

**ON-FARM SEED PRIMING TO ALLEVIATE THE EFFECT OF SALINE
STRESS DURING GERMINATION OF PEARL MILLET
(*PENNISETUM GLAUCUM* L.)**

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Abstract

Crop tolerance to salinity is of high importance particularly in the arid and semi-arid regions and pre-sowing seed treatments have been used to overcome the effect of salt stress problem. In the present experiment, on-farm primed (pearl millet seed soaked in distilled water at 25°C in the dark for 10 hr) and dry seeds were then germinated in petri dishes under six levels of salinity (control (0), 40, 80, 120, 160 and 200 mM NaCl) under laboratory conditions. The investigation was carried out in a completely randomized design (CRD) with factorial arrangement and each treatment was replicated four times. The results showed that there were significant differences ($P < 0.05$) for mean germination time (MGT) and time taken for 50% germination (T50) in all salinity levels. Salinity stress decreased the seed germination traits however, on-farm priming improved it. The value of germination index (GI), seed vigor index (SVI) and germination stress index (GSI) decreased by increasing salt stress and value was recorded in case of dry seed sown as compared to primed. Results also indicated that maximum reduction in germination percentage (GP) was 12.61 and 8.57%, however; increase in T50 was 129 and 69% in dry and primed seed respectively, sown at highest level of salinity (120 mM) compared to control treatment. These findings suggest that on-farm priming is an effective method to increase seed germination grown under moderate salt stress.

Introduction

Soil salinization is a global issue of dryland agriculture and improper management of irrigation methods and farming practices it is also spreading in irrigated (Abraha and Yohannes 2013). Salinity present in the soil has a physiological affect by decreasing the water uptake and by lowering osmotic potential of plant (Paparella *et al.* 2015, Ibrahim 2016). It delays the germination of seed as well as final germination percentage and subsequently affects the plant growth (Rahman *et al.* 2000). Many strategies have been being applied to induce stress tolerance in crops; among them, at germination stage, seed priming is easy, low-cost, an effective and practically applicable technique to enhance rapid and uniform germination especially under an adverse environment (Paparella *et al.* 2015). It is pre-sowing treatment for water uptake (imbibition) and it precedes the first stages of germination, however prevents the rupturing of seed coat and ultimately protrusion radicle (Farooq *et al.* 2009, Silva *et al.* 2015).

Soaking of seed in water, surface drying and same day sowing is termed as on-farm priming. The priming treatment consists of safe limit and it is considered that seed or seedling should not be damaged during premature germination. It is considered that on-farm priming is a low cost technology, and provide immediate benefit and also access to other benefits (Harris *et al.* 2001). It

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is especially useful and practically adoptable for the poor farmers holding the marginal lands for cultivation (Harris *et al.* 1999, 2005). Furthermore, on-farm priming enhances the germination as well as emergence and provides other benefits like establishment of crop stand (Murungu *et al.* 2004). On-farm seed priming is very simple technique as compared to techniques used in crops of temperate region like halo-priming and matri-priming. The main purpose of on-farm seed priming is just rapidly soak the seed and sow it in hydrated condition to give rise the germination as well as emergence (Pill 1995).

Pearl millet (*Pennisetum glaucum* L.) is a cross-pollinated cereal crop and grown for fodder as well as for grain purposes, especially in Asia and eastern Africa (Martel *et al.* 1997, Kapila *et al.* 2008). The cultivated area of pearl millet in Pakistan is of 486 thousands hectares and annual production of 299 thousands tones (Anonymous 2015-16). The seed has high nutritional value (Khairwal *et al.* 1999), containing large number of vitamins and minerals including 7-11% proteins, 60-70% carbohydrates, 2-7% crude fibre and 1.5-5% fat (Singh *et al.* 2012). The green fodder of pearl millet is used as a feed for livestock all over the country (Arif *et al.* 2010). It is an annual crop and considered tolerant to adverse environmental conditions (Rachie and Majumdar 1980).

