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Full Length Research Paper

Responses of phenological and physiological stages of spring safflower to complementary irrigation

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In order to investigate impact of complementary irrigation on phenological stages, chlorophyll content, radiation absorption and extinction coefficient, as well as some aspects concerning the yield of spring safflower, a split-plot experiment based on randomized complete block design with three replication was conducted at the Ardabil Research Center For Agriculture And Natural Resources, Ardabil, Iran in 2009. Experimental treatments included irrigations (non- irrigation, irrigation at the head appearance and irrigation at the flowering stage) and safflower cultivars (Jila, native Isfahan and Pi-537636). Irrigation levels showed significant effects on flowering initiation, flowering period, seed filing period, maturing, number of head, number of seed per head, shoot dry weight, seed yield, extinction coefficient, radiation absorption and chlorophyll content. There were significant differences among cultivars in terms of head appearance, flowering, flowering period, maturing, number of head per plant, number of seed per head, shoot dry weight, seed yield per plant and seed oil content. Also, interactions between irrigation and cultivar were significant for flowering period and seed yield. The highest flowering period was observed by applying irrigation at the head appearance for the native Isfahan cultivar which caused the highest seed yield at the flowering stage. Complementary irrigation increased chlorophyll content and radiation absorption and decreased extinction coefficient at both stages compared to no irrigation.

Key words: Safflower, complementary irrigation, absorption of radiation, extinction coefficient.

INTRODUCTION:

Water availability is an important factor affecting plant growth and yield, mainly in arid and semi-arid regions, where plants are often subjected to periods of water deficit. The occurrence of morphological and physiological responses, which may lead to some adaptation to drought stress, may vary considerably among species (Levitt, 1980). Water stress influences plant growth at various levels; from cell to community, the quantity and quality of plant growth depends on cell division and enlargement, and differentiation and all of these events are affected by water stress. Water stress is a very important limiting factor at the initial phase of plant growth and establishment. Reduction in water availability reduces the number of leaves per plant, as well as individual leaf size and leaf longevity. The yield components such as grain yield, grain number, grain size, and floret number, decrease under pre-anthesis drought stress in sunflower. Seed yield and yield components are severely affected by water deficit. Water stress reduced the head diameter, 100-achene weight and yield per plant in sunflower (Shao et al., 2008).

Safflower (*Carthamus tinctorius* L.) is a plant adapted to moderate drought climates with rather low rates of available water. Safflower was primarily cultivated for its pharmaceutical usages but nowadays it is cultivated to produce edible oil and seed (Mc Pherson et al., 2004). The importance of oil crops such as safflower has increased in recent years, as this plant successfully acclimatizes to rain-fed conditions and is highly drought resistant due to its extended root system enabling access

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to water down to 4 m in the soil (Knowles, 1989; Weiss, 2000).

Favorable growth and production of safflower, however, depends on sufficient water supply. Several studies indicate that optimum yields are obtained under irrigation conditions (Ozturk et al., 2008). Increasingly drought stress at the susceptible growing stages restricts yield so complementary irrigation can lead to the optimum yield (Oweis and Hachum, 2001). Complementary irrigation consists of applying limited amounts of water at the humidity deficit to maintain plant growth and stable seed yield (Oweis et al., 1999). Optimum complementary irrigation in the rain-fed lands is conducted at the flowering stage on three main basis (Oweis, 1997): 1) water is used merely for improving crop yield grown as rain-fed (and has traditional yield without irrigation); 2) precipitation is considered the most important water source; in the absence of precipitation, complementary irrigation provides sustained yield and 3) these irrigation schedules at the critical stages allow achieving to some degree, the optimum yields (rather than maximum) with the lowest available water.

