

**RESPONSIVENESS INDEX OF SORGHUM (*SORGHUM BICOLOR* (L.)
MOENCH) GROWN UNDER CADMIUM CONTAMINATED SOIL
TREATED WITH PUTRESCINE AND MYCORRHIZA**

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Abstract

Effect of polyamine (putrescine) and mycorrhiza combination on putrescine and mycorrhiza responsiveness index was studied in sorghum. It is evident that the average putrescine and mycorrhiza responsiveness index was non-significantly enhanced by 48.40, 48.43 and 46% when exposed to higher dose of polyamine (putrescine) and mycorrhiza combination (T5) as compared to lower dose of polyamine (putrescine) and mycorrhiza combination (T4) at 30, 60 and 90 DAS of interval. Similarly, when plants were exposed to higher dose of polyamine (putrescine) and mycorrhiza combination (T11) than its putrescine and mycorrhiza responsiveness index was non-significantly enhanced by 11.03, 13.22 and 13.18% as compared to T10. When treatment, T17 was compared to T16, the putrescine and mycorrhiza responsiveness index increased significantly by 7.13, 6.73 and 5.35% at proposed DAS. So, the combination of putrescine and mycorrhiza showed the best combination for the mitigation of cadmium toxicity for the putrescine and mycorrhiza responsiveness index.

Introduction

Plants growing in metal contaminated soils harbor a diverse group of microorganism (Idris *et al.* 2004, Zarei *et al.* 2008) that were capable of tolerating high concentration of metal and providing a number of benefits to both the soil and the plant. Among the microorganisms involved phytoremediation of heavy metal, the rhizosphere bacteria deserve special attention because they can directly improve the phytoremediation process by changing the metal bioavailability through altering the soil pH, release of chelators (Organic acid and siderophores), oxidation/reduction reaction (Gadd 2000, Khan *et al.* 2009, Kidd *et al.* 2009, Ma *et al.* 2011a). Similarly the metal tolerant mycorrhizal fungi were also been frequently reported in hyper-accumulators growing in metal polluted soils indicating that these fungi have evolved a heavy metal-tolerance and that they may play important role in phytoremediation of the site (Gohre and Paszkowski 2006, Miransari 2011). Plant associated-microbes can also immobilize the heavy metals in the rhizosphere through metal reduction reaction. For example, inoculation of Cr-resistant bacteria *Cellulosimicrobium cellulans* to seeds of green chilli grown in Cr (VI) contaminated soils decreased Cr uptake into the shoot by 37% and root by 56% compared with uninoculated control. This study indicated that bacteria reduced the mobile and toxic Cr (IV) to nontoxic and immobile Cr (III) in the soil. The plant-associated microbes may also contribute in plant metal uptake through biosorption mechanism (Rajkumar *et al.* 2010). Biosorption can be defined as the microbial adsorption of soluble/insoluble organic/inorganic metals by a metabolism dependent, active process (Ma *et al.* 2011a). Several authors have pointed out that bacterial biosorption mechanism accounted for reduced plant metal uptake. Sorghum is one of the fundamental staple nourishments for the world's

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poorest and most sustenance unsecured individuals over the semi-dry tropics. Sorghum belongs to the grass family Graminae. It is a short-day C4 plant. The ideal photoperiod which actuates flower formation is in the vicinity of 10 and 11 hours. Cadmium is a chemical element with symbol Cd and atomic number 48. This soft, bluish-white metal is chemically similar to the two other stable metals in group 12, zinc and mercury. Because of a strong demand for cadmium worldwide, particularly in the nickel-cadmium battery industry, approximately 30,000 t of cadmium is released into the atmosphere each year, with an estimated amount of 4,000 - 13,000 t coming from industrial activities (Gohre and Paszkowski 2006). The strong affinity of Cd²⁺ ions for sulhydryl groups of several compounds and phosphate groups involved in plant metabolism might explain the greater toxicity. Polyamines (PAs), widely present in living organisms, are now regarded as a new class of growth substances which include spermidine (Spd, a triamine), spermine (Spm, a tetramine), and their obligate precursor putrescine (Put, a diamine) which play a pivotal role in the regulation of plant development. Mycorrhizal roots may act as a barrier against metal transport, reducing metal transfer and enhancing root/shoot Cd ratios. This effect is attributed to metal adsorption on hyphal walls, since chitin has an important metal-binding capacity [3]. Glomalin, a glycoprotein produced by arbuscular mycorrhizal fungi (AMF), may have a metal chelating function diminishing metal availability for plants.

Materials and Methods

A pot experiment was conducted in the nethouse of Department of Plant Physiology, Institute of Agricultural Sciences, Banaras Hindu University located in the south-eastern part of Varanasi city which is at 25°18' N latitude, 83°03' E longitude and at an altitude of 75.7 m above mean sea level.

Chemicals and mycorrhiza were available in the laboratory of stress physiology and plant tissue culture, Department of Plant Physiology, Institute of Agricultural Sciences, Banaras Hindu University, Uttar Pradesh.