The investigation was conducted in the controlled conditions of laboratory and the objective was to improve the seed germination potential of pearl millet crop under salinity stress by using on-farm seed priming technology.

Materials and Methods

This investigation was done to check the effect of on-farm priming on the germination of pearl millet seeds sown under different salinity levels. The planned study was carried out in the laboratory of Department of Environmental Sciences of COMSATS University Islamabad (CUI), Vehari Campus in the Province Punjab of Pakistan. The seeds of pearl millet (YBS-83) were collected from the Maize and Millet Research Institute Sahiwal as this cultivar is registered and approved for cultivation and considered very suitable for grain as well as forage production. Thirty seeds were placed in each Petri dishes and filter paper (Whatman No. 1) was placed on both sides of seed. The soaked seed of pearl millet in distilled water at 25°C in the dark for 10 hrs and then surface dried considered as on-farm priming seed (Harris *et al.* 2002). The solution of NaCl of different concentration *i.e.* (control (0), 40, 80, 120, 160 and 200 mM NaCl) were prepared by dissolving calculated amount of NaCl and solution of 10 ml of relevant concentration was added to each Petri dish (Asgharipour and Rafiei 2011). The Petri dishes were washed first with the tap water, and then distilled water was used for rinsing followed by hot air sterilization at 170°C for 4 hrs (Muhammad and Hussain 2010). The Petri dishes were placed in the laboratory at room temperature ($23 \pm 5^\circ\text{C}$) and relative humidity of $50 \pm 5\%$ (ISTA 2011). The experiment setup had factorial arrangement in a completely randomized design and each treatment was replicated four times. Association of Official Seed Analysis (AOSA 1990) method was followed for counting daily seed germination until the germination become constant.

Soon after the first day of sowing of seed, the germinated seeds were counted on daily basis at 11.00 am and seed containing 10mm radical length were considered as germinated (Goertz and Coons 1989). The counting of seed germination was continued till the germination constant. The final counting of germinated seed was considered to calculate the germination percentage (GP). The GP was calculated by the formula as given by ISTA (2009). Time Taken for 50% germination was calculated by using the formula of Farooq *et al.* (2005).

Mean germination time was calculated according to the equation described by Ellis and Roberts (1981). In order to calculate germination index (GI), Maguire equation (Maguire 1962) was used. From the 2nd day to 7th once in 24 hours the germinated seeds were counted and GI or speed of germination (SG) was calculated. Seed Vigor Index (SVI) was calculated by following the formula of Abdul-Baki and Anderson (1970). Germination Stress Index (GSI) was determined by the formula given below and refers to salt tolerance (Bouslama and Schapaugh 1984).

$$\text{GSI} = \frac{\text{Speed of germination at stress condition (SGc)}}{\text{Speed of germination at control condition (SGc)}} \times 100$$

Data collected during the course of study was analyzed statistically by using Fisher's ANOVA technique and LSD test of probability at 5% was used to compare the mean values of treatments (Steel *et al.* 1997).

Results and Discussion

On-farm seed priming also known as seed soaking is an effective technology for a rapid and uniform germination which results in good establishment of crop stand. It is very simple soaking technique to start metabolic activities before start of germination. Although salinity stress decreased the germination percentage (GP), however, non-significant ($p > 0.05$) differences were observed among the salinity treatments up to 160mM both in dry and primed seed in term of GP. Maximum GP (88 and 86%, respectively) was recorded in the control (water) treatment, while minimum GP (80 and 75%) was observed at highest treatment of salinity (200 mM) for primed and dry sown seed, respectively (Table 1). On-farm priming technology play important role in improving the germination under salt stress (Abro *et al.* 2009). The seed water uptake is important factor for germination and increasing concentration of salinity in the growth medium caused decrease in the uptake of water (Gonzalez and Ramirez 1999, Mensah and Ihenyen 2009). Gulnaz *et al.* (1999) also observed that pre-sowing seed soaking mitigate the effect of salt on germination of wheat. Highly positive correlation coefficient ($r = 0.98^{**}$) was found between GP and GI (Table 3) and regression value also show strongly positive association i.e. $R^2 = 0.99$ (Fig. 1a).

