HOW THE GERMINATION PERFORMANCE AND SEEDLING ESTABLISHMENT UNDER ABIOTIC STRESSES ARE IMPROVED BY SEED PRIMING ?

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ABSTRACT

One of the most promising treatments carried out with the intent of reducing the time needed between sowing and the seedling emergence, and to increase seed tolerance to adverse conditions during seed germination and seedling establishment is the osmoconditioning, also called priming. This review reveals major changes showed in abiotic-stressed plants as function of seed priming treatments, with a must approach about physiological and biochemical changes promoted by seeds priming. Many studies cited in this review have reported improvements in seed physiological quality and seedling vigor by seed priming under abiotic stresses conditions and its has been related with repair and build-up of nucleic acids, increased synthesis of proteins, osmotic adjustment mechanisms, repair of membranes and reduced lipid peroxidation, resulting from enhanced antioxidative activities. However, further studies are necessary to explain how the physiological and biochemical changes caused by seed priming improve the plant adaptations to environmental stressful conditions.

Keywords: osmoconditioning, seeds physiological quality, performance improvement, stressful conditions, physiological and biochemical mechanisms.

INTRODUCTION

Low quality lots of seeds frequently require more time for germination and seedling emergence. This makes the seedlings more sensitive to climate adversities, reducing the final percentage of emergence, and normally resulting in irregular seedling emergence. Therefore, many studies have been carried out with the intent of reducing the time needed between sowing and the seedling emergence, and to increase seed tolerance to adverse conditions during germination. One of the most promising treatments is the osmoconditioning, also called seed priming, which consists of controlled hydration of seeds to a certain level to facilitate the initial germination process without the occurrence of root protrusion [1].

The key basis of all pre sowing is to hydrate the seed under controlled conditions, so that they become physiologically active and thus they are able to initiate repair mechanisms and detoxify the system [2]. A number of research workers used different types of salts in pre-sowing soaking treatments to the seeds of various crops to get either better establishment of seedling or better plant stand and yield under varied environmental conditions [3].

There are many seed priming techniques, such as hydropriming (the soaking of seeds in water prior to sowing), hardening (alternate soaking of seeds in tap water and drying before sowing), osmoconditioning (seeds are generally pre-treated with polyethylene glycol 6000), osmohardening (similar to hardening but in the presence of NaCl), and hormonal priming (soaking of seeds in plant growth regulator solutions). These treatments have been used to accelerate emergence of roots and shoots, more vigorous plants, and better drought tolerance in many field crops [4], like wheat, sugar beet, maize, soybean and sunflower [5,6].

Since the discovery of seed priming in plants in the 1970s [7], this phenomenon has been demonstrated in different plant species, especially under abiotic stress conditions. Hence, priming appears to be a common feature of the plant’s immune system that offers protection against wide spectrum of environmental stresses [8]. Improvements in germination performance under abiotic stress conditions have been reported in literature as direct responses of seed priming, like the increased maximum tolerance to water stress by this treatment [9]. In addition, the carrot seeds priming in PEG-6000 solutions increased the emergence of seedlings in the field and performance in sub- and supra-optimal temperatures [10] and the osmoconditioning of sweet sorghum seeds mitigated the harmful effects caused by salt and thermal stress during germination and root growth [11].

The beneficial effects of priming have been verified on germination and seed emergence, as well as seedling establishment of many crops, particularly under abiotic stress, as reported by Reference [12]. These authors showed enhances in water uptake, percentage and velocity of sorghum seed germination. This behavior has been associated with repair and build-up of nucleic acids, increased synthesis of proteins and repair of membranes [1].

Seed priming also enhances the activities of anti-oxidation in treated seeds [13]. Therefore, the improved seed quality promoted by seed priming has been attributable principally to reduced lipid peroxidation, resulting from enhanced antioxidative activities [14]. In addition to benefic effects cited previously, recent progress made in molecular aspects of the seed priming has related this treatment with the accumulation of dormant mitogen-activated protein kinases, chromatin modifications and alterations of primary metabolism [15].

SEED AGEING AND SALINITY INDUCES MEMBRANE DAMAGES BY OXIDATIVE STRESS

Biochemical and physiological deterioration during seed aging has been studied mostly under accelerated aging conditions using high temperature and high seed water content. Some studies indicate that lipid peroxidation and the degradation of membrane phospholipids are major causes of seed aging under accelerated aging conditions [13]. However, plants contain numerous antioxidant compounds, both enzymatic and non-enzymatic, which act to prevent oxidative damage by scavenging free radicals before they attack membranes or other seed components [16]. Some protective mechanisms involving free radical and peroxide scavenging enzymes, such as catalase (CAT), guaiacol peroxidase (POX), ascorbate peroxidase (APX) and superoxide dismutase (SOD), have been evaluated within the mechanism of seed aging (13,17,18).

