Genotypic Variation for Response to Phosphorus Fertilization in Common Bean Mutants

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ABSTRACT

Common bean is a source of protein. However, its productivity among smallholder farmers is low and thus cannot sustain the demand. One of the constraints limiting bean production is phosphorus (P) deficiency. Use of efficient P utilizing genotypes is an option which is feasible to small scale farmers. One way of creating genetic variability is through mutation breeding which generates mutants genetically different from the parental genotype in one or few traits. The specific objectives of the study were (i) to evaluate seed yield potential of bean mutants in P limiting soils and (ii) to investigate the association of seed yield to nodulation among the evaluated bean mutants. The experiment was laid as a 5 (genotypes) x 3 (phosphorous levels) factorial design in the screen house. The genotypic and P effects were significant ($p < 0.05$) for vine length, nodulation, pod yield, seed yield, dry shoot biomass and root biomass. SZ3-3-B-B2 out-performed the parent in both seed yield per hectare and nodulation. The genotype x P interaction was highly significant ($P < 0.001$) for seed yield and the mutant SZ3-3-B-B2 produced a significantly higher yield at a fertilization rate of 30 Kg/ ha P than the parental genotype. It can be deduced that SZ3-3-B-B2 was the most efficient P utilizing genotype. In addition, the amount of phenotypic variation (r2) explained for the association between nodulation and yield was found to be 0.10. These results suggest that there is a possibility that artificial inoculation can enhance the performance of the evaluated bean mutants.

Keywords: *Mutation, Beans, Phosphorus, Nodulation, Seed yield.*

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1. INTRODUCTION

Common bean (*Phaseolus vulgaris* L.), is one of the most important pulse crops in Africa and Latin America. It originated in Latin America and has two primary centers of origin in the Mesoamerican and Andean regions that are easily distinguished by molecular means [1]. Common bean accounts for around 50 % of the grain legumes consumed globally [2] and it is high in protein, starch, dietary fiber, potassium, molybdenum, thiamine, vitamin B6, folic acid and selenium, which can be eaten as dry seed or as green beans. In Eastern and Southern African countries, beans are a cheaper source of protein especially among the resource poor people [3].

In Zambia, most farmers grow landraces that are favoured for their colour and taste. However, these landraces have low yield potential and are susceptible to pests and diseases. The average yield of the landraces in Zambia is between 0.3 to 0.5 tons per hectare, which is low when compared to the yield potential of improved cultivars [3] . Contributing factors to this low yield include the inherently low yielding potential of landraces, biotic and abiotic stresses. Abiotic stresses include drought and low soil fertility. Among the soil fertility factors, inadequate Phosphorus (P) is one of the major abiotic factors limiting bean productivity in the tropics [4]. It is important for biological nitrogen fixation (BNF), which occurs in the root nodules, as it provides the basis for the formation of useful energy. This energy is essential in the production of ammonia (NH_3) which is later on converted to amino acid such glutamine for plant growth and development [5, 6]. Common bean production on soils deficient in P and without P fertilizer application, contribute to low BNF and yield losses [4]. To improve productivity and reduce cost of fertilizer, use of genotypes which are efficient at utilizing available phosphorous should be identified or bred. In addition, P- efficient genotypes are a viable option for small scale farmers.

Genetic improvement of common bean for P use efficiency needs adequate genetic variability. Mutation through the use of gamma rays is one of the tools previously used to create genetic variability for a wide range of traits in bean [7, 8]. Among the common bean mutants, some promising genotypes based on preliminary yield trials were evaluated for their yield performance in varying levels of P- fertilizer application. There is therefore need to determine whether seed yield performance is correlated with nodulation in these mutants so as to establish weather artificial inoculum may be required to enhance performance. The specific objectives of the study were therefore (i) to evaluate seed yield potential of bean mutants in P- limiting soils and (ii) to investigate the association of seed yield to nodulation among the evaluated bean mutants.

2. MATERIALS AND METHODS

2.1. Germplasm Used

Solwezi (SZ) parental line and its four candidates derived mutants, SZ3-3-B-B2, SZ3-2-B-B1, SZ7-4-B-B and SZ9-B-B-B2 were used. Tembo and Munyinda [7] has described the development of these mutants. Briefly, the mutants were developed by exposing the parent Solwezi to 150 Gray gamma rays followed by an advancing and selection process up to a stable F8 stage.

