Tolerance of Some Wheat Varieties to Boron Toxicity

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Boron (B) toxicity is an important problem in low rainfall and highly alkaline soils of central and southern part of Iran. We evaluated B toxicity tolerance of 10 Iranian wheat varieties in a greenhouse experiment. Experimental design was factorial Completely Randomized Design (CRD) with 10 wheat varieties × six B levels (0, 2.5, 5, 10, 20 and 40 mg B kg^{-1} soil) in three replications. The results showed a great range of tolerance among wheat varieties. Arg was the most tolerant one and Chamran showed the least tolerance. It seems that different mechanisms involved in B toxicity tolerance, namely exclusion of B from root, redistribution of B within leaves and integration of these two mechanisms. Significant negative linear correlation observed between shoot B concentration and shoot dry weight (r = 0.85, $p < 0.01$) and positive linear correlation between shoot B concentration and shoot dry weight reduction percentage (r = 0.82, $p < 0.01$).

Keywords: boron toxicity, wheat varieties, boron tolerance

Introduction

Boron (B) is one of the essential micronutrients for plant growth, widely distributed in lithosphere and hydrosphere. B is known to be unusual micronutrient in that the range between toxicity and deficiency is narrow and at physiological and common soil pHs exists as un dissociated boric acid (Camacho-Cristóbal et al. 2008; Yau and Ryan 2008; Herrera-Rodríguez et al. 2010; Reid 2010).

Both B deficiency and toxicity can suppress plant growth. B deficiency can be easily corrected but B toxicity is a major micronutrient disorder affecting yield of many crops especially cereals around the world (Kalayci et al. 1997; Savić et al. 2013). Excessive B in soil and irrigation water causes B toxicity in crops and more over it is a common nutritional problem in low rain fall and dry areas especially in alkaline soils of Australia, North Africa and West Asia (Nable et al. 1997; Camacho-Cristóbal et al. 2008). B toxicity is probably first reported in the united states but its importance was not sufficiently recognized until the 1980s when significant yield reduction observed in South Australia. Two main factors determining accumulation of high B level in soils are irrigation water (the most important contributor) and soils which inherently have high levels of B (Nable et al. 1997; Yau and Ryan 2008).

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Since ameliorating of B toxic soils is impractical in the most circumstances selecting or breeding of crop cultivars with high B toxicity tolerance is the only practical solution to increase yield in high B soils. Different works showed a wide range of B tolerance between and within plant species such as barely (Rehman et al. 2006), chickpea (Ardıc et al. 2009), tomato (Güneş et al. 2009), canola (Öztürk et al. 2010), lentil (Hobson et al. 2006), maize (Hakki et al. 2007), *Brassica rapa* (Kaur et al. 2006), oilseed rape (Savić et al. 2013) and wheat (Karaman et al. 2012).

Karaman et al. (2012) reported that bread wheat genotypes are more B tolerant than durum wheat. Furlani et al. (2003) evaluated the response of four wheat genotypes to different B levels and concluded that IAC 287 and IAC 24 genotypes were more tolerant to B toxicity. Using 70 durum wheat genotypes in a greenhouse experiment to study genotype variation to B toxicity Torun et al. (2006) reported that dry matter production of all genotypes, except four, were markedly decreased by applied B. Kalayci et al. (1997) concluded that the most B tolerant wheat cultivars were of local origin. Breeding accessions of barely from Iran and Afghanistan were the most tolerant among seven WANA countries (Yau 2002).

Central and southern parts of Iran are typical arid and semi-arid areas with low rain fall. The soils of this area are characterized by low organic matter (OM) content, high soil pH, high calcium carbonate content and coarse texture, which make them susceptible to induce B toxicity to plants, besides high levels of B in irrigation water and B toxicity symptoms reported from this area (Rajaie et al. 2009). In spite of wheat culture importance in central and southern parts of Iran no study on screening of B toxicity tolerance of Iranian wheat varieties has been reported. The current work was conducted to study differential responses of 10 Iranian wheat varieties to B toxicity in greenhouse conditions to select some cultivars having certain mechanisms controlling B toxicity for culture in soils containing toxic levels of B and find possible varieties for B toxicity tolerance breeding programs.

