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Foliar applied iron and zinc improves the growth, physiological and yield related traits of wheat under drought

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Abstract

Water is the basic unit for sustaining healthy plant life. Limited or unavailability of water leads to ultimate plant death. Drought severely affects the yield of wheat, especially when it occurs at the critical growth stages like tillering and anthesis. Therefore, attempts have been and being made to develop measures to combat the drought effects in wheat. This study investigated the sole and/or integrated use of Iron (Fe) and Zinc (Zn) spray on growth and yield of Wheat under drought stress. Drought was created at the tillering and anthesis stages by withholding the irrigation. At both these stages, different doses of Zn and Fe were applied alone and/or in combination. Results revealed that drought stress significantly ($P<0.05$) reduced the 1000- grain weight (30 vs. 42g), number of grains per spike (27 vs. 62), spike length (11 vs. 17cm), number of spikelets per spike (22 vs. 9) physiological (leaf chlorophyll content, stomatal conductance & photosynthetic rate) and water related traits. Combine application of Fe and Zn to wheat under drought significantly increased the 1000 grain weight (40g), seed yield (5350 kg/ha), relative water content, leaf osmotic potential, leaf water potential, protein content (16%) and spike length (16cm). The Zn was more effective in mitigating the ill impacts of drought as compared to the Fe alone. While, combine spray of both Zn and Fe, gave best results as compared to the sole application of either nutrient. Results of this study lead us to conclude that both Zn and Fe have the ability to compensate the detrimental effects caused by drought stress and their combine use can be more useful as compared to their sole usage.

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Introduction

Wheat (*Triticum aestivum* L.) is considered as one of the most important cereal crop and staple food in many countries of the world including Pakistan. It is used to make roti, nans, bread, cookies, pasta and other products. It is the cheapest source of human calories and protein content in the average diet (Lev-Yadun *et al.*, 2000). Among the all cereal crops, wheat is the most favorite food for human being and as feed for animals also (Vaughan and Judd, 2003).
Wheat grains contain 9.4-14 g/100g protein, 1.8-2.3 g/100g fiber, 1.2-2.5 g fat and have 69.1-75.4 g/100g available carbohydrates (Ken, 2004). Wheat provides about 55 percent of the carbohydrates and 20% of the food calories as consumed globally (Breiman and Graur, 1995). Wheat is grown under different variety of soil and climatic conditions in Pakistan. From the total acreage, about one third falls under rain fed areas where annual amount and intensity of rainfall is very low (Khanzada *et al.*, 2001). Drought stress makes threats to the agricultural production in the majority of the areas of world (Abolhasani and Saeidi, 2004), as 20,000,000 km^2 of the lands throughout the world lies in semi-arid regions (Alyari and Shekari, 2000).

Drought causes a drastic effect on plant productions each year in the different regions of the world. There are many reasons for the low availability of water to the plants not only due to high salinity conditions and high temperature of the surroundings but also due to the deviation in the normal rainfall pattern of an area (Baybordi, 2004; Soriano *et al.*, 2004). Drought stress is characterized by reduction of water content, turgor loss, very low leaf water potential, leaf stomata opening and closing, reduction in cell enlargement and decline in growth occurs. In the harsh conditions of water deficiency photosynthesis process in plants may stop which leads to the interruption of metabolism and at last the death of plant (Jaleel *et al.*, 2008). Shortage of water in soil also causes very slow movement of micro-nutrients like as Iron and Zinc in soil and plants. This disturbs the growth and many physiological processes of plants (Chimenti *et al.*, 2002).

The chlorophyll production and quantity of abscisic acid are affected by the utilization of Zn and Fe (Jiang and Huang, 2002). The increase in the quantity of chlorophyll ultimately enhances yield during the light reaction of photosynthesis (Conversion of carbon dioxide and water into glucose). Starch accumulation in the grain can be suppressed due to the limited availability of water during the process of assimilates translocation at the time of grain filling (Yang *et al.,* 2004). In the same way, grain weight and spike length are also adversely affected due to water deficit condition (Wang *et al.*, 2005). Less grain weight under drought conditions is the main reason of reduced seed yield of wheat (Baybordi, 2004).