The amount of radiation that may be absorbed by a plant is strongly related to vegetation cover or leaf area index (LAI), canopy structure, and solar zenith angle. The efficiency of utilizing the absorbed photosynthetically active radiation (PAR) for biomass production, hereafter termed radiation-use efficiency (RUE), can change with variation in leaf chlorophyll content, plant growth stage, and field management practices and environmental stress intensity (Wang et al.,2001). Alvino et al. (2000) reported that water deficit affected RUE by altering LAI, radiation absorption and canopy structure. Radiation interception is variable throughout the growing period of crops (Watiki et al., 1993) and is influenced mainly by the green leaf area duration and canopy extinction coefficient (K). Many studies indicate that the variability of K for a given species is determined by the effect of environmental constraints (like drought) on its canopy through the modification of angle, spatial distribution and optical properties of leaves (Jeuffroy and Ney, 1997). Irrigated crops allow RUE to remain relatively stable throughout the crop cycle; however water deficits decrease RUE, especially during grain filling. Are then transpiration based stress factors or the water supply typically applied to the unstressed RUE valid for crops that rarely reach full canopy cover in such arid environments? RUE in the canopy is directly limited by the rate of photosynthesis of well-watered leaves; previous photosynthesis studies show that water stress reduces the maximum photosynthetic rate and, at the canopy level, a lower LAI occurs. The lower LAI arises by two mechanisms: (1) the production, then senescence of leaves (a profligate mechanism) and (2) leaf area development (leaf appearance and leaf expansion) that is driven on the basis of available water (a conservative mechanism) (O'Connell et al., 2004).

With regards to precipitation limitation, high rates of evapotranspiration and other cultivation limiting factors under the rain-fed condition, it is of great importance to select drought tolerance cultivars using drought stress studies. This work was done to gain the favorable yields by choosing the suitable irrigation timing and impact on the seed yield of the spring safflower cultivars.

MATERIALS AND METHODS

This experiment was performed at the Ardabil Research Center for Agriculture and Natural Resources, Ardabil, Iran. Site of the experiment was located at the elevation of 1350 m from the sea level (38°15' N, 48°15' E). Soil texture was loamy-clay. Work was arranged as split-plot based on the randomized complete block design with three replications. Irrigation levels as main factor comprised of no irrigation, irrigation at head appearance and irrigation at the flowering stage based on the plant physiological stages (Tanaka et al., 2002); were placed as the main-plots and safflower cultivars (Jila, native Isfahan and PI-537636) were arranged as sub-plots. Each sub- plot was comprised of six growing rows at 25 cm distance, each 4 m long. Plants were grown 10 cm apart on the rows. The site was under seed legumes cultivation a year before. Planting was done manually in mid- March, weeds were controlled mechanically and Diazinon pesticide was used against *Acanthi Philus Helianthi* Rossi. Phenological aspects (degree-day from planting to head appearance, flowering initiation, flowering period, seed filing period and maturity) were recorded. Chlorophyll content in apical fully extended leaves was determined using chlorophyll meter device (Konica-Minolta, SPAD). Rate of the radiation absorption was measured in a clear, sunny day during 12 to 14 h using sunscan (Delta- TDevices, England) device. Intercepted light percent was calculated as follows (Hashemi- Dezfuli, 1990): $[(I_0-I_i)/I_0] \times 100$; Where: I_0= rate of the radiation at the upper levels of the canopy and I_i= rate of the radiation at the lower levels of the canopy. Extinction coefficient (k) was calculated as follows: (Gallo et al., 1993): $I_i = I_0 e^{-KLAI}$, $k = -\ln (I_i/I_0)/LAI$.

To determine yield and yield components, lateral rows were removed as edge effects, and harvest was analyzed only for the middle rows. Seed oil percent was determined using a seed analyzer device (Inframatic 8620- percon, Germany). Data were analyzed in SAS 2003 and graphs were drawn using excel software.

RESULTS AND DISCUSSION

Phenological traits

Required time to head appearance was not affected by irrigation, because complementary irrigation was performed beyond this stage. A significant difference (P<0.05) was observed among the cultivars in terms of head appearance: PI-537636 genotype showed the highest degree-day (759.99) and native Isfahan showed the lowest (698.90); there was no significant difference between PI-537636 and Jila cultivars (Table 1). All cultivars differed from each other in terms of appearance of phenological stages. This means that genetic features play an important role in determining the length of each stage, in addition to the environmental factors (Shaneiter et al., 1981; Zheng et al., 1993). Flowering initiation was significantly different (P<0.01) among the irrigation treatments. Irrigation caused the highest (1071.12) and the lowest (918.88) degree-day

Table 1. Impact of irrigation levels and cultivars on the phenological stages (degree days). Averages with the same letter do not show significant difference with each other.

