International Journal of Chemical Studies

P-ISSN: 2349–8528
E-ISSN: 2321–4902
IJCS 2017; 5(6): 1140-1143

Received: 20-09-2017
Accepted: 23-10-2017

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Seed priming to alleviate the effect of salinity stress in rice

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Abstract
The study to evaluate the effect of seed priming with hydrogen peroxide (H_2O_2), jasmonic acid (JA) and sodium nitroprusside (SNP) on seed germination and vigour of three different rice varieties of salt tolerant (TNAU Rice TRY 3), salt sensitive (ADT (R) 49) and moderately tolerant (CO 43) varieties at different salt concentrations (100, 150 & 200 mM) revealed that seed priming with SNP @ 80 µM recorded maximum germination in all the varieties when compared with hydroprimed and unprimed control seeds irrespective of the salt concentrations. The aim of the study was to mitigate the ill-effects of salinity and improves the performance of the seedling, under salt stress condition, and it was evident that seed priming with SNP 80 @ µM was found to be effective for mitigating the effect of salt stress in rice even under higher salt concentrations.

Keywords: rice, seed priming, germination, SNP

Introduction
At global level rice is one of the most important cereals preferred for human consumption and it supplies up to 80 per cent of daily energy intake which serves as the staple food for more than one-third of the world's population (Khush, 1997) [8]. Even though, rice is cultivating under various soils and climate conditions but it is extremely sensitive to different abiotic stresses especially salinity, sodicity, drought, light, temperature *etc*., (Grover, 1999) [7]. Though rice is considered as a salt sensitive crop (Flowers *et al*., 1985) [6], it is one of the most widely grown crops in coastal areas frequently inundated with saline sea water during high tidal period (Akbar *et al*., 1972) [2]. Soluble salts levels in the soil has been adversely affected the growth of the most of the crops and it is considered as salt affected soil. Increased salinity problems in the soil due to salt water interventions and poor drainage leads to soil less fit for crop growth. For sustaining life on earth, to overcome these problems, reclamation or least minimizing the effect of salinity or sodicity is most important and necessary. Seed priming is one of the seed quality enhancement techniques, where the seeds are partially soaked and subsequently dried back for invigourative effect that expresses on field emergence and extended up to yield (Austin *et al*., 1969) [4]. Hegarty (1970) [11] also opined that priming would improves the velocity of germination and seedling emergence even under sub-optimal environmental conditions. Seed priming is a simple and low cost technique in which controlled hydration of seeds followed by redrying is done to break dormancy, improve germination and seedling establishment (Afzal *et al*., 2009) [3]. Farooq *et al*. (2006) [5] revealed that seed priming techniques promoted germination, yield and grain quality of rice. Compared the different priming *viz*., hydropriming, halopriming and osmopriming were most suitable for several vegetable crops (Nirmala and Umarani, 2008) [10]. For the ultimate goal of crop improvement, it is necessary to understand the mechanism of plant responses to salinity stress.
In plants, hydrogen peroxide (H_2O_2) is one of the major and the most stable reactive oxygen species (ROS) and regulates basic processes such as acclimation, defense and development (Slesak *et al*., 2007). Exogenous H_2O_2 treatments simultaneously enhanced multiresistance to heat, chilling, drought and salt stresses in maize seedlings (Gong *et al*., 2001). Jasmonic acid (JA) is a naturally occurring growth regulator found in higher plants. Several physiological roles have been described for this compound during plant development and in response to biotic and abiotic stress. Jasmonates antagonistically regulate the expression of salt stress inducible proteins, associated with salt stress in rice (Moons *et al*., 1997). Similarly, Sodium nitroprusside (SNP) is used as releasing compound of nitric oxide. At lower concentration of NO which induces the biochemical and physiological processes in plants (Neill *et al*., 2003).

Moreover, all the seed priming chemicals which is used for the experiments enhances the plant metabolism in the crop plants to withstand even under salinity stress.
Keeping in this view the role of these priming agents *viz*., H_2O_2, JA and SNP on improving vigour under salt stress conditions and beneficial effects in various crops, the present study was conducted to evaluate and compare the effects among the seed priming agents on growth and alleviation of the salinity stress in rice plants.

