Identification of Phosphorus Efficient Rice Cultivars under Low P Nutrition through Hydroponic based Screening

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ABSTRACT: Phosphorus (P) is one of the major nutrients required by plants. A higher portion of P in lowland rice soils is found in unavailable forms due to fixation. Therefore, continuous application of P fertilizer to rice (*Oryza sativa* L.) is needed to obtain a satisfactory yield. Identification and subsequent cultivation of high yielding rice varieties which can withstand low level of P is a better alternative to the continuous P application. The objective of this study was to categorize Sri Lankan rice varieties according to their response to P deficient conditions. Forty eight rice varieties including three old improved varieties and 45 new improved varieties were evaluated at deprived (10 μ M P) and sufficient (50 μ M P) P levels in a nutrient solution culture. Multiple plant traits; number of tillers, root and shoot dry weights and P content in shoot tissues were assessed at 52 days after planting. Rice variety Bg 94-1 gained higher biomass and P uptake (i.e. shoot P content- mg/plant). Simultaneously, At402 had lower biomass gain and shoot P content. Rice varieties were grouped into two distinct clusters based on their responses to P deficiency such as biomass gain, P uptake and number of tillers per plant. This study showed that At 405, Bg 94-1, At 307, Bg 304, Bg 300 and At 354 are promising rice varieties with higher response to low level of P supply.

Keywords: Deficiency, germplasm evaluation, phosphorus, screening, tolerance

INTRODUCTION

Rice is the most important food crop in Asia, and the food security mainly depends on the production of rice. As such rice cultivation plays a big role in Asia generating income directly and indirectly (Dawe, 2000). Increase in the rate of rice production shows a diminishing trend with urbanization, climate change and soil problems. Therefore, the increase in rice production should be obtained through increasing productivity of rice per unit area cultivated (Redfern *et al.*, 2012). One solution for increasing production efficiency (i.e. productivity) in rice is through the development of higher-yielding and nutrient efficient varieties (Kush, 1995). Sixteen essential elements are required for proper growth of rice; mainly nitrogen (N), phosphorus (P) and potassium (K) are supplied in rice fields as inorganic fertilizers in greater quantities (De Datta, 1981). Unavailability of adequate amount of P in soil, retarded plant

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growth and development resulting significant yield losses (Dobermann and Fairhurst, 2000). Phosphorus requirement and acquisition are higher during the early growth stages (Vinod and Heuer, 2012).

Applied P is quickly converted to unavailable forms adhering to soil particles and can lead to deficiency in available P in soil. Also P deficiency in soil can occur due to the low P content of the parental material, low pH and soil with high P-fixing characteristics (Rose and Wissuwa, 2012). Phosphorus deficiency is one of the major limiting factors for crop production in highly weathered soils (Sanchez and Salinas, 1981). Some soil properties, such as soil pH, clay, Fe and Al contents are closely related to the P sorption capacity of the soils. Due to high P fixation capacity, a greater amount of external P should be applied to reach higher productivity (Yost *et al.*, 1979). Excessive application of P fertilizers can cause P contamination of freshwater bodies. This triggers the eutrophication in freshwater systems (Tirado and Allsopp, 2012). Therefore, it is important to use a balance fertilizer for sustainable rice farming. Application of high amounts of P fertilizers for two seasons per year has increased the accumulation of P in Sri Lankan soils (Sirisena and Suriyagoda, 2018). Therefore, the most economical and environmental friendly alternative is to introduce rice varieties which perform well under low P conditions.

Rice varieties differ significantly in their P requirement under P deficient condition. To identify P efficient varieties shoot and root dry weight can be used as a criterion for screening (Fageria.1998). Kottearachchi *et al.* (2013) has reported that there is a significant difference in root length and shoot dry weight between rice varieties grown under low P hydroponic system although there was no such difference in high P supplied condition. Meanwhile, Aluvihare *et al.* (2015) has identified Bg94-1, Bg403 and At362 as P deficiency tolerant varieties. The aim of this work was to identify the variation in P deficiency tolerance in Sri Lankan rice varieties under controlled P levels in hydroponic nutrient medium, and thereby identify the best performing varieties having deficiency tolerance at vegetative stage.

METHODOLOGY

The experiment was conducted in a glasshouse at the Agricultural Biotechnology Center, University of Peradeniya, Sri Lanka. Minimum and maximum temperatures inside the glasshouse during this period were 26°C and 38°C, respectively. Forty eight locally bred rice varieties, forty-five new improved and three old improved rice varieties under different maturity age groups were screened in Yoshida nutrient solution with low (10 μ M) and high (50 μ M) P concentrations (Table 1).