Many management practices are used to improve the drought tolerance in wheat like stubble or residual mulches (Ahmad *et al.*, 2015), use of osmolytes (Raza *et al.*, 2012a) or solutes like spray of potassium (Raza *et al.*, 2012b), gibberelic acid and amino acid application and use of micronutrients (Iron, Zinc, Copper, etc). Studies on micronutrients have proved that they can be used efficiently in crop plants for mitigating the impact of different kinds of stresses like water stress. Iron and Zinc are among those micronutrients. Both the micro and macro nutrients are importance for plant nutrition and low quantity of any nutrient can badly effect the production of crop (Murphy *et al.*, 2008). Unbalanced application of phosphorus and nitrogen produce the low quality of grain and these fertilizers have also reduced the micronutrients uptake by plants (Maralian, 2007-2008). So, there is a need to check the effects of micronutrients application on plants. Micronutrients can also increase dry matter production, total straw yield, and grain yield of wheat (Asad and Rafique, 2002).

Many researchers have reviewed Zinc deficiency in the soils of Pakistan (Hadi *et al.*, 1997). Amongst the micronutrients, Zinc scarcity takes place both in humans and crop plants (Hotz and Brown, 2004). Latest reports indicated that almost 5000, 000 children die during 5 years of age per annum due to insufficiency of low Zn and Fe in their diet (Black *et* *al*, 2008). Zinc sulphate fertilizer is the best one among the fertilizers needed by plants due to its performance on stomatal adjusting according to the changing environment and uptake of nutrients and ions by plants. So, there is need to pay attention during application of such kind of fertilizers either water is sufficient or inadequate (Karam *et al.*, 2007; Babaeian *et al.*, 2010).

Unbalanced use of synthetic fertilizers and prolonged drought are the main factors for low amount of Iron and Zinc in the soil solution. Seed yield and protein percentage in the plants increases with the application of Iron (Yahyavi *et al.*, 2004). Seed yield improved due to the utilization of Iron, which affects photosynthesis in plants, and ultimately there will enhance of effectiveness of dry matter substances and reallocation of photosynthates into the economic portion of wheat. The growers in the arid areas face the problem of Iron deficiency due to high pH of soils. Pakistani soils are high in pH which results reverse effects of Iron and Zinc with the other nutrients and consequently in the presence of other plant nutrients Zn and Fe availability to plants is less. Under drought stress conditions, we believe that the yield losses can be minimized by the sufficient supply of Zn and Fe. The objective of present work was to study the possible role of Zn and Fe applied on wheat foliage at different growth stages under drought, in mitigation of stress in terms of physiological components and nutrients uptake.

Materials and methods

To explore the objective as outlined above, a field experiment on clay-loam soil was conducted at agronomic research area, University College of Agriculture and Environmental Sciences, The Islamia University of Bahawalpur to investigate the effect of foliar application of iron and zinc on wheat growth, physiology and yield under drought stress. Soil analysis carried out prior to sowing revealed EC = 2.04 dsm-^{1}, pH = 8.6, O.C(%) = 0.79, soil P_2O_5 = 5 ppm, K_2O = 115 ppm, N= 0.109, field capacity =21% W/W, wilting point = 10% W/W and the volume weight of the soil was 1.22 g.cm^{3}.

The experiment was laid out in randomized block design with four replications. Treatments included were T_0=Control, T_1=Drought, T_2=Zn (150 g ha^{-1}), T_3=Fe (150 g ha^{-1}), T_4=Zn+Fe (150+150g ha^{-1}), T_5= Zn+Fe (200+150 g ha^{-1}) and T_6= Zn+Fe (150+200 g ha^{-1}).The foliar spray of Fe and Zn was done at tillering and anthesis after giving drought at both stages. Zinc was applied as zinc sulphate and Iron as Iron oxide both were applied through foliar application and at a rate of 150g/ha. Additionally, the wheat crop received P and N at a rate of 112 kg ha^{-1} o and 150 kg ha^{-1}, respectively. Total amount of P was mixed in soil while soil preparation, whereas N was applied in two splits: at the time of soil preparation and at heading stage of wheat crop. Wheat seeds were sown in each plot with size of 5m x 3m with hand drill having 22.5 cm row to row distance. Leaf chlorophyll contents were measured by chlorophyll meter. Stomatal resistance/conductance measurements was made with an automatic porometer MK-3 (Delta-T Devices, Burwell Cambridge, England) Hertford, Herts, England) while Photosynthetic rate was recorded by using the an infrared gas analyzer (Li-COR-LI 6250). Relative water content (RWC) was calculated by means of the following formulas, RWC (%) = FW- DW/ TW- DW X 100.