Materials and Methods

Experiments were carried out at Department of Seed Science and Technology, Tamil Nadu Agricultural University, Coimbatore during 2015. The seeds of three different rice varieties of salt tolerant (TNAU Rice TRY 3), salt sensitive (ADT (R) 49) and moderately salt tolerant (Co 43) varieties were taken for the study.
The seeds were primed with H_2O_2, JA and SNP at different concentrations of 0.25, 0.50 & 0.75%), (50, 75 & 100 µM) and (40, 80 & 100 µM) in the respective solution for the duration of 12 hours @ 1:1 ratio of seeds to solution. After that, seeds were removed from the solutions and shade dried at room temperature to bring back its original moisture content for assessing the seed quality parameters.
The germination test was conducted by following procedure outlined in using between paper method in order to assess salt stress effect on seed germination. Germination paper used for conducting germination test was pre-soaked in various concentration of NaCl solution *viz*., 0, 100, 150 and 200mM concentrations. Four replicates of 100 seeds each were germinated in a seed germinator maintained at 25 ± 2 ºC temperature and 95 ± 3% RH. After fourteenth day, the seedlings were evaluated and the normal seedlings were counted and expressed in percentage (ISTA, 2011) [12].
At the time of germination count in roll towel method, ten normal seedlings were selected at random from each replication and used for measuring the root and shoot length. The values were calculated and expressed in centimetre.
The seedlings used for growth measurement were placed in a paper cover and dried in shade for 24 h and then kept in an oven maintained at 85±2 ºC for 24 h. The dried seedlings were removed from the hot air oven and cooled in the desiccators over silica gel. Dry weight was recorded and the mean values were expressed in mg/10 seedlings.
The speed of germination was calculated and the results were expressed in number (Maguire, 1962). Vigour index was computed and the mean values were expressed in whole number (Abdul-Baki and Anderson, 1973) [1]. The data obtained from each of the experiments were subjected to an analysis of variance and treatment differences tested for significance (P= 0.05). Wherever necessary, the per cent values were transformed to arc-sine values before analysis. The critical differences (CD) were calculated at 5 and 1 per cent probability level (Gomez and Gomez, 1984) [14].

Result and Discussion

Germination is an important index for salt tolerance and significant differences were observed due to priming treatments and varieties under stress conditions. While observing the effect of different salinity stresses imposed using sodium chloride at 100, 150 and 200 mM levels on germination, a greater reduction in unprimed seeds was noticed at the salinity levels of 200 mM in all the varieties. Salinity is reported to decrease as well as delay the germination of most of the crops. Lower levels of salinity delay germination, whereas higher levels not only reduce the germination percentage but also can inhibit the seedling emergence (Ghoulam and Fares, 2001) [15].
The treatments of SNP as seed priming significantly increase the germination, length of root and shoot, speed of germination, drymatter production and vigour index as compared to hydropriming and unprimed seeds. Salinity stress has negative effects on growth parameters and application of SNP could alleviate the harmful effect of salinity in sunflower plants (Nejadalimoradi *et al*., 2014) [19].
Out of the various concentrations used SNP 80 µM showed best results and increased the germination percentage by (80, 82 & 81%), root length (9.2, 8.4 & 8.6 cm), shoot length (4.1, 3.0 & 3.5 cm), speed of germination (6.90, 6.73 & 6.84), drymatter production (0.163, 0.120 & 0.140 g10seedlings^{-1}) and vigour index (1164, 912 & 992) in the salt tolerant variety (TNAU Rice TRY 3), salt sensitive variety (ADT (R) 49) and moderately salt tolerant variety (Co 43) respectively and were found to be superior over control at 200 mM stress level as illustrated in (Fig. 1& 2). The increase in seedling vigour index and seedling dry weight was due to increased germination percentage, root length and shoot length of seedlings. Similar increase in germination and seedling growth parameters were observed due to seed priming in sesame (Suma, 2005) [16] and black gram (Surulirajan, 2007) [17].

