# **ORIGINAL ARTICLE**



# Growth and Yield Parameters of *Triticale* as Influenced by Methanol Foliar Application Under Water Stress

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Triticale is mainly grown for feed grain, and biomass production for thatching straw and common human utilization. A combined analysis with split plot layout in two years of 2016 and 2017 with 5 replications was used to evaluate yield and yield components of triticale under different methanol concentrations and irrigation managements in Isfahan, Iran. Irrigation treatments consisted of irrigation on the basis of 70%, 80%, 90% and 100% crop water requirements, and methanol treatments as foliar application on the basis of 15% methanol concentration, 30% methanol concentration and control treatment (0%). Methanol application influence a hundred grain weight was significant. The maximum plant height, number of tillers, LAI, LAD, a hundred grain weight, grain yield, biological yield, harvest index and protein content was achieved in irrigation on the basis of 100% crop water requirement. The maximum plant height, number of tillers, LAI, LAD, one hundred grain weight, grain yield, biological yield, harvest index and protein was obtained in 2017. Foliar methanol application with 15% concentration had obtained the maximum plant height, LAI, LAD, a hundred grain weight, biological yield, SPAD and protein percentage. The results of this experiment suggest that methanol can aid in alleviating the effects of drought stress on triticale in climatic condition of Isfahan. It is concluded that triticale cultivars performed superior in 2017, with 15% concentration of methanol application and irrigation on the basis of 100% crop water requirement.

Key words: Triticale; methanol; irrigation; LAI; LAD; SPAD

Triticale is a man-made crop developed by crossing wheat (Triticum turgidum or Triticum aestivum) with rye (Secale cereale) (Kamyab et al. 2009). Compared with wheat, triticale has superior characteristics such has higher protein content (Kamyab et al. 2009), less requirement for nitrogen fertilization and more tolerance to salinity and drought stresses (Ahmed and Abdel-Hady 2006; Moharrery et al. 2015; Giunta et al. 2017). Heidari et al. (2016) reported that triticale seems to be an alternative to other small grain cereals, particularly wheat and barley for cultivation under unfavorable conditions or in the low-input agricultural systems. It has been reported that under drought stress conditions and problematic soil regions, triticale showed distinct yield superiority and had adaptive advantages over wheat (Pfeiffer 2003; Lelley 2006; McKenzie et al. 2014; Roques et al. 2016). Agronomic traits such as grain yield and its components are the major selection criteria for evaluating drought tolerance under field conditions (Fayaz and Arzani 2011; Soleymani et al. 2011; Soleymani et al. 2012; Shahrajabian et al. 2013; Sun et al., 2019a; Shahrajabian et al., 2020a,b; Sun et al., 2021a,b,c). The closely linked of LAI, leaf area duration (LAD), and SPAD (Chlorophyll index) with high grain yield have been reported for some crops such as spring barley (Janusauskaite and Auskalniene 2014) and spring triticale (Janusauskaite et al. 2017). Many scholars have reported that SPAD measurements can be used as a single and rapid tool to detect and select stable and high yield of plants (Nakano et al. 2010; Kendal 2015; Janusauskaite et al. 2017). Methanol spray is a method that increases CO<sub>2</sub> fixation in plants and methanol may act as a carbon source for plants (Nonomura and Benson 1992). Methanol is the simplest alcohol and can be produced through anaerobic metabolism by some bacteria (Khaki-Moghadam and Rokhzadi 2015). Furthermore, methanol is emitted from leaves of C3 plants (Fall and Benson 1996). In a research, plant biomass and SPAD chlorophyll content were increased by foliar spraying of soybean plant with 15% (v/v) methanol (Saadpanah et al. 2013). The decline in intercellular CO<sub>2</sub> is a key limiting factor photosynthesis under drought stress conditions, and plant productivity may be promoted by increasing the availability of CO<sub>2</sub> in leaves through applying a carbon source (Khaki-Moghadam and Rokhzadi 2015). Consequently, foliar methanol application in arid and semi-arid regions maybe useful in increase of productivity and plant growth. Khaki-Moghadam and Rokhzadi (2015) found that, foliar application of methanol under drought stress conditions at flowering stage may increase growth and yield of safflower, whereas under fully watered conditions the application of lower doses of methanol maybe more effective in yield improvement than use of higher dose. Nonomura and Benson (1992) stated that foliar sprays of aqueous 1-50% methanol increased growth and development of C<sub>3</sub> crop plants in arid environment. Most of triticale cultivars have a high grain yield but perform poorly with regard to biomass yield (Pronyk and Mazza 2011). Deficit irrigation means as the application of water below full crop water requirements, which is one of the choice to sustain productivity (Faroog et al. 2009; Soleymani and Shahrajabian 2013; Shahrajabian et al. 2017; Soleymani and Shahrajabian 2017; Sun et al., 2019b; Shahrajabian et al., 2020c,d; Sun et al., 2021c,d). Supplemental and managing irrigation, even in semi-arid and arid climates, is one of the crucial means which guarantee stable and reliable yields. In center of Iran, because of limited rainfall, water shortage and stress, major loss in crop productivity happens. The aim of this research was to evaluate the influence of methanol application and irrigation management on yield and yield components of triticale in semi-arid region of Iran.

