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Full Length Research Paper

Effects of seed priming and water potential on seed germination and emergence of wheat (*Triticum aestivum* L.) varieties in laboratory assays and in the field

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Poor crop establishment is a major problem in wheat production due to low soil moisture. Two experiments were undertaken to determine the effects of seed priming on seed germination and seedling emergence of wheat varieties. The first experiment determined the effects of water potentials (0, -0.01, -0.1, -0.2, -0.5 and -1.5 MPa), seed hydro-priming treatments including non-primed, primed (without seed drying), primed and 12 h drying on seed germination of wheat varieties. The second experiment determined the optimum seed soaking duration in wheat for maximum emergence. The factors used were seed treatment (soaking for 0, 1, 2, 4, 8, 12, 16, 20 and 24 h and 12 h soaking and 12 h drying) and variety. In the first experiment, there was a significant (P < 0.01) interaction between seed treatment and water potentials. Seed germination percent for all the treatments decreased as water potentials decreased. The non-primed seeds had the greatest decrease in germination percent as water potential was lowered. At low water potentials (-0.2 and -0.5 MPa), priming improved the germination of all the varieties. In the second experiment, there was a significant (P < 0.05) seed treatment effect on wheat emergence. Priming resulted in an increased final percent emergence and lower time to 50% emergence when compared with the non-soaked seed. It was concluded that priming wheat seed up to 12 h may be recommended were soil water potential is low enough to limit emergence. Delay of planting after soaking for 12 h did not affect the final emergence.

Key words: Seed priming, wheat, water potential, germination rate.

INTRODUCTION

Low wheat (*Triticum aestivum*) yields experienced by resource limited small-scale farmers in tropical low altitude (<500 m.a.s.l) areas of Zimbabwe are the result of short winter seasons, late planting, lack of inputs and poor stands (Murungu and Madanzi, 2010). Good crop establishment is an essential pre-requisite for the efficient use of resources such as water and light, hence plant stand is a major determinant of yield (Parera and Cantiliffe, 1999; Witcombe and Harris, 1997). Finch-Savage et al. (2004) showed that conditions after sowing had a large influence on the emergence and seedling vigour in sorghum and argued that speed of germination and emergence was an important determinant of successful establishment. Rapidly germinating seedlings could emerge and produce deep root systems before the upper layers of the soil dry out, hardened or become dangerously hot.

Recently, there has been renewed interest in seed priming (also known as hydro-priming) to improve establishment (Finch-Savage et al., 2004; Harris, 1996). This approach consists of soaking seeds in water (usually overnight), surface drying and planting the same day. This decreases the time that the seed spends in the seedbed simply imbibing water. Once sown, seeds spend significant amounts of time just absorbing water from the soil. So, by reducing the imbibition time to minimum (through seed priming), germination rate of seed can be increased and seedlings emergence improved (Hartman et al., 2002).

In recent years, seed priming has been tested in over

1000 trials in India, Pakistan, Nepal, Bangladesh and Zimbabwe on a range of crops including maize (*Zea mays*), sorghum (*Sorghum bicolor*), rice (*Oryza sativa*), wheat and Chickpea (*Cicer arietinum*) (Harris et al., 1999, 2001). Smallholder farmers in these areas reported that emergence was faster, less re-sowing was required and that plants were more vigorous. In wheat, mean yield increases in six large on-farm trials of 5% have been reported in Nepal (Harris et al., 2001). This approach is termed 'on-farm seed priming" to distinguish it from the energy-intensive, high technology seed hardening or seed conditioning process available to farmers in high input temperate agriculture and horticulture (Hardgree, 1996; Parera and Cantliffe, 1999)

The use of on-farm seed priming in wheat production among smallholder farmers is relatively unknown. Preliminary work on seed priming with other crops, for example pigeon pea *(Cajanas cajan*), cowpea (*Vigna unguiculata*) and cotton *(Gossypium hirstutum*) has confirmed the utility of identifying adverse reactions to priming conditions (Finch-Savage et al., 2004; Harris et al., 1999). In this study, *in vitro* germination studies were undertaken to determine the response of wheat to seed priming. *In vitro* studies are useful for initial screening to avoid detailed and expensive field studies. Seed priming has been shown to result in depressed emergence percentages in cotton, implying that different crop species differ in their response to priming (Murungu et al., 2004).

