



## Effects of pre-sowing seed treatments on seed germination and salinity tolerance of Rice (*Oryza sativa* L.) seedlings

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Article published on December 30, 2016

**Key words:** Rice (*Oryza sativa*), Seed pretreatment, Salinity tolerance, Seed germination

### Abstract

The experiment was conducted to investigate the effect of different pre-sowing seed treatments on seed germination and salinity tolerance of rice seedlings during 2015-2016 at Faculty of Agriculture, University of Ruhuna, Sri Lanka. Rice variety At 362 was used for these experiments. Various organic and inorganic treatments such as coconut water, fermented milk water, fermented rice water, Ascorbic acid, KCl, KNO<sub>3</sub>, ZnSO<sub>4</sub>, CaCl<sub>2</sub> and water (control) were used as treatments. Complete Randomized Design (CRD) used as experiment design with three replicates. Cumulative germinated seed count was taken to interpret germination percentage. Under saline condition plant height, number of leaves, root volume and shoot dry weight of seedlings were taken as growth parameters. Results revealed that highest seed germination was observed in KNO<sub>3</sub> primed seeds (95%) and lowest was recorded in Ascorbic acid treated seeds (82%). Other chemicals such as CaCl<sub>2</sub>, ZnSO<sub>4</sub> and KCl also showed positive response for seed germination compared to control treatment. The results indicate from plant growth under saline condition revealed 2% CaCl<sub>2</sub> enhance the salinity tolerance of initial growth of rice seedlings. The highest plant height, number of leaves, root volume and shoot dry weights were recorded in 2% CaCl<sub>2</sub>. However considering with control treatment other chemical solutions have given positive effect on plant growth and development under saline conditions.

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## Introduction

Seed priming is the induction of a particular physiological state in plants by the treatment of natural and synthetic compounds to the seeds before germination which is a simple and low cost hydration technique use for better crop stand and higher yields in a range of crops including rice (Farooq *et al.* 2006a, 2009; Kaymak *et al.* 2009). Hydropriming is the recommended seed priming method before germinating rice seeds in Sri Lanka. However seed priming by using different chemical solutions (Osmopriming) has made some considerable effect on plant growth. Seed priming has been successfully demonstrated to improve germination, seed emergence, uniform and vigorous crop stand in seeds of many field crops, such as vegetables and small seeded grasses, wheat, sugar beet, maize, soybean and sunflower (Parera & Cantliffe, 1994) under less than optimum field conditions.

Moreover, rice seeds primed with KCl and CaCl<sub>2</sub> reported early emergence and seedling growth, better crop stand, increased kernel, yield, harvest index, and improved quality (Farooq *et al.*, 2006a) Salinity is one of the most serious factors limiting the productivity of agricultural crops, with adverse effects on germination, plant vigor and crop yield.

Seed priming technique widely use as a beneficial mode for controlling of plant abiotic stress under low cost conditions. Several studies have been done by researches relevant to this area. Considering to drought stress; using of KNO<sub>3</sub> and CaCl<sub>2</sub> solutions have given higher growth rate of rice seedlings (Galahitigama *et al.*, 2016). According to Sirvitepe *et al.*, 1999 seed priming with different salts, especially NaCl have shown higher germination rate under saline conditions. Seed priming with NaCl also showed higher growth and yield in tomato plants when salt treatments were applied with seed sowing. (Cano *et al.*, 1991) similarly Mohommadi, 2009 found that among the seed priming treatments, primed with Potassium nitrate showed higher germination rate in Soya bean. Considering these views the main objective this study was to investigate the effect of

different organic and inorganic osmopriming treatments on seed germination and salinity tolerance of paddy seedlings. Through this it was expected to find proper solution for salinity control of paddy seedlings at initial stage.

## Materials and methods

*Experiment (1): Pre sowing seed treatments on seed germination*

*Experiment site*

A lab experiment was conducted Department of Crop Science, Faculty of Agriculture under protected house conditions during 2015-2016.

*Experiment design*

The trial was laid out in Complete Randomized Design (CRD) with three replicates. Rice variety At 362 (Samba variety) was used for this experiment.

*Treatments*

Seeds were soaked overnight separately in water (control) and different chemicals and organic solutions (coconut water, fermented milk water, fermented rice water, Ascorbic acid 1%&2%, KCl 1&2%, KNO<sub>3</sub> 1&2%, ZnSO<sub>4</sub> 1&2%, CaCl<sub>2</sub> 1&2% used as treatments. The ratio of seed weight to solution volume was 1:5 (gmL<sup>-1</sup>) (Farooq *et al.* 2006a). Before seeds kept in pet radishes, seeds were toughly washed with water. Then seeds were put in a pet radish (100seeds/petradish) for germination. Germinated seeds were calculated within first 15 days. Data were statistically analyzed using analysis of variance (ANOVA) statistical method and mean separation was done using Dunnett's test.