2.2. Location and Soil Characteristics

The research was conducted at the University of Zambia, screenhouse in Lusaka (15°23'S and 28°25'E, at 1250m above sea level). The P- limiting soil (about 2.93 mg/ kg) used was purposely chosen from a P deprived area. It was then dried in the screenhouse for two days and thereafter sieved using a 2 mm sieve.

2.3. Experimental Design, Layout and Trial Management

The experiment was laid as a 4 (Genotypes) x 3 (levels of Fertilizer) factorial completely randomized design (CRD). The experimental plot was a 15 cm diameter pot filled with 3 kg of soil. Before filling the pots, the soil was thoroughly mixed with an inorganic fertilizer, Triple Super Phosphate (TSP) [46% P_2O_5, 12% Ca, 1% S]. The quantity of TSP added was calculated to achieve the soil P levels at three levels 0 kg/ ha, 30 kg/ ha and 60 kg/ ha P respectively. These were coded as low P (L), medium P (M) and high P (H) respectively. The control (0 kg/ ha P) had no TSP added. In each pot, four seeds were planted which were later thinned to two plants per pot. The plants in a pot were watered weekly. Standard cultural practices such as weeding and irrigation were followed.

2.4. Data Collection and Analysis

Data was collected on nodulation, plant height, shoot dry mass, dry root mass, shoot biomass, pod weight, root biomass, and yield. The data was collected on each and every plant at various growth stages. At harvest, mature pods were first removed from the plants. The bean stem vines were cut from the bottom just below the soil level. After measuring the plant height, the shoot biomass was oven dried at 65 °C for 48 hours. The root biomass was assessed by removing the roots from the flower pots, washing them gently in water to expose the root nodules. Thereafter, the roots were dried in an oven set at 65 °C for 48 hours. The dried shoots and root were then weighed using an electronic beam balance. Prior to drying the roots, root nodules were counted and scored using the modified protocol by CRS [9]. The scores ranged from 1 to 5 with 1 (low count- less than 10 nodules) to 5 (very high- more than 25 nodules).

The analysis of variance was performed on all measured parameters. Correlation analysis was also done for mean grain yield and nodulation. All the data was performed in GenStat statistical software [10] using the General Linear Model (GLM). The analysis was based on the factorial ANOVA model:$Y_{ijk} = \mu + \alpha_i + \beta_j + (\alpha\beta)_{ij} + \varepsilon_{ijk}$ where μ is the overall mean, α_i is the effect of the ith phosphate fertilization level, β_j is the effect of the jth genotype, $(\alpha\beta)_{ij}$ is the combined effect of the ith phosphate fertilization level and jth genotype, and ε_{ijk} is the random error effect.

3. RESULTS

The genotypic main effect was significant ($p< 0.001$) for all the traits studied in Table 1. With phosphorus and interaction main effects, significant variations were obtained on all the traits studied except for vine length and nodulation respectively.

Table-1. Mean squares for analysis of variance for measured parameters in a phosphorous limiting trial evaluated during the 2015/ 2016 cropping season at the University of Zambia, screenhouse.

SOV	DF	VL	NOD	PYHA	SYHA	DRYSB	DRYRB
G	4	460846***	4.42***	30910.4***	25640.2***	16.65***	16.65***
F	2	319.6ns	6.67***	76695.0***	54786.8***	3.49***	3.49***
G x F	8	952.2***	0.22ns	9364.9***	6319.6***	5.98***	5.99***
Error	27	137.9	0.15	619.2	927.6	0.050	0.05

*,**,*** Significant at α = 0.05, 0.01, 0.001 respectively, SOV- Source of variation, F- Fertilizer level, G- Genotype, VL- Vine length, NOD- Nodulation, PYHA- Pod yield (kg/ ha), SYHA- Seed yield (kg/ ha), DRYSB-Dry shoot biomass, DRYRB- Dry root biomass.

