



PHYSIOLOGICAL AND BIOCHEMICAL BASIS OF PRE-SOWING SOAKING SEED TREATMENT-AN OVERVIEW

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Abstract

The proper germination of a seed in field is a crucial phenomenon because during this period seed has to face various kinds of stresses that may be biotic or abiotic. To get the better performance of seeds in respect to seed germination and healthy seedling establishment of many crops is subjected to various kinds of pre-treatments of seed with water and different chemicals, *i.e.*, Ca(NO₃), KNO₃, NH₄(SO₄)₂, NaCl, KCl, KH₂PO₄, CaCl₂ and Mg(NO₃)₂ in the form of pre-sowing / soaking and seed hardening (hydration/ dehydration) treatment. Study of literature educates that this type of pre-sowing soaking/ hardening treatments improves rate of germination by encouraging the metabolic process in the seed. Therefore, the treated seeds not only reduce the duration of time for seed germination but also improve the post germination phases of plant's life and finally the yield even under adverse condition because these types of treated seeds are found to be more stress resistant also.

Key words: Agriculture, Crop, Germination, Imbibition, Priming, Salt

Introduction

Rapid germination and emergence of healthy seedlings is an important step for successful establishment of plants. While water deficit during germination stage results in a reduction or complete inhibition of seed germination and seedling establishment (Kaya *et al.*, 2006). Reduction in germination percentage, amylase activity and solubilization of sugar content in endosperm of wheat seeds, germinating under low temperature was reported (Anaytullah and Bose, 2007). However, late sowing of wheat seed reduces rate of germination and seedling emergence in the field due to very low temperature in the month of December to January in North Indian condition (Anaytullah, 2007). Under drought condition, performance of seed germination and healthy seedling establishment inhibited due to the reduction of water potential, which results decline in water uptake by plants (Farooq *et al.*, 2009). Oxidative damage caused by the production of reactive oxygen species (ROS) is another major problem under drought stress (Gill and Tuteja, 2010). It is essential to overcome the adverse effects of drought stress for better crop yields (Ashraf and Rauf, 2001). For improving seed germination and

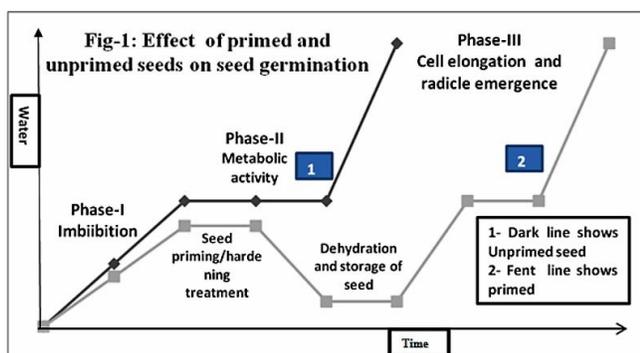
healthy seedling establishment, various possible approaches have been used *i.e.* seed hardening, priming, soaking and seed coating (Basra *et al.*, 2003). Seed priming treatment is use to accelerate seed germination, seedling growth and yield in most of the seeds under normal and stress conditions (Ashraf and Foolad, 2005; Iqbal and Ashraf, 2006; Mohammadi, 2009). It is recorded from the various literature that pre-sowing/soaking and seed hardening (hydration / dehydration) treatment to the seeds with different chemicals like Ca(NO₃), KNO₃, NH₄(SO₄)₂, NaCl, KCl, KH₂PO₄ and Mg(NO₃)₂ at various concentrations that promote seed germination and healthy seedling establishment via promoting the water uptake, amylase activity, protease activity, solubilization of nitrogen from endosperm to growing embryonic axis which improves the other biochemical parameters *i.e.* nitrate reductase activity, chlorophyll content in different crop (Anaytullah and Bose, 2007; Bose *et al.*, 1982; Mondal and Basu, 1984; Bose and Mishra, 1999; Bose and Mishra, 2001; Bose and Pandey, 2003 and Sharma and Bose, 2006).