#### MATERIALS AND METHODS

Two year experiments were conducted at agricultural research farm of Faculty of Agriculture, Islamic Azad University, Isfahan (Khorasgan) Branch, Isfahan, Iran (Latitude 32°40′ N, longitude 51°58′ E, and 1570 m elevation) in order to investigate the influence of different methanol concentrations and irrigation on the basis of crop water requirement during 2016, 2017. A combined analysis with split plot layout in two years of 2016 and 2017 with 5 replications was used. Irrigation treatments consisted of irrigation on the basis of 70%, 80%, 90% and 100% crop water requirements, and

methanol treatments were foliar application on the basis of 15% methanol concentration, 30% methanol concentration and control treatment (0%). On the basis of soil analysis, the organic carbon was 1.2% and 1.0% at depth of 0-30 cm and 30-60 cm, respectively; moreover, at both depths soil texture was clay (Table 1). Mean temperature changes from 23th October to 22th October at the Isfahan experimental field in 2016 has shown in Table 2. The soil preparation consisted of mouldboard ploughing, followed by disking and smoothing with a land leveler. Autumn triticale (ET-84-8) was sown at a density of 400 viable seeds per m<sup>2</sup> with skillful workers on Oct 23th. On the basis of soil analysis, nitrogen fertilizer was used from urea source (80 kg N ha<sup>-1</sup>). Half of the nitrogen was applied during pre-sowing cultivation of soil and the remaining amount of nitrogen fertilizer was applied at the tillering stage of triticale. Each plot (10 m<sup>-2</sup>) consisted of six rows, with 17.5 cm spacing. The crop was harvested in the stage of full maturity, and grains yield was measured and adjusted at 14% moisture. Weed control, diseases and pest management were carried out in accordance with the crop development as required. The foliar treatments were applied at tillering stage. Control plots did not receive any foliar application. Chlorophyll index (SPAD, soil plant analysis development) was measured by using chlorophyll meter Minolta SPAD 502. Measurements were performed from the middle of the leaf blade just before harvesting using a portable meter. The plant height of main shoot was measured with a ruler before Plants were harvested at physiological harvest. maturity, oven-dried at 72°C, and after that yield and yield components including grain yield and one hundred grain weight (g) were determined. Leaf area meter was measured by leaf area meter (Delta T Device, UK). LAD was measured using the following formula (Janusauskaite et al., 2017):

$$LAD = \frac{L1 + L2}{2(T2 - T1)}$$
(1)

Where La and L2 are the first and second measurements of green leaf area, and T1 and T2 represent the time of the first and second measurements. The separated grain weighed, and harvest index (%) was calculated according to the

following formula:

$$HI (\%) = \frac{Grain yield}{Biological} \times 100$$
(2)

The amount of nitrogen was calculated by Kjeldahl analysis from dry and ground samples (Bremner and Breitenbeck 1984), and then nitrogen was multiplied by 6.26 to determine protein content. After collection of data related to measured properties, variance analysis was done by SAS statistical software.

