Can Hydro-Priming Improve Germination Speed, Vigour and Emergence of Maize Landraces under Water Stress?

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Abstract: We evaluated whether hydro-priming could improve vigour characteristics and seedling emergence of local maize (*Zea mays* L.) landraces compared to two commercial hybrids under water stress at the University of KwaZulu-Natal, Pietermaritzburg. Seeds from local landraces were produced and characterized according to kernel colour, white (Land A) and purple (Land B), and compared to two hybrids, SC701 and SR52, which are popular amongst local farmers. Seeds from each variety were soaked in water for 0 hours (unprimed or control), 12 hours and 24 hours, and germinated in a germination chamber at 25 °C for 8 days. Parameters measured included final germination, mean germination time (MGT) and germination velocity index (GVI). Seedling emergence was performed in seedling trays, using pine bark wetted to 25% or 75% of field capacity (FC), for 21 days in a temperature-controlled glasshouse (25 °C day; 15 °C night; 60% RH). Parameters measured included final emergence, mean emergence time (MET), root and shoot lengths, and leaf area. Priming landraces for 12 and 24 hours reduced MGT by 9% and 7%, respectively, compared to 5% in hybrids for both 12 and 24 hours priming. GVI of landraces was improved by 40% following 12 hours of priming. GVI of hybrids was 11% and 7% slower than landraces after priming seeds for 12 and 24 hours, respectively. Priming seeds for 24 hours improved emergence at 25% FC. Priming seeds for 24 hours reduced MET for all varieties. Priming seeds for 12 and 24 hours increased leaf area by 33.8% and 29%, respectively. Hydro-priming seeds for 12 and 24 hours, respectively, improved GVI, reduced MGT and improved emergence and MET of maize landraces under water stress. Performance of hybrid seeds remains superior to that of landraces even after seed treatment to improve germination and vigour. The positive response of landraces to seed treatment, and improved performance under water stress conditions, suggest that there is a need to identify genes for vigour in landrace maize.

Key words: Emergence, germination, hybrids, hydropriming, landraces, water stress.

1. Introduction

Good crop establishment is essential for efficient water use [1] and is a major constraint to crop production in the semi-arid tropics [2-4]. This is particularly true for maize (*Zea mays* L.) which does not tiller [5]. Good germination and emergence are important for achieving good crop establishment and maximum possible plant populations in the field, more

so under adverse growing conditions. As such, speed of germination and emergence is important for successful establishment [6].

Technology that enhances germination and emergence is thus important in mitigating deleterious effects of poor crop establishment due to drought. Such technology would allow farmers to achieve good crop stands and ultimately good yields. Seed priming is one such technology which has been developed to enhance germination characteristics of seeds [7]. Its purpose is to partially hydrate the seeds to a point where germination processes are initiated but not completed

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[8, 9]. Primed seeds exhibit rapid germination and emergence under field conditions [10].

A variety of methods have been used to study the effect of seed priming on germination and growth rate of maize. These include osmopriming (soaking seeds in osmotic solutions such as polyethylene glycol), halopriming (soaking seeds in salt solutions), hydro-priming (soaking seeds in water), hormonal-priming and matri-priming [11-14]. Priming maize seed using polyethylene glycol (PEG) or potassium salts (K_2HPO_4 or KNO_3) accelerated germination in a chilling germinator ($10^\circ C$) [15]. Soaking seed in 2.5% potassium chloride (KCl) for 16 hours reduced coleoptile and radicle length, while soaking seed in 20 ppm GA_3 for 30 min improved some germination traits [16].

Hydro-priming (henceforth referred to as priming) is a simple low-cost method of seed priming that requires no sophisticated equipment and gives results which are easy to see [7]. Nagar et al. [17] observed a significant improvement in field emergency and seedling characteristics after hydro-priming maize for 16 hours. In a series of experiments, Harris et al. [3] showed that hydro-priming greatly improved establishment and vigour of upland rice, maize and chickpea, and resulted in faster development, earlier flowering and maturity and higher yields. This simple, low-cost, low-risk intervention also had positive impacts on the wider farming system and livelihoods and the technology proved highly popular with farmers [3, 18].