Treatment	Heading	Flowering	Flowering period	Seed filling period	Maturity
Irrigation levels					
Non-irrigation	756.13[a]	918.88[b]	189.34[c]	315.07[b]	1481.27[b]
Irrigation at heading	742.78[a]	1071.12[a]	398.29[a]	369.71[ab]	1784.48[a]
Irrigation at flowering	724.91[a]	950.46[b]	269.33[b]	542.16[a]	1761.94[a]
Cultivar					
Jila	759.99[a]	972.20[b]	248.26[b]	424.24[a]	1648.03[b]
Native Isfahan	698.90[b]	1031.04[a]	407.40[a]	291.80[b]	1730.24[a]
PI-537636	764.93[a]	937.21[b]	201.31[c]	510.89[a]	1649.41[b]

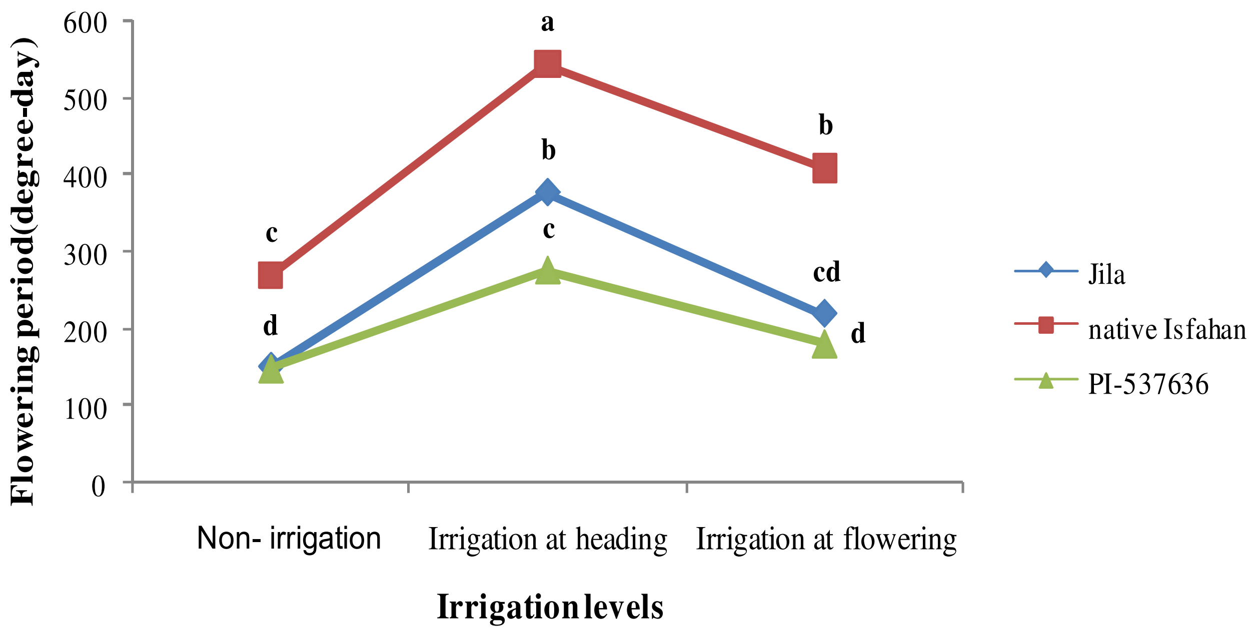

Figure 1. Flowering period as affected by irrigation and cultivars of safflower.

in the head appearance stage until flowering. Also, there was insignificant difference between no irrigation and irrigation at the flowering stage (Table 1). Since the plants under no irrigation and irrigation treatments at the flowering stage had been subjected to drought stress, they entered rapidly the flowering stage. Cultivars showed significant differences ($P<0.05$) in terms of flowering initiation: native Isfahan possessed the highest degree-day (1031.04) while PI-537636 had the lowest (937.21); Jila was placed at the same level with these cultivars. Asheri et al. (1975) observed significant differences of this trait by studying various safflower germ plasms, as well.

