
EFFECT OF SEED PRIMING ON GERMINATION PERFORMANCE AND YIELD OF OKRA (*ABELMOSCHUS ESCULENTUS* L.)

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ABSTRACT:- To overcome the problem of slow and erratic emergence in okra, two experiments, i.e., laboratory and field were carried out during 2013. In the laboratory experiment, okra seeds were primed in PEG-8000 (polyethylene glycol) and Mannitol solutions (0, -0.4, -0.8, -1.2, -1.6, -2 and -2.4 Mpa osmotic potential) for 6, 12, 18 and 24 hours while dry seeds were used as control. Same primed seeds were used in the field experiment. Priming proved effective in improving percent germination and reducing the mean germination time (MGT) as compared to unprimed seeds. Priming agents, its osmotic potential and duration significantly affected germination percentage, which increased as osmotic potential lowered from 0 to -1.2 Mpa while further lowering osmotic potential to -1.6 Mpa and below adversely affected germination. Similarly, germination percentage significantly enhanced as priming duration increased from 6h to 12h but further increase in duration from 12h to 18h caused insignificant increase in germination percentage. Increase in priming duration beyond 18h, however, resulted in non-significant decrease in percent germination. Amongst priming agents, PEG gave better germination than Mannitol, however, priming agents were ineffective in lowering MGT. Increase in osmotic potential from -2.4 to 0 Mpa and duration from 6h to 24h caused significant reduction in MGT. Field experiment results showed significant improvement in yield from primed seeds as compared to unprimed seeds. Seeds primed with PEG gave significantly a higher yield than Mannitol. Maximum yield was observed at -1.2 Mpa osmotic potential. Like MGT, days to 50% emergence in the field were unaffected by priming agent but significantly affected by osmotic potential, reduction in days to 50% emergence occurred as osmotic potential increased from -2.4 to 0 Mpa

Key Words: Abelmoschus esculentus; Seed Priming; Polyethylene Glycol; Mannitol; Osmotic Potential; Yield; Yield Components; Pakistan.

INTRODUCTION

Seed priming is a pre-sowing seed treatment in which seed is allowed to imbibe enough water to start pre-germinative metabolic processes but insufficient for radicle protrusion be-

cause with radicle protrusion seed loses its desiccation tolerance (Posmyk et al., 2001). After hydration, treated seeds are dried back to their initial moisture content. This drying back process is called hydration dehydration and is practiced to

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minimize deterioration in seed during storage. Priming improves seed germination performance by starting early processes of germination but not cell division (Yuan et al., 2010). Metabolism that occurs during priming is not enough to cause radicle emergence (McDonald, 2000).

There are two types of seed priming, in one type water penetrate freely into seed which is called hydro-priming while in other type seed hydration is controlled. If controlled hydration is achieved through the addition of solute to water then it is called osmopriming or if a solid matrix is used to provide controlled seed hydration then it is called solid matrix priming (Pill and Necker, 2001).

Seed priming improves germination performance of seed (Kausar et al., 2009). Osmopriming strengthens the antioxidant system and increases seed germination potential, resulting in an increased stress tolerance in germinating seeds (Chen and Arora, 2011). Response of seed to priming is affected by priming duration, osmotic potential of priming solution (Arif et al., 2008), priming agent (Farooq et al., 2005) and oxygen supply to seed (Nascimento, 2003).

In okra seed, slow and erratic emergence is the main problem which results in low fertilizer efficiency, unsynchronized harvesting and ultimate low yield. The main goal of this research work was to overcome the problem of slow and erratic emergence in okra seed through seed priming and to find out optimum seed priming duration, osmotic potential and priming agent for okra seed priming.

