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DOI: <https://dx.doi.org/10.4314/acsj.v28i2.3>



EFFECT OF PRIMING REGIMES ON SEED GERMINATION OF FIELD CROPS

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(Received 19 December 2019; accepted 22 June 2020)

ABSTRACT

Germination, early emergence and stand establishment of crops are major yield determining factors in semi-arid and arid rainfed areas. Farmers in marginal and low input areas tend to have poor germination due to poor and shallow soils. The objective of this study was to evaluate the effect of priming regimes on seed germination of field crops. A study was conducted at Lupane State University, Biotechnology Laboratory in 2019. Treatments included seed priming techniques, namely hydropriming, halopriming (2% NaCl solution) osmopriming (10% PEG 6000), and solid matrix priming (18% volume/weight sand); and timing at 6, 12, 24 hours for maize and 2, 4, 6 hours for cowpea, sorghum and millet. Solid matrix priming, followed by hydropriming significantly ($P < 0.05$) improved germination parameters (germination percentage, daily germination, peak value and germination speed) of all crops. Halopriming and osmopriming were similar in their ineffectiveness, and resulted in the least germination parameter values. Six hour solid matrix priming in 18% v/w sand was the most effective method among most crops; suggesting that solid matrix priming is an effective and possibly low cost technology, with potential to improve germination of field crops.

Key Words: Seed hydration, semi-arid, solid matrix priming

RÉSUMÉ

La germination, l'émergence précoce et l'établissement des peuplements sont des facteurs déterminants du rendement dans les zones pluviales semi-arides et arides. Les agriculteurs des zones marginales et à faible apport ont tendance à avoir une mauvaise germination en raison de sols pauvres et peu profonds. L'objectif de cette étude était d'évaluer l'effet des régimes d'amorçage sur la germination des semences des cultures de champs. Une étude a été faite à Lupane State University, laboratoire de biotechnologie en 2019. Les traitements comprenaient des techniques d'amorçage des semences, à savoir l'hydroprimage, l'halopriming (solution à 2% de NaCl), l'osmopriming (10% PEG 6000) et l'amorçage à matrice solide (18% volume / poids de sable) ; et chronométrage à 6, 12, 24 heures pour le maïs et 2, 4, 6 heures pour le niébé, le sorgho et le mil. L'amorçage de la matrice solide, suivi de l'hydropriming, a significativement amélioré ($P < 0,05$) les paramètres de germination (pourcentage de germination, germination quotidienne, valeur maximale et vitesse de germination) de toutes les cultures. L'halopriming et l'osmopriming étaient similaires dans leur inefficacité et ont donné les valeurs de

paramètres de germination les moins élevées. L'amorçage de la matrice solide sur six heures dans du sable à 18% v / w était la méthode la plus efficace parmi la plupart des cultures; suggérant que l'amorçage à matrice solide est une technologie efficace et peut-être à faible coût, avec un potentiel pour améliorer la germination des cultures de champs.

Mots Clés: Hydratation des graines, semi-aride, amorçage à matrice solide

INTRODUCTION

In arid and semi-arid areas of many sub-Saharan African countries, soil moisture stress affects seed germination and subsequently seedling emergence. In such areas, the moisture required for germination is usually available only for a short time (Musemwa *et al.*, 2013). Management of such soils is a great challenge and yet land preparation is poor under suboptimal soil moisture supply. Hence, techniques that help to enhance germination rates and seedling establishment are imperative.

Seed priming is a technique that can be used to increase the ability of seed to germinate under various environmental conditions (Nawaz *et al.*, 2013; Lutts *et al.*, 2016). Seed priming is a pre-sowing strategy that influences seedling development, by promoting pre-germination metabolic activity prior to emergence of the radicle and plumule (Nawaz *et al.* 2013). It is a simple and low-cost hydration technique, in which seeds are soaked in a priming solution; followed by drying of seeds that initiates germination related processes without radicle emergence. Common priming techniques include osmopriming (polyethylene glycol, glycerol, sorbitol or mannitol), halopriming (salt solutions), hydropriming (water), solid matrix priming (solid, insoluble matrix, such as vermiculite, peat, sand or charcoal) and hormonal priming (Gibberellic acid), salicylic acid, ascorbate or kinetin) (Lutts *et al.*, 2016). However, no priming method is universally effective across crops and sometimes even within varieties of a given crop (Tzortzakis, 2009; Lutts *et al.*, 2016;). Currently, priming is not routinely done in many parts of Africa

perhaps owing to absence of evidence of compelling benefits. The objective of this study was to evaluate the effects and timing of different seed priming techniques on promotion of germination of field crops.

