

DIFFERENTIAL GROWTH AND PHOSPHORUS UPTAKE BY WHEAT CULTIVARS AT DIFFERENT P LEVELS

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ABSTRACT

A solution culture study was planned to evaluate genetic variations among ten advance wheat genotypes. Wheat genotypes differed significantly ($P < 0.05$) in shoot and root dry matter production, root: shoot ratio, P uptake and shoot P utilization efficiency. Shoot P uptake was strongly correlated ($R^2 = 0.85$, $R^2 = 0.87$) with shoot and root dry matter production. Shoot dry yield plant⁻¹ varied 1.00 to 2.77 (g) at 25 μM (deficient) while 1.20 to 3.27 (g) at 250 μM (adequate) P levels, respectively. Genotype NRL-1120 produced highest shoot dry matter (SDM) at deficient P level while genotype Yecora produced the highest SDM at adequate P level. Unlike SDM, there was 13% reduction in root dry matter (RDM) with the increase in P levels from 25 to 250 μM in the growth medium. More or less all the wheat genotypes produced similar RDM at adequate & deficient P levels. Among ten wheat genotypes NRL-1105, NRL-1120 and Yecora are the P-efficient wheat genotypes while NRL-1101, NRL-1129 and WL-711 are the P- inefficient wheat genotypes.

Key words: Hydroponic, genotypic difference, phosphorus, phosphorous use efficiency & wheat

INTRODUCTION

Cereals make up around 50% of global food production at present (FAO, 2013). Wheat alone accounts for one fifth of human's food, providing 21% of the calories and 20% of the protein to more than 4.5 billion people in 94 developing countries (Braun *et al.*, 2010). Wheat (*Triticum aestivum* L.) is a staple food in Pakistan and provides 37% of the total calories intake in Pakistan (FAO, 2013). It contributed 2.1% of the country's GDP and 10% of the value added in agriculture. In Pakistan, wheat is cultivated on 9.26 million ha with an average yield of 2752 kg ha⁻¹ (Anonymous, 2015-2016). The average wheat yield in Pakistan is lower than other wheat producing countries due to widespread nutrient deficiency (Rashid and Awan, 2002).

Phosphorus deficiency is a common nutritional problem affecting crop production globally including Pakistan (Fairhurst *et al.*, 1999). Billions hectares cultivated soils worldwide ($> 30\%$ of world's arable land) are deficient in available P and could not sustain adequate plant growth. Low use efficiency of applied P (15-20%) in soils makes P fertilization not only uneconomical but also environmentally unsafe practice (Vance *et al.*, 2003). The ever rising prices of P fertilizers in the world market besides increasing concern of environmental degradation (Vance *et al.*, 2003) call for multidimensional solutions to tackle the problem, instead of relying upon conventionally available high input approaches. The P balance of Pakistani soils is negative (5-10 kg ha⁻¹ yr⁻¹) (Ahmad and Rashid, 2003). Shah *et al.* (2016) reported that more than 90% soils in Pakistan are considered deficient (contain < 10 mg kg⁻¹ P) in available-P. Furthermore, the world reserves of inexpensive P (rock phosphate; mined for production of P fertilizer) will be depleted in the very near future (Herring and Fantel, 2003; Steen, 1998). It is of primary importance to screen out crop genotypes that can grow well in soils having low content of available phosphorous. A few solution culture studies revealed genotypic variability for P absorption and utilization in chickpea, cotton and wheat (Alam *et al.*, 2003; Richardson *et al.*, 2011). Little efforts have been made to mark a distinct "P-efficient" or "P-inefficient" cultivar, most probably due to difficulties in defining "efficiency", that varies due to the definition used (Gourley *et al.*, 1994).

Several studies have employed hydroponics system with different supply of P to select P-efficient cultivars or examine the morphological and physiological responses (Wang *et al.*, 2010). This study is conducted to compare differential response in P use efficiency of selected wheat genotypes grown in hydroponic system, in attempt to understand underlying mechanism of P efficiency of wheat.

