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Variations in grain mineral concentrations of Turkish wheat landraces germplasm

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RESEARCH ARTICLE

Abstract

The study aimed to examine and interpret variability of some mineral contents in wheat genotypes throughout growing seasons. A collection of 86 landraces originated from different provinces in Turkey and 14 registered cultivars were studied during two successive years for variability of some mineral contents (Fe, Zn, B, K, Mn, Mo, Cu, Mg and Ca) in grain. Both correlation coefficients and genotype trait (GT)-biplot analysis were used to examine and interpret variability of mineral contents in wheat genotypes throughout growing seasons. According to the correlation analysis, there were positive relationships among Fe and Zn, Mn, Mg and Cu contents. Furthermore, the relationship between the concentrations of the elements indicated that the correlation coefficients were consistent with the results of the GT-biplot analyses. The highest variations were obtained from Fe and Zn concentrations along with the longest vectors from GT-biplot origin. Fe, Zn and Mn contents were the ideal traits, according to mineral contents scaling GT-biplot. These traits were recorded as the best representative of the overall mineral contents along the most powerful to discriminate genotypes. Pure line 22 was superior with respect to Fe, Zn and B composition, while pure line 47 was superior regarding Ca contents. Comparison between pure lines and modern cultivars led to the conclusion that Fe, Zn and Mn content of many pure lines were usually higher than those of modern cultivars. Moreover, mean grain concentrations of Fe, Zn and Mn in pure lines from landraces were significantly higher than all cultivars, 9.25, 14.82 and 6.75%, respectively. Therefore, some pure lines could be recommended to use as genetic material to enhance the genetic basis of bread wheat breeding programmes all over the world.

Keywords: Turkey, wheat landraces, mineral contents, biplot, correlations, breeding

1. Introduction

Wheat cultivation performed essential part and led enormous changes in the lives of people for centuries in Turkey and neighbouring countries (Köksel and Cetiner, 2015). Recent archaeological excavations in Gobekli Tepe of Sanliurfa Province have a potential to shed light on the periods prior to the known date of agriculture, especially on the domestication of wheat (Bird, 1999). Different bread wheat landraces were used in Turkey for a long time in last decades. The diversity of Turkish wheat landraces has received great attention since the beginning of the 20th century (Karagoz and Zencirci, 2005). Exploration and collection missions were mounted and the collected germplasm was evaluated in different countries (Gokgol, 1939).

Cereal crops – such as wheat – are still an important source of minerals and other nutrients for humans all over the world. Mineral deficiencies with, such as, Fe, I and Zn are mostly caused due to inadequate levels in peoples' diet (Welch and Graham, 1999). Fe deficiency ranks among the most widespread nutrient deficiencies, estimated to be suffered over two billion people worldwide (Stoltzfus and Dreyfuss, 1998). Zn is an essential trace mineral influencing gene expression as well as cell development and replication (Hambridge, 2000). Henderson *et al.* (2003) concluded that cereals and cereal products provide 44% of the daily intake of Fe (15% from bread), 27% of Mg (13% from bread), 25% of Zn (11% from bread) and 31% of Cu (14% from bread).

The breeding of semi-dwarf, high-yielding crop cultivars called 'green revolution' instead of increasing applications of fertilisers and other agrochemicals which had increased the grain yield notably since the mid-1960s. This has undoubtedly contributed to alleviating global food shortages and famine that would have otherwise occurred at a much larger scale (Fan *et al.*, 2008). Unfortunately, plant breeding has been historically oriented toward higher agronomic yield rather than the nutritional concentration (Morris and Sands, 2006; Welch and Graham, 1999).

The solutions to micronutrient malnutrition may include supplementation, and diversification of diet, as well as bio fortification of crops either by agronomic or genetic methods (e.g. plant breeding); the latter is considered to be the most effective for resource-poor populations in the developing countries (Welch and Graham, 1999). In the light of recent studies, sufficient genetic variation in germplasm mineral concentrations of major crops belongs to both landraces and their wild relatives. The genetic variation can easily be explored in breeding strategies to combine high nutrient density accompanying high-yielding traits (Cakmak *et al.*, 2000; Graham *et al.*, 1999; Monasterio *et al.*, 2007).