Maize landraces are still being grown by subsistence farmers in KwaZulu-Natal, South Africa under a rainfed system, which according to Rowland [19] is a risky environment. The risk is related to rainfall amount and distribution [7] during the time of planting. Farmers normally sow their maize either in late spring, before the onset of the rains, or with the first rains. The former crop usually suffers from a dry seedbed, resulting in poor emergence. The latter crop may suffer from rains that usually peter out early. In either case, the result is poor crop establishment leading to poor

yields due to reduced plant populations.

Earlier work [20] showed that landraces may have the same viability but not vigour as hybrids since landraces were slower to germinate and emerge than hybrids. The aim of this study was to evaluate whether hydro-priming can be used to improve germination speed and emergence of local maize landraces under water stress conditions. The performance of landraces was compared to two popular hybrids, SC701 and SR52.

2. Material and Methods

2.1 Planting Material

Seed for the maize landraces was initially donated by local farmers in KwaZulu-Natal, South Africa, and multiplied at the University of KwaZulu-Natal in the previous year (2007). Maize landraces were characterized according to kernel colour, two of which were selected for this study; white (Land A) and purple (Land B). Two hybrids, SC701 and SR52, were used in the study for purposes of comparing the landraces' performance.

2.2 Seed Priming Procedure

Seeds were soaked in distilled water for 12 (P12) and 24 hours (P24), respectively. After soaking, the seeds were surface dried.

2.3 Laboratory Germination

Three replicates of 25 seeds from each variety and priming treatment were germinated between double layered, moistened paper towels [21]. The paper towels were rolled, put into zip-lock bags and incubated in a germination chamber at $25^\circ C$ [22] for 8 days. Radicle protrusion was the criterion of germination. Observations for final germination percentage, based on normal seedlings, were made according to AOSA [21] guidelines. Root and shoot length, root: shoot ratio, fresh and dry mass were measured.

Mean time to germination (MGT) was calculated according to Eq. (1) by Ellis and Roberts [23]:

$$MGT = \frac{\sum D_n}{\sum n} \quad (1)$$

Where:

MGT = mean germination time,

n = the number of seed which were germinated on day D, and

D = number of days counted from the beginning of germination.

Germination speed was calculated based on Maguire's [24] Eq. (2):

$$GVI = G1/N1 + G2/N2 + \dots + Gn/Nn \quad (2)$$

Where:

GVI = germination velocity index G1, G2...Gn = number of germinated seeds in first, second... last count.

N1, N2...Nn = number of sowing days at the first, second... last count.

2.4 Total Soluble Sugars

0.1 g freeze-dried seed tissue was added to a test tube. To each test tube, 10 mL of 80% ethanol was added and homogenized for 30 sec using Ultraturax and incubated for 1 hour at 80 °C in a water bath. Thereafter, the test tube were stored at 4 °C for 24 hours whereafter they were centrifuged (10,000 rpm for 15 min at 4 °C). Extract was filtered through glass wool and dried down overnight in a Savant Vacuum drier. Dried samples were diluted up to 2 mL by adding ultra pure water and centrifuged for 15 min and filtered through 0.4 micron nylon syringe filter into sample vials. Soluble sugars were determined using High-Performance Liquid Chromatography (HPLC) (Shimadza) as reported by Aung et al. [25, 26].

2.5 Seedling Emergence

Three replicates of 10 seeds from each variety and priming treatment combination were planted in seedling trays using pine bark as growing media at 25% and 75% field capacity (FC), respectively, over a period of 22 days in a temperature controlled (25 °C, 60% humidity) glasshouse. Trays were weighed and

watered at two-day intervals to maintain field capacities. Data collected included daily emergence for 14 days, leaf area, root and shoot lengths and root and shoot mass (fresh and dry).

Mean time to emergence was calculated using Eq. (3) by Bewley and Black [27]:

$$MET = \frac{\sum (fx)}{\sum f} \quad (3)$$

Where MET = mean emergence time,

f = number of newly germinating seeds at a given time (day), and

x = number of days from date of sowing.

2.6 Statistical Analysis

Data collected was analyzed using GenStat® Version 11 statistical package. Means were separated using LSD ($P = 0.05$).

