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WHEAT BIOCHEMICAL RESPONSE TO CADMIUM TOXICITY UNDER *FUNNELIFORMIS MOSSEAE* AND *PIRIFORMOSPORA INDICA* SYMBIOSIS

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Abstract

Shahabivand S., Aliloo A.A., Maivan H.Z., 2016: Wheat biochemical response to cadmium toxicity under *Funneliformis mosseae* and *Piriformospora indica* symbiosis [Kviečių, esančių simbiozėje su *Funneliformis mosseae* ir *Piriformospora indica*, biocheminis atsakas į kadmio toksinį poveikį]. – Bot. Lith., 22(2): 169–177.

The effects of root endophytic fungus *Piriformospora indica* and arbuscular mycorrhizal fungus *Funneliformis mosseae* were investigated on some biochemical parameters in leaves and roots of *Triticum aestivum* cv. 'Sardari39' under Cadmium (Cd) stress. The experiment was carried out with inoculation treatments of *F. mosseae*, *P. indica*, *F. mosseae* + *P. indica* and no-inoculation (control) at four Cd concentrations (0, 0.3, 0.6 and 0.9 mM Cd). The results revealed that in non-inoculated plants, H_2O_2, malondialdehyde (MDA) and proline contents increased in leaves and roots in response to increasing soil Cd concentrations. However, guaiacol peroxidase (GPX) activity in roots and superoxide dismutase (SOD) activity in leaves and roots increased at lower Cd concentrations, while at higher Cd concentrations the rate decreased. In the presence of *P. indica* and *F. mosseae* + *P. indica*, H_2O_2 and MDA contents decreased, and proline accumulation increased in wheat leaves and roots. Also, the presence of *P. indica* increased GPX and SOD activities in leaves and roots. The study concluded that *P. indica* and *F. mosseae* were able to maintain an efficient symbiosis with wheat plants in soil at high Cd concentrations. However, the impact of *P. indica* in alleviating Cd stress was more noticeable than that of *F. mosseae*.

Keywords: cadmium, mycorrhiza, stress, wheat.

INTRODUCTION

Cadmium (Cd) is a heavy metal with a high toxicity to plants and humans. One of the important entries of the metal to food chain is crops. Roots easily absorb the metal, and then accumulate through the plants from roots to seeds (Metwally et al., 2005). Cd pollution is a global environmental hazard and nearly 30×10^3 tons of Cd enters to the environment annually through various anthropogenic sources (Junqing et al., 2010). Reduction in growth, leaf roll, chlorosis and necrosis are some of Cd toxicity symptoms in plants (Faizan et al., 2011). The element induces oxidative stress via lipid peroxidation, hydrogen peroxide generation and ion leakage (Cherif et al., 2011). Cadmium also induces activity of antioxidant enzymes, and mostly causes their imbalance (Chen et al., 2010). Similarly, high rates of the non-enzymatic antioxidants such as proline are induced by this toxin (Zhao, 2013). Arbuscular mycorrhizal (AM) fungi (such as *Glomus mosseae*) are important soil microorganisms forming symbiotic associations with most of the vascular plant families. The stress amelioration effects of mycorrhizae in metal-contaminated soils have been recognized in various plant species (Andrade et al., 2010). Improved nutritional status and altered metal uptake are among the most related benefits of mycorrhizal association to

