

Variation in Grain Zinc and Iron Concentration in Wheat Germplasm of Diverse Origin

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ABSTRACT

Micronutrient malnutrition, resulting from dietary deficiency of important minerals such as zinc (Zn) and iron (Fe), is a widespread foodrelated health problem. In a recent initiative of CGIAR's HarvestPlus challenge program is embarked upon to address this issue through the development of biofortified cultivars with elevated levels of these micronutrients in common wheat (*Triticum aestivum* L.). Genetic enhancement mainly depends on existence of genetic variability available in the gene pool. Hence, the magnitude of variability for grain Zn and Fe concentrations were studied in 600 wheat core-collection accessions of diverse origin. Grain Zn concentrations among the accessions ranged from 16.85 to 60.77 mg kg⁻¹ and Fe concentrations ranged from 26.26 to 68.78 mg kg⁻¹. The highest levels of Zn concentrations were observed in a Chinese spring bread wheat accessions "HONG DUAN MANG" and highest Fe concentration was observed in a accession originated from Spain 'ANDALUCIA 344'. Top ranking accessions with high Zn and Fe concentrations are being evaluated for multi-locational testing to study the expression of these micronutrients in target countries, also these accessions are being used as potential donor for further germplasm improvement at International Maize and Wheat Improvement center (CIMMYT), Mexico. There was a highly significant and positive correlation between Zn and Fe concentrations (r=0.81; P<0.01), indicating simultaneous improvement of both of the micronutrients would be effective and high Fe and Zn sources identified in this study provide a valuable genetic resource for breeding cultivars with high Zn and Fe concentrations.

Keywords: Bread wheat, durum wheat, core-collection, genetic variation, zinc and iron

INTRODUCTION

Micronutrient malnutrition arising from zinc (Zn) and iron (Fe) deficiency has emerged as a serious health problem worldwide, affecting more than two billion people worldwide (Welch and Graham 2004). The most prevalent deficiencies for these micronutrients occur particularly among pregnant women and children in resource poor families of developing countries (WHO 2002). Of these, Fe is of the most importance, with approximately three billion people suffering from Fe deficiency related anemia (United Nations System Standing Committee on Nutrition 2004; The World Bank 2007). The global average prevalence of Zn deficiency has been estimated to be 31%, with the most severe expressions in Africa and South Asia. Its deficiency is known to significantly increase the risk of many diseases viz., diarrhea, pneumonia, and malaria, therefore Zn deficiency has been linked to the morbidity and mortality of children.

A major factor causing Zn and Fe deficiency is their low bioavailability in diets based on cereals and legumes. Thus, it is important to improve the micronutrient quality of staple foods by increasing the levels of Zn and Fe concentrations. Bread wheat (Triticum aestivum L.) is a highly consumed cereal crop. On a worldwide scale, it contributes approximately 30% of the total cereal production, making it a major source of minerals for many people (McKevith 2004). Wheat provides 21% of the food calories and 20% of the protein to more than 4.5 billion people in 94 developing countries (Braun et al. 2010) Wheat is also one of the most important food crops in South Asia, as this region is being targeted for initial release and commercialization of biofortified wheat, especially in the area around Indo-Gangetic plains, a region with high population densities and high micronutrient malnutrition. So it is imperative to develop wheat cultivars with improved Zn and Fe concentrations to alleviate malnutrition.

Sustainable solutions to malnutrition will only be found by closely linking agriculture to nutrition and health and by formulating agriculture, nutrition and health policies to reflect this need (Graham et al. 2007; The World Bank 2007). Genetic enhancement of crop cultivars with elevated levels of these micronutrients would be cost effective sustainable way of solving global micronutrient malnutrition problem (White and Broadley 2009). Consequently, the Consultative Group on International Agricultural Research (CGIAR) launched the HarvestPlus initiative (http://www. harvestplus.org), a program focusing on breeding food crops with high micronutrient contents. Under this initiative, CIMMYT (Centro Internacional de Mejoramiento de Maiz y Trigo, http://www.cimmyt.org) and partner institutions, are attempting to develop high yielding, disease resistant wheat germplasm with enhanced levels of Zn and Fe (Velu et al. 2010). Success in crop improvement through breeding depends on the existence of genetic variation for the target traits in the available gene pool. Hence, a representative sample of 600 gene bank wheat accessions were evaluated for grain mineral elements that could be explored in future wheat breeding programs. The main objective of the present study was to investigate the variation for micronutrient concentration in wheat grain among 600 wheat accessions of diverse origin.