Materials and Methods

In order to compare B toxicity tolerance of some Iranian wheat varieties a greenhouse experiment was conducted with 10 wheat varieties, namely Verinac (V1), Hamoon (V2), Chamran (V3), Arg (V4), S7811 (V5), Pishtaz (V6), Parsi (V7), Arta (V8), Karkhe (V9) and Yavares (V10) (two last were durum wheat) and six B levels (0, 2.5, 5, 10, 20 and 40 mg B kg^{-1} soil). Seeds were prepared from Seed Propagation Research Institute of Iran. The soil used in this study was from Iranian southern part (28°32′48″ N and 57°51′04″ E), sandy loam in texture and 8% clay content (Bouyoucos 1962). Some soil chemical properties (Sparks et al. 1996) were as: EC = 3.5dS m^{-1}, pH (saturation paste) = 7.9, saturation percentage = 27.9%, organic carbon content = 0.62%, cation exchange capacity = 8.12 cmol$_c$ kg^{-1}, calcium carbonate equivalent = 4.90% and hot water soluble B (HWSB) = 0.9 mg kg^{-1}.

Three kg of air dried soil replaced in plastic bags and B treatments as H$_3$BO$_3$ solution added to each one. In addition 35 mg N kg^{-1} soil as urea, 50 mg K kg^{-1} soil as K$_2$SO$_4$,

5 mg Fe kg^{-1} soil as FeEDHHA, 10 mg Zn kg^{-1} soil as ZnSO$_4$ and 10 mg Mn kg^{-1} soil as MnSO$_4$ were added to each pot uniformly. Soil in each bag reached to field capacity by adding distilled water and mixed thoroughly and translocated to poly ethylene pots. Two 35 mg N/kg^{-1} soil as urea top dressed in three weeks intervals. Ten wheat seeds of each variety were planted in every pot and thinned to 5 uniform plants two week after planting. Day/night temperature was adjusted between 29/23 °C and 200 μmol m^{-2} s^{-1} irradiance with 12 hours photoperiod. Soil moisture controlled between 70–90% of field capacity (FC) by weighting pots and adding distilled water daily. Fifty-four days after sowing plants were cut from soil surface and shoot fresh weight determined, then washed thoroughly in tap and distilled water and oven dried in 65 °C for 48 hours. Roots separated from soil and washed in tap and distilled water precisely to avoid any contamination and root dry weight and root B concentration determined. For B concentration measurement, 0.5 g of each of the shoot and root sample was dry-ashed in a muffle furnace at 500 °C for 6 h (Cottenie 1980). The ash was then dissolved in 0.1N HCl and B was determined colorimetrically (420 nm) using the azomethine-H method (Keren et al. 1996). Values were expressed as mg kg^{-1} DW.

Statistical analysis

Recorded data were processed by the analysis of variance (ANOVA) in a factorial experiment on the basis of Completely Randomized Design (CRD) with 10 wheat varieties × six B levels (0, 2.5, 5, 10, 20 and 40 mg B kg^{-1} soil) in 3 Replications and finally 180 pots. The data were analyzed by SAS 9.2 and Sigma Plot 12 computer soft wares. Treatment means were compared using LSD (*t*-test). Slicing method was used for interaction analysis.