Table 1. Effect of salinity on GP, MGT and T50 parameters under dry and on-farm primed seed of pearl millet.

NaCl levels (mM)	GP		MGT		T50	
	Dry	On-farm priming	Dry	On-farm priming	Dry	On-farm priming
Control (water)	85.83 a	87.50 a	2.71 de	2.55 e	0.94 d-f	0.83 f
40	85.00 a	85.83 a	2.90 de	2.70 de	1.06 d-f	0.91 ef
80	82.50 ab	85.00 a	3.05 c-e	2.93 de	1.17 b-f	1.17 b-f
120	80.00 ab	82.50 ab	3.56 a-c	3.19 cd	1.54 bc	1.18 b-f
160	77.50 ab	80.83 ab	3.85 ab	3.52 bc	1.63 b	1.37 b-e
200	75.00 b	80.00 ab	4.04 a	3.80 ab	2.15 a	1.40 b-d
LSD value	10.06		0.52		0.47	

Means followed by the same letter are not significantly different at $p < 0.05$ level (LSD test). GP = Germination percentage, MGT = Mean germination time, T50= Time taken for 50% germination, LSD = Least significant difference.

Mean germination time (MGT) of both dry and on-farm primed sown seed increases significantly with the increasing salinity levels. The MGT differs non-significantly ($p > 0.05$) between dry and primed sown seed in each salinity treatment (Table 1). Maximum MGT (4.04 and 3.80 days) was recorded at highest salinity levels (200mM) and minimum MGT (2.71, 2.55 days) was observed in control in dry and primed seed sown, respectively. As far as taken for 50% germination (T50) is concerned, it increased with increasing the salinity concentration (salinity levels). The difference of T50 between dry and primed seed was non-significant, except at highest level (200mM) of salinity where maximum T50 (2.15 days) was recorded in dry seed.

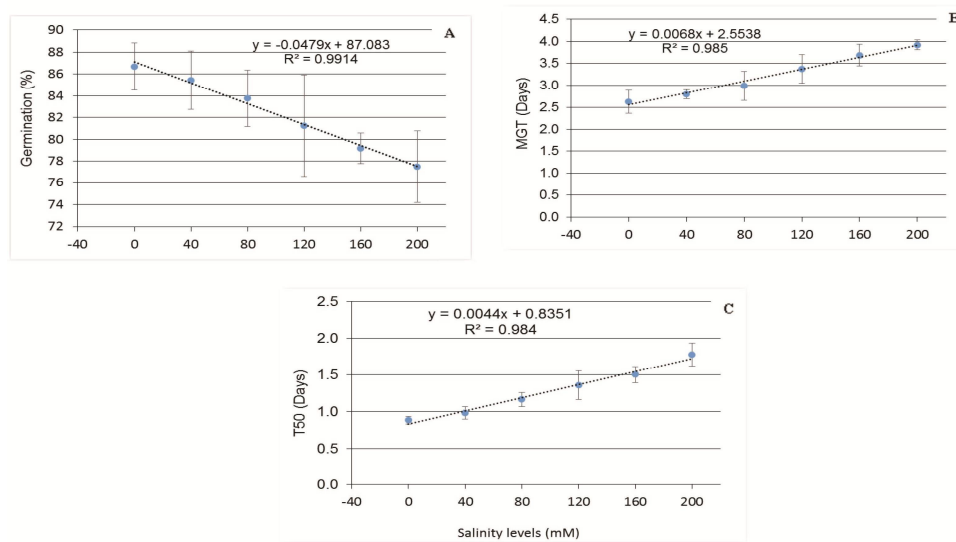


Fig. 1. Relationship of germination (A), MGT (B) and time T50 (C) with salinity levels of pearl millet seed.

Overall higher T50 value was observed in dry seed as compared to primed seed at all the salinity treatment (Table 1). The NaCl salinity affects the seed germination by decreasing water uptake of seed and ultimately delayed the MGT (Okcu *et al.* 2005). Langeroodi and Noora (2017) also found that seed hydration improved the seed vigor by decreasing the T50 as well as MGT.

