# Screening for genotypic variation in phosphorus-uptake efficiency in cereals on Australian soils

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## Abstract

A glasshouse pot trial was conducted to screen for P-uptake efficiency in 18 cereal genotypes by growing plants on two soils with high total P content but low plant available P. Genotypic variation in plant growth and P uptake were apparent in both soils at two harvests made at 21 and 35 days after sowing. A significant P fertilizer response was observed on both soils. Total P uptake by the genotypes was closely correlated with shoot biomass, suggesting that shoot biomass may be a reliable parameter to select for P-uptake efficiency in short-term pot bioassays. This variation may provide opportunities for developing molecular markers for wheat breeding programs to improve P-uptake efficiency in wheat.

## Media summary

Significant genotypic variation in phosphorus-uptake efficiency was found in hexaploid wheat and other cereal germplasm. This may provide opportunities to explore genetic variation in wheat breeding programs.

## Key Words

Cereals, genotypic variation, phosphorus, soil, uptake-efficiency, wheat

## Introduction

Grain producers in Australia currently spend an estimated $450 million annually on phosphorus (P) fertilizer. However, only 10-20 % of this applied P is directly used by crops in the year of application and subsequent usage of the residual P rarely exceeds 50% (Bolland and Gilkes 1998). Soluble P fertilizers applied to soil react with soil constituents and are readily “fixed” as adsorbed P, sparingly-soluble P-precipitates (Al-P, Fe-P or Ca-P) or converted to organic P forms. These fixed pools of P are largely unavailable to crop plants. This is a problem in many Australian soils, particularly in acidic and alkaline soils. Such soils can accumulate large amounts of P, yet plant growth can be limited by poor P availability. The development of crop plants that are better able to acquire P from fixed-P forms, as well as using the P taken up more effectively, will result in more efficient use of P fertilizer with resulting yield benefits and cost savings.

Relatively little information is available on the P-uptake efficiency of Australian wheat cultivars, particularly in soil. Few studies have assessed the variation in P efficiency of wheat (Batten, 1986; Batten and Khan 1987; Jones et al 1989; Osborne and Rengel, 2002). In one of the most comprehensive screens published so far (Osborne and Rengel, 2002), over 100 cereal genotypes (mainly Western Australian lines) were screened in washed sand and supplied with nutrient solution that contained different levels of soluble P. This study established that, under these defined conditions, there was significant variation in P nutrition characteristics among wheat cultivars. However, it needs to be established whether similar variation in P-uptake efficiency occurs in a range of Australian agricultural soils. The aim of this study was to develop a protocol to establish the extent of variation in P-uptake efficiency among genotypes of wheat and other cereals.

## Methods

*Materials*

The cereal genotypes included 12 hexaploid and 3 synthetic hexaploid wheat lines, 1 rye line, 1 triticale line and 1 durum line.

The soils used for the screen were a Red Ferrosol from Robertson, NSW (pH, 5.6; total P, 2500 mg P/kg soil; resin P, 2.1 mg P/kg soil) and a Red Kandosol from Grenfell, NSW (pH, 5.2; total P, 200 mg P/kg soil; resin P, 5.8 mg P/kg soil). The soils were collected from the 0-10cm layer, air dried and sieved through a 4 mm screen. For the pot size used, 2.8 L soil (2.7 kg for the Robertson and 3.3 kg for the Grenfell soil) was measured into plastic bags. There were two P application rates (-P, 0 kgP/ha; +P, 100 kg P/ha for Grenfell soil and 700 kg P/ha for Robertson soil). Each treatment was replicated 4 times per genotype. Finely ground triple superphosphate (20.7% P, passed through a 2 mm sieve) was added to the soil in the plastic bags of the +P treatments. Basal nutrient solutions to provide 80 kg N/ha, 50 kg K/ha, 20 kg Mg/ha, 20 kg Ca/ha, 5 kg Zn/ha, 2 kg Cu/ha, 1 kg Mo/ha and 2 kg B/ha were added to each soil bag and mixed thoroughly with the soil. The mixed soils were put into 20 cm ? 15 cm pots, and then watered to 65% of field capacity.

*Management*

Eight even-sized seeds of each genotype were sown into each pot at a depth of 2 cm. After sowing, additional water was added to achieve 65% field capacity. A complete randomized block design was used. Pots were randomly located on benches within each block in a naturally-lit glasshouse. The temperature regime was maintained at a day/night temperature of 20/10?C throughout the experiment. Seedlings were thinned to 5 uniform plants per pot at the 1-leaf stage. Pots were watered to 65% of field capacity every 2 days at early stages and every day at later stages. Additional N (50 kg N/ha as $NH_4NO_3$) was top-dressed at tillering. Weeds were removed from pots manually.

