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Seed Priming and Water Potential Effects on Germination and Emergence of Common Bean (*Phaseolus Vulgaris* L.)

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Abstract

The aim of the study was to evaluate use of on-farm seed priming under artificial drought as a strategy to improve stand establishment in common bean grown under dry-land conditions. A laboratory experiment aimed at determining the effects of different water potentials (0, -10, -100, -200, -500, -1500 kPa) and different seed treatments (non-priming; 12-hour priming; 12-hour priming and 12-hour drying; 12-hour priming and 24-hour drying) on germination of three common bean varieties (PAN329, Contenta, Black bean). A field experiment was also carried out to determine the effects of different priming periods (0; 1; 2; 4; 8; 12; 16; 20 and 24 hours; 12-hour soaking and 12-hour drying; 12-hour soaking and 24-hour drying; 24-hour soaking and 24-hour drying) on emergence of varieties PAN329 and Contenta. The highest germination rate was observed in water potentials of -100 and -200 kPa in PAN329 and Contenta while Black bean achieved higher germination at -500 kPa in the laboratory experiment. In the second experiment, soaking seed between 20 and 24 hours resulted in the highest emergence for PAN329 and Contenta respectively. It can be concluded that where water is limiting seed priming can be recommended to help improve stand establishment.

Key Words: Emergence, stand establishment, water potential, seed priming

1.0 Introduction

Smallholder farmers grow common bean (Phaseolus vulgaris (L.) Merr) in Zimbabwe and other Southern African countries during the wet season from mid-January onwards under rain fed conditions. One of the major constraints to production of common bean is poor stand establishment especially under dry-land conditions (Harris et al., 2001). Reasons for this include soil moisture deficit during the critical period of germination and emergence (Wortmann et al., 1998), low seed quality, inadequate seedbed preparation, untimely sowing, poor sowing technique and adverse soil properties like soil crusting (Harris et al, 2001).

Poor crop stand is caused primarily by poor and non-uniform germination. Germination is the transformation of an embryo within a seed into a seedling (Balasubramaniyan and Palaniapan, 2004). Uniform germination leads to even crop stands and hence higher yields. This is because there will be effective utilization of water, light and soil nutrients. Uneven crop stands lead to poor yield due to direct competition between the different stages of crop growth resulting in the older crops outcompeting younger plants for resources like moisture, light and nutrients and reduced efficiency of utilizing these resources (Harris, 1996). The process of seed germination is a most critical stage in the production of any crop because it influences the subsequent crop stand and hence yield (Almonsourii et al., 2001). Fast germination allows rapid development of seedlings' roots while soil conditions around the seed are still conducive.

Seed imbibition rate and rate of germination generally decrease when water potential of soil is low (Williams and Shaykewich, 1971). During imbibition, a seed absorbs a certain amount of water for it to germinate and this is called hydration and it is species specific (Hadas and Russo, 1974). If there is inadequate water, the first phase of germination leaves the seed with a low water content that will unnecessarily prolong the length of the second phase of breakdown of food reserves thereby delaying onset of the third phase in which the seedling resumes growth and hence delayed germination (Bradford, 1986).

However, uniform germination seldom occurs in rainfed agricultural systems due to unpredictable and erratic rainfall. As a result a lot of research has been done so as to find ways to ensure rapid and uniform germination of seeds in semi-arid conditions and these include scarification on-farm seed priming, which is the soaking of seed in water over night before planting. According to Bradford (1986), priming of seeds is very beneficial to enhance faster germination and emergence of a wide range of crop species. When primed seeds are sown, time between sowing and emergence will be reduced because a lot of processes would have taken place already and uniform germination is usually achieved. This is also important for synchronizing growth when conditions are favorable thereby improving yields (Harris et al., 2001). No extra seed will be required therefore it is economic to the farmer. The farmer will avoid unnecessary debts of buying extra seed to cover for poor germination and emergence.

Research was done previously on priming of different crops and was shown to minimize resowing and the plants were more vigorous. Surveys done on priming indicated that a 22 % yield increase was achieved in cereals like maize (Zea mays) in Zimbabwe and India as well as a 37 %vield increase in wheat (Triticum aesitvum) in India and Pakistan (Harris et al, 2001). When priming of soybeans was done it was recommended that priming can be useful where the water potential is low enough to limit emergence though soybean largely responded negatively to priming at higher to medium water potential (Murungu et al., 2005). In another study with cotton (Gossypium hirstum), seed priming resulted in depressed emergence percentages indicating the need to determine the responses of various crop species to priming (Murungu et al., 2004). Research from Centre for Arid Zone Studies (CAZS), in Wales, produced maximum lengths of time for which seeds are to be primed and these vary from crop to crop (Harris et al., 2004). Over-soaking of seed might be as good as not soaking or even worse, because the testa will burst due to over-imbibition. But for the process of priming to be successful farmers have to dehydrate the seeds before sowing so as to reduce the risk of damage to the testa and to make handling of seeds easier. The surface dried seeds can be stored for a long time after priming even up to one year (Harris et al., 2001). But at the same time, the post-priming duration can affect the performance of the primed seeds.

