Seed priming for alleviation of heavy metal toxicity in plants: An overview

Rajkumar Prajapati, Sunita Kataria & Meeta Jain*
School of Biochemistry, Devi Ahilya Vishwavidyalaya, Khandwa Road, Indore 452 001, Madhya Pradesh, India
*Email: mjjainmeeta@gmail.com

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ABSTRACT
Heavy metal (HM) toxicity is vital environmental constraint that limits crop productivity worldwide. Several physiological processes necessary for plant survival have been found to be affected by HM toxicity. In recent farming, advanced mechanisms are being developed to overcome from the stresses to enhance the yield. The seed priming is an affordable method for plants to survive under abiotic and biotic stresses. Priming is useful for commercial seed lots by seed technologists to increase the vigor of the seeds in terms of germination potential and enhance the tolerance against various stresses. It also removes the pollution threats by minimizing the uses of chemical fertilizers. The seeds having deprived of quality in terms of seed germination and seedling characters ultimately affect the growth, photosynthetic performance and yield of the plants under HM stress. On the other hand seed primed with various seed priming methods such as hydropriming, hormonal priming, chemical priming, biopriming, magnetopriming and nanopriming perform well under HM toxicity. Seed priming methods have been considered as a unique approach to get rid of HM stress by enhancing the seed germination, seedling vigor, rate of photosynthesis, biomass accumulation and thus increase the crop productivity. The present review provides an overview of different seed-priming methods and their role in alleviation of adverse effects of HM stress in plants.

Introduction
Plants live in a dynamic environment where many adverse conditions in the form of abiotic stresses such as cold, drought, salinity, heavy metal (HM) toxicity and UV-B stress are generated which affect their growth and development (1). Among various stress factors, heavy metal toxicity is considered a potential environmental enemy in natural and agriculture ecosystems. The increase in industrialization has escalated heavy metal pollution resulting in environmental, agricultural and human health problems (2, 3). Though HMs are naturally present in the soil; anthropogenic and geologic activities further increase their concentration, thus they cause harmful effects on organisms (3, 4). Toxic metal contaminated soil and water poses a severe risks to public and food safety (3, 4). Among these heavy metals, nickel (Ni), manganese (Mn) and zinc (Zn) are considered as essential metals which are required in small amount and play a significant role in numerous physiological processes of plants (5, 6). Nonessential metals such as cadmium (Cd), mercury (Hg) and lead (Pb) do not have important biological role. Instead, these metals when accumulate in higher concentrations disturb the physiological, structural and biological functions (2, 6). The general detrimental effects of HMs are chlorosis, distorted nutrient assimilation, reduction in growth and photosynthesis; lower biomass accumulation and senescence, which eventually can cause the death of the plants (7–9). In the current scenario, it is quite a challenge to overcome/repair the damage brought by HMs stress in plants.

Heavy metals are now considered as the second most pollutants, including pesticides, CO₂ and SO₂, as they are most hazardous pollutant which probably may beat solid and atomic waste (8). Natural activities like volcano eruption and rocks erosion have contributes in rising the release of toxic elements to the atmosphere; though, increased human activities such as mining, refining and painting have enhanced...
their concentration in the biosphere (10, 11). Around 70% of the heavy metals and their compounds reach to our body and food is considered as the main route (10, 11). The interruption of nature’s geochemical cycle of metals by human results in addition to one or more of heavy metals in the soil and water which is dangerous to human health, plants and animals (12–14). At the cellular level, toxic effect of HMs is mediated by producing reactive oxygen species (ROS) and causing oxidative stress. Activation of various antioxidative defence mechanisms and reduction in the uptake of heavy metal are the defense mechanisms used by the plants to sustain their growth and productivity under HM toxicity (11). General effects of heavy metal stress on the plants are illustrated in Fig. 1.

During past few years various seed priming methods have been developed to overcome the toxic and adverse effects of HM on crop plants and agricultural systems, to fulfil the global food demand. In this review seed priming is discussed in the context of their potential for alleviation of metal stress.