Drought stress shortened flowering period and the lowest period was obtained from irrigation during the head appearance stage. Palmer et al. (1995) obtained the same results with soybean experiments. Cultivars had different flowering duration (Table 1). Also, significant interaction ($p<0.05$) was observed between irrigation and cultivar for this trait, and native Isfahan showed the highest degree-day (543.47) in irrigation of head appearance. In contrast, PI-537636 showed the lowest rate without irrigation (Figure 1). Flowering duration showed significant difference with number of head per plant, seed yield and radiation absorption (Table 4).

Seed filling period differed between irrigation treatments ($P<0.01$) and irrigation in the flowering stage had the highest seed filling period (542.16) while no irrigation caused the lowest one (3015.04) (Table 1). Impact of drought stress on declining of seed filling period in crop plants was reported previously by Vieira et al. (1992). This study also found significant differences ($P<0.01$) for seed filling period among the cultivars (Table 1), in that this period was the shortest in native Isfahan (291.8 degree-day), observed via lowest 1000 seed weight of the cultivar.

Table 2. Impact of irrigation levels and cultivars on yield and yield components. Averages with the same letter do not show significant difference with each other.

Treatment	Head per plant	Seed per head	Shoot weight (g per plant)	Seed weight (g per plant)	Oil percent
Irrigation levels					
Non-irrigation	4.98^{b}	12.61^{b}	8.16^{c}	2.68^{b}	29.16^{a}
Irrigation at heading	5.86^{a}	24.76^{a}	11.96^{b}	4.82^{a}	29.46^{a}
Irrigation at flowering	5.41^{ab}	25.18^{a}	14.89^{a}	5.23^{a}	30.17^{a}
Cultivar					
Jila	5.10^{b}	16.73^{b}	10.84^{b}	3.63^{b}	28.18^{b}
Native Isfahan	6.11^{a}	24.52^{a}	12.91^{a}	4.59^{a}	29.20^{b}
PI-537636	5.03^{b}	21.29^{a}	11.36^{b}	4.44^{a}	30.41^{a}

Seed filling period was positively correlated with seed yield and chlorophyll content and negatively correlated with extinction coefficient (Table 4).

Irrigation treatments had significant effect ($P<0.01$) on required time to maturity: irrigation in the head appearance stage led to the highest (1784.48) and no irrigation caused the lowest (1481.27) degree-day. No significant difference was observed between irrigation in the head appearance and irrigation in the flowering stage (Table 1), so drought stress shortened the plant growth period. In other words, one of the available mechanisms for plant facing drought stress is to reduce plant growth period to cope with the unfavorable growing conditions and to attain maturity stage faster. Cultivars had significant difference ($P<0.01$) for required degree-day until maturity (Table 1). Native Isfahan cultivar had the highest degree-day (1730.24) and possessed the longest growth period compared to the other cultivars.

Yield and yield components

Irrigation in the head appearance led to the highest head per plant (5.86) and non-irrigation produced the lowest one (4.98), so irrigation in the head appearance stage increased number of head 15.02% (Table 2). Thus, the closer irrigation application to the head appearance stage, the more heads and eventually yields are produced. Abel (1976) found that drought stress decreases number of heads per plant. We observed that the number of head per plant was not same for the cultivars (Table 2) and native Isfahan had the highest head compared to the other cultivars (Table 2). Ghorpade et al. (1993) and Khidir (1974) found significant variation in the number of head per plant in safflower. Number of head per plant had a positive and significant correlation with the seed yield (Table 4).

