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Standardization of priming duration of okra seed

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Abstract
Seed priming promote rapid and more uniform seed germination. In this study the pre-sowing effects of seed priming treatments on some physiological viz. seed germination, seedling vigour and seedling dry weight of okra were investigated. Seeds were subjected to seed priming methods, namely, hydropriming and osmoprimig. Hydropriming comprising a total of 9 treatments of different priming durations (16hrs – 72hrs) along with control (Non-primed). Hydropriming for 24h significantly increased the seed germination, seedling vigour, seedling length seedling dry weight and seeds primed with PEG 6000 (–0.5M Pa) for 24 hours exhibited best results in terms of characters like speed of germination, germination percentage, seedling length and dry weight, vigour index-I and II in okra cv. P-8.

Keywords: hydropriming, osmopriming, poly ethyl glycols, crd, speed of germination, seedling length and dry weight, vigour index

Introduction
Okra [*Abelmoschus esculentus* (L.) Moench] is a native crop of Tropical Africa that belongs to the family Malvaceae. For its robust nature, dietary fibers and distinct seed protein balanced in both lysine and tryptophan amino acids; it is also called "a perfect villager's vegetable" (Kumar et al., 2010) [8]. Okra seeds germinate very slowly and unevenly although they are viable seeds. Reduced, delayed, and erratic emergence is a serious problem in okra cultivation caused by seed hardness as it creates problems in rapid germination and uniform field stand (Purquerio et al., 2010) [12]. The hard seed coat restricts the water imbibition and uniform growth and development of the embryo and as a result interferes with seed germination (Mereddy et al., 2015) [9]. The problem of low germination due to the hard seed coat in okra can be overcome by seed priming. Seed priming is the process of controlled hydration of seeds which is potentially able to promote rapid and more uniform seed germination and plant growth (Sharma et al., 2014) [16]. Priming allows some of the metabolic processes necessary for germination to occur without germination taking place. Seed priming induced synchronized germination, increased seed vigor, and growth of seedlings under stressful conditions i.e. increase in germination and emergence rate (Bajehbaj, 2010) [4]. Different seed priming methods has been used to enhance germination and seed vigor of okra. Among them, Hydro-priming i.e. seed soaking in pure water and re-drying to original moisture content before sowing; Osmo-priming i.e. soaking the seed in a solution of osmoticum. The experiment aimed to study the Standardization of priming duration both hydropriming and osmopriming for overcoming the germination hindrance of okra seeds.

Materials and Methods
The present investigations was carried out in the Laboratory of Department of Seed Science and Technology, Dr Y S Parmar University of Horticulture and Forestry, Nauni, Solan, HP during the year of 2017-18. The details of materials used and techniques employed during the course of investigations are presented below.
The present studies were conducted as two different experiments:

1. Standardization of hydropriming duration of okra seed: For hydro priming treatment, seeds were completely immersed in water in ratio of 1:4 (one part of seeds and 4 parts of water). 2000 seeds was taken for each treatment of hydropriming. After hydro priming, the seeds were shade dried to reduce the moisture content to 8 per cent. The okra seeds (2000 seeds each for treatments) were hydroprimed for 8 hours intervals up to 72 hours. The treatment details are as below:

Table 1: Treatment details for Hydropriming

Treatments	Hydropriming durations
T_1	Hydropriming for 16 hours
T_2	Hydropriming for 24 hours
T_3	Hydropriming for 32 hours
T_4	Hydropriming for 40 hours
T_5	Hydropriming for 48 hour
T_6	Hydropriming for 56 hour
T_7	Hydropriming for 64 hour
T_8	Hydropriming for 72 hour
T_9	Control

Methodology of hydropriming

Hydro-priming of involves complete immersion of okra seeds 2000 for each treatments in water for different duration (16 , 24, 32, 40, 48, 56, 64 and 72 hours) before sowing and may followed by the shade dried to reduce the moisture content to 8 per cent.