MATERIALS AND METHODS

A hydroponic experiment was carried out to study the effect of P deficient and adequate levels on the growth of wheat. The seeds of ten wheat genotypes (including checks Yecora = P efficient and WL 711= in-

efficient) were surface sterilized with sodium hypochlorite and germinated on moist filter papers in Petri dishes in an incubator at 20 ± 1 °C until ready for transplanting. Five days old seedlings were transplanted in foam plugged holes of thermopole sheet floating on a continuously aerated modified Johnson nutrient solution (Johnson *et al.*, 1957) contained in two stainless steel tubs of 20 L capacity. Two seedlings were transplanted in one hole and each hole was considered as one repeat. Treatments were arranged according to completely randomized factorial design with three replications. Two phosphorus levels were established by using ammonium phosphate ($\text{NH}_4\text{H}_2\text{PO}_4$) salt; adequate ($250 \mu\text{M}$) and deficient ($25 \mu\text{M}$) P levels. The pH of the solution was maintained at 5.5 ± 0.5 with HCl or NaOH (Table 1). The plants were initially grown in nutrient solutions containing half strengths of all macro and micronutrients until day 10 after which the full-strength solutions were used. The nutrient solutions were replaced with fresh mixtures on days 10, 15, 19, 24, 28 and 32 following transplantation. Harvesting of the plants was carried out on day 35 after transplantation. The tissue samples were then air dried on blotting paper and later dried in a forced draft oven at 70 ± 1 °C for 48 hours (until constant weight). Dried samples of shoots and roots were ground in a mechanical grinder (MF 10 IKA, Werke, Germany) to pass through a 1 mm sieve. A 0.5 g portion of uniformly mixed plant sample was digested in diacid mixture of nitric acid and perchloric acid (3:1) at 150°C (Miller, 1998). Phosphorus concentration in shoot and root digest was estimated using vanadate-molybdate colorimetric method (Chapman and Pratt, 1961). Phosphorus contents (mg P plant^{-1}) were calculated in root and shoot by multiplying P concentration in the respective tissue with its dry matter and on whole plant basis for shoot and root P uptake. Phosphorus utilization efficiency ($\text{g}^2 \text{SDM mg}^{-1} \text{P}$) was calculated by the following formula (Siddiqui and Glass, 1981).

$$\text{Phosphorus Utilization Efficiency} = \frac{1}{\text{Shoot P concentration}} \times \text{Dry matter}$$

$$\text{P – Stress factor (\%PSF)} = \frac{(\text{SDM (adequate/sufficient P)} - \text{SDM (deficient/stress P)})}{\text{SDM (adequate P)}} \times 100$$

The data were subjected to statistical treatments using computer software “MS Excel” and “MSTAT-C” (Russell and Eisensmith, 1983). Completely randomized factorial design was employed for analysis of variance (ANOVA). Least square method of regression / linear correlation was used to calculate regression and correlation coefficients among different parameters. The differences among the treatment means were compared by using LSD test (Steel *et al.*, 1997).

RESULTS AND DISCUSSION

A hydroponic experiment was carried out to study the severity of phosphorus (P) deficiency symptoms on leaves, shoot and root dry matter production, and shoot and root concentration of P in 10 wheat (*Triticum aestivum* L.) genotypes. The results indicated that at low P level, first visual symptom of P deficiency appeared as development of dark-green color in leaves and then reduction in shoot elongation and leaf size of most genotypes. As the P deficiency stress progressed, the oldest leaves became chlorotic and showed desiccation starting from the leaf tips. Severity and development time of these leaf symptoms greatly differed among wheat genotypes.

Shoot and Root Biomass: Growth behavior of the ten wheat genotypes was remarkably different at the deficient and adequate levels of P supply in the growth medium (Fig. 1-8). It was revealed by significant interactive effect ($P < 0.05$) of P rates and genotypes on shoot dry matter (SDM), root dry matter (RDM) and root: shoot ratio (RSR) (Table 3). Shoot dry matter of wheat genotypes decreased 19 % when P supply was reduced from $250 \mu\text{M}$ to $25 \mu\text{M}$, in the growth medium. In deficient P supply, NRL 1120, NRL 1105 and Yecora produced higher SDM while minimum production was observed in NRL 1119 followed by NRL 1124. At deficient P supply, reduced SDM is attributed to enhanced ability of plants to invest in root biomass which jeopardizes yield potential of above ground portions (Gill *et al.*, 2002). The maximum reduction in SDM production due to P deficiency was observed in NRL 1101 and WL 711 while minimum reduction was observed by NRL 1120 and NRL 1105. Such interactions are important for crop cultivar development (Kang, 1998). Genotypic differences for SDM under differential P levels were also reported by Gill *et al.*, (1994) and Yaseen *et al.*, (2008). The data regarding root dry mater (Table 2, 3) showed significant difference among the wheat genotypes, the maximum root dry matter was recorded 1.09, 1.06 and $0.98 \text{ g plant}^{-1}$ in NRL 1120, NRL 1105 and Yecora, while the minimum was observed NRL-1124 and NRL-1101 in deficient P supply. Root dry matter (RDM) decreases 13% with the increase in P level from 25 to $250 \mu\text{M}$ in the growth medium. All the wheat genotypes produced statistically non significant RDM at adequate and deficient