host plants under metal stress (Ali et al., 2015). It has been shown that in AM-inoculated plants, the activity of antioxidant enzymes such as catalases (CAT) and superoxide dismutases (SOD) significantly increases in response to metal stress, also the proline content, which involves in the cellular defence against oxidative stress, induces by both mycorrhizal association and stresses (Sarkar et al., 2016). Along with AM fungi, the endophytic fungus *Piriformospora indica* colonizes the roots of many plant species, which promotes their growth, seed yield and tolerance to stresses (Lareen et al., 2016). It has been reported that *P. indica* protects barley plants from high salt concentrations (Murphy et al., 2015). Vahabi et al. (2015) found enhanced glutathione reductase (GR) activity in leaves of *Arabidopsis* plants colonized by *P. indica.* The activation of the antioxidant enzyme systems is a major target of *P. indica* in leaves of barley and *Arabidopsis thaliana* (Baltruschat et al., 2008; Oelmüller et al., 2009). The colonization of maize plants by *P. indica* leads to increased growth (due to its growth-promoting abilities) and enhanced antioxidant capacity (Kumar et al., 2009). The similar result has been also reported by Sun et al. (2010) on Chinese cabbage at drought stress. Reports on the effects of interactions of *P. indica* and *F. mosseae* on plant biochemical and physiological changes are scarce under metal stress conditions. Therefore, this study was carried out to investigate the interaction effects of *P. indica* and *F. mosseae* on MDA, H_2O_2, proline contents and the activity of antioxidant enzymes of GPX and SOD in *Triticum aestivum* cv. 'Sardari39' under different soil Cd levels.

MATERIALS AND METHODS

Plant materials. Wheat seeds (*Triticum aestivum* cv. 'Sardari39') were obtained from the Dryland Agricultural Research Institute, Maragheh, Iran. The seeds were surface sterilized by soaking in 1% NaClO for 20 minutes, then, rinsed with distilled water five times and allowed to vernalize between layers of filter paper wetted with sterile distilled water at +2°C for one month.

Soil preparation. The experimental soil was collected from the surface horizon of the campus farm of Maragheh University. It contained sand (65%), silt (23%), clay (12%); pH 7.3 and EC 1.3 ds/m. The soil samples were air-dried, sieved to pass 2 mm and were steam sterilized (100°C for 1 h, three consecutive days) by autoclaving to eliminate native AM fungus propagules as well as other microorganisms. After sterilization, the soil was spiked with $CdCl_2$ to obtain the concentrations as follows: 0, 0.3, 0.6 and 0.9 mM Cd. Samples then were incubated at 20°C for one month allowing metal to distribute into various fractions and to equilibrate with soil solid phase.

Fungal materials. *Piriformospora indica* was cultured in Petri dishes on a modified Kaefer medium (Sherameti et al., 2005). The plates were placed in a temperature-controlled growth chamber at 25°C for two weeks. The liquid culture was kept in shaker incubator at 100 rpm for 15 days at room temperature. The amount of 50 ml liquid culture was added to each pot that was treated with *P. indica*. *Funneliformis mosseae* inoculum consisted of spores, soil, hyphae and infected maize root fragments (supplied from the Department of Biology, Maragheh University, Maragheh, Iran). The inoculated dosage was 50 g of inoculums per pot, containing approximately 20 spores/g soil.

Planting and growth conditions. The experiment was carried out under greenhouse conditions and consisted of a completely randomized 4 × 4 factorial design. Pots were filled with 5 kg of sterilized sandy soil that contained Cd of four concentrations (0, 0.3, 0.6, 0.9 mM Cd), each treatment was replicated four times. The fungal treatments were: (1) inoculation of the AM fungus *Funneliformis mosseae* (50 g of inoculated soil); (2) inoculation of the root endophytic fungus *Piriformospora indica* (50 ml of liquid culture); (3) inoculation of *F. mosseae* and *P. indica* together (co-inoculation); (4) no-inoculation (control). Mycorrhizal and endophytic fungal inoculums were placed 2 cm below wheat seeds at sowing time. Non-AMF and non-endophytic fungal treatments received the same weight of autoclaved soil and growth mixture. The experimental pots were placed in a growth chamber under conditions of 14 h of light, 10 h of darkness, 20–28°C, relative humidity 50–65%, and light intensity 5000 Lux. Ten and twenty days after the sowing time, the tap water was replaced with one-half and full-strength Hoagland solution (with half P content), respectively. Plants were harvested after 45 days. Roots and leaves of the harvested wheat samples were rinsed with tap water

to remove soil particles and then carefully washed with deionized water. The root samples were stored in water for 1 h to study colonization, and for biochemical analysis, the samples of leaves and roots were stored in liquid N_2 immediately.

