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Genotypic variation in P efficiency of selected Iranian cereals in greenhouse experiment

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Abstract

A factorial completely randomized block design experiment with three replications was carried out in greenhouse to evaluate cereal genotypic variation in phosphorus (P) acquisition and utilization efficiency in a calcareous soil with low available P (4.6 mg P kg^{-1} soil) and high total P (1260 mg P kg^{-1} soil). Treatments consisted of eight bread wheat (*Triticum aestivum* L.), three durum wheat (*Triticum durum* L.), three barley (*Hordeum vulgare* L.), one rye (*Secale cereale* L.), one oat (*Avena sativa* L.) and one triticale (*X Triticosecale* L.) genotypes at two levels of P fertilizer (0 and 84 mg P kg^{-1} soil). Genotypes showed significant differences in chlorophyll meter reading, number of tillers, shoot P concentration and content (the total amount of P per shoot in pot), and shoot dry weight (SDW). Phosphorus efficiency (relative shoot dry weight) significantly differed among genotypes and ranged from 0.42 for barley (genotype M-80-16) to 0.97 for bread wheat Azadi. Shoot P concentration increased significantly from 1.9 to 4.7 mg g^{-1} DW and shoot P content from 13.2 to 46.1 mg P pot^{-1} by applying P. With no P supply (P_0), durum wheat Yavarus with 20.1 mg P pot^{-1} and barley line M-80-16 with 5.8 mg P pot^{-1} had the highest and lowest P content, respectively. Bread wheat Azadi (0.45) and durum wheat Yavarus (0.43) had the highest relative P content; therefore, they were efficient in P acquisition. Oat produced the highest dry weight per unit of P taken up and hence was efficient in P utilization. There was no correlation between P efficiency and shoot P concentration of genotypes (r= 0.12), but the relationship between P efficiency and shoot P content (total amount of shoot P per pot) was highly significant (r= 0.66**), suggesting that shoot P content is a reliable parameter in screening cereal genotypes during vegetative growth.

Keywords: P acquisition; P utilization; wheat; barley; rye; triticale

Introduction

Phosphorus (P) is an essential plant nutrient. Its peculiar behavior, both in calcareous soils (high Ca^{2+}) and in acidic soils (high Al^{3+} and/or Fe^{3+}), requires regular annual applications of this element to maintain optimum crop yield. The annual consumption of P fertilizer is more than 39 million tons (Heffer, 2008), ranking second after nitrogen. In Iran, the annual P fertilizer consumption is about 800,000 tons (Malakouti et al., 2008). Because of the complexity of P chemistry in soil, only about 20% of the total amount of P fertilizer is utilized by the first crop and the remaining 80% is fixed in soil in an unavailable form (Grotz and Guerinot, 2002). Excessive P accumulation in soils results in low productivity and low seed protein because of interaction with other nutrients especially micronutrients. Also, transport of soil particles loaded with P into lakes and surface waters cause eutrophication (Malakouti et al., 2008). Hence, various approaches that can increase utilization of P fertilizer in the soil-plant system have been considered, including selection of genotypes with increased P acquisition and utilization efficiency (Marschner, 1995).

Differential capacity of different plant genotypes to acquire and utilize nutrients has encouraged researchers to study nutrient efficiency as influenced by both nutrient absorption by roots and utilization in plants. The relative importance of these strategies depends on specific element and on plant species (Marschner, 1998). Gahoonia and Nielsen (1996) demonstrated that from plants nutrition point of view, a genotype efficient in P absorption is the one which can both dissolve soil P and absorb it efficiently. Batten (1992) considered selection of efficient varieties as supplementary to, or even as a replacement for, fertilizer application in agriculture. Mahon (1983) suggested that any progress in this field would depend on the progress in our knowledge about plant genetic variability. However, Gahoonia et al., (1994) have reported that besides genetic potential, environmental factors would also be important.

