YYÜ TAR BİL DERG
(YYU J AGR SCI)
2016, 26(4): 529-537

Geliş Tarihi (Received): 13.08.2016
Kabul Tarihi (Accepted): 16.11.2016

Araştırma Makalesi/*Research Article (Original Paper)*

Wheat Biofortification Through Zinc Foliar Application and Its Effects on Wheat Quantitative and Qualitative Yields under Zinc Deficient Stress

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Abstract: Wheat has a special role in providing the required energy and protein of people in developing countries like Iran. Wheat is confronted with nutritional limitations such as little zinc (Zn) and low biological value of protein, which results in hidden hunger and malnutrition. Hence, this research was conducted to study the effect of Zn foliar application on Kohdasht wheat variety performance yield and its quality in randomized complete block design (RCBD) with three replications. The treatments were non-use Zn sulfate (C; control and/or Zn deficient stress condition) and the use of Zn sulfate (spraying 0.5 g L^{-1} at stem elongation and grain filling stages (T1) and spraying 0.5 g L^{-1}at stem elongation and 2 g L^{-1} at grain filling stages (T2)). The result indicated that Zn foliar application increased grain Zn content and yield by improving grain number per spike. Moreover, in Zn treatments, amino acids contents and total Lysine and Threonine were increased that resulted in higher grain protein content in compared to Zn deficient stress condition. On the other hand, Zn application reduced phytic acid (PA) content and molar ration of PA to Zn. It can be concluded that Zn foliar application could yield alleviate malnutrition by increasing grain Zn content and protein, and by decreasing PA and molar ratio of PA to Zn under Zn deficient stress condition. Therefore, fertilizer strategy (e.g., agronomic biofortification) appears as short-term solution to alleviate malnutrition problem.

Keywords: Amino acids, Bread wheat, Phytic acid to Zn molar ratio, Protein, Zinc application

Yapraktan Çinko Uygulaması ile Buğdayda Biyo-zenginleştirme ve Bunun Çinko Eksikliği Stresindeki Buğday'ın Nicel ve Kalitatif Verimi Üzerine Etkileri

Özet: Buğday, İran gibi gelişmekte olan ülkelerdeki insanların enerji ve protein ihtiyacını karşılamada özel bir role sahiptir. Buğday, gizli açlık ve yetersiz beslenme ile sonuçlanan çinko (Zn) azlığı ve proteinin düşük biyolojik değeri gibi beslenme kısıtlamaları ile karşı karşıya bulunmaktadır. Bu nedenle, bu araştırma, yapraktan Zn uygulamasının Kohdasht buğday çeşidinin verim ve kalitesine etkisi incelemek amacıyla tesadüd blokları deneme deseninde üç tekerrürlü olarak yürütülmüştür. Uygulamalar, kontrol Zn (K; kontrol ve / veya Zn eksik stres durumu) ve Zn sülfatın kullanılması [kök uzama ve tahıl dolum aşamalarında 0.5 g/L püskürtme (Tl) ve kök uzama aşamasında 0.5 g / L ve tahıl dolum aşamasında 2 g / L püskürtme (T2)] şeklinde planlanmıştır. Sonuçlar, yapraktan Zn uygulanmasının, başak başına tahıl sayısını artırarak, tahıl Zn içeriğini ve verimini arttırdığını göstermiştir. Ek olarak, Zn uygulamalarında, amino asitler içeriği ve toplam Lizin ve Treonin miktarları, Zn eksik stres koşuluna kıyasla daha fazla bulunmuş ve bu da daha yükske tane protein içeriğine neden olmuştur. Öte yandan, Zn uygulaması, fitik asit (PA) içeriğini ve PA'nın Zn'ye molar oranını düşürmüştür. Yapraktan Zn uygulanmasının Zn eksikliğindeki stres koşulları altında Zn içeriği ve proteini artırarak ve PA'nın Zn'ye molar oranını azaltarak yetersiz beslenmeyi azaltabileceği sonucuna varılabilir. Bu nedenle, yetersiz beslenme sorununu hafifletmek için gübre stratejisi (örn. Agronomik biyo-zenginleştirme) kısa vadeli bir çözüm olarak karşımıza çıkmaktadır.

