NARDI FUNDULEA, ROMANIA ROMANIAN AGRICULTURAL RESEARCH, NO. 40, 2023
www.incda-fundulea.ro; doi.org/10.59665/rar4037 Print ISSN 1222-4227; Online ISSN 2067-5720

ENHANCING GRAIN YIELD AND ZINC CONTENT IN BREAD WHEAT USING ZINC AND NITROGEN APPLICATION UNDER SUPPLEMENTARY IRRIGATION TREATMENTS

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ABSTRACT

Wheat (*Triticum aestivum* L.) is one of the crops that has great importance in human and animal nutrition. Nutrient management can improve the nutritional value and grain yield in bread wheat. Therefore, this study was conducted to assessment the effect of Zinc (Zn) levels as well as the interactions with Nitrogen (N) and irrigation regimes on yield, yield components and Zn content in bread wheat grain. To this purpose, an experiment was performed in a split-split plot design with four replications, which supplementary irrigation (no irrigation, irrigation at stem elongation and irrigation at the stage of grain filling) as the main plot, Zn levels as split-plot (no spraying of Zn and Spraying Zn with a concentration of 5%) and nitrogen consumption levels (nitrogen-free, half the normal dosage and normal nitrogen consumption) as split-split factors. The results showed that the grain yield and yield components affected by the applied treatments. The highest grain yield (646 g/m^2) and Zn concentration (32.69%) observed in spraying Zn at 5% concentration supplemented with irrigation at the grain filling stage with normal N application. Also, Zn content of grain was increased by Zn foliar application. Furthermore, the grain yield significantly correlated with Zn content, Harvest index (HI), seed filling rate (SFR), thousand seed weight (TKW) and seed number per spike (SNS) traits. The highest grain yield was obtained in supplementary irrigation in grain filling stage indicating importance of supplementary irrigation in this stage. In general, the results of this study showed that the grain yield and quality in bread wheat could be increased by supplementary factors.

Keywords: bio-fortification, bread wheat, Zn, irrigation, foliar application.

INTRODUCTION

Wheat is one of the crops that have been great importance in human and animal nutrition for thousands of years (Shiferaw et al., 2013). Fertile regions of warm climate in the north of the Iran, including the plains of Gorgan, Gonbad Kavus, Mazandaran and Moghan are important areas of wheat production in the country (Faramarzi et al., 2010).

Wheat is an economical source of energy, especially for the people of the Third World regions (Mughal et al., 2020). Wheat growing areas in developing countries are affected by water (Moayedi et al., 2010) and nutrient (Clair and Lynch, 2010) deficit conditions.

Efficient agronomical methods such as proper utilization of available water including interaction of water and nutrient usage that allow the development of plant cultivation in arid areas (Hajjarpoor et al., 2021). Nutrients can be provided to the plant in the shortest possible time by feeding through the leaves. Foliar nutrition is a good way to reduce the use of chemical fertilizers and reduce their environmental risks. Nitrogen is the fourth major constituent of plant dry weight and interacts with water consumption (Hopkins, 2004). Zn acts as a cofactor for enzymes is also needed for the biosynthesis of a large number of proteins (Alloway, 2008). Zn is essential for the synthesis of tryptophan, a precursor to the synthesis of melatonin, nicotinic acid, and auxin (Pedler et al., 2000).

Received 30 November 2022; accepted 23 February 2023. First Online: February, 2023. DII 2067-5720 RAR 2023-108

The importance of Zn in human cell metabolism is determined by the effects of Zn deficiency, including decreased immune system function, reduced repair and regeneration, and neurological disorders (Baltaci et al., 2018).

Supplementary irrigation can lead to sustainable yield in wheat and is a determining factor in increasing wheat yield (Yaghini and Narimani, 2020). The importance of nutrients such as N and Zn as an essential element for plant nutrition and its decisive role in plant growth and especially in increasing wheat yield has been proven (Toor et al., 2021). The exact determination of nitrogen in water shortage conditions has not yet been confirmed. In dry areas, the amount of fertilizer is applied based on the humidity regime of the region, so in these areas, the amount of nitrogen fertilizer should be given in a balanced way, because lack or excess of nitrogen fertilizer reduces water use efficiency (Zhong and Shangguan, 2014).