Although a lot of work had been done on priming of other crops, very little has been done on common bean. Mannitol and other Polyethylene Glycerols (PEGS) have been used to impose water stress on plants and seeds by decreasing the osmotic potential of the growing medium without interferences from soil physical conditions such as soil clods, temperature and crusting (Muchena and Grogan 1977). In this study mannitol will be used to help understand how bean seeds may respond to physical conditions at planting. Murungu et al., (2005) used a similar approach using mannitol to study how different varieties of soybean respond to low water potentials. This study therefore, intends to determine the effect of priming on germination and emergence of common beans as well as to establish the most ideal water potential and

priming and post-priming duration to improve stand establishment in common bean under simulated conditions and under field conditions.

2.0 Materials and Methods

Two experiments were conducted at Midlands State University in Gweru, Zimbabwe between April and June 2008. Experiment 1, was carried out to determine the effect of water potential and seed priming on germination of common bean varieties. Experiment 2, determined the optimum hours for soaking common bean in water and the effects of delaying sowing after priming the seeds on emergence.

Description of varieties used

Three varieties, PAN 329, Contenta and a black bean landrace were used in this trial. PAN 329 variety is a cream speckled, kidney-shaped sugar bean type variety which is medium sized. Contenta is a large-sized variety that is red kidney bean variety. The black bean landrace is a small-sized variety that is black in colour, kidney-shaped variety.

Seed germination tests

Germination tests were carried out for the three common bean varieties under standard conditions. Eight sub-samples of 50 seeds each were placed in moist and rolled paper towels. The paper was then blind folded on the middle and the seeds were completely covered by the lower half of blotter paper. The papers were placed in an incubator at 25 °C. Number of seeds that germinated was determined after 7 days.

Experiment 1: Water potential, priming period and variety effect on common bean germination

A 6 x 3 x 4 factorial laboratory experiment was laid out in a Completely Randomised Design (CRD) with three (3) replicates. The experiment was conducted using 6 different water potentials (0, - 10, - 100, - 200, - 500, -1500 kPa), three varieties (PAN 329, Contenta and Black bean) and four seed priming treatments (non-priming; 12-hr priming; 12-hr priming and 12-hr drying; 12-hr priming and 24-hr drying). The seeds were first primed by soaking the seeds in tap water and surface dried by blotting with a cloth. Seed soaking and drying were staggered to ensure that all seeds were incubated at the same time. Drying for the periods stipulated above was done at room temperature. Mannitol was used to effect the different water potentials that were used and distilled water was used as a control (0 kPa). Mannitol is an inert non-electrolyte chemical. After the priming and drying period seeds were placed in Petri dishes containing different water potentials (effected by Mannitol) and then incubated at a temperature of around 25 oC for eight (8) days. A total of 144 Petri dishes were incubated. The concentration of the different water potentials were obtained using the following formula suggested by Thill, et al., (1979):

g = PVm / RT (1)

Where g = grams of solute;

P = osmotic pressure

V = volume in litres

m = molecular weight of chemical used (Mannitol) R = 0.0825 atmospheres per degree per mole T = absolute temperature

Seed germination was assessed every morning at 1000 hours for a continuous period of eight (8) days. Seeds were considered to have germinated when a shoot of 5 mm had developed. Germinated seeds were counted and recorded to calculate germination rate and final percentage germination. The following formula was used to calculate the germination rate:

$$G = X/Y + (X2 - X1)/Y2 + \dots (Xn - Xn-1)/Yn, \quad (2)$$

where G = germination rate, X = percent seedling germination on the first count, Y = number of counts from planting to the first count, X2 = percentage seedling germination on the second count, Y2 = counts from planting to second cound, Xn = percent seedling emergence on the nth count and Yn = number of counts from planting to the nth count (Maguire, 1962 ; Murungu et al., 2005).

Experiment 2: Effects of different priming period and different postpriming drying periods on emergence of common beans

A field experiment was carried out to determine the effects of post-priming duration on emergence of common beans. The experiment was a 13×2 factorial laid out in a split-plot design with variety as the main plot factor and priming period as the sub-plot factor. The seed material used in the experiment was first carried out in the

laboratory where, 100 seeds of each of two (2) different common bean varieties (PAN 329 and Contenta) were soaked in tap water for different periods in 500 ml beakers at room temperature. Seeds were soaked for varying periods which are: 0, 1, 2, 4, 8, 12, 16, 20, and 24 hours, 12 hours + 12 hours drying, 12 hours + 24 hours drying, 24 hours + 12 hours drying and 24 hours + 24 hours drying. Soaked seeds were then ferried to the field and sown in 1 m² plots. Soaking was staggered so that sowing was done at the same time. The seeds were sown in rows 0.45 m apart and 0.07 m between seeds and the plots were watered after sowing and left for eight days. The total area sown was 140 m².