Anahtar kelimeler: Amino asitler, Çinko uygulaması, Ekmeklik buğday, Fitik asit Zn molar oranı, Protein

Introduction

Malnutrition arising from micronutrients deficiency such as Zn is considered as one of the present problems of humane societies especially in developing countries like Iran. Some of the side effects of Zn deficiency in humane societies are stunting in children, high susceptibility to infectious, impaired mental, increased morbidity and mortality (Cunningham-Rundles vd. 2005). It is estimated that one third of the World's population especially children are suffering from Zn deficiency (WHO 2002) and annually, nearly 450 thousand children below 5 years old die due to the effect of Zn deficiency (Bryce vd. 2005).

In Central and West Asia, wheat provides about 50 percent of people's daily energy requirements which reaches over 70 percent in rural areas (Cakmak vd. 2004). Regretfully, this plant naturally has little Zn content and it will be even less if planted in Zn poor soils (Cakmak vd. 2004). Moreover, there are antinutrient combinations in wheat such as PA that interferes with micronutrients absorption in human body. All of these factors along with low variety of diet result in low intake of daily requirements for Zn. Improvement of Zn content in wheat grain by bifortification is a proper approach for providing Zn requirements of body and reducing the malnutrition (Cakmak vd. 2010; Abdoli vd. 2014; Abdoli vd. 2016; Saha vd. 2017). Biofortification can be carried out by agronomic and genetic biofortification methods (Sadeghzadeh 2013). In agronomic method, the amount of plant's Zn requirement is provided Zn application such as soil or foliar application. Providing Zn requirements of plant improves metabolic processes like protein biosynthesis and carbohydrates (Bharti vd. 2013; Nawaz vd. 2015). Also, besides increasing the yield, it increases Zn concentration in grain as a result of which PA content is decreased (Khan vd. 2008; Singh and Singh-Shivay 2015). All these changes decrease in PA to Zn molar ratio in grain and raise the possibility of Zn absorption in body. For example, Ozturk vd. (2006) and Yang vd. (2011) stated that Zn spraying at grain filling stage had the most effect on increasing Zn content in grain and reducing PA to Zn molar ratio. Furthermore, due to the effect of Zn use in wheat and rice, increase of Zn content in grain, decrease of PA content, decrease of PA to Zn molar ratio and Zn bioavailability in body has been reported (Mabesa vd. 2013; Imran vd. 2015).

Considering the importance of wheat in providing people's energy and protein requirements (about 50 percent), it seems that quantitative and qualitative improvement of wheat protein is necessary to reduce the negative effects of malnutrition arising from low protein intake especially among low income and vulnerable segments of society. Nowadays, the role of Zn in protein biosynthesis has been proved (Passerini vd. 2007; Shu vd. 2008). In this regard, increase of free amino acids content in Zn deficiency and decrease of protein biosynthesis content has been reported by Cakmak vd. (1989). Increase of protein due to Zn application has been reported by Bybordi and Malakouti (2007) and Pourgholam vd. (2013). Zn acts as a cofactor in the structure of RNA polymerase. Furthermore, Zn is from the structural components of ribosome and in its absence, their function and protein biosynthesis is decreased.

Besides the quantitative yield of protein, its quality or its biological value is also important. Some of the factors influencing biological value of wheat protein are amino acids balance and essential amino acids content (Šramková vd. 2009). Wheat protein has limitation on the amount of Lysine and Threonine amino acids. In order to improve the quality of cereal grain protein, a lot of attempts have been made by reformers to improve the quality of wheat protein and increase Lysine and Threonine amino acids content such as discovering corn and barely with high Lysine and introducing them as a specific genotype. Unfortunately, along with these positive changes, negative characteristics happen like yield reduction and/or sensitivity to pests or disease. Zn foliar application was able to increase essential amino acids content in wheat protein. Moreover, Mishra and Abidi (2010) and Bharti vd. (2013) have reported an increase in Methionine content due to the effect of Zn application in agricultural plants.

The outbreak of malnutrition due to Zn and protein deficiency in developing countries like Iran and the role of wheat in people's nutrition in the country indicate the necessity of improving the quantitative and qualitative yield of wheat. On the other hand, Zn foliar application is a promising short-term approach to improve Zn concentrations in grains and can also help in alleviation of Zn deficiency related health problems in the developing world. Accordingly, the aim of the present study was to determine the effects of Zn foliar application on (i) grain yield and (ii) some of the parameters influencing the quality of wheat grain such as Zn concentration, phytic acid, protein content and amino acids and (iii) correlations of these components in bread wheat grain under zinc deficient stress condition.