Irrigation also had a significant effect on seed number per head ($P<0.01$). Irrigation in the flowering stage caused the highest (25.18) and non-irrigation caused the lowest (12.61) seed number per plant (Table 2), and increased seed per head by 99.7%. It seems that irrigation in this stage affects positively the number of fertile flowers and consequently, the number of seed per head. Abel (1976) showed that seed number per head is decreased as a result of drought stress. Reduction in seed number resulting from drought stress has been reported in common bean (Ramirez-Vallejo and Kelly, 1998) and in soybean (Cox and Jollif, 1986). In our study, native Isfahan had the highest (24.52) and Jila had the lowest (16.73) seed per head (Table 2). Ashri (1974) stated that seed number per head is different within the Iranian cultivars.

Irrigation in the flowering stage caused the highest (14.89) and non-irrigation caused the lowest (8.16 gr/plant) shoot weight (Table 2). Decrease in shoot weight under the non-irrigation conditions was due to lower plant growth. Native Isfahan possessed the highest (12.91) and Jila, the lowest (10.84) shoot weight (Table 2). Sankar et al. (2008) reported biomass loss under drought circumstances as well.

Seed yield was affected by irrigation treatments. Sufficient water precipitation resulted in the increase in number of flower heads per plant and number of seeds per plant and hence, increased plant yield. According to Leonard and French (1969), plant yield was affected by irrigation. Fereres and Fernandez (1986) observed large variation in yield among the sunflower cultivars. Also, Gonzales et al. (1994) reported different yields as a function of irrigation amounts. There were significant interactions ($P<0.01$) between irrigation and cultivar for seed yield: PI-537636 genotype possessed the highest (6.04 $gr.m^{-2}$) and Jila the lowest (2.5 $gr.m^{-2}$) seed yield with non-irrigation treatment (Figure 2). There were positive correlations between seed yield and number of head per plant, flowering duration, seed filling period, chlorophyll content and radiation absorption, and negative and significant correlation with extinction coefficient (Table 4).

Seed oil percent was not affected by irrigation treatments. Seed oil content, considered a quantitative trait, is controlled by multiple genes so, it is less likely for these genes to be affected by water deficit conditions. Hang and Evans (1985) found that drought stress did not

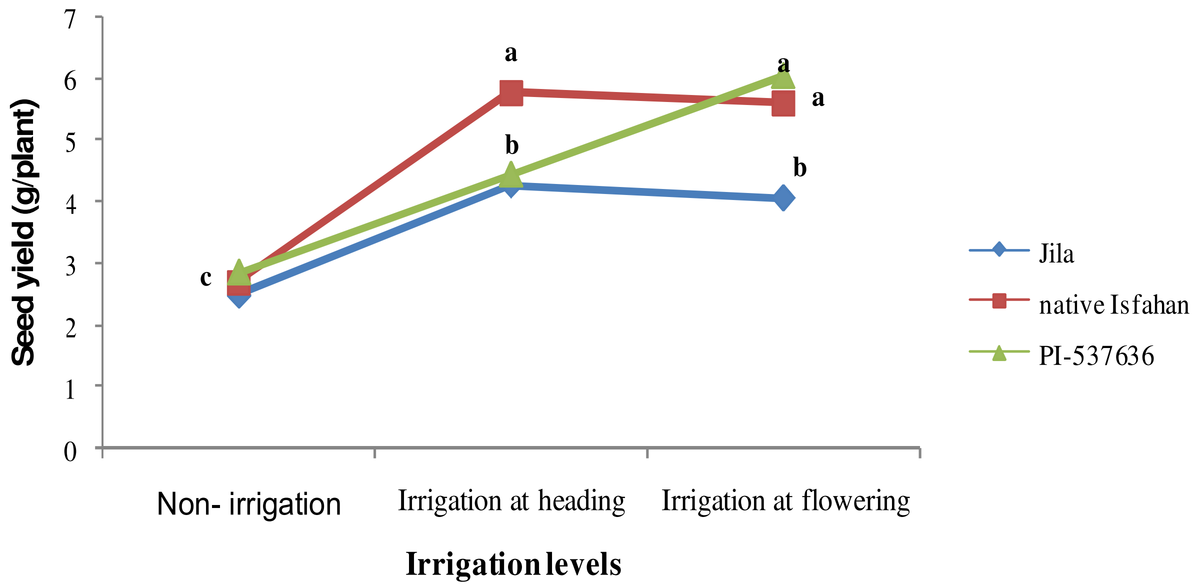

Figure 2. Seed yield as affected by irrigation and cultivars of safflower.