Studying different bread and durum wheat genotypes, Ozturk et al., (2005) found large differences among varieties with respect to P efficiency and stated that P acquisition was the most important factor in P efficiency. They also used the relative shoot dry weight as P efficiency index and showed that under insufficient P conditions, the total plant P and shoot dry weight can be used as reliable indices for evaluation of P efficiency. Gahoonia and Nielsen (1996) showed that winter wheat and spring barley varieties are more P efficient than winter barley, with the differences related to root hair length and root exudates. With respect to P efficiency, when a new wheat variety was compared with a native one, under both sufficient and deficient P conditions the new variety was more efficient based on root morphology, P retranslocation and optimum P utilization (Horst et al., 1993). In Australia, Liao et al., (2005) studied 18 cereal species in two soil types with low and high total P, proposing that total P absorbtion can be used as a criterion in identifying P-efficient species. Similarly, in spring wheat plants, there were significant differences among varieties with respect to P uptake and use efficiency (Gill et al., 2004). Gunes et al., (2006) used 25 winter wheat varieties from Turkey and reported significant differences among them with respect to P efficiency, with durum varieties more P efficient than bread ones. No correlation was found between plants grown in the greenhouse and those grown in the field. Osborne and Rengel (2002b) studied 99 wheat, 8 triticale and 4 rye varieties using four indices of P efficiency [shoot dry weight at deficient P supply, relative shoot dry weight, P uptake efficiency and P utilization efficiency (shoot dry weight per unit of shoot P)]. Under deficient P conditions, rye and triticale genotypes were more efficient (better P uptake and P utilization) than wheat varieties. There were also some differences among wheat varieties.

In Iran, highly calcareous soils are frequently used for wheat production but, despite of relatively high total soil P, crops often suffer from P deficiency. As mentioned above, it is well-documented that plant genotypes differ greatly in adaptation to P deficiency stress. However, there are no published reports on the P efficiency of Iranian cereals. Considering the importance of selecting P-efficient cereal cultivars and making a more efficient use of indigenous soil P, the present study was undertaken to evaluate P efficiency in several Iranian cereal species.

Materials and Methods

Soil

Among 1500 soil samples which had been collected for "Wheat Project" at Soil and Water Research Institute, 8 soil samples with physical and chemical similarities and with low available P (<5 mg P kg^{-1}) were selected. Soils were analyzed for total P by sodium carbonate fusion (Olsen and Sommers, 1982) and Olsen methods (Olsen et al., 1954) (Table 1). Ghazvin 1 with the highest total P was selected for our studies (Table 2).

Table 1. Olsen and total P (mg kg^{-1}) in selected soils.

Soil	Hamedan1	Hamedan 2	Tehran	Ghazvin1	Ghazvin 2	Karaj	Esfahan 1	Esfahan 2
Olsen-P	5	4.2	4.8	4.6	4.3	4.5	2.9	2.4
Total-P	880	720	1125	1260	1257	903	1130	1053

Table 2. Chemical properties of Ghazvin 1 soil.

pH	EC	O.C	Olsen-P	Av-K	Fe	Mn	Zn	Cu	B
	(dS m^{-1})	(g kg^{-1})	←			(mg kg^{-1})			→
7.8	0.95	2.5	4.6	276	1.46	2.80	0.50	0.80	0.40

Plant species

We tested eight bread wheat cultivars (Roshan, Azadi, Qods, Pishtaz, C-81-4, M-81-13, N-81-9, and S-80-18), three durum wheat cultivars (Aria, Yavarus, and D-81-15), three barley cultivars (Karoon, Torshe, and M-80-16), and one cultivar of rye, oat, and triticale. These selected species and cultivars have been produced in Iranian breeding programs and are cultivated throughout Iran.

Greenhouse Experiment

The experiments were factorial in completely randomized blocks with 17 cultivars and two P fertilizer levels (0 and 84 mg P kg^{-1}soil as KH_2PO_4) in three replications. After sieving (2 mm), the soils were placed in plastic pots (10 kg each), amended with basal nutrients and mixed thoroughly. Basal nutrients were (mg kg^{-1} dry soil) 50 N (as urea), 50 K (as K_2SO_4), 30 Mg (as $MgSO_4.7H_2O$), 10 Fe (as sequestrene-138), 15 Zn (as $ZnSO_4$), 2.5 Cu (as $CuSO_4.5H_2O$), 15 Mn (as $MnSO_4.H_2O$), 2.5 B (as H_3BO_3). After 4 weeks of planting, the N fertilizer was re-applied as topdressing. Fourteen seeds were used in each pot and the stands were thinned to 7 after seedlings established.