MATERIALS AND METHOD

Laboratory Experiment

This experiment was carried out to overcome the problem of slow and erratic emergence in okra seeds through seed priming. In this experiment the effect of seed priming on germination percentage and mean germination time (MGT) was studied in the laboratory of Horticulture Department, Khyber Pukhtunkhwa Agricultural University, Peshawar, during 2013. In this experiment, okra seeds of variety Sabz pari were primed in Polyethylene glycol 8000 (PEG 8000) and Mannitol solutions at 21° C with osmotic potentials 0, -0.4, -0.8, -1.2, -1.6, -2 and -2.4 Mpa for 6, 12, 18 and 24 hours while unprimed dry seeds were used as control. Osmotic potentials of priming solutions were determined according to Michel (1983). Aquarium pump was used for adequate supply of oxygen to the priming solution. After priming, seeds were rinsed with water and dried at room temperature to initial moisture content (11%). After drying, 20 seeds for each treatment in three repeats were placed in Petri dishes on blotting paper moistened with water. Petri dishes were kept in an incubator at 21o C and germination count was done at 6h interval till final germination, the experiment lasted for about one week. Mean germination time (MGT) was determined by the following formula of Ellis and Roberts (1981).

$$MGT = \sum n \times g / N$$

where,

n = Number of seeds germinated on time g

N= Total number of germinated seeds.

Field Experiment

This experiment was conducted during 2013 at Agricultural Research Farm of Khyber Pukhtunkhwa Agricultural University, Peshawar, Pakistan, to study the role of seed priming in controlling the problem of slow emergence under field conditions and in enhancing yield of okra. Seeds were primed in PEG 8000 and Mannitol solutions. On the basis of the 1st experimental results, 12h priming duration was used for 0 Mpa osmotic potential and 18h priming duration was used for osmotic potential -0.4, -0.8, -1.2, -1.6, -1.2 and -2.4 Mpa while dry seeds were used as control. After priming, the seeds were rinsed with water and then dried to initial moisture content at room temperature. After drying, 100 seeds of each treatment were sown on April 20, 2013 in well prepared plots of 6.75 m² with seed to seed distance of 15 cm and row to row distance of 45cm. Data was recorded on days to 50% emergence and yield.

Experimental Design

In laboratory experiment, 64 treatments (four durations, two priming agents and seven levels of the priming agents with the eighth one as control i.e., dry seeds) with three repeats were analyzed in completely randomized design (CRD), with three factors.

The field experiment was laid out in Randomized Complete Block Design (RCBD), with two factors for analyzing 16 treatments replicated thrice (two priming agents with seven

levels of each while dry seeds were used as the eighth level.

RESULTS AND DISCUSSION

Laboratory Experiment

Seed Priming Effect on Percent Germination

Analysis of the data revealed that priming agents, osmotic potential, duration and interaction regarding priming agent and osmotic potential significantly affected percent germination of okra, while interaction of priming agent \times duration, osmotic potential \times duration and interaction of the three (priming agent, osmotic potential and duration) had insignificant effect on percent germination.

It is evident from data that seed priming improved seed germination percentage as compared to control (Figure 1). Data also showed that PEG as a priming agent, germinated more okra seeds (78.4%) at -1.2 Mpa osmotic potential (Table 1). Osmotic

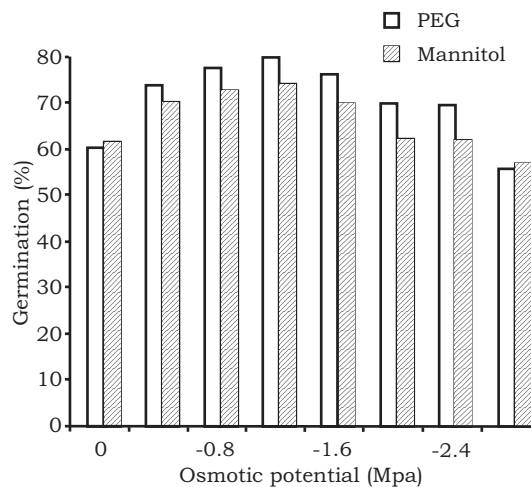


Figure 1. Percent germination of okra seeds as affected by seed priming agents at various osmotic potential

Table 1. Mean germination time (MGT) of okra seeds as affected by seed priming, osmotic potential of priming solution and duration of priming