Root colonization. The percentage of fungal root length infection was estimated by visual observation of fungal colonization after clearing washed root in 10% KOH and staining with 0.05% trypan blue in lactic acid. Quantification was carried out by using the grid-line intersect method.

MDA and H_2O_2 determination, and assay of proline content. Malondialdehyde (MDA) was measured by the colorimetric method. Concentration of H_2O_2 was measured by the method of Velikova et al. (2000). Concentration of H_2O_2 was given on a standard curve. Proline was measured according to the method of Bates et al. (1973). Proline content was calculated from a standard curve.

Enzyme extraction and activity assay. For SOD and GPX extraction, leaf and root samples (0.5 g) were homogenized in ice cold 0.1 M phosphate buffer (pH 7.5) containing 0.5 mM EDTA with pre-chilled pestle and mortar. Each homogenate was transferred to centrifuge tubes and was centrifuged at 4°C for 15 min. at 15000 g. The supernatant was used for enzyme activity assay. SOD (EC 1.15.1.1) activity was estimated by recording the decrease in absorbance of superoxide-nitro blue tetrazolium complex by the enzyme. Absorbance was recorded at 560 nm and one unit of enzyme activity was taken as the quantity of enzyme reducing the absorbance reading of samples to 50% compared to tubes lacking enzymes. Guaiacol peroxidase (EC 1.11.1.7) was measured as the increase in the absorbance at 470 nm when guaiacol polymerizes to tetraguaiacol. Protein content of samples was determined by the method of Bradford (1976). Bovine serum albumin was used as a standard.

Statistical analysis. SAS software (ver. 9.1(was used for the analysis of variance by using GLM procedure, and the comparison of means was performed by Duncan's multiple range tests at 0.05 levels.

RESULTS

In *P. indica*-inoculated plants, root colonization was reduced slightly by increasing soil Cd, whereas in *F. mosseae*-inoculated plants, root colonization was significantly decreased at 0.6 and 0.9 mM Cd compared to 0 and 0.3 mM Cd (Fig. 1). Root colonization in co-inoculated plants significantly decreased only at 0.3 mM Cd compared to control.

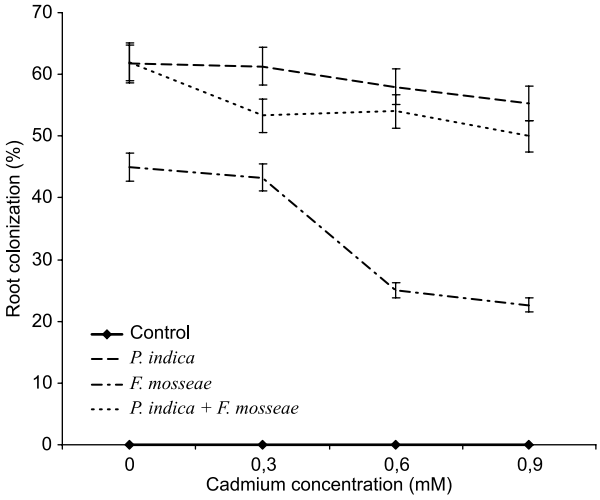

Fig. 1. Root colonization percentage (± SE) in wheat cv. 'Sardari39' by *P. indica* and *F. mosseae* by increasing Cd in the soil

In non-inoculated plants, the Cd concentration significantly ($p < 0.01$) increased MDA (except for roots at 0.3 mM Cd), H_2O_2 and proline contents in wheat leaves and roots. Presence of *P. indica* and *P. indica* + *F. mosseae* significantly reduced MDA and H_2O_2 contents, whereas it increased proline accumulation in leaves and roots, except for root MDA at 0.3 mM Cd, and root H_2O_2 at 0 mM Cd. Presence of *F. mosseae* significantly decreased MDA of leaves and roots (except for roots at 0.3 mM Cd), H_2O_2 of leaves at 0 and 0.3 mM Cd and H_2O_2 of roots at 0.3 mM Cd, whereas it increased the proline of leaves at 0, 0.3 and 0.6 mM Cd and root proline contents at 0 and 0.3 mM Cd in the soil (Tables 1, 2, 3).

