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Effect of pre-sowing invigoration seed treatments on germination behaviour and seedling vigour in Wheat (*Triticum aestivum* L.) Seeds.

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Abstract
The experiment was conducted in Post Graduate Laboratory, Department of Genetics and Plant Breeding, Sam Higginbottom University of Agriculture, Technology and Sciences, Allahabad (U.P.), in order to standardize the suitable method of priming for Wheat seeds (var. DBW-17). Four methods of priming viz., T_1-Unprimed (Control), T_2 and T_8 Hydro-priming with Distilled water, T_3 and T_9 Osmo-priming with PEG 20%, T_4 and T_8 Halo-priming with NaCl 1%, T_5 and T_{11} Halo-priming with $CaCl_2$ 1%, T_6 and T_{12} Organic priming with Tulsi leaf extract 5% and T_7 and T_{13} Organic priming with Neem leaf extract 5% hydration for 8 and 12 hours respectively. It was found that among all the priming showed significance difference with the control and the highest germination percentage (%), germination index, energy of emergence, seedling length (cm), seedling fresh weight (mg), seedling dry weight (mg) and vigour indices were observed in $CaCl_2$ priming for 12 hours. This study also showed that Seed priming with $CaCl_2$ were found to increase the seedling characters. The seeds treated with $CaCl_2$ (1%) hydration 12 hours followed by $CaCl_2$ (1%) hydration 8 hours and PEG (20%) hydration 8 hours recorded numerically higher values but for energy of emergence and for germination index second best was Neem leaf extract 5% hydration 12 hrs and for dry and fresh weight PEG (20%) hydration 8 hours was higher value compared to control. The study helps to improve the seedling character, growth of seeds with the help of seed priming treatments which are cost effective, economic, non-toxic and eco-friendly sources.

Keywords: Wheat, Priming Methods, PEG, NaCl, $CaCl_2$, Tulsi leaf extract, Neem leaf extract

Introduction
Wheat is a crop widely cultivated for its seed, a cereal grain which is a worldwide staple food. There are many species of wheat which together make up the genus *Triticum*; the most widely grown is common wheat (*T. aestivum*). Poor germination and low seed vigour are the major problems faced in the recent days. When eaten as the whole grain, wheat is a source of multiple nutrients and dietary fiber, and is associated with lower risk of several diseases, including coronary heart disease, stroke, cancer and type 2 diabetes. In a small part of the general population, gluten – the major part of wheat protein can trigger coeliac disease, non-coeliac gluten sensitivity, gluten ataxia and dermatitis herpetiformis. Seed priming has been found out to be very successful method to overcome these problems. Priming allows some of the metabolic processes necessary for germination to occur without germination take place. In priming, seeds are soaked in different solutions with high osmotic potential. This prevents the seeds from absorbing in enough water for radicle protrusion, thus suspending the seeds in the lag phase. Seed priming has been commonly used to reduce the time between seed sowing and seedling emergence and to synchronize emergence. In seed priming, the osmotic pressure and the period for which the seeds are maintained in contact with the membrane are sufficient to allow pre-germinative metabolic processes to take place within the seeds up to a level limited to that immediately preceding radicle emergence. Priming is an invigouration technique that enhances the physiological and biochemical events in seeds during suspension of germination by low osmotic potential and negligible matric potential of the imbibing medium. Such treatments include water soaking (Bradford, 1986) [4], priming in which hydration is controlled in an osmoticum such as polyethylene glycol (PEG) or a salt solution. (Heydecker and Coolbear, 1977) [10]: Pretreatment of seeds by various methods in order to improve seed germination rate, percentage germination, and improve uniformity of seedling emergence by controlling the water available in the seed. The pretreatment initiates the early stages of germination, but does not permit radicle protrusion, and then the seeds are dried until needed. Seed priming has been successfully demonstrated to improve germination and emergence in

