DOI: 10.2478/cerce-2018-0011
Available online: www.uaiasi.ro/CERCET_AGROMOLD/
Print ISSN 0379-5837; Electronic ISSN 2067-1865

Original Article

Cercetări Agronomice în Moldova
Vol. LI , No. 2 (174) / 2018: 5-15

EVALUATING THE POTENTIAL EFFECT OF SEED PRIMING TECHNIQUES IN IMPROVING GERMINATION AND ROOT SHOOT LENGTH OF MAIZE SEED

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Received Jan. 03, 2018. Revised: Apr. 24, 2018. Accepted: May 02, 2018. Published online: June 27, 2018

ABSTRACT. The present research was conducted under laboratory conditions. The purpose of research was to investigate the potential of priming with press mud, peat moss, sand, gunny bags, compost, farm yard manure and moringa leaf extract (MLE) on seedling growth and germination capacity of maize seed. Untreated or non-primed seeds were used as a control treatment. Priming treatments improved germination capacity, stand establishment and seedling vigor, compared with control. Priming with moringa leaf extract enhance germination and seedling vigor of maize seed, compared with the control and other seed primed treatments. In moringa leaf extract primed seeds, root and shoot growth was improved. Overall, moringa leaf extract primed maize seeds performed better than all other treatments and it could be related by seedling vigor enhancement and lowering the mean germination time, due to imbibition of higher quantity of water and earlier enzymatic activity. The results propose that moringa leaf extract priming treatment had the potential to enhance germination, stand establishment and early growth of maize seeds.

Keywords: seed invigoration; moringa leaf extract; solid matrix priming; matriconditioning; osmotic priming; corn.

Abbreviations: MLE - moringa leaf extract; FGP - final germination percentage; MGT - mean germination

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time; FDG - first day of germination; LDG - last day of germination; GI - germination index; T50 - time to attain 50% germination; RL - root length; SL - shoot length; RL/SL - root to shoot ratio.

INTRODUCTION

The rationale for early planting seed treatments is to activate seed's own resources and to combine them with external resources to maximize growth in stand establishment and yield. Physiological and non-physiological treatments are important to remove several soil, climatic and water constraints. Physiological seed treatments that may enhance the seed performance, primarily based on seed hydration techniques, which includes presoaking, humidification, matri-conditioning, wetting and dying and pre-germination. Others used may promote germination or remove blocks to germination demands, either its chemical or may be physical stimuli in addition to seed hydration, which includes scarification of seed, treatment with chemicals, coating and seed inoculation with beneficial microbes.

Seed priming is a technique controlling hydration and drying, that results in more rapid germination when the seeds are re-imbibed. Seed priming technique has been found to be a feasible technology to improve seedling emergence in some field crops, such as cotton, common maize, rice and wheat (Murungu *et al.,* 2004; Farooq *et al.,* 2006; 2008a,b). In sweet corn, seed priming commonly used to reduce the time between seed sowing and seedling emergence and to improve the percentage of emergence (Chiu *&* Sung, 2002).

Seed priming has been shown to improve seed performance under sub-optimal temperature conditions (Lin & Sung, 2001). Priming increases, the environmental range suitable for germination, and provides faster and synchronous seedling emergence (McDonald, 1999). Seed priming is widely used for enhancing seed performance by improving the rate and uniformity of germination and decreasing seed sensibility to external factors (Corbineau & Cõme 2006). The benefits of seed priming had been reported, including improving stand establishment in semi-arid condition (Clark *et al.*, 2001) and at drought stress (Kaur *et al.*, 2002), enhancing seed with low vigor (Bittencourt *et al.*, 2005), improving dormancy breakdown (Farooq *et al.*, 2005), or increasing productivity (Hussein *et al.*, 2007).

The appropriate water holding capacity, consistency, and high porosity of peat moss are qualities that have contributed to its worldwide use as an ingredient of growing substrates and as a carrier for commercial bacterial inoculants (Eudoxie & Alexender, 2011). Rapidly decreasing reserves of exploitable non-renewable peat moss had led the price to increase, which will ultimately limit its use (Tariq *et al.,* 2012).

Poor seedling establishment was the major obstacle in sweet corn production (Zhao *et al.,* 2007). Sand priming significantly improved field emergence performance of all super sweet corn cultivars, and there was marked improvement by priming at 20ºC for 24 hrs (Zhao *et. al.*, 2009). In the present experiment, a priming method was developed using sand as a priming solid matrix (sand priming). Field emergence percentage (FEP) and field emergence speed (FES) are vital indices for field performance assessment since they directly affect crop yield and quality (Zhao *et al.*, 2007). Therefore, the effect of sand priming on improving the field emergence performance of super sweet corn was investigated. Many studies suggested that membrane system integrity, α-amylase activity and embryo protein contents were considered to be important indices of germination performance (Liu *et al.*, 1999; Black & Bewley, 2000; Farooq *et al.*, 2006). To further elucidate the effect of sand priming on the field emergence, we also analyzed the performance of super sweet corn, membrane system integrity, α-amylase activity and protein content.

