

Original Research Article

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Studies on Seed Priming with Hydrogen Peroxide for Mitigating Salt Stress in Rice

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ABSTRACT

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The study to evaluate the effect of seed priming with hydrogen peroxide (H₂O₂) (0.25, 0.50 and 0.75%) on seed germination and vigour of three different rice varieties of salt tolerant (TNAU Rice TRY 3), salt sensitive [ADT (R) 49] and moderately salt tolerant (Co 43) varieties at different salt concentrations (100, 150 and 200mM) revealed that seed priming with hydrogen peroxide @ 0.25% recorded maximum germination in all the varieties when compared with hydroprimed and unprimed seeds irrespective of the salt concentrations. This treatment improved the germination performance of the salt sensitive rice variety (ADT (R) 49) also under higher NaCl concentrations.

Introduction

Salt stress imposes a major environmental threat to agriculture. Due to varying climatic conditions, the plants had undergone various abiotic stresses. Among these, salt stress has a major constraint in the delta regions. Especially, NaCl is one of the most abundant and powerful salt which causes nutrient deficiency and also create toxicity to the plant metabolism (Azevedo *et al.*, 2006). In addition to that, when the plants were exposed to the high salinity conditions, there having a capability to produce more Reactive Oxygen Species such as superoxide (O₂•-), hydrogen peroxide (H₂O₂) and hydroxyl radicals (OH•) (Implay, 2003). ROS formation leads to

changes in intercellular redox homeostasis and it is now widely accepted that redox signals are key regulators of plant metabolism, morphology and development (Bowler and Fluhur, 2000; Foyer and Noctor, 2003). H₂O₂ may plays an important role in signal transduction for abiotic stress tolerance, although H₂O₂ is toxic at high concentrations (Uchida, 2003). To improve the stress tolerance in plants, controlling the free radicals is the better way (Bor *et al.*, 2003). There is an increased production of hydrogen peroxide leads to improve the survival of the crop plants.

Among various techniques, seed priming is the effective way to mitigate the effect of stress condition in different plants. Generally, seed priming maintains the uniformity and rate of emergence of the crop (Bakht *et al.*, 2010). The general purpose of priming is to initiate the germination process but not completed due to partial hydration of seeds. Specifically under stress conditions, it induces the germination changes and which is to maintain the germination rate and uniformity in the seedling emergence (Ashraf and Fool and, 2005). Seed priming with H₂O₂ having the capacity to enhance the multi-resistance to heat, drought, chilling and salt stress (Uchida *et al.*, 2002).

Keeping this view, a study was conducted to evaluate the effect of seed priming with H₂O₂ in the rice seeds of salt tolerant, salt sensitive and moderately salt tolerant varieties under salt stress conditions.

Materials and Methods

Experiments were carried out at Department of Seed Science and Technology, Tamil Nadu Agricultural University, Coimbatore. 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 H₂O₂ at different concentrations of 0.25, 0.50 and 0.75% 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°C temperature and 95 ± 3% RH. After fourteenth day, the seedlings were evaluated and the normal seedlings were counted and expressed in percentage (ISTA, 2011).

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 seedling 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°C 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.

Statistical analysis

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).

Results and Discussion

Seed germination is the most critical stage in seedling establishment which determines the successful crop production. Pre sowing seed quality enhancement causes early, enhanced germination and growth rate of seedlings. During seed priming, the amount of water