For recoding of leaf water potential (-MPa), an entirely stretched youngest leaf (fourth from the top) was cut out from every plant and calculations were made with a water potential apparatus (Chas W. Cook Div., England). To compute the leaf osmotic potential (-MPa), a fraction of the leaf used for water potential purpose will be frozen for two weeks. After that, frozen leaf thawed and crushed using metal rod to take out the frozen sap. After centrifugation (8000 x *g*) for 4 minutes, the sap will be used for osmotic potential determination in a Vapor Pressure Osmometer (Wescor 5520, Logan, USA).

At maturity when moisture content in the seed was about 14%, area of one square meter in each plot was randomly selected using a quadrant. Thereafter, the plants in this area were manually harvested to estimate, grain yield biological yield, no. of spikes per surface unit, number of grains per spike, number of spikelets per spike, length of the spike, height of the plant and stem, and thousands kernels.

The grains from each treatment were oven-dried at 70 °C and ground to pass a 1mm sieve. Dried ground form of wheat grain (0.1 g) was taken in digestion tubes and 5ml of concentrated sulphuric acid (H_2SO_4) was added to each digestion tube and incubated overnight at 25°C. Then 1 ml of hydrogen peroxide (H_2O_2) (34%) was poured into the sides of the digestion tube, ported the tubes in a digestion block and heated at 350°C, until fumes were produced. They were continued to heat for half an hour. The digestion tubes were removed from the block and cooled down. Then 1 ml of H_2O_2 was added and the tubes were kept back into the digestion block. The above step was repeated until the cooled digested material was colorless. Thereafter about 0.1 g was used for the determination of total N content by Kjeldahal's method and later converted into protein content by multiplying N content with a factor of 5.70 (ISO, 2009). The K^+ in the digests was determined with the help of flame photometer and phosphorus was determined by spectrophotometer at 420 nm using standard curve method (Lindner, 1944). Collected data regarding all the parameters was analyzed by using Fisher's analysis of variance technique and LSD test at 5% probability was used to compare the differences among treatments (Steel *et al.,* 1997).

Results

Growth & Physiological Related Parameters

Results revealed that no. of plants m^{-2}, plant height and number of fertile tillers m^{-2} were not significantly affected under drought stress by foliar application of Iron (Fe) and Zinc (Zn) (Table 1).

Table 1. Variance analysis and means compares of drought stress and effects of foliar application of Iron and Zinc on wheat (*Triticum aestivum* L.) growth components.

Treatments	Plant Population (m^{-2})	No. of fertile tillers(m^{-2})	Plant height (cm)	Spike length (cm)	Number of spikelets per spike
T_0=Control	210 a	421a	98 a	16.83 a	21.67 a
T_1=Drought	198 b	396 b	86 b	10.50 g	9.33 g
T_2=Zn(150 g ha^{-1}) as $ZnSO_4$+Drought	200 b	403 b	93ab	12.50 e	13.33 e
T_3=Fe(150g ha^{-1}) as Fe_2O_3+ Drought	206 ab	415 ab	93 ab	11.58 f	11.33 f
T_4=Zn+Fe (150+150g ha^{-1}) +Drought	205 ab	413 ab	91 ab	13.67 d	15.33 d
T_5=Zn+Fe (200+150g ha^{-1}) +Drought	204 ab	413 ab	95 a	16.00 b	19.67 b
T_6=Zn+Fe (150+200g ha^{-1}) +Drought	200 ab	418 ab	93 ab	15.00 c	17.33 c

Significant results at 0.05 % probability.

Spike length was significantly affected by foliar applied micronutrient treatments (Table 1). Highest spike length was obtained in control treatment (T_0) and minimum in drought stressed treatment (T_1).