MATERIALS AND METHODS

A set of 600 bread and durum wheat accessions originated from more than 25 countries representing major wheat growing regions of the world were drawn from the genetic resources collection in the gene bank of CIMMYT-Mexico. These accessions were evaluated in a randomized complete block design (RCBD) with two

Table 1 Core-collection accession with high	Zn and Fe concentrations ((>mean + 2 SD), Co	d. Obregon, Mexico.
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Accession	Origin	Zn concentration	Zn content	Fe concentration	Fe content
		(mg kg ⁻¹)	(µg grain ⁻¹)	(mg kg ⁻¹)	(µg grain⁻¹)
HONG DUAN MANG ⁽¹⁾	China	60.77	1.50	50.95	1.27
AKSEREZ ⁽³⁾	Turkey	60.58	1.57	61.63	1.60
ANDALUCIA 344 ⁽³⁾	Spain	59.69	1.51	68.78	1.73
GRAY JD1024 ⁽³⁾	Afghanistan	54.18	1.68	58.53	1.83
PI316094 ⁽³⁾	USA	53.62	2.01	56.82	2.13
W-68 ⁽¹⁾	Pakistan	53.27	1.13	47.38	1.00
W71 ⁽³⁾	Pakistan	52.30	1.08	52.64	1.08
HONG HUA MAI ⁽¹⁾	China	51.50	1.56	44.05	1.33
PI264736 ⁽³⁾	USA	50.95	1.80	56.62	1.99
SARI BASAK ⁽¹⁾	_	49.95	1.36	46.39	1.27
GRAY JD1004 ⁽³⁾	Afghanistan	49.59	1.51	57.06	1.74
NOVOMICHURINKA ⁽¹⁾	-	49.53	1.39	49.79	1.41
AKRUSAM ⁽³⁾	Turkey	49.27	1.22	55.44	1.37
CI7648 ⁽³⁾	Russia	49.16	1.47	52.70	1.58
ARRANCADA 13720 ⁽¹⁾	Spain	49.11	1.08	44.39	0.98
W-77 ⁽¹⁾	Pakistan	49.03	1.09	46.66	1.04
W-61 ⁽¹⁾	Pakistan	48.64	1.00	50.87	1.05
DURAZIO RIJO ⁽¹⁾	Portugal	48.42	1.26	49.70	1.32
PI316089 ⁽³⁾	USA	48.39	1.48	56.02	1.72
PI324072 ⁽¹⁾	USA	46.93	1.60	48.77	1.66
W-76 ⁽¹⁾	Pakistan	46.83	1.17	49.80	1.24
MISTO 792 ⁽¹⁾	Ethiopia	46.59	1.26	48.27	1.31
OUED ZENATI 368 ⁽³⁾	-	46.07	1.67	54.10	1.97
W-69 ⁽¹⁾	Pakistan	45.58	1.06	50.40	1.18
W-36 ⁽³⁾	Pakistan	44.72	1.35	54.53	1.65
CI3735 ⁽¹⁾	Russia	44.64	1.59	45.60	1.65
GRIFONE 235 ⁽³⁾	-	44.57	1.07	58.23	1.40
RED SCHLANSTETTER RIMPAU ⁽²⁾	Germany	41.85	1.10	52.73	1.39
YEMEN-DW ⁽²⁾	Yemen	39.40	0.91	53.99	1.21
W-82 ⁽²⁾	Pakistan	39.13	0.98	54.29	1.37
ADUR ⁽²⁾	Austria	39.10	1.23	51.91	1.63
NS51.43 ⁽²⁾	_	33.53	1.01	54.86	1.66
PI352426 ⁽²⁾	USA	30.60	1.07	65.80	2.38
Mean		30.41	0.97	39.65	1.27
Minimum		16.85	0.49	26.26	0.63
Maximum		60.77	2.06	68.78	2.38
SD±		6.92	0.24	6.09	0.26

1, 2 & 3 are accessions with >mean + 2 SD for Zn concentration, Fe concentration and common for both Zn and Fe concentrations, respectively.

replications at CIMMYT- Cd Obregon, Mexico. Each accession was grown in 2 rows of 2 m length at 0.75 cm spacing between the rows at Cd Obregon research station in Mexico. The experiment received 120 kg ha⁻¹ N (50% basal and 25% top dressed twice at third and 7 weeks after sowing), 60 kg ha⁻¹ P₂O₅ and 40 kg ha⁻¹ K₂O. Plants were harvested at physiological maturity, threshed in a non-mineral contaminating threshing machine and the grains cleaned of any glumes and inert matters. Grain samples were analyzed for mineral concentration at Waite Analytical Services, University of Adelaide, Australia, based on the nitric/perchloric acid digestion method using an inductively coupled plasma optical emission spectrometer (ICP-OES) (Zarcinas *et al.* 1987).

RESULTS AND DISCUSSION

The concentrations of 15 minerals in wheat grain were determined. For brevity, only the data of two trace elements (Zn and Fe) that are of particular importance to human nutrition are presented in this paper. Among the 600 accessions, grain Zn varied by 3.6-fold, ranged from 16.85 to 60.77 mg kg⁻¹ with the mean of 30.41 mg kg⁻¹ and, grain Fe concentrations varied by 2.6-fold, ranging from 26.26 to 68.78 mg kg⁻¹ with the mean of 39.65 mg kg⁻¹ (**Table 1; Fig. 1**). Analysis of variance showed highly significant differences existed among entries for Zn concentration and contents (Table **2**). Nine accessions $(50.95-60.77 \text{ mg kg}^{-1})$ had > 50 mg/kgZn. The highest Zn concentrations were observed in a Chinese accessions 'HONG DUAN MANG' followed by a Turkish accession 'AKSEREZ', and these accessions are being used as potential donor for germplasm improvement at CIMMYT. Top ranking accessions with high Zn and Fe concentrations are being evaluated in replicated trials in



Fig. 1 Frequency distribution of grain Zn and Fe concentrations in core-collection accessions.