3. Results

3.1 Laboratory Germination

3.1.1 Final Germination

Priming had a highly significant effect ($P < 0.001$) on final germination. Results for final germination showed there was a significant interaction ($P < 0.05$) between priming and variety (Table 1). With the exception of Land B, priming did not increase final germination in the other three varieties. Maximum germination (100% for Land A and 98.67% for both hybrid varieties) was achieved in the unprimed treatment. For both priming treatments (P12 and P24), final germination fell by an average 8% in the hybrids compared to 4% in the landraces. Land B attained maximum germination (98.67%) when seeds were primed for 24 hours (P24).

3.1.2 Mean Germination Time (MGT)

Priming had an effect ($P < 0.001$) on mean germination time (MGT). For all varieties, priming reduced MGT. There was a highly significant interaction ($P < 0.001$) between variety and priming in MGT (Table 1). Hybrids germinated faster than landraces when seeds were not primed. The effect of

Table 1 Germination attributes of landraces (Land A and Land B) and hybrids (SC701 and SR52) for unprimed (UP), 12 hours (P12) and 24 hours (P24) seeds.

	Variety	Germination	MGT	GVI	Root length (mm)	Shoot length (mm)	Root:Shoot	Fresh mass (g)	Dry Mass (g)
Unprimed	Land A	100a	4.7ab	32.99e	113d	89.5g	1.265abc	1.402c	0.314c
	Land B	97.33abc	4.7ab	34.63de	170.2ab	139.5b	1.227abcd	1.805a	0.304c
	SC701	98.67ab	4.767a	32.79e	135.5cd	102.8fg	1.345ab	1.498bc	0.242d
	SR52	98.67ab	4.5bc	38.46cd	132cd	106f	1.252abc	1.558bc	0.379a
Mean		98.67 ^a	4.667 ^a	34.72 ^b	137.7 ^a	109.5 ^b	1.272 ^a	1.566 ^b	0.3097 ^b
P12	Land A	93.33bc	4.3de	46.84a	114.5cd	163.5a	0.721e	1.895a	0.323bc
	Land B	92c	4.233e	48.35a	109.4d	111.4ef	0.983cde	1.601b	0.298c
	SC701	94.67abc	4.367d	44.43ab	191.7a	133.9bc	1.444a	1.934a	0.373ab
	SR52	86.67d	4.4cd	39.85bc	122.7cd	128.4bcd	0.974de	1.586bc	0.408a
Mean		91.67 ^c	4.325 ^b	44.87 ^a	134.6 ^a	134.3 ^a	1.03 ^b	1.754 ^a	0.3503 ^a
P24	Land A	93.33bc	4.367d	40.17bc	123cd	123.8cde	1.012cd	1.599bc	0.314c
	Land B	98.67ab	4.3de	49.12a	132.3cd	129.4bc	1.053cd	1.454bc	0.297cd
	SC701	97.33abc	4.3de	48.43a	147.7bc	135.8bc	1.09bcd	1.545bc	0.276cd
	SR52	82.67d	4.467cd	34.6de	129.3cd	113def	1.147bcd	1.489bc	0.4a
Mean		93 ^b	4.358 ^b	43.08 ^a	133.1 ^a	125.5 ^a	1.076 ^b	1.522 ^b	0.3217 ^b
LSD ($P = 0.05$) Variety × Priming		6.422	0.1206	5.371	33.47	15.42	0.2847	0.1988	0.05503
LSD ($P = 0.05$) Priming		3.211	0.0603	2.685	16.74	7.71	0.1424	0.0994	0.02751

MGT = mean germination time; GVI = germination velocity index (germination speed). Numbers with different letters in the same column differs significantly at LSD ($P = 0.05$).

priming on MGT was more pronounced in the landraces than the hybrids. Priming landraces for 12 and 24 hours reduced MGT by 9% and 7%, respectively, compared to a reduction of 5% for hybrids in both cases.

3.1.3 Germination Velocity Index (GVI)

In addition, priming had a highly significant effect on germination speed, increasing GVI in all varieties. There was a highly significant interaction ($P < 0.001$) between variety and priming with respect to GVI (Table 1). Hybrids germinated 5% faster than the landraces when seeds were not primed. However, when seeds were primed for 12 and 24 hours, landraces germinated 11% and 7% faster than the hybrids, respectively. Over-all, priming seeds for 12 hours had the greatest effect on landraces, improving their GVI by 40% when compared to unprimed seeds.