Treatment in which zinc (T_2) and iron (T_3) were applied solely produced more lengthy spikes than T_1. Combine application of micronutrient (T_4, T_5 and T_6) produced more lengthy spikes as compared to their sole application (T_2 and T_3). Among the combine micronutrient treatments, T_5 produced lengthiest spike.

Foliar applied micronutrient showed significant effects on number of spikelets per spike (Table 1). Maximum number of spikelets per spike (21.67) was obtained in control treatment (T_0) and minimum in drought stressed treatment (T_1). Treatment in which zinc (T_2) and iron (T_3) were applied solely produced more number of spikelets per spike than T_1. Combine application of micronutrient (T_4, T_5 and T_6) produced more number of spikelets per spike as compared to their sole application (T_2 and T_3). Among the combine micronutrient treatments, T_5 produced maximum number of spikelets per spike (19.67).

Table 2. Variance analysis and means compares of drought stress and effects of foliar application of Iron and Zinc on wheat (*Triticum aestivum* L.) physiological components.

Treatments	Leaf chlorophyll contents (%)	Stomatal conductance (mmolm^{-2}s^{-1})	Photosynthetic rate (μmolm^{-2}s^{-1})	Leaf relative water contents (%)	Leaf osmotic potential (MPa)	Leaf water potential (MPa)
T_0=Control	30.00 a	360a	15.50 a	84.50 a	1.80 a	1.12 a
T_1=Drought	14.00 g	220 g	7.03 g	40.00 g	1.11 g	0.22 g
T_2=Zn(150 g ha^{-1}) as $ZnSO_4$+Drought	20.00 e	290 e	12.03e	65.00 e	1.42 e	0.65 e
T_3=Fe(150g ha^{-1}) as Fe_2O_3+ Drought	17.50f	275 f	11.80 f	60.00 f	1.35 f	0.58 f
T_4=Zn+Fe (150+150g ha^{-1}) +Drought	22.13 d	320 d	12.80 d	70.80 d	1.62 d	0.85 d
T_5=Zn+Fe (200+150g ha^{-1}) +Drought	27.50 b	338 b	14.20 b	78.50 b	1.72 b	0.98 b
T_6=Zn+Fe (150+200g ha^{-1}) +Drought	23.50 c	330 c	13.40 c	75.50 c	1.68 c	0.92 c

Significant results at 0.05 % probability.

A significant effect of foliar applied micronutrient was observed on leaf chlorophyll contents (Table 2). More leaf chlorophyll content was obtained in control treatment (T_0) and minimum in drought stressed treatment (T_1). Solely applied zinc (T_2) and iron (T_3) produced more leaf chlorophyll content (%) than T_1. Combine application of micronutrient (T_4, T_5 and T_6) gave more leaf chlorophyll content than their sole application (T_2 and T_3). Among the combine micronutrient treatments, T_5 produced maximum leaf chlorophyll content.

Stomatal conductance (mmolm^{-2}s^{-1}) was significantly affected by foliar applied micronutrient (Table 2). The higher stomatal conductance (mmolm^{-2}s^{-1}) was obtained in control treatment (T_0) and minimum in T_1. Sole application of zinc (T_2) and iron (T_3) resulted in more stomatal conductance (mmolm^{-2}s^{-1}) than T_1. Combine application of micronutrient (T_4, T_5 and T_6) produced more stomatal conductance (mmolm^{-2}s^{-1}) as compared to their sole application (T_2 and T_3). Among the combine micronutrient treatments, T_5 produced highest stomatal conductance (mmolm^{-2}s^{-1}). Table.2 indicates that foliar applied micronutrient had statistically significant effect on photosynthetic rate of wheat. The maximum photosynthetic rate (μmolm^{-2}s^{-1}) was observed in control treatment (T_0) and minimum in T_1. Sole application of zinc (T_2) and iron (T_3) resulted in higher photosynthetic rate (μmolm^{-2}s^{-1}) than T_1. Combine application of micronutrient (T_4, T_5 and T_6) showed more photosynthetic rate (μmolm^{-2}s^{-1}) as compared to their sole application (T_2 and T_3). Among the combine micronutrient treatments, T_5 produced highest photosynthetic rate (μmolm^{-2}s^{-1}).