The first harvest was made at the 3-leaf stage, 21 days after sowing (DAS), when 2 plants were randomly selected from each pot and cut off at the soil surface. Shoots were dried at 70?C for 48 hours, weighed, and then milled to a fine powder in a puck mill. Shoot material was digested in a sulfuric acid/hydrogen peroxide mixture, and P concentrations were measured by the molybdate blue method. The second harvest was made when the reference wheat line, Westonia, was at the 5-leaf stage (35 DAS). Whole plants were harvested and shoot dry weights were obtained. Roots were recovered from the soil by using a water spray and a 2 mm sieve. The cleaned root samples were sub-sampled for root length measurement as described by Liao et al (2004), and the remainder of the root samples dried. Dry weights of all root sub-samples were recorded separately, so that total root length and weights per plant could be calculated. Plant tissue P concentrations were measured as for harvest 1.

**Results**

Shoot and root biomass of all 18 cereal genotypes increased significantly when grown on either the Robertson or Grenfell soil when P fertilizer was applied. This suggests that both soils were P-deficient for the cereal genotypes studied. There were significant differences in shoot biomass, root biomass and root length between the genotypes grown on both soil types at a given P rate. Table 1 shows the genotypic variations in plant growth and P uptake parameters of all the genotypes grown on the Robertson soil with or without P addition. The Grenfell soil provided similar results (data not presented).

Under P-limiting conditions, shoot biomass of the 18 genotypes ranged from 0.06 to 0.13 g/plant and 0.10 to 0.31 g/plant when grown on Robertson and Grenfell soil, respectively (see Table 1 for the Robertson soil). When P fertilizer was supplied, shoot biomass ranged from 0.62 to 1.17 g/plant and 0.60 to 1.13 g/plant for Robertson and Grenfell soil, respectively. A significant genotypic difference in root/shoot ratio was found for both soils. Tissue P concentrations and total P uptake differed significantly between genotypes for a given P treatment on both soils. On the Robertson soil, shoot P concentrations ranged from 1.07 to 1.96 mg P/g DW and 4.01 to 5.08 mg P/g DW for the -P and +P treatments, respectively, when the plants were harvested at 35 DAS (Table 1). The differences in plant biomass and tissue P concentrations between genotypes resulted in significant genotypic differences in total P uptake.

Plant P uptake was closely correlated with seed P content when the genotypes were grown on Robertson soil in the absence of P fertilizer. On the Grenfell soil, however, seed P content correlated with shoot P uptake at the first harvest only. When P fertilizer was added, seed P content had no significant impact on plant P uptake (Table 2). Positive correlations were found between shoot biomass and total P uptake, suggesting that biomass may be a reliable parameter for selecting P-uptake efficiency under various P availabilities.

The responsiveness of the genotypes to P addition (ratio of shoot biomass in the +P treatment to shoot biomass in the -P treatment) differed significantly. At the first harvest (21 DAS), the P responsiveness of genotypes ranged from 1.2-4.2, and was independent of soil type. The P responsiveness of the genotypes increased with plant growth stage, and ranged from 6-19 for the Robertson soil and from 4-9 for the Grenfell soil at the second harvest (35 DAS). The rates of P application in the +P treatment were much higher than the rates used by farmers in Australia, so a further study is currently being conducted to examine the responsiveness of genotypes to more typical P levels.

**Table 1. Shoot dry weight (SDW, g/plant), root dry weight (RDW, g/plant), root to shoot ratio (RDW/SDW), root length (RL, m/plant), shoot P concentration (SPC, g P/kg DW), and total P uptake (TP, mg P/plant) by 18 cereal genotypes at harvest 2 (35 DAS) grown on Robertson soil in a glasshouse.**

| Genotype | SDW | RDW | RDW/SDW | RL | SPC | TP |
|---|---|---|---|---|---|---|
| | | | **-P (0 kg P/ha)** | | | |
| Synthetic Hexaploid 1 | 0.073 | 0.121 | 1.67 | - | 1.78 | 0.303 |
| 2 | 0.088 | 0.138 | 1.55 | 23.5 | 1.31 | 0.337 |
| 3 | 0.079 | 0.134 | 1.71 | - | 1.96 | 0.356 |
| Hexaploid 4 | 0.096 | 0.131 | 1.36 | - | 1.22 | 0.356 |
| 5 | 0.108 | 0.167 | 1.59 | - | 1.49 | 0.394 |
| 6 | 0.043 | 0.095 | 2.19 | - | 1.83 | 0.234 |
| 7 | 0.072 | 0.111 | 1.57 | 17.6 | 1.39 | 0.283 |
| 8 | 0.110 | 0.125 | 1.14 | 22.0 | 1.07 | 0.308 |
| 9 | 0.047 | 0.113 | 2.63 | 22.3 | 1.82 | 0.297 |
| 10 | 0.067 | 0.122 | 1.83 | 21.9 | 1.39 | 0.254 |