Osmotic potential (Mpa)	Duration (h)				Mean
	6	12	18	24	
0	21.8 ^{efg}	12.7 ^m	7.7 ^p	0 ^r	10.5 ^h
-0.4	22 ^{efg}	14.5 ^l	10.2 ^o	3 ^q	12.4 ^g
-0.8	22.5 ^{def}	15.5 ^{kl}	11.8 ^{mmo}	7 ^p	14.2 ^f
-1.2	24 ^{cd}	18.2 ^{ij}	15.2 ^{kl}	10.3 ^o	16.9 ^c
-1.6	24 ^{cd}	20.5 ^{gh}	16.5 ^{jk}	10.8 ^{no}	17.9 ^d
-2	25.5 ^{bc}	21 ^{fgh}	19.3 ^{hi}	12.5 ^{mn}	19.6 ^c
-2.4	25.8 ^b	23.5 ^{de}	19.5 ^{hi}	14.7 ^l	20.9 ^b
Unprimed	28.2 ^a	28.3 ^a	28.8 ^a	27.8 ^a	28.3 ^a
Mean	24.2 ^a	19.3 ^b	16.1 ^c	10.8 ^d	

LSD at 5% level of probability for osmotic potential=0.87

LSD at 5% level of probability for duration=0.61

LSD at 5% level of probability for osmotic potential × duration=1.75

Means followed by same letters do not differ significantly

potential of PEG solution lesser or above than -1.2 Mpa reduced germination, in other words osmotic potential below or above -1.2 Mpa negatively affected seed germination of okra. However, reduction for PEG solution at osmotic potentials immediately below or above -1.2 Mpa i.e., at -0.8 and -1.6 Mpa was non-significant. Like PEG, Mannitol also showed highest germination (72.7%) at -1.2 Mpa but was significantly lower than that recorded in PEG at this osmotic potential. The data, however, indicated that PEG at -0.4 Mpa (72.6%) was as effective as Mannitol at -1.2 Mpa (72.7%). Data also showed that drop in osmotic potential (0 to -1.2 Mpa) improved germination but further drop in

osmotic potential (-1.2 to -2.4 Mpa) caused reduction in germination.

Priming significantly improved germination percentage as compared to unprimed. The possible reason for improved germination through priming may be synthesis of proteins and leaching of growth inhibitors (Khan et al., 1977; Bray et al., 1989), repair of deteriorative DNA in seeds (Di Girolamo and Barbanti, 2012) and activation of antioxidant enzymes which lower peroxidation in seeds (Pukacka and Rajajczak, 2005). The finding that priming improved percent germination is in line with Arif et al. (2008) who had reported that priming improved germination.

Osmotic potential significantly affected germination percentage. Moosavi et al. (2009) had also reported that osmotic potential of priming solution effect percent germination. Yaseen et al. (1994) reported that high osmotic potential of priming solution caused rapid penetration of water into seeds thereby rupturing seed coat which encouraged auto-oxidation and leakage from seed resulting in poor germination. Very low osmotic potential (-1.6 and below) has resulted in poor germination of okra seeds in present study. According to Okamoto and Joly (2000) solution potential lower than certain level may cause hypoxia (low oxygen regime) which may in turn impair germination.

Priming duration affected percent germination because seeds of each species need specific amount of water to get into lag phase of germination in which all the pre-germinative metabolic processes occurs. So, if priming is done for short period then seed would not get enough water that is required for getting seed into

lag phase of germination. On the other hand priming for prolonged period will allow excess of water that may exceed the quantity required for the initiation of lag phase of germination and radicle protrusion will occur due to which seed lose its desiccation tolerance thereby results in loss of seed viability (Dekkers et al., 2015; Pereira et al., 2014). According to Okamoto and Joly (2000) prolonged submergence also causes hypoxia which may reduce germination percentage. Arif et al. (2008) had also reported that seed priming duration affected germination percentage. Results exhibited lowest germination (64.53%) for 6h seed priming. Highest germination (69.43%) was recorded for 18h which was statistically at par to 24h seed priming. Increasing priming duration beyond 24h may impair okra germination (Figure 2).

In priming agents better germination performance was observed for PEG 8000 than Mannitol. It might be due to the inert nature and comparatively larger molecular size of PEG which cannot penetrate in seed to cause toxicity (Pill et al., 1991).