The present study was carried out to evaluate the compatibility of polyamines (putrescine) and mycorrhiza in the mitigation of induced toxic effect of cadmium at 30, 60 and 90 DAS of sorghum. The pot size for the experiment was in the diameter of 30 and 25 cm in height with capacity for 10 kg of soil, having a small hole at the bottom. Soil was collected from Experimental Farm, Institute of Agricultural Sciences, Banaras Hindu University. Pots containing soil mix (Soil + FYM in 3 : 1) were inoculated with seeds of sorghum and kept under nethouse condition with consistent care and precaution. According to plan of work, targeted pots were inoculated with Endomycorrhiza *Glomus* sp. The source of *Glomus* sp. was Tata Energy Research Institute (TERI), New Delhi. Heavy metal stress was created in plants by the exogenous application of cadmium nitrate in soil. Two best concentrations of heavy metals on the basis of initial screening were selected, i.e., 0.07 % per 10 kg of soil and 0.15 % per 10 kg of soil. Putrescine was applied at the rate of 2.5 and 5.0 mM through foliar spray at the seven DAS of interval. Various observations were taken at three stages such as 30, 60 and 90 DAS after sowing in the concerned pots.

Sorghum seeds were soaked in distilled water overnight before the day of sowing and then 10 seeds were sown in each pot. The depth of sowing was 5 cm. The pots were watered daily and the soil was fertilized with FYM in order to ensure healthy growth of the seedlings.

The solution was made at two successive concentrations are of 2.5 and 5.0 mM and putrescine was applied at the rate of 2.5 and 5.0 mM through foliar spray at the seven DAS of interval. The experiment was laid out in completely randomized design (CRD). There were 18 treatments including control. Each treatment was replicated five times.

Results and Discussion

Effect of polyamine (putrescine) and mycorrhiza combination on putrescine and mycorrhiza responsiveness index was studied in sorghum. It is evident that the average putrescine and mycorrhiza responsiveness index was non-significantly enhanced by 48.40, 48.43 and 46% when exposed to higher dose of polyamine (putrescine) and mycorrhiza combination (T5) as compared to lower dose of polyamine (putrescine) and mycorrhiza combination (T4) at 30, 60 and 90 DAS of interval. Similarly, when plants were exposed to higher dose of polyamine (putrescine) and mycorrhiza combination (T11) than its putrescine and mycorrhiza responsiveness index was non-significantly enhanced by 11.03, 13.22 and 13.18% as compared to T10. When treatment, T17 was compared to T16, the putrescine and mycorrhiza responsiveness index increased significantly by 7.13, 6.73 and 5.35% at proposed DAS. So, the combination of putrescine and mycorrhiza showed the best combination for the mitigation of cadmium toxicity for the putrescine and mycorrhiza responsiveness index (Fig.1).

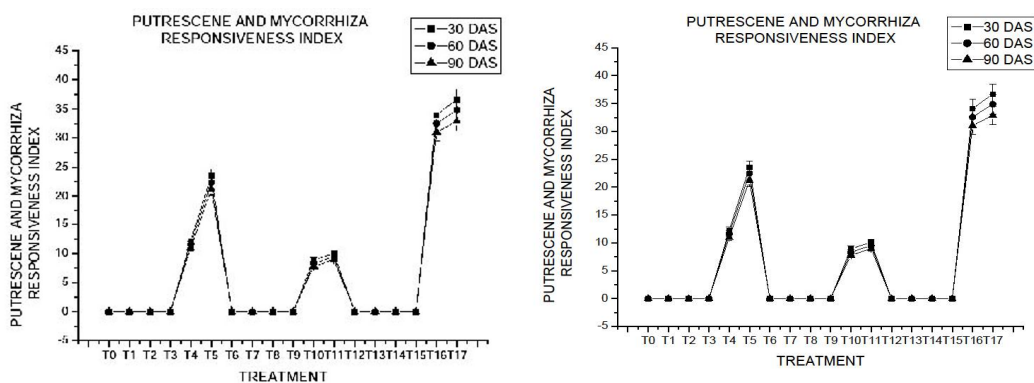


Fig. 1. Putrescine and mycorrhiza responsiveness index of sorghum during Kharif 2015-16 and 2016-17 [left to right]