Materials and Methods

A field experiment was carried out on cv. kohdasht of bread wheat (*Triticum aestivum* L.) in loamy soil in Agricultural Research Station, University of Maragheh, Iran (37º 22′ N latitude, 46º 16′ E longitude and altitudes of 1542 m) during cropping season year of 2013. Soil samples were taken at a depth of 0-30 cm in order to determine the physical and chemical properties of the soil (Table 1). The field was prepared by applying plow, disc, and leveling yields and then was divided into plots with $2 \times 1.4\ m^2$ size. Disinfected seeds were planted with a density of 500 seeds per square meter at a distance of 20 cm between rows on 11^{th} April 2013. Climate data of the study area during growing season for 2013 were presented in Table 2. The treatments include the use (C: control and/or Zn deficient stress condition (non-use zinc), T1: foliar Zn application (spraying) by dose 0.5 g/L at the stem elongation + 0.5 g/L at grain filling stages, and T2: foliar Zn application by dose 0.5 g/L at the stem elongation + 2 g/L at grain filling stages). The experiment was conducted in a randomized complete block design (RCBD) with three replications. Foliar Zn treatments (as $ZnSO_4.7H_2O$) were applied along with 0.01% (v/v) Tween as surfactant and nitrogen at the rate of 1% urea. In order to better absorption of zinc from the leaves, not only a few drops of Tween were added to solution as surfactant, also spraying yields was carried out at the end of the day using a backpack sprayer pump applying 100 mL of solution per square meter.

The wheat crop was harvested from a net plot of $0.6\ m^2$ after leaving a 0.2 m border on each side. At maturity, ten plants were randomly harvested from each plot to measure the number of grain per spike and thousand grain weight. Samples of grains were analyzed for the quality traits.

The Zn concentration of grain was measured by Liu vd. (2006) method. To this end, 0.5 g of very finely ground grain weighted and transferred to digestion tube and 5 mL of dense nitric acid was added into it and was kept for 1 h at 100°C. When samples got cooled, 2.5 mL dense perchloric acid was added and was kept for 3 hours at 200°C. Then, when samples got cooled, samples were filtered using filter paper and then its volume was brought to 25 mL with distilled water. Finally, its Zn concentration was measured using the atomic absorption spectrophotometer (Shimadzu AA-6300, Japan).

For phytate measurement, 60 mg finely-ground grain samples were extracted with 10 mL of 0.2 N HCl at room temperature for 2 h under continuous shaking. Phytate in the extract was determined by indirect method that uses absorption of pink color developed by un-reacted Fe (III) with 2,2′-bi-pyridine (Haug and Lantzsch 1983) at 519 nm with a Elisa (BioTek, Powre Wave XS2, USA). Molar concentration of phytate and Zn in grain was used to calculate phytate to Zn ratio (phytate: 660.04 g/mol; Zn: 65.4 g/mol).
Standard No. 994/12 AOAC method was applied to measure the amount of amino acids (Wendt-Thiex 2000). In this regard, 15 out of 20 amino acids which take part in the structure of grains proteins including 8 essential amino acids (Valine, Isoleucine, Lysine, Leucine, Threonine, Methionine, Phenylalanine and Histidine) and 7 non-essential amino acids (Alanine, Serine, Aspartic acid, Glutamate acid, Tyrosine, Arginine and Glycine) were determined using a calibrated amino acid analyzer (Model A200). It should be noted that the peaks of the mentioned amino acids were obtained by a single injection into the device. Since the determination of the essential amino acid tryptophan needs to inject another mobile phase which is costly, therefore the mentioned amino acid measurement is neglected due to the financial constraints.

Grain protein content was also measured using Kejeldahl-N (5.7×N) (Association of Official of Analytical Chemist 1975). All data were subjected to one-way analysis of variance (ANOVA) using SAS software (Version 8.0) and data comparisons were conducted using Duncan test at the 5% significance level.

Table 1. Physical and chemical characteristics of the soil at a depth of 0 to 30 cm

Soil texture	Clay	Silt	Sand	N	P	K	Zn	Cu	Fe	pH	EC*
	(%)			(%)	(mg/kg)						(ds/m)
Loam	17.2	35.2	47.6	0.1	8.43	102.2	0.41	0.5	2.39	6.81	0.49

*Electrical conductivity of soil saturation extract.