Further analysis revealed that SZ3-2-B-B1, SZ3-3-B-B2 and SZ7-4-B-B out-performed the parent in nodulation per plant Table 2. However, only SZ3-3-B-B2 out-performed the parent in both grain yield per hectare and nodulation capacity. There was a general increase in performance across genotypes in all measured parameters with an increase in P- levels Table 3.

Table-2. Means of genotypic parameters' response across fertilizer levels in a phosphorous limiting trial evaluated during the 2015/16 cropping season at the University of Zambia, screenhouse.

Genotype	V.L	NOD	PYHA	SYHA	DRYSB	DRYRB
PARENT	114.6	1.4	381	334	7.08	2.06
SZ3-2-B-B1	82.6	2.7	352	254	3.38	4.34
SZ3-3-B-B2	46.3	2.9	496	400	6.22	5.76
SZ7-4-B-B2	236.7	2.0	375	301	6.07	3.74
SZ9-B-B-B2	116.1	1.3	438	333	5.14	3.29
$LSD_{(\alpha=0.05)}$	21.88	0.7	46.4	56.8	0.28	0.42

V.L- Vine length, NOD- Nodulation, PYHA- Pod yield (kg/ ha), SYHA- Seed yield (kg/ha), DRYSB-Dry shoot biomass, DRYRB- Dry root biomass.

Table-3. Means of parameters' response on fertilizer levels across genotypes in a phosphorus limiting trial evaluated during the 2015/16 season at the University of Zambia, screenhouse.

F.L	V.L	NOD	PYHA	SYHA	DRYSB	DRYRB
0	116.7	1.400	328	256	4.274	4.167
30	116.4	2.067	432	347	6.215	4.063
60	124.6	2.733	465	371	6.245	3.284
$LSD_{(\alpha=0.05)}$		0.5557	35.9	44.0	0.2165	0.3222

F.L- Fertilizer level, V.L- Vine length, NOD- Nodulation, PYHA- Pod yield (kg/ha), SYHA-Grain yield (kg/ha).
DRYSB-Dry shoot biomass, DRYRB- Dry root biomass.
SYHA- Seed yield (kg/ ha).

Further analysis on significant G (Genotype) x F (Fertilizer) interaction Figure 1 for mean yield performance reviewed that the mutant SZ3-3-B-B2 out-performed the parental genotype at both 30 kg P/ ha and 60 Kg/ ha. No significant differences were obtained in mean yield performance when SZ3-3-B-B2 and its parent were compared at an application rate of 0 kg/ ha P. Apparently, the correlation between yield and nodulation across genotypes was low ($r = 0.31$, $P= 0.05$).