MATERIALS AND METHODS

A laboratory experiment was conducted at the Lupane State University, Biotechnology laboratory under incubator conditions (25 °C and 80% relative humidity). Treatments included halopriming (sodium chloride) at 2% concentration, hydropriming (distilled water), osmopriming (Polyethylene glycol 6000), and solid matrix priming (18% v/w 1 mm sand); and six different crop varieties (2 Maize, 2 Cowpea, 1 Sorghum, 1 Pearl Millet). A two factorial (5 × 6) experiment with an added control (non-primed), priming techniques was setup in the laboratory. Due to differences in germination periods of the different crops, the incubation experiments were done separately to compare the varieties of maize and small grains. The maize experiment was a three factorial (5 × 4 × 2), an added control (non-primed) with 5 priming techniques and four incubation periods of 0, 6, 12, 24 hours and two varieties of maize (ZM401 and PGS51). Each treatment was replicated three times. Small grain crops were compared using a three factorial (5 × 4 × 4) experiment with five priming techniques and 4 incubation periods 0, 2, 4, and 6 hours replicated three times. The added control of non-primed treatments represented a zero level for each variety and incubation period. All the experiments were setup following a Completely Randomised Design, replicated thrice.

All seeds used were of selected and certified nature, purchased from Prime Seed Company on the basis of its availability and widespread use in semi-arid areas. The difference in germination period used in the research came from initial trial runs that showed the smaller seeded crops to germinate prematurely during priming periods longer than 6 hours.

Standard laboratory germination tests were conducted according to the standards of International Rules for Seed Testing (ISTA, 2007). Various germination parameters were observed across the different crops calculated as follows:

$$\text{Germination \%} = \frac{\text{Germinated seeds}}{\text{Total seeds}} \times 100$$

..... Equation 1

$$\text{Mean daily germination (MDG)} = \frac{\text{Germinated seeds}}{\text{Number of days}} \times 100$$

..... Equation 2

$$\text{Peak value} = \frac{\text{Highest number of germinated seeds}}{\text{Number of seeds}} \times 100$$

..... Equation 3

$$\text{Germination value} = \frac{\text{Peak value}}{\text{Mean daily germination}}$$

..... Equation 4

$$\text{Germination seed} = \frac{n1}{d1} + \frac{n2}{d2} + \frac{n3}{d3} \dots \dots \frac{nx}{dx}$$

..... Equation 5

Where:

n = number of seeds germinated, d = number of days

The Shapiro-Wilk procedure was used to test for normality of the data prior to analysis. Data were analysed using Analysis of Variance (ANOVA) in GENSTAT software, version 14 (Payne *et al.*, 2010). Significant mean values were separated with Fischer's Protected Least Significant Difference (LSD) at 5% level.