A significant effect of foliar applied micronutrient was observed on all water related traits of wheat i.e. leaf relative water content (%), leaf osmotic and leaf water potential (MPa) (Table 2). Higher water related parameters were obtained in control treatment (T_0) and minimum in drought stressed treatment (T_1). Solely applied zinc (T_2) and iron (T_3) showed more water related traits than T_1. Combine application of micronutrient (T_4, T_5 and T_6) gave more values of relative water content (RWC), leaf osmotic potential and leaf water potential than their sole application (T_2 and T_3). Amongst the combine micronutrient treatments, T_5 showed higher rates of all water related traits of wheat.

Yield Related Parameters

Results showed that contents of phosphorus and potassium in wheat grain were not significantly affected under drought stress by foliar application of Iron (Fe) and Zinc (Zn) (Table 3).

A significant effect of foliar applied micronutrient was observed on number of grains per spike (Table 3). The highest number of grains per spike was obtained in control treatment (T_0) and minimum in drought stressed treatment (T_1). Solely applied zinc (T_2) and iron (T_3) produced more number of grains per spike than T_1. Combine application of micronutrient (T_4, T_5 and T_6) gave more number of grains per spike than their sole application (T_2 and T_3). Among the combine micronutrient treatments, T_5 produced maximum number of grains per spike. 1000-grain weight was significantly affected by foliar applied micronutrient (Table 3). The maximum 1000-grain weight was obtained in control treatment (T_0) and minimum in T_1. Sole application of zinc (T_2) and iron (T_3) resulted in more 1000-grain weight than T_1. Combine application of micronutrient (T_4, T_5 and T_6) produced maximum 1000-grain weight as compared to their sole application (T_2 and T_3). Among the combine micronutrient treatments, T_5 produced highest 1000-grain weight. Foliar applied micronutrient also showed significant effects on grain yield of wheat (Table 3).

Table 3. Variance analysis and means compares of drought stress and effects of foliar application of Iron and Zinc on wheat (*Triticum aestivum* L.) yield and yield components.

Treatments	No. of grains per spike	1000 grain weight (g)	Grain Yield (kg/ha)	Phosphorus content (%)	Potassium content (%)	Protein content (%)
T_0 = Control	62.33 a	42.33 a	5500.00 a	0.38 a	2.3 d	11.55 g
T_1= Drought	27.66 g	30.33 g	3916.67 g	0.38 a	2.7 ab	12.57 f
T_2= Zn(150g ha^{-1}) as $ZnSO_4$+Drought	39.66 e	33.66 e	4350.00 e	0.39 a	2.2 d	13.81 d
T_3=Fe(150g ha^{-1}) as Fe_2O_3+Drought	33.66 f	32.00 f	4016.67 f	0.41 a	2.3 cd	13.14 e
T_4=Zn+Fe(150+150g ha^{-1}) +Drought	44.66 d	35.66 d	4716.67 d	0.42 a	2.4 bcd	14.32 c
T_5=Zn+Fe(200+150g ha^{-1}) +Drought	57.33 b	40.33 b	5350.00 b	0.43 a	2.7 abc	16.05 a
T_6=Zn+Fe(150+200g ha^{-1}) +Drought	51.00 c	37.66 c	5016.67 c	0.39 a	2.9 a	14.91 b

Significant results at 0.05 % probability.

The maximum grain yield (5500.00 kg/ha) was obtained in T_0 and minimum (3916.67 kg/ha) in T_1. Treatment in which zinc (T_2) and iron (T_3) were applied solely produced more grain yield than T_1. Combine application of micronutrient (T_4, T_5 and T_6) yielded more grain than their sole application (T_2 and T_3). Among the combine micronutrient treatments, maximum grain yield (5350.00 kg/ha) was obtained from T_5.

Grain protein content was significantly affected by foliar applied micronutrient (Table 3). Maximum grain protein content was obtained in T_5 and minimum inT_0.