Wheat is an important source of minerals and especially providing Fe, Zn, Cu and Mg to the diet of Turkish people. There is an urgent need for development of wheat varieties with improved protein, Fe and Zn content in Turkey (Köksel and Cetiner, 2015).

In the present study, pure lines selected from different Turkish bread wheat landraces as well as registered cultivars were used. The main objectives were: (1) to determine the amount of genetic variation considering numerous mineral contents (Fe, Zn, B, K, Mn, Cu, Mg and Ca) over two growing seasons; and (2) to compare landraces (86 pure lines) and cultivars (14 bread wheat) based on their mineral contents via genotype trait (GT)-biplot techniques.

2. Material and methods

Field experiment

In total 100 bread wheat genotypes (86 Turkish landraces pure lines and 14 registered bread wheat cultivars) were used as the experimental plant material. The pure lines were selected from bread wheat landraces by pure line selection method during 2002-2005 growing seasons at the Central Anatolian Region of Turkey (Akcura, 2006). Other experiment materials were 14 registered cultivars (Bagci-2002, Bayraktar-2000, Bezostaja-1, Dagdas-94, Demir-2000, Gerek-79, Gun-91, Karahan-99, Kenanbey, Konya-2002, Seval, Tekirdag, Tosunbey and Zencirci-2000) which are most commonly grown in Turkey. The field experiments were carried out under rain-fed conditions at Canakkale Onsekiz Mart University Dardanos Field Experiment Area, Canakkale, Turkey, in 2012 and 2013 growing seasons.

The experimental area was fallow before each growing season. Before sowing, randomised soil samples (0-30 cm depth) were collected from the field; soil texture was loam. Soil pH recorded 7.9, measured in saturated soil. Organic matter was 1 g/kg of soil, free lime (calcium carbonate; $CaCO_3$) was 43 g/kg of soil. Plant-available K and P in the soil were 2.4 kg/da, 41.30 kg/da, respectively. Plant-available Fe, Zn, Mn and Cu in the soil were 3.2, 4.8, 2.36 and 1.00 mg/kg, respectively. The plant materials (100 genotypes) were sown in 4 rows of 2 m long incomplete block design with two replications. Sowing was done on first week of October in both growing seasons. Weeds were controlled manually. Fertilisation was 27 kg/ha N and 69 kg/ha P_2O_5 at sowing, 43 kg/ha N was applied at the end of tillering stage at both growing seasons. Experimental plots were harvested at similar dates between June 16 and June 28 in both years.

The total mean rainfall during the 2012 and 2013 growing seasons in Canakkale (latitude: 40°7'N; longitude: 26°23'E; altitude: 6 m above sea level) was 505 and 688 mm, respectively. The long-term rainfall (means of 52 growing seasons) for Canakkale was 584 mm. Grain samples were dried and cleaned before measuring mineral concentrations. All analyses were performed on the complete set (86 pure lines, 14 cultivars with two replicates) of samples in both seasons.

Measurement of mineral concentration of bread wheat grain

Samples of threshed grain and straw were dried at 70 °C for 48 h in an air-forced oven, for the of mineral concentration analyses. Dried samples were ground with a mill (Arcelik, Istanbul, Turkey). Later, about 0.3 g ground samples were digested in mixture 4:1 (HNO_3:$HClO_4$) in a closed microwave system (Jones and Case, 1990). Concentrations of Zn, Fe, Mn, B, Cu, Mg, K, Mo, and Ca were read by atomic absorption spectrophotometer (GBC Scientific Equipment Ltd., Dandenong, VIC, Australia) according to Isaac and Kerber (1971). Measurements of mineral concentrations were compared using the certified values of the related minerals in the reference grain samples (BCR-189 wheat whole meal flour) for each set of measurements.

Data analysis

Variance analyses were run on data obtained from 86 pure lines and 14 standard cultivars. In pooled analysis experiments, years were random, while genotypes were fixed. A linear correlation analyses was applied pairwise to all the parameters studied across the growing seasons. Analysis of variance and linear correlations were performed using SAS (SAS Institute, Cary, NC, USA).