One of the recommended strategies is supplementary irrigation and optimal nitrogen consumption, both of which are limiting factors for wheat yield in rainfed areas (Oweis and Hachum, 2006). Wang et al. (2004) in their study of the effects of supplementary irrigation on wheat concluded that a timely irrigation phase increased nitrogen uptake and decreased residual nitrogen in the soil.

Tavakoli and Oveys (2004) concluded in their experiment that the crop response to nitrogen at the level of 60 kg/ha was significant. The maximum water use efficiency is obtained from 60 kg/ha nitrogen and one third of full irrigation.

The importance of nitrogen in increasing yield has been proven, but the exact amount of nitrogen in the climatic conditions of the region and the cultivars used have not yet been clarified. Nitrogen is a key factor in achieving optimal performance in cereals. Wheat usually needs a lot of absorbable nitrogen during its growing season. Increasing nitrogen consumption increases leaf area, tiller formation, leaf area index and leaf area durability and this increase leads to the production of more dry matter and grain yield (Yiotis et al., 2021). Hera et al. (2018) in a study investigated the effect of Zn foliar application on different wheat traits under drought stress conditions. The results showed that foliar application eliminated the negative impact of dehydration stress. Former studies showed that Zn foliar spray enhanced water use efficiency and grain yield in wheat (Karim et al., 2012; Umair Hassan et al., 2020; Anwar et al., 2021).

The main goal of this study is to investigate the effect of Zn levels as well as the interactions with N and irrigation regimes on yield, yield components and Zn content in wheat grain.

MATERIAL AND METHODS

Plant material and experimental conditions

In this study, a bread wheat breeding line named 91-17, which is released named Kalateh and is one of the best spring genotypes in the region was used as plant material. The study was conducted in the research farm of Imam Khomeini Agricultural high school located in Aliabad Katoul in Golestan province, Iran during 2018-2019 and 2019-2020 cropping seasons. Aliabad Katoul with an average annual rainfall of 500-600 mm, an altitude of 140 meters above sea level and an average temperature of maximum 18°C and minimum 3°C at latitude 36’54’’N and longitude 54’52’’E has a temperate and humid climate (Table 1).

Before the sowing of the plants, different samples of the soil from a depth of 0-30 cm were collected by zigzag method and sent to the laboratory for analysis, the results and characteristics of the soil showed in the Table 2.

Table 1. Maximum, minimum, and mean monthly air temperature, rainfall and reference evapotranspiration (ET0) in 2018-2019 and 2019-2020

Year	Month	T_{max} (°C)	T_{min} (°C)	T_{mean} (°C)	Rain (mm)	ET_o (mm)
2018	December	16.0	7.8	11.9	51.2	38.3
2019	January	15.2	4.7	10.0	120.7	36.3
	February	13.6	4.3	9.0	158.9	30.7
	March	16.7	5.9	11.3	148.5	61.9
	April	18.5	9.7	14.1	93.2	56.9
	May	25.8	13.8	19.8	38.0	113.7
	June	33.4	20	26.7	7.8	215.6
	December	15.2	5.8	10.5	30.3	25.2
2020	January	14.3	4.6	9.5	26.3	33.5
	February	14.5	3.4	8.9	91.6	41.1
	March	16.3	6.7	11.5	58.3	51.2
	April	16.2	8.1	12.2	101.1	40
	May	24.8	13.2	19.0	60.4	121.3
	June	33.8	18.6	26.2	13.2	219.9

Table 2. Physiochemical characteristics of the soil of experimental field (0-30 cm depth)

Soil texture	K (ppm)	P (ppm)	Total N (%)	Organic matter (%)	PH	EC (dSm^{-1})
Silty-Clay-Loam	228	4.2	0.16	1.56	7.6	1.4

Experimental design

The experiment was performed in the field using a split-split plot design arranged in a completely randomized block with four replications in which supplementary irrigation levels (I_0, I_1 and I_2) as the main plot, Zn levels (Z_0 and Z_1) as split-plot and nitrogen levels (N_0, N_1 and N_2) as split-split factors (Table 3).