The value for correlation coefficient between all possible combinations of parameters was calculated (Table 2). The results of correlation coefficient indicate that MGT has a highly significant positive association with T50 (0.93**) and negative with GI (-0.97**), GP (-0.98**) and SVI (-0.95**). Furthermore, highly negative association of T50 was observed with GI, GP and SVI. The regression analysis indicates strong relationship of MGT ($R^2 = 0.99$) and T50 ($R^2 = 0.94$) with increasing salinity levels (Fig. 1 b and c).

Germination index (GI) called as speed of germination (SG), increased when the treatment of NaCl salinity decreased. In case of primed seed no significant difference of GI was recorded with increasing the salinity stress, however, reduction in GI of dry seed was not significant up to 160mM but at highest salinity level (200mM) it differed significantly ($p < 0.05$) with respect to control (water) treatment. Germination index better indicated for the vigor of seed (Wang *et al.* 2004) and salinity stress at germination stage significantly affect the GI (Cokkizgin 2012). Besides other benefits, on-farm priming directly play role in producing more vigorous seedling and better

tolerance to stress environment (Harris *et al.* 1999). Germination index has a significant and positive correlation with GP (0.98**) and SVI (0.94**).

Table 2. Pearson correlation coefficient (r) for analyzed variables of pearl millet.

	GP	T50	MGT	GI	SVI	GSI
GP	1.00	-	-	-	-	-
T50	-0.97**	1.00	-	-	-	-
MGT	-0.98**	0.93**	1.00	-	-	-
GI	0.98**	-0.97**	-0.97**	1.00	-	-
SVI	0.94**	-0.90**	-0.95**	0.94**	1.00	-
GSI	-0.11 ^{NS}	0.08 ^{NS}	0.10 ^{NS}	-0.18 ^{NS}	-0.06 ^{NS}	1.00

*,** Significant at 0.05 and 0.01 probability levels, respectively.

Table 3. Effect of salinity on GI, SVI and GSI parameters under dry and on-farm primed seed of pearl millet.

NaCl levels (mM)	GI		SVI		GSI	
	Dry	On-farm priming	Dry	On-farm priming	Dry	On-farm priming
Control (water)	33.20 ab	41.43 a	102406 b-d	161548 a	-	-
40	31.33 a-c	41.15 a	106797 bc	139328 b	104.33 a	88.41 ab
80	30.65 a-c	38.09 a-c	90379 cd	115066 cd	88.41 b	73.15 b-d
120	29.11 a-c	37.71 a-c	86564 de	112803 cd	84.84 bc	71.80 c-e
160	27.59 bc	36.34 a-c	61402 f	88442 ef	60.69 d-f	55.492 e-g
200	26.20 c	35.72 a-c	44445 g	71928 fg	43.02 g	45.21 fg
LSD value	6.74		20246		16.11	

Means followed by the same letter are not significantly different at $p < 0.05$ level (LSD test). GI = Germination index, SVI = Seed vigor index, GSI = Germination stress index.

Seed vigor index (SVI) improved by on-farm priming and salinity treatments had a significant differences in terms of SVI ($p < 0.05$). With increasing the salinity levels the SVI decreased both in dry and primed sown seed. Regression analysis indicate significant relation ($R^2 = 0.97$) of increasing salinity treatments with SVI (Fig. 2b). These results of current study (GI and SVI) are in agreement with the finding of Ozdener and Kutbay (2008), and they reported that GP and rate of germination were reduced by increasing salinity levels and it may be due to osmotic stress caused by reduced water uptake or ion toxicity (Khan *et al.* 2008). On-farm priming is considered as key technology to increase the seed vigour and early growth of seedling (Harris *et al.* 2001).

