Vol. 25 No. 2 1997 Cereal Research Communications

The effect of rye genetic information on zinc, copper, manganese and iron concentration of wheat shoots in zinc deficient soil

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Abstract

Experiments were conducted to study the genetic variability of Zn, Fe, Mn and Cu concentration in the shoot of wheat and other cereals. Cultivars of rye showed higher Zn efficiency than the wheats. There was a considerable variability for the four elements between rye and wheat varieties. By utilisation of disomic wheat-rye addition lines it could be demonstrated that the rye chromosomes 2R and 7R may improve the Mn and Fe concentrations of wheat, chromosome 1R the Zn and 5R the Cu, respectively. Even in a particular 4B/5R wheat-rye translocation line the Zn, Fe and Mn concentrations were significantly modified from 5.8 to 6.7, 92 to 171 and 123 to 236 µg/g DW, respectively. Both wheat and rye positively respond to Zn supply in Zn concentration of shoots, although the effect in rye is less pronounced. The average increases in Zn concentration in wheat and rye reached 851 % and 689 %, respectively, and correlated with a subsequent decrease of Cu>Fe>Mn. Among the wheats there were remarkable differences in the uptake of Zn additionally applied. The English variety 'Avalon' showed the best Zn utilisation.

Key words: micronutrient density, wheat-rye chromosome additions, rye translocations, wheat

Introduction

More than two billions of people, especially in many Third World countries, suffer not only by lack of adequate quantities of food, but also by food that does not meet the minimum daily nutritional requirements. It is particularly true for malnutrition resulting from inadequate intakes of available forms of trace elements, e.g. Fe and Zn (Bouis, 1993, Graham & Welch, 1996). World-wide, cereal grains comprise more than 50 % of the edible dry matter consumed. Wheat is one of the primary staple foods eaten by impoverished inhabitants. However, often it contains very low amounts of adequate and available quantities of iron (Fe) and zinc (Zn), and to less extent copper (Cu) or manganese (Mn) (Salunkhe & Deshpande 1991). Therefore, an increase of micronutrient concentrations and an improvement of their bioavailability is of great importance for many regions throughout the world. A side-effect of higher concentrations of micronutrients in seeds could be an increase of agronomic performance of such wheats by better seedling vigour and better resistance to fungal root diseases when re-sown on deficient soils (Rengel & Graham 1995, Graham & Welch 1996). The natural variation for micronutrient density of wheat

tissue and seeds is not well documented. Beusichem & Graham (unpubl. data) gave first data considering Fe loading of seeds. In their study there was a phenotypic variation by about 50 % in Fe concentration. Other results (Graham et al. 1992, Cakmak et al. 1996a,d) on Zn density of shoots and seeds of tetraploid and hexaploid wheats show a comparably low degree of variation when tested under Zn-sufficient and Zn-deficient soil conditions.

The present investigation should reveal further genetic diversity among wheat accessions and rye, which is known as a highly Zn efficient crop (Cakmak et al. 1996c,d; Schlegel et al. 1996) under Zn-deficient conditions. Zn deficiency was simulated in order to detect Zn-efficient genotypes for large Zn-deficient cropping areas of Turkey. Besides Zn efficiency, a high Zn, Cu, Fe and Mn accumulation in the tissue is desirable. Therefore, the translocation of the trace elements into the shoots was a main subject of the present study. Moreover, the investigation should also contribute to a genetic characterisation of several aneuploid tester lines in order to utilise them for targeted experiments of alien introgression.

Material and Methods

The wheat, rye, wheat-rye addition and translocation lines used in the experiment are described by Schlegel et al. (1991, 1996). The aneuploid material was microscopically checked for the presence of the alien chromosome and correct chromosome number applying the C-banding procedure after Schlegel and Gill (1984).

Pot experiments were carried out under greenhouse conditions of Adana (Turkey) with 2.2 kg soil/plastic pot. All experiments were carried out in three replications. Zn-deficient soil from Eskisehir (Turkey) were used, where Zn deficiency is widespread (Cakmak et al. 1996a). Plants were supplied with (10 mg Zn/kg soil), or grown without, Zn supply for 38 days. All pots were randomised every 4-5 days and watered daily to about field capacity using deionised water. Concentrations of Cu, Fe, Zn and Mn in shoots were measured by Atomic Absorption Spectroscopy (AAS) after ashing samples at 550 °C, and dissolving ash in 3.5 % HCl (Cakmak et al. 1996a). For statistical analysis the t-Test and multiple correlation tests were applied.