3.1.4 Germination Vigour Traits

Furthermore, there was a highly significant interaction ($P < 0.001$) between variety and priming

for germination vigour traits such as root and shoot lengths and fresh mass (Table 1). Root length for landraces fell by 20% (P12) and 9% (P24) as compared to the maximum root length reached when seeds were not primed. Land B, in particular was negatively affected by priming. Hybrids increased root length in response to priming. Their roots were 28% and 7% longer than the landraces when seeds were primed for 12 and 24 hours, respectively. Priming increased shoot length for all varieties. For landraces, seeds primed for 12 and 24 hours, respectively, had about 22% and 10% longer shoots than the unprimed seeds. While for hybrids, primed seeds were 25% (P12) and 19% (P24) longer than unprimed seeds. Overall, landraces responded better than hybrids to priming with regard to shoot length by 7% (P12) and 1.7% (P24). Lastly, priming had a significant effect ($P < 0.05$) on dry mass. Landraces had a marginal increase ($< 1\%$) when seeds were primed for 12 hours. Hybrids, however, increased dry mass by 25% and 8% when seeds were primed for

12 and 24 hours, respectively.

3.1.5 Total Soluble Sugars

There was no significant interaction ($P > 0.05$) between variety and priming, with respect to total soluble sugars (TSS) (Fig. 1). Hydro-priming did not have a significant ($P > 0.05$) effect on TSS. Priming resulted in a decline in TSS in both hybrids and Landrace B. However, there were significant differences ($P < 0.05$) between varieties with respect to TSS. Landraces had higher TSS content compared to hybrids. Overall, based on mean values, landraces had 19% more TSS than hybrids.

3.2 Seedling Emergence

3.2.1 Seedling Emergence

There were no differences ($P > 0.05$) in seedling emergence (Fig. 2) with respect to variety, priming and field capacity. There was no significant interaction ($P > 0.05$) between the three treatment factors. SR52 was adversely affected when seeds were primed for 24 hours. Emergence improved under water stress when seeds were primed for 24 hours. There was no significant ($P > 0.05$) three way interaction with respect to mean emergence time (MET) (Fig. 3). However, there was a highly significant interaction ($P < 0.001$) between priming and field capacity (Fig. 3). MET was reduced when seeds were primed for 24 hours in all varieties (Fig. 3). Priming seeds for 12 hours improved emergence under optimum conditions (75% FC) and not under water stress (25% FC).

3.2.2 Root and Shoot Length

There was no significant three way interaction ($P > 0.05$) between variety, priming and field capacity for all seedling characteristics (Table 2). With respect to root length, priming and field capacity both had significant effects ($P < 0.05$) while their interaction was also significant ($P < 0.05$) (Table 2). Under water stress conditions (25% FC), root length increased by 4% (P24) and 16% (P12) in response to priming. In particular, landraces increased root length under water stress by 4% (P24) and 21% (P12). Although the interaction between priming and field capacity had no effect ($P >$

Fig. 1 Total soluble sugars concentration for landraces (Land A and Land B) and hybrids (SC701 and SR52) in response to hydro-priming seeds for 0 hours (UP), 12 (P12) and 24 hours (P24).

Fig. 2 Seedling emergence for landraces (Land A and Land B) and hybrids (SC701 and SR52) grown at 25% FC and 75% FC after seeds were either not primed or primed for 12 (P12) and 24 (P24) hours.

Fig. 3 Mean emergence time (MET) for landraces (Land A and Land B) and hybrids (SC701 and SR52) grown at 25% FC and 75% FC after seeds were either not primed or primed for 12 (P12) and 24 (P24) hours.

Table 2 Seedling characteristics of landraces (Land A and Land B) and hybrids (SC701 and SR52) at 25% and 75% field capacity, respectively.