P levels. This increase in RDM was possibly due to stimulation effect of P deficiency on root growth under P stress conditions (Haynes *et al.*, 1991; Gill *et al.*, 1994; Yaseen *et al.*, 1998, 2004). Increase in root biomass production under P stress was also reported by many workers they reported increase in root biomass due to more translocation of photosynthates from shoot to root under P stress which probably can help plant to absorb more P under limited supply in the growth medium. Likely NRL-1101 and NRL-1124 has maximum root shoot ratio at deficient P supply while NRL-1120 had minimum at both P levels. Shoot dry matter was strongly negative correlated ($R^2 = -0.909$) with root: shoot ratio (Fig. 4).

Table1.The composition of the Long Ashton complete nutrient Solution used in this study (Johnson *et al.*, 1957).

Reagents	Stock Sol. molarity	μM	mL/ 20litre
KH_2PO_4	1	25& 250	2 & 20 ml of mM
KNO_3	1	1500	30
$\text{Ca}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$	1	2000	40
MgSO_4	1	500	10
H_2BO_3	0.1	10	2
Na_2MoO_4	0.001	0.1	2
K_2HEDTA^*	0.1	25	5
Zn HEDTA	0.1	2	0.2
Mn HEDTA	0.01	1	2
Cu HEDTA	0.001	0.5	10
Fe HEDTA	0.1	100	20

* Hydroxy Ethylene Diaminetriacetic Acid (HEDTA)

Table 2.Growth performance of wheat genotypes grown at adequate and deficient levels of P supply.

Treatments	Plant height (cm)		Tillers (NO)		Root length (cm)		Fresh Shoot Yield (g plant ⁻¹)	
	Deficient P supply	Adequate P supply	Deficient P supply	Adequate P supply	Deficient P supply	Adequate P supply	Deficient P supply	Adequate P supply
NRL-1101	15.77 ij	21.30 f	3.00 fg	5.33 a-e	81 a	57 ef	10.77 i	21.30 d
NRL-1105	25.83 abc	27.20 a	6.67 abc	7.00 ab	70 bc	65 cd	25.83 ab	27.20 a
NRL-1117	18.47 gh	24.57 bcd	3.67 efg	6.00 a-d	69 bc	53 fg	18.47 ef	24.57 bc
NRL-1119	18.37 gh	25.17 bc	3.33 efg	4.00 d-g	71 b	52 fg	18.37 ef	25.17 b
NRL-1120	25.23 bc	27.50 a	4.67 c-g	6.00 a-d	71 b	62 de	25.23 b	27.50 a
NRL-1123	17.00 hi	24.37 cde	3.33 egf	4.67 c-g	65 cd	51 g	17.00 fg	24.37 bc
NRL-1124	15.10 j	19.13 g	2.67 g	4.00 d-g	67 cd	56 fg	15.10 gh	19.13 e
NRL-1129	14.90 j	22.67 ef	2.67 g	4.67 c-g	78 a	57 ef	11.90 i	22.67 cd
WL-711	14.40 j	23.20 de	2.33 g	5.00 b-f	80 a	56 fg	14.40 h	23.20 cd
Yecora	24.50 bcd	26.27 ab	6.67 abc	7.33 a	70 bc	65 cd	24.50 bc	26.27 ab
Mean	18.96 B	24.14 A	3.93 B	5.40 A	72.27 A	57.40 B	18.16 B	24.14 A
LSD Value	1.821		2.098		5.594		1.93	

Means followed by different letters in the same column are significantly different from each other at 5% level of significance.