Table 3. P Treatments and their level

Factors	Treatments	
Irrigation	No irrigation	I_0
	Irrigation at stem elongation	I_1
	Irrigation at the stage of grain filling	I_2
Zn	No spraying on Zn	Z_0
	Spraying Zn with a concentration of 5%	Z_1
Nitrogen	Nitrogen-free	N_0
	Half the normal dosage	N_1
	Normal Nitrogen consumption	N_2

To conduct this research, first in November, the earth was ploughed with a reversible plough and in December, the earth was disked in two stages. After ploughing the earth by the Pythagorean method, the plan was executed by wooden nails and thread, then the recommended amount Phosphorus fertilizers (220 kg/ha), potash (130 kg/ha) and a quarter of nitrogen (62.5 kg/ha), (considering that the area of the plots was 8 m^2, the amount of nitrogen fertilizer in each stage, the recommended amount of 50 g/plot and half of the recommended amount (25 g/plot) based on soil analysis, was poured into the plots before planting and planted on December 11, 2018 and December 2, 2019. sowing was done by a linear grain milling machine at the rate of 135 kg/ha of the seeds. The distance between the repetitions was 3 m and the distance between the main plots was 1.5 m and the sub-plots were 1 m.

Nitrogen fertilizer was used in four stages (before planting, tillering, stem elongation and seed filling stage) as road, irrigation by pressurized system and type tape and also for Zn foliar application of 2-liter manual sprayers. Weed control was done in the tillering stage by 2,4-D and MCPA for broadleaf grasses and clodinaphobic propargyl for narrow leaves. Also, in April and May, the field was treated in two stages with tilt fungicides. And follicle spraying was done, to fight pests in May, diazinon and phenytoin was used.

Measurement of the traits

The measured traits were included plant height (PH), days to maturity (DM), seed filling rate (SFR), thousand seed weight (TKW), seed number per spike (SNS), Zn content of grain (ZNg), Zn content of shoots (ZNs), grain yield (GY), harvest index (HI).

To measure yield and yield components after removing the margins, plants harvested from 1 m^2 inside the plots and the grain yield was measured based on g/m^2. To measure PH, from the soil surface to the tip of the main spike without measuring the awn was recorded at physiological maturity and measured in centimetres. HI was calculated by dividing grain yield by biological yield. The following equation was used to calculate grain filling rate.

Grain filling period / grain yield = grain filling rate

To measure the SNS, a sample consisting of 5 main spikes that were randomly sampled from each experimental plot and the desired traits were measured for each of the 5 samples and their average was used for statistical calculations. The Zn content in the leaves and seeds was determined by atomic absorption spectrometry and dry ash methods (Ferreira et al., 2010).

Statistical analysis

Analysis of variance (ANOVA) was performed by SAS ver. 9.4, the means were compared by LSD at 5 level of probability and graphs and tables were drawn using Microsoft Excel 2019.

RESULTS AND DISCUSSION

Grain yield and its components

Results showed that the planting date, plant density, cultivar types, as well as the interaction effects of cultivar types × planting date, cultivar types × plant density, and cultivar types × planting date × plant density had a significant effect on the trait of the number of lateral branches (Table 3).

Results of the analysis of variance showed that yield and yield components affected by the applied treatments (Table 4). DM, SNS and SWS affected by irrigation and nitrogen. Zn foliar, N and irrigation significantly affected on grain Zn content in bread wheat in the present study.

Grain yield and Zn content increased by Zn foliar application and supplementary irrigation in grain filling stage under different treatments. However, changes in these traits were varied in N application under other treatments (Figure 1). The highest grain yield (646 g/m^2) and Zn concentration (32.69 mg/kg) observed in $Z_1N_2I_2$ treatment (Zn 5% + normal N + irrigation in grain filling stage) (Figure 1).