The GT-biplot was constructed by plotting the first two principal components (PC1 and PC2) derived from subjecting the genotype-mineral content matrix to singular value decomposition (Yan and Kang, 2003) of the trait-centred and standardised data. This methodology uses a biplot to show the factors (G and GT) that are important in genotype evaluation and that are also the sources of variation in multiple trait data (Yan and Kang, 2003). In the present study, genotype-focused scaling was used in visualising for genotypic comparison, with mineral content-focused scaling for mineral content comparison. Furthermore, the symmetric scaling was preferred in visualising the 'which-won-what' pattern of the multiple traits data. The tester vectors that originated from biplot origin and reach markers of the mineral contents were used to visualise among mineral contents (Yan and Kang, 2003). GT-biplot analyses were done using GenStat software (VSN International, Hemel Hempstead, UK).

3. Results and discussion

Variation of mineral concentration

Descriptive information on seed mineral concentrations of bread wheat landraces and cultivars are given in Supplementary Table S1. For landraces, Fe and Zn concentrations were of the same stature (35.53-53.08 and 22.66-38.57 mg/kg, respectively). Among landraces, L22, L81 and L80 had the highest Fe concentration (53.08, 52.26 and 52.04 mg/kg, respectively) as well as Zn concentration (38.57, 37.43 and 37.10 mg/kg, respectively). For cultivars, the range of Fe and Zn contents were between 35.81 (Karahan-99) and 42.69 mg/kg (Tekirdag), and 23.97 (Bayraktar-2000, Gun-91, Kenanbey and Seval) and 31.27 mg/kg (Bezostaja-1), respectively. Similarly, Mn concentrations of both genotype groups showed the same differences. Mn concentrations of landraces ranged between 30.92 (L29) and 48.58 mg/kg (L69). Landraces L69 (48.58 mg/kg), L18 (48.43 mg/kg), L63 (47.94 mg/kg), L77 (47.94 mg/kg) and L48 (47.94 mg/kg) contained the highest Mn concentrations. Mn concentrations of cultivars were found between 31.84 (Seval) and 33.80 mg/kg (Konya-2002) with a mean value of 32.97 mg/kg (Supplementary Table S1).

B, Cu, and Mo contents of landraces were between 8.63 (L39) and 15.77 mg/kg (L23), 4.12 (L65) and 6.69 mg/kg (L83), 0.854 (L34 and L78) and 1.782 mg/kg (L3), respectively. B, Cu, and Mo contents of cultivars were ranged between 7.73 (Bezostaja-1) and 13.12 mg/kg (Gerek-79), 4.34 (Bagci-2002) and 6.56 mg/kg (Kenanbey), 0.873 (Seval) and 1.823 mg/kg (Demir-2000), respectively. Thus, the B, Cu, and Mo content for both landraces and cultivars showed similar values (Supplementary Table S1).

Furthermore, the content of some macro elements (K, Mg and Ca) were determined (Supplementary Table S1). Among landraces K, Mg and Ca concentrations ranged between 2.25 (L77) and 5.41 g/kg (L57), 1.02 (L38) and 1.69 g/kg (L22), 0.34 (L3 and L66) and 0.55 g/kg (L47), respectively. For cultivars, the highest K, Mg and Ca concentrations were determined from Gun-91 (5.05 g/kg), Gerek-79 (1.60 g/kg) and Gerek-79 (0.52 g/kg), respectively. The macro element concentrations of landraces were found similar to those of the cultivars (Supplementary Table S1).

The results showed that there were highly significant variations among landraces for some mineral concentrations. Especially a wide variation in grain contents of Fe, Zn and Mn in landraces was found (Supplementary Table S1). The sufficient genetic variability in Turkish wheat landraces can be used to develop wheat cultivars with increased Fe and Zn concentrations in grain. Most landraces had higher Fe, Zn and Mn contents than the registered cultivars. Similarly, Garvin *et al.* (2006) showed that grain Zn and Fe concentrations decreased significantly with the date of cultivar release in a set of 14 USA wheat cultivars from production eras spanning more than a century. In addition, the green revolution has unintentionally contributed to decreased mineral density in wheat grain (Fan *et al.*, 2008; Zhao *et al.*, 2009). No significant differences in B, K, Cu, Mg, Ca and Mo contents were found between landraces and cultivars in our study.

Overall mean of Fe, Zn and Mn concentrations in our results were similar to previous studies on bread wheat genotypes (Harmankaya *et al.*, 2012; Hussain *et al.*, 2011; Murphy *et al.*, 2008; Zhao *et al.*, 2009).