*Experiment 2: Pre sowing seed treatments on salinity tolerance of rice seedlings*

Pot experiment was conducted under laboratory condition. Seeds were primed by using above mentioned solvents. 7 days after germination, seeds were transplanted in small pots (diameter: 3 1/4 inch and height: 2.8 inch) filled with paddy soil. Pots were placed in small trays to maintain water level. Two weeks after planting salinity was induced by using 50mM and 100mM NaCl solutions.

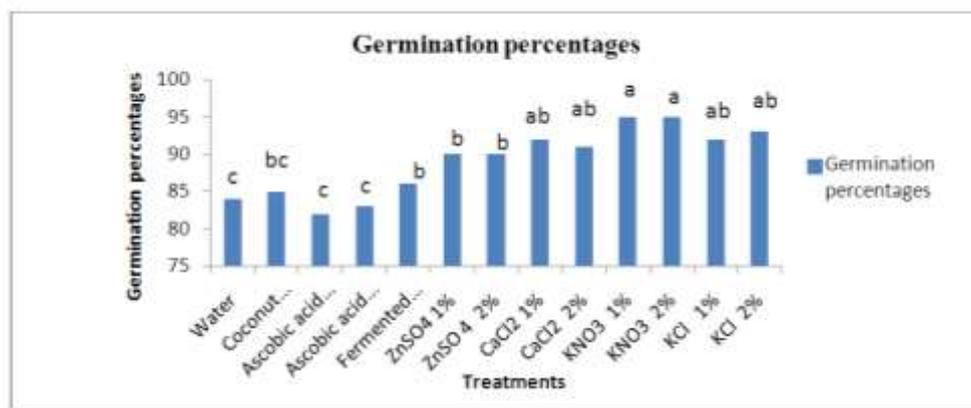
### Measurements

Data was collected two weeks after induction of salinity. Plant height, number of leaves, root volume and shoot dry weights were measured as growth parameters. The plant height was recorded from base to the tip of longest leaf. Oven dried plants under 65°C for 72 hours were used for shoot dry weight measurements. Data were statistically analyzed using analysis of variance (ANOVA) statistical method and mean separation was done using Dunnett's test.

### Results and discussion

#### Experiment 1: Effect of seed osmopriming on seed germination

According to the results revealed from germination test there is a significant effect from seed osmopriming on seed germination. The highest seed germination was recorded in  $\text{KNO}_3$  primed seeds (95%) and lowest was recorded in Ascorbic acid treated seeds (82%). In here other chemicals such as  $\text{CaCl}_2$ ,  $\text{ZnSO}_4$  and  $\text{KCl}$  also showed positive response for seed germination. When considering to organic solutions fermented rice water and coconut water treated seeds have given higher germination percentage compared to hydro priming. (Fig. 1).



**Fig. 1.** Effect of seed osmopriming on seed germination.

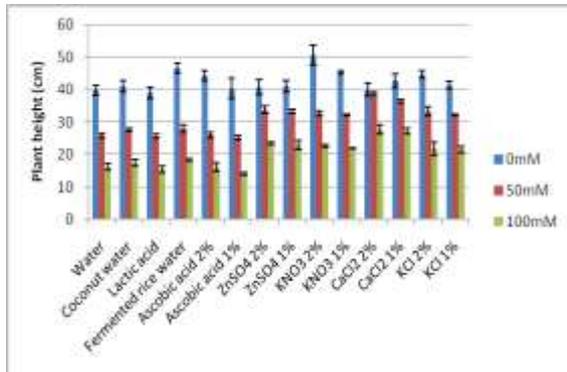
Generally, seed germination entails three distinct phases. (Imbibition, lag phase and radicle growth and emergence) When the imbibition process starts, it allows some physiological and biochemical processes to active. The past researches were made some evidences where priming boosts the rate of imbibition process. According to the Farahani *et al.*, 2011, the rate of hydration increased dramatically after seed primed by chemical reagents which could contribute to facilitate emergence phase of rice. Furthermore priming process prolongs the lag phase where the most of biochemical reactions are occur. Through this it ensure the vigor of young seedlings. Priming changes the interior condition of the embryo also. In here it has reported that  $\text{KNO}_3$  priming increased the embryo length in tetraploid watermelon seeds (Nerson *et al.*, 1985). Through this it facilitates the germination process of seeds. According to the Sung & Chang, 1993 seed priming make some changes in

biochemical content of the seed and improves membrane integrity and enhances physiological activities at seed germination. The improvement in emergence of primed seed may be due to the fact that priming induces a range of biochemical changes in the seed that are required to initiate the germination process i.e., breaking of dormancy, hydrolysis or metabolism of inhibitors, imbibition and enzymes activation (Ajouri *et al.*, 2004). This may be the reason for higher germination percentage recorded in primed seeds compared to hydro primed seeds.