Polyethylene glycols (PEG) and mannitol have been used by several investigators to impose water stress on plants and seeds, by decreasing the osmotic potential of the growing medium (Murungu et al., 2005; Lawlor, 1970). This technique has the advantage that the water potential of the medium can be controlled independently of other physical conditions such as soil crusting or temperature. This can help one understand how seeds may respond to physical conditions at planting. Muchena and Grogan (1977) used a similar approach (with mannitol instead of PEG) to study how different sized maize seeds respond to low water potentials. They observed greater final percent germination of small seeds than large seeds at low water potentials. They suggested that this was because smaller seeds required less water for germination owing to their smaller volume.

For many small-scale farmers in the semi-arid tropics, severe labour constrains during the peak planting period may hinder the sowing of wheat seed after soaking, resulting in seeds drying or delayed planting of the primed seed. Wheat seed is usually expensive and in short supply and farmers are unwilling to discard primed seed that would have dried up. It was therefore important to investigate the effects of unforeseen delays in sowing after seed priming. It has been demonstrated (Murungu et al., 2003) that while the water imbibed by the seed during seed soaking can induce germination, it may not result in emergence of maize seed. Therefore, the water imbibed by the seed during soaking needs to be augmented by soil moisture in the seedbed to ensure optimal emergence.

This study was carried out to better understand the response of wheat to seed priming. The interaction of wheat varieties and water potentials with different priming periods was also investigated. Effects of delays in sowing after seed soaking on germination and emergence were also estimated. It was hypothesized that seed priming will result in high emergence percentages.

MATERIALS AND METHODS

Two experiments involving wheat were conducted at Midlands State University, Zimbabwe. Experiment 1 was carried out to determine the effect of water potential and seed priming on the germination of wheat varieties. Experiment 2 investigated the optimum hours for soaking wheat in the water and the effects of delaying sowing after priming the seeds on emergence.

Seed germination tests

Germination of four wheat varieties (Dande, Insiza, Kana and S95063) was determined under standard conditions using rolled paper towels as described later. Eight sub-replicates of 50 seeds each were folded and placed in the middle and seeds were covered completely with the lower half and the paper were rolled and placed in an incubator at 25°C. The number of seeds that germinated was determined 7 days later.

Experiment 1: Water potential, seed treatment and variety effect on wheat germination

A laboratory experiment using 4 varieties (Dande, Insiza, Kana and S95063), 6 different water potentials (0, -0.1, -0.1, -0.2, -0.5 and -1.5 MPa) and 4 seed treatments (primed, non-primed and primed seed dried for 12 and 24 h after soaking) was conducted using a 4 x 6 x 4 factorial laid out in a randomized complete block design (RCBD) with 3 replicates. Seed was primed by soaking in tap water for 12 h and then surface dried by blotting with a cloth. Seed soaking was staggered so that all seeds were incubated at the same time. Drying of the soaked seeds was done at room temperature (25°C). Ten seeds were placed in each Petri dish lined with filter paper. Treatments were applied randomly to Petri dishes. Mannitol was used to effect the different water potentials, while distilled water was applied for the control (0 MPa). Mannitol is considered to be an inert non-electrolyte. The concentrations of the solution were obtained from the formula suggested by Thill et al. (1979):

$$g = PVm / RT$$

Where, g = gram of solute, P = osmotic pressure, V = volume in litres, m = molecular weight of chemical used, R = 0.0825 atmospheres per degree per mole and T = absolute temperature.