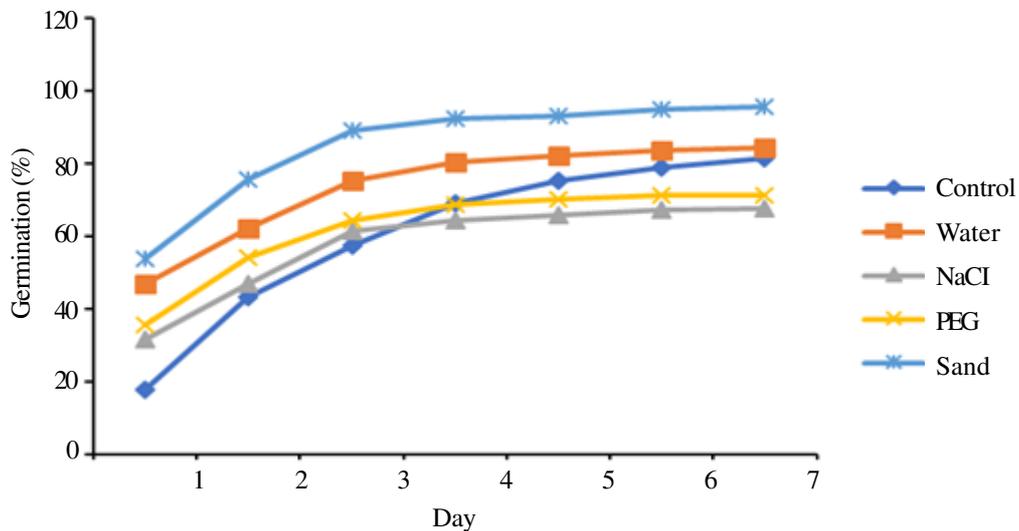
RESULTS

There was generally significant interaction effects (P<0.05) between priming, priming technique, genotype across the two main time intervals (day 1 and 7) (Table 1). All crops primed with sand had the highest germination percentage from day 1 to day 7, followed by distilled water (Fig. 1). For cowpea varieties, CBC 2 and IT18, the highest germination percentage was observed in seeds primed with sand compared to other techniques. Seeds of sorghum variety Macia, primed with PEG, had the highest germination percentage; followed by sand and then distilled water (Fig. 2). Pearl millet variety Okashana primed with distilled water had the highest germination percentage in all the non-maize crops evaluated. However, there was no significant difference (P>0.05) in the germination percentage rates of Okashana seeds primed with water, NaCl, PEG and sand (Fig. 2).

Maize variety PGS51 had its highest germination percentage for seeds primed with distilled water. There was no significant difference (P>0.05) in germination percentage of seeds primed with NaCl, PEG and those primed with sand.

TABLE 1. Analysis of variance for day 1 and day 7 germination under priming, priming technique and varieties

Source of variation	Degrees of freedom	Day 1 Mean squares	Day 7 Mean squares	F probability	F probability
Priming	1	36.9	<.001	3.1	0.023
Priming*technique	3	18.3	<.001	16.3	<.001
Priming*variety	10	36.6	<.001	22.4	<.001
Priming*agent*variety	15	4.0	<.001	5.6	<.001
Residual	60				
Total	89				



$P < 0.001$, LSD 10.47, CV 28.6 NaCl = sodium chloride, PEG = polyethylene glycol

Figure 1. Germination percentages of crops treated with different priming techniques during the 7 day period.

Maize variety ZM401 had the least germination percentage observed in unprimed seeds; and the highest in seeds primed with sand, although there was no significant difference ($P > 0.05$) in priming with sand, PEG, NaCl and distilled water on germination percentage (Fig. 2). Priming ZM401 maize variety in distilled water for 12 hours resulted in the highest germination (100%); whereas

PGS51 in sand for 6 hours resulted in the highest germination (96%) (Table 2).

The interaction of priming, genotype and duration had a significant effect on the germination of CBC 2, IT 18, Macia and Okashana which were primed for 2, 4 and 6 hours (Table 3). Overall, the non-primed control seed had the greatest germination percentage, followed by 6 hours priming