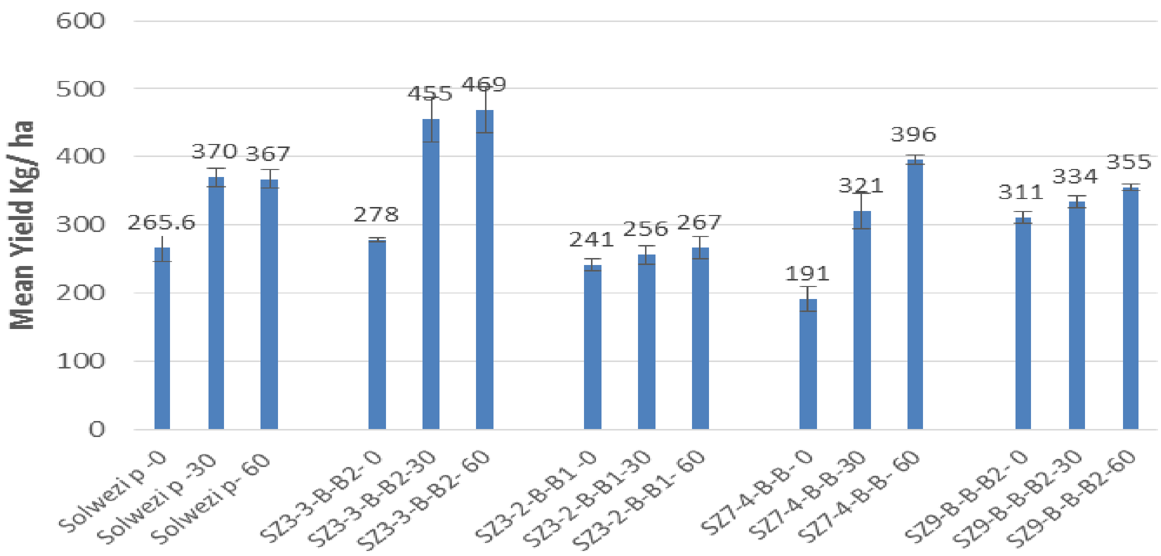

Figure-1. Mean performance of genotypes at different levels of phosphorous.
Experiment conducted at the University of Zambia screenhouse during 2015/16 season Solwezi p- parent; SZ3-3-B-B2, SZ3-2-B-B1, SZ7-4-B-B and SZ9-B-B-B2- Solwezi mutants; 0, 30, 60- fertilizer application rate of 0, 30 and 60 kg/ ha P, respectively; Bars shown- standard error.

4. DISCUSSION

Phosphorous is an important key element in bean production and productivity. Sufficient genetic variability is important for genetic enhancement of BNF and efficient P utilization in common bean. One way of enhancing variation within a bean genotype is through the use of gamma-ray generated mutants, which genotypically differ in one or few traits when compared to parental genotype. This entails the creation of new alleles which become fixed after several generations of selfing. This research focused on identifying high yielding genotypes under P- limiting soils and to investigate the possibility of using artificial inoculum to enhance the productivity of bean mutants. With mutation breeding the key aim is to create mutants which out performs the parental genotype in one or few traits of interest for varietal evaluation and release or as parents in a breeding program [7, 8]. In this study SZ3-3-B-B2 out- performed the parental genotype including other mutants across P- fertilization levels Table 2 in mean nodulation and yield. Further analysis of the interaction between G x F showed that SZ3-3-B-B2 significantly surpassed the parental mutant at a fertilizer rate of 30 Kg P. Additional increase in P did not significantly increase seed yield. This suggests that 30 Kg P was the appropriate rate for the parental mutant and SZ3-3-B-B2. Failure for SZ3-3-B-B2 to perform significantly better than the parent at 0 kg/ ha P implies that phosphorous is key in bean productivity and that the efficiency of P utilization varies among genotypes [4, 11].

The correlation between nodulation and yield was found to be low (r=0.31), giving a low of phenotypic variation explained value (r^2) of 0.10. Previous studies have demonstrated that yield is a function of nitrogen fixation but does not necessarily correlate with a number of nodules [5]. It has been found out that the effectiveness of nodules depends on the quantity of leghemoglobin present in the nodules [12]. Leghemoglobin plays a role in promoting nitrogen fixation and ultimately yield. Since no direct measurements were done to measure the quantity of leghemoglobin, we hypothesis that generally the quantity of leghemoglobin across genotypes was too low to enable a higher correlation factor between nodulation and yield. Previous work has demonstrated that nodule initiation but not necessarily functioning (ability to fix nitrogen) depends on P supply in common bean [5]. Thus, nitrogen fixation in beans depends on both P availability and the bean genotype [13]. Other authors have found that artificial inoculation further enhances nodulation, nitrogen uptake, growth and yield response [14, 15]. From the computed low value of the phenotypic variation explained (r^2= 0.10) in an association between nodulation and yield, there is a likelihood that artificial inoculation can enhance the productivity of the evaluated bean mutants.

5. CONCLUSION

The application of P enhanced the performance of common bean mutants as observed in increased mean performance on all measured parameters as fertilizer levels increased. Amongst the mutants studied, SZ3-3B-B2 was identified as the best performing genotype in utilizing P with regard to seed yield. Therefore SZ3-3-B-B2 can be considered for evaluation for variety release or can be used as a parent in a breeding program. Furthermore, it was revealed that there was a low correlation (r = 0.31) between nodulation and seed yield, giving r^2 value of 0.1. With that revelation, further research using artificial inoculum can be carried out to determine whether inoculation can enhance the performance of these bean mutants.

REFERENCES

[1] M. W. Blair, G. Iriarte, and S. Beebe, "QTL analysis of yield traits in an advanced backcross population derived from a cultivated Andean× wild common bean (Phaseolus vulgaris L.) cross," *Theoretical and Applied Genetics*, vol. 112, pp. 1149-1163, 2006. Available at: https://doi.org/10.1007/s00122-006-0217-2.