seeds of many crops specially vegetables and small seeded grasses. Seed priming has presented promising, and even surprising result, for many seed including the legume seeds (Bradford, 1986) [4]. Short time hydration treatment, e.g. hydro priming, humidification (incubating seed at high relative humidity) have been widely used to increase seed vigour and extend longevity in many plant species (Powell *et al.,* 2000) [21]. Potassium chloride is the most widely used source of potassium for agricultural crops, and Cl is considered an essential micronutrient for optimal growth (Fixen, 1993) [8]. Potassium chloride has been introduced as the osmoticum to enhance germination, emergence and growth of Poaceae plants (Misra & Dwivedi, 1980) [18]. Halo-priming of seeds in pre-sowing treatments in an osmotic solution allows seeds to absorb water, but restricts radicle occurrence through testa until the primed seeds are sown for germination under salt stress conditions. Primed seeds usually show improved germination parameters (Hardegree & Van Vactor, 2000) [11]. The present study was done under objective of to evaluate the effects of pre-sowing invigouration seed treatments on Germination behavior and Seedling Vigour in Wheat (*Triticum aestivum* L.) Seeds and access the suitable priming method and its duration for wheat. It is reported that pigeon pea seeds treatment with $CaCl_2$ or KNO_3 generally exhibited improvement in proteins, free amino acid and soluble sugars during germinating under salt stress.

Materials and Methods

The present study entitled "Effect of pre-sowing invigouration seed treatments on germination behavior and seedling vigour in Wheat (*Triticum aestivum L.*) Seeds." under Post graduate laboratory of Seed Science and Technology was conducted in the Department of genetics and plant breeding, Naini Agricultural Institute, Sam Higginbottom University of Agriculture Technology and Sciences, Allahabad during 2016-2017. Allahabad is located in South Eastern part of Uttar Pradesh, India. The site of experiment is located at 25.57^0N latitude, 81.56^0N longitude and 98 meters above mean sea level. The lab experiment was analyzed by using C.R.D. (Complete Randomized Design) with four replications and 13 treatments under laboratory condition with control, one hydropriming, two organic priming and three halopriming in two duration of hydration 8 and 12 hours. Seeds treated with Distilled water for 8 hours, Polyethylene glycol (PEG) @ 20% for 8 hours, Sodium chloride (NaCl) @ 1% for 8 hours, Calcium chloride ($CaCl_2$) @ 1% for 8 hours, Tulsi leaf extract @ 5% for 8 hours, Neem leaf extract @ 5% for 8 hours, Distilled water for 12 hours, Polyethylene glycol (PEG) @ 20% for 12 hours, Sodium chloride (NaCl) @ 1% for 12 hours, Calcium chloride ($CaCl_2$) @ 1% for 12 hours, Tulsi leaf extract @ 5% for 12 hours, Neem leaf extract @ 5% for 12 hours. Afterwards, primed seeds were allowed to dry back to their original moisture content under shade to access the parameters. Seed quality parameters include speed of germination, Energy of emergence, Germination percentage, root length, shoot length, seedling length, seedling dry weight, seedling vigour index length and seedling vigour index mass.

The standard for chemical preparation, amount of distilled water use 100 ml. The chemical was added to 100 ml of distilled water with constant stirring. The volume of solution was finally constituted to one litre, then it became 100ppm stock solution of each chemical. The flasks containing chemicals was covered with muslin cloth to avoid any contamination. For the preparation of NaCl (5%) solution 50 (g) NaCl was a taken in a measuring flask and made up to 1000 ml distilled water, while for (1%) $CaCl_2$ solution 10 (g.). $CaCl_2$ was taken in a measuring flask and made up to 1000 ml with distilled water. For preparation of PEG (20%) solution 200(g) PEG was a taken in a measuring flask and made up to 1000 ml distilled water.

After preparation of solution of PEG, NaCl, $CaCl_2$, Tulsi leaf extracts Neem leaf extracts hydration, wheat seeds was soaked in required solution for 8 & 12 hour at 25^0C temperature. Simultaneously seeds was soaked in distilled water. After 8 & 12 hr of soaking the solution was drained out from the beaker and pre-soaked seeds were air dried to original weight and then placed for germination in laboratory under controlled condition. Then seeds were placed in four replication in completely randomized design (CRD) in between paper method for germination in laboratory under controlled condition.

The observation on the characters *viz.,* Germination percent (ISTA 2004) [12], Speed of germination, Energy of emergence(%)(Ruan *et al*., 2002) [23], Root length (cm), Shoot length (cm), Seedling length (cm), seedling fresh weight (g), seedling dry weight (g), Seedling vigour index I^{st}, Vigor index II^{nd} (Baki and Anderson 1973) [1] were recorded. The experimental data recorded were subjected to statistical analysis for calculating analysis of variance, range, mean, critical difference and coefficient of variation (Fisher 1936) [8].

Result and Discussion

According to the results, all studied traits were affected by the treatments and there was completely significant difference between control (unprimed seeds) and primed seeds (Table-1).