Relationship among mineral concentration

Although the genotype genotype environment (GGE)-biplot methodology was originally proposed for analysing multi-environmental trials data for a given trait, it is equally applicable to all types of two-way data that assume an entry-by-tester structure, such as a genotype-by-trait two-way dataset (Yan and Kang, 2003). Further information about the discriminating power of mineral contents, together with a representation of their mutual relationships, can be obtained by the mineral content-vector view of the GT-biplot. The mineral content-vectors are the lines that originate from the biplot origin and reach markers of the traits (Figure 1). In this case, a long mineral content-vector reflects a high capacity to discriminate the genotypes. Mg, Zn and Fe concentrations were the most discriminating with the longest vectors from the origin. Cu, Mn, Mo, Ca and K concentrations were moderately discriminating while B was least discriminating with the smallest vector (Figure 1).

The correlation coefficients between 9 test mineral contents are given in Table 1. Among the mineral contents, Fe content was positively correlated with Zn and Mn content. The association of Zn with Mn was positive and significant. Mn was positively correlated with Ca, Mg content positively correlated with Cu content. Fe content

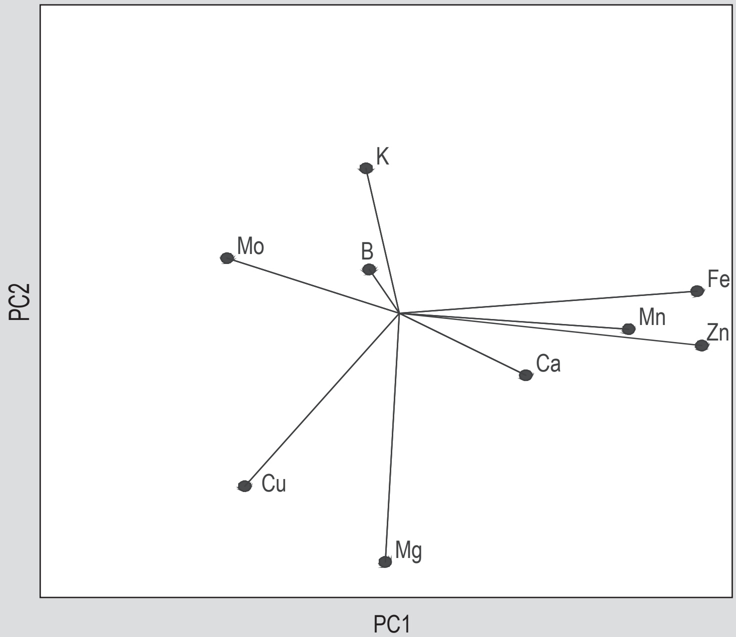

Figure 1. Genotype trait-biplot based on mineral content-focused scaling for mineral contents.

was negatively correlated with Cu and Mo, Zn content negatively correlated with Mo content (Table 1).

The vector view of the GT-biplot (Figure 1) provides a succinct summary of the interrelationships among the mineral contents. Since the cosine of the angle between the vectors of any two traits approximates the correlation coefficient between them, this view of the biplot is best for visualising the interrelationship among traits (Akcura, 2011; Yan and Kang, 2003).

GT-biplot, which was based on mineral content-focused scaling, was portrayed to estimate the pattern of mineral contents (Figure 1). Considering the angles between mineral vectors, Fe, Zn and Mn concentrations were positive significantly correlated (Figure 1). In addition, between Zn and Mn was a significant and positive relationship. All three traits showed a weak but positive correlation with Ca concentration. Similarly, Mg and Cu were significant positively associated with each other. Among traits, considering the angles between vectors, Fe and Cu concentrations were significant negatively correlated. Other associations between traits were not significantly correlated. It is remarkable that the nine vector lines in Figure 1 approximate the whole correlation matrix well (Table 1).