Results

The effect of B levels, variety and B levels × variety on shoot and root dry weight and shoot and root B concentration were significant. Increasing B levels up to 5 mg kg^{-1} did not affect shoot dry weight but higher B levels decreased shoot dry weight significantly (Table 1). In Chamran variety the second B level (2.5 mg kg^{-1}) reduced shoot dry weight significantly that shows this variety is very sensitive to B toxicity. From the other side varieties like Arg, Karkhe, Arta, Pishtaz, S7811 and Hamoon showed significant dry weight reduction with increasing B level up to 20 mg kg^{-1}, meaning that these varieties are relatively tolerant to B toxicity. Other varieties are between these two categories. Arg variety showed significantly higher shoot dry weight in different B levels than other varieties that shows B toxicity tolerance in this variety (Table 1). These results confirmed by shoot fresh weight results (data not showed). Root dry weight results were similar to shoot dry weight although it seems that B levels affect shoots more than roots. Increasing B levels up to 5 mg kg^{-1} did not affect root dry weight but higher B levels decreased root dry weight significantly. Arg variety produced significantly higher root dry weight than other varieties in different B levels (Table 1).

Table 1. Shoot dry weight (g pot⁻¹), root dry weight (g pot⁻¹), shoot B concentration (mg kg⁻¹) and root B concentration (mg kg⁻¹) of 10 wheat varieties as affected by B levels

	B1 (0)	B2 (2.5)	B3 (5)	B4 (10)	B5 (20)	B6 (40)	Mean
Shoot dry weight (g pot⁻¹)							
Verinac	1.024±0.07Aabc	1.058±0.02Aabc	1.017±0.13Acd	0.737±0.10Bbc	0.604±0.07Bbc	0.190±0.03Ccd	0.772CDE
Hamoon	0.762±0.12Ac	0.84±0.01Ac	0.789±0.13Ad	0.816±0.07Aabc	0.469±0.1Bbc	0.286±0.03Bbcd	0.660E
Chamran	1.392±0.04Aa	1.082±0.06Babc	0.744±0.11Cd	0.581±0.12CDc	0.447±0.02Dc	0.080±0.03Ed	0.712DE
Arg	1.278±0.32Bab	1.263±0.08Ba	1.600±0.04Aa	1.115±0.23BCa	0.966±0.15Ca	0.522±0.14Da	1.124A
S7811	1.081±0.12Aabc	0.984±0.04Abc	0.980±0.15Acd	1.055±0.1Aab	0.557±0.03Bbc	0.347±0.11Babc	0.834BCD
Pishtaz	1.024±0.07Aabc	1.113±0.12Aab	1.011±0.06Acd	0.867±0.14ABabc	0.596±0.11BCbc	0.439±0.05Ccd	0.842BC
Parsi	1.343±0.03Aa	1.280±0.16Aa	1.357±0.12Aab	0.928±0.08Bab	0.520±0.07Cbc	0.180±0.04Dbc	0.935B
Arta	0.876±0.07Abc	0.943±0.11Abc	0.799±0.11ABd	0.739±0.07ABbc	0.563±0.04BCbc	0.336±0.08Cabc	0.709DE
Karkhe	1.117±0.21Aabc	1.066±0.09Aabc	1.182±0.05Abc	0.912±0.09ABabc	0.695±0.07Bab	0.278±0.06Cb	0.875BC
Yavares	1.234±0.13Aab	1.108±0.07ABab	0.938±0.11Bcd	0.838±0.05Babc	0.523±0.07Cbc	0.340±0.02Cabc	0.830BCD
Mean	1.133a	1.074a	1.042a	0.859b	0.594c	0.300d	
Root dry weight (g pot⁻¹)							
Verinac	0.229±0.02Ac	0.242±0.04Ab	0.226±0.01Ac	0.161±0.02ABd	0.171±0.02ABbcd	0.079±0.01Bde	0.185F
Hamoon	0.350±0.09Abc	0.346±0.03Ab	0.333±0.09Ac	0.421±0.06Aab	0.194±0.03Bcd	0.121±0.01Bcde	0.294CD
Chamran	0.553±0.06Aab	0.416±0.07ABab	0.284±0.09BCc	0.164±0.03CDd	0.117±0.01Dde	0.049±0.01De	0.264CDE
Arg	0.716±0.19Aa	0.535±0.04Ba	0.734±0.08Aa	0.505±0.1BCa	0.388±0.04CDa	0.254±0.05Da	0.522A
S7811	0.275±0.03Ac	0.233±0.01Ab	0.275±0.03Ac	0.304±0.08Abcd	0.165±0.01Acd	0.210±0.04Aab	0.244DEF
Pishtaz	0.546±0.03Aab	0.558±0.08Aa	0.515±0.06ABb	0.375±0.02Babc	0.224±0.05Cbc	0.159±0.01Cbc	0.396B
Parsi	0.348±0.01Abc	0.372±0.05Aab	0.353±0.01Abc	0.242±0.02ABcd	0.190±0.02BCbcd	0.053±0.01Ce	0.259CDE
Arta	0.392±0.05Abc	0.405±0.14Aab	0.384±0.03Ac	0.303±0.01Abcd	0.283±0.04ABb	0.139±0.03Bbcd	0.312C
Karkhe	0.209±0.03Ac	0.276±0.02Ab	0.301±0.02Ac	0.255±0.06Acd	0.169±0.02ABcd	0.075±0.01Bde	0.214EF
Yavares	0.334±0.03Abc	0.251±0.01ABb	0.228±0.03ABc	0.220±0.03ABcd	0.183±0.01Bcd	0.142±0.04Bbcd	0.226EF
Mean	0.395a	0.363a	0.359a	0.295b	0.208c	0.128d	