Measurements

The chlorophyll intensity in flag leaves was evaluated by a chlorophyll meter (Minolta SPAD-502) 47 days after planting. Three readings were taken for each leaf (basal 1/3, middle part and top 1/3). Chlorophyll evaluation was performed for all flag leaves in each pot and the results were expressed as the average for each replication. Eight-week old plants were harvested, dried in 80 ^{o}C and weighed for shoot dry weight (SDW). P concentration (PC) was assayed using the vanadomolybdate method (Westerman, 1990), and total P uptake (TP = SDW * PC) and P efficiency indices were calculated:

P acquisition efficiency (PACE):

$$PACE = \left(\frac{TPinP_0}{TPinP_{84}} \right) \tag{1}$$

P utilization efficiency (PUTE):

$$PUTE = \left(\frac{SDW}{TP} \right) \tag{2}$$

P efficiency (PE):

$$PE = \left(\frac{SDWinP_0}{SDWinP_{84}} \right) \tag{3}$$

Statistical analyses were performed by SAS software and graphs were prepared by Excel.

Results and Discussion

Genotypes showed significant differences in the chlorophyll meter reading, number of tillers, shoot P concentration and content (the total amount of shoot P per pot), and shoot dry weight (Tables 3 and 4).

Chlorophyll meter reading (Chl)

In the P_0 treatment, the chlorophyll meter reading ranged from 36 for Torshe to 48 for Yavarus (Table 4) confirming the difference in color. The average chlorophyll reading for bread wheat, oat and triticale was about 42 and the reading for rye was 39. For all genotypes except Torshe, application of P resulted in an increase in chlorophyll readings of flag leaves (average increased from 42 to 45). It is generally reported that under P deficiency, cell expansion is restricted more than chlorophyll synthesis resulting in more chlorophyll per unit leaf area (Marschner, 1995). However, such a pattern was observed only in Torshe cultivar.

Anthocyanin accumulation (purple color), as one of the visual symptoms of P deficiency in plants, was observed only in the old leaves of M-80-16 barley. This finding is in agreement with reports by Ozturk et al., (2005) and Elliot et al., (1997) who classified plant genotypes by visual symptoms, without finding specific P-deficiency symptoms in wheat leaves.

Number of tillers (Till)

In P_0, the number of tillers was the lowest (3.3) in triticale and the highest in rye (21). In response to P application, the average number increased by 68%; for bread wheat Roshan and M-81-13, the increase was more than 100% (Table 4). Similar results were reported by Horst et al., (1993) who found an increase in number of tillers from 15 to 37 in response to P application at a rate of 60 mg kg^{-1}soil. These results indicate that in addition to nitrogen, P can also be used to increase tillering and increase crop density.

Shoot dry weight (SDW)

Oat species and S-80-18 and Azadi wheat cultivars produced the highest and durum wheat Aria the lowest amount of shoot dry matter (Table 4). In response to P application, the amount of dry matter was increased in all species significantly (averaged from 6.9 to 9.8 g pot^{-1}). Bread wheat M-81-13 durum Aria and barley M-80-16 gave the highest response to P application for the respective species. A similar range in genetic variability of wheat and barley cultivar responses was reported previously (Batten, 1986; Gahoonia et al., 1996; Manske et al., 2000; Osborne and Rengel, 2002a,b; Gill et al., 2004; Lian et al., 2005; Ozturk et al., 2005; Gunes et al., 2006).

P concentration in plants (PC)

On average, shoot P concentration was the lowest (1.2 mg g^{-1} SDW) for oat and the highest (2.6 mg g^{-1} SDW) for Yavarus (Table 4). With no use of P (P_0), the average P concentration in plants was 1.9 mg g^{-1} SDW and was increased to 4.7 mg g^{-1} SDW in response to P application. In control plants (P_0), the amount of P in bread and durum wheat varieties was higher than the amount in barley, rye and oat (2.0 vs <1.5 mg g^{-1} SDW). This indicates that barley, rye and oat can utilize P better with lower P concentrations in their tissues as compared to wheat varieties. However, it indicates that wheat varieties used in this study are more efficient in P absorption. Under optimal conditions, the sufficient P level in plants is about 4.0 mg g^{-1} dry weight (Reuters and Robinson, 1997). In our study, where P was applied, its concentration was increased in all genotypes significantly. This is in agreement with the results reported by Osborne and Rengel (2002a,b), Liao et al., (2005) and Ozturk et al., (2005).

Table 3. Mean squares of chlorophyll meter reading (Chl); number of tillers (Till); shoot dry weight (SDW); P concentration (PC); total P in plants per pot (TP).