Table 3. Impact of irrigation levels and cultivars on chlorophyll content, absorption of radiation and extinction coefficient. Averages with the same letter don't show significant difference with each other.

Treatment	Chlorophyll	Absorption of radiation	Extinction coefficient
Irrigation levels			
Non-irrigation	55.04[b]	49.33[b]	0.78[a]
Irrigation at heading	71.44[a]	56.67[a]	0.64[b]
Irrigation at flowering	73.76[a]	54.11[a]	0.60[b]
Cultivar			
Jila	68.2[a]	51.11[a]	0.69[a]
Native Isfahan	67.07[a]	54.11[a]	0.67[a]
PI-537636	64.98[a]	54.89[a]	0.65[a]

significantly affect seed oil percent of safflower. Sing et al. (1997) reported that seed oil percent is not affected by irrigation levels. However, cultivars studied here showed significant difference ($P<0.05$) for seed oil percent and the highest (30.41%) amount belonged to PI-537636 genotype (Table 2). Previous studies also showed that seed oil content is different among cultivars of safflowers (Arsalan et al., 1997; Koutroubas et al., 2009).

Chlorophyll content, radiation absorption and extinction coefficient

Irrigation had a significant impact on leaf chlorophyll content ($P<0.01$) such that it increased this trait and the highest value (73.76) was obtained using irrigation at flowering (Table 3). Irrigation increased chlorophyll content by 34% compared with no irrigation. Synerri et al. (1993) reported that drought stress caused hydrolysis of tilakoid proteins and decline of chlorophyll content. Chandrasekar et al. (2000) also found that drought decreases chlorophyll content. Fotovat et al. (2007) stated that wheat leaf chlorophyll content was decreased significantly by exerting severe drought stress relative to medium.

Radiation absorption was affected significantly by irrigation treatments ($P<0.01$) such that the highest (56.67) and the lowest (49.33 %) radiation absorption was observed as a result of irrigation at head initiation and without irrigation, respectively (Table 3). Irrigation at head appearance increased light absorption by 15%. It seems that the increase in the number and area of the leaves caused by irrigation led to increase in the radiation absorption. Cultivars studied here did not show significant difference for this trait (Table 3).

Impact of irrigation treatments was significant on extinction coefficient ($P<0.01$) and the highest rate (0.78) was

Table 4. Simple correlation among the studied traits.

	Head per plant	Seed yield	Flowering period	Seed filling period	Chlorophyll content	Absorption of radiation	Extinction coefficient
Head per plant	1.00						
Seed yield	0.56 **	1.00					
Flowering period	0.81 **	0.65 **	1.00				
Seed filling period	0.15 ns	0.66 **	0.15 ns	1.00			
Chlorophyll content	0.28 ns	0.63 **	0.33 ns	0.53 **	1.00		
Absorption of radiation	0.62 **	0.54 **	0.51 **	0.28 ns	0.12 ns	1.00	
Extinction coefficient	-0.25 ns	-0.8 **	-0.34 ns	-0.77 **	-0.5 **	-0.4 *	1.00

ns, * and ** are in significant and significant at (P<0.05) and (P<0.01), respectively.

observed without irrigation. Irrigation at flowering stage decreased extinction coefficient by 30% (Table 3). Irrigation likely increased the number and duration of the leaves, hence the decrease in the extinction coefficient. This trait showed negative and significant correlation with seed yield, seed filling period, chlorophyll content and radiation absorption (Table 4).

Conclusions

In this study we found that limitation of sufficient irrigation (complementary irrigation) caused yield loss via early flowering decrease in flowering period, seed filling period and maturity stage, and also decrease in chlorophyll content, radiation absorption and increase extinction coefficient. Since irrigation at flowering increased yield by 95%, in water limitation conditions, irrigation is of great importance to improving yield. With regards to the differences among the cultivars related to yield, native Isfahan was classified as a superior cultivar.

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