All seedling characters *viz.* Germination percent, Speed of germination, Energy of emergence(%), Root length (cm), Shoot length (cm), Seedling length (cm), seedling fresh weight (g), seedling dry weight (g), Seedling vigour index I^{st}, Vigor index II^{nd} were affected by $CaCl_2$ 1% hydration for 12 hours concentration and significantly recorded maximum.

Significantly higher germination percent (98.5%) reported in treatment T_{11} $CaCl_2$ 1% hydration 12 hours followed by T_5 (97.50%) primed with $CaCl_2$ 1% hydration 12 hours. Minimum germination percent recorded by T_1(92.00%) with unprimed control (Table 2).

Table 1: Analysis of variance for seedling characters in wheat.

Characters	Mean Square	
	Treatments (Df=6)	Error (Df=21)
Germination Percentage	15.20**	3.41
Germination Index	24.7**	1.79
Energy Of Emergence	17.88**	3.85
Root Length	4.79**	0.88
Shoot Length	6.52**	0.49
Seedling Length	17.15**	1.44
Seedling Fresh Weight	138399.43**	21033.65
Seedling Dry Weight	911.81**	138.66
Seed Vigour Index I^{st}	205487.51**	18377.44
Seed Vigour Index Ii^{nd}	9707098.01**	1338398.50

* And ** significant at 5% and 1% level of significance, respectively.

Higher germination index (26.88) reported in treatment T_{11} $CaCl_2$ 1% hydration 12 hours followed by T_{12} (34.12) primed with Tulsi leaf extract. Minimum germination index recorded by T_1 (26.73) with unprimed control (Table 2). Maximum

energy of emergence (77.50%) recorded by T_{11} primed with $CaCl_2$ followed by T_9 (76.75%) primed with PEG(20%) hydration 12 hrous. Minimum recorded in T_1 control (70.75%) (Table 2). (Misra & Dwivedi, 1980; Al-Mudaris and Jutzi, 1997; Elouaer *et al*., 2012) [18, 7]. Was also found similar result for germination%, speed of germination and energy of emergence. It is evaluated the sorghum seeds soaked in $CaCl_2$ solution increased the activity of total amylase and proteases in germinating seeds under salt stress. Maximum root length (13.18cm) recorded by T_{11} treatment $CaCl_2$ (1%) hydration 12 hours followed by T_5(11.19cm) primed with $CaCl_2$ (1%) hydration8 hours. Minimum root length recorded by T_1 (9.12cm) with control. Maximum shoot length (12.64cm) recorded by T_{11} treatment primed with $CaCl_2$ (1%) hydration 12 hours and it followed by T_5 (12.6cm) primed with $CaCl_2$ (1%) hydration 8 hours. The shortest shoot length founded in T_3 PEG (20%) hydration 8 hours (8.58cm). Maximum seedling length (24.35cm) recorded by T_{11} primed with $CaCl_2$ (1%) hydration 12 hours followed by T_{10} (23.85cm) primed NaCl (1%) hydration 12 hours. Shortest seedling length recorded in T1 unprimed control (17.7 cm) (Table 2). (Demir and Oztokat 2003) [6] Also found that root and shoot lengths increased in seeds due to salt priming as compared to non-primed seeds. Halo-priming of seeds in pre-sowing treatments in an osmotic solution allows seeds to absorb water, but restricts radicle occurrence through testa until the primed seeds are sown for germination under salt stress conditions. Primed seeds usually show improved germination parameters (Hardegree & Van Vactor, 2000) [11].