Sources	df	MS				
		Chl	Till	SDW	PC	TP
Block	2	7.1^{ns}	2.4^{ns}	1.6^{ns}	0.01^{ns}	30^{ns}
Genotype	16	80^{**}	201^{**}	4.4^{**}	0.1^{**}	71^{**}
P	1	204^{**}	293^{**}	213^{**}	20^{**}	$27{,}520^{**}$
Genotype*P	16	8.1^{**}	13.4^{**}	3.1^{**}	0.04^{**}	56^{**}
Error	66	3.4	2.6	1.0	0.01	29

**:Significant at P=0.01; ns: not significant

Total P content (TP) and P acquisition efficiency (PACE)

With no P application (P_0), the total P content was highest in Yavarus (durum wheat) and Azadi (bread wheat) and lowest in barley M-80-16. With no addition of P (P_0), the average total P content was 13.2 mg pot^{-1} and was increased to 46 mg pot^{-1} in response to P application (Table 4).

Plant genetic potential plays an important role in determining total P uptake at any level of P supply, so in order to exclude this factor the relative total P uptake (treatment P_0/treatment P_{84}) was used to estimate P acquisition efficiency (PACE). The average PACE for all genotypes used in our study was 0.29 mg g^{-1} SDW. The values for all bread wheats and durum wheat Yavarus were higher than those for barley, oat, rye and triticale. Hence, non-bread species tested produced high dry weight but absorbed less P, suggesting they are more efficient in P utilization than P acquisition. Wheat cultivars Azadi and Yavarus were ranked more efficient and barley M-80-16 was ranked inefficient in P acquisition compared with other genotypes (Table 5). In similar studies, Batten (1986) using 23 wheat genotypes and Osborne and Rengel (2002a) using 106 cereal genotypes also found large genotypic differences in P content. The differences were attributed to root size, root morphology and changes in the rhizosphere. Manske et al., (2000) suggested that P acquisition efficiency is more important than P utilization efficiency. However, Ozturk et al., (2005) stated that the two types of P efficiency are equally important. Gahoonia et al., (1996) reported that the differences in P efficiency among wheat and barley cultivars are due to their ability in P absorption. Fageria and Baligar (1997, 1999) used total P absorption and shoot dry weight as two important criteria in screening wheat and rice cultivars. According to these studies it is evident that there are wide differences among cereal species with respect to P acquisition efficiency. Hence, it is doubtful that a uniform P fertilizer program based on soil test will be appropriate for all cereal species.

Table 4. Effects of P application on chlorophyll meter reading (Chl), number of tiller (Till), shoot dry weight (SDW), P concentration (PC), and on total P in plants per pot (TP).

Genotype	Chl		Till		SDW (g pot^{-1})		PC (mg g^{-1}DW)		TP (mg pot^{-1})	
	P_0	P_{84}	P_0	P_{84}	P_0	P_{84}	P_0	P_{84}	P_0	P_{84}
Roshan	43.10	45.53	5.33	13.00	7.53	9.21	2.1	5.5	15.75	51.08
Azadi	47.36	50.90	3.66	4.66	8.03	8.28	2.1	4.8	17.22	39.63
Qods	46.76	48.26	3.66	7.00	6.96	8.35	1.8	4.1	12.41	34.37
Pishtaz	41.66	45.33	5.00	5.33	7.48	10.76	2.1	4.8	16.18	51.65
C-81-4	38.73	41.70	8.33	12.33	7.19	10.31	2.1	4.5	15.09	46.18
M-81-13	42.30	44.36	3.00	9.00	5.93	10.47	2.1	4.4	12.90	46.43
N-81-9	42.50	43.26	3.33	4.00	6.40	10.47	2.2	4.4	14.05	46.90
S-80-18	42.93	48.13	6.33	7.33	8.14	10.24	2.0	4.4	16.25	45.12
Aria	43.56	43.90	3.33	5.33	4.13	7.65	2.2	4.6	9.33	35.97
D-81-15	38.90	44.70	3.33	5.66	6.65	9.51	2.0	5.2	13.74	49.40
Yavarus	48.03	52.56	4.00	6.33	7.62	9.60	2.6	4.9	20.15	47.01
Karoon	42.06	47.80	5.33	6.33	6.59	11.38	1.4	4.2	9.46	47.74
M-80-16	40.40	44.93	4.00	12.00	4.24	10.17	1.4	4.7	5.85	47.84
Torshe	36.03	33.33	15.33	19.33	6.88	8.90	1.8	5.9	12.88	53.03
Rye	38.90	40.03	21.00	31.33	7.38	10.94	1.5	4.8	11.62	53.69
Oat	43.73	48.60	6.66	10.00	8.39	10.14	1.2	4.1	10.35	41.94
Triticale	41.36	43.56	3.33	3.66	7.44	9.70	1.6	4.5	11.88	44.62
Mean	42.28	45.11	6.17	9.56	6.88	9.77	1.9	4.7	13.24	46.09
LSD(for Genotype)	3.08		2.60		1.61		0.30		3.88	
LSD(for P)	0.72		0.63		0.39		0.1		2.1	
LSD(for Gen*P)	2.12		1.86		1.14		0.4		6.22	
CV	4.2		20.5		11.9		10.9		18.2	