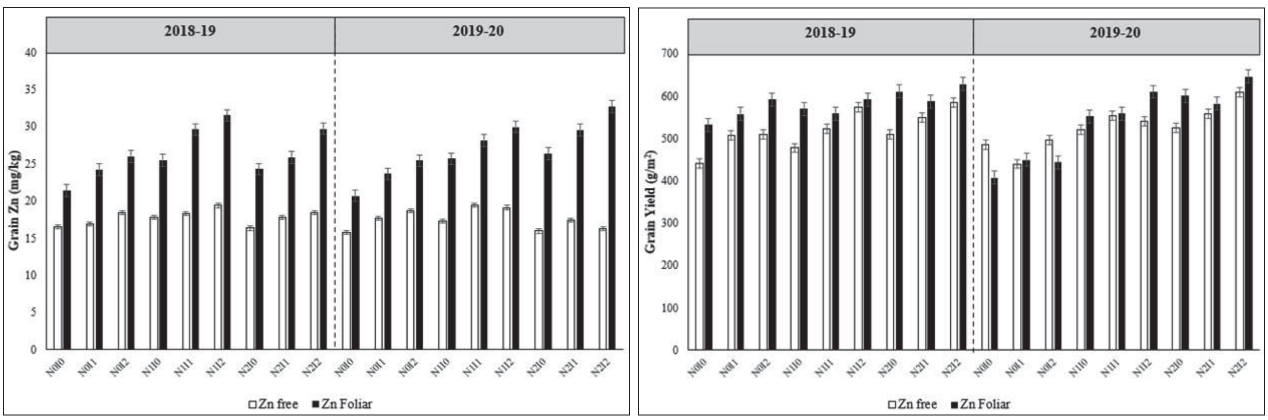

Figure 1. Grain yield and Zn concentration in bread wheat under different N and irrigation treatments during 2018-19 and 2019-20 cropping seasons

The Zn concentration in the wheat grain and shoots increased with application of Zn foliar in comparison with control treatment. Totally, the Zn concentration in grain was higher than the shoots (Figure 2). Meanwhile, grain and shoot Zn content showed no significant changes under different irrigation treatments (Figure 2).

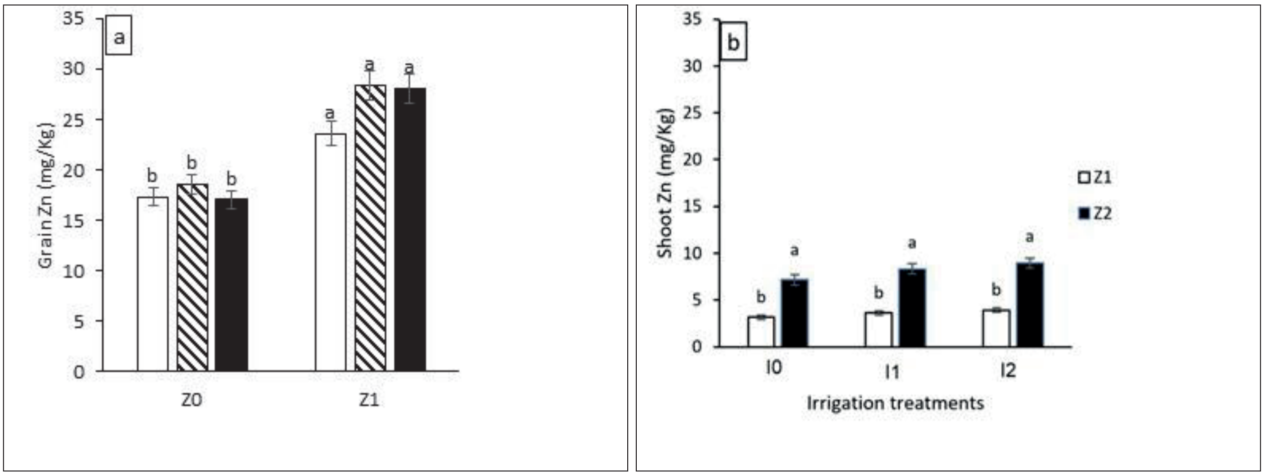

Figure 2. Zn application on Zn content in the bread wheat: a) Grain and b) Shoot

The interaction of N and Zn application on Zn content in the grain and shoots revealed the response of the bread wheat to the applied treatments (Figure 1). The grain Zn content under Zn application was higher than no application of Zn in all of the N treatments (Figure 1). In contrast, the shoot Zn concentration was significantly higher with application of normal N under Zn foliar application (Figure 1).

Pearson correlation coefficients of the survived traits revealed the relationship of the traits under the applied traits. Grain yield significantly correlated with HI, SFR, TKW, SNS, GZn and SZn. Furthermore, grain Zn content in addition to correlation with Y, was correlated with SZn, SFR and TKW traits (Table 5).