Each Petri dish was filled with 12 ml of mannitol solution to give the respective water potentials. The dishes were incubated at 25°C for 8 days. Seed germination was assessed at 10 a.m. every morning. Seeds were considered to have germinated when a shoot of 5 mm had developed. Germinated seeds were counted and recorded to calculate the germination rate and final percent germination. The following formula was used to calculate the germination rate:

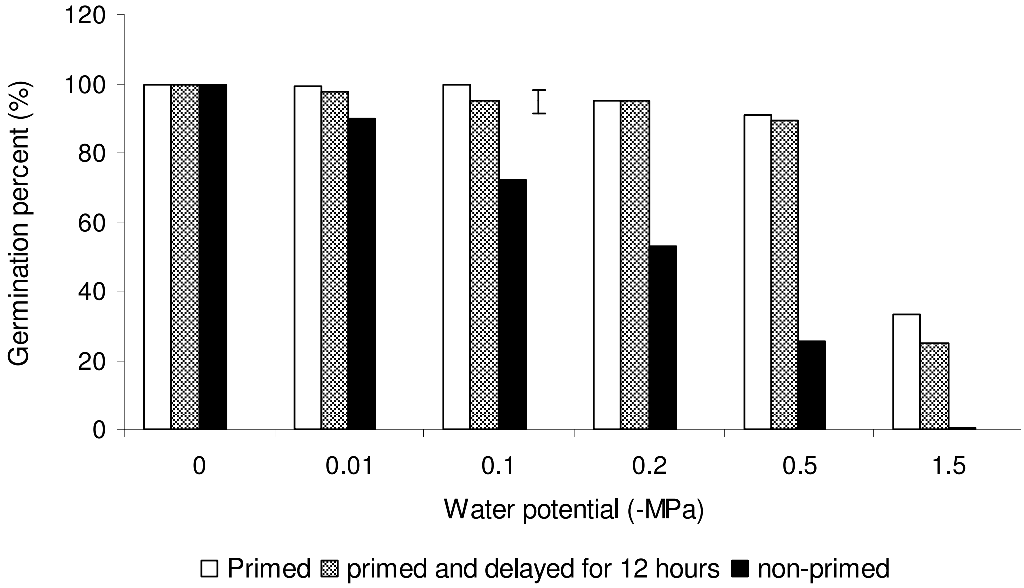

Figure 1. Wheat seed germination percent after different seed treatments at various water potentials. Error bars represent the LSD; means across variety are presented.

$$G = X / Y + (X_2 – X) / Y_2 + ... (X_n – X_n – 1) /Y_n$$

Where, g = germination rate, X = percent seedling germination on the first count, Y = counts from planting to the first count, X_2 = percent seedling germination on the second count, Y_2 = counts from planting to second count, X_n = percent seedling emergence at n^{th} count and Y_n = counts from planting to n^{th} count (Maguire, 1962; Lusembo et al., 1995). The data were subjected to analysis of variance.

Experiment 2: Effects of different soaking times and drying periods on wheat emergence

Two trials were carried out, one in the laboratory and the other in the field. In the laboratory trial, samples of 100 wheat seed were soaked in tap water in 500 ml beakers at room temperature of about 25°C. This method of seed priming is common in smallholder farms in the south-eastern parts of Zimbabwe (Finch-Savage et al., 2004). Factors used were nine seed treatments (priming for 0, 1, 2, 4, 8, 12, 16, 20 and 24 h) and four varieties (Dande, Insiza, Kana and S95063). The 9 x 4 factorial experiment was laid out in an RCBD with three replicates. The weight of each sample, before and after soaking, was recorded using a scientific analytic scale (Avery Bekerel Model).