Table 2. Monthly climate parameters of the study area during growth season for 2013

Months	Temperature (˚C)			Precipitation (mm)	Relative Humidity (%)		
	Max	Min	Average		Max	Min	Average
April	18.9	6.1	12.5	21.8	70	26	48
May	21.4	8.8	15.1	20.8	71	28	50
June	29.0	14.9	21.9	1.8	54	18	36
July	19.8	33.6	26.7	0.0	45	17	31

Source: Meteorological Office, Maragheh, Iran.

Results and Discussion

Zn sulfate foliar application increased the amount of wheat grain yield compared to Zn deficient stress condition. So, due to the effect of Zn application by dose 0.5 g/L at stem elongation and grain filling stages (T1) and by dose 0.5 g/L at stem elongation and 2 g/L at grain filling stage (T2), grain yield increased from 118 g/m^2 in Zn deficient stress condition to 263 and 304 g/m^2, respectively (Table 3). Zn application considerably increased number of grain per spike and thousand kernel weight (Table 3). Regarding the role of Zn in various metabolic processes like protein biosynthesis and carbohydrates, foliar application led to formation of spikes with more numbers of spikelet per spike and floret per spikelet. By influencing the mentioned metabolic processes, Zn presence at spraying stages until maturity resulted in better meeting the need of spike and its components yield (Abdoli vd. 2014; Abdoli and Esfandiari 2014). Zn application in treatments T1 and T2 increased number of grain per spike by 26.2 and 38.4 percent, respectively, compared to Zn deficient stress condition. Increase of grain number per spike along with the increase of thousand kernel weights in treatment T2 are the reasons for increasing grain yield by 122.9 and 157.6 percent compared to Zn deficient stress condition, respectively (Table 3). Pointing to the role of Zn in reducing the problem of spike sterility, Bagci vd. (2007) stated that this element helps to increase grain number in wheat by removing limitations. Increase of grain yield due to the effect of Zn foliar spraying in wheat has also been reported by Khan vd. (2008), Abdoli vd. (2014), Singh and Singh-Shivay (2015) and Abdoli vd. (2016).

Zn application increased Zn content per wheat grain. Treatment T2 resulted in more Zn aggregation per wheat grain, and grain Zn content reached from 14.6 mg/kg dry matter in Zn deficient stress condition to 23.2 and 29.5 mg/kg dry matter in treatments T1 and T2, respectively. Due to the effect of treatments T1 and T2, Zn content was increased to 58.9 and 102.5 percent respectively compared to Zn deficient stress condition (Table 3). Zn application improved Zn absorption and accumulation of Zn in different plant parts (Cakmak vd. 1989; Saha vd. 2017). Increase of Zn content in aboveground organs and grain due to Zn application has been reported by Nan vd. (2002), Balali and Malakouti (2002) and Abdoli vd. (2014). Zn foliar application at grain filling stage had more effect on grain Zn concentration in compared to application at the stem elongation stage (Abdoli vd. 2014). Cakmak vd. (1989) stated that spraying at the last stages of wheat growth (such as milky and dough stages) have more effects on Zn content in grain and some parts of endosperm compared to soil application method. Also, Ozturk vd. (2006), Yang vd. (2011) and Li vd. (2014) demonstrated that the most Zn content in grain was gained when spraying Zn at the beginning of grain development (milky and dough stages).

Phytic acid content was decreased due to Zn application, and reached from 63.7 mg/g in Zn deficient stress condition to 40.7 and 22.2 mg/g in treatments T1 and T2, respectively (Table 3). Considering the negative effects of PA on absorption of micronutrients such as Zn in body and the specific role of wheat in people's nutrition, researchers try to decrease PA content in grain (Yang vd. 2011; Mabesa vd. 2013; Imran vd. 2015). Motesharezadeh and Savaghebi (2012) believed that increase of grain yield due to the effect of Zn application leads to attenuation of phosphate and PA content in grain and this is followed by a reduction in PA. Zn sulfate foliar application in wheat grains considerably decreased PA to Zn molar ratio compared to Zn deficient stress condition. Thus, through Zn foliar application, this ratio reached from 43.3 in Zn deficient stress condition to 21 and 7.4 in treatments T1 and T2 respectively. In other words, due to the effect of treatments T1 and T2, PA to Zn molar ratio in wheat grain was decreased by 55.6 and 84.4 percent respectively (Table 3). PA to Zn molar ratio is used for evaluating Zn bioavailability in body. Normally, in ratios higher than 15, Zn absorption considerably is decreased. Also, if affected by decrease of PA to Zn molar ratio, Zn bioavailability can't be less than 15. In this study, only in treatment T2, PA to Zn molar ratio was decreased to less than 15. Furthermore, reduction of this ratio was due to a simultaneous decrease in PA content and an increase in Zn in grain. In this regard, Bharti vd. (2013) stated that soil application along with Zn spraying led to increase of Zn content in grain (80 percent) and 23.2 percent decrease of PA content. Yang vd. (2011) mentioned that spraying at grain filling stage has the most effect on the increase of Zn content in grain and decrease of indicator of PA to Zn molar ratio. Other researchers emphasize that Zn application leads to its increase and accumulation in grain and aboveground organs (Nan vd. 2002; Balali and Malakouti 2002).