#### RESULTS

The influence of year was not significant on any experimental characteristics except for LAD. Irrigation treatment has meaningful effects on plant height, number of tiller, LAI, LAD, one hundred grain weight, grain yield, biological yield, harvest index and Protein, but SPAD was not significantly affected by irrigation treatment. Among all experimental characteristics, just one hundred grain weight influenced by methanol concentration, while no significant influence of methanol concentrations on other experimental characteristics was found. The analysis of variance showed that the effect of interactions between irrigation × year, and also irrigation × methanol on LAD and grain yield was statistically significant. Aside from LAD and one hundred grain weight, no experimental characteristics influenced by interaction between methanol and year (Table 3). The maximum value for plant height, number of tillers and LAI was related to 2017, followed by 2016. Moreover, no significant differences were found between treatments. The maximum and the minimum LAD was achieved in 2017 and 2016, respectively; the meaningful difference was found between 2016 and 2017. Plantation on 2017 had obtained the highest grain yield (517.36 g/m<sup>-2</sup>), biological yield (1813.39 g/m<sup>-2</sup>) and Harvest index (0.29). No meaningful differences were found between 2016 and 2017. On the one side, the highest value for SPAD and Protein was obtained for 2016 (46.03), and 2017 (13.53%); on the other side, no significant differences were found between treatments (Table 4). The maximum plant height, number of tillers and LAI was obtained for irrigation on the basis of 100% of crop water requirement, followed by irrigation on the basis of 90% of crop water requirement. Irrigation on the basis of 100% of crop water requirement had no meaningful differences with irrigation on the basis of 90% of crop water requirement. The maximum number of LAD, one hundred grain weight (6.03 g), grain yield, biological yield and harvest index (0.31%) was related to irrigation on the basis of 100% of crop water requirement. Although, the highest number of Harvest index was achieved in irrigation on the basis of 100% of crop water requirement followed by 90%, 80% and 70%, but no significant differences were found between treatments. The maximum value for SPAD was related to irrigation on the basis of 90% of crop water requirement (46.58); Moreover, it had just meaningful difference with irrigation on the basis of 70% of crop water requirement. The protein percentage of triticale differed between various irrigation treatments. The maximum and the minimum protein percentage was related to irrigation on the basis of 100% or crop water requirement (14.59%) and irrigation on the basis of 70% of crop water requirement (10.63%), which had no meaningful differences with each other (Table 5). Water shortage during reproductive stage of plants can lead to a decrease of photosynthesis rate and consequently reduction of number of tillers, leading to a lower number of seed and see weight and finally may lead into lower grain yield. In another word, the significant reduction of photosynthesis and production of assimilates arise

because of reduced water availability which indicated a decline in allocation of assimilates to see and subsequent decrease in seed weight. The maximum value of plant height, LAI and LAD was related to application of methanol 15%, which had no meaningful differences with other treatments. Although the maximum number of tillers was achieved in control treatment (0%), no significant differences were found between treatments. The maximum number of one hundred grain weight was related to application of methanol 15% (5.89 g), which had meaningful differences with other treatments. The maximum grain yield and biological yield was achieved in usage of control treatment (0% methanol), and methanol 15%; although, no meaningful differences were found between treatments. This shows that biomass yield, just as grain yield, is a multiplicative trait with different components trait contributing to it. One the one hand, the maximum harvest index was related to control treatment (0.30%); on the other hand, the minimum one was related to application of methanol 15% (0.27%), but no meaningful differences was shown between these two treatments. Application of methanol 30% and 15% had obtained the highest SPAD (45.70), and protein (13.78%). Like, former experimental characteristics, no meaningful differences were found between treatments (Table 6).

Depth (cm)	EC (dS/m)	рН	CaCO₃	Organic carbon (%)	Nitrogen (ppm)	Available P (ppm)	Available K (ppm)	Sand %	Silt %	Clay %	Soil texture
0-30	2.5	7.8	35	1.2	0.12	15.6	400	11	41	48	Clay
30-60	2.43	7.9	38	1.0	0.10	10.2	380	11	37	52	Clay

Table 1. Soil analysis of agriculture research field in Isfahan (0-30 cm).