where, DAS = Days after sowing. Data are in the form of mean \pm SEM. S = Significance at $p \leq 0.05$ and $p \leq 0.01$, NS = Non significant at $p \leq 0.05$ and $p \leq 0.01$ using Origin 6.1. T0 = Control. T1 = Control + mycorrhiza. T2 = Control + 2.5 mM putrescine. T3 = Control + 5 mM putrescine. T4 = Control + 2.5 mM putrescine + mycorrhiza. T5 = Control + 5 mM putrescine + mycorrhiza. T6 = 0.07% $\text{Cd}(\text{NO}_3)_2$. T7 = 0.07% $\text{Cd}(\text{NO}_3)_2$ + mycorrhiza. T8 = 0.07% $\text{Cd}(\text{NO}_3)_2$ + 2.5 mM putrescine. T9 = 0.07% $\text{Cd}(\text{NO}_3)_2$ + 5 mM putrescine. T10 = 0.07% $\text{Cd}(\text{NO}_3)_2$ + 2.5 mM putrescine + mycorrhiza. T11 = 0.07% $\text{Cd}(\text{NO}_3)_2$ + 5 mM putrescine + mycorrhiza. T12 = 0.15% $\text{Cd}(\text{NO}_3)_2$. T13 = 0.15% $\text{Cd}(\text{NO}_3)_2$ + mycorrhiza. T14 = 0.15% $\text{Cd}(\text{NO}_3)_2$ + 2.5 mM putrescine. T15 = 0.15% $\text{Cd}(\text{NO}_3)_2$ + 5 mM putrescine. T16 = 0.15% $\text{Cd}(\text{NO}_3)_2$ + 2.5 mM putrescine + mycorrhiza and T17 = 0.15% $\text{Cd}(\text{NO}_3)_2$ + 5 mM putrescine + mycorrhiza.

Effect of mycorrhiza on mycorrhizal responsiveness index was studied in sorghum grown under the cadmium stress. Data were recorded at 30, 60 and 90 days after sowing (DAS). It is evident that the average mycorrhizal responsiveness index was significantly enhanced by 10.61, 10.61 and 10.62% when exposed to heavy metal stress along with mycorrhiza (T7) as compared to control plus mycorrhiza (T1) at 30, 60 and 90 DAS of interval. Similarly, when plants were exposed to higher dose of heavy metal (T12) then its mycorrhizal responsiveness index was significantly reduced by 87.26, 86.83 and 87.26% as compared to control (T1) on the dates of proposed interval. So, the mycorrhiza showed the positive combination for the mitigation of cadmium toxicity for the mycorrhizal responsiveness index (Fig. 2).

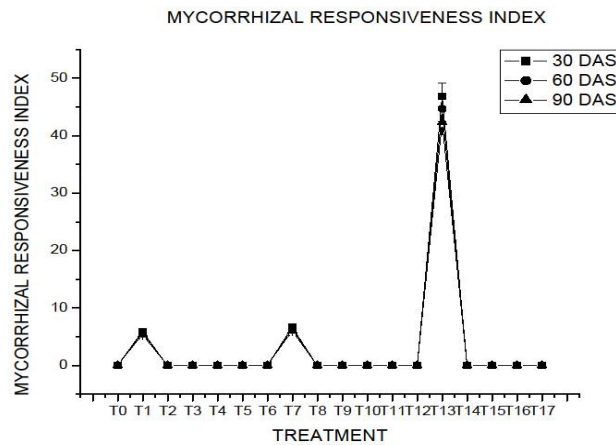


Fig. 2. Mycorrhizal responsiveness index of sorghum grown under Cd-stress

Effect of polyamine (putrescine) was studied in putrescine responsive index sorghum grown under the cadmium stress. Data were recorded at 30, 60 and 90 days after sowing (DAS). It is evident that the average putrescine responsive index was significantly enhanced by 48, 38 and 27% when exposed to putrescine (T8) as compared to control (T2) at 30, 60 and 90 DAS of interval. Similarly, when plants were exposed to higher dose of putrescine (T9) then its putrescine responsive index was significantly enhanced by 18, 19.1 and 27% as compared to control (T3) on the dates of proposed interval. When treatment, T14 was compared to T2, the putrescine responsive index increased significantly by 79.6, 78.82 and 80.34% at proposed DAS. In comparison to T3, the exogenous application of putrescine (T15) showed mitigating effect by increasing putrescine responsive index by 76.40, 76.44 and 77.82% on proposed DAS. So, the putrescine at the rate of 2.5 and 5 mM showed the positive combination for the mitigation of cadmium toxicity for the mycorrhizal responsiveness index (Fig. 3).

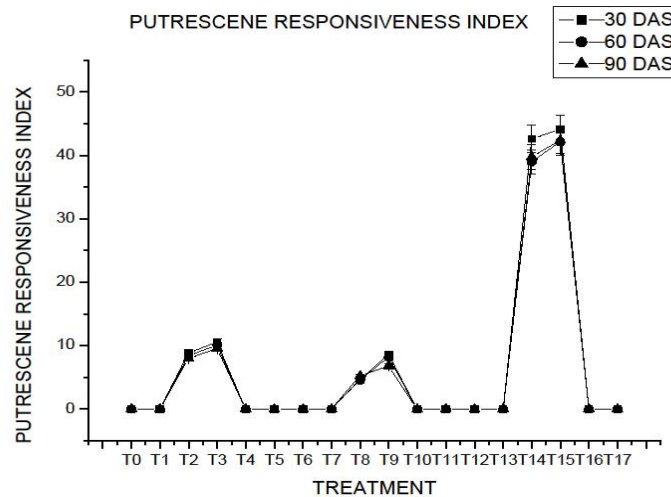


Fig. 3. Putrescine responsiveness index of sorghum grown under Cd-stress.

The combination of putrescine and mycorrhiza showed the best combination for the mitigation of cadmium toxicity for the putrescine and mycorrhiza responsiveness index.

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