Analyzed data showed considerable variation regarding GSI (Table 2), and the noted index was significantly reduced for by increasing salinity stress treatment. The highest value of GSI was found in control treatment and the lowest value is related to highest salinity treatment (200mM)

both in dry and primed sown seed (Table 2). Furthermore, GSI exhibited a strong association ($R^2 = 0.96$) with increasing the salinity levels (Fig. 2c).

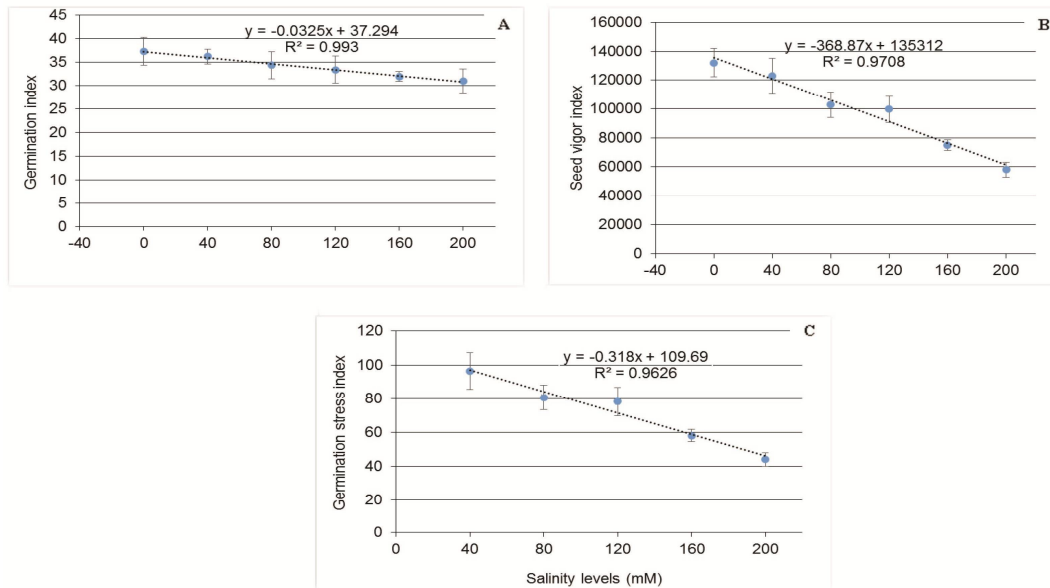


Fig. 2. Relationship of germination index (A), seed vigor index (B) and germination stress index (C) with salinity levels of pearl millet seed.

Germination stress index (GSI) is a criterion for measuring stress tolerance (Bousslama and Schapaugh 1984) and it is considered an appropriate value for estimation under stress condition (Sapra *et al.* 1991). The value of GSI is used in to observe the effect of treatment, as in the present experiment it was significantly reduced at higher salinity level (200mM) compared to lower salinity level both in primed and dry sown seed.

On-farm seed priming is safe, very effective and easily can be easily adopted by the farmers having poor resources and it has potential to benefit the poor farmers. It has been observed from results of present study that pearl millet is moderately tolerant to salinity at germination stage and emphasis the positive effect of on-farm priming on seed germination parameters. Further thorough investigations would warrant unveiling the tolerance of pearl millet at later on stages of plant when grown in moderately saline field.

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References

Abdul-Baki AA and Anderson JD 1970. Viability and leaching of sugars from germinating barley. *Crop Sci.* pp. 131-34.