Results

Out of a larger experiment with several other wheat-related cereals, in this study several wheat genotypes were compared with rye cultivars concerning the micronutrient concentrations in the shoots (µg/g DW). In Tab. 1 the mean concentrations of Zn, Cu, Fe and Mn in shoots are given for 14 wheat and six rye accessions which were tested in Zn-deficient substrate. Largest amounts of the four microelements were found for Mn, followed by Fe, Cu and Zn, both for wheat and rye. While the variation for the concentrations was comparative low in wheat, rye showed a wider range of diversity, especially for Mn and Fe. Average values for Zn, Mn and Cu are slightly higher in rye, compared to wheat. Multiple correlation analysis did not reveal statistically significant correlation coefficients between the concentrations of Zn, Cu, Fe and Mn. However, in rye there is a significant correlation between the Fe and Cu concentrations. In both species there is a negative correlative tendency between the Mn and Zn accumulation (Tab. 2).

Table 1 Comparison of different wheat and rye cultivars for zinc, iron, manganese and copper concentrations in the shoot (µg/g DW) under Zn-deficient soil conditions

Genotype	Zn	sd	Fe	sd	Mn	sd	Cu	sd
WHEAT								
Avalon	6.2		100		120		16	
Besostaya 1	5.6		105		160		10	
Bolal	5.1		126		178		21	
Borenos	6.5		89		124		16	
Carola	5.8		105		177		17	
Cheyenne	6.8		99		110		16	
Chinese Spring.	6.3		100		148		18	
Dagdas	5.5		120		203		18	
Fakon	7.1		149		123		17	
Gerek 79	6.6		123		174		11	
Giza	6.8		64		125		15	
Holdfast	7.9		130		158		19	
Seri	6.6		133		184		11	
Viking	5.7		91		136		17	
Mean	**6.3**	0.7	**110**	21.9	**152**	29.0	**16**	3.2
RYE								
Dominant	7.4		101		128		20	
Graser	5.7		107		227		17	
Imperial	7.6		46		126		14.	
Inbred	8.9		142		129		28	
Pico	6.3		120		179		24	
Pluto	6.2		86		149		17	
Mean	**7.00**	1.2	**100**	32.6	**157**	40.3	**20**	5.4

Because some of the rye genotypes showed the highest Mn, Fe, Cu and Zn concentrations of all, it was hypothised that they could contribute to wheat improvement in micronutrient density in tissues. Two sets of disomic wheat-rye chromosome additions were investigated for their capability to accumulate micronutrients in shoot tissue. Indeed, the individual rye chromosomes contributed to different extents to micronutrient concentrations. Significant effects were found for the chromosomes 2R and 7R on Mn and Fe concentrations, the chromosome 1R on Zn concentration and for the chromosome 5R on Cu concentration, when they are compared to the wheat controls 'Holdfast and 'Chinese Spring'. The chromosome 1R of 'King II' may even significantly decrease the Fe concentration of wheat (see Tab.3). For determination of segmental chromosome effects of rye, several wheat-rye translocation lines were included in this study. The following translocations between the chromosomes 1R/1A, 1R/1B, 1R/1D, 4B/2R, 5R/5A and 4B/5R were

Table 2 Comparison of correlation coefficients between Zn, Cu, Fe and Mn concentrations in wheat and rye genotypes grown under Zn-deficient conditions

	Copper	Iron	Manganese
WHEAT			
Zinc	+0.10	+0.21	-0.40
Copper	-	+0.13	-0.01
Iron	-	-	+0.44
RYE			
Zinc	+0.49	+0.19	-0.77
Copper	-	+0.90**	-0.12
Iron	-	-	+0.23

** Significant at P=5 % level

Table 3 Comparison of two wheat-rye addition series (1R-7R) in comparison to the wheat controls 'Holdfast' (Ho) and 'Chinese Spring' (Cs) for zinc, iron, manganese and copper concentrations in the shoot (μg/g DW) under Zn-deficient soil conditions

	Zn		Fe		Mn		Cu	
	Ho -Ki	Cs -Im	Ho -Ki	Cs -Im	Ho -Ki	Cs -Im	Ho -Ki	Cs -Im
Ho	**7.9**		**130.0**		**158.0**		**19.7**	
Cs		**6.3**		**100.3**		**148.7**		**18.7**
Im		7.6		46.0		126.0		18.7
1R	5.8	*7.4*	*77.0*	92.3	136.0	151.7	15.3	19.7
2R	6.1	5.6	*159.0*	92.3	*222.0*	131.0	*22.7*	18.3
3R	7.2	5.9	116.0	109.0	173.7	139.0	17.0	20.7
4R	6.8	6.4	127.0	94.7	157.3	135.7	18.0	18.0
5R	6.8	5.2	131.7	112.7	166.5	151.7	19.0	*23.3*
6R	-	5.7	-	84.3	-	125.7	-	17.0
7R	6.9	6.7	*154.3*	74.7	*211.7*	126.3	17.3	18.0