#### Experiment 2: Effect of osmopriming treatments on salinity tolerance of rice (*Oryza sativa*) seedlings

According to the results, seed osmo-priming treatments were shown significant effect on salinity tolerance in young plants. The highest plant height, number of leaves and shoot dry weight were recorded in  $\text{CaCl}_2$  2%.

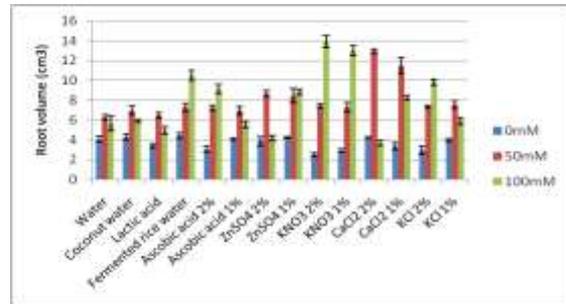
The lowest values were given in lactic acid compared to control treatment. Furthermore potassium salts treated seeds were also showed positive effect on salinity stress. When salinity level increases the values of all the parameters were reduced (Fig. 2).



**Fig. 2.** Plant heights changes under different salinity levels.

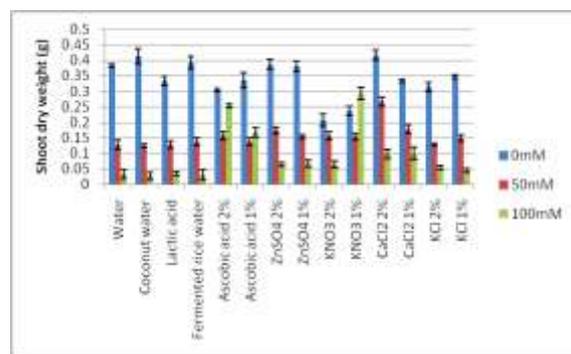
Salinity stress mainly affects to early growth of plant seedlings. Sometimes salinity reduces the seed germination also. According to the Dood and Donovan, 1999 excessive ions might be toxic for mobilization of reserves; radicle emergence and early growth of seedlings. Different studies have been done to identify the salinity effects on growth performances of field crops. As a result of these studies, it revealed that considerable changes occur in plant physiology have create negative impact on plant growth. In here salinity stress create a growth retardation in plant due to reduction of water up take or ionic imbalance owing to toxic effects of sodium (Na<sup>+</sup>) and chloride (Cl<sup>-</sup>) ions. Simultaneously salinity upsets the plant hormone level and reduces the utilization of seed reserves. (Ahmad and Bano, 1992). Leaf area reduced consistently and significantly as salinity increase. Salinity stress also affect to the chlorophyll content in plant. In some plants chlorophyll content increased due to the deposition of NaCl in the chloroplast. (Ex: pearl miller, mustard, sugar beet and cabbage). However it has been documented that the chlorophyll content decreased in plants such as tomato, radish, rice and Pea (Jamil *et al.*, 2012). According to Flowers and Yeo, 1981 rice is sensitive especially at young seedling stage, where varying degree of mortality occurs at 50 mM NaCl and about 50% of 14 days old

seedlings may die in most salt sensitive varieties within ten days of salinity stress. Therefore salinity strongly affects seed germination and early seedling growth, photosynthesis and protein synthesis (Fig. 3).



**Fig. 3.** Root volume changes under different salinity levels.

A similar pattern was observed in root volume and shoot dry weight also. In here the salinity level increasing, the length of root highly reduced but number of roots enhanced. However total shoot weight was abridged significantly when increasing the salinity. This may be due to slowing down the water absorption by the plant. Demir and Arif (2003) also obtained similar results. They have found that the root growth was more badly influenced by salinity than shooting growth (Fig. 4).



**Fig. 4.** Shoot dry weights changes under different salinity levels.

Seed priming can use as a method for reduce the negative impact on salinity for plants. Seed priming with CaCl<sub>2</sub>, KCl and KNO<sub>3</sub> reduced the Na<sup>+</sup> uptake of plants and/or increased the uptake of K<sup>+</sup>/Ca<sup>2+</sup> compared to control under salinity. Using these chemicals alleviate the NaCl stress on early seedling growth and improved salt tolerance is associated with

increased absorption of essential nutrients and restricted absorption of toxic elements (Chipa and Lal, 1995; Wahid and Shabbir, 2005). According to the Shannon and Francois, 1977 seed priming by using CaCl<sub>2</sub> has given better results might be due to the influence of Ca<sup>2+</sup> on membranes and enhanced antioxidant proteins like Cu/Zn SOD and other stress induced dehydrins proteins. Similar type experiment has been done for sunflower seeds which primed using CaCl<sub>2</sub> given substantial effect on plant growth. (Kathiresan *et al.*, 1984).