Phosphorous uptake and Utilization efficiency: Genotypes showed marked differences in P uptake at deficient and adequate levels of P. All genotypes differed significantly ($P < 0.05$) for their shoot P uptake at deficient P level (Table 4). NRL 1120, Yecora and NRL 1105 had a maximum 2.54, 2.40 and 2.30 mg plant⁻¹ P uptake recorded while minimum 0.52, 0.76 and 0.92 mg plant⁻¹ was observed in NRL 1101, WL 711 and NRL 1129. In case of root P uptake NRL 1120, Yecora and NRL 1105 had the highest 10.69, 9.32 and 8.68 mg plant⁻¹ P uptake at deficient P

level while the lowest 2.58, 2.75 and 3.47 mg plant⁻¹ was recorded in NRL 1124, NRL 1101 and NRL 1129. This suggested that greater P uptake was associated with genotypes having larger root system. The importance of a larger root system become even greater in soil grown plants, where P supply is diffusion limited (Gill *et al.*, 2002; Kosar *et al.*, 2002 and 2003).

Table 3. Growth performance of wheat genotypes in terms of dry matter of shoot and root grown at adequate and deficient levels of P supply.

Treatments	Shoot Dry Matter (g plant ⁻¹)		Root Dry Matter (g plant ⁻¹)		Dry Root shoot ratio	
	Deficient P supply	Adequate P supply	Deficient P supply	Adequate P supply	Deficient P supply	Adequate P supply
NRL-1101	1.13 jk	2.50 de	0.68 j	1.02 c-f	0.60 ab	0.41 efg
NRL-1105	2.67 bcd	2.70 bc	1.06 bcd	1.07 bc	0.40 fg	0.40 fg
NRL-1117	2.20 g	2.40 ef	0.95 g	1.00 efg	0.43 def	0.42 efg
NRL-1119	2.30 fg	2.40 ef	0.96 g	1.01 def	0.42 efg	0.42 efg
NRL-1120	2.77 b	2.80 b	1.09 b	1.10 b	0.39 fg	0.39 fg
NRL-1123	1.40 i	1.80 h	0.75 i	0.85 h	0.54 c	0.47 d
NRL-1124	1.00 k	1.20 j	0.65 j	0.70 j	0.65 a	0.58 bc
NRL-1129	1.77 h	2.57 cd	0.84 h	1.05 bcd	0.47 d	0.41 efg
WL-711	1.93 h	2.60 cd	0.88 h	1.05 bcd	0.46 de	0.40 fg
Yecora	2.37 efg	3.27 a	0.98 fg	1.22 a	0.41 efg	0.40 fg
Mean	1.96 B	2.43 A	0.88 B	1.01 A	0.48 A	0.43 B
LSD Value	0.165		0.05218		0.05218	

Means followed by different letters in the same column are significantly different from each other at 5% level of significance.

Table 4. Phosphorus uptake, P utilization efficiency and P Stress factor of wheat genotypes grown at adequate and deficient P levels.

Treatments	P uptake Shoot (mg plant ⁻¹)		P uptake Root (mg plant ⁻¹)		PUE (mg SDM g ⁻¹ P)		PSF (%)
	Deficient P supply	Adequate P supply	Deficient P supply	Adequate P supply	Deficient P supply	Adequate P supply	
NRL-1101	0.52 j	3.75 e	2.75 l	9.10 de	0.31 h	0.69 c	49.45
NRL-1105	2.30 g	10.21 a	9.32 d	12.36 ab	0.58 fg	0.59 efg	5.02
NRL-1117	1.54 h	4.82 d	6.40 hi	9.05 de	0.55 g	0.60 efg	24.83
NRL-1119	1.38 hi	5.03 d	7.51 gh	8.75 def	0.63 def	0.66 cd	27.02
NRL-1120	2.54 fg	10.05 a	10.69 c	11.31 bc	0.69 c	0.70 c	8.24
NRL-1123	1.18 hij	6.25 c	4.16 jk	7.96 efg	0.24 i	0.31 h	30.23
NRL-1124	1.39 hi	7.27 b	2.58 l	5.25 ij	0.11 j	0.13 j	21.08
NRL-1129	0.92 hij	3.34 e	3.47 kl	8.33 d-g	0.55 g	0.80 b	47.50
WL-711	0.76 ij	3.18 ef	4.44 jk	7.79 fg	0.64 cde	0.87 a	37.93
Yecora	2.40 g	9.63 a	8.68 d-g	12.63 a	0.61 def	0.85 ab	6.73
	1.49 B	6.35 A	6.00 B	9.25 A	0.49 B	0.62 A	
LSD value	0.7268		1.24		0.0521		

Means followed by different letters in the same column are significantly different from each other at 5% level of significance.