P utilization efficiency (PUTE)

PUTE represents the amount of dry matter produced per unit of P absorbed [Eq. (2)]. Any species able to maintain metabolic activities at low tissue P concentration and produce more dry matter per unit of P absorbed is considered efficient in P utilization. Among species used in our study, this index ranged from 0.38 (for Yavarus) to 0.81 g SDW mg^{-1} P (for oat). Barley, rye, oat and triticale were more efficient than bread and durum wheats (Table 5). In all species PUTE decreased with P application, suggesting that under P limitation alternative pathways develop in plants to maximize the use of limited available P. Fageria et al., (1988) reported that with the increase in P application, utilization efficiency in rice cultivars reduced significantly. In another study Fageria and Baligar (1999) found that new wheat cultivars had higher P utilization efficiency than older ones. Horst et al.,

(1993) found that wheat cultivars whose productivity is based on the number of seeds per spike suffered little under P limitations. Osborne and Rengel (2002b) found wide variation in P utilization among genotypes of wheat, rye and triticale.

Table 5 Efficiency indices in different cereal genotypes.

Genotype	PACE	PUTE (g SDW/mg P)		PE
		P_0	P_{84}	
Roshan	0.31	0.47	0.18	0.82
Azadi	0.45	0.46	0.21	0.97
Qods	0.36	0.56	0.24	0.83
Pishtaz	0.31	0.46	0.20	0.70
C-81-4	0.32	0.47	0.22	0.70
M-81-13	0.29	0.46	0.23	0.58
N-81-9	0.30	0.46	0.22	0.63
S-80-18	0.36	0.50	0.23	0.80
Aria	0.26	0.44	0.21	0.54
D-81-15	0.28	0.48	0.19	0.70
Yavarus	0.43	0.38	0.20	0.79
Karoon	0.20	0.76	0.24	0.58
M-80-16	0.12	0.73	0.21	0.42
Torshe	0.24	0.54	0.17	0.77
Rye	0.23	0.66	0.20	0.68
Oat	0.25	0.81	0.24	0.83
Triticale	0.28	0.63	0.22	0.77
Mean	0.30	0.55	0.21	0.71
LSD	0.12	0.07		0.20
CV	25.7	17.3		17.2

PACE: P acquisition efficiency; PUTE: P utilization efficiency; PE: P efficiency.

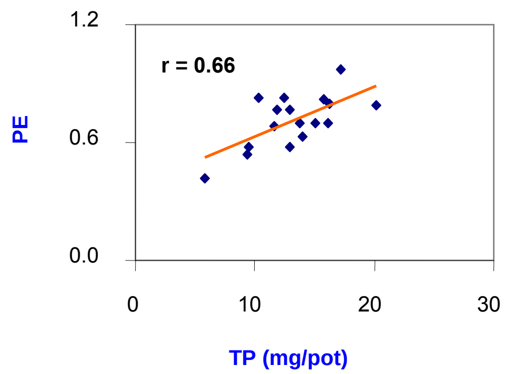

Figure 1. Correlations between P efficiency (PE) and total P content in plants (TP).

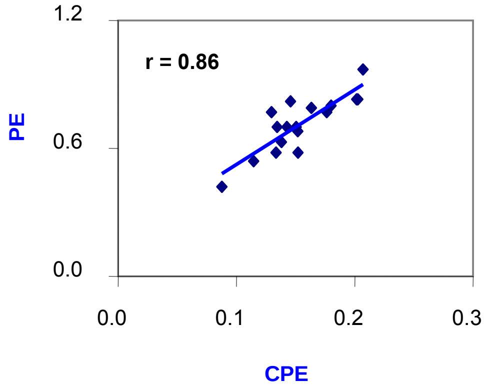

Figure 2. Correlations between calculated P efficiency (CPE) and experimentally determined P efficiency (PE).