In the field experiment, seed treatments from the laboratory experiment were used for sowing but with additional seed treatments of delayed sowing for 12 and 24 h after 12 and 24 h of soaking, respectively. Thus, the treatments were as follows: priming for 0, 1, 2, 4, 8, 12, 16, 20 and 24 h, 12 h soaking + 12 h drying, 12 h soaking + 24 h drying, 24 h soaking + 12 h drying and 24 h soaking + 24 h drying. No seed germinated after application of the earlier mentioned treatments. Soaking times were staggered so that seeds were sown at the same time. The experimental design was a 13 x 4 factorial laid out in an RCBD with three replications. The plots were watered five days after sowing. The plots measured 1 x 1 m and inter-row spacing was 0.25 m. Small trenches, 20 to 30 mm deep, were opened using hand hoes and 100 seeds were planted per row. The study area had a sandy-loam soil and day-time temperatures averaged at 23.3°C, while night temperatures averaged at 17.2°C during the course of the experiment.

Seedlings visible above the soil surface were considered to have emerged and these were counted and recorded for ten days. Seedling emergence was recorded each day in the morning at 8 a.m. After ten days, the seedlings were uprooted for dry mass determination. The seedlings from each plot were placed in the oven for four days at 75°C and then were removed and weighed.

For the different soaking hours experiment, weight increase for each seed treatment was expressed as a percent of the dry weight of the seeds. The time to 50% emergence ($t_{0.5}$) was estimated from cumulative emergence curves by noting the time when emergence crossed the 50% emergence line on a graph. The $t_{0.5}$ value was based on the total number of seeds that emerged from each treatment, and not the total number sown (Mabika, 1994). Weight increase, $t_{0.5}$, final emergence and dry weight measurements were subjected to analysis of variance (ANOVA) using the Genstat Discovery statistical package.

RESULTS

Germination tests

From the standard germination tests carried out on the seeds, 100% of SC95063, 100% of Kana, 99.5% of Insiza and 99.3% of Dande germinated.

Wheat final germination percent and rate of germination

Water potential and seed treatment significantly ($p < 0.01$) affected the final germination percentage. There was a significant ($p < 0.01$) interaction between the seed treatment and water potential on the final germination. Germination percent for all the treatments decreased as

Table 1. Germination rate of the different wheat varieties. Means across water potentials and seed treatments are presented.

Variety	Germination rate (number/day)
Insiza	59.3
S95063	56.8
Dande	64.9
Kana	58.8
LSD	3.1

water potentials decreased. However, non-primed seeds had the greatest decrease in germination percent as water potential was lowered when compared to the primed seed (Figure 1). Non-primed seeds were not able to germinate at -1.5 MPa. There was no significant ($p > 0.05$) difference on the final germination percent for non-primed seed at 0 and -0.1 MPa (Figure 1).

Seed treatment, variety and water potential all had significant ($P < 0.01$) effects on the germination rate of wheat. Variety Insiza showed the lowest germination rate, while Dande had the highest germination rate (Table 1). Germination rate decreased as the potential was lowered for all the seed treatments. There was a significant water potential × seed treatment interaction such that the germination rate of non-primed seed was much more sensitive to low water potential than the primed seeds (Figure 1). Primed and delayed seeds for 12 h had the highest germination rate at all the water potentials. As water potential was lowered, decrease in the germination rate was much greater for non-primed seed when compared to the other seed treatments (Figure 2).

Effects of soaking duration on wheat seed weight increase

Soaking time had significant ($P < 0.01$) effects on weight increase, while variety had no significant ($P > 0.05$) effects on weight increase. As the hours of soaking increased, the weight increased for all the wheat varieties. Seed weight increased by about 70% at 12 h of soaking and stabilized at 90% after 20 h of soaking (Figure 3).

Effect of soaking duration on wheat emergence

Seed soaking duration significantly ($P < 0.05$) affected the emergence, while variety did not significantly ($P > 0.05$) affect emergence. The seed treatment × variety interaction was also not significant ($P > 0.05$). As soaking duration was increased, it resulted in a decline in percent emergence. Soaking wheat seed for more than 12 h resulted in an emergence percent lower than 70% with only 55% emergence after 24 h of soaking (Figure 4).