Table 3. The effect of Zn application on grain yield, number of grain per spike, thousands of grain weight, Zn concentration of grain, phytic acid content, phytate to Zn molar ratio and protein content of wheat.

Treatments	Grain yield (g/m^2)	Number of grain per spike	Thousands of grain weight (g)	Zn concentration of grain (mg/kg)	Phytic acid content (mg/g)	Phytate to Zn molar ratio	Protein content (%)
C†	118 ^{b}	37.0 ^{c}	26.2 ^{b}	14.6 ^{b}	63.7 ^{a}	47.3 ^{a}	14.9 ^{b}
T1	263 ^{a}	46.7 ^{b}	26.9 ^{b}	23.2 ^{ab}	40.7 ^{ab}	21.0 ^{b}	18.5 ^{a}
T2	304 ^{a}	51.2 ^{a}	30.9 ^{a}	29.5 ^{a}	22.2 ^{b}	7.4 ^{b}	17.2 ^{a}
F test	*	**	**	*	*	*	**
CV (%)	19.5	2.65	3.27	16.8	29.7	37.9	4.38

† C: control and/or Zn deficient stress condition (non-use zinc), T1: Zn application by dose 0.5 g/L at stem elongation + 0.5 g/L at grain filling stages, T2: Zn application by dose 0.5 g/L at stem elongation + 2 g/L at grain filling stages.
Means in the same column followed by the same letter are not significantly different at $P < 0.05$ according to Duncan's test. Ns, * and **: Non significant, significant at 5% and 1% levels of probability, respectively.

Due to the effect of Zn sulfate foliar application, grain protein content was increased. Total protein content was 14.9 percent in Zn deficient stress condition which reached 18.5 and 17.2 percent in treatments T1 and T2 (Table 3) due to the effect of Zn sulfate foliar application. Nowadays, the role of Zn in protein metabolism has been proved so that Zn deficiency in most of plants causes disorder in some metabolic processes like RNA metabolism and protein synthesis (Sharma vd. 1981; Kitagishi and Obata 1986; Cakmak vd. 1989). Also, Kitagishi vd. (1987) believe that Zn deficiency causes disorder in RNA synthesis and structure and ribosome in plants. On the other hand, an increase in the amount of RNA activity due to Zn deficiency has been reported (Sharma vd. 1981). Furthermore, aggregation of free amino acids and amides due to Zn deficiency compared to Zn deficient stress condition has been reported (Kitagishi and Obata 1986; Cakmak vd. 1989). In this regard, referring to aggregated amino acids in plant textures due to Zn deficiency, Cakmak vd. (1989) stated that free amino acids content is decreased in textures by adding this element (Zinc) to environment. In return, protein content of plant textures was increased through adding Zn to environment which is the indicator of Zn role in protein biosynthesis. Increase of grain protein content due to the effect of Zn application has been reported by some of the researchers like Bybordi and Malakouti (2007) and Pourgholam vd. (2013).

In this study, 15 amino acids changes including 7 non-essential amino acids (Alanine, Serine, Aspartic acid, Glutamic acid, Tyrosine, Arginine and Glycine) and 8 essential amino acids (Valine, Isoleucine, Lysine, Leucine, Threonine, Methionine, Phenylalanine and Histidine) in wheat grain were explored. Zn sulfate application caused an increase in the mentioned amino acids content (except Isoleucine in treatment T1) in wheat grain (Table 4). It should be noted that total essential amino acids in Zn deficient stress condition was 5888 mg/100 g of wheat grain which reached 6095 and 6299 mg/100 g due to the effect of treatments T1 and T2. In other words, treatments T1 and T2 increased the studied total amino acids by 3.51 and 6.98 percent respectively compared to Zn deficient stress condition (Table 4). Due to the effect of Zn sulfate foliar spraying, total Lysine and Threonine was increased in treatments T1 and T2 compared to Zn deficient stress condition. It should be mentioned that total of these two amino acids in Zn deficient stress condition was 1298 mg/100 g of wheat grain which reached 1371 and 1411 respectively due to the effect of treatments T1 and T2. In other words, treatments T1 and T2 increased total Lysine and Threonine by 5.62 and 8.71 percent compared to Zn deficient stress condition (Table 4).

