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## IDENTIFICATION OF NITROGEN-USE-EFFICIENT AND DEEP ROOTED WHEAT GENOTYPES

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### ABSTRACT

Global food security is at high risk due to climate change. Wheat being the leading cereal of Pakistan finds a pivotal position in food security. Adaptation strategies to combat the adverse effects of climate change must focus on identification and use of genotypes that are efficient in the use of available nitrogen. In a sand culture study, fifteen wheat genotypes were compared under non-stressed (irrigated) conditions to identify nitrogen efficient genotypes having deep root system. Significant ( $P \leq 0.05$ ) differences were observed amongst genotypes for all the traits being studied. The results revealed that Farkhr-e-Sarhad took up maximum nitrogen ( $47 \text{ mg plant}^{-1}$ ) and minimum nitrogen was taken up by accession 11172 ( $31 \text{ mg plant}^{-1}$ ). Maximum rooting depth of 76 cm was attained by accession 11277 and minimum of 43 cm was attained by NRL-0517. The study generated information on nitrogen uptake potential and rooting depth of Pakistani wheat genotypes. It provided baseline data for the exploitation of deep rooting and high nitrogen uptake capability towards the development of wheat varieties.

**Keywords:** climate change, nitrogen uptake, rooting depth, root yield, wheat

### INTRODUCTION

The biggest challenge of this century is to secure the nations by securing food under climate change as in future farmers will be growing crops in an environment different from today. Despite the technological advances, wheat yield potential had become stagnant. Scientists from across the globe are under taking serious efforts to break this stagnancy in yield. One key theme to raise yield potential of wheat will be to increase photosynthetic capacity and efficiency. However, plants with increased photosynthetic rate and a larger biomass are likely to require more efficient if not larger root systems (Reynolds *et al.*, 2011). Response of plants to climate change will be governed by resource availability at critical stages of growth and nitrogen (N) plays a vital role. Plant roots are key contributors towards the survival of plants under adverse conditions, maintaining supplies of water and nutrients. Increasing levels of  $\text{CO}_2$  increase growth and yield in  $\text{C}_3$  plants until sufficient N is available in soil indicating that N uptake is a bottleneck for yield enhancement under climate change (Zhang *et al.*, 2013).

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Pakistan is the 7<sup>th</sup> largest wheat producer in the world with annual production of 22 million tons (FAO STAT, 2010). Improved wheat varieties form an integral component of production technology in Pakistan as well as in other developing countries of the region. Green revolution introduced high yielding, fertilizer responsive and semi-dwarf wheat varieties through CIMMYT in many parts of the world including Pakistan that revolutionized cereal production (Borlaug, 1968). Root size of post green-revolution wheat genotypes is small compared to that of drought tolerant landraces. Their root system may be too small for optimum uptake of water and nutrients and maximum grain yield (Waines and Edahi, 2007). This tends to support the view that direct selection for only above-ground organs might also indirectly select for a small root system, especially under well-irrigated and well-fertilized growing conditions, as at CIMMYT breeding stations, where there would be no selection advantage for a larger root system. Recently selected landraces had been employed in CIMMYT's drought crossing programs that are providing initially promising results (Trethowan and Reynolds, 2007).

Roots, the hidden half of plants, had not been considered in making selections for yield improvement in wheat with few exceptions, e.g. Hurd *et al.* (1972); Hurd (1974); Richards and Passioura (1989). Root research is a key area offering enormous potential to improve yield and sustainability. Contribution of deep roots towards drought resistance in wheat has been reported by many workers, e.g. Sayar *et al.* (2007); Manschadi *et al.* (2007); Palta *et al.* (2011). Studies of Kage and Ehlers (1996) and King *et al.* (2003) suggest that water uptake optimization of cereals is obtained mainly by deep root systems with high specific root length. Taking into account the persisting difficulty of screening root traits, the main contribution of root research to breeding can be via a detailed characterization of few selected accessions to be used as potential parents for crosses (Blum, 2011). Local cultivars, landraces and wild relatives are valuable resources for traits related to abiotic stress resistance (Araus *et al.*, 2007; Trethowan and Mujeeb-Kazi, 2008; Habash *et al.*, 2009). Keeping in view the role of N uptake in maintaining plant productivity under climate change and potential contribution of deep roots in water uptake under water limited conditions, a study was designed to identify potential wheat genotypes for further use in wheat breeding program.