	Variety	Root length (mm)	Shoot length (mm)	Root: Shoot	Root DM (g)	Shoot DM (g)	Leaf area (cm ²)
Unprimed	Land A	55.33 ^{cde}	204.3 ^d	0.276 ^{bcd}	0.2733 ^{bcdefg}	0.1367 ^{abcde}	36.2 ^{def}
25FC	Land B	56 ^{cde}	185 ^d	0.304 ^{abc}	0.2167 ^{fgh}	0.1133 ^{bcde}	35.8 ^{def}
	SC701	65 ^{abcd}	187 ^d	0.4061 ^a	0.3133 ^{abcde}	0.0967 ^{cde}	35.8 ^{def}
	SR52	45 ^e	177.3 ^d	0.2627 ^{bcd}	0.3367 ^{abcd}	0.0767 ^e	29.2 ^f
P12	Land A	66.67 ^{abc}	261.7 ^{abcd}	0.2547 ^{bcd}	0.1767 ^h	0.1533 ^{abcd}	51.9 ^{bcde}
25FC	Land B	68.33 ^{ab}	224.7 ^{cd}	0.3033 ^{abc}	0.21 ^{fgh}	0.14 ^{abcde}	45.8 ^{bcdef}
	SC701	62.33 ^{bcd}	259.3 ^{abcd}	0.2416 ^{bcd}	0.3967 ^a	0.15 ^{abcd}	49.2 ^{bcdef}
	SR52	60.67 ^{bcd}	250.7 ^{abcd}	0.243 ^{bcd}	0.3467 ^{abc}	0.14 ^{abcde}	47.3 ^{bcdef}
P24	Land A	53.33 ^{de}	234.3 ^{bcd}	0.2288 ^{bcd}	0.2 ^{gh}	0.1267 ^{bcde}	43.3 ^{cdef}
25FC	Land B	62.67 ^{bcd}	198.7 ^d	0.3204 ^{ab}	0.2567 ^{cdefgh}	0.12 ^{bcde}	33.4 ^{ef}
	SC701	57.33 ^{bcd}	294.7 ^a	0.1942 ^d	0.27 ^{bcddefgh}	0.1467 ^{abcde}	61 ^{abc}
	SR52	58.33 ^{bcd}	206.3 ^d	0.289 ^{abcd}	0.36 ^{ab}	0.0833 ^{de}	34.6 ^{ef}
Unprimed	Land A	58.67 ^{bcd}	215.7 ^d	0.2721 ^{bcd}	0.21 ^{fgh}	0.1333 ^{abcde}	44.3 ^{cdef}
75FC	Land B	63b ^{cd}	226.7 ^{cd}	0.2783 ^{bcd}	0.1967 ^{gh}	0.1267 ^{bcde}	37.5 ^{def}
	SC701	62.67 ^{bcd}	284.7 ^{ab}	0.2243 ^{bcd}	0.29 ^{bcddefgh}	0.1367 ^{abcde}	59.5 ^{abc}
	SR52	53.33 ^{cde}	217 ^{cd}	0.2459 ^{bcd}	0.3267 ^{abcd}	0.09 ^{de}	41.7 ^{cdef}
P12	Land A	65 ^{abcd}	296.7 ^a	0.2233 ^{bcd}	0.2233 ^{efgh}	0.18 ^{ab}	59.9 ^{abc}
75FC	Land B	57.67 ^{bcd}	199 ^d	0.2941 ^{abcd}	0.2467 ^{defgh}	0.1133 ^{bcde}	36.1 ^{def}
	SC701	63.33 ^{bcd}	250.7 ^{abcd}	0.2555 ^{bcd}	0.3033 ^{abcdef}	0.1633 ^{abc}	61.3 ^{abc}
	SR52	60.33 ^{bcd}	300 ^a	0.203 ^{cd}	0.2933 ^{bcdefg}	0.2033 ^a	76.8 ^a
P24	Land A	65.33 ^{bcd}	249.7 ^{abcd}	0.2834 ^{bcd}	0.2833 ^{bcdefg}	0.1533 ^{abcd}	53.4 ^{bcde}
75FC	Land B	61.67 ^{bcd}	258.7 ^{abcd}	0.239 ^{bcd}	0.23 ^{efgh}	0.18 ^{ab}	55.8 ^{bcd}
	SC701	67 ^{abc}	300.7 ^a	0.2235 ^{bcd}	0.2767 ^{bcdefg}	0.1733 ^{ab}	66.3 ^{ab}
	SR52	75.67 ^a	273.3 ^{abc}	0.2773 ^{bcd}	0.2667 ^{bcdefgh}	0.1733 ^{ab}	65.4 ^{ab}
LSD _(P=0.05) Var*Priming*FC		12.159	57.24	0.10798	0.09336	0.07179	20.68

FC = Field Capacity. Numbers with different letters in the same column differ significantly at LSD ($P = 0.05$).