### Conclusion

Results illustrated that seed priming by using chemicals has given better results in enhancing the seed germination and salinity tolerance compared to hydro priming. Among them priming by using KNO<sub>3</sub> is preeminent for increasing of seed germination and CaCl<sub>2</sub> solution make better enhancement of seedling performance under saline condition.

### References

- Ahmad J, Bano M.** 1992. Effect of sodium chloride on physiology of cotyledons and mobilization of reserved food in *Cicer arietinum*. Pakistan Journal of Botany **24**, p.40-48.
- Ajourri AA, Haben, Becker M.** 2004. Seed priming enhances germination and seedling growth of barley under conditions of P and Zn deficiency. Journal of Plant Nutrition and Soil Science **167**, p.630-636.
- Cano EA, Bolarin MC, Perez-Alfocea F, Caro M.** 1991. Effect of NaCl priming on increased salt tolerance in tomato. Journal of Horticultural Science **66**, P. 621-628.
- Chhipa BR, Lal P.** 1995. Na/K ratios as the basis of salt tolerance in wheat. Australian Journal of Agriculture Research **46**, p. 533-539.
- Demir M, Arif I.** 2003. Effects of different soil salinity levels on germination and seedling growth of safflower (*Carthamus tinctorius*). Turkish Journal of Agriculture and Forestry **27**, p. 221-227.
- Dood GL, Donovan LA.** 1999. Water potential and ionic effects on germination and seedling growth of two cold desert shrubs. American Journal of Botany **86**, p.1146-53.
- Farahani HA, Moaveni P, Maroufi K.** 2011. Effect of hydro priming on germination percentage in sunflower (*Helianthus annuus* L.) cultivars, Advances in Environmental Biology **5(8)**, p. 2253-2257.
- Farooq M, Basra SMA, Hafeez K.** 2006a. Seed invigoration by osmohardening in coarse and fine rice (*Oryza sativa* L.). Seed Science Technology **34**, p.181-187.
- Flowers TJ, Yeo AR.** 1981. Variability in the resistance of sodium chloride salinity within rice (*Oryza sativa* L.) varieties. New Phytologist **88**, p. 363-373.
- Galahitigama GAH, Wathugala DL.** 2016. Pre-sowing seed treatments enhanced the Drought tolerance of Rice (*Oryza sativa* L.) Proceedings of the International symposium on Agriculture and Environment, 13<sup>th</sup> January 2016, Faculty of Agriculture, University of Ruhuna, Sri Lanka p. 130-135.
- Jamil M, Bashir S, Anwar S, Bibi S, Bangash A, Ullah, Rha ES.** 2012. Effect of salinity on physiological and biochemical characteristics of different varieties of rice. Pakistan Journal of Botany **44**, p.7-13.
- Kathiresan K, Kalyani V, Ganarethinam JL.** 1984. Effect of seed treatments on field emergence and early seedling growth of melons early growth and some physiological processes of sunflower (*Helianthus annuus* L.) Field Crop Research **9**, p. 215-217.
- Kaymak HC, Guvenc I, Yaralı F, Donmez MF.** 2009. The effects of bio-priming with PGPR on germination of radish (*Raphanus sativus* L.) seeds under saline conditions. Turkish Journal of Agriculture **33**, p.173-179.

**Mohammadi GR.** 2009. The effect of seed priming on plant traits of late-spring seeded soy bean (*Glycine max* L.). American-Eurasian Journal of Agriculture & Environmental Science **5(3)**, 322-326.

**Nerson H, Govers A.** 1986. Salt priming of muskmelon seeds for low temperature germination. Scientia Horticulture **28**, p. 85-91.

**Parera CA, Cantliffe DM.** 1994. Pre-sowing *Brassica* seed to improve emergence under different temperatures and soil moisture conditions. Crop Science **27**, p. 1050-1053.

**Shannon MC, Francois LE.** 1977. Influence of seed pre-treatment on salt tolerance of cotton during germination. Agronomy Journal **69**, p. 619-622.

**Sivritepe HO, Eris A, Sivritepe N.** 1999. The effect of NaCl priming on salt tolerance in melon seedlings. Acta Horticulturae **49**, p.277-284.

**Sung JM, Chang YH.** 1993. Biochemical activities associated with priming of sweet corn seeds to improve vigor, Seed Science and Technology **21**, p. 97-105.

**Wahid A, Shabbir A.** 2005. Induction of heat stress tolerance in barley seedlings by presowing seed treatment with glycine betaine. Plant Growth Regulators **46**, p.133-141.