The polygon view of a GT-biplot explicitly displays the which-won-what pattern, and hence is a succinct summary of the genotype trait interaction pattern of a multiple traits data set (Figure 2). The polygon is formed by connecting

Table 1. Correlation coefficients between grain mineral contents in wheat across growing seasons.[1]

	Fe	Zn	Mn	B	Cu	Mo	K	Mg	Ca
Fe	1	0.69**	0.27**	-0.01	-0.33**	-0.27**	-0.06	-0.04	0.18
Zn	0.69**	1	0.52**	-0.06	-0.15	-0.21*	-0.03	0.06	0.11
Mn	0.27**	0.52**	1	-0.03	-0.08	-0.16	-0.02	-0.07	0.19*
B	-0.01	-0.06	-0.03	1	0.07	0.02	0.11	-0.04	0.05
Cu	-0.33**	-0.15	-0.08	0.07	1	0.16	-0.03	0.24*	-0.09
Mo	-0.27**	-0.21*	-0.16	0.02	0.16	1	0.1	-0.06	-0.14
K	-0.06	-0.03	-0.02	0.11	-0.03	0.1	1	-0.11	0.01
Mg	-0.04	0.06	-0.07	-0.04	0.24	-0.06	-0.11	1	0.11
Ca	0.18	0.11	0.19*	0.05	-0.09	-0.14	0.01	0.11	1

[1.] * *P*<0.05; ** *P*<0.01.

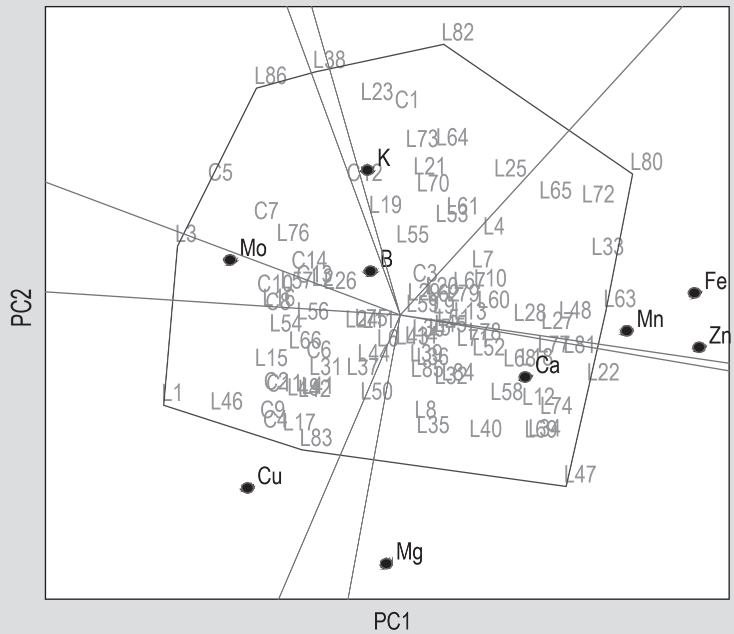

Figure 2. Polygon views of the genotype trait-plot based on symmetrical scaling for the which-won-what pattern for genotypes and mineral contents. Details of genotypes are presented in Supplementary Table S1.

the markers of the genotypes that are further away from the biplot origin so that all other genotypes are contained in the polygon (Kaya *et al.*, 2006). The rays in Figure 2 are lines that are perpendicular to the sides of the polygon or their extensions. This view helps identify genotypes with the highest values for one or more traits. Landraces L47, L22, L63, L80, L82, L38, L86, C5, L3, L1 and L83 were the vertex genotypes (Figure 2). Vertex genotypes are usually the best in their sectors (Yan and Kang, 2003). The scores of traits Fe, Zn and Mn, are located in the L80 sector, suggesting that cultivar L80 had highest or near-highest values for these three mineral concentrations. Similarly, landraces L22, L81, L63 which were on the polygon can be evaluated in the same sector. These genotypes were second or third to L80 for these traits. Similarly, landrace L83 was the highest in Cu concentration. Landrace L47 had the highest Mg content. Landrace L3 had the highest Mo content (Figure 2). In our study, most of the mineral variations of Turkish wheat genotypes were explained by the biplot.