Another major constraint to seed germination and seedling establishment is soil salinity, which is a limiting factor to crop production in arid and semi arid regions of the world [19]. Soil salinity may affect the germination of seeds and seedling establishment either by creating an osmotic potential external to the seed and root preventing water uptake, or through the toxic effects of Na^+ and Cl^- ions on the germinating seed and plant growth [5]. In addition to ionic and osmotic components, salt stress, e.g. accelerated aging of seeds, also leads to oxidative stress through an increase in reactive oxygen species (ROS), such as superoxide ($O_2^{\bullet -}$), hydrogen peroxide (H_2O_2) and hydroxyl radicals ($OH^{\bullet}$) [20,21].

WATER UPTAKE AND GERMINATION IS ENHANCED IN ABIOTIC-STRESSED SEEDS OF DIFFERENT VIGOR LEVELS AS FUNCTION OF THE OSMOCONDITIONING

Reference [12] evaluated water uptake, seed germination and growth of sorghum seedlings from artificially aged and osmoconditioned seeds, under conditions of salt stress. The sorghum behavior of seed water uptake indicated that water uptake by the seed was directly related to water availability, matric potential of the substrate, osmotic potential of the solution that moistens the substrate, temperature and intrinsic characteristics of the seed. In this experiment, the priming promoted an increase in the water-uptake rate only in seeds with greater vigor – in other words, those not subjected to accelerated aging. Increase in the water-uptake rate might result from an accumulation of solutes from the onset of seed metabolism during priming, resulting in greater seed swelling when rehydrated [1,7].

Increases in the seedling growth correlated with higher water uptake by primed seed have resulted in higher seedling growth [4]. For instance, the hydropriming of chickpea seeds resulted in three to four times more growth with respect to root and shoot length in comparison with seedlings obtained from non-primed seeds in drought condition [22]. Similarly, the results obtained by Reference [12] in sorghum seeds with greater vigor subjected to salt stress with NaCl at 0.1 M confirmed that effects of seed priming were more notable when the environment was unfavorable.

Many authors have observed better improvement of seeds physiological quality when the seeds are under specific stress conditions, such as sub- or supra-optimal temperatures, water deficit or high salinity of the medium (10,11,23,24). On the other hand, in seeds with less vigor subjected to salt stress, those not subjected to priming presented a higher water-uptake rate than those that were osmoconditioned [12]. This greater uptake of water may be correlated to the fact that aged and deteriorated seeds were more sensitive to soaking damage, because the cell membranes were weakened, lost their integrity and became more susceptible to rapid water movement into the cell [25].

Reference [26] subjected maize seeds to hydropriming, hormonal priming with 100 mg L^{-1} gibberellic acid (GA_3) or indole-3-acetic acid (IAA) solutions and halopriming with 50 mM $CaCl_2$ or 50 mg L^{-1} ascorbate (ASA) for 24 h. Seed priming enhanced germination of spring maize under cool conditions and the performance was better in seeds subjected to $CaCl_2$ or ASA. According these authors this could be associated to the effect of higher reducing and total sugars as well as higher α-amylase activity. In a recent study, Reference [27] conducted laboratory experiments on freshly harvested pyrethrum (*Tanacetum cinerariifolium*) seeds to investigate the effects of light and seed hydropriming on germination, and shoot and root growth at 25 °C. The responses of hydroprimed and unprimed seeds to salt and drought stress were determined at osmotic potentials of 0 (distilled water), −0.3, −0.6, −0.9, −1.2 MPa in NaCl and PEG-6000. In this work the researchers showed that hydropriming shortened the delay of mean germination time at all osmotic potentials. It also improved the germination percentage in distilled water (from 52% to 59%) and resistance to salt stress with nearly double germination (from 16% to 29%) at the highest salt concentration.

OSMOCONDITIONING OF AGED SEEDS PROMOTES ACCUMULATION OF ORGANIC AND INORGANIC SOLUTES IN NaCl-STRESSED SEEDLINGS

Plants usually develop mechanisms that allow the maintenance of growth under salt-stress conditions; these mechanisms tend to limit salt absorption into the plant in order to avoid toxic ion concentrations in the cytosol [28]. In general, salt-tolerant plants perform osmotic adjustments through the accumulation of osmotically active solutes, such as ions in the vacuole and organic solutes, which allow water uptake, cell enlargement and plant growth under salinity [29]. The osmolytes that usually participate in osmotic adjustment vary between species and plant developmental stage and consist of sugars (glucose and fructose), alcohols (glycerol) and amino quaternary compounds (glycinebetaine and proline) [30].

Reference [31] verified that osmopriming of low physiological quality sorghum seeds (non-aged) provided an attenuation of salinity negative effects (100 mM NaCl) on seedling growth. According them Na^+ and Cl^- accumulation in shoot of NaCl-stressed sorghum seedlings from primed seeds indicates an osmotic adjustment induced by seed priming, which was efficient in reducing the osmotic stress caused by salinity. Furthermore, these authors confirmed that proline was the main contributing organic solute to osmoregulation in seedling shoot subjected to salinity, and its levels increased due to seed priming. Thus, the observed results in literature suggest that changes in inorganic and organic solute concentration in both shoot and roots could be induced by seed priming as a function of salt stress tolerance caused by this treatment, although the changes in these organs are poorly related to each other.