Table 1. (cont.)

Shoot B concentration (mg kg⁻¹)

Verinac	174.5±14Eab	410.6±13Dcd	546.4±20Db	1022±33Cd	2330±84BBc	4362±42Aa	1474BC
Hamoon	307±4Ea	576±3Dab	657±20Dab	1460±66Ca	2837±92Ba	3767±45Ade	1601B
Chamran	237.2±4Fab	485.9±8Ebc	786.2±9Da	1117±48Ccd	1809±23Bd	2896±104Ag	1222E
Arg	134.5±3Eb	254.2±14DEc	331.9±20Dc	567.5±33Cf	975.1±18Bf	1759±41Ah	670.4G
S7811	125.3±4Eb	286.9±11Dde	339.5±20Dc	793.6±43Ce	1259±47Be	3306±52Af	1018F
Pishtaz	276.1±15Ea	703.7±3Da	746.3±81Da	1183±10Cac	2958±75Ba	4814±36Aa	1780A
Parsi	184.1±9Fab	409.9±8Ecd	692±30Da	1135±14Ccd	2833±104Ba	3627±52Ae	1480C
Arta	220.1±10Fab	431±19Ec	760.3±33Da	1287±38Cb	1847±81Bd	3692±196Ae	1376D
Karkhe	226.7±14Fab	479.5±13Ebc	775.8±23Da	1142±44Cbcd	2446±100Bb	3876±66Ad	1491C
Yavares	215.6±8Fab	470±11Ebc	723.5±19Da	1191±43Cbc	2247±39Bc	4199±126Ac	1508C
Mean	210.1f	450.8e	636d	1089c	2157b	3630a	

Root B concentration (mg kg⁻¹)

Verinac	75±1.1Da	79.5±4.1Da	81.9±2.9Dbcde	157.3±4.5BCab	212.2±11.6Be	475.1±13Ad	180.2D
Hamoon	69±3.7Dab	59.07±2.1Dab	99.37±3.1Cabc	97.53±3.6Cfg	243.1±5.7Bd	377.8±14.7Af	152.1E
Chamran	39.6±0.8Fcd	62.87±1.8Eab	104.8±3.2Dab	171.8±8.6Ca	359.8 22.9±Ba	582.2±9.2Ac	220.2B
Arg	31.7±2.4Dd	54.3±2.7Db	52.33±1.6Df	77.17±2.9Cg	179±7.5Bf	378.2±17.3Af	128.8F
S7811	56.9±3.1Dabc	71.3±3.1Dab	75.23±7.2Ddef	103.5±2.4Cef	232±8.4Bde	560.7±16.1Ac	183.3D
Pishtaz	36.73±1.7Ecd	51.23±3DEb	67.43±7.4Def	116±5.2Cdef	283.2±16.2Bc	734±28.6Aa	214.8B
Parsi	47.03±5.5Ebcd	64.13±7DEa	79.17±3.7Dcde	129.2±7.7Ccd	245.6±8.6Bd	630.8±7.8Ab	199.3C
Arta	46.3±1.6Ebcd	82.43±2.6Dab	76.80±4Dcde	119.2±5.7Ccdef	288.2±14.6Bbc	472.2±10.9Ad	180.9D
Karkhe	63.2±2.5Eab	70.67±6.5Eab	97.17±3.6Dbcd	124.9±2.3Ccde	361.7±4.5Ba	717.2±2.8Aa	239.1A
Yavares	52.80±4.8Eabcd	72.27±3.2Dab	111.7±3.7Ea	141.4±3Cbc	311±6.5Bb	423.8±7.3Ae	185.5D
Mean	51.83f	66.78e	84.59d	123.8c	271.6b	535.2a	