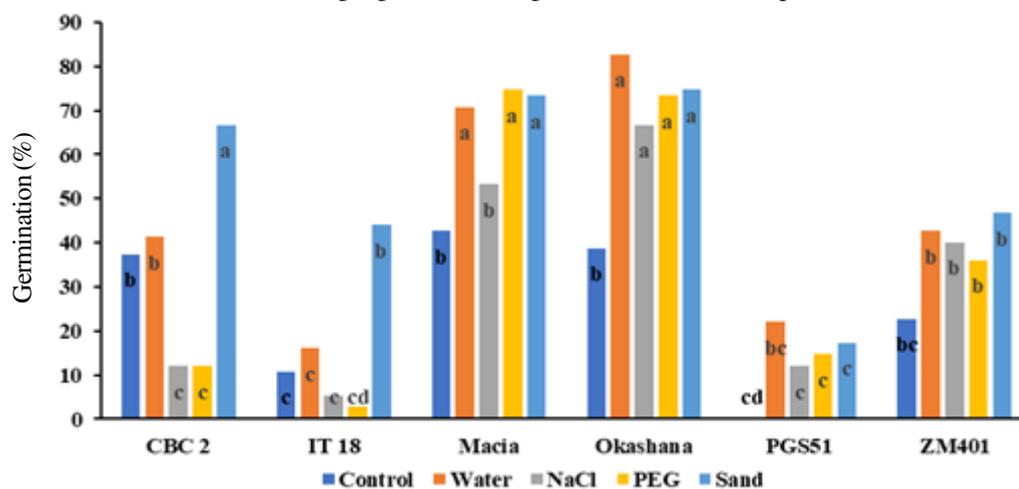


Figure 2. Germination percentage on day 1 crops treated using different priming techniques.

TABLE 2. Germination response for maize varieties to interaction of priming, variety and duration of incubation

Priming	Variety	0 hours	6 hours	12 hours	24 hours
Control	PGS51	58.7 ^b	-	-	-
	ZM401	49.3 ^{bc}	-	-	-
Water	PGS51	-	89.3 ^a	93.3 ^a	90.7 ^a
	ZM401	-	98.7 ^a	100.0 ^a	98.7 ^a
NaCl	PGS51	-	82.7 ^{ab}	82.7 ^{ab}	84.0 ^{ab}
	ZM401	-	97.3 ^a	96.0 ^a	93.3 ^a
PEG	PGS51	-	88.0 ^a	80.0 ^{ab}	85.3 ^a
	ZM401	-	90.7 ^a	96.0 ^a	74.7 ^{ab}
Sand	PGS51	-	96.0 ^a	68.0 ^b	94.7 ^a
	ZM401	-	92.0 ^a	97.3 ^a	88.0 ^a
Mean		54.0	91.8	89.2	88.7
P0.013					
LSD 14.73					

Means were compared using LSD = not done

TABLE 3. Variety germination response to interaction of priming, variety and duration

Priming	Variety	0 hours	6 hours	12 hours	24 hours
Control	CBC2	77.3 ^a	-	-	-
	IT 18	57.3 ^{ab}	-	-	-
	Macia	84.0 ^a	-	-	-
	Okashana	88.0 ^a	-	-	-
Water	CBC2	-	74.7 ^{ab}	77.3 ^a	62.7 ^{ab}
	IT 18	-	33.3 ^c	50.7 ^b	37.3 ^b
	Macia	-	92.0 ^a	93.3 ^a	88.0 ^a
	Okashana	-	76.0 ^a	82.7 ^a	97.1 ^a
NaCl	CBC2	-	66.7 ^{ab}	40.0 ^b	29.3 ^c
	IT 18	-	34.7 ^c	17.3 ^c	19.3 ^c
	Macia	-	81.3 ^a	68.0 ^{ab}	77.3 ^a
	Okashana	-	82.7 ^a	92.0 ^a	85.3 ^a
PEG	CBC2	-	54.7 ^b	41.3 ^b	35.3 ^c
	IT 18	-	17.3 ^c	12.0 ^{cd}	18.0 ^c
	Macia	-	88.0 ^a	93.3 ^a	93.3 ^a
	Okashana	-	84.0 ^a	93.3 ^a	94.7 ^a
Sand	CBC2	-	54.7 ^b	72.0 ^{ab}	94.7 ^a
	IT 18	-	73.3 ^{ab}	69.3 ^{ab}	81.3 ^a
	Macia	-	90.7 ^a	89.3 ^a	94.7 ^a
	Okashana	-	88.0 ^a	97.3 ^a	89.3 ^a
Mean		76.7	64.2	68.1	68.7
P0.047					
LSD21.92					

Means were compared using LSD = not done

TABLE 4. Crop mean daily germination, peak value, germination value and germination speed response to priming