However, the effect of *P. indica* in reducing MDA and H_2O_2 contents, and in increasing proline accumulation was more noticeable than that of *F. mosseae* in both leaves and roots. The highest H_2O_2 and MDA contents were observed in non-inoculated plants at 0.9 mM Cd, whereas the least MDA and H_2O_2 resulted in the presence of *P. indica* at 0 mM Cd in wheat leaves and roots (Tables 1, 2). Also the highest and the lowest proline accumulation in both leaves and roots were recorded in the presence of *P. indica* at 0.9 mM Cd and in non-inoculated wheats at 0 mM

Table 1. Mean comparison of MDA (nM/gFW) contents affected by mycorrhiza and cadmium various concentrations in roots and leaves of wheat cv. 'Sardari39'

Fungi treatments	Roots				Leaves			
	Cadmium concentration (mM)				Cadmium concentration (mM)			
	0	0.3	0.6	0.9	0	0.3	0.6	0.9
Control	33.6 f	29.3 fg	68.5 c	96.5 a	176.4 h	302.7 c	319.9 b	339.1 a
Piriformospora indica (P)	24.8 g	27.0 g	53.1 e	66 cd	149.6 j	210.1 g	227.6 f	242.3 e
Funneliformis mosseae (G)	27.3 g	28.5 g	61.8 d	87.5 b	160.8 j	212.5 g	272.9 d	310.0 c
P + G	24.9 g	27.1 g	54.5 e	67.9 c	149.9 j	212.7 g	233.5 f	248.7 e

Same letters at rows and columns are not different at $p \leq 0.01$; MDA: malondialdehyde.

Table 2. Mean comparison of H_2O_2 (mM/gFW) contents affected by mycorrhiza and cadmium various concentrations in roots and leaves of wheat cv. 'Sardari39'

Fungi treatments	Roots				Leaves			
	Cadmium concentration (mM)				Cadmium concentration (mM)			
	0	0.3	0.6	0.9	0	0.3	0.6	0.9
Control	0.029 de	0.045 c	0.063 b	0.081 a	0.068 f	0.105 c	0.127 b	0.174 a
Piriformospora indica (P)	0.019 e	0.029 de	0.043 c	0.061 b	0.032 g	0.059 f	0.080 e	0.09 de
Funneliformis mosseae (G)	0.024 de	0.030 d	0.061 b	0.077 a	0.040 g	0.067 f	0.120 b	0.164 a
P + G	0.023 de	0.032 d	0.045 c	0.063 b	0.033 g	0.059 f	0.085 de	0.094 cd

Same letters at rows and columns are not different at $p \leq 0.01$.

Table 3. Mean comparison of proline (mM/gFW) contents affected by mycorrhiza and cadmium various concentrations in roots and leaves of wheat cv. 'Sardari39'

Fungi treatments	Roots				Leaves			
	Cadmium concentration (mM)				Cadmium concentration (mM)			
	0	0.3	0.6	0.9	0	0.3	0.6	0.9
Control	0.298 f	0.464 d	0.534 c	0.566 c	0.522 j	1.038 g	1.252 f	1.353 de
Piriformospora indica (P)	0.463 d	0.566 c	0.664 b	0.723 a	0.847 h	1.296 ef	1.463 a–c	1.517 a
Funneliformis mosseae (G)	0.409 e	0.533 c	0.568 c	0.564 c	0.721 i	1.231 f	1.391 cd	1.408 b–d
P + G	0.463 d	0.556 c	0.638 b	0.737 a	0.845 h	1.255 f	1.416 b–d	1.485 ab

Same letters at rows and columns are not different at $p \leq 0.01$.