Establishment and maintenance of plants

Seeds were imbibed in water for 24 h and germinated. Seedlings were transferred in to plastic buckets filled with full strength Yoshida nutrient solution (Yoshida, 1976) supplemented with 50 μ M of P (optimal) or 10 μ M P (deprived) concentrations (Kekulandara *et al.*, 2016). Sodium dihydrogen phosphate monohydrate (NaH₂PO₄.H₂O) was used as the P source. Six seedlings per bucket were maintained with equal spacing. The pH of the solution was maintained at 5.6 - 5.8 and it was replaced once a week. The experimental design was Complete Randomized Design with 3 replicates.

Variety	Duration	Recommendation	Pedigree
	(months)		
New Improve	ed Varieties		
Bg 250	2 1/2	Drought/flood escaping	Farmer field selection
Bg300	3	General cultivation	Bg 367-7//IR 841/Bg 276-5
Bg304	3	General cultivation	Co 10/IR 50//84-1587/Bg 731-2
Bg305	3	General cultivation	Bg 1203/Bg 1492
Bg310	3	Saline prone areas	Bg 300/Pokkali
Bw272-6B	3	Low country wet zone	BW 259-3/BW 242-5-5
At303	3	General cultivation	At 66-2/Bg 276-5
At306	3	General cultivation	OB 2273/At 05
At307	3	General cultivation	Bg 2225-1/Bg 96-3298
At 308	3	General cultivation	Bg 2225-1/Bg 2426-2
Bg366	3 1/2	General cultivation	Bg300/94-2236//Bg300/Bg304
Bg352	3 1/2	General cultivation	Bg 380/Bg 367-4
Bg369	3 1/2	Saline prone areas	Bg 94-1/Nonabokra
Bg360	3 1/2	General cultivation	84-3346/IR36//Senerang
•	2.1/2	T 1 1 1 1 1	Bg797/Bg300//85-1580/
Bg357	3 1/2	Island wide cultivation	Senerang M-17
Bg358	3 1/2	General cultivation	Bg 12-1/Bg1492
Bg94-1	3 1/2	General cultivation	IR 262/Ld66
Bw361	3 1/2	General cultivation	IR 36/Bw 267-3-11M
Bg359	3 1/2	Wet zone	88-5089/Bg 379-2
Bw364	3 1/2	Wet zone	IR 36/Bw 267-3-11M
Bw363	3 1/2	General cultivation	IR 36/BW 267-311M
Bw351	3 1/2	Low country wet zone	Bg 90-2/Bg 401-1
Bw 367	3 1/2	General cultivation	Bg 358/Bw 361
At353	3 1/2	Saline prone area	Bg 94-1(R)/Bg400-1//Bg 94-1
At354	3 1/2	Saline prone area	Bg 94-1/Pokkali
At 362	3 1/2	General cultivation	At 85-2/Bg 380
Ld368	3 1/2	Wet Zone	Ld 4-9-11/Ld 99-17-4
Ld365	3 1/2	Wet zone	Selection of Ld 355
Bg380	4	Dry &Intermediate zone	Bg 90-2* ⁴ /Ob 677
Bg403	4	General cultivation	83-1026/Bg 379-2
Bg406	4	Northern region	Bg 73-797/Ptb 33/Ob 678
Bg450	4 1/2	General cultivation	Bg 12-1 * ² /IR 42
Bg454	4 1/2	General cultivation	MR 1523/87-519
Bg455	4 1/2	Submergence areas	Ob2547/CR9413//IR46/Ob 2552
Bg379-2	4 1/2	Low country wet zone	IR 2071-586/Bg 400-1
Bg 400-1	4 1/2	General cultivation	Ob 678//IR 20/H-4
Bg11-11	4 1/2	General cultivation	Engkatek/#2H-8
Bw400	4 1/2	Saline and acid soils	Bw 259-3/Bw 242-5-5
Bw400 Bw451	4 1/2	Low country wet zone	By 259-57By 242-5-5 Bg 400-1/Bg 11-11
Bw451 Bw452	4 1/2	General cultivation	Hondarawala 502/C 104
Bw452 Bw453	4 1/2	Low country wet zone	IR 2071-586/Bg 400-1
At401	4 1/2	Costal Saline area	Bg 94-1/Pokkali
A1401			6
At402	4	Southern province	IR4432-52-6-4/Bg90-2//76-