P efficiency (PE)

Researchers used different indices to estimate P efficiency, e.g. absolute yield and the amount of P absorbed under P-limited conditions, relative shoot dry weight, P acquisition and utilization efficiency, rate of P absorbed per unit of root weight or root length, etc. (Gahoonia and Nielsen, 1996; Fageria and Baligar, 1999; Rengel, 1999; Monasterio et al., 2002; Osborne and Rengel, 2002a,b). Considering all theses variations in terminologies and methodologies, it is hard to find a general consensus on P efficiency definition. However, the prevailing opinion is using the ratio of yield under P-limited over that under P-sufficient conditions (Rengel, 1999), which takes into account both the P acquisition and P utilization efficiencies. Hence, in our study, the relative shoot dry weight (RSDW) was adopted as P efficiency index (PE) [Eq. (3)].

Using the above criteria, barley M-80-16 with 0.42 had the lowest and bread wheat Azadi with 0.97 the highest P efficiency indices. Other species, such as Qods, Roshan, S-80-18, and oat were placed in the same statistical group as Azadi cultivar, i.e. P-efficient (Table 5). There was a significant positive correlation ($r = 0.66^{**}$) between P efficiency index and total P content in plants per pot (Figure 1). Similar findings have been reported by Ozturk et al., (2005) with a positive correlation of r=0.81. However, such a correlation was weak with respect to P concentration, which is in agreement with studies of Jones et al., (1992) and Baligar (1999). In most studies good correlations have been found between total P absorbed and P efficiency (Jones et al., 1992; Gill el al., 1994; Fageria and Baligar, 1997; Baligar, 1999).

P efficiency index (relative shoot dry weight) and the one calculated by Monasterio et al., (2001) used the products of P acquisition efficiency (PACE) by P utilization efficiency (PUTE) under deficient P (CPE = PACE x PUTE; where CPE: calculated P efficiency). Relationship between P efficiency and CPE showed a significant ($p< 0.01$) positive correlation (r=0.86) (Figure 3). Such a high positive correlation indicates that the relative shoot yield includes both P acquisition and P utilization indices; thus, it can be an appropriate index for P efficiency evaluation, which is also in agreement with the findings of Ozturk et al., (2005) and Liao et al., (2005). Nonetheless, with this index it is possible that a species that produces low absolute shoot yield under P-limited condition (P_0) and does not respond to P application be classified as P efficient. In any case, in order to prevent such species to be upgraded, perhaps it would be better to pay attention to absolute productivity under P-limited condition (P_0) conditions as it has also been suggested by Gerloff (1978) and Lynch (1998) who has proposed screening under P-limited condition for low input systems and screening under sufficient P for high input ones. They have used the term “fertilizer responder” for the latter group of plants. Therefore, on the basis of absolute index, one can consider species like wheat (Azadi and S-80-18) and oat as P efficient are suitable for low input systems, and genotypes of wheat (Pishtaz, C-81-4, M-81-13, N-81-9 and S-80-18), barley (Karoon and M-80-16) along with oat and rye as “fertilizer responder” are suitable for high input systems. It is obvious that both wheat S-80-18 and oat can be grown under the two systems (Table 4). Therefore, by changing the indices, the ranking of cultivars and varieties in screening will also change. These findings are also supported by those reported by Osborne and Rengel (2002b) who found that among 111 cereal genotypes, none was superior in all efficiency indices. Therefore, depending on the goals the screening processes of cultivars and varieties could well be different.

Conclusions

As it is expected, with respect to P acquisition and utilization, a great deal of variations exists among different cereals species. It seems that wheat genotypes (bread and durum) tested in present study had higher shoot P concentration and content than other cereals (oat, barley, rye, and triticale), suggesting that P acquisition ability should be the most important mechanisms for high P efficiency in such genotypes. In contrast with wheat, other tested cereals (oat, barley, rye, and triticale) produce high biomass at low shoot P concentration, indicating importance of better P utilization in these varieties. A general conclusion can be reached when much more accessions are tested and further research is needed to reveal P acquisition (root architecture, root exudations, and mycorrhizal association) and utilization efficiency mechanisms (internal mechanisms associated with better use of absorbed P at cellular level) in cereals. It is hoped that in the future studies on finding efficient cereal cultivars and varieties, the details of efficiency mechanisms and gene responsible for coding the enzymes involved be worked out. By using gene transfer techniques more efficient plants can be produced and new steps will be taken towards the management of P fertilizer application. By reducing both the amount of P fertilizers used and the costs involved, a positive approach will be taken towards both sustainable agriculture and environmental protection. Perhaps for humanity to cope with population explosion dilemma on one hand, reducing both fertilizer consumption and their costs on the other, another green revolution can be achieved. Of course, this time, not in the direction of producing more by using more fertilizers but in the direction of producing more with least amount of fertilizer.

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