Table 2: Mean Comparison of Germination and Vigour Traits in Wheat

Tretments	Germination (%)	Energy of emergence	Germination index	Root length (cm)	Shoot length (cm)	Seedling Length (cm)	Seedling fresh weight (mg)	Seedling dry weight (mg)	Vigour index I	Vigour Index II
T_1	92	70.75	26.73	9.12	10.26	17.7	875	70.87	1628.4	6510.87
T_2	93.25	71	28.42	9.78	9.49	19.01	1200	97.2	1772.68	9084.07
T_3	93.75	73	28.68	10.29	8.58	20.55	1450	117.45	1926.56	11001.83
T_4	96.25	75	32.19	10.56	9.23	21.64	1350	109.3	2082.85	10532.55
T_5	97.5	76.25	33.5	11.19	12.06	23.25	1400	113.4	2266.87	11060.12
T_6	95.5	74.5	32.21	10.24	11.03	19.73	1175	95.17	1884.21	9106.2
T_7	92.75	74.25	30.19	11.28	9.59	20.87	1218	98.6	1935.69	9216.02
T_8	95.25	75.5	31.9	8.4	11.87	17.75	1025	83.02	2169.80	7916.29
T_9	95	76.75	32.14	10.97	8.90	22.84	1231	99.71	1690.68	9462.69
T_{10}	94.75	74.75	32.29	10.97	9.55	23.85	1425	115.42	2259.78	10936.04
T_{11}	98.5	77.5	35.72	13.18	12.64	24.35	1587	128.54	2398.47	12661.19
T_{12}	96.5	76.5	34.12	10.95	10.67	21.85	1225	99.22	2108.52	9582.11
T_{13}	97.25	73.25	32.67	11.09	10.19	21.28	1287	104.24	2069.48	10156.72
MEAN	95.25	74.55	31.6	10.72	10.32	21.21	1266.34	102.53	2007.50	9788.51
SE(d)	1.3	1.38	0.66	0.66	0.49	0.84	102.55	8.32	95.85	818.04
S.Em	2.6	0.98	0.67	0.42	0.35	0.60	72.51	5.88	67.78	578.44
C.D at 5%	2.64	2.8	1.91	1.8	1.00	1.71	207.43	16.84	193.89	1654.65

T1 -UNTREATED (CONTROL) T2 -DISTIL WATER (8 HOURS) T3 -PEG6000 (20%) (8 HOURS) T4 -NaCl (1%) (8 HOURS) T5 -CaCl2 (1%) (8 HOURS) T6 -TULSI EXTRACT(5%) (8 HOURS) T7 -NEEM EXTRACT(5%) (8 HOURS) T8 -DISTIL WATER (12 HOURS) T9 -PEG6000 (20%) (12 HOURS) T10- NaCl (1%) (12 HOURS) T11-CaCl2 (1%) (12 HOURS) T12-TULSI EXTRACT(5%) (12 HOURS) T13-NEEM EXTRACT(5%) (12HOURS)

Maximum seedling fresh weight (1587mg) reported by T_{11} treatment primed with $CaCl_2$ (1%) hydration 12 hours followed by T_3 (1450mg) primed with PEG (20%) hydration 8 hours. Lowest value of seedling fresh weight found in T_1 unprimed control (875mg). Maximum seedling dry weight (128.54mg) recorded by T_{11} primed with $CaCl_2$ (1%) hydration 12 hours followed by T_3 (117.45mg) primed with PEG (20%) hydration 8 hours. Lowest value of seedling dry weight found in T_1 unprimed control (70.87mg) (Table 2). (Jafar *et al.* 2012; Ashraf and Rauf 2001; Toklu *et al*., 2015) [13, 2, 24] also reported to the results regarding root and shoot fresh weights are in agreement with those of who reported that fresh and dry weights of seedlings from haloprimed seeds were significantly higher, as compared to other unprimed seeds. Maximum seedling vigour index I^{st} (2398.47) recorded by T_{11} primed with $CaCl_2$ (1%) hydration 12 hours followed by T_5 (2266.87) primed with $CaCl_2$ (1%) hydration 8 hours Minimum seedling vigour index I^{st} recorded by T_1 (1628.4) in control (Unprimed) (Table 2) The osmo-priming, halopriming has positive effect on the seed germination and their consequences. They help to release in enzymes and accelerate seed metabolism and physiological activities (Jie *et al*., 2002) [14]. Maximum seedling vigour index II^{nd} (12661.19) recorded by T_{11} primed with $CaCl_2$ (1%) hydration 12 hours and it was followed by T_5 (11060.12) primed with $CaCl_2$ (1%) hydration 12 hours. Minimum seedling vigour index II^{nd} recorded by unprimed T_1 (6510.87) in control (Table 2). It has been reported that primed seeds showed better germination pattern and higher vigour level than non- primed (Ruan *et al*., 2002) [23]

Conclusion

It is concluded from the present study that the different concentration of priming treatment showed significant effect on seed germination and seed vigour parameters. Priming with $CaCl_2$ (1%) hydration 12 hours increased germination (%) and seed vigour in wheat. Second best priming is $CaCl_2$ (1%) hydration for 8 hours and PEG (20%) hydration 12 hours. In all priming method halopriming with 12 hours hydration showed best result in comparison to organic priming.

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