Table	2.	Mean	temperat	ure changes	from 23th	October	to 22th	October	at the	Isfahan	experimental	field	in 2	2016
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Month	Mean maximum temperature (°C)	Mean minimum temperature (°C)	Mean monthly temperature (°C)	The maximum temperature (°C)	The minimum temperature (°C)	Monthly rainfall (mm)
23th Oct-23th Nov	16.15	5.31	10.73	24.00	-3.18	11.70
23th Nov-23th Dec	10.62	-2.01	4.30	18.12	-8.23	5.80
23th Jan-23th Feb	9.76	-3.93	2.92	15.26	-8.67	1.00
23th Feb-23th Mar	9.72	0.97	5.35	16.64	-6.99	18.00
23th Mar-23th Apr	17.54	2.80	10.17	23.30	-3.42	35.50
23th Apr-23th May	17.86	5.26	11.56	23.00	-0.43	26.72
23th May-23th June	23.46	10.12	16.79	29.94	2.90	26.70
23th June-23thJuly	31.64	15.18	23.41	35.00	12.05	0.21
23th July-23th Aug	35.62	16.65	26.14	36.40	15.00	0

S.O.V	df	Plant height	No. Tiller	LAI	LAD	100 Grain weight	Grain Yield	Biological Yield	HI	SPAD	Protei n
Year	1	3.125 <sup>ns</sup>	0.429 <sup>ns</sup>	0.0001 <sup>n</sup> s	0.046*	2.219 <sup>ns</sup>	10829.013 <sup>ns</sup>	92306.722 <sup>ns</sup>	0.0005 <sup>n</sup> s	170.786	26.257 <sup>n</sup> s
Block*Year	4	387.541	0.783	0.501	0.004	0.620	17943.930	80801.611	0.007	107.450	25.995
a	3	2943.717 *	16.129* *	6.674**	0.216**	16.452* *	297893.532* *	1877382.167* *	0.0118 <sup>n</sup> s	199.061 <sup>ns</sup>	57.082 *
a* Year	3	116.236 <sup>ns</sup>	1.404 <sup>ns</sup>	0.742 <sup>ns</sup>	0.010*	0.777 <sup>ns</sup>	48642.088*	111051.426 <sup>ns</sup>	0.005 <sup>ns</sup>	108.967 ns	12.982 <sup>n</sup> s
block*a (Year)	12	917.449	0.772	0.552	0.002	4.037	9168.560	80196.852	0.005	53.606	5.829
b	2	1148.791 <sup>ns</sup>	0.211 <sup>ns</sup>	0.729 <sup>ns</sup>	0.001 <sup>ns</sup>	21.079* *	14355.680 <sup>ns</sup>	198968.167 <sup>ns</sup>	0.006 <sup>ns</sup>	26.523 <sup>ns</sup>	24.344 <sup>n</sup> s
a*b	6	1303.606 <sup>ns</sup>	1.327 <sup>ns</sup>	0.376 <sup>ns</sup>	0.009*	0.434 <sup>ns</sup>	21913.421*	163750.500 <sup>ns</sup>	0.009 <sup>ns</sup>	48.530 <sup>ns</sup>	16.290 <sup>n</sup> s
b* Year	2	688.041 <sup>ns</sup>	0.334 <sup>ns</sup>	0.577 <sup>ns</sup>	0.020*	6.028*	4817.180 <sup>ns</sup>	318124.389 <sup>ns</sup>	0.004 <sup>ns</sup>	105.575 <sup>ns</sup>	0.482 <sup>ns</sup>
a*b* Year	6	513.930 <sup>ns</sup>	0.275 <sup>ns</sup>	0.843*	0.005 <sup>ns</sup>	1.470 <sup>ns</sup>	11428.810 <sup>ns</sup>	208549.537 <sup>ns</sup>	0.003 <sup>ns</sup>	98.210 <sup>ns</sup>	22.047 <sup>n</sup> s
Error	32	640.951	1.214	0.344	0.003	1.433	9007.153	132085.56	0.007	170.786	10.242

Table 3. Analysis of variance for experimental characteristics.