Table 1. Rice varieties used for the study

At4054Dry and Intermediate zonesAt 402/Basmathi 442 zonesLd4084General cultivationAt 01/Ld 98-152Old Improved VarietiesFigure 100 (100 - 100	Table 1. cont	t		
Ld4084General cultivationAt 01/Ld 98-152Old Improved VarietiesFree Control of Contro	At405	4	Dry and Intermediate	At 402/Basmathi 442
Old Improved VarietiesGeneral cultivationMurungakayan 302/MasH44 1/2General cultivationMurungakayan 302/MasH73 1/2General cultivationPachchapefumal/Mas//H-5			zones	
H44 1/2General cultivationMurungakayan 302/MasH73 1/2General cultivationPachchapefumal/Mas//H-5	Ld408	4	General cultivation	At 01/Ld 98-152
H7 3 1/2 General cultivation Pachchapefumal/Mas//H-5	Old Improve	d Varieties		
1	H4	4 1/2	General cultivation	Murungakayan 302/Mas
H10 3 General cultivation Pachchaperumal/Mas/H-5	H7	3 1/2	General cultivation	Pachchapefumal/Mas//H-5
	H10	3	General cultivation	Pachchaperumal/Mas/H-5

Plant trait measurements

Physiological traits of varieties were assessed by evaluating multiple plant attributes closely related to P deficiency. Three plants were harvested as replicates from each pot at 52 days after planting. Number of tillers, roots and shoot dry weight, and shoot P content were taken at the time of harvesting.

Phosphorus analyses

Shoots and roots were air dried for two days and oven dried for 48 hours at 60° C. Weight of shoots and roots were measured and 5 g of each sample was ground and made into ash at 200 °C for 2 h followed by 450 °C for 2 h. The ash was dissolved in 6% HNO₃ and P concentrations were measured by colorimetric assay using the molybdovanadophosphate method using a spectrophotometer (Kitson and Melon, 1944). Shoot P content (SPC) was calculated by multiplying shoot P concentration with shoot dry weight.

Data analysis

Measured and calculated data were analyzed using GLM procedure in SAS to determine the main effects of variety, P concentration and interactions among them. Means were compared using least significant differences at P = 0.05. All the variables of P deprived condition were subjected to hierarchical cluster analysis using MINITAB statistical software to observe the grouping of varieties.

RESULTS AND DISCUSSION

The effects of P concentration in the medium, variety and their interaction on shoot dry weight, root dry weight and total dry weight were significant (P<0.05). At 50 μ M P level shoot and root dry weight among varieties were similar whereas at 10 μ M P level, dry weights were significantly different among varieties. Therefore, shoot, root and total dry weights were compared separately at 10 μ M P level for all the varieties tested. Bg 94-1 showed the highest biomass gain in all three aspects. Similarly P content was compared among all the varieties studied. Bg 94-1 showed a higher amount of shoot P content proving its ability to perform in low P availability. Simultaneously, At402 has the lowest biomass gain in shoot, root and total dry weights. Low shoot P content recorded in At 402 confirms the poor performance in P uptake and utilization efficiency(Figure 2).

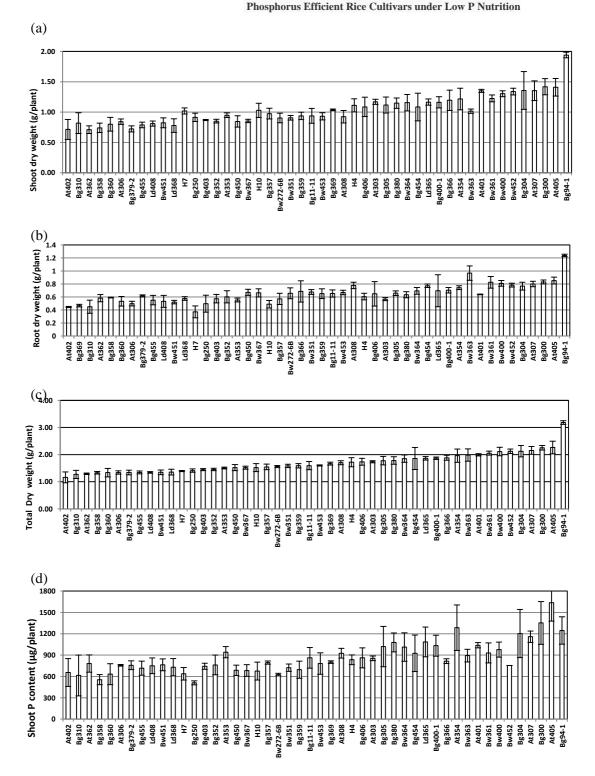


Figure 1. Variation of mean (a) shoot dry weight, (b) root dry weight (c) total dry weight and (d) shoot P content among the varieties grown at low P condition (10 μ M) Grouping of varieties