[2] W. J. Broughton, G. Hernandez, M. Blair, S. Beebe, P. Gepts, and J. Vanderleyden, "Beans (Phaseolus spp.)–model food legumes," *Plant and Soil*, vol. 252, pp. 55-128, 2003. Available at: https://doi.org/10.1023/a:1024146710611.

[3] V. M. Mwale, J. M. Bokosi, and C. M. Masangano, "Yield performance of Dwarf Bean lines under researcher designed farmer managed system in three Bean Agroecological zones of Malawi," *African Journal of Biotechnology*, vol. 7, pp. 2847-2853, 2008.

[4] E. Otabbong, D. N. Mubiru, and T. Basamba, "Development of common bean (Phaseolus vulgaris l.) production under low soil phosphorus and drought in sub-Saharan africa: A review," *Journal of Sustainable Development*, vol. 7, pp. 1913-9071, 2014. Available at: https://doi.org/10.5539/jsd.v7n5p128.

[5] S. Kouas, N. Labidi, A. Debez, and C. Abdelly, "Effect of P on nodule formation and N fixation in bean," *Agronomy for Sustainable Development*, vol. 25, pp. 389-393, 2005. Available at: https://doi.org/10.1051/agro:2005034.

[6] FIS, "Define biological nitrogen fixation (BNF) and explain its importance Oregon State University. Available: https://forages.oregonstate.edu/. [Accessed 22/ 03/ 2019]," 2019.

[7] L. Tembo and K. Munyinda, "Clustering bean mutants based on heterotic grouping," *African Crop Science Journal*, vol. 1, pp. 1 – 7 2015.

[8] L. Tembo, L. Pungulani, P. H. Sohati, J. C. Mataa, and K. Munyinda, "Resistance to callosobruchus maculatus developed Via Gamma Radiation in Cowpea," *Journal of Agriculture and Crops*, vol. 3, pp. 65-71, 2017.

[9] CRS, *Nodule scoring chart – pulse*. Australia: Murdoch University, 2015.

[10] R. W. Payne, D. A. Murray, R. A. Harding, D. B. Baird, and D. M. Soutars, *An introduction to genstat for windows* 13th ed. Hemel Hempstead, UK: VSN International, 2010.

[11] D. Silva, J. Esteves, J. Gonçalves, C. Azevedo, T. Ribeiro, A. Chiorato, and S. Carbonell, "Evaluation of common bean genotypes for phosphorus use efficiency in Eutrophic Oxisol," *Bragantia*, vol. 75, pp. 152-163, 2016.

[12] C. Hultman, "Abundance of root nodules on common bean, Phaseolus vulgaris – a comparison between Swedish fields with and without a recent history of common bean cultivation," Masters Thesis in Biology. Faculty of Natural Resources and Agricultural Sciences. Department of Soil and Environment, 2018.

[13] B. Adnane, J. J. Drevon, K. Oufdou, B. Mandri, M. Faghire, and C. Ghoulam, "Nodule phosphorus requirement and O2 uptake in common bean genotypes under phosphorus deficiency," *Acta Agriculturae Scandinavica, Section B-Soil & Plant Science*, vol. 61, pp. 602-611, 2011. Available at: https://doi.org/10.1080/09064710.2010.533188.

[14] G. K. Gicharu, N. M. Gitonga, H. Boga, R. C. Cheruiyot, and J. M. Maingi, "Effect of inoculating selected climbing bean cultivars with different rhizobia strains on nitrogen fixation," *Online International Journal of Microbiology Research*, vol. 1, pp. 25- 31, 2013.

[15] V. P. Da Costa Neto, J. B. S. Mendes, A. S. F. De Araújo, F. De Alcântara Neto, A. Bonifacio, and A. C. Rodrigues, "Symbiotic performance, nitrogen flux and growth of lima bean (Phaseolus lunatus L.) varieties inoculated with different indigenous strains of rhizobia," *Symbiosis*, vol. 73, pp. 117-124, 2017. Available at: https://doi.org/10.1007/s13199-017-0475-6.