Cd, respectively (Table 3). *P. indica* and *P. indica* + *F. mosseae*-inoculated plants contained the lowest MDA and H_2O_2 (except for root H_2O_2), and the highest accumulation of proline in leaves and roots compared to that in control and *F. mosseae*-inoculated plants (Tables 1–3).

The application of Cd to soil caused alterations in the GPX and SOD activities (Table 4 and 5). In non-inoculated plants, GPX activity in leaves significantly increased between 0–0.6 mM Cd. Presence of *P. indica* significantly ($p < 0.01$) increased GPX activity at 0, 0.3 and 0.6 mM Cd in wheat leaves compared to non-inoculated wheat plants (Tables 4–5). Presence of *F. mosseae* significantly increased GPX activity in leaves at 0.3 mM Cd, whereas it decreased the GPX activity in leaves at 0.6 and 0.9 mM Cd compared to that of non-inoculated plants (Table 4). In co-inoculated plants, GPX activity in leaves increased at 0 and 0.3 mM Cd compared to non-inoculated plants. The highest and the lowest GPX activities in leaves were observed in the presence of *P. indica* at 0.6 mM Cd, and in non-inoculated plants at 0 mM Cd, respectively (Table 4). In non-inoculated wheats, GPX activity in roots significantly increased between 0.3– 0.6 mM Cd, whereas it decreased between 0.6–0.9 mM Cd. In the presence of *P. indica* and *F. mosseae*, GPX activity in roots significantly increased only at 0 and 0.3 mM Cd. In non-inoculated plants, no significant difference was found in SOD activity in leaves at 0.3 mM Cd compared to 0 mM Cd, whereas by increasing soil Cd, it was decreased (Table 5). In roots of non-inoculated plants, at 0.3 mM Cd significant increase in SOD activity compared to that of 0 mM Cd was observed. However, between 0.3–0.6 mM

Table 4. Mean comparison of GPX (U/mg prot. min) activity affected by mycorrhiza and cadmium treatments in roots and leaves of wheat cv. 'Sardari39'

Fungi treatments	Roots				Leaves			
	Cadmium concentration (mM)				Cadmium concentration (mM)			
	0	0.3	0.6	0.9	0	0.3	0.6	0.9
Control	2.23 g	2.42 fg	2.83 be	2.51 ef	0.136 h	0.251 f	0.575 b	0.603 b
Piriformospora indica (P)	2.80 b–d	2.97 ab	2.91 ab	2.36 fg	0.208 fg	0.412 d	0.673 a	0.606 b
Funneliformis mosseae (G)	2.60 c–e	3.13 a	2.74 b–e	2.55 d–f	0.181 gh	0.334 e	0.496 c	0.455 cd
P + G	2.71 c–d	3.11 a	2.79 b–d	2.39 fg	0.217 fg	0.461 cd	0.616 b	0.579 b

Same letters at rows and columns are not different at $p \leq 0.01$; GPX: guaiacol peroxidase.

Table 5. Mean comparison of SOD (U/mg prot. min) activity affected by mycorrhiza and cadmium treatments in roots and leaves of wheat cv. 'Sardari39'

Fungi treatments	Roots				Leaves			
	Cadmium concentration (mM)				Cadmium concentration (mM)			
	0	0.3	0.6	0.9	0	0.3	0.6	0.9
Control	35.27 c	44.98 ab	44.32 ab	35.48 c	33.6 de	34.8 cd	30.3 e	29.6 f
Piriformospora indica (P)	44.39 ab	45.48 ab	49.20 a	47.84 ab	42.6 a	43.4 a	31.3 ef	31.3 ef
Funneliformis mosseae (G)	38.29 c	44.32 ab	46.10 ab	43.11 b	42.7 a	36.8 bc	28.9 f	28.4 f
P + G	48.46 a	47.29 ab	47.61 ab	42.92 b	39.1 b	37.2 bc	29.6 f	29.4 f

Same letters at rows and columns are not different at $p \leq 0.01$; SOD: superoxide dismutase.