Despite increasing grain protein content due to the effect of Zn foliar application in wheat grain, protein quality is also of specific importance (Šramková vd. 2009).

Table 4. The effect of Zn application on amino acids contents (mg/100 g) of grain of wheat.

Amino acid contents (mg/100 g)	Treatments			F test	CV (%)
	C†	T1	T2		
Non-essential amino acids					
Alanine	678 [c]	704 [b]	726 [a]	**	0.07
Serine	969 [c]	977 [b]	999 [a]	**	0.05
Aspartic acid	653 [c]	709 [a]	703 [b]	**	0.08
Glutamic acid	5399 [c]	5463 [b]	5612 [a]	**	0.04
Tyrosine	501 [c]	580 [b]	603 [a]	**	0.22
Arginine	364 [c]	368 [b]	436 [a]	**	0.20
Glycine	345 [c]	360 [b]	370 [a]	**	0.09
Essential amino acids					
Valine	620 [c]	637 [b]	648 [a]	**	0.06
Isoleucine	531 [b]	529 [c]	533 [a]	**	0.06
Lysine	404 [c]	468 [b]	486 [a]	**	0.23
Leucine	1461 [c]	1536 [b]	1641 [a]	**	0.11
Threonine	893 [c]	904 [b]	924 [a]	**	0.07
Methionine	852 [c]	878 [b]	900 [a]	**	0.06
Phenylalanine	741 [c]	743 [b]	772 [a]	**	0.07
Histidine	386 [c]	401 [a]	395 [b]	**	0.08
Lysine + Threonine	1298 [c]	1371 [b]	1411 [a]	**	0.10
Total Non-essential amino acids	8908 [c]	9161 [b]	9449 [a]	**	0.06
Total Essential amino acids	5888 [c]	6095 [b]	6299 [a]	**	0.07
Total amino acids	14796 [c]	15256 [b]	15748 [a]	**	0.06

† C: control and/or Zn deficient stress condition (non-use zinc), T1: Zn application by dose 0.5 g/L at stem elongation + 0.5 g/L at grain filling stages, T2: Zn application by dose 0.5 g/L at stem elongation + 2 g/L at grain filling stages.
Means in the same column followed by the same letter are not significantly different at $P < 0.05$ according to Duncan's test. Ns, * and **: Non significant, significant at 5% and 1% levels of probability, respectively.

Also, regarding the increase of grain protein content, the effect of Zn foliar application on amino acids combination in total protein and its nutritional value was studied. Nutritional value of protein depends on amino acids content and balance especially essential amino acids. In wheat protein, essential amino acids content of Lysine and Threonine is limited and this very factor results in reduction of its nutritional value. To improve protein quality of cereal grain, a lot of attempts were made by reformers in past so as to improve protein quality of wheat and to increase amino acids content of Lysine and Threonine such as discovering corn and barely with high Lysine and introducing them as specific genotype. Unfortunately, along with these positive changes, negative characteristic such as yield reduction and sensitivity to disease and pest appeared in them. Zn foliar application was able to increase essential amino acids content in wheat protein. In spite of existing limitation on Lysine and Threonine in wheat, after Leucine, Threonine content was the most in all of the studied treatments. Among the studied essential amino acids, Lysine and Histidine had the least content in each of the three components. But Lysine content compared to other amino acids was increased more due to Zn foliar application and its content got close to the content of other essential amino acids. This can help to improve biological value of protein through creating balance among essential amino acids (Table 4). Furthermore, the increase of total Lysine and Threonine along with essential amino acids is one of the positive effects of Zn foliar application (Table 4). It has been proved that Zn element is an important cofactor for RNA polymerase for facilitating polymerization of amino acid which is coded by mRNA. It should be noted that Mishra and Abidi (2010) and Bharti vd. (2013) has reported the increase of Methionine content due to the effect of Zn application in agricultural plants.