Treatment in which zinc (T_2) and iron (T_3) were applied solely produced more grain protein content than T_1. Combine application of micronutrient (T_4, T_5 and T_6) produced more grain protein content as compared to their sole application. Among the combine micronutrient treatments, T_5 produced highest grain protein content.

Discussion

Water deficit stress affects the growth, physiological, yield and quality parameters of wheat. Application of micronutrients (Fe and Zn) either alone or in combination was reported to be an effective technique to minimize the adverse effects of drought. Spike length of wheat adds enormous share in ultimate economic yield because it has considerable influence on number of spikelets, grain size and number of grains per spike. Drought affected wheat plants produced smaller spikes as compare to the proper irrigated plants (Taheri *et al.*, 2011). It is due to less turgor pressure, leads to less cell division and less growth (Jaleel *et al.*, 2009). Abbas *et al.* (2009) found that foliar applied Zinc (Zn) has a positive effect on spike length of wheat under water deficit conditions due to the role of Zn is in auxin production which is involve in cell division and cell elongation. Fe also has a significant effect on spike length of wheat, as Fe is helpful in chlorophyll formation which results in more photosynthesis and more growth. Combine application of Zn and Fe can result in the formation of longer spikes (Kholdebarin and Islamzadeh, 2002; Monjezi *et al*., 2013; Jam *et al*., 2012). It might be due to synergistic effect of both nutrients (Gaffar *et al*., 2011).

Chlorophyll concentration in plant leaves display its capacity to make photosynthates (Zhang *et al.,* 2007). Process of photosynthesis is badly affected by concentration of chlorophyll. This concentration is mostly lower under drought stress conditions due to the reduction in activity of chlorophyllase enzyme which is mainly involved in the formation of chlorophyll (Ashraf *et al.,* 2013). Simultaneous application of both iron and zinc produced more concentration of chlorophyll in wheat crop as compared to drought stress. Similar results were also propsed by Khakwani *et al*. (2013) in fully irrigated crop (more chlorophyll concentration) as compared to water deficit conditions. Iron is directly involved in the formation of chlorophyll.

Stomatal conductance is an imperative measurement of plant responses under drought or low availability of water (Baloch *et al*., 2012). Synergistic effect of both iron and zinc is helpful for crop plants in water stress conditions (Gaffar *et al*., 2011). These nutrients are involved in the controlling of stomatal aperture by producing auxin in this way a resistance may be created under drought stress conditions. Process of photosynthesis computes the overall growth and yield of crop plants. Under severe water conditions and low relative water content of leaves there is very slow rate of photosynthesis (Alkhaldi *et al*., 2012). Stomatal conductance is directly related to the formation of glucose in photosynthesis.

Relative water content of plant leaves is the indication of presence of water in tissues of plant leaves. Computation of drought is also carried out through it (Sanchez-Blanco *et al*., 2002). Control treatment showed higher values of relative water content due to more leaf area and cell volume in relation to drought (Stikic *et al*., 2003). Combine use of zinc and iron showed better results than their sole application and drought. Low RWC is also attributed with the root systems of various cultivars and their capability to absorb the soil moisture in water deficit conditions (Khakwani *et al*., 2013). Leaf water and osmotic potentials have positive influences on the overall growth and physiology of crop plants. Leaf water potential shows the potential energy of water in leaf tissues. Simultaneously applied micronutrients improved the water related traits of wheat due to their synergistic effects as zinc is directly involved in the production of auxins in plants and iron is the main functional element in the formation of chlorophyll. Changing water potential of various varieties is might be due to the alteration in osmotic pressure of crop plants (Siddique *et al*., 2000).

Potential of wheat production is estimated by the number of spikelets per spike. Number of spikelets per spike of wheat add huge share in final economic yield. Less number of spikelets per spike under water deficit conditions is mainly due to less spikelets primordial formation during tillering stage or may be due to death of floret at the terminal and basal ends of the spike (Maqbool *et al*., 2015). Abbas *et al*. (2009) noted significant effect of different levels of Zn on number of spikelets per spike in wheat under water deficit conditions. Zn is more efficient on the formation of spikelets and/or the increase in the number of spikelets per spike due to role of Zn in the production of growth promoting hormone auxin and its role in many metabolic activities (Hafeez *et al*., 2013). Foliar applied Fe also increased the number of spikelet per spike in drought affected wheat as compared to untreated plants due to more photosynthesis (Abbas *et al*., 2009). However combined application of Fe and Zn produced more number of spikelets per spike, might be due to synergistic effects of both nutrients (Zeidan *et al*., 2010).

