absorbing polymers on ornamental plants in future research.

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