## **MATERIALS AND METHODS**

During winter 2012-13, a sand culture experiment was conducted under glass house conditions at Nuclear Institute for Food and Agriculture, Peshawar, Pakistan. Mean daily temperature during the season was 20°C and mean daily relative humidity was 25%. Fifteen wheat genotypes were tested in four replicates in a completely randomized design. Genotypes tested included commercial cultivars and heat tolerant accessions (Table 1). Large sized plastic pots (length-24 inches, diameter-5 inches) were filled with 10 kg of coarse sand and water content was kept around field capacity throughout the experiment. Five plants of almost equal size were maintained in each pot. Hoagland and Arnon (1950) nutrient solution (1/2 strength) was applied @ 0.5 L pot<sup>-1</sup> to meet nutritional requirement of plants at 30 days interval.

**Table 1.** Brief description of wheat genotypes

Genotypes	Distinct Features	Released By	Year of Release
Bathoor	High yielding, disease resistant Potential yield: 6-7 tons ha <sup>-1</sup>	Nuclear Institute for Food and Agriculture (NIFA), Peshawar, Pakistan	2008
Fakhr-e-Sarhad	Drought tolerant, lodging resistant Potential yield: 7-8 tons ha <sup>-1</sup>	NIFA, Peshawar, Pakistan	1998
Barsat	High yielding, low water requiring, moderately resistant to wheat rusts	NIFA, Peshawar, Pakistan	2010
Tatara	Drought tolerant, durable rust resistant, shattering resistant Potential yield: 5-6 tons ha <sup>-1</sup>	NIFA, Peshawar, Pakistan	1996
Chakwal-86	High yielding, drought tolerant	Barani Agricultural Research Institute, Chakwal, Pakistan	1986
Parwaz-94	High yielding, semi-dwarf variety, resistant to yellow rust, Potential yield: 5-6 tons ha <sup>-1</sup>	Wheat Research Institute, AARI, Faisalabad, Pakistan	1994
PR-98	High yielding, suitable for rain fed areas	Cereal Crops Research Institute, Pirsabak, Pakistan	Candidate variety
PR-102	Suitable for irrigated areas, aphid resistant	Cereal Crops Research Institute, Pirsabak, Pakistan	Candidate variety
NRL-0517 (NIFA Lalma)	High yielding, drought tolerant Zinc and Boron efficient, semi dwarf	NIFA, Peshawar, Pakistan	2014
PM-376	-	-	-
11172 11163 11277 18723 11144	Breeding lines from Australia under trials for heat and drought tolerance	Australia	Not available

Source: <http://wheatatlas.org/country/varieties/PAK/0>, <http://wheatpedigree.net>

At physiological maturity (140 days after sowing), data on above ground plant parameters was recorded. Plant height was assessed using a wooden meter rod from standing plants. Chlorophyll content was measured using SPAD-Chlorophyll meter (SPAD-502, Konica Minolta, Japan) from fully expanded leaves. Leaf area plant<sup>-1</sup> was measured using AccuPAR PAR/LAI ceptometer (model LP-80, Decagon Devices, Inc., Pullman, USA). At maturity, plants were removed from pots and threshed to assess grain weight per spike. Roots were washed out of sand using water and spread over a wooden board to assess their depth using wooden meter rod. Plant samples were dried in an oven at 65°C for 48 hours (Pietsch *et al.*, 2007). Samples were processed and analyzed for N content and uptake, following the methods described by Jackson (1962). Data were analyzed using MSTAT-C software to perform analysis of variance and means were compared using least significant difference values at 5% level of significance.

## RESULTS AND DISCUSSION

Significant ( $P \leq 0.05$ ) differences were found among genotypes for all the traits under study (Table 2 and 4). Genotypes can be broadly classified into two groups based on plant height, i.e. dwarf (<70 cm) and semi-dwarf (70-100 cm). This grouping can have implications for rooting depth and for uptake of water and

nitrogen as was indicated by Waines and Edahi (2007) that dwarf plants have generally shallow root system with limited capacity for water and N uptake. Plant height varied from 64-89 cm. Accession 11277 was the shortest genotype (64 cm) and NRL-0517 was the tallest (89 cm). Entries PM-376, 11172 and 11277 belonged to dwarf group (plant height < 70 cm), while others belonged to semi-dwarf group (70-100 cm).