In another research, Reference [32] soaked wheat seeds in aerated solution of ascorbate, salicylic acid, kinetin and $CaCl_2$ for 12 h and showed that as an index of salinity tolerance, seed priming treatments also improved the leaf K^+ contents with simultaneous decrease in Na^+ concentration, osmopriming being the best treatment. Similarly, this research related maximum total phenolic contents, total soluble proteins (TSP), α-amylase and protease activities with the osmoprimed treatment (with $CaCl_2$) of seeds followed by ascorbate priming. According this study, economic analysis also indicated that osmopriming is more viable with maximum net return and benefit-to-cost ratio.

ACTIVATION OF ANTIOXIDANT ENZYMATIC SYSTEM BY SEED PRIMING

A study assessing the growth, lipid peroxidation and activity of antioxidative enzymes in sorghum seedlings grown under salt stress from artificially aged and primed seeds showed that priming of low physiological quality seeds (aged seeds) has provided an attenuation of salinity negative effects (100 mM NaCl) on seedling growth [33]. The results obtained by these authors suggest that osmoconditioning induced an increase in catalase (CAT) and guaiacol peroxidase (GPX) activities and these enzymes protected the seedlings against oxidative damage caused by seed accelerated aging and salinity in nutrient solutions.

In a recent research carried out by Reference [34], it was investigate the importance of catalase in oxidation protection during accelerated ageing and repair during subsequent priming treatment of sunflower (*Helianthus annuus* L.) seeds. The inhibition of catalase by the addition of aminotriazol during priming treatment reduced seed repair indicated that catalase plays a key role in protection and repair systems during ageing, which was associated with H_2O_2 accumulation as showed experimentally by the authors in biochemical quantification and $CaCl_3$ staining. They verified that catalase activity decreased at the level of gene expression, protein content and affinity, as well as interestingly, priming induced catalase synthesis by activating expression and translation of the enzyme.

It is important to remember that CAT has been considered an enzyme of great relevance in protecting plants against reactive oxygen species [20]. This is one of the main enzymes that remove H_2O_2, which may lead to the formation of hydroxyl radical ($^{\bullet}OH$), causing lipid peroxidation and cell membrane peroxidation, thus damaging plant growth [35].

The lowest lipid peroxidation levels in abiotic-stressed seedlings from osmoconditioned seeds can be related to seed priming capacity to act as a damage repairer of seed deterioration resulting from lipid peroxidation [1]. Reference [14] observed that this improvement might be associated with the conversion of lipids into carbohydrates and the antioxidant activity promoted by priming through increased activity of enzymes, such as isocitrate lyase (IL) and malate synthase (MS) in the first process, and SOD, CAT and APX, peroxidase (PO) and glutathione reductase (GR) in the second process, respectively.

PRE-SOWING SEED TREATMENT USING PLANT GROWTH REGULATORS AND HORMONES

Of various strategies being employed these days to mitigate the adverse effects of salinity stress, pre-sowing seed treatment with some plant growth regulators has gained much importance for being one of the most economical approaches of growing crops on salt affected soils [36]. However, another works have showed that various priming agents have variably under a variety of abiotic stresses and in different crop species. Pre-sowing seed treatment of triacontanol (TRIA), for example, did not alter the studied attributes by Reference [37], except that under non-saline conditions TRIA application increased the POD activity significantly in both cultivars.

Seed priming with hormones has been an efficient method for increasing seed vigor as well as seedling growth under stressful conditions. These responses have been associated to the activation of antioxidant systems in a range of crops [38]. Reference [39] working with *Vicia faba*, reported improvement in salinity tolerance due to the application of salicylic acid during seed priming was associated with enhanced CAT, ascorbate peroxidase (APX), POD and glutathione reductase (GR) activities. In a recent study, Reference [40] verified that seeds of

Agropyron elongatum primed with gibberellin (GA) and abscisic acid (ABA) exhibited induced CAT and SOD activities under drought conditions when compared to unprimed seeds). Another hand, results described by Reference [38] showed that hormonal priming with methyl jasmonate, salicylic acid or CEPA (chloroethylphosphonic acid), an ethylene (ET) releaser, does not induce the antioxidant activity of superoxide dismutase, catalase, ascorbate peroxidase or glutathione reductase in maize seedlings subjected to salt stress.

CONCLUSIONS

The beneficial effects of priming have been proven on germination and emergence of seed, as well as seedling establishment of many crops, particularly under abiotic stress. On the other hand, although there has been an immense amount of recent research involving abiotic stress enhances associated with seed priming, few studies have addressed combined stresses.

The improvements in seed physiological quality and seedling vigor by seed priming under abiotic stresses conditions has been related with repair and build-up of nucleic acids, increased synthesis of proteins, osmotic adjustment mechanisms, repair of membranes and reduced lipid peroxidation, resulting from enhanced antioxidative activities.

This review article has presented many results from recent researches, however further studies are necessary to explain how the physiological and biochemical changes caused by seed priming improve the plant adaptations to environmental stressful conditions.

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