Data represent means ± standard error of 3 independent replications.

Mean separation by LSD test, $P < 0.05$. Capital letters are used to indicate variation between the plant growth indices of each cultivar in different B levels, and small letters are used to indicate variation of plant indices of different cultivars in each B level.
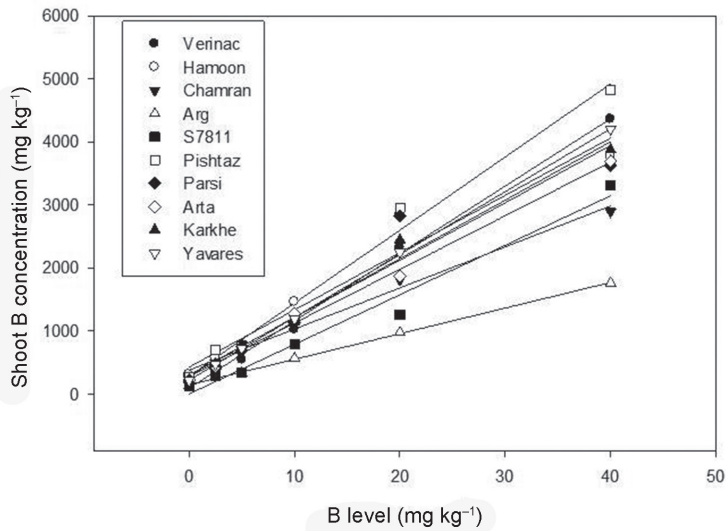
Figure 1. Effect of B levels on shoot B concentration in different wheat cultivars

Shoot and root B concentration increased significantly with increasing B levels (Table 1). Mean shoot and root B concentration of all genotypes correlated significantly to B application levels (Figs 1 and 2).
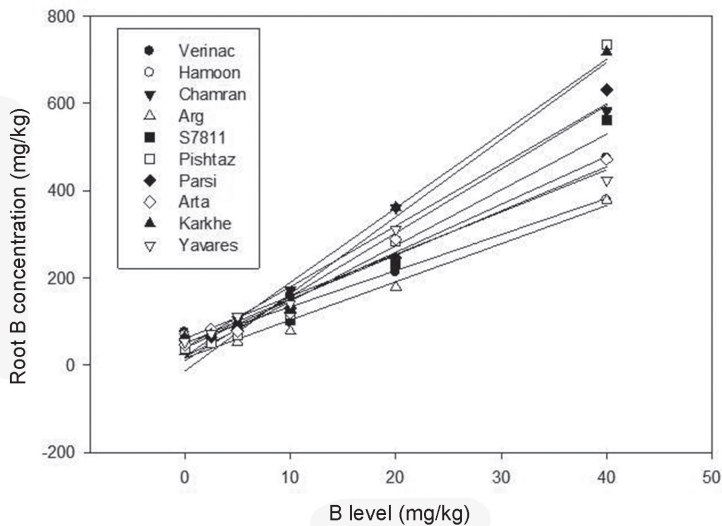