Effect of soaking period on time to 50% ($t_{0.5}$) emergence and dry mass

Significant effects ($P < 0.05$) on $t_{0.5}$ but not on dry weight of various seed treatments were recorded. Interaction of variety and seed treatment was not significant ($P > 0.05$). Non-primed seeds took much time to reach $t_{0.5}$ than seeds which were primed (Table 2). The seeds which were soaked and delayed for 12 h had the least number of days to reach $t_{0.5}$.

DISCUSSION

Seed priming improved germination percent at -0.2, -0.5 and at -1.5 MPa. At these water potentials, primed seeds had a higher percent germination than non-primed seeds. Non-primed seeds faced moisture stress conditions at lower water potentials because seed moisture was lost to the highly concentrated germination media, which is consistent with previous findings (Murungu et al., 2004). The greater final percent germination of primed wheat seeds at low water potential may potentially confer it with an advantage when planted in drier seedbeds. At 0 and -0.1 MPa, there was little difference in the germination percent for all the seed treatments, presumably because water availability for seed uptake was not limiting.

Generally, germination rate decreased as water potentials decreased, but it was more pronounced in non-primed seeds. It is probable that as the water potential was lowered, water from seeds was lost to the germination media by osmosis. The differences in germination rate among the wheat varieties may be due to genetic factors, although it is possible that differences in seed quality resulting from environmental factors (differences in temperature and humidity) may also be responsible.

With increase in soaking duration, seeds imbibed water up to 20 h for all the varieties and there was no further seed weight increase beyond 20 h of soaking. This may be due to the fact that the seed had reached its imbibition saturation point. From the field investigations, seed soaked for 20 and 24 h had the lowest final percent emergence. Solutes required for germination may have been leached out of the seed during the long hours of

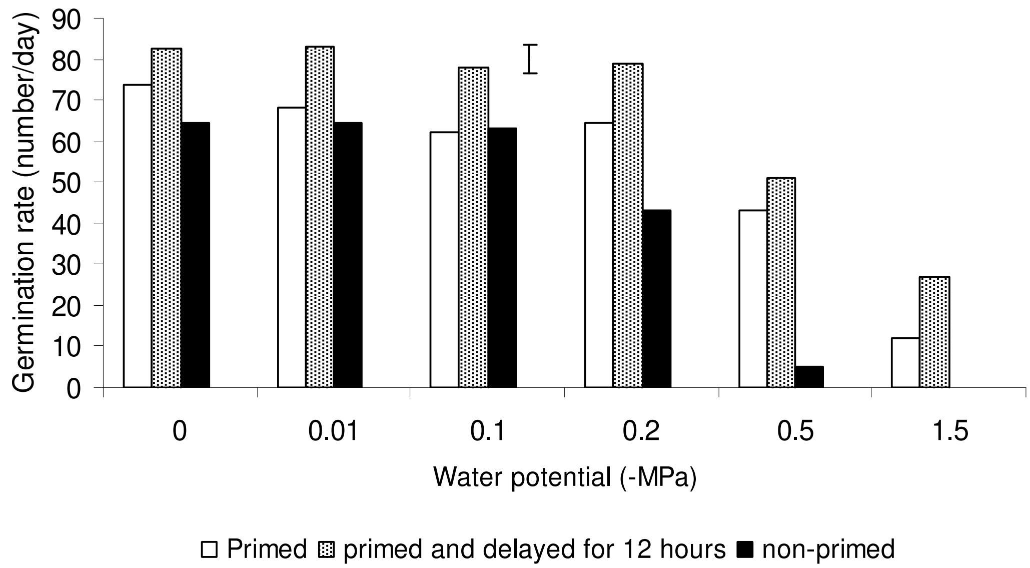

Figure 2. Germination rate of different seed treatments at various water potentials. Error bars represent the LSD; means across varieties are presented.