In general, biplot analysis clearly showed that grain yield and Zn content under Zn foliar application was higher that control (Zn-free) in the two cropping seasons. This difference was more obvious in grain Zn content that grain yield.

Table 4. Analysis of variance (Mean Squares) for the survived traits in bread wheat

SOV	df	PH	DM	SFR	TKW	SNS	ZNg	ZNls	Y	BY	HI
Y	1	1469.44**	4923.36**	49.69**	216.82**	2190.24**	0.36 ns	0.06 ns	11953.78 ns	266944.44**	0.044**
R(Y)	6	56.76	6.23	11.42	3.39	140.60	9.10	3.09	24368.46	171620.37	0.000
I	2	27.38*	10.69**	15.90*	9.42**	294.74**	149.00**	20.33**	30438.21**	116267.36*	0.000ns
Y*I	2	2.42 ns	3.76*	1.01 ns	0.27 ns	50.38 ns	1.76 ns	0.23 ns	872.09 ns	45850.69 ns	0.002ns
R*I	9	45.69	3.04	3.40	4.09	65.68	5.74	0.07	5328.88	33304.40	0.000
Z	1	2.25ns	1.36 ns	37.86**	33.93**	15.47 ns	2934.48**	756.34**	51000.69**	71111.11 ns	0.009*
Y*Z	1	0.03 ns	1.36 ns	17.85*	10.62**	1.07 ns	4.55 ns	0.10 ns	20784.03 ns	173611.11*	0.000ns
I*Z	2	0.02 ns	1.34 ns	1.53 ns	5.42*	26.03 ns	35.66**	3.51*	2095.67 ns	24079.86 ns	0.000ns
Y*I*Z	2	37.34*	0.05 ns	1.44 ns	0.82 ns	7.48 ns	0.96 ns	0.19 ns	3100.46 ns	64079.86 ns	0.001ns
R*Z(I)	9	13.76	1.40	7.13	1.06	58.29	3.97	2.18	7423.88	40543.98	0.001
N	2	212.71**	15.77**	58.36**	23.50 **	258.11**	115.08**	9.31**	110945.67**	901475.69**	0.001ns
Y*N	2	57.01**	9.67**	11.67*	2.41 ns	72.13 ns	7.18 ns	0.50 ns	23862.38*	272934.03**	0.001ns
I*N	4	3.09 ns	0.21 ns	0.70 ns	2.62 ns	18.46 ns	0.33 ns	1.41 ns	720.74 ns	14288.19 ns	0.000ns
Y*I*N	4	4.67 ns	0.19 ns	1.04 ns	1.74 ns	11.73 ns	0.60 ns	0.40 ns	1440.63 ns	2777.78 ns	0.000ns
Z*N	2	1.90 ns	0.05 ns	3.25 ns	0.17 ns	54.54 ns	75.37**	7.44**	4131.38 ns	40954.86 ns	0.000ns
Y*Z*N	2	3.55 ns	0.09 ns	4.47 ns	1.83 ns	3.75 ns	22.54**	0.88 ns	10011.09 ns	26788.19 ns	0.001ns
I*Z*N	4	7.79 ns	0.34 ns	1.25 ns	0.49 ns	3.89 ns	3.05 ns	0.91 ns	2053.92 ns	11892.36 ns	0.000ns
Y*I*Z*N	4	9.74 ns	0.21 ns	1.54 ns	0.31 ns	3.77 ns	2.10 ns	0.26 ns	2355.90 ns	7881.94 ns	0.001ns
Error	87	8.30	0.82	3.68	1.45	30.36	4.30	1.09	5966.468	31079.18	0.002
CV (%)		2.95	0.54	14.32	3.07	10.03	9.36	17.82	14.27	11.73	13.08

ns, * and **: non-significant, significant at 5 and 1 level of probability, respectively.

Table 5. Pearson correlation between the traits

Traits	PH	DM	SFR	TKW	SNS	GZn	SZn	Y
DM	0.853**							
SFR	0.006	-0.318						
TKW	-0.4*	-0.706**	0.703**					
SNS	-0.37*	-0.686**	0.621**	0.786**				
GZn	0.102	0.0332	0.522**	0.391*	0.137			
SZn	0.047	0.0143	0.42*	0.372*	0.0684	0.906**		
Y	0.25	-0.0691	0.961**	0.558**	0.504**	0.539**	0.42*	
HI	-0.59**	-0.799**	0.533**	0.723**	0.619**	0.362*	0.344*	0.347*

*, $p < 0.05$; **, $p < 0.01$.