Number of grains per spike is the main factor which particularly contributes to the economic yield of wheat. The number of grains per spike is the major factor which affects the grain yield. Under water deficit stress number of grains decreased mainly due to the dehydration of pollen grain (Khanzada *et al*., 2001). Micronutrients had a significant effect on the number of grains per spike (Jam *et al.*, 2012). Foliar application of micronutrients enhanced the number of wheat grains per spike (Khan *et al.*, 2010). Zeidan *et al.* (2010) reported that micronutrients Fe and Zn alone application in different treatments increase the number of wheat grains per spike than the control treatment through positively affecting the fertility of flowers (Monjezi *et al.*, 2013).

1000-grain weight indicates the degree of the formation of grain in wheat crop. Treatment in which proper irrigation was applied produced more 1000-grain weight than drought affected plants. More 1000-grain weight with proper irrigations is due to more translocation of assimilates towards grain because of availability of sufficient amount of water in root zone (Maqbool *et al.*, 2015). Alone application of either Fe or Zn give higher 1000-grain weight than without application of these nutrients (Zeidan *et al.*, 2010).The increase in 1000-grain weight of wheat by the simultaneous application of Fe and Zn has been reported by Ziaeian and Malakoti (2002) and El-Majid *et al.* (2000). The increase in grain weight was resulted from the positive effects of these micronutrients on post-anthesis photosynthesis and the allocation of dry matter to grains which finally led to the increase in 1000-grain weight. Similar results were reported by Monjezi *et al.* (2013) and Jam *et al.* (2012).

Seed yield is the result of various yield contributing parameters like spike length, number of spikelets per spike, number of grains per spike, and grain weight. Drought stress decreases the grain yield of wheat. Less grain yield under water deficit stress is due to less translocation of photosynthates from the leaves and as drought stress faster the maturation process. More seed yield of wheat with foliar application of Fe is mainly due to its role in enzymatic reactions and photosynthesis, which ultimately result in higher seed yield (Habib, 2009). More seed yield with application of Zn is due to its role in production of protein, carbohydrates, cell metabolism, protection of cell organelles and other processes linked to make the plants well-matched with water shortage conditions (Yari *et al.*, 2005). These results are in lined with the findings of Yilmaz *et al.* (1997), El-Majid *et al.* (2000) and Zeidan *et al.* (2010). Synergistic effect of both Zn and Fe results more grain yield as compared to their sole application (Gaffar *et al.*, 2011).

Drought stress at both tillering and anthesis stages shortened the grain filling stage of wheat, which in turn, increased the grain protein as compared to control treatment due to the increased accumulation of hydrocarbons (Monjezi *et al.*, 2013). Drought affected plants treated with Zn has more grain protein contents due to the positive effect of Zn on important plant processes, such as nitrogen metabolism and nitrogen uptake by the plants (Potarzycki and Grzebisz, 2009). Drought affected plants treated with Fe also has more grain protein contents than control treatment. Seadh *et al.* (2009) reported that foliar applied Fe @ 500 ppm on wheat enhanced grain protein content over untreated wheat plants. Micronutrients (Iron and Zinc) either alone or in a combination had significant effect on grain protein content of wheat (Zeidan *et al.*, 2010). Similar results were found by Jiang *et al.* (2002) and Monjezi *et al.* (2013). It is obvious that the foliar application of Iron and Zinc increased the grain protein contents but at the same time reduced the concentration of phosphorus and potassium contents of wheat grain due to low uptake of these nutrients through roots (Zeidan *et al.*, 2010).

Conclusions

Foliar applied Iron and Zinc significantly improved the growth, physiology, yield and grain protein contents of wheat (*Triticum aestivum* L.) under drought stress conditions. Combined application of Zn + Fe (200+150 g ha^{-1}) under drought at tillering and anthesis performed better in improving growth, physiological, yield related parameters and quality of wheat.

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