**Table 2.** Growth, development and yield traits of wheat genotypes

Genotypes	Plant height (cm)	Grain weight (spike <sup>-1</sup> )	Dry weight (g plant <sup>-1</sup> )	Leaf area (cm <sup>2</sup> plant <sup>-1</sup> )	Chlorophyll content (%)	Rooting depth (cm)	Root dry weight (g plant <sup>-1</sup> )	Root N uptake (mg plant <sup>-1</sup> )	Total N uptake (mg plant <sup>-1</sup> )
Bathoor	81abcde	1.80abc	2.65abcd	77de	44abc	56cdef	1.65cd	5.0def	38cd
Fakhr-e-Sarhad	80abcde	1.95a	2.68abcd	97abc	43abc	65bc	2.05bc	7.01bc	47a
Barsat	84abcd	1.70abcd	2.25bcd	110a	44abc	52efg	2.08bc	6.7bcd	40bcd
Tatara	76cdef	1.88ab	2.33bcd	87cd	40cd	47fg	1.28def	4.2ef	45ab
Chakwal-86	72efg	1.55cdef	3.10ab	70de	37d	47fg	1.73bcd	5.5cde	41bcd
Parwaz	73efg	1.48defg	3.00abc	70de	46a	54ef	1.58cde	6.7bcd	44abc
PR-98	78bcdef	1.48defg	2.00d	89bcd	43abc	43g	0.93f	3.2f	36def
PR-102	74defg	1.58bcde	2.18cd	75de	43abc	64bcd	1.33def	5.2cde	39cd
NRL-0517	89a	1.33efg	2.18cd	107ab	45ab	43g	1.05ef	3.2f	32ef
PM-376	68fg	1.43defg	2.30bcd	70de	43abc	59cde	1.50cdef	5.2cde	36def
11172	64g	1.20g	2.40bcd	60e	47a	55def	2.78a	9.7a	31f
11163	75cdef	1.28efg	2.88abc	84cd	46a	61cde	1.68cd	6.7bcd	40bcd
11277	64g	1.25fg	3.48a	81cd	41bcd	76a	2.05bc	6.5bcd	37de
18723	85abc	1.40defg	2.93abc	74de	44abc	64bc	1.73bcd	6.5bcd	41bcd
11144	88ab	1.25fg	1.95d	58e	44abc	71ab	2.3ab	8.0ab	32ef
LSD (0.05)	10.7	0.32	0.874	19	5.3	9.4	0.58	1.8	5.5

Grain weight spike<sup>-1</sup> ranged from 1.20-1.95 g. Accession 11172 had the lowest (1.20 g) and Fakhr-e-Sarhad had the highest (1.95 g) grain weight spike<sup>-1</sup>. Dry weight plant<sup>-1</sup> ranged from 1.95-3.48 g. Accessions 11144 and 11277 attained 1.95 g and 3.48 g dry weight plant<sup>-1</sup>, respectively. Leaf area plant<sup>-1</sup> varied from 58-110 cm<sup>2</sup>. Accession 11144 reached the value of 58 cm<sup>2</sup> for leaf area plant<sup>-1</sup> and cv. Barsat got 110 cm<sup>2</sup>. Chlorophyll content ranged between 37-47%. Chakwal-86 possessed the lowest content of 37%, while accession 11172 had the highest content of 47%.

Genotypes under study exhibited variation in maximum rooting depth (43-76 cm). Genotype PR-98 and NRL-0517 attained rooting depth of 43 cm each. Accession 11277 had 76 cm deep roots and was at par with 11144 (71cm). Deep rooting (71-76 cm) attributes of accession 11277 and 11144 make them a suitable choice to be used as parent in breeding for water limited environments. Root dry weight plant<sup>-1</sup> was found in the range of 0.93-2.78 g. Genotype PR-98 has shallow root depth (43 cm) and it attained lowest dry weight plant<sup>-1</sup> (0.93 g). The accession 11172 attained the highest root dry weight plant<sup>-1</sup> (2.78 g). This

accession belonged to dwarf plant height group and entries in this group had usually higher root dry weight plant<sup>-1</sup> except PM-376. A higher root dry weight is usually an indicative of more root surface area and root volume in most of the cases. These features are desirable for water and nutrients uptake (Himmelbauer *et al.*, 2004). The same accession also had highest values of chlorophyll content indicating that it is a good accumulator of assimilates in vegetative parts but as it possessed lower grain weight, it seems not be a good converter of assimilates into reproductive parts. It needs to be crossed with other high grain yielding parent to take advantage of these traits. Using this accession in breeding programs has certain merits towards sustainable agriculture as on account of its high root weight; it is going to contribute more towards soil organic matter contents in the long run (Kell, 2011).

Root N uptake was found in the range of 3.2-9.7 mg plant<sup>-1</sup>. Genotypes PR-98 and NRL-0517 exhibited lowest N uptake (3.2 mg plant<sup>-1</sup>). These entries had poor rooting depth and root dry weight that contributed towards lower N uptake. Accession 11172 achieved the highest value of N uptake (9.7 mg plant<sup>-1</sup>). This accession belonged to dwarf group and had attained highest value of root dry weight plant<sup>-1</sup>. Total N uptake varied from 31-47 mg plant<sup>-1</sup>. Accession 11172 had the lowest value of N uptake plant<sup>-1</sup> (31 mg) while Fakhr-e-Sarhad had the highest value of 47 mg plant<sup>-1</sup>. Correlation analysis among parameters studied revealed that rooting depth has positive and significant correlation with dry weight per plant and root dry weight. Highly significant correlation was observed between grain weight per spike and total N uptake (Table 3).