Data collection on emergence was done from sowing. Seedlings visible above the soil surface (about 20 mm) were considered to have emerged and these were counted and recorded every day at 0900 hours for a continuous period of 10 days. The recorded data was used to calculate the rate of emergence and the percentage emergence. Rate of emergence and percentage emergence were calculated using the above formulas.

Data analysis

Analysis of variance (ANOVA) was done using Genstat Discovery Edition (Version 3) and means were separated using the Least Significant Difference (L.S.D.) method.

3.0 Results

Experiment 1: Effects of water potential and priming duration on germination of three common beans varieties

Germination tests

From the germination tests carried out on the seeds, 90 % of PAN 329 variety germinated, 81 % for Contenta and 73 % for Black bean. The germination percentages were varied with Black bean achieving the lowest germination percentage.

Effects on germination rate

There was no significant interaction (P>0.005) between the three main factors of water potential, priming duration and variety throughout the study. There was also no significant interaction (p>0.05) between variety and priming period as well as between priming period and water potential. However, there was significant interaction (p<0.001) between variety and water potential on germination rate common beans (Figure 1). of Germination percentage was highest at -200 kPa and -100 kPa for Contenta and PAN 329. Black bean showed the highest germination rate at -500 kPa and lowest germination rate at -100 kPa. Overally Black bean had the lowest germination rate compared to the other two varieties. The two main factors of variety and water potential also showed significant differences (P<0.001) on germination rate, whereas the priming duration main factor did not show any significant difference (P>0.05).

Figure 1: Effect of variety and water potential on germination rate of common beans. Error bars represent the standard error of the difference of means (71 d.f) Midlands State University Journal of Science, Agriculture and Technology Volume 3(1), 2012



Effects on germination percentage

There was no significant interaction (P>0.005) between the three main factors of variety, water potential and priming duration on germination percentage. There was however, significant interaction between variety and water potential on germination percentage. Germination percentage increased from 0 kpa to – 100 kpa and started falling after -200 kpa. The highest germination percentage was achieved at – 100 Kpa by PAN 329 and Contenta, whereas the variety Black bean achieved highest germination percentage at -500 Kpa (Figure 2). There was no significant interaction between variety and priming duration as well as between priming duration and variety. There was significant differences (P<0.001) between variety and water potential main effects on germination percentage due to priming duration.

Figure 2: Effect of variety and water potential on germination percentage of common bean. Error bars represent the standard error of the difference of means (71 d.f).



Experiment 2: Effect of priming period on field emergence rate and percentage of two common bean varieties

Effect on field final emergence rate under field conditions

There was a significant interaction (p<0.005) between variety and priming period on emergence rate of the two common beans varieties (Figure 3). There was an increase in emergence rate with an increase in the duration of priming with highest emergence rate being achieved by Contenta after being soaked for 20 hours while PAN 329 achieved the highest emergence rate after being soaked for 24 hours. There was decreased emergence rate due to drying when compared to the priming duration that resulted in the highest emergence rate by the two varieties.

Figure 3: Effect of priming duration on the emergence rate of common beans under field conditions. Error bars represent the standard error of the difference of means (48 d.f)



Effect on emergence percentage under field conditions

There was significant interaction (P<0.001) between variety and priming duration on emergence percentage of common bean (Figure 4). There was an increase in the emergence percentage of common bean as the priming duration was increased. The highest emergence percentage was obtained when the variety Contenta was primed for 20 hours and that of PAN 329 was achieved

after beans were soaked for 24 hours. Soaking beans for between 12 and 24 hours and then drying them achieved higher emergence percentage compared to beans soaked for between 0 and 16 hours for between the two varieties.

Figure 4: Effect of priming period on emergence of two common beans varieties. Error bars represent the standard error of the difference of means (48 d.f)





4.0 Discussion

Germination of three common bean varieties at different water potentials There was good germination percentage and rate at water potential of -100 kPa and -200 kPa in Contenta and PAN 329 while the highest germination percentage and rate was obtained at -500 kPa for black bean variety. The differences in germination rates between the three varieties reflect their differences in seed size with the smaller seeded Black bean performing well at lower moisture potential. The high performance of the smaller seeded variety at low water potential may confer an advantage to small sized seed when planted in drier soils. These findings are consistent with work found in soybean where small sized seed achieved higher stand counts than larger sized seed (Madanzi, 2004).