Solid matrix priming was carried out with compost, press mud or gunny bags that are cheaper, as compared to calcined clay and Micro Cell E. First, of all 500 g seeds were mixed with 1 kg sterilized compost or press mud and 350 mL of distilled water in closed plastic containers. The containers were placed under shade at room temperature for 24 hrs. For gunny bag matriconditioning the seeds were placed within two saturated gunny bags for 24 hrs under shade (Bennett & Waters, 1987). Solid matrix priming helps to improve potential matrix (Cantliffe, 1997), while pre-soaking seed treatment with plant growth regulators in vegetables improve the growth (Nakamura *et al.*, 1982). The organic waste materials, mainly of plant and animal origin, provide a good source of organic matter and nutrients to improve soil productivity (Sarwar *et al.*, 2007; Sharma *et al.*, 2005). Animal manures (poultry and cattle) have also been effectively used as fertilizers for centuries and recognized as the most considerable natural fertilizers because of its low cost and high content of macro- and micronutrients for crop growth (Sarwar *et al.*, 2008; Sharpley & Smith, 1995; Sloan *et al.*, 2003). Composting is a useful strategy for the sustainable recycling of organic wastes (Tuomela *et al.*, 2000). Composting has become a preferred method for municipalities and industries to recycle a variety of organic by-products into a safer and more stabilized material for application to soil (Butler *et al.*, 2001; Carr *et al.*, 1995). The organic waste material used for the composting varies in their nutrient and mineral composition, as it affects the rate of decomposition of the compost and the composition of matured compost (Adegunloye *et al.*, 2009).

Application of plant growth regulators or nutrients during pre-soaking, priming and other pre-sowing treatments in many crops have improved seed performance that results in overall plant growth and productivity particularly under adverse conditions, such as temperature extremes or salinity (Taylor & Harman, 1990; Pill & Finch-Savage, 1998; Afzal *et al.,* 2008; Bakht *et al.,* 2011). Typical responses of priming are faster and closer spread of time to emergence, over all seedbed environments and wider temperature range of emergence, which leads to better crop stand, hence improved the yield and harvest quality, especially under suboptimal and stress growing conditions in the field (Halmer, 2004). Furthermore, the primed seeds often germinate and emerge more rapidly than non-primed seeds, especially at low temperatures (Bodsworth & Bewley, 1981; Murray, 1990; Zheng *et al.,* 1994). Although proper exogenous application of plant hormones along with nutrients, antioxidants, organic and inorganic chemicals promotes plant growth and development, however, these are not cost effective and out of the reach of farmers. So, efforts should be made to explore the cheaper and best alternatives of expensive priming agents. Among different natural sources used to extract plant growth regulators, moringa (*Moringa oleifera* L.) is gaining a lot of attraction (Foidl *et al.,* 2001). Moringa belongs to family Moringaceae. There are about 13 species of moringa of which *M. oleifera* is most widely grown. Since leaves of moringa are rich in zeatin, it can be used as natural source of cytokinin (Fuglie, 1999; 2001). In addition, moringa leaf is also rich in ascorbates, carotenoids, phenols, potassium and calcium, which have plant growth promoting capabilities and often applied as exogenous plant growth enhancers (Foidl *et al.,* 2001).

MATERIAL AND METHODS

Plant material

Seeds of spring maize hybrid Pioneer P-1543 were obtained from Pioneer seeds (Pvt.) Ltd., Sahiwal, without any chemical treatment. Pioneer P-1543 is a yellow kernel double cross hybrid.

Seed priming treatments

Following seed priming techniques were used to observe the performance of spring maize hybrid P-1543: T1 = Control; T2 = Priming with press mud; T3 = Priming with peat moss; T4 = Priming with sand; T5 = Priming with gunny bags; T6 = Priming with compost; T7 = Priming with farm yard manure (FYM); T8 = Priming with moringa leaf extract (MLE).

Seed riming protocol

T1 = Control non-primed; T2 = Solid matrix priming with saturated press mud of sugar mill for 24 hrs; T3 = Matriconditioning with saturated peat moss for 24 hrs; T4 = Matriconditioning with saturated sand for 24 hrs; T5 = Solid matrix priming with saturated gunny bags for 24 hrs; T6 = Solid matrix priming with saturated compost for 24 hrs; T7 = Matriconditioning with saturated farm yard manure for 24 hrs; T8 = Osmotic priming, seeds were primed using aerated low water potential with moringa leaf.