In multi-environmental trials, the ideal genotype is located in the first concentric circle in the biplot (Kaya *et al.*, 2006). Desirable genotypes are those located close to the ideal genotype. Thus, starting from the middle concentric circle, concentric circles were drawn to help visualise the distance between genotypes and the ideal genotype. The ideal genotype can be used as a benchmark for selection. Genotypes that are far away from the ideal genotype can be rejected in early breeding cycles (Figure 3) while genotypes that are close to it can be considered in further tests (Yan and Kang, 2003). Placed near the first concentric circle, landraces L47 and L22 can thus be used as benchmarks for evaluation of bread wheat genotypes. Landraces L69, L74, L58, L81, L77, L18, L68, L40 and L52 were located near the ideal genotype, thus they are considered desirable genotypes (Figure 3). Interestingly, ideal genotypes (landraces 47 and 22) were the vertex genotypes which were the best with several mineral contents (such as Ca, Zn, Fe and Mn) in the previous biplot (Figure 2). Undesirable genotypes were those far away from the first concentric circle; most of them were cultivars, namely, Demir-2000, Gun-91, Zencirci-2000, Konya-2002, L3 and L86 (Figure 3).

In GGE-biplot, the ideal environment is representative and has the highest decimating power (Yan and Kang, 2003). In GGE-biplot, the ideal test environment should have large PC1 scores and small (absolute) PC2 scores (Kaya *et al.*, 2006). Although such an ideal trait may not exist in reality, it can be used as a reference for genotype evaluation based on multiple traits. We can evaluate ideal trait in GT-biplot, similarly to the ideal genotype. Such an ideal trait is represented by an arrow pointing to it (Figure 4). A trait is more desirable if it is located closer to the ideal trait. On the other hand, more desirable traits are strongly positively correlated with ideal trait in biplot. Thus, using the ideal trait as the centre, concentric circles were drawn to help visualise the distance between each trait and the ideal trait (Yan and Kang, 2003). In ideal trait evaluation, Figure 4 indicates that Fe, Zn and Mn fell into the centre of concentric circles. These are ideal traits in terms of being the most representative of the overall traits and the most powerful to discriminate genotypes. Being nearest

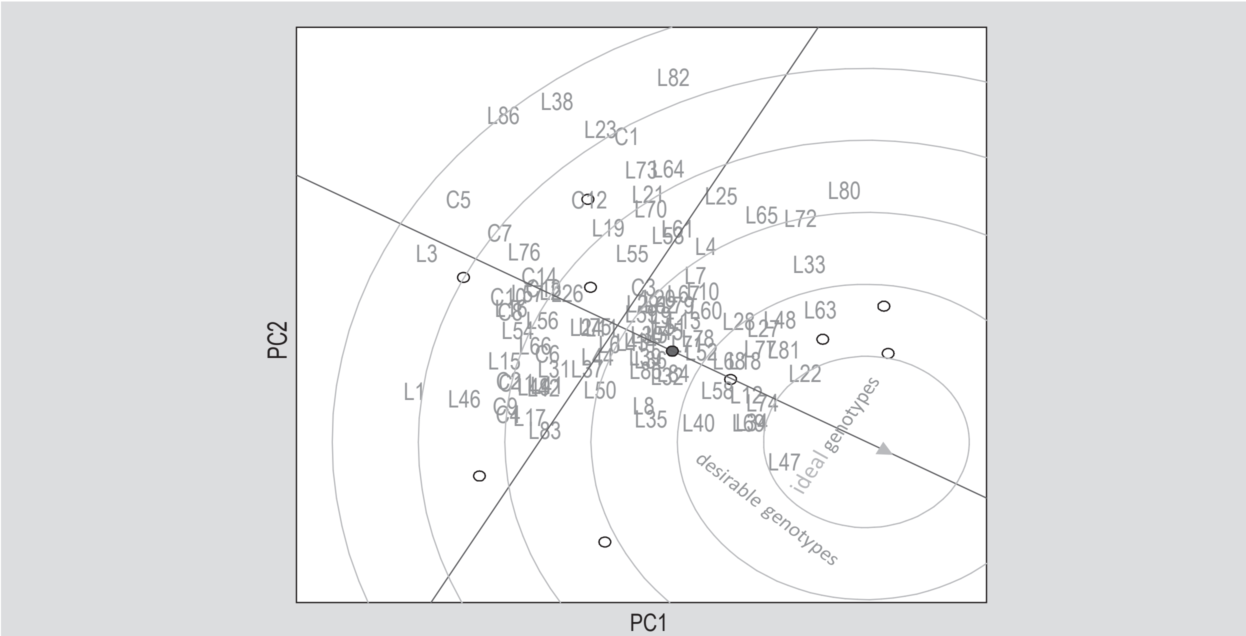

Figure 3. Genotype trait-biplot based on genotype-focused scaling for comparison of the genotypes with the ideal genotype.