0.05) on shoot length (Table 2), priming, on its own, had a highly significant effect ($P < 0.001$) on shoot length. There were no differences in root and shoot dry mass (Table 2).

3.2.3 Leaf Area

Leaf area development showed no significant three way interaction ($P > 0.05$) between variety, priming and field capacity (Table 2). Field capacity had a highly significant effect ($P < 0.001$) on leaf area (Table 2), reducing it by about 23% under water stress. Nonetheless, priming had a significant effect ($P < 0.001$) on leaf area (Table 2); leaf area increased by 33.8% and 29% in response to priming seeds for 12 and 24 hours. Landraces increased their leaf area under water stress (25% FC) by 34% (P12) and 6.5% (P24) while the hybrids increased by 48.5% (P12) and 47%

(P24), respectively.

4. Discussion

Priming of seed has been effectively used to enhance the vigour and emergence of seedlings under both optimal [28, 29] and sub-optimal conditions [30, 31]. The objective of this study was to determine whether or not hydropriming can be used to improve vigour, with respect to germination attributes and seedling emergence under water stress, in landraces and thus improve crop establishment.

Priming had a negative effect on final germination of all varieties. Rapid uptake of water during priming may have caused imbibition injury, resulting in failure of seeds to germinate. There are similar instances in the literature reporting imbibitional injury in seeds,

including maize [32-36]. Although most of these reports show imbibitional damage at low temperatures, imbibitional damage at higher temperatures, although less severe, can also reduce germination [5].

Priming improved germination speed and reduced MGT. Primed seeds germinated faster and more uniformly than unprimed seeds. Although priming reduced root lengths in landraces, it increased shoot lengths, fresh mass and dry mass; suggesting that a greater part of seed reserves were channeled to the shoots which is crucial for early establishment and photosynthesis. Overall, priming improved vigour of the seeds, with landraces performing well when seeds were primed for 12 hours. These results are similar to others reported in literature [3].

Wahid et al. [37] observed an increase in soluble sugars concentration in response to priming sunflower achenes. They concluded that priming-induced improvements in germination and seedling growth were associated with greater substrate availability for germination. However, contrary to this expectation, results showed an unclear pattern with respect to TSS. In most instances, priming resulted in reduced substrate availability, more so in hybrids. The higher TSS in landraces may explain their positive response to priming.

Successful crop establishment determines plant density, uniformity and management options [38] and depends not only on the rapid and uniform germination of the seed, but also on the capacity of the seed to emerge under water stress [39]. Alleviating the deleterious effect of water stress at this stage can increase chances for attaining a good crop [40].

Priming increased seedling emergence under both optimum and water stress conditions. Priming for 12 hours improved emergence of the landraces at 75% FC while priming for 24 hours resulted in better emergence for all varieties at 25% FC. Priming for 24 hours resulted in reduced MET under water stress. Priming also resulted in increased roots and shoots lengths as well as increased leaf area in the landraces. Thus,

priming resulted in improved crop establishment and healthier seedlings. Ghassemi-Golezani et al. [14] reported that hydropriming improved seedling emergence rate and percent in lentil; Harris et al. [3] reported enhanced seedling establishment and early vigour of upland rice, maize and chickpea after hydropriming; Kibite and Harker [41] reported that seed hydration improved uniformity of seedling emergence of wheat, barley and oat seeds.

5. Conclusion

Good crop establishment is a prerequisite for successful crop production especially under water stress conditions. Seeds responded better to priming for 12 hours when conditions were optimum while priming for 24 hours improved emergence, reduced MET and improved seedling characteristics under water stress. Farmers may use hydro-priming, combined with a slightly higher seeding rate, when planting early and late in the season. Hydro-priming maize landraces for various periods of time may be used a low-cost technology to enhance emergence and vigour characteristics of landraces under both optimum and water stress conditions. However, there is need for further research to evaluate whether these initial benefits may contribute to improved yields under water stress.

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