Biofortification through agronomic approaches has been applied to enhance grain yield and nutrients content including Zn enhancement in bread wheat (Kutman et al., 2010; Ram et al., 2015; Cakmak and Kutman, 2018; Ning et al., 2019). Foliar Zn application has been used to in enriching the grain with Zn in bread wheat during grain-filling (Cakmak et al., 2010; Cakmak and Kutman, 2018). Foliar application of Zn combined with irrigation regimes and N application has been validated in Zn enrichment and bioavailability in grain (Ning et al., 2019).

Nitrogen causes cell division, enlargement, increases leaf area, durability, the number of tillers and its survival. Nitrogen is the only element whose deficiency greatly reduces wheat yield (Anagholi et al., 2006). In wheat, the amount of nitrogen in the grain is mainly affected by the supply of nitrogen after pollination and the amount of nitrogen that is stored before pollination and returns to the grain. After pollination, grain size and amount of nitrogen are significantly reduced due to nitrogen deficiency (Khanjani and Bahrani, 2017).

There is a significant correlation between usable water and nitrogen, because the increase in nitrogen leads to further development of wheat roots and therefore the usable water in the plant root area increases and as a result, moisture stress decreases. However, if there is not enough water, increasing the use of nitrogen fertilizer increases the moisture stress applied to the plant and reduces plant yield and water use efficiency (Ercoli et al., 2008).

Irrigation during the critical stages of wheat growth can increase wheat photosynthesis, dry matter production and remobilization in wheat and thus increase grain yield significantly higher than unirrigated wheat (Saeedipour and Moradi, 2011; Boughdiri et al., 2014; Mohamed et al., 2015).

In the present study, the effects of fertilizers and irrigation treatments on grain yield and Zn concentrations showed no noticeable differences in different years, suggesting a low influence of environmental factors such as rainfall and temperature on applied treatments (Wang et al., 2017).

Cakmak et al. (2010) reported that foliar application of Zn significantly increased the Zn content of the grain. This increase was greater than in the early stages of growth, especially when foliar spraying took place late in the growing season. Karim et al. (2012) claimed that foliar application had no significant effect on grain yield under drought stress conditions. However, under drought stress, it increased grain yield by 15% and also the grain Zn content significantly. Nasiri Majd et al. (2015) studied the effect of Zn foliar application on yield and yield components of wheat in irrigated wheat cultivars (Chamran, Falat and Star) in Ahvaz. They reported that foliar application of Zn by 0.5% per hectare increased grain yield compared to the control and also increased grain and Zn content.

Montoya et al. (2020) reported an increasing in grain Zn concentrations (by an average of 14%) using Zn-N co-fertilization in on a rainfed winter wheat under field experiment. They also showed the synergistic effect between Zn and N biofortification using a natural organic Zn complex, which in the present study we by using a foliar application. Similar result reported by Yaseen and Hussain (2021) when they applied Zn on two standard and Zn-biofortified wheat cultivars.

Regard to the increasing of micronutrient malnutrition issues especially Zn, increasing grain Zn concentration in main crops would be a promising approach to enrichment of food crops such as bread wheat. Therefore, the findings presented in this study may have important consequences for human nutrition.

CONCLUSIONS

The grain yield and quality in bread wheat were affected by different factors including Zn foliar application, N and complementary irrigation. The grain zinc content increased significantly by foliar application of zing during two cropping seasons. The grain yield increased by zinc, nitrogen and complementary irrigation application. The efficiency of the Zn foliar application was not generally affected by cropping season, which could be generalizable for future. Furthermore, the zinc content of grain and shoot was correlated with grain yield. In general, the results of this study showed that grain yield and quality in bread wheat could be increased by supplementary factors.

ACKNOWLEDGEMENTS

We would like to thank Mr. Aqil Khosravi, the esteemed director of Imam Khomeini Agricultural Conservatory in Aliabad Katoul, for his efforts in conducting this research.

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