- Abraha B and Yohannes G 2013. The role of seed priming in improving seedling growth of maize (*Zea mays* L.) under salt stress at field conditions. *Agric. Sci.* **4**(12): 666-672.
- Abro SA, Mahar AR and Mirbahar AA 2009. Improving yield performance of landrace wheat under salinity stress using on-farm seed priming. *Pak. J. Bot.* **41**(5): 2209-2216.
- Anonymous 2015-16. Pakistan Economic Survey. Finance Divisions, Economic Advisor's Wing, Islamabad. pp. 28-29.
- AOSA 1990. Rules for testing seeds, USA. *J. Seed Technol.* **12**: 1-112.
- Arif A, Muhammad I, Jamil K, Khan AR, Hassan S and Kiramat K 2010. Comparative studies of different pearl millet (*Pennisetum americanum* L.) varieties as affected by different yield components. *Elec. J. Environ. Agric. Food Chem.* **9**(9): 1524-1533.
- Asgharipour MR and Rafiei M 2011. Effect of salinity on germination and seedling growth of lentils. *Aust. J. Basic App. Sci.* **5**(11): 2002-2004.
- Bousslama M and Schapaugh WT 1984. Stress tolerance in soybean. Part I: Evaluation of three screening techniques for heat and drought tolerance. *Crop Sci.* **24**: 933-937.
- Cokkizgin A 2012. Salinity stress in common bean (*Phaseolus vulgaris* L.) seed germination. *Nat. Bot. Horti. Agrobi. Cluj-Napoca* **40**(1): 177-182.
- Ellis RA and Roberts EH. 1981. The quantification of ageing and survival in orthodox seeds. *Seed Sci. Technol.* **9**: 373-409.
- Farooq M, Basra SMA, Ahmad N and Hafeez K 2005. Thermal hardening: a new seed vigor enhancement tool in rice. *J. Integ. Plant Biol.* **47**: 187-193.
- Farooq M, Basra, SMA, Wahid A, Khaliq A and Kobayashi N 2009. Rice seed invigoration: A review. *In: Lichtfouse E (ed). Organic Farming, Pest Control and Remediation of Soil Pollutants: Sustainable Agricultural Reviews.* Springer Science, Amsterdam.
- Goertz SH and Coons JM 1989. Germination response of tepary and navy beans to sodium chloride and temperature. *Hort. Sci.* **24**(6): 923-925.
- Gonzalez LM and Ramirez R 1999. Water uptake by rice seeds at high salt concentrations, as a possible of cultivar tolerance. *Cultivos Tropicales* **20**(1): 35-37.
- Gulnaz A, Iqbal J and Azam F 1999. Seed treatment with growth regulators and crop productivity. II. Response of critical growth stages of wheat (*Triticum aestivum* L.) under salinity stress. *Cereal Res. Commun.* **27**(4): 419-426.
- Harris D, Breese WA and Kumar RJVDK 1999. On-farm seed priming in semi-arid agriculture: development and evaluation in maize, rice and chickpea in India using participatory methods. *Exp. Agric.* **35**(1): 15-29. <https://doi.org/10.1017/S0014479799001027>
- Harris D, Breese WA and Rao JVDK 2005. The improvement of crop yield in marginal environments using 'on-farm' seed priming: nodulation, nitrogen fixation and disease resistance. *Aust. J. Agric. Res.* **56**: 1211-1218.
- Harris D, Joshi A, Khan PA, Gothkar P and Sodhi PS 2001. On-farm seed priming: using participatory methods to revive and refine a key technology. *Agric. Sys.* **69**(1): 151-164.
- Harris D, Tripathi RS and Joshi A 2002. On-farm seed priming to improve crop establishment and yield in dry direct-seeded rice. *Direct seeding: Research Strategies and Opportunities*, International Research Institute, Manila, Philippines, pp. 231-240.
- Ibrahim EA 2016. Seed priming to alleviate salinity stress in germinating seeds. *J. Plant Physiol.* **192**: 38-46. <https://doi.org/10.1016/j.jplph.2015.12.011>
- ISTA 2009. Handbook of Seedling Evaluation, 3rd Ed. ISTA Germination Committee, Zurich, Switzerland. 520 pp.
- ISTA. 2011. Rules Proposals for the International Rules for Seed Testing 2011 Edition. International Seed Testing Association. Secretariat, Zürichstrasse 50, CH-8303 Bassersdorf, Switzerland. 53 p.
- Kapila RK, Yadav RS, Plaha P, Rai KN, Yadav OP, Hash CT and Howarth CJ 2008. Genetic diversity among pearl millet maintainers using microsatellite markers. *Plant Breed.* **127**: 33-37.