Figure 2. Effect of B levels on root B concentration in different wheat cultivars

A great range of shoot and root B concentration observed in different wheat varieties. Mean shoot and root B concentration in different B levels ranged between 670.4–1780 mg kg^{-1} and 128.8–239.1 mg kg^{-1}, respectively, for different wheat varieties (Table 1). There was a wider range for B concentration in shoot than root. The least shoot and root B concentration observed in Arg and the highest shoot and root B concentration were in Pishtaz and Karkhe respectively. Significant correlation (r = 0.93, $p < 0.01$) observed between shoot and root B concentration and a negative significant correlation (r = 0.85, $p < 0.01$) observed between shoot B concentration and shoot dry weight. Similarly significant negative correlation (r = 0.66, $p < 0.01$) observed between root B concentration and root dry weight.

Mean of shoot and root dry weight reduction percentage at two last B levels (20 and 40 mg kg^{-1}) ranged between 41.81–81.01% and 31.82–84.99%, respectively (Table 2), which shows large variation in B toxicity tolerance among wheat varieties. Based on shoot dry weight data tolerance of different wheat varieties are as:

Arg > Arta > Pishtaz > Hamoon > Karkhe > S7811 > Verinac > Yavares > Parsi > Chamran

Arg variety with minimum dry weight reduction for B highest levels had significantly higher shoot and root dry and fresh weight and significantly lower shoot and root B concentration than other varieties and can be considered as the most tolerant variety to B toxicity among experimental varieties. From the other side Chamran showed the highest dry weight reduction for highest B levels and can be considered as the most sensitive variety to B toxicity. Between two-tested durum wheat cultivar Karkhe is tolerant to B toxicity and can be cultivate in high B soils. A significant correlation observed between shoot B concentration and shoot dry weight reduction percentage (r = 0.82, $p < 0.01$).

Table 2. Reduction percentage of shoot and root dry weight in two highest B levels in different wheat varieties

	Shoot		Root		Mean	
	B5 (20)	B6 (40)	B5 (20)	B6 (40)	Shoot	Root
Verinac	41.02	81.45	25.18	65.36	61.23	45.27
Hamoon	38.47	62.41	44.72	65.56	50.44	55.14
Chamran	67.91	94.11	78.84	91.14	81.01	84.99
Arg	24.44	59.18	45.74	64.56	41.81	55.15
S7811	48.47	67.87	39.88	23.76	58.17	31.82
Pishtaz	41.83	57.13	59.06	70.90	49.48	64.98
Parsi	61.28	86.62	45.35	84.76	73.95	65.55
Arta	35.76	61.68	27.74	64.51	48.72	46.12
Karkhe	37.87	75.11	19.01	64.22	56.44	41.62
Yavares	57.59	72.47	45.31	57.39	65.03	51.35

Discussion

In this study, the negative effect of high B levels on shoot and root growth varied among wheat varieties. Large variations in B toxicity tolerance among wheat varieties were seen for plant responses (Table 1). The second B level (2.5 mg kg^{-1}) reduced shoot dry weight of Chamran variety significantly that shows this variety is very sensitive to B toxicity. There is increasing evidence showing that genotypes relatively sensitive to B toxicity are also relatively tolerant to B deficiency and vice versa (Yau 2002). On the other hand, this variety is B efficient and can be cultivated in low B soils and used as genetic material to breed varieties tolerant to B deficiency. Shoot and root dry weight of Arg variety in different B levels was significantly higher than other verities, and percent of shoot dry weight reduction in two highest B levels were minimum in this variety that shows this variety is the most tolerant to B toxicity among tested varieties. This variety could be cultivated in high B soils and could be recommended for breeding process for B toxicity tolerant genotypes. Metwally et al. (2012) concluded that high B levels reduced growth rate of B susceptible wheat Gemmeza 9 while increased tolerant sakha 93. They concluded adverse effect of high B levels on wheat growth seems to be cultivar dependent. Karaman et al. (2012) observed large genotypic variation among wheat genotypes to B toxicity. Variation of wheat varieties tolerance to B toxicity have been showed by others (Kalayci et al. 1997; Furlani et al. 2003; Torun et al. 2006).