Extract (MLE) solution for 24 hrs (Bennet & Waters, 1987). During priming period, continuous fresh air was supplied through plastic pipes connected with an aeration pump. During priming seed weight to solution, volume ratio I: 10 (w/v) was maintained (Farooq *et al.*, 2005). After treatment, seeds were rinsed carefully with distilled water and seeds were dried back near to their original moisture level under shade (Lee & Kim, 2000). After priming, the seeds were used for raising primed nursery and direct planting. The untreated dry seeds were used as control and raised control nursery.

To obtain MLE, fresh leaves were collected from the mature trees of moringa planted in the Agronomic Research area at the University of Agriculture, Faisalabad. Juice was extracted according to the method of Foidl *et al.* (2001), with the use of a locally manufactured juice extractor. The juice extracted from moringa leaves was diluted with distilled water to make a 3% concentration.

Post priming operations

After osmo-conditioning or matriconditioning, seeds were given three washings with distilled water (Khan, 1992) and redried to original weight with forced air under shade. Seed moisture contents were determined by using three seed samples, according to the recommendations of Ellis *et al.* (1985). Seed moisture was maintained at 15%. Four replicates of 12 seeds each were germinated in 9 cm diameter Petri dishes on two moistened filter papers at 12 ± 3°C in an incubator (Sanyo, England). The filter papers were moistened everyday with distilled water throughout the experiment. A seed was considered germinated on radicle visibility after counting on daily basis. The experiment was assesed day to day. Number of emerged seeds were recorded daily, according to the seedling evaluation. Mean germination time (MGT) and time to attain 50% germination (T50) were calculated on the basis of equations of Ellis & Roberts (1981) and Coolbear *et al.* (1980), respectively. Emergence index (EI) was calculated according to the handbook of the Association of Official Seed Analysts (1983), as per following formulae:

$$EI = \frac{\text{No. of germinated seeds}}{\text{Days of first count}} + \ldots\ldots\ldots + \frac{\text{No. of germinated seeds}}{\text{Days of final count}}$$

Final emergence percentage (FEP) was taken at the end of experiment. It represents the ratio, in percentage of final number of emerged seedlings to total seeds planted.

$$FEP\,(\%) = \frac{\text{Final number of seedlings emerged}}{\text{Total number of seeds sown}} \times 100$$

After 12 days, root and shoot lengths were measured by using scale, while seven plants were selected randomly from each petri dish. Data was analyzed statistically for significance by applying LSD test for separation of treatment means using SAS 9.2.

RESULTS AND DISCUSSION

Final germination percentage, mean germination time, first day of germination, last day of germination, germination index, time to gain 50% germination, root length of seedlings and shoot length of seedlings shows significant difference among treatments, whereas the ratio of root and shoot length shows non-significant behavior. Priming with moringa leaf extract, seeds shows maximum final germination percentage, followed by priming with gunny bags and farm yard manure, whereas minimum germination percentage was recorded under peat moss priming, which is similar to control (non-prime) and priming with press mud, sand and compost (*Table 1*). Alternatively, minimum mean germination time was observed in moringa leaf extract primed seeds, while maximum germination time was recorded under primed seeds with farm yard manure that is statistically similar to all other treatments (*Table 1*). First day of germination also have great importance for seedling establishment. Minimum days to start of germination was seen in seeds that are primed with moringa leaf extract and the maximum days to start germination was recorded in non-primed seeds, followed by priming with press mud, gunny bags and farm yard manure. However, priming with peat moss, sand and compost shows average days between maximum and minimum days to start emergence (*Table 1*). Minimum final days to complete germination was recorded under moringa leaf extract primed seeds, while maximum days for germination observed under sand primed seeds, which is statistically similar to all other treatments except the moringa leaf extract primed seeds (*Table 1*). Highest germination index was recorded under moringa leaf extract primed seeds, followed by seeds that are primed with press mud, peat moss, sand and farm yard manure, while lowest germination index was observed under control non primed seeds (*Table 1*).