Mexico, also in target countries to study their Genotype \times Environment interaction for expression of these micronutrients.

A large and highly significant difference among the accessions was also found for Fe concentration and content. Thirty-five accessions ($50.12-68.78 \text{ mg kg}^{-1}$) had grain Fe concentrations >50 mg kg⁻¹, 'ANDALUCIA 344' accession originated from Spain had highest grain Fe, followed by a accession from USA 'PI352426'' and then, Turkish accession

 Table 2 Analysis of variance for grain Zn and Fe concentrations and contents.

Source of	df	Mean squares				
variation		Fe		Zn		
		Concentration (mg kg ⁻¹)	Content (µg grain ⁻¹)	Concentration (mg kg ⁻¹)	Content (µg grain ⁻¹)	
Genotype	599	74.08**	0.13**	95.69**	0.11**	
Block	1	137.57	0.06	6186.43	4.10	
Error	599	11.68	0.02	20.98	0.02	

*, ** Significant at 5% and 1% probability levels, respectively.

sion 'AKSEREZ' (second ranked for Zn concentrations). This was expected considering a highly significant and positive correlation between Fe and Zn concentration (r=0.81; P<0.01) (**Fig. 2**). A similar relationship between Fe and Zn has been reported in maize (Maziya-Dixon *et al.* 2000), rice (Zhang *et al.* 2004) and wheat (Ortiz-Monasterio *et al.* 2007; Velu *et al.* 2010), indicating that selection for either Fe or Zn will lead to correlated selection response for the other.

In total, 89 accessions showed grain Fe concentrations above mean + 1 SD for Fe and 19 accessions had above mean + 2 SD. For Zn, 83 accessions showed above mean + 1 SD and 27 accessions had above mean + 2 SD. Of these, there are 13 accessions are common for both Zn and Fe concentrations with mean + 2 SD, indicating close relationship between the Zn and Fe concentrations. Table 1 provides total amount of Zn and Fe per seed (content). Large variation was observed for Zn $(0.49-2.06 \ \mu g \ grain^{-1})$ and Fe content (0.63-2.38 µg grain⁻¹) indicates large differences for seed weights among accessions. For example, the accession 'PI 352426' with a second highest concentration of Fe $(>65.80 \text{ mg kg}^{-1})$ also had high seed weight (or seed size) and had, consequently the highest total amount of Fe (2.38 μ g grain⁻¹). A similar trend was observed for Zn. There was a highly significant correlation between the concentration and content of Zn (r=0.65; P<0.01) and Fe (r=0.45; P<0.01) indicating accessions with high Zn and Fe concentrations also had greater grain weight, suggesting that higher grain Zn and Fe concentrations are not necessarily related to small grain size or weight.

Table 2 shows the Pearson correlations between Zn and Fe concentrations and content. A strong positive correlation was found between Zn and Fe concentrations (r=0.81; P < 0.01) and contents (r=0.85; P < 0.01) indicating simultaneous improvement is possible as these minerals accumulation controlled by common genetic factors. A very strong correlation between grain Fe and Zn concentrations with grain protein was also shown previously by Ortiz-Monasterio et al. (2007) indicating grain protein may be sink for Zn and Fe. Recent studies have shown that the Gpc-B1 (Grain protein content-B1) locus from wild emmer wheat affects both grain protein content and the concentrations of these minerals in grain (Distelfeld et al. 2007). This locus encodes an NAC transpiration factor (NAM-B1) that accelerates senescence and increases remobilization of nutrients (Fe, Zn, N) from leaves to developing grains (Uauy et al. 2006). This positive correlation between Zn and Fe concentrations and, with protein content would be useful information to develop wheat cultivars with high protein with highest Zn and Fe concentrations.

Current research results showed the existence of large variability for Zn and Fe concentrations among a diverse range of core-collection accessions. Top ranking accessions with high Zn and Fe concentrations would serve as a potential donor for further germplasm improvement with elevated micronutrients. A high positive correlation between Zn and Fe indicates simultaneous selection would be highly effective. Genetic enhancement of grain Zn and Fe concentrations in wheat is being integrated with genetic improvement of grain yield, disease resistance and other agronomic traits at CIMMYT (Velu *et al.* 2010). In the pursuit of this objec-



Fig. 2 Relationship between grain Zn and Fe concentrations in corecollection accessions.

tive, studies are underway to examine the association of grain Zn and Fe with grain yield and other agronomic traits.

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