Shoot and root B concentration increased significantly with increasing B levels and large variation in shoot and root B concentration observed in different wheat varieties. Significant increase in B concentration with increasing B levels and significant difference among wheat genotypes reported by Karaman et al. (2012).

Relative B toxicity tolerance of Arg, Karkhe, Arta, Pishtaz, S7811 and Hamoon varieties seems logical because they were evolved in dry areas where B toxicity is a problem which resulted to B toxicity tolerance of these varieties (Savić et al. 2013). Tolerant genotypes originated from regions where high B concentration has been reported in the soil and ground water (Nable et al. 1997). Yau (2002) reported that barley accessions from Iran and Afghanistan were the most tolerant among seven west Asia and north Africa countries.

There are some reports on less B toxicity tolerance of durum wheat compared to bread wheat (Kalayci et al. 1998; Karaman et al. 2012) but durum wheat Kharkhe showed relative B toxicity tolerance and can be cultivated in areas with high B levels.

Different mechanisms and/or genes are involved to induce B toxicity tolerance. Based on significant correlations between shoot B concentration and shoot dry weight reduction (r = 0.82, $p < 0.01$) and between shoot and root B concentration (r = 0.92, $p < 0.01$) it can be concluded that exclusion mechanism results to reduced concentration of B in B tolerant varieties. Kaur et al. (2006) in study of 19 *Brassica rapa* genotypes reported that tolerant genotypes used exclusion mechanism. Ardıc et al. (2009) concluded that the ability of chick pea Gokce to reduce shoot B concentration by efflux of B from roots is the reason of improved growth of this genotype in B toxic condition. The primary mechanism for tolerance to B toxicity is the ability to maintain low concentrations of B in plant tissues.

B toxicity tolerant varieties generally accumulate lower concentrations of B than sensitive ones. Active efflux is physiological reason for reduced B accumulation in tolerant varieties (Schnurbusch et al. 2010). Reid (2010) reported that wheat tolerance to B toxicity correlated with reduced tissue accumulation of B. It can be concluded from the least shoot and root B concentration of Arg that B excludes from whole plant. However a closer look to shoot and root B concentration (Table 1) and reduction percentage of shoot dry weight with B levels (Table 2) shows in varieties like Pishtaz and Hamoon that exhibit remarkable B toxicity tolerance, B concentration is very high and this suggest a mechanism rather than exclusion. Torun et al. (2006) in a study of 70 durum wheat genotypes reported that some varieties showed a good relation between B concentration and decrease in dry weight but ultimately there was no significant relationship, and suggested that an internal mechanism is involved.

It seems that the mechanism is redistribution of B within leaf. Reid and Fitzpatrick (2009a and b) showed that redistribution of B from toxic place, symplast (cytoplasm) into less toxic place, apoplast (cell wall) reduces B toxicity. It seems that in some varieties like Arta, Karkhe and S7811 both mechanisms are involved to covey B toxicity tolerance. Existence of different B toxicity tolerance mechanisms give us chances to breed varieties with improved tolerance greater than that identified in this study.

Whether the best tolerant variety to B is also effective one for grain yield and how much B is accumulated in the grain needs more prolonged experiments. To identify the nature of different tolerance to B some physiological and molecular approaches will be required.

In conclusion there was a great range in B toxicity tolerance among studied wheat varieties. Arg was the most tolerant one and Chamran showed the least tolerance. It seems that different mechanisms involved in B toxicity tolerance of wheat varieties. In some varieties (like Arg) exclusion of B from root, in some varieties (like Pishtaz and Hamoon) redistribution of B were the primary mechanisms, and in some varieties (like Arta, Karkhe, and S7811) both mechanisms are involved. The findings of this work are good base for future experiments in which one can concentrate on the two varieties Arg and Chamran as model plants suitable for experiments on different aspects of B tolerance.

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