Crop varieties can be classified in to two different groups according to their responses to nutrient supply such as efficient or inefficient in nutrient deficient condition and responder or non- responder in nutrient sufficient condition (Ortiz-Monasterio et al., 2001). The dendrogram drawn using shoot and root dry weights, shoot P concentrations and contents and number of tillers under low P condition at 52 days after planting showed two distinct clusters. The varieties At405, Bg94-1, At307, Bg304, Bg300 and At354 clustered in to Cluster 2 while all other 44 varieties clustered in to Cluster 1 at similarity level at 33% (Figure 2). Varieties belonging to each cluster are shown in Table 2 and the means of each parameter are shown in Table 3. Means of all the P efficiency related parameters of Cluster 2 are greater than those of Cluster 1 revealing that the varieties in Cluster 2 are tolerant to P deficiency. Bg94-1, Bg304, At405 and At354 varieties have previously been identified as early tillering varieties at low P available condition confirming their efficiency for P uptake and use (Kekulandara et al, 2016). Although varieties At354 and Bg300 included in phosphorus deficiency tolerant category in this study, they were grouped under susceptible group by Aluvihare et al. (2016). Similarly, At362 was identified as P deficiency tolerant variety by Aluvihare et al. (2016) while an opposite response was observed in the present experiment. Bg 94-1 has been identified as a tolerant variety in both the studies. Each cluster consists of short, medium and long age rice varieties. It indicates that clustering has been made irrespective of the maturity age of the variety proving that there is no effect of the age of the variety on P deficiency tolerance.

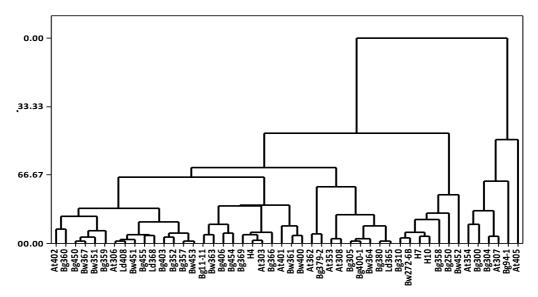


Figure 2. Dendrogram constructed by using shoot and root dry weights, number of tillers and shoot P concentration and contents at 10 µM P supply

Cluster 1			Cluster2		
At 303	At308	At306	Bg403	At354	
Bw400	At353	Bg455	Bw451	Bg94-1	
At401	Bg400-1	Bw351	Bg359	Bg300	
Bw364	Ld365	Bg352	H10	At405	
Bg380	Bg454	Ld408	Bg250	A307	
Bg360	Bw361	Bg450	Bg358	Bg304	
Bg406	At362	Bg310	Bg369		
H4	Bg379-2	H7	Bg366		
Bw363	Bg272-6B	At402			
Bw452	Bg11-11	Bw367			
Bw453	Bg305	Bg357			
Ld368	U U				

Table 2. Varieties in different clusters

Table 3. Mean values of variables in each cluster

	Cluster 1	Cluster 2
Shoot dry weight (g/plant)	0.97 ± 0.08	$1.45{\pm}0.16$
Root dry weight (g/plant)	0.63 ± 0.05	$0.87{\pm}~0.09$
Total dry weight (g/plant)	1.60 ± 0.10	2.32 ± 0.16
Shoot P concentration $(\mu g/g)$	$825.7 \pm 78.9 $	918.8±113.3
Shoot P content (µg/plant)	801.8 ± 103.5	1313.3 ± 248.2
Number of tillers per plant	1.71 ± 0.20	2.44 ± 0.33

Regression analysis

Scatter plot drawn for total dry weight of varieties against their time taken for matutity under low P conditions is shown in Figure 3. It descriminates Bg300, Bg94-1, At405 and Bw452 producing greater biomass from the age groups of 3 month, 3½ month, 4 month and 4½ month age groups respectively at low P supplied environment. Scatter plot drawn against maturity age and the mean P content in shoots is shown in Figure 4. Varieties Bg300, At354, At 405 and Bg380 were the superior in 3 month, 3½ month, 4 month and 4½ month age groups, rspectively.