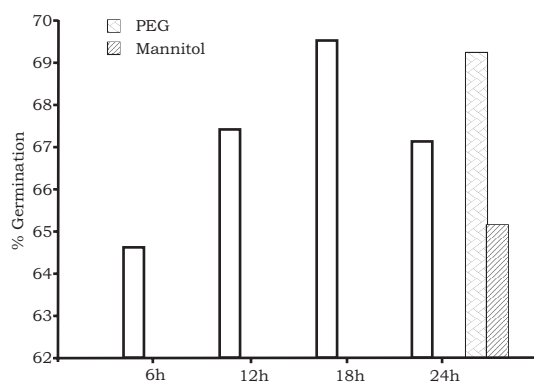


Figure 2. Percent germination of okra seeds as affected by seed priming duration

Seed Priming Effect on Mean Germination Time (MGT)

Analysis of the data revealed that osmotic potential, duration and their interaction had significant effect on MGT, whereas priming agents had insignificant effect on MGT. The data indicated that seed priming does reduce seed MGT as compared to control (Table 1). Priming with pure water had reduced MGT from 28.3h as in dry seeds (unprimed) to 10.5h.

Length of priming duration is important, priming for 24h has reduced mean germination time (MGT) to 10.8h as compared to 24.2h in priming for 6h. The data revealed that osmotic potential less than zero delayed MGT (Table 2). Seeds at -2.4 Mpa osmotic potential delayed germination as compared to that at 0 Mpa and other osmotic potentials.

About 5.88%, higher germination was found by priming with PEG than Mannitol (Figure 2). As concerned priming duration, results showed significant increase in percent germination as duration increased from 6h to 12h, however, further increase of duration from 12h to 18h caused insignificant increase while increasing priming duration from 18h to 24h caused insignificant decrease in percent germination.

Table 2. Days to 50% emergence as affected by seed priming

Priming Agent	Osmotic potential (Mpa)							Unprimed	Mean
	0	-0.4	-0.8	-1.2	-1.6	-2	-2.4		
PEG	4.33	5.00	6.00	6.00	7.00	8.00	8.00	7.66	6.50
Mannitol	4.00	4.66	5.66	6.33	6.33	8.00	8.00	8.00	6.37
Mean	4.1 ^d	4.8 ^{cd}	5.8 ^{bc}	6.1 ^{bc}	6.6 ^{ab}	8 ^a	8 ^a	7.83 ^a	

LSD at 5% level of probability for osmotic potential=1.58

Priming significantly reduced mean germination time (MGT) over unprimed seeds. Varietf et al. (2010) reported that priming activate and synthesize hydrolytic enzymes e.g. lipases, amylases and proteases which mobilize storage materials in seed. On rehydration quick emergence take place because all pre-germinative processes had already taken place. Pukacka and Ratajczak (2005) had reported that priming activates antioxidant enzymes which lower per oxidation in seed thereby maintaining seed vigour which may result in quick germination. Bray et al. (1989) and Davison and Bray (1991) reported protein synthesis in seeds whereas Burgass and Powell (1984) and Varietf et al. (2010) reported that priming cause repair of damaged DNA, due to protein synthesis and repair in seed during priming seed become invigorated which result in reduced MGT.

Field Experiment **Seed Priming Effect on Days to 50% Emergence.**

Mean values of the data showed that osmotic potential of priming solution had significant effect on days to 50% emergence while priming agents and their interaction with osmotic potential had no significant effect on days to 50% emergence (Table 1).

It indicated that unprimed dry seeds and seeds primed at lower osmotic potential i.e., from -2 to -2.4 Mpa took significantly more days to germination as compared to seeds primed at high osmotic potential except those seeds primed at -1.6 Mpa osmotic potential from which the difference was insignificant (Table 2). Priming seeds with pure water took significantly minimum days (4.1) to

germination as compared to seeds primed at osmotic potential lower than 0 Mpa except -0.4 Mpa osmotic potential wherein insignificant increase in days to emergence was observed. It is very logical, that at zero osmotic potential water rapidly penetrated in seed and activated germination processes. Whereas, at lower osmotic potential, water penetration into the seed slowed down and delayed germination. Quick emergence at 0 Mpa osmotic potential is not as important as the overall germination which is evident where priming at -1.2 Mpa had enhanced overall germination to 75.60% compared to 59.79% with pure water (Figure 1).