As shown in Figure 1, correlations between Zn concentration in grain and protein content (r = 0.46*), and total amino acids (r = 0.39*) were significant for the bread wheat with Zn application. By contrast, Zn concentration in grain and phytic acid content (r = -0.54**) and PA to Zn molar ratio (r = -0.79**) were negatively correlated. These results are in agreement with Yang vd. (2011), Esfandiari vd. (2016) and Saha vd. (2017) who found a significant negative correlation between grain Zn concentration and PA content.

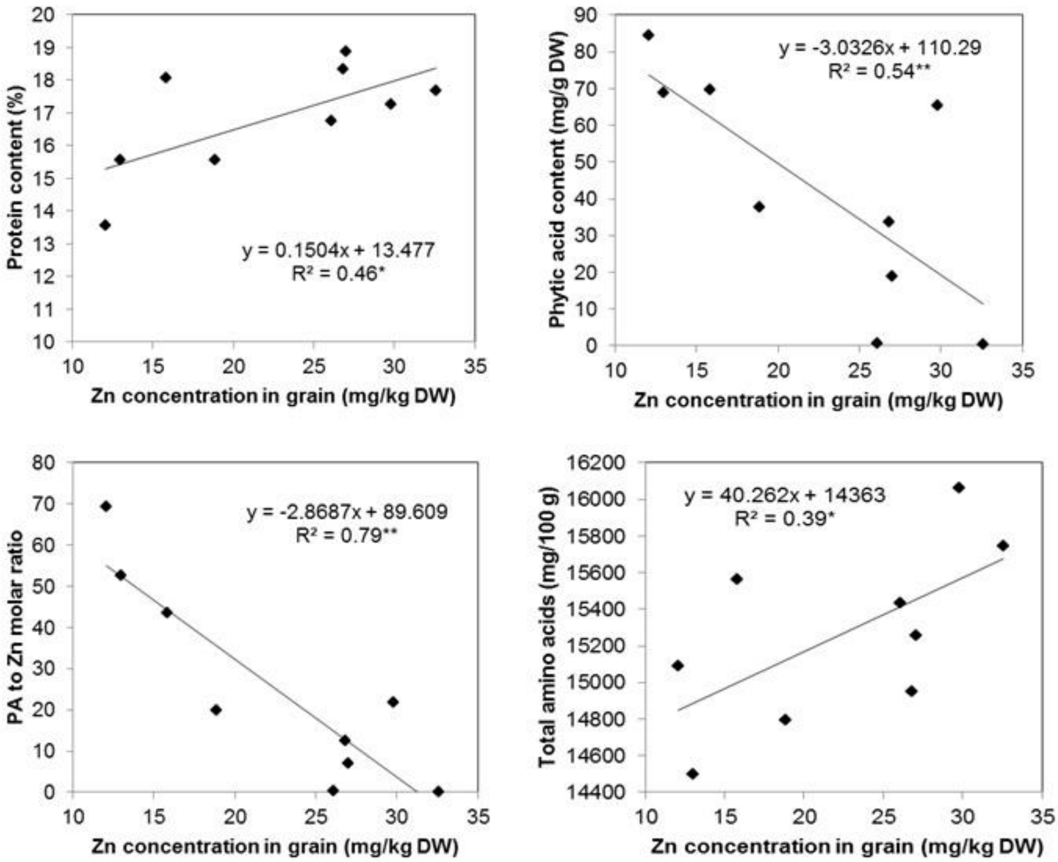

Figure 1. The relationship between Zn concentration in grain with protein content, phytic acid content, phytate to Zn molar ratio and total amino acids of bread wheat.

Conclusion

In conclusion, Zn sulfate foliar application along with increasing grain yield (quantitative aspect of production), improves the quality of produced crop through increasing grain Zn content, decreasing PA and molar ratio of PA to Zn. Of course, quantitative and qualitative aspects of wheat were improved compared to Zn deficient stress condition and treatment T1 by increasing content of Zn use in treatment T2. Zn application increased grain protein and amino acids content especially essential amino acids. Therefore, regarding the dominant agricultural condition in Iran and malnutrition, Zn foliar application is the suiTable approach for increasing yield, increasing grain Zn, decreasing PA and molar ratio of PA to Zn and improving biological value of wheat.

Acknowledgements

The work reported here is a part of the research project by title 'biofortification of wheat by zinc and iron'. The funding for this research was provided by Ministry of Science, Research and Technology (MSRT), Iran.

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