Effect of Priming/Pre-Sowing Soaking/hardening treatment of seed with water

When a dry seed is soaked in water, the uptake of

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water start in tirphasic stages (Bewley, 1997) in which imbibition is the first stage where due to low water potential the rapid water uptake starts. Mc Donald, (2000) reported, DNA and mitochondria are repaired and proteins are synthesized using exist (mRNA). Study regarding the synthesis of RNA in primed seed confirmed by reverse transcription Polymerase chain reaction (PCR) analysis in cotton (Shinde, 2008). While in second stage, water uptake decreased in comparison to the first stage in seed. During this phase, embryo prepared for germination along with the synthesis of mitochondria and proteins by mRNA. The third stage of imbibition process is identified with a rapid increase of water and completed with the rupturing of seed coat and emergence of radical (fig.1).



(Source: Based and authors' work, 2018)

Fig.1: Effect of primed and unprimed seeds on germination

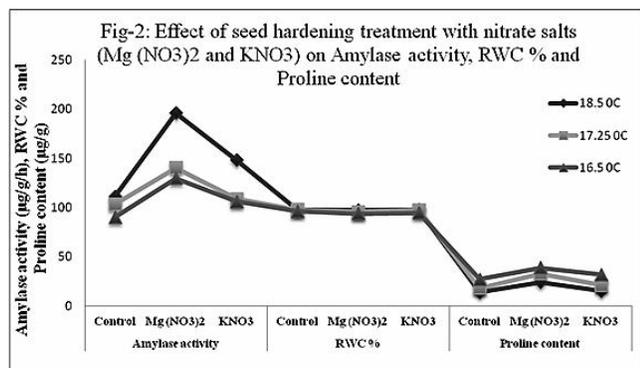
Stage first and second is the base phase of seed priming treatment where seed is just few steps away from radical protrusion. It has been found that pre-sowing soaking /hardening of seeds with distilled water, increased germination percentage and seedling vigor in wheat and rice crop (Al Ansari, 1997; Jaiswal *et al.*, 1997 and Suksoon *et al.*, 1998). Job *et al.* (2000) reported that enzymes involved in the mobilization of storage proteins are either synthesized or activated during seed priming treatment. Enzymes which are involved in the mobilization of carbohydrate (α -Amylase and α -Amylase) and lipid mobilization are also activated during seed priming (Sung and Chang, 1993). Issam *et al.* (2012) reported that seed priming can increase antioxidant activities as well as decrease lipid peroxidation during seed germination. Effects of priming are also associated with a wide range of metabolic event (Shehab *et al.*, 2010). Due to seed priming, antioxidant enzymes, such as superoxide dismutase, peroxidase, and catalase and osmoprotectants compound, such as proline, soluble sugar and soluble protein are producing (Farhad *et al.*, 2011). Activation of protective enzymes can be reduced ROS base oxidative damages (Posmyk *et al.*, 2009). While, osmoregulatory compound can help to increase water uptake by

increasing water potential inside the seed (Farooq *et al.*, 2009).

Effect of Pre-implanting/Pre-Sowing Soaking/Hardening treatment of Seed with various salts