1 \* significant at 0.05 significance in F-tests, \*\*significant at 0.001 in F-tests, <sup>ns</sup> non-significant.

2 A= Irrigation on the basis of Crop Water Requirement, B= Methanol

#### Table 4. Mean comparison for experimental characteristics in different years.

Year	Plant height (cm)	Number of tillers	LAI	LAD	100 grain weight (g)	Grain yield (g/m²)	Biological yield (g/m²)	HI	SPAD	Protein (%)
2016	100.33a	4.41a	3.24a	0.45b	4.71a	492.83a	1741.78a	0.28a	46.03a	12.32a
2017	100.75a	4.57a	3.25a	0.50a	5.06a	517.36a	1813.39a	0.29a	42.95a	13.53a
Commor	n lattars with	in each colui	nn do no	diffor sign	nificantly					

Common letters within each column do not differ significantly.

Table 5. Mean comparison for experimental characteristics on the basis of crop water requirements.

Irrigation treatment	Plant height (cm)	Number of tillers	LAI	LAD	100 grain weight (g)	Grain yield (g/m <sup>-2</sup> )	Biological yield (g/m <sup>-2</sup> )	HI	SPAD	Protein (%)
%70	84.77b	3.10b	2.37c	0.31b	3.93b	327.61c	1346.1c	0.25a	39.55b	10.63b
%80	95.66ab	4.69a	3.29b	0.52a	4.28b	500.94b	1713.8b	0.30a	45.54ab	12.43a
%90	109.77a	5.07a	3.58ab	0.53a	5.29a	567.33a	2023.3a	0.28a	46.58a	14.04a
%100	112.00a	5.09a	3.72a	0.55a	6.03a	624.50a	2027.2a	0.31a	46.30ab	14.59a

Common letters within each column do not differ significantly.

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Methanol concentrations (%)	Plant height (cm)	Number of tillers	LAI	LAD	100 grain weight (g)	Grain yield (g/m <sup>-2</sup> )	Biological yield (g/m <sup>-2</sup> )	н	SPAD	Protein (%)
0	94.33a	4.58a	3.05a	0.47a	4.72b	531.25a	1782.3a	0.30a	43.79a	11.81a
15	108.00a	4.50a	3.39a	0.49a	5.89a	501.25a	1866.2a	0.27a	43.99a	13.78a
30	99.29a	4.39a	3.29a	0.47a	4.04b	482.79a	1684.3a	0.28a	45.70a	13.18a

Common letters within each column do not differ significantly.