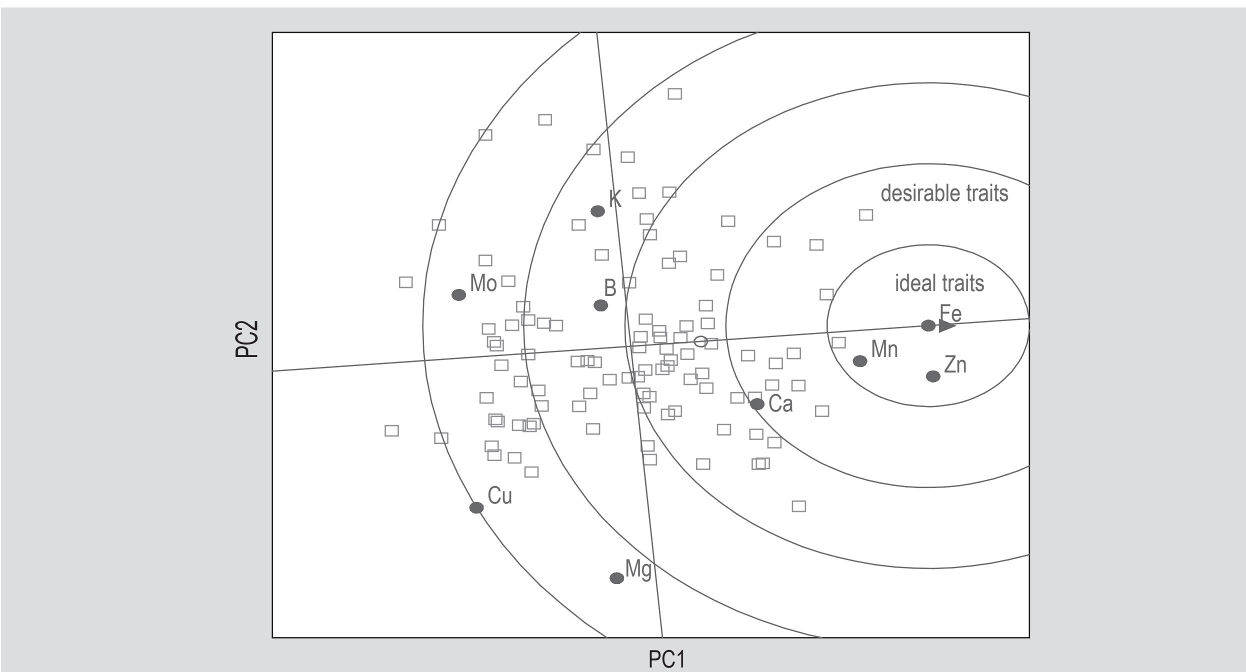

Figure 4. Genotype trait-biplot based on trait-focused scaling for comparison the traits (mineral contents) with the ideal traits.

to the first concentric circle, Ca is close to the ideal trait. Therefore, Fe, Zn and Mn content should be regarded to select higher mineral concentrations.

4. Conclusions

In this study, one hundred bread wheat genotypes (86 landraces and 14 cultivars) were evaluated in respect to nine mineral concentrations. After comparing landraces and cultivars, most of landraces revealed to have higher Fe, Zn and Mn contents than all registered cultivars. The concentration of Fe was the most effective trait according to the GT-biplot analysis. The relationship between the concentrations of the elements expressed as correlation coefficients was consistent with the results of the GT-biplot analyses.

The results of the GT-biplot analyses are more interpretable. The main advantage of the biplot is its graphical presentation of the data, which greatly enhances the ability to understand the patterns among the mineral contents. When evaluating mineral concentrations, GT-biplot can be used to evaluate genotypes based on multiple minerals to identify both ideal genotypes and ideal minerals. Especially, a wide variation was determined in grain contents of Fe, Zn and Mn in landraces. This variability can be used to develop enriched Fe and Zn wheat cultivars in wheat breeding programmes all over the world.

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Supplementary material

Supplementary material can be found online at http://dx.doi.org/10.3920/QAS2016.0886.

Table S1. Mineral compositions (mg/kg and g/kg) in grain of Turkish bread wheat landraces and cultivars.

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