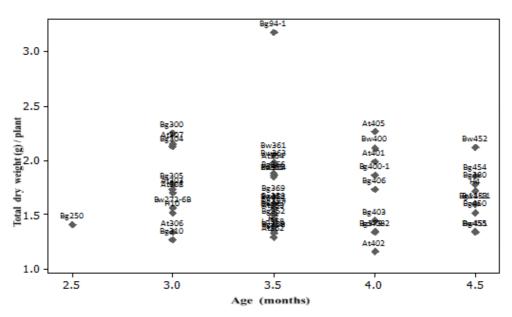


Figure 3. Distribution of varieties based on total dry weight gained at low P availability according to their maturity age classes

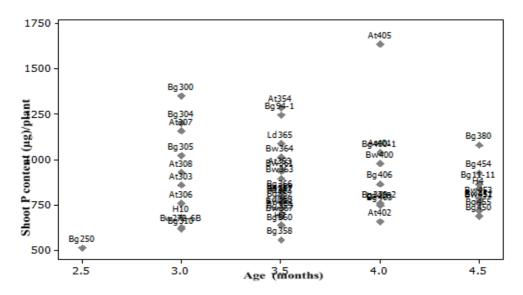


Figure 4. Distribution of varieties based on shoot P content at low P availability against their maturity age classes

Comparison of varietal performance under high and low P availability

The scatter plot drawn for biomass gained at low P and high P availability illustrates that varieties Bg94-1, At405, Bg300, Bg304 and At307 performed well in both at low and high P availability, whereas H7, H4, Bg455, At306, At402 and Bg11/11 performed poorly in both conditions (Figure 6). However, some varieties such as Bg250, Bg358, Bg310 and Bg369 have gained comparatively higher dry weight in high P availability compared to P deprived condition. Most of the short age varieties have shown higher dry weight at 52 days after planting at high P supply. It is clear that dry weight of all the varieties has reduced by more than half at low P condition compared to high P supply confirming the importance of P in plant growth and the occurrence of P deficiency at 10 μ M P supply (Figure 5).

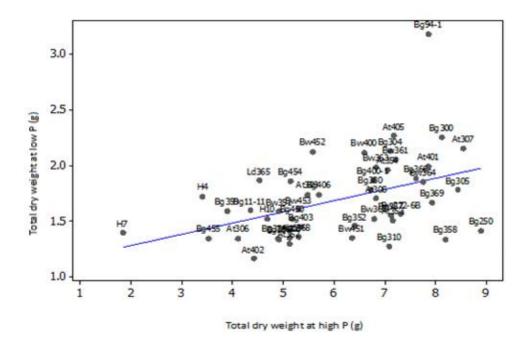


Figure 5. Varietal performances on dry weight gained at low P vs high P availability at 52 days after planting.

Efficient genotypes possess high P uptake efficiency, whereas responders show high utilization efficiency. As nutrient uptake and utilization processes are interdependent from each other, it is difficult to distinguish a responder from an efficient cultivar. Therefore, efficient screening method is important to select cultivars for low-nutrient tolerance (Agrama, 2006). Deficiency symptoms of plants can be studied well using hydroponics, where the presence or absence of nutrient components can be controlled precisely. Studies under controlled conditions in a hydroponic solution generally involve giving precise P deficiency stress on seedlings over in the field which may show a considerable level of spatial and temporal variation in P content in the medium. In addition, it makes easier to observe deficiency symptoms that occur in the roots, which is difficult to observe in soil-grown plants (Salisbury and Ross, 1992). Thus, hydroponic based screening system gives more reliable data than nutrient experiments conducted in soil although it is not the real practice adopted in rice cultivation.

CONCLUSIONS

There is a significant variation in genotypes for P deficiency tolerance in rice. The P deficiency tolerance ability in varieties did not show any correlation with their maturity age classes. Bg300, Bg304 and At307 in 3 month age group, Bg94-1, At354 in 3½ month age group and At405 in 4 month age group performed well in both P uptake and dry matter production. Bg94-1 performed extraordinarily in biomass gain and P accumulation. Simultaneously, At402 showed poor performance in biomass gain in both shoots and roots. Low amount of shoot P content recorded in At402 confirms its poor P uptake and utilization efficiency under P deprived conditions.

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