It is interesting to note that P uptake in shoots at deficient P level differed significantly. This indicates that wheat genotypes which accumulated more P in their shoots from a deficient growth medium were more tolerant to P deficiency stress. Haynes *et al.*, (1991) also observed similar responses. This differential P uptake had close link with differences in P uptake of shoot and roots which were mainly associated with the differences in root P concentration ($R^2=0.808$ and $R^2=0.378$). Fig .8 and 3, respectively, in the genotypes grown at deficient P level (data

is not given) probably because the magnitude of differences in P concentration were much wider compared to differences in their root dry matter. Schenk and Barber (1979) and Coltman *et al.* (1985) reported that differences between genotypes in P absorption were due to morphological and physiological root characteristics. Blume (1988) has already reported similar results. It increased significantly with decreasing P supply in the root medium, which can be attributed to translocation of photosynthates from shoot to root. Similarly increased P uptake with decreasing P supply was reported by (Gaume *et al.*, 2001; Gill *et al.*, 2002). The differences for phosphorus utilization efficiency (PUE) and phosphorus stress factor (PSF) (Table 4). Shoot dry matter yield was correlated ($R^2 = 0.40$) with shoot P utilization efficiency (Fig. 1) were also observed among these genotypes. PUE significantly increased with increasing P supply when P supply was raised from 25 to 250 μM in the growth medium. The greater reduction in PUE due to P deficiency was observed in NRL-1101 while, lowest was observed in NRL-1105 and NRL-1120. Data regarding phosphorus stress factor, minimum PSF was recorded in NRL 1105, Yecora and NRL-1120 while maximum was observed in NRL 1101, NRL 1129 and W1-711. Four out of ten genotypes depicted in (Table 4) PSF higher than 30% and the rest more than 30%. Only three genotypes NRL 1105, Yecora and NRL-1120 showed PSF $< 10\%$. The results of this study provide useful information about the genetic differences in wheat with regards to P relation. Therefore, involvement of genotypes with better P absorption and utilization mechanism in the breeding programme could be useful to evolve wheat genotypes that can perform better on P deficient soils.

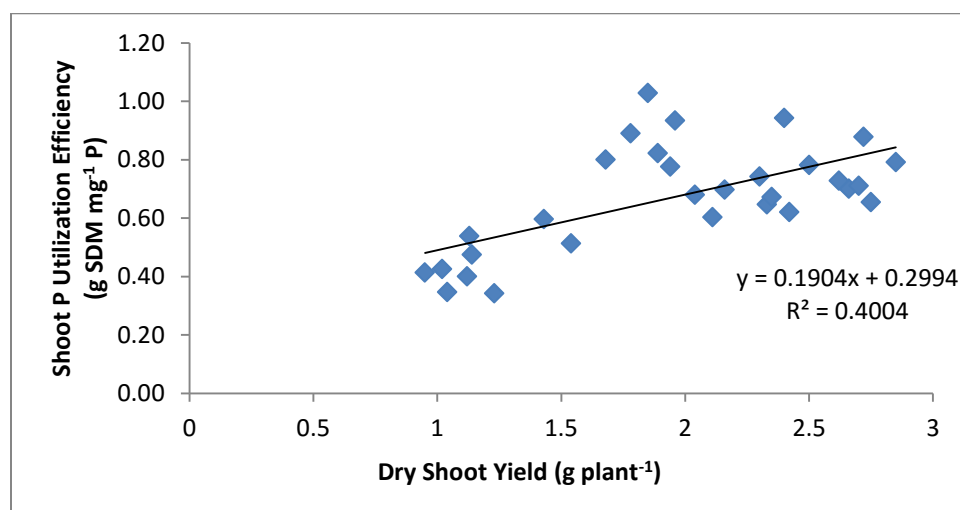


Fig. 1. Relationship between Dry shoot yields and shoot P utilization efficiency of wheat genotypes at deficient P supply in culture solution.