Experiment 2: Standarsization of osmopriming duration of okra

Method of osmopriming: Seed priming was carried out in the laboratory of the Department of Seed Science and Technology, Dr YS Parmar University of Horticulture and Forestry, Nauni, Solan during 2017-18. First of all, 2000 seeds per treatment were counted, weighed and put into sterilized plastic petri plates after words seed priming is done with polyethylene glycol 6000 (PEG-6000). The osmotic potentials of -0.5 and -1.0 M Pa were prepared by dissolving 202.13 and 295.713 g of PEG-6000 in one liter of water, respectively (Nienow and Bujaski, 1991) [11]. The petri plates were kept at room temperature (26 ^{0}C). For the different duration (16, 24, 32, 40, 48 hours). After seed priming for different duration, the seeds were taken out and washed with tap water 3-4 times. The primed seeds were then dried in shade to their original moisture content of 8 per cent before storage at room temperature.

Table 2: Treatment details for Osmopriming

T_1	Priming with PEG-6000(-0.5 M Pa) for 16 hours
T_2	Priming with PEG-6000(-0.5 M Pa) for 24 hours
T_3	Priming with PEG-6000(-0.5 M Pa) for 32 hours
T_4	Priming with PEG-6000(-0.5 M Pa) for 40 hours
T_5	Priming with PEG-6000(-0.5 M Pa) for 48 hours
T_6	Priming with PEG-6000(-0.5 M Pa) for 56 hours
T_7	Priming with PEG-6000(-0.5 M Pa) for 64 hours
T_8	Priming with PEG-6000(-0.5 M Pa) for 72 hours
T_9	Priming with PEG-6000(-1.0 M Pa) for 16 hours
T_{10}	Priming with PEG-6000(-1.0 M Pa) for 24 hours
T_{11}	Priming with PEG-6000(-1.0 M Pa) for 32 hours
T_{12}	Priming with PEG-6000(-1.0 M Pa) for 40 hours
T_{13}	Priming with PEG-6000(-1.0 M Pa) for 48 hours
T_{14}	Priming with PEG-6000(-1.0 M Pa) for 56 hours
T_{15}	Priming with PEG-6000(-1.0 M Pa) for 64 hours
T_{16}	Priming with PEG-6000(-1.0 M Pa) for72 hours
T_{17}	Control

Seed quality parameters

Speed of germination

High speed of germination is an indication of vigourous seed lot. Number of germinated seeds are counted every day from the first day and the cumulative index is made by the formula (Maguire, 1962),

Speed of germination = $\Sigma(n/t)$

Where n is the number of seeds newly germinating at time t and t is days from sowing.

Germination (%)

The germination test was carried out as per ISTA procedure (Anonymous, 1985). Four hundred seeds from each treatment were taken and the test was carried out in four replications, having 100 seeds each. The seeds were allowed to germinate using between paper method at 25 °C. The germination count was taken on 4th day of the test. Germination percentage was worked out by using the following formula:

$$\text{Germination (\%)} = \frac{\text{Number of normal seedlings germinated}}{\text{Total number of seeds kept for germination}} \times 100$$

Seedling length (cm)

Ten normal seedlings, selected at random at first count were used to work out the seedling length. Seedling length was worked out by taking the total length of seedlings from the tip of the primary leaf to the tip of primary root with the help of scale and expressing the mean value in centimeters (cm).

Seedling dry weight (mg)

Ten normal seedlings selected for measuring seedling length were used to work out seedling dry weight. Seedlings were put in butter paper pocket and kept in oven at 60°C for 48 hours. Seedling dry weight was recorded and the mean value was expressed in milligrams (mg)

Seed vigour index-I

Seed vigour index-I was calculated as per the formula given by Abdul-Baki and Anderson (1973).

Seed vigour index-I = Germination (%) × Seedling length (cm)

Seed vigour index-II

Seed vigour index-II was calculated as per the formula given by Abdul-Baki and Anderson (1973).