At high water potential of 0 kPa and – 10 kPa in PAN 329 and Contenta resulted in low germination and this may be attributed to too much availability of moisture which in turn caused leaking of solutes required for germination out of the seed or removal of growth promoters on the testa (Murungu et al, 2005). Low water potentials of –500 kPa and –1500 kPa gave low germination rates in PAN 329 and Contenta. At these water potentials, the solute potential is very high such that less water moves into the seed to promote germination. Murungu et al., (2004) produced almost similar results in cotton and maize.

Percentage germination was different in all the three varieties with PAN 329 achieving the highest germination percentage followed by Contenta while Black bean achieved the lowest. These differences might be caused by genetic differences between these varieties and also due to environmental factors like the way in which the seeds were produced, seed longevity and storage. Seed size might also be one of the factors causing the differences in performance of the three varieties as it influenced moisture required for seed germination. PAN 329 and Contenta have relatively large seed sizes compared to black bean therefore water imbibition is high due to increased surface area for absorption of water in these large seed-sized varieties. Muchena and Grogan, (1977) observed that the quantity of water uptake by a seed depends on seed size and also on the hydrability of the seed contents. Therefore low germination rate in black bean may be attributed to its small sized seeds and also on the hydrability of the contents of the seeds.

There was no difference (P>0.05) on germination percentage or rate due to priming period in the laboratory experiment. This may be attributed to immediate triggering of the germination process once the seed starts to imbibe water even before the seed reached its imbibition saturation point. It can also be attributed to the fact that the after drying period did not result in reversal of the germination process due to loss of moisture. These results differ from the ones obtained in soybean where germination decreased with increased soaking duration probably due to leakage of solutes (Murungu, et al., 2005). Although the need for drying in common beans was necessitated by fears in damage of the seed coat if the seed was sown immediately after soaking, observations were that common beans have a fairly stronger seed coat compared to soybean.

Optimum seed priming period and post priming drying duration on emergence of common beans. From the field experiment (experiment 2) it was shown that seeds soaked for 20 and 24 hours had the highest final emergence

percentage. The rate of emergence was also more that double that of dry seed confirming results by other researchers who found out that priming reduces the time seed emerges from the soil in the field (Murungu et al., 2004; Harris at al., 2001). This suggests that these soaking periods are the optimum periods for seed priming. Under these conditions the first phase of germination would have begun and continued soaking would produce adverse effects due to leaking of solutes important for germination (Shereena and Salim, 2006) and washing away of important fungicides hence emergence will be lowered. On-farm seed priming trials in sorghum conducted by Harris et al., (1999) reported that priming seeds for about 24 hours decreased time to 50 % emergence by about 12 hours. The highest emergence rate was achieved by PAN 329, confirming the results found in the standard germinations test where the variety achieved 90 % germination probably indicating that it had higher vigor compared to Contenta. Soaking for a short period of time is not sufficient to trigger germination and over-soaking (above 24 hours) can lead to very low emergence therefore the optimum priming period is between 20 and 24 hours. Owing to their sizes and chemical composition, the two common varieties require more time to absorb moisture to achieve optimum germination compared to maize which requires around 8 hours (Murungu et al., 2004) and soybeans which requires about 16 hours (Murungu et al., 2005). Although treatments which were subjected to post priming storage achieved lower germination percentage and rate, than seed soaked for 20 hours

without post storage, they performed better than bean seed soaked for less than 20 hours indicating that in varieties prone to cracking due to soaking, reasonable emergence rates may be achieved with drying the seed to improve handling despite the fact that the varieties used in this study had stronger seed coats. Similar results were found in soybean where post priming storage resulted in better emergence rate than some treatments where soybean seed was soaked without drying particularly soaking soybean for between 16 to 20 hours (<u>Murungu et al., 200</u>5).

5.0 Conclusions and Recommendations

Seed priming improved the germination and emergence of PAN 329 and Contenta in the laboratory and in the field respectively. Placing primed seeds in water potentials of -100 kPa and -200 kPa gave the highest rate of germination and soaking in the laboratory. Therefore priming of common bean might only be recommended if the soil water potential is low enough to limit emergence but not under severe moisture deficit and soaking should be done 20 to 24 hours before sowing thus on-farm seed priming can be practiced to improve emergence of common bean. Primed seeds should not be left to dry for a long time before sowing since their emergence will be reduced. Generally primed seed had faster emergence rate than unprimed seed indicating that seed priming can be used to increase the rate of seed emergence in the field.

It is advised that priming of common beans be done in those varieties with large sized seeds like Contenta and PAN 329. Small-sized varieties like black bean should not be primed since they do not respond positively to priming.

Further studies need to be done using more common bean varieties so that a composite conclusion can be drawn. Also, studies need to be done on how solute leakages can be minimized in higher water potentials. Future studies can also look into effects of priming on agronomic characteristics such as days to flowering, days to physiological maturity and grain yield.

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