Findings from the study indicated that accessions offer a valuable source of germplasm on account of their desirable root traits for further exploitation in breeding program. Rooting depth of accessions was found in the range of 55-76 cm while other genotypes had root depth of 43-65 cm. Root dry weight plant<sup>-1</sup> of accessions was 1.68-2.78 g and was better than other genotypes in the present study that varied between 0.93-2.08 g plant<sup>-1</sup>. It suggests that accessions are a valuable asset for use in breeding for sustainable agriculture based on their root dry weight as well because root residues may ultimately contribute towards long term development of soil fertility. It is emphasized that use of accessions in breeding program for coming decades can be a part of technology for climate smart agriculture. Efforts should be made to develop varieties that return more residues to soil through their greater root weights; it means we are returning more towards the soil in terms of organic residues (Gregory *et al.*, 2010; Raza *et al.*, 2015). Maintaining higher organic matter content is a viable option to mitigate adversaries of climate on soil and crop as organic matter acts as a buffer under drought and helps soils to improve their water holding capacity (Pathak *et al.*, 2012).

Roots offer enormous potential for crop improvement under changing climate (Araus *et al.*, 2008; Habash *et al.*, 2009; Wu and Cheng, 2014). We have demonstrated existing variability among accessions and cultivated genotypes through this study. This variability in root traits needs to be exploited to favor the farming community. As rooting traits have been ignored by CIMMYT and national breeding programs over the last decades, we have found that rooting depth and weights have greatly reduced in current and upcoming varieties. This trend will not favorable to sustainable wheat production in climate vulnerable zones of

Pakistan as well as in the region. We need a partial shift in our wheat breeding strategy that shall focus both on grain yield as well as on root traits. Optimization of rooting depth in wheat genotypes is a research question worth pursuing and it may vary with region, climate and soil type. Modeling offers tremendous potential to identify suitable plant traits and their optimum values for developing ideotypes for various climate change scenarios for future.

**Table 3.** Correlation among studied parameters

Parameter	Rooting depth	Total N uptake	Grain wt. spike <sup>-1</sup>	Chlorophyll content	Plant height	Leaf area plant <sup>-1</sup>	Root dry weight plant <sup>-1</sup>
Dry wt./plant	0.329*	0.063	-0.079	-0.016	-0.050	-0.133	0.321*
Rooting depth		0.0001	-0.235	0.038	-0.126	-0.231	0.429**
Total N uptake			0.552**	-0.087	0.233	0.235	0.134
Grain wt./spike				-0.162	0.225	0.248	-0.096
Chlorophyll content					0.160	-0.141	0.157
Height						0.158	-0.200
Leaf area							-0.165

**Table 4.** Analysis of variance (Mean sum of squares)

SOV	DF	Plant height	Grain weight Spike <sup>-1</sup>	Dry weight plant <sup>-1</sup>	Leaf area plant <sup>-1</sup>	Chlorophyll content	Rooting depth	Root dry weight plant <sup>-1</sup>	Root N uptake	Total N uptake
Replication	3	94.77	0.05	0.22	278.02	11.22	35.58	0.23	1.17	16.55
Treatment	14	253.89	0.23	0.81	954.22	27.17	391.32	0.95	11.95	89.30
Error	42	56.41	0.05	0.38	181.57	13.70	43.64	0.16	1.58	15.12
CV (%)		9.8	14.55	23.99	16.75	8.54	11.58	23.57	20.99	10.09

## CONCLUSION

The present study was conducted to identify wheat genotypes for higher rooting depth and N uptake potential. Accession 11277 and 11144 were identified as the deepest rooting entries and their high rooting depth can be exploited in breeding program for water limited environments. It is suggested to cross them with locally well adapted, high yielding varieties to evolve new genotypes. Fakhr-e-Sarhad and Tatara maintained the highest N uptake per plant and can be a suitable choice of parents for developing varieties that can maintain higher productivity under changing climatic conditions especially under elevated levels of CO<sub>2</sub>. The untapped potential of roots in overcoming the current and forthcoming problems cannot be overlooked. It is emphasized that roots shall be targeted for wheat improvement for ensuring national and global food security under climate change. Root research is a neglected area that needs to be incorporated in the ongoing national wheat breeding programs to develop new wheat varieties having high water and nutrient use efficiency for ensuring food security. This study provided preliminary information on rooting depth and N uptake potential of selective genotypes. Further detailed characterization of

wheat genotypes under field conditions on their water uptake in conjunction with root traits are needed to confirm the findings from this pot experiment. Additional studies with diverse germplasm from national and international sources will help us to identify more potential sources for use in wheat breeding program dedicated towards the development of varieties that can perform better under water limited conditions in a changing climate.

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