Priming	MDG	PV	GV	GS
Control	2.97 ^b	4.28 ^{ab}	12.73 ^b	32.77 ^{bc}
Water	3.01 ^b	5.70 ^a	18.23 ^a	41.75 ^a
NaCl	2.59 ^c	4.12 ^b	12.26 ^b	33.70 ^b
PEG	2.59 ^c	4.60 ^{ab}	13.77 ^b	35.70 ^b
Sand	3.23 ^a	5.19 ^a	17.12 ^a	44.34 ^a
P	<0.001	0.002	<0.001	<0.001
LSD	0.19	0.98	3.22	2.99

Means were compared using LSD. MDG = mean daily germination, PV=peak value, GV = Germination value, GS = Germination speed

duration. However, there was great variation in germination percentage depending on the genotype and priming technique (Table 3).

Priming technique had a significant effect ($P < 0.05$) on mean daily germination, peak value, germination value and germination speed (Table 4). Priming all crop varieties with sand resulted the highest mean daily germination, crop germination speed and germination value (Table 4).

DISCUSSION

Across all the priming techniques evaluated, all the crop genotypes had a starting point advantage compared to non-primed ones (Fig. 1). This is a critical element in the crop field where there is need for rapid germination and emergence to be able to compete with weeds and for the scarce soil resources. Notably, the significant enhancement of germination parameters due to sand priming (solid matrix) could be because seeds were slowly provided with water and thus allowed for a slow and controlled imbibition to occur, prompting rehydration cell membrane repair mechanisms to operate efficiently and adequately (Mercado and Fernandez, 2002). Furthermore, this decreased electrolyte leakage, thus allowing for more rapid germination (Zhao *et al.*, 2009). Other priming techniques evaluated were primarily liquid formulations ie PEG, water, salt solution which allow for rapid imbibition. Immersion methods cause rapid water uptake compared to solid osmotic media that rehydrates seed slowly. Rapid, uncontrolled imbibition is often associated with more electrolyte leakage since desiccation damaged cell membranes need time to reconstitute (Pallaoro *et al.*, 2016) which could explain the slower rate of germination and related parameters.

The positive contribution of solid matrix conforms to the results of Zhao *et al.* (2009) and Singh *et al.* (2015) who noted that seed priming with sand or vermiculite improved field emergence of maize and carrot

respectively compared to PEG non-primed controls. Priming with sodium chloride had a lower germination percentage and other seed parameters than water and PEG. This could be attributed to the lower water potential caused by the salt or ionic effects (Zhang *et al.*, 2015). Similarly, PEG had relatively lower germination parameter values due to its hydrophilic nature that reduced imbibition. CBC2 and IT18 seemed more sensitive to these two priming techniques compared to cereals, Macia and Okashana. This is contrary to the positive role of sodium chloride that was noted by Eskandari and Kazemi (2011), who observed that cowpeas had the highest germination when primed with the salt compared to other halo-priming techniques.

All primed maize seed (PGS51 and ZM401) had germination percentages greater than that of their non-primed counterparts from day 1 to day 7 duration of incubation, regardless of priming technique. Water was a highly effective priming agent, second to sand in all the crop genotypes, with no adverse effects to germination parameters. This is important given that water is more readily available for use prior to planting. However, it is important for farmers to get the duration of priming right to prevent pre-germination or inadequate imbibition. Priming CBC 2, IT 18, Macia and Okashana for 6 hours in sand gave seeds maximum time to absorb water, which was sufficient to initiate and maximise germination. Priming maize longer than 12 hours had no additional benefit. Across all crops, priming with sand resulted in improved mean daily germination and germination speed (GS) (Table 4). Generally, priming (except with sodium chloride) was an effective means of promoting rapid germination of the crops.

CONCLUSION

Overall, sand priming for 6 hours hastens seeds germination, giving primed seedlings competitive advantage in the field. Where sand is not available, water is the next best priming

option. All cereals responded positively to priming whereas cowpea did not respond well regardless the technique used. It is important to identify the most effective technique and ideal duration for a given crop genotype. Further research on the role of priming under field conditions on abiotic factors such as salinity should be investigated.

ACKNOWLEDGEMENT

The authors thank the laboratory technicians in the Department of Crop and Soil Sciences at Lupane State University, Zimbabwe for the technical assistance.

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