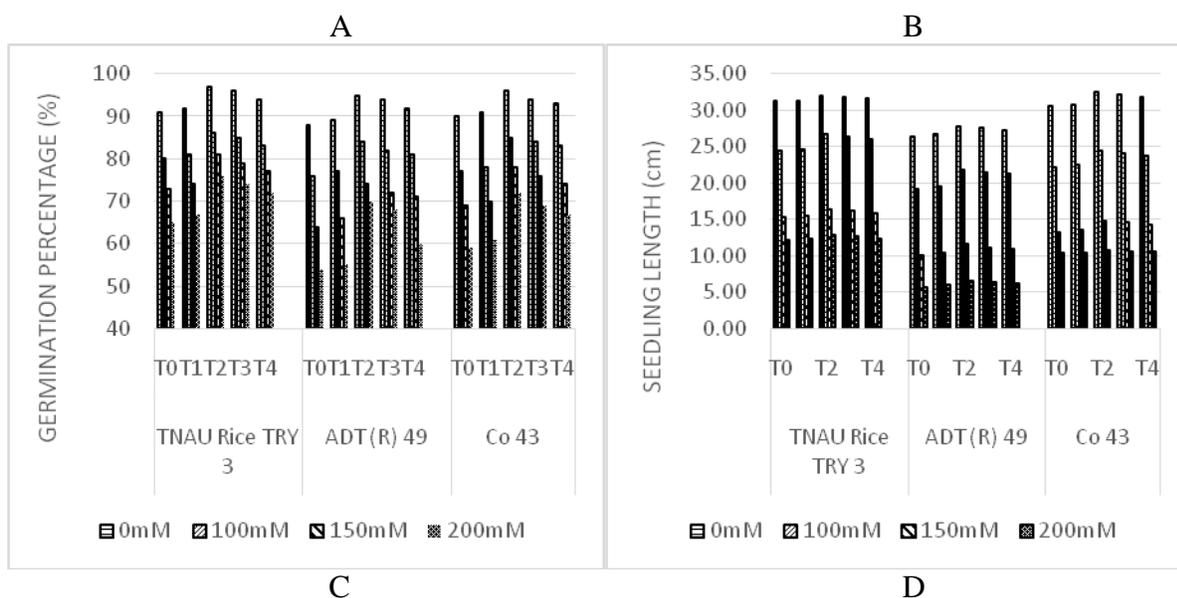
absorption is controlled so as to allow necessary metabolic activities for germination to take place but prevent radicle emergence by limiting the seed water content, different physiological activities occur within the seed at different moisture level. Salinity is reported to decrease as well as delay the germination of most of the crops. In this present study, seed priming with H₂O₂ (0.25, 0.50 and 0.75 %) had been increased the germination over control and hydropriming in all the three rice varieties with varied salt concentrations media including higher salt concentrations (200 mM) and results were illustrated in figure 1A. The germination percentage of seeds and salt concentrations were inversely proportional.

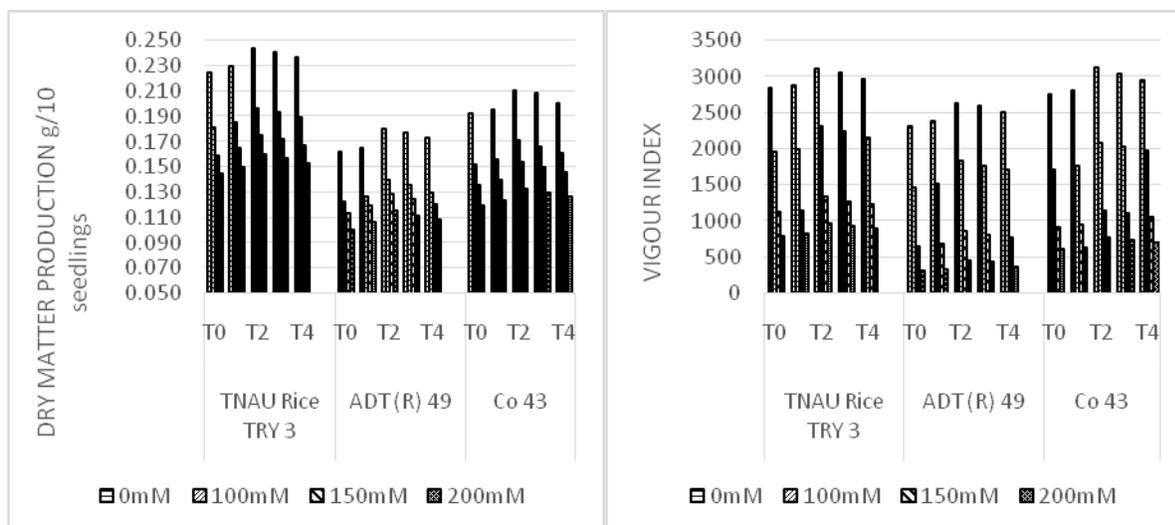
The germination percentage was significantly decreased with increased salt concentration in all the three varieties. In salt sensitive variety [ADT (R) 49], seed priming with 0.25% H₂O₂ enhanced the seed germination by 13% than the unprimed control and 11% than hydroprimed seeds even at higher salt concentration of 200 mM. Similarly, in