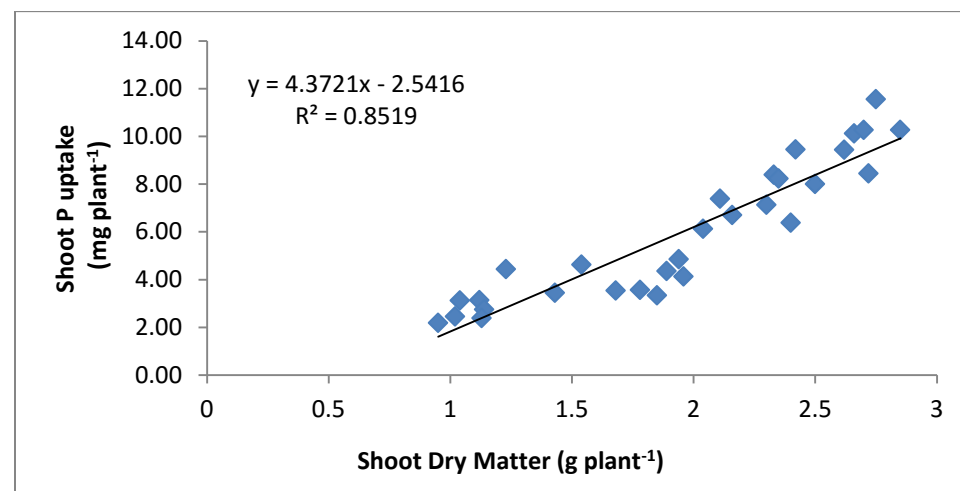


Fig. 2. Relationship between Dry shoot yields and shoot P uptake of wheat genotypes at deficient P supply in culture solution.

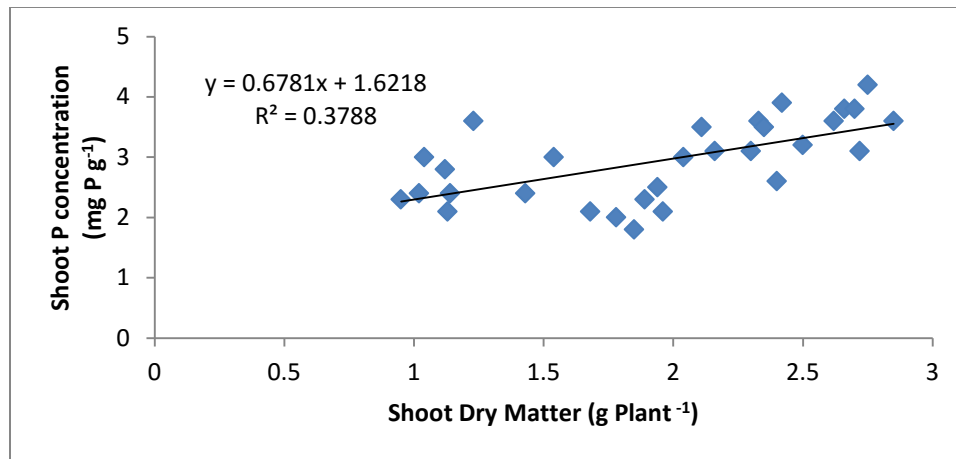


Fig. 3. Relationship between Dry shoot yield and shoot P concentration of wheat genotypes at deficient P supply in culture solution.