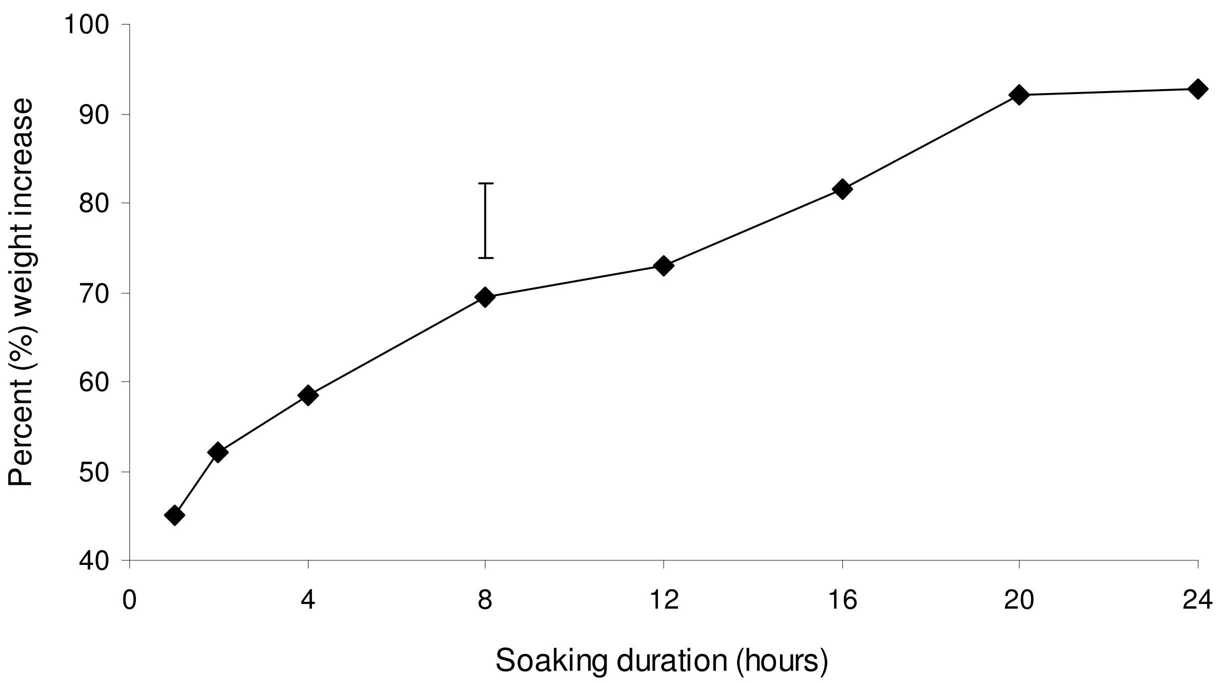

Figure 3. Seed moisture content of wheat seeds after soaking in distilled water for various hours. Means across varieties are presented. Error bar represents the LSD.

soaking (Rahoui et al., 2010). Another reason could be that soaking for longer periods may have resulted in excess water been trapped between cotyledons causing the embryo to suffocate due to lack of oxygen (Finch-Savage et al., 2004). Furthermore, excess water uptake might have led to physiological seed fracturing.

Murungu et al. (2004) reported that seed priming had adverse effects on cottonseed emergence in the field, while laboratory experiment showed priming enhancing germination of cottonseed. The discrepancy between primed and non-primed cottonseed germination in the laboratory and field was explained by the vast difference between polyethylene glycol (PEG) environment in the laboratory and field seedbed environment. Solute leakage