**Seed priming methods**

Seed priming is a pre-sowing treatment that offers the possibility to improve post-harvest seed quality and allows the break of dormancy leading to the increase germination rate, speed and uniformity. The seed priming involves the initiation of germination metabolism by controlling the hydration of seeds and activating various metabolic processes before radical protrusion (15, 16). Some natural and synthetic compounds which are used to prime the seeds before germination, induce biological alteration in plants (8, 17). Further, seed priming methods have become a promising approach in protecting plants from biotic and abiotic stresses (8, 17) by quicker germination and a harmless priming method which is preferred for increased osmotic adjustment ability of the seeds, seedling establishment and crop yield under non-stress as well as stress conditions (26). Karalića and Selović (27) found an improvement in photosynthetic activity and carbohydrate metabolism in hydroprimed maize seeds grown under Cd stress. Kumar and Boss (28) also suggested that hydroprimed seeds may conquer the phytotoxic effects of the HM stress in the germinating seeds of rice due to increase in α-amylase activity. It has also been suggested that the inhibitory effect of mercuric chloride (HgCl\(_2\)) on seed germination and seedling growth of wheat can be alleviated by hydropriming (21).

**Hormonal-priming**

In this kind of priming seeds are pre-treated with different hormones such as auxins, cytokinins, gibberellins, salicylate etc. which stimulate the seedling’s growth and development (8, 28–30). In
agronomical crop management practices, phytohormones as regulator of heavy metal absorption have been used to alleviate the HM toxicity (8, 30). It was found that seed germination in Cd contaminated areas can be efficiently improved using hormonal priming with auxin, cytokinin, gibberellin, abscisic acid and ethylene in pigeon pea (Cajanus Cajan) (31). It was reported that Salicylic acid (SA) and 24-epibrassinolide increase the relative water content and HM tolerance index in Brassica juncea (32). Further, pre-treatment of seeds with the combination of 24-epibrassinolide and SA, lessen the adverse effects of Pb stress through alteration in antioxidative defense response and enhanced osmolyte contents in B. juncea (33). It was reported that SA and sodium hydrosulfide (NaHS) pretreatments decrease the adverse effects of Pb stress in Zea mays plants through increase in glycine betaine (GB) and nitric oxide (NO) contents and by regulation of genes participating in methionine metabolism (34). Under Cr stress, seed priming with SA improved the morphological, physiological, biochemical and metabolic parameters of rice (35). Osmopriming with polyethylene glycol (PEG) and hormo-priming with gibberelic acid (GA) improved germination and early seedling growth of white clover in a heavy metal-contaminated soil (36).

Chemical priming

In this kind of priming, seeds are pre-treated with various chemicals such as selenium (Se), Silicon (Si), paclobutrazol, calcium chloride, potassium phosphate, chitosan, putrescine, butenolide, nitric oxide, hydrogen peroxide, hydrogen sulfide, melatonin, defensins and polynamines to improve germination rate and enhance abiotic stress tolerance (37-41). Defensins are specifically reported to increase Zn tolerance at higher concentrations (50 and 100 mM) in wheat germinated grains through alleviating the oxidative stress by up regulation of the antioxidant enzymes (42). This could be an unexpected role for defensins which opens up new horizons for the investigation of defensin mechanisms of action (42). Melatonin has been found to ameliorate copper toxicity through improvement in copper sequestration, activating the ROS scavenging, changing the gene expressions, increasing the levels of glutathione and phytochelatin and further improved the distribution of nutrient elements which were disturbed by Cu²⁺ (43). Cadmium induced inhibition of growth, chlorophyll contents, gas-exchange attributes and photosynthetic efficiency in Vicia faba has been reported to be alleviated by calcium chloride (44). The significant reduction in boron toxicity by reducing the oxidative damage and increased activities of antioxidative enzymes has been shown by calcium (45). Pre-soaking of seeds with Se (5, 10 and 20 μM) alleviated the negative effect of cadmium (Cd) on growth and due to a decrease in oxidative injuries caused by Cd (46).

Silicon (Si) has been reported to alleviate the toxicity of various heavy metals in crops plants (47, 48) through an increase in the pH of the growth media (49), stimulation of the antioxidative enzymes (50, 51) and reduction of metal uptake (48, 52). Further, it was reported that Si lessens the Cd toxicity in wheat seedlings by increasing the plant growth and antioxidant capacity by reducing the uptake of Cd and lipid peroxidation (52).