Seed vigour index-II = Germination (%) × Seedling dry weight (mg)

Results and Discussion

Standardization of hydropriming duration of okra seeds

Seed quality parameters: Maximum germination (83.40), speed of germination (24.52), seedling length (10.90), seedling dry weight (21.50mg), seed vigour index-I (909.00), seed vigour index-II (1786.10.) were recorded with hydro priming duration of 24 hrs. The possible reason for enhancing germination, speed of germination, seedling length, seedling dry weight, seed vigour index I and II at 24 hrs duration might be due to fact that during seed priming, there is a rapid initial uptake of water leading to ignition of metabolic processes making condition favorable for earl and fast germination 9 (Arif, 2005). Further, space is developed in the primed seed that facilities water uptake ther by accelerating the speed of germination (Argerch and Bradford, 1989) [2]. Moreover, physiological and biochemical changes might have increased physiological activity of the embryo and mobilized the food reserves into the growing seedling length (Doijode and Raturi, 1987) [5] that might have resulted in increased seedling length, seedlings dry weight and ultimately enhanced seedling vigour. On other hands minimum values recorded for the said parameters at higher priming duration may be due to adverse effect of prolonged soaking because of decrease in DNA repair activity (Van Pijlen *et al*., 1996) [19].

Table 3: Results of standardization of hydropriming duration of okra seeds

Treatment	Germination (%)	Speed of germination	Seedling length (cm)	Seedling dry weight (mg)	SVI-1	SVI - 2
T1	82.355	23.240	10.300	20.900	846.370	1,717.040
T2	83.400	24.520	10.900	21.250	909.000	1,786.100
T3	81.000	22.470	10.125	19.600	818.900	1,587.450
T4	79.500	21.000	9.300	18.920	739.150	1,504.150
T5	76.750	20.120	8.550	18.420	663.250	1,427.750
T6	76.000	18.750	7.750	17.250	658.500	1,395.000
T7	74.250	18.250	7.000	16.500	649.750	1,307.500
T8	72.750	17.500	6.000	15.250	639.750	1,237.500
T9 (Control)	71.250	17.750	5.250	14.750	625.000	1,100.000
C.D	2.014	2.334	1.297	2.616	13.403	133.418

Standarsization of osmopriming duration of okra

Seed quality parameters: Maximum speed of germination (39.66) for priming was observed when seeds were primed with PEG-6000(-0.5 M Pa) for 24 hour and minimum speed of germination (18.50) was recorded with non primed seeds. The possible reason for fastest speed of germination of the seeds primed with PEG 6000 –0.5 MPa for 24 hrs at 7 days after priming is may be due to fact that seeds primed with polyethylene glycol may have rapidly imbibed and revived the seed metabolism resulting in higher and faster germination rate and reduction in the inherent physiological heterogeneity in germination. Almost identical results have been reported by Rowse (1995). Maximum Germination (95.35%) for priming was observed when seeds were primed with PEG-6000(-0.5 M Pa) for 24 hour and minimum germination percentage was recorded with non-primed seeds. Possible reason for increasing seed germination after priming might be due to stimulation of hydrokytic enzymes, which bring about breakdown of stored food material into sugers and thus promoting germination through cell division. This result may be confirm by Singh (1999) in tomato seeds. The poor per cent germination in non-primed seeds might be due to decrease in water uptake by the seed due to low water potential of the germination medium, improper activation of enzyme activities (Ashraf 1988). Maximum seedling length (16.90) for priming was observed when seeds were primed with PEG-6000(-0.5 M Pa) for 24 hour and minimum seedling length was recorded with non-primed seeds. It may be due to enhanced enzymatic activities of catalase, peroxidase, amylase and invertase in the seeds treated with PEG 6000 -0.5 MPa for 24 hrs. There may be an increase in protein, sugar and RNA content which have resulted in quicker germination and better growth resulting into more seedling length. Almost identical results have been expressed in PEG treated seed by Singh (1984) [17]. Maximum seedling dry weight (29.56) for priming was observed when seeds were primed with PEG-6000(-0.5 M Pa) for 24 hour and minimum seedling dry weight was recorded with non primed seeds. The increased dry weight due to osmopriming may be due to beneficial effects of osmopriming on seed structure, biochemistry, enzyme activities and organic substances in germinating seeds as reported by several workers.