Significant values compared to the wheat controls (at P=5%) are written in italics

considered (for detailed description see Schlegel et al. 1996). One of them, the 4B/5R translocation line of the variety 'Viking', was higher in shoot Mn and Fe concentrations than all the other *Agropyron*, wheat-*Haynaldia* and wheat-*Aegilops* translocations and even all other wheats, ryes, oats, triticales, several wheat hybrids (for description of material see Schlegel et al. 1996). The averages of 74 entries were at 101.4 (Fe) and 145.0 μg/g DW (Mn) (unpubl. data). In the 4B/5R translocation line (Viking-trans) the Cu content was not remarkably improved. But

the zinc, iron and manganese concentration rose from 5.8 to 6.7, 92 to 171 and from 123 to 236 µg/g DW, respectively (cf. Fig. 1).
The experiment was simultaneously used for studying the genotype response to zinc application (10 mg Zn/kg soil). Both wheat and rye positively responded in Zn concentration of shoot, particularly the wheat (Fig. 2). The increases of about 850 % in wheat and about 690 % in rye correlated with a subsequent decrease of Cu>Fe>Mn. Among the wheats there were remarkable differences. The English

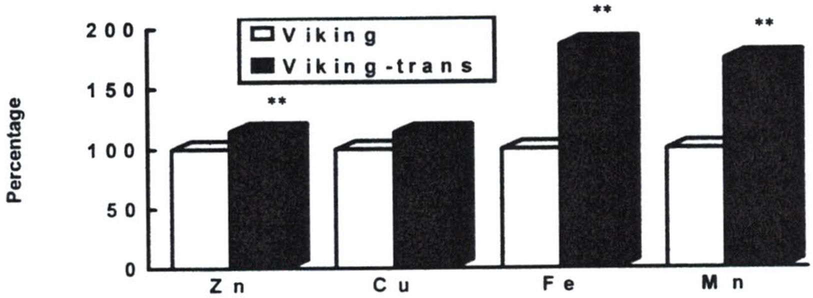

Figure 1 Relative increase of zinc, copper, iron and manganese concentrations of shoots (µg/g DW) of hexaploid wheat affected by the presence of a translocated chromosome 5RL segment as compared to the control 'Viking'; plants were grown under Zn-deficient conditions; ** significant difference at P=5 %

variety 'Avalon' surprisingly showed the highest increase in Zn concentration, followed by the Turkish varieties 'Bolal' and others. The lowest Zn concentration was found in 'Holdfast'. Although the rye varieties are less differentiated, the wheat-rye addition lines reveal that individual chromosomes of rye may improve the Zn utilisation of wheat (Fig. 3a,b). Again the rye chromosomes 1R, 2R and 7R of 'King II' and chromosome 5R of 'Imperial' seem to be critical for that behaviour.

Discussion

Despite the differences in resistance to Zn deficiency described in this paper and elsewhere (Schlegel et al. 1996), the wheat and rye cultivars did not strongly differ in Zn concentration in shoot (Tab. 1). Different approaches for revealing genetic variability in Zn have to be taken in account for gaining additional genetic variability. Induced mutations or alien genetic information could contribute to broadening variation.
Since the average differences between wheat and rye cultivars are not heavily deviate, rye cannot be determined as a general donor to increase micronutrient density in wheat, although its importance for improvement of Zn efficiency is irrefutable (high yield performance under Zn deficiency). Nevertheless, the maximum Zn, Cu, Fe and Mn concentrations of rye genotypes and/or populations are as high

as in wheat or mostly higher than wheat (Tab. 1, Cakmak et al 1996d). This is not only true for the whole rye genome but also for individual chromosomes. Single chromosomes from the rye variety 'King II' significantly increased the Zn, Cu, Fe and, particularly, Mn contents of wheat shoots.

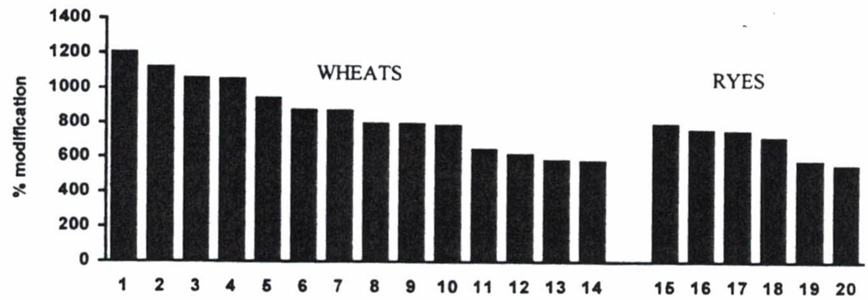

Figure 2 The relative increase or decrease (as a percentage) in concentrations of Zn in the shoot after zinc supply to the Zn-deficient soil; wheat varieties (1-Avalon, 2-Bolal, 3-Seri, 4-Giza, 5-Gerek, 6-Chinese Spring, 7-Dagdas, 8-Besostaya 1, 9-Carola, 10-Viking, 11-Borenos, 12-Fakon, 13-Cheyenne, 14-Holdfast) and rye varieties (15-Graser, 16-Dominant, 17-Pluto, 18-Pico, 19-Imperial, 20-Inbred line)