A number of seed priming techniques have been developed, such as hydropriming, halopriming, osmopriming, thermopriming and biopriming (Ashraf and Foolad, 2005). The mechanism of seed priming treatments is not fully understood yet, but it has been reported that many physiological and biochemical changes take place during seed treatments (Ghiyasi *et al.*, 2008), which stimulate the seeds to start their germination steps before sowing. The optimum temperature for germination of maize seed in presence of water soaked condition was about 30-35°C. At higher temperature *i.e.* 40-50°C, a major portion of nitrogen was leached out to the medium from the endosperm. Soaking of seed at optimum temperature supported maximum growth of seedling and formation of ethanol soluble and insoluble nitrogen contents and higher nitrate reductase activity in primary leaves (Bose and Srivastava, 1982) and also reported that the effect of temperature on seed germination, protease activity and nitrogen solubilization in endosperm and its translocation towards the embryo. Gupta *et al.* (1983a) reported that nitrate reductase activity was induced by nitrate during the first 20 hours of wheat germination and nitrate reductase activity was induced by nitrate without a lag phase in seed germinated in water for 24, 32, and 48 hours. Enzyme α -amylase solubilize store foods via either inducing the *de novo* synthesis or increasing the activities of existing enzymes (Sung and Chang, 1993 and Lee and Kim, 2000), thereby producing germination metabolites in requisite amounts and transport solubilize food reserves to developing root and shoot parts where it helps in improving their growth and development. Priming is one of the physiological methods, which improves seed performance and faster and synchronized germination (Sivritepe and Dourado, 1995). Lee and Kim, (2000) reported that seed priming induces *de novo* biosynthesis of α -amylase, a key metabolic event in producing vigorous seedlings (Saha *et al.*, 1990). Similar results were observed by Bose and Tandon (1981); Bose *et al.*, (1992); Bose *et al.*, (1993); Bose and Mishra (1997) they reported that nitrate in general and $Mg(NO_3)_2$ in particular improved the germination percentage but the extent of improvement depends upon the cationic partners of the nitrate salts. They suggested that among nitrates of K, Ca and Mg, the later one is found better to show its carryover effect in maize. Nitrate application ($Mg(NO_3)_2$ and KNO_3) in form of seed-soaking /hardening treatment is found to improve not only seedling and root growth as

a fresh and dry weight but also increased the relative water content %, amylase activity ($\mu\text{g/g/h}$ fresh weight) and proline content ($\mu\text{g/g}$ dry weight) of wheat seeds under low temperature (Fig.2) which helps in osmotic adjustment and adaptive response in wheat seeds (Anaytullah and Bose, 2007).



(Source: Based and authors' work, 2018)

Fig. 2: Effect of seed hardening treatment with nitrate salts

Application of $\text{Mg}(\text{NO}_3)_2$ for osmopriming improves the process of germination via activating of various enzymes (Hexokinase, Phosphofruktokinase, Enolase) of respiratory cycle and nitrogen metabolism (Decarboxylase, Nitrate reductase). Moosavi *et al.* (2009) concluded that primed seeds significantly exhibited higher germination percentage, speed of germination, root length and seed vigor, total seed protein, peroxides (POD) and polyphenoloxidase (PPO) enzymes. Hilhorst, (1990) reported that nitrate (NO_3) acts as dormancy breaking agent during germination via stimulating the pentose phosphate pathway by producing NADPH_2 . There are reports that hydration of seed, but not exceeding the lag phase with priming permits early DNA replication (Bray *et al.*, 1989), increased RNA and protein synthesis (Ibrahim *et al.*, 1983), facilitate ATP availability (Mazor, 1984), faster embryo growth (Chang *et al.*, 2000), repair of deteriorated seed (Saha, 1990) and decrease leakage of metabolites in comparison to control (Styer and Cantliffe, 1983). Bose *et al.* (1982) observed that $\text{Ca}(\text{NO}_3)_2$ reduced the leakage of electrolytes from the seeds during germination than $\text{NH}_4(\text{SO}_4)_2$, NH_4NO_3 and control. Further, it is reported that nitrate act as a signaling molecules and favors the germination and this signaling interact either with abscisic acid or gibberellic acid pathway (Alboresi, 2005).

Conclusion

Primed seed (either hydro priming/nitrate priming) improved seed germination by increasing metabolic processes of seed over unprimed seed. After an

imbibition, the primed seed probably takes off from the third stage of germination. Seed priming also help in synchronization of all the metabolic activity in a seed lot. Hence, ensure synchronous seed germination as well as healthy seedling establishment. Including above mentioned beneficial effect, seed priming with nitrate may also act as a local long term signaling molecule which may have the capacity to induce genes related to the stress tolerance/resistance in different crops. Therefore, plant can perform better in adverse (stress) conditions.

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