Priming at high osmotic potential (0 Mpa) significantly minimized (4.1) days to 50% emergence as compared to unprimed seeds (7.8) and seeds primed at low osmotic potential (Table 2). Farooq et al. (2010) and Khan et al. (1977) had reported that at high osmotic potential seeds absorb sufficient amount of water to start pre-germinative metabolic processes so on reimplantation seeds are ready to quick emergence because all the processes necessary for germination had already taken place.

Effect of Seed Priming on Yield ($t\ ha^{-1}$)

Mean values of the data showed that osmotic potential and priming agents significantly affected yield but their interaction was found insignificant (Table 3).

Data about the effect of osmotic potential on yield indicated that as osmotic potential falls from 0 to -1.2 Mpa, a significant increase of $1.1\ t\ ha^{-1}$ (14.7-13.6) occurred while further fall in osmotic potential (-1.6, -2.0 and -2.4 Mpa) resulted in significant

Table 3. Okra yield as affected by seed priming(t ha⁻¹)

Priming agents	Osmotic potential(Mpa)								Mean
	0	-0.4	-0.8	-1.2	-1.6	-2	-2.4	Unprimed	
PEG	13.8	13.9	14.4	14.9	14.2	13.05	12.5	12.4	13.6 ^a
Mannitol	13.5	13.9	14.4	14.6	13.9	12.60	12.2	12.3	13.4 ^b
Mean	13.6 ^c	13.9 ^c	14.4 ^{ab}	14.7 ^a	14 ^{bc}	12.82 ^d	12.4 ^e	12.4 ^e	

LSD at 5% level of probability for osmotic potential=0.40

Means followed by same letters in each column and row do not differ significantly

reduction. Maximum yield (14.7 and 14.4 t ha⁻¹) was observed at -1.2 and -0.8 Mpa, respectively followed by (14 t ha⁻¹) at -1.6 Mpa which was statistically at par with the yield at -0.8, -0.4 and 0 Mpa osmotic potential while minimum yield (12.4 t ha⁻¹) was recorded for unprimed seeds and seeds primed at -2.4 Mpa osmotic potential followed by 12.8 t ha⁻¹ at -2 Mpa osmotic potential. In priming agents more yield was recorded in plants raised from seeds primed with PEG 8000 as compared to Mannitol.

Seed priming at each osmotic potential except -2.4 Mpa osmotic potential significantly improved yield over unprimed seeds. Mohammadi (2009) had also reported that priming caused increase in yield.

Yield is the final objective and is a function of multiple factors which affect crop from their sowing to harvest. Seed priming with PEG, significantly enhanced final yield in okra crop. Since high and smooth germination is the first step in the later crop growth and yield. Seed priming with PEG had generally encouraged smooth germination over Mannitol (Table 1) and pure water (0 Mpa) treatment due to which more yield was obtained from plants raised from PEG primed seeds. Present

result is in agreement with Bakht et al. (2010) who also reported that priming agents affected yield. Significantly more yield was obtained from plants primed at -1.2 Mpa osmotic potential as compared to plants from seeds primed at other osmotic potential. This is because more seeds germinated at this osmotic potential. Too rapid or too slow penetration of moisture into seeds impairs the potential of seeds to germinate. It could be due to membrane rupturing and hampering food supply to germinating embryo.

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AUTHORSHIP AND CONTRIBUTION DECLARATION

S.No	Author Name	Contribution to the paper
1.	Dr. Inayat-Ur-Rahman	Conceived the idea, Wrote abstract, Methodology, Did SPSS analysis, Conclusion, Technical input at every step, Overall management of the article, Data collection, Data entry in SPSS and analysis, Results and Discussion, Introduction, References
2.	Dr. Shamsher Ali	Methodology, Did SPSS analysis, Conclusion, Technical input at every step, Overall management of the article, Data collection, Data entry in SPSS and analysis, Results and Discussion, Introduction, References
3.	Dr. Mukhtar Alam	Conclusion, Technical input at every step, Overall management of the article, Data collection, Data entry in SPSS and analysis, Results and Discussion, Introduction, References
4.	Dr. Abdul Basir	Conclusion, Technical input at every step, Overall management of the article, Data collection, Data entry in SPSS and analysis, Results and Discussion, Introduction, References
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6.	Dr. Hidayat Ullah	Data collection, Data entry in SPSS and analysis, Results and Discussion, Introduction, References
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