Accumulation of Fe in shoots of Zn-deficient plants was already reported. It was discussed that high Fe concentration in shoots of Zn-deficient plants is a result of either enhanced Fe uptake or a 'concentration effect' due to reduced shoot growth (Cakmak et al. 1994, Walter et al. 1994). Also Mn is accumulated in Zn-deficient shoot tissue of different plant species (Cakmak, unpubl. data). Manganese efficiency seems to be controlled by genes of rye chromosome 2R (Graham 1987). The same chromosome was determined in our study as critical for Mn accumulation. The efficiency is related to the enhanced uptake and translocation to the shoot (Graham 1988). Similar to manganese Fe efficiency is related to the Fe translocation to the shoot. Such type of linkage would be highly appreciated for selection. A positive genetic potential is carried on the chromosomes 2R and 5R for Cu uptake, and the chromosomes 2R and 7R for Fe and Mn uptake. The chromosomes 2R and 5R also showed the highest copper efficiency (Werner et al. 1990). Thus, Cu-efficient genotypes may also improve the Cu density in the tissue. The effect of the chromosomes 2R and 7R on Fe and Mn concentrations is not yet documented, and it seems not to be related to Fe efficiency, after own data. The rye 'King II' and its chromosome additions to wheat did not receive best scores for Fe efficiency, when they were grown under Fe-deficient conditions (Block & Schlegel, 1993, unpubl.). On the other hand, the multiple increased Fe and Mn concentrations in the 4B/5R wheat-rye translocation line of the wheat variety 'Viking', compared to the Zn-sufficient plants and to the controls, suggest an association at least between zinc inefficiency and iron accumulation.

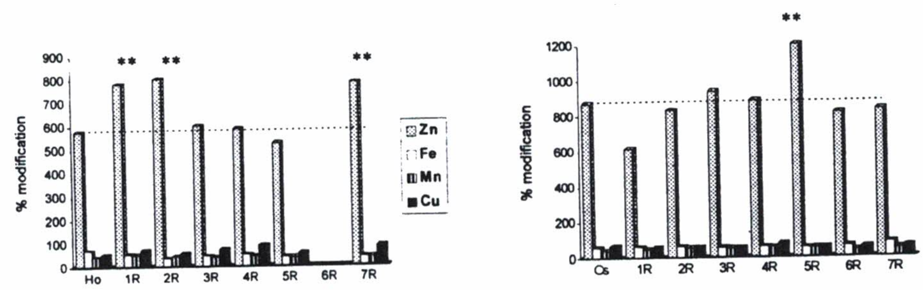

Figure 3 The relative increase or decrease in concentrations of Zn, Fe, Mn and Cu in the shoot after zinc supply to the Zn-deficient soil; Holdfast-King (left) and Chinese Spring-Imperial (right) wheat-rye addition lines (1R-7R) and the wheats 'Holdfast' (Ho) and 'Chinese Spring' (Cs) as controls

The same translocation line was previously determined to be high Cu- and Fe-efficient when it was grown under deficient conditions (Schlegel et al. 1991). High Cu or Fe efficiency was accompanied by a strong exudation of phytosiderophores such as 2'-deoxymugineic acid (DMA=2843 µmol/l) and mugineic acid (MA=48 µmol/l) (Schlegel & Römheld 1993, unpubl.). In the same experiment Cu- and Fe-efficient plants showed exudation of DMA to a lower extent (1154 µmol/l). Because the chromosome 5R was not identified as contributor to high Fe and Mn concentrations by using the addition lines in this study, but critical by the 5R translocation of 'Viking' (Fig. 1), it is concluded that either 5R chromosomes of other rye genotypes carry such alleles (the rye donor of Viking-trans is not known) or the genetic background of 'Viking' mediates a high acquisition of Fe and Mn. The close positive correlation between the Fe and Mn contents in rye is remarkable (r=0.90**), just as the trend to a negative correlation between the Zn and Mn contents both in rye and wheat.
The present investigation exhibited the existence of wheat and rye genotypes which show significant increases of concentrations of microelements in shoot tissue in Zn-deficient soil. When there is a high accumulation of Zn in shoot tissue also a higher Zn concentration in the grains can be expected (Welch 1986). It encourages to larger screenings for related plant material. The purposeful utilisation of individual rye chromosomes is suggested.

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Acknowledgement: This work was supported by NATO's Scientific Affairs Division and in part by TÜBITAK (The Scientific and Techn. Res. Council of Turkey)

Received 10 February, 1997, accepted 12 March, 1997

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