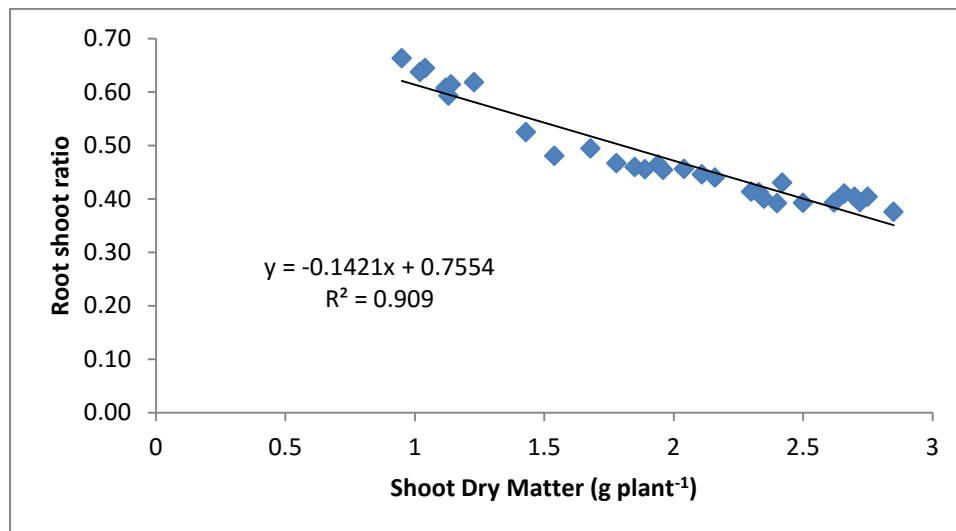


Fig. 4. Relationship between Dry shoot yields and root shoot ratio of wheat genotypes at deficient P supply in culture solution.

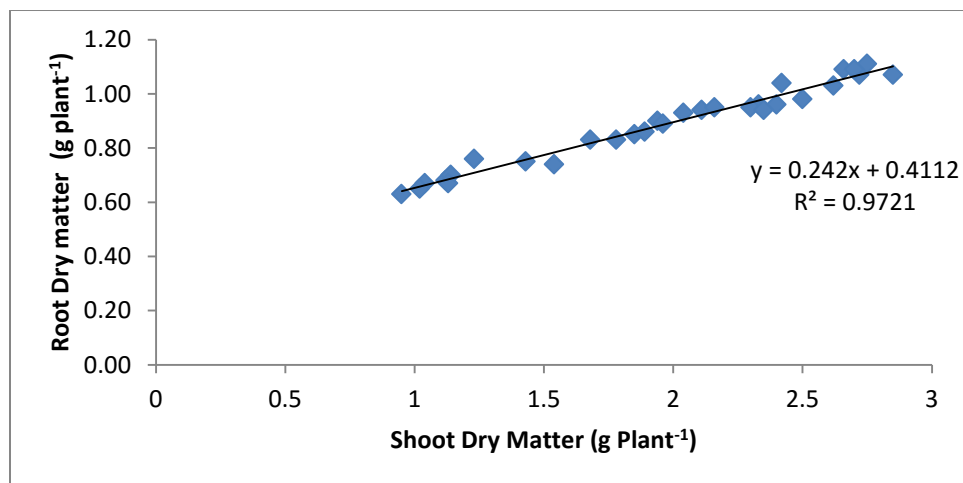


Fig. 1. Relationship between Dry shoot yields and root dry matter of wheat genotype at deficient P supply in culture solution

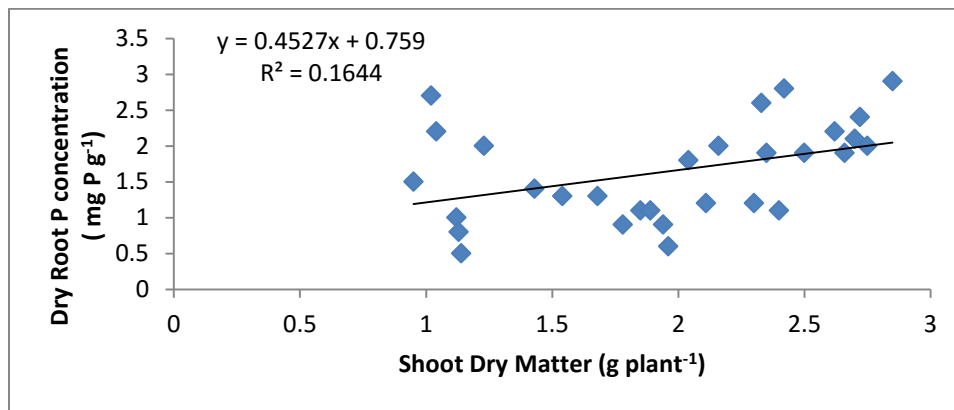


Fig. 6. Relationship between Dry shoot yields and dry root P concentration of wheat genotypes at deficient P supply in culture solution.