#### DISCUSSION

Water stress is the major limiting factor in crop production in the world. Yield is the principle selection index used under drought stress conditions. Like the results of this experiment, the influence of the weather conditions in different years on grain yield was confirmed by the studies on other cereals (Gonzalez et al. 2010; Peymaninia et al. 2017). The maximum value of plant height, number of tillers, LAI, LAD, one hundred grain weight, grain yield, biological yield, harvest index and protein content was achieved in irrigation on the basis of 100% crop water requirement. The result of this research is in agreement with Khaki-Moghadam and Rokhzadi (2015). Reduction in plant height under drought stress was reported by Guttieri et al. (2001) and Dencic et al. (2000). Leaf extension can be limited under water stress conditions in order to get a balance between the water status of plant tissues and the water absorbed by plant roots (Passioura, 1996). Reduced plant height as the result of water deficit, is a general response of plants to reduced water availability. Khaki-Moghadam and Rokhzadi (2015) also reported that water deficit and drought stress may delay development of plants, leading to plant height reduction. Guttieri et al. (2001) also observed that moisture deficit induced reduction in 100-grain yield wheat due to reduction in the number of grains per spike. Plant growth occurs by cell elongation and cell division, which are very sensitive to drought stress. Cell elongation is inhibited by a reduction in turgor pressure resulting from water deficiency. Water deficit also impairs cell division. Thereby, disruption of cell elongation and division can explain the observed reduction in plant height and growth. Ahmadi and Joudi (2007), Krcek et al. (2008) and Bijanzadeh and Emam (2017) also reported that grain yield is reduced depending on the degree of water deficit. The improvement of HI leads to more efficient redistribution of dry matter into grain and in turn increases grain yield (Madic et al. 2005; Moradi et al. 2010; Shahrajabian et al. 2017). HI can be used an indirect selection criterion for improving grain yield in cereals under moisture stress conditions (Fayaz and Arzani 2011; Shahrajabian et al. 2011). In agreement with results of this experiment, Ahmed (2011) and Khaki-Moghadam and Rokhzadi (2015) observed that the SPAD chlorophyll index was lower in their experiments under water deficit stress. The highest plant height, number of tillers, LAI, LAD, one hundred grain weight, grain yield, biological yield, harvest index and protein was obtained in 2017. Foliar methanol application with 15% concentration had obtained the highest plant height, LAI, LAD, one hundred grain weight, biological yield, SPAD and protein percentage. Leaf area index (LAI) is a good indicator of crop state and is closely linked to other crop and soil variables such as biomass, grain yield, nutrition status and crop nitrogen uptake (Janusauskaite et al. 2017). Significant reaction of safflower to methanol concentration in limited irrigation management indicated that the deleterious effects of drought stress can be alleviated by methanol application, to increase CO2 concentration. Nakano et al. (2010) ascertained that grain yield of wheat was significantly and positively correlated with the LAI and SPAD. In this experiment triticale positively respond to methanol concentration in limited irrigation management. As a consequence, treating triticale with methanol can promote net photosynthesis leading to improved yield (Nonomura and Benson 1992; Khaki-Moghadam and Rokhzadi 2015). Percival et al. (2008) and Ling et al. (2011) mentioned that leaf chlorophyll content is an important indicator of physiological status in plant, and the variation in leaf chlorophyll content is considered to be a plant response to environmental stress. Furthermore, Nonmura and Benson (1992) and Rowe et al. (1994), Hernandez et al. (2000) and Zbiec et al. (2003) reported that foliar spraying of methanol increased growth and yield of various C<sub>3</sub> plants. The yield increased caused by the applied measures can be explained by a better management of irrigation of the plants, which found an important solution in enhanced tillering, grain yield and one hundred grain weight. Gowda et al. (2011) also have recently reported that grain yield, plant height, spikes per square meter and thousand-kernel weight are key variables that allowed the prediction of early biomass yield. The results of this experiment suggest that methanol can aid in alleviating the effects of drought stress on triticale in climatic condition of Isfahan. It is concluded that triticale cultivars performed superior in 2017, with 15% concentration of methanol application and irrigation on the basis of 100% crop water requirement.

## CONCLUSION

Water stress is the major limiting factor in crop production in the world. Yield is the principle selection index used under drought stress conditions. The highest value of plant height, number of tillers, LAI, LAD, one hundred grain weight, grain yield, biological yield, harvest index and protein content was achieved in irrigation on the basis of 100% crop water requirement. Plant growth occurs by cell elongation and cell division, which are very sensitive to drought stress. Cell elongation is inhibited by a reduction in turgor pressure resulting from water deficiency. Water deficit also impairs cell division. Thereby, disruption of cell elongation and division can explain the observed reduction in plant height and growth. The highest plant height, number of tillers, LAI, LAD, one hundred grain weight, grain yield, biological yield, harvest index and protein was obtained in 2017. Foliar methanol application with 15% concentration had obtained the highest plant height, LAI, LAD, one hundred grain weight, biological yield, SPAD and protein percentage. Significant reaction of safflower to methanol concentration in limited irrigation management indicated that the deleterious effects of drought stress can be alleviated by methanol application, to increase CO2 concentration. The yield increased caused by the applied measures can be explained by a better management of irrigation of the plants, which found an important solution in enhanced tillering, grain yield and one hundred grain weight. The results of this experiment suggest that methanol can aid in alleviating the effects of drought stress on triticale in climatic condition of Isfahan. It is concluded that triticale cultivars performed superior in 2017, with 15% concentration of methanol application and irrigation on the basis of 100% crop water requirement.

#### CONFLICTS OF INTEREST

All authors have declared that they do not have any conflict of interest for publishing this research.

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