moderately salt tolerant variety (Co 43), 15% germination improvement was recorded than the unprimed control and 14% than hydroprimed seeds. The salt tolerant variety (TNAU Rice TRY 3), recorded 11% germination improvement than the unprimed control and 9% improvement than hydroprimed seeds at higher salt concentration of 200 mM.

Similar results were found in rice (Uchida *et al.*, 2002) and in wheat (Wahid *et al.*, 2007) both revealed that at lower levels of H₂O₂ increases the growth and physiological phenomena by preventing oxidative damage and enhanced the capacity to withstand in high salinity conditions. According to, Azevedo Neto *et al.*, (2005) in maize suggested that H₂O₂ induced an increase in salt tolerance during subsequent exposure to salt stress by the incorporation of hydroponics solution. In addition to that Sriniegn *et al.*, (2015) revealed that salinity can retard the germination under higher salt concentration in tomato seedling.

Fig.1 The effect of H₂O₂ on growth of rice seedlings of primed and unprimed seeds under salt stress conditions. A) Germination percentage (%), B) seedling length (cm), C) Dry matter production g/10 seedlings and D) Vigour index





T₀ -Control, T₁ -Seed priming with water,
T₃ -Seed priming with H₂O₂ @ 0.50%,

T₂ -Seed priming with H₂O₂ @ 0.25%,
T₄ -Seed priming with H₂O₂ @ 0.75%

Similarly there was a decreased seedling length with increased salinity condition. The seedling length was observed maximum at lower salinity level and constantly decreased with higher salinity level. According to, Hakim *et al.*, (2010) any crops growing under salinity condition there may be a reduction in seedling length. Similar results were observed in indica rice varieties there was a gradual decrease in the seedling length might be due to effect of NaCl salt (Anbumalarmathi and Mehta, 2013). In the present study also decreased seedling length was recorded due to higher salt concentration. However the seeds primed with H₂O₂ @ 0.25% concentration recorded an increased seedling length in all the three rice varieties of salt tolerant (TNAU Rice TRY 3), salt sensitive (ADT (R) 49) and moderately salt tolerant (Co 43) at different salt concentrations. But the stability had been maintained even in sensitive variety and results were illustrated in figure 1B.

The seedling vigour and dry matter production showed inverse relationship with salt concentrations. Generally the salt tolerant (TNAU Rice TRY 3) and moderately salt tolerant varieties recorded maximum dry matter production and seedling vigour when

compared with salt sensitive variety and results were illustrated in figure 1C and D. Similar results were observed in rice and wheat, the seedling length and dry weight were decreased with increased salt stress (Jamil and Rha, 2007).

From the present study it is clearly evident that H₂O₂ can be an important signal molecule for abiotic stress tolerance, and usage of minimal concentration enhances the physiological attributes of the rice crop even under salt stress conditions. It also conferredly increased the tolerance level of salt tolerant (TNAU Rice TRY 3), salt sensitive (ADT (R) 49) and moderately salt tolerant (Co 43) rice varieties.

Hence seed priming with H₂O₂ @ 0.25% could be recommended for mitigating the effect of salt stress in rice even under higher salt concentrations.

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