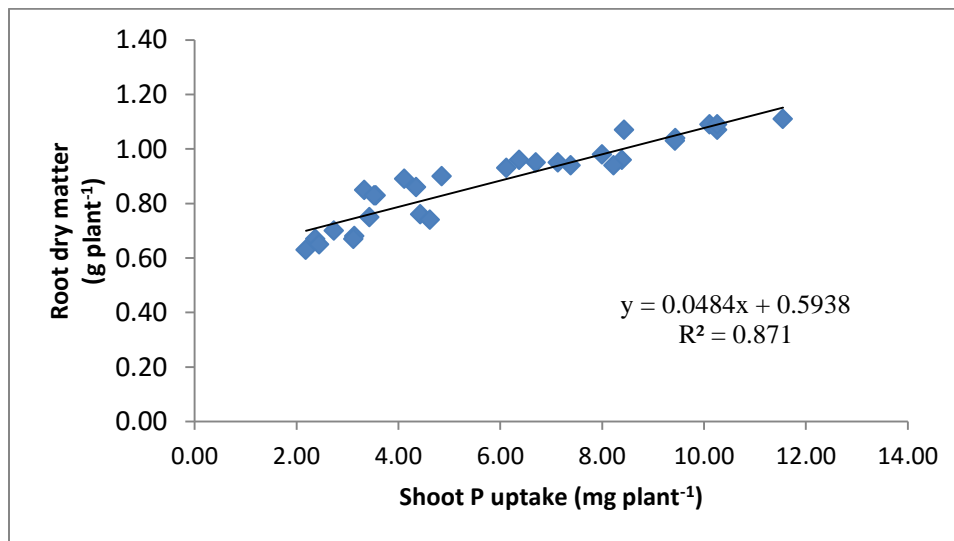


Fig. 2. Relationship between Dry root yields and shoot P uptake of wheat genotype at deficient P supply in culture solution.

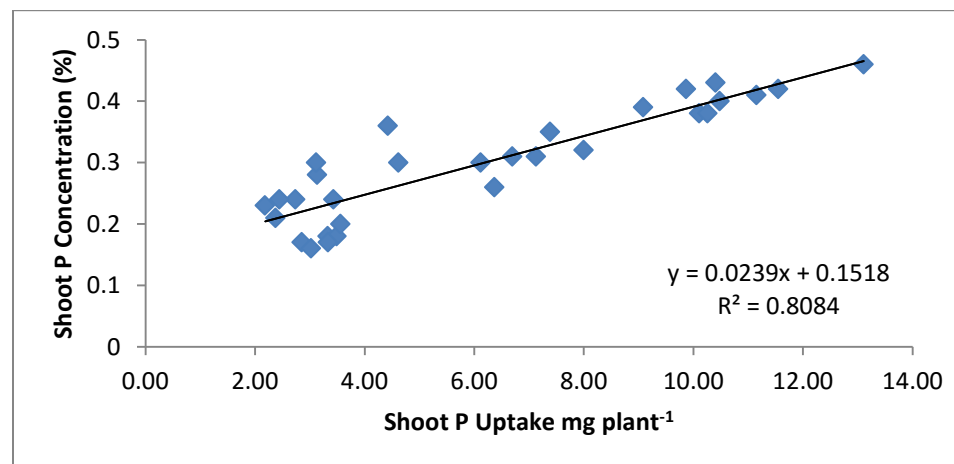


Fig. 8. Relationship between shoot P uptake and shoot P concentration of wheat genotypes at deficient P supply in culture solution.

Conclusion

Positive genotypic variations existed among wheat genotypes for P acquisition (differences in P uptake) and its utilization to produce biomass (use efficiency). Cultivars efficient in P acquisition and utilization (NRL 1105, Yecora and NRL-1120) produced more growth than inefficient cultivars (NRL 1101, NRL 1129 and WL-711). Efficient cultivars translocated more P from roots toward shoot, while inefficient cultivars retained more proportion of total P in their roots. Efficient P acquisition and utilization from roots toward shoot was the major adaptive mechanism identified in P efficient cultivars which must be studied further under field conditions.

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