Table 1 - Effect of different seed invigoration techniques on stand establishment

Treatments	FGP (%)	MGT (d)	FDG (d)	LDG (d)	GI
Control (non-prime)	88.67 bc	10.30 a	6 a	11 a	5.46 c
Priming with press mud	88.78 bc	10.38 a	6 a	11 a	7.92 ab
Priming with peat moss	83.79 c	10.69 a	5 b	11 a	7.25 abc
Priming with sand	88.22 bc	11.06 a	5 b	12 a	6.90 abc
Priming with gunny bags	91.97 b	10.59 a	6 a	11 a	6.39 bc
Priming with compost	88.84 bc	10.92 a	5 b	11 a	6.34 bc
Priming with farm yard manure	91.42 b	11.21 a	6 a	12 a	7.62 abc
Priming with moringa leaf extract	99.67 a	7.34 b	4 c	8 b	8.77 a
LSD	6.49	1.37	0.63	1.31	1.99

Note: within a column, any two means not sharing a letter in common differ significantly at $P < 0.05$.
FGP – final germination percentage; MGT – mean germination time; FDG – first day of germination, LDG – last day of germination, GI – germination index.

Lowest time to gain 50% germination was recorded under moringa leaf extract primed seeds, followed by seeds primed with sand; however, the highest time was observed in primed seeds with peat moss, followed by primed seeds with farm yard manure, gunny bags, compost and control non-primed seeds (*Table 2*). Maximum root and shoot length was also observed in primed seeds with moringa leaf extract (MLE), while minimum root and shoot length was observed under peat moss primed seeds and sand primed seeds respectively (*Table 2*). Statistically root to shoot ratio shows non-significant effects between the treatments (*Table 2*).

Seed priming was observed to improve seed performance under sub-optimal temperature conditions (Lin & Sung, 2001). Priming also increases the environmental range better for germination, provides faster and synchronous seedling germination (McDonald, 1999). Seed priming enhance seed vigor (Bittencourt *et al.* 2005), improving dormancy breakdown (Farooq *et al.* 2005), or increasing productivity (Hussein *et al.*, 2007). The major issue for getting low maize yield is poor stand establishment. Improved stand establishment and increase in yield has been observed in many crops due to priming treatments with different seed priming agents (Murungu *et al.,* 2004; Farooq *et al.,* 2006; 2008a,b). Seed priming reduces the time between seed sowing and emergence; however, germination improved as compared to control (Chiu *et al.,* 2002). Furthermore, the primed seeds often germinate and emerge more rapidly than non-primed seeds, especially under low temperatures (Bodsworth & Bewley, 1981; Murray, 1990; Zheng *et al.,* 1994).

Table 2 - Effect of different seed invigoration techniques on stand establishment and seed vigor

Treatments	T50 (days)	RL (cm)	SL (cm)	RL/SL
Control (non-prime)	8.38 abc	7.12 b	1.67 b	4.29
Priming with press mud	8.00 bc	7.16 b	1.71 b	4.26
Priming with peat moss	9.13 a	6.38 b	1.48 b	4.47
Priming with sand	7.72 c	7.07 b	1.45 b	5.01
Priming with gunny bags	8.16 abc	8.01 b	1.77 b	4.49
Priming with compost	8.36 abc	7.04 b	1.67 b	4.25
Priming with farm yard manure	8.86 ab	8.02 b	1.79 b	4.56
Priming with moringa leaf extract	5.50 d	10.95 a	2.96 a	3.81
LSD	0.98	1.84	0.57	NS

Note: within a column, any two means not sharing a letter in common differ significantly at $P< 0.05$.

T50 – time to gain 50 % seedling germination; RL – root length; SL – shoot length; RL/SL – root to shoot ratio.

The observed findings showed that priming with moringa leaf extract may increase one or more parameters at cellular level (carbohydrate and proteins) or water content inside the dry seed and the start of enzymatic activity, which ultimately enhance germination by increasing final germination percentage, germination index, root length and shoot length, whereas lowering mean germination time (MGT), time to gain 50% germination (T50), first day of germination (FDG) and last day of germination (LDG) (Afzal *et al.*, 2009). MLE contains natural plant growth enhancer compounds, such as zeatin, Ca, and K, while magnetic water may also increase enzymatic activity and carbohydrate metabolism. These results were similar to the findings of Ali *et al.* (2011), that MLE treated maize have higher root growth and shoot growth.

Exposure of seeds to almost all priming techniques enhanced the germination of maize seeds and stand establishment of maize seedlings as well. The moringa leaf extract treatment are graded at the highest, followed by treatments of peat moss, press mud, sand, gunny bags, compost, farm yard manure, compared with the control non-prime. The impact of moringa priming on maize seed germination and seedling growth was found almost similar, but its best selection depends on its environment-friendly property and its cheapness along with common availability to the poor farmers.

CONCLUSION

Most of the treatments with priming have the capacity to enhance germination, early stand establishment and higher vigor of maize seeds. Generally, priming with moringa leaf extract (MLE) performed better due to higher final germination percentage (FGP), germination index (GI), root length (RL), shoot length (SL) and lower mean germination time (MGT), time to gain 50% germination (T50), first day of germination (FDG) and last day of germination (LDG) with all other treatments.

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