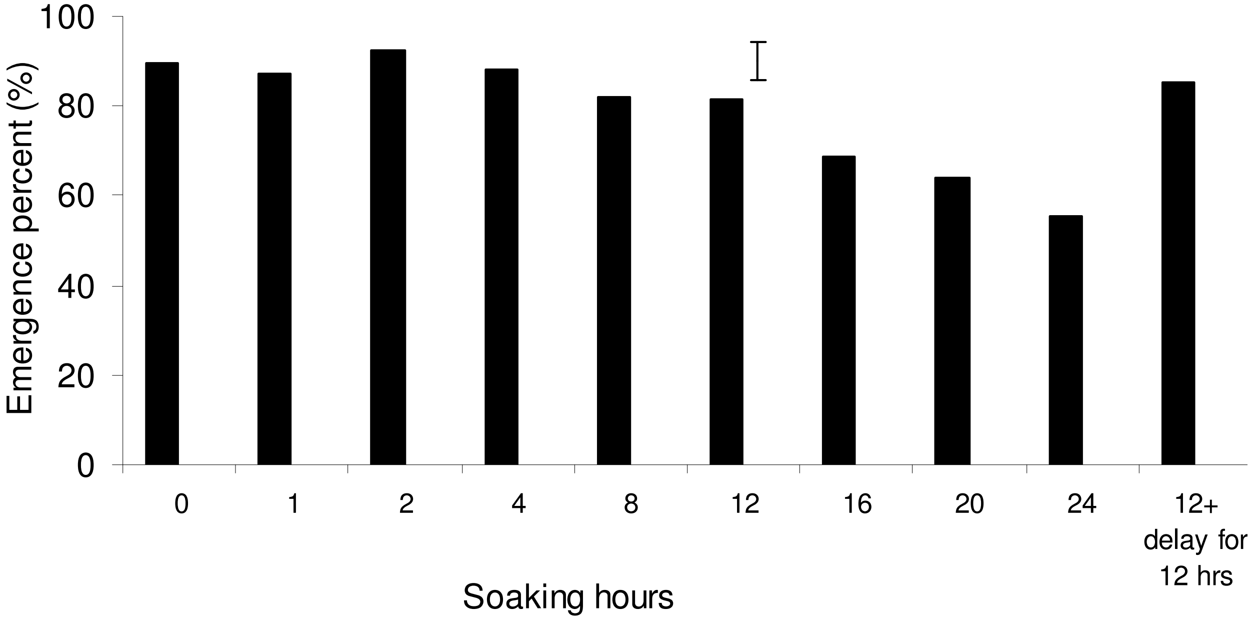

Figure 4. Effects of soaking period on emergence of wheat. Error bar represents the LSD; means across variety are presented.

Table 2. Effect of soaking duration on time to 50% emergence ($t_{0.5}$) and dry mass of wheat.

Soaking duration (h)	Time to 50% emergence (days)	Dry mass (g/plant)
0	5.545	3.23
1	4.21	2.53
2	4.26	2.46
4	4.69	2.25
8	3.64	2.64
12	3.91	2.31
16	4.15	2.85
20	3.82	2.25
24	3.85	2.82
12 + delay of 12 h	3.18	3.29
LSD	0.82	ns

Means across variety are presented.

during soaking (Hegarty, 1978) could have limited the primed cottonseed emergence under field conditions. This difference may also be due to the fact that, in the field, soil moisture was not limiting since the plots were watered regularly. In the laboratory experiment, water was a limiting factor at low water potential, and resulted in high germination percent for the primed seeds. This highlights the need to supplement laboratory results with actual field experiments.

Priming reduced the time to 50% emergence. The primed seeds spent less time imbibing water from the soil. For germination to begin, seeds have to imbibe water from the surrounding soil. The primed seeds had sufficient moisture to start the germination process unlike the un-primed seed that required time to imbibe moisture from the soil. Murungu et al. (2003) recorded similar results where priming lowered time to 50% emergence from 3.9 to 2.9 days in cotton. Hardgree (1996) also discovered that priming native perennial grasses reduced the time to 50% germination by 4.8 days. Also Harris et al. (1999) reported that priming decreased time to 50% emergence by about 12 h in on-farm seed trials of priming in sorghum.

Conclusions

At low water potentials (-0.2 and -0.5 MPa), seed priming improved the germination of wheat. Delaying sowing by 12 h after priming, gave high germination percent at most of the water potentials in the laboratory experiment. Priming also resulted in higher emergence under the field conditions, reducing time to 50% emergence. It was concluded that priming wheat seed up to 12 h may be recommended where soil water potential is low enough to limit emergence. Delaying planting after soaking by 12 h did not affect final emergence.

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