Table 3: Results of standardization of osmopriming duration of okra seeds

Treatment	Germination (%)	Speed of germination	Seedling length (cm)	Seedling dry weight (mg)	SVI-I	SVI - II
T1 Priming with PEG-6000(-0.5 M Pa) for 16 hour	92.50	36.93	16.15	22.66	1493.80	2,509.0
T2 Priming with PEG-6000(-0.5 M Pa) for 24 hour	95.35	39.66	16.90	29.56	1620.95	3063.10
T3 Priming with PEG-6000(-0.5 M Pa) for 32 hour	90.83	36.18	16.20	27.80	1469.27	2377.42
T4 Priming with PEG-6000(-0.5 M Pa) for 40 hour	90.22	32.30	15.35	27.20	1385.51	2336.26
T5 Priming with PEG-6000(-0.5 M Pa) for 48 hour	86.72	29.13	14.18	26.38	1229.30	2288.20
T6 Priming with PEG-6000(-0.5 M Pa) for 56 hour	85.25	28.25	13.0	25.35	1160.0	2150.0
T7Priming with PEG-6000(-0.5 M Pa) for 64 hour	83.0	27.000	12.000	24.37	1075.000	2055.0
T8Priming with PEG-6000(-0.5 M Pa) for 72	82.500	25.250	11.0	22.62	1007.50	1995.0
T9Priming with PEG-6000(-1.0 M Pa) for 16 hour	83.20	26.85	13.730	25.15	1145.33	2672.76
T10 Priming with PEG-6000(-1.0 M Pa) for 24 hour	90.38	37.0	16.08	30.38	1452.78	2151.49
T1 Priming with PEG-6000(-1.0 M Pa) for 32 hour	89.38	34.70	12.63	23.83	1128.32	2098.99
T12 Priming with PEG-6000(-1.0 M Pa) for 40 hour	88.16	28.0	11.88	23.00	1046.75	2026.08
T13 Priming with PEG-6000(-1.0 M Pa) for 48 hour	85.91	25.38	11.23	22.23	965.41	1910.96
T14 Priming with PEG-6000(-1.0 M Pa) for 56 hour	85.0	24.50	9.50	21.11	907.50	1867.50
T15 Priming with PEG-6000(-1.0 M Pa) for 64 hour	83.50	23.25	8.75	19.80	832.50	1,785.0
T16 Priming with PEG-6000(-1.0 M Pa) for 72 hour	80.50	21.50	7.50	18.25	782.50	1600.0
Control	77.0	18.50	6.250	17.00	687.50	1425.0
C.D	3.298	2.988	3.124	2.881	102.058	152.29

This present are in line with those of Khalil *et al.* (1997) [7] who reported that plant raised from seed preconditioned in PEG-8000 exhibited higher dry weight compared to plants raised from untreated seeds which supports our findings. Improvement in seedling dry weight has also been reported by Nagarajan *et al.* (2003) [10] who were of the opinion that priming treatments increased the activities of dehydrogenases (an indicator of seed viability) and peroxidises (free radical scavenging enzyme). It appears that not only hydration but enzymatic activities of catalase, peroxidise, amylase and invertase got increased in PEG treated seeds besides increasing the protein, sugar and RNA content (Saxena (1976 and 1980) [13, 15] The poor seedling dry weight in non-primed seeds might be due to decrease in water uptake by the seed due to low water potential of the germination medium, improper activation of enzyme activities (Ashraf *et al.* 2002)

[3]. Maximum seed vigour index (1620.95) for priming was observed when seeds were primed with PEG-6000(-0.5 M Pa) for 24 hour and minimum seed vigour index –I was recorded with non primed seeds. Higher seed vigour index is may be due to due to the maintenance of controlled but sufficient hydration of seed to a level that permits pregerminative metabolic activity to proceed but prevent the actual radicle emergence (Thakur *et al*., 1997) [18] in bell pepper and increased stand establishment in wheat (Yari *et al*. 2011) [20].

Conclusion

Priming of okra seeds might be the best option to overcome the reduced and delayed germination in okra seeds caused by seed hardness. Priming of seed before sowing facilitates the plant growth and development and its yield. Okra seed priming with different treatments on seed germination and seedling vigor revealed that the Hydropriming for 24 hrs and seeds primed with PEG 6000 (–0.5M Pa) for 24 hours exhibited best results in terms of seed quality characters in okra cv. P-8. So, seed priming is a useful technique for improving the germination percentage, germination rate, seedling growth, seed vigiour index I and II.

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