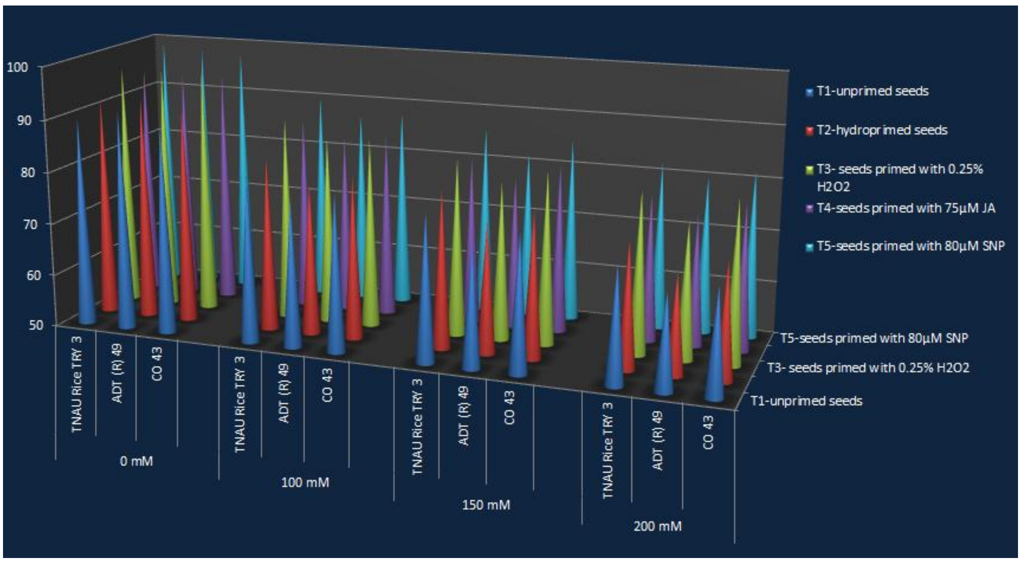

Fig 1: Effect of seed priming on growth of seedling on germination percentage

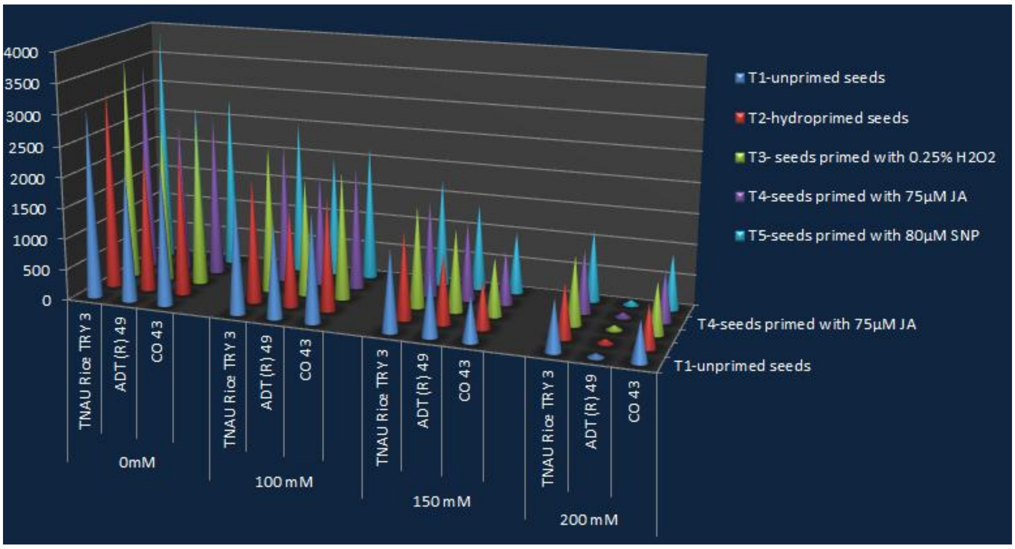

Fig 2: Effect of seed priming on growth of seedling on vigour index

In the present investigation, there was improved seed germination and seedling growth by SNP due to the production of Nitric oxide (NO). SNP also named as nitric oxide donor and NO is an atmospheric gaseous molecule, soluble in water and lipids. At lower concentrations NO has been involved in the regulation of physiological processes of crop plants.

The germination percentage was significantly decreased with increased salt concentration in all the three varieties. Seed priming with 80 µM SNP enhanced the seed germination than the unprimed control (13, 17 & 15 per cent) and hydroprimed seeds (10, 15 & 12 per cent) of TNAU Rice TRY 3, ADT (R) 49 and CO 43 respectively. Even though, there was reduction with increased concentrations of salinity levels in all the physiological parameters, the results has been proved that, the stability was maintained in all the three varieties at 200 mM salt concentration by the seed priming with SNP followed by H_2O_2 and JA.

In addition to that, when the plants were exposed to high salinity conditions, they developed the capability to produce more Reactive Oxygen Species (ROS). The present study revealed that, H_2O_2 is one of the ROS which is having the ability to counteract the effect of salinity and improves the survival of the crop plants under salt stress condition. According to Bor *et al*., 2003 [13], to improve the stress tolerance in plants, controlling the free radicals is the better way. Similarly, by the usage of minimal concentration of priming agents, the growth parameters of rice crop even under salt stress condition was enhanced even in the salt sensitive variety (Hemalatha *et al*., 2017) [18]. Likewise, JA also a phytohormone, which increased the salt tolerance by enhancing the accumulation of nontoxic metabolities and also act as a potential growth regulator to improve the growth of the seedlings under salinity conditions.

Conclusion

Various studies on plant responses to stress condition have some contradictory or inconclusive results regarding the differential responses in salt tolerant and salt sensitive varieties. The present study revealed that seed priming with SNP produced desirable results, both promoting the germination as well as increased the seedling growth and seedling vigour. It is concluded that, the ill-effects of salinity stress could be mitigated to considerable extend by advocating seed priming with SNP under increased salinity conditions due to releasing of NO after the seed priming with SNP. The salt tolerant variety TNAU Rice TRY 3 performed better than other varieties under salt stress conditions. The seed priming treatment improved the germination performance of the salt sensitive variety ADT (R) 49 when compared with salt tolerant and moderately salt tolerant varieties due to priming treatments. Hence seed priming with SNP @ 80 µM could be recommended for mitigating the effect of salt stress in rice even under higher salt concentrations.

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