Biological priming or bio-priming

It consists of coating the seeds with a bacterial bio-control agent such as Pseudomonas aureofaciens Kluvyer AB254 and after that the seeds were hydrated for 20 hrs under warm conditions (23°C) in a self-sealing plastic bag in moist vermiculite or on moist germination blotters and before radicle emergence the seeds were removed (53). Applying beneficial microorganisms to the seeds during priming may further improve the development of the crop and it may help the plant to be healthy for long duration (54). It was reported that application of plant–microbe synergy to restore lands, contaminated with HMs is a promising technique (55). It has been suggested the benefits of heavy metal tolerant-plant growth promoting (HMT-PGP) bacterial strains (PGPB) such as Alcaligenes faealis MG257493.1, Bacillus cereus MG257494.1 and A. faealis MG966440.1; as they perform various functions like increased plant growth and removal or detoxification of HM from the soil (56). Further, these three HMT-PGBP strains were employed for alleviation of the toxic effects of heavy metal’s on Sorghum bicolor and increased its growth characteristics (57).

Magnetopriming (MP)

MP involves treatment of seeds using a magnetic field before sowing. There is a great impact of such physical treatment on multiple levels from morpho-structural aspects to changes in gene expression (22). Magnetopriming with static magnetic field (SMF) enhance the speed of germination, plant height, leaf area, photosynthetic pigment, efficiency of PS II, rate of photosynthesis and yield of soybean plants under salt and UV-B stress (20, 22). It was observed that MP mitigates the adverse effect of cadmium stress by reducing the level of malondialdehyde, H₂O₂ and O₂⁻ and increased the growth and photosynthetic parameters, NO content and nitric oxide synthase activity (58). It has been reported that the mechanism of alleviation of adverse effects of salt and HMs stress by MP is related to NO signaling (22, 58). It was found that metallothionein and receptor for activated C kinase 1 (RACK1) play a critical role in the ROS mediated signal transduction pathway to enhance the seed germination and seedling vigor in magnetoprimed tomato seeds (59).

Nanopriming

It includes the use of several metal nanoparticles (NPs), such as AuNPs, AgNPs, FeNPs, CuNPs, ZnNPs, ZnONPs and TiO₂NPs etc. Nanoparticles based on carbon (e.g., fullerene and carbon nanotubes) are also useful as seed priming agents for promoting growth characteristics and improve the stress tolerance in crop plants. Nano-TiO₂ alleviates the Cd toxicity in the plants by increase in the growth and photosynthesis of plants (60). Nano-scale hydroxyapatite can alleviate the Cd toxicity in Brassica juncea (61). It was also found that Si NPs protect maize seedlings against arsenic (As) toxicity through enhancing the activities of antioxidiant
enzymes like ascorbic acid peroxidase (APX), dehydroascorbate reductase (DHAR), superoxide dismutase (SOD), glutathione reductase (GR) and limiting the accumulation of As and ROS (62). Nano-priming with AgNPs enhanced the α-amylase activity, resulting in higher soluble sugar content for supporting seedling growth. Furthermore, these NPs stimulate the up-regulation of aquaporin genes in germinating seeds (63). It was found that seed priming with Si NPs positively increase the growth, biomass and yield of wheat plants under cadmium contaminated soil (18).

Conclusion
These methods of seed-priming are the best way to grow the crops under unfavorable conditions and overcome the germination related problems, reduce germination time, improve crop yield and when crops are grown in HM contaminated areas. Significant amount of work has been carried out on the importance of seed-priming techniques for different crop plants affected by the heavy metal stress. The data presented in this mini review can be useful for developing agro-ecological technology based on the exogenous application of seed priming agents to improve tolerance under HM contamination which may contribute to the agricultural or ecological sectors, and explore ways for further improvisation. This low input technique is sustainable and will help in reclamation of HMs contaminated soils, thus increasing the quality of seeds and crop yield in such areas.

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SK and MJ drafted the outline of review; writing -original draft preparation by RP and SK; writing-review and editing and revision by SK and MJ.

Conflict of interest
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