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|---|---|---|---|---|---|---|
| 11 | 0.059 | 0.111 | 1.93 | - | 1.44 | 0.278 |
| 12 | 0.103 | 0.161 | 1.57 | - | 1.44 | 0.367 |
| 13 | 0.176 | 0.219 | 1.25 | 39.1 | 1.28 | 0.582 |
| 14 | 0.116 | 0.142 | 1.22 | 24.1 | 1.61 | 0.422 |
| 15 | 0.086 | 0.149 | 1.75 | 24.8 | 1.39 | 0.339 |
| Rye 16 | 0.082 | 0.147 | 1.83 | 29.8 | 1.58 | 0.371 |
| Triticale 17 | 0.058 | 0.100 | 1.74 | 19.4 | 1.62 | 0.260 |
| Durum 18 | 0.134 | 0.246 | 1.29 | 28.6 | 1.37 | 0.394 |
| $LSD_{0.05}$ | 0.028 | 0.036 | 0.28 | 6.1 | 0.33 | 0.086 |
| | | | **+P (700 kg P/ha)** | | | |
| Synthetic Hexaploid 1 | 0.931 | 0.468 | 0.50 | - | 4.34 | 5.550 |
| 2 | 1.115 | 0.494 | 0.45 | 100.7 | 4.77 | 6.559 |
| 3 | 0.747 | 0.441 | 0.59 | - | 4.35 | 4.597 |
| Hexaploid 4 | 0.620 | 0.310 | 0.51 | - | 4.84 | 4.064 |
| 5 | 0.949 | 0.398 | 0.42 | - | 4.37 | 5.217 |
| 6 | 0.827 | 0.430 | 0.53 | - | 4.02 | 4.482 |
| 7 | 0.833 | 0.349 | 0.42 | 62.9 | 4.56 | 4.836 |
| 8 | 0.742 | 0.402 | 0.54 | 85.1 | 4.60 | 4.903 |
| 9 | 0.859 | 0.390 | 0.45 | 70.6 | 4.51 | 5.125 |

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|---|---|---|---|---|---|---|---|
| 10 | 0.904 | 0.429 | 0.48 | 90.8 | 4.28 | 4.966 |
| 11 | 0.771 | 0.424 | 0.55 | - | 4.59 | 4.892 |
| 12 | 0.929 | 0.473 | 0.52 | - | 4.01 | 5.128 |
| 13 | 1.047 | 0.503 | 0.48 | 74.6 | 4.11 | 5.751 |
| 14 | 1.159 | 0.505 | 0.43 | 78.0 | 4.01 | 6.087 |
| 15 | 0.909 | 0.524 | 0.58 | 89.7 | 4.46 | 5.449 |
| Rye 16 | 1.172 | 0.514 | 0.44 | 77.2 | 5.04 | 7.541 |
| Triticale 17 | 0.944 | 0.379 | 0.40 | 73.9 | 5.08 | 6.097 |
| Durum 18 | 0.869 | 0.392 | 0.45 | 70.8 | 4.51 | 5.049 |
| $LSD_{0.05}$ | 0.198 | 0.058 | 0.10 | 20.7 | 0.66 | 0.680 |

**Table 2. Correlations between seed P content (seed P), shoot dry weight (SDW, 1 and 2 designate harvest 1 and 2, respectively), root dry weight (RDW) and shoot P accumulation (SP) and total P uptake (TP) for 18 cereal genotypes grown on Robertson and Grenfell soil with and without P fertilizer. Values are $r^2$ for linear regressions (n=18). $^{*}$Significant at 0.05 level; $^{**}$ significant at 0.01 level; $^{***}$significant at 0.001 level.**

| P treatment | Parameter | Robertson soil | | | Grenfell soil | | |
|---|---|---|---|---|---|---|---|
| | | SP1 | SP2 | TP2 | SP1 | SP2 | TP2 |
| -P | Seed P | 0.718$^{***}$ | 0.567$^{***}$ | 0.504$^{**}$ | 0.269$^{*}$ | 0.020 | 0.058 |
| | SDW1 | 0.941$^{***}$ | 0.699$^{***}$ | 0.571$^{**}$ | 0.806$^{***}$ | 0.363$^{**}$ | 0.429$^{**}$ |
| | SDW2 | - | 0.829$^{***}$ | 0.631$^{**}$ | - | 0.907$^{***}$ | 0.912$^{***}$ |
| | RDW2 | - | 0.699$^{***}$ | 0.631$^{***}$ | - | 0.462$^{**}$ | 0.701$^{***}$ |
| +P | Seed P | 0.051 | 0.001 | 0.001 | 0.035 | 0.043 | 0.049 |

| | | | | | | |
|---|---|---|---|---|---|---|
| SDW1 | 0.718*** | 0.245* | 0.248* | 0.870*** | 0.477** | 0.592** |
| SDW2 | - | 0.805*** | 0.801*** | - | 0.750*** | 0.783*** |
| RDW2 | - | 0.306* | 0.403* | - | 0.155 | 0.321* |

## Conclusion

Plant growth and P uptake by a diverse range of cereal genotypes differed considerably, indicating that breeding for P-uptake efficiency in wheat is possible. The close correlations between shoot biomass and plant P uptake suggests that shoot biomass is a reliable parameter for screening wheat genotypes in soils for P-uptake efficiency.

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