- Khairwal IS, Rai KN, Andrews DJ and Harinarayana G 1999. Pearl Millet Breeding. Oxford and IBH, New Delhi, India.
- Khan MA., Ungar IA and Showalter AM 2000. Effects of salinity on growth, water relations and ion accumulation of the subtropical perennial halophyte, *Atriplex griffithii* var. *stocksii*. *Ann. Bot.* **85**(2): 225-232.
- Langeroodi ARS and Noora R 2017. Seed priming improves the germination and field performance of soybean under drought stress. *J. Animal Plant Sci.* **27**(5): 1611-1621.
- Maguire JD 1962. Speed of germination aid selection and evaluation for seedling emergence and vigor. *Crop Sci.* **2**: 176-177.
- Martel E, Denay E, Siljak-Yakovlev S, Brown S and Sarr A 1997. Genome size variation and basic chromosome number in pearl millet and fourteen related *Pennisetum* species. *J. Hered.* **88**: 139-143.
- Mensah JK and Ihenyen J 2009. Effects of salinity on germination, seedling establishment and yield of three genotypes of mung bean (*Vigna mungo* L.) in Edo State, Nigeria. *Nig. Ann. Nat. Sci.* **8**(2): 17-24.
- Muhammad Z and Hussain F 2010. Effect of NaCl salinity on the germination and seedling growth of some medicinal plants. *Pak. J. Bot.* **42**(2): 889-897.
- Murungu FS, Chiduzo C, Nyamugafata P, Clark LJ, Whalley WR and Finch-Savage WE 2004. Effects of 'on-farm seed priming' on consecutive daily sowing occasions on the emergence and growth of maize in semi-arid area of Zimbabwe. *Field Crops Res.* **89**(1): 49-57.
- Okcu G, Kaya MD and Atak M 2005. Effects of salt and drought stresses on germination and seedling growth of pea (*Pisum sativum* L.). *Turk. J. Agric. Fores.* **29**(4): 237-242.
- Ozdener Y and Kutbay HG 2008. Effect of salinity and temperature on the germination of *Spergularia marina* seeds and ameliorating effect of ascorbic and salicylic acids. *J. Environ. Biol.* **29**(6): 959-964.
- Paparella S, Araújo SS, Rossi G, Wijayasinghe M, Carbonera D and Balestrazzi A 2015. Seed priming: state of the art and new perspectives. *Plant Cell Rep.* **34**: 1281-1293.
- Pill WE 1995. Low water potential and presowing germination treatments to improve seed quality. *In*: Basra, AS (ed.), *Seed Quality: Basic Mechanisms and Agricultural Implications*. Haworth Press, New York, pp. 319-359.
- Rachie KO and Majumdar JV 1980. Pearl Millet. Pennsylvania State University Press, University Park, Pennsylvania.
- Rahman MS, Matsumuro T, Miyake H and Takeoka Y 2000. Salinity-induced ultrastructural alternations in leaf cells of rice (*Oryza sativa* L.). *Plant Prod. Sci.* **3**(4): 422-429. <https://doi.org/10.1626/pps.3.422>
- Sapra VT, Savage E, Anaele AO and Beyl CA 1991. Varietal differences of wheat and triticale to water stress. *J. Agron. Crop Sci.* **167**1: 23-28.
- Silva CB, Marcos-Filho J, Jourdan P and Bennett MA 2015. Performance of bell pepper seeds in response to drum priming with addition of 24-epibrassinolide. *Hort. Sci.* **50**: 873-878.
- Singh KP, Mishra A and Mishra HN 2012. Fuzzy analysis of sensory attributes of bread prepared from millet-based composite flours. *LWT-Food Sci. Technol.* **48**(2): 276-282.
- Steel RGD, Torrie JH and Dickey DA 1997. Principles and Procedure of Statistics. McGrawHill book Co., USA. pp. 178-18.
- Wang YR, Yu L, Nan ZB and Liu YL 2004. Vigor tests used to rank seed lot quality and predict field emergence in four forage species. *Crop Sci.* **44**(2): 535-541.

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