Cd there was no difference, but between 0.6–0.9 mM Cd significant decrease resulted in SOD activity in roots. Presence of *P. indica* significantly increased SOD activity in leaves at 0 and 0.3 mM Cd, and SOD activity in roots at 0 and 0.9 mM Cd compared to non-inoculated wheats. Presence of *F. mosseae* significantly increased SOD activity in leaves at 0 mM Cd and SOD activity in roots at 0.9 mM Cd. Co-inoculated plants significantly increased SOD activity in leaves at 0 mM Cd and SOD activity in roots at 0 and 0.9 mM Cd compared to non-inoculated plants. In inoculated and non-inoculated plants, there was no difference in SOD activity in leaves at 0.6 and 0.9 mM Cd (Table 5). The SOD activity in roots did not alter in inoculated and non-inoculated plants between 0.3–0.6 mM Cd.

DISCUSSION

In response to increased Cd concentrations in the soil, root colonization was reduced in *F. mosseae*, *P. indica* and co-inoculated plants. However, *F. mosseae* was more sensitive than *P. indica* at 0.6 and 0.9 mM Cd in the soil. This indicated that elevated concentrations of Cd in the soil seemed to exert harmful effects on spore production of *F. mosseae*. This is in agreement with the results of Redon et al. (2009), who showed that total AM fungal spore numbers decreased with increasing concentrations of heavy metals Cd, Zn and Pb in contaminated agricultural soils. Besides the heavy metals, the other soil properties such as organic matter, soil type and pH influence spore production of AM fungi in the rhizosphere (Islas et al., 2016). The results revealed significant colonization differences between fungal genera treatments and the plant. Also, the responses of the treatments were remarkably different under cadmium toxicity. Results of this study indicate that Cd exposure resulted in oxidative stress measured in terms of MDA content and H_2O_2 generation in wheat leaves and roots; moreover there was an up-regulation of scavenging enzymes such as SOD and GPX to counter Cd-induced stress. MDA is an indicator of lipid peroxidation and links to peroxidation of polyunsaturated fatty acids in the membrane thereby releasing ROS like H_2O_2 (Gill & Tuteja 2010). The increased MDA content in response to Cd exposure is one of the mechanisms of Cd toxicity (Ahmad et al., 2011). According to Luo et al. (2011), the Cd-treated perennial ryegrass exhibited a greater level of malondialdehyde and activity of the peroxidase, catalase, and superoxide dismutase relative to the control. Such a substantial increase in H_2O_2 in response to Cd has also been reported in roots of bread wheat and it

was correlated to oxidative stress in roots (Huang et al., 2015). Increased levels of MDA and H_2O_2 indicated that Cd exposure results in generation of ROS, which are highly toxic molecules and cause cellular damage in plants (Mishra et al., 2014). In this work, we observed decreased H_2O_2 and MDA contents in wheat leaves and roots colonized with *P. indica* and *F. mosseae*. Fungal inoculation notably decreased H_2O_2 in roots and leaves, indicating a lower accumulation of H_2O_2 in fungi-inoculated plants. Fester & Hause (2005) resulted that in the root cells of AM roots locally induced accumulations of H_2O_2 were limited both in the intra-cellular AM hyphae and at the intercellular hyphal surface. Rangel with coworkers (2014) found phytoprotective effect of arbuscular mycorrhizal fungi species against arsenic toxicity in tropical leguminous species. The overall level of MDA was lower in *P. indica*-colonized plants compared to *F. mosseae*-inoculated and control plants, and, thus, the fungus could partially counteract this stress response. We suggested that *P. indica* could prevent or retard the degradation of lipids by preventing excess ROS formation under stress conditions.

Chelating effects of proline with Cd has been demonstrated in plants which form a nontoxic Cd-proline complex (Irfan et al., 2013). In this study, with the increasing soil Cd concentration, the enhanced concentrations of proline were observed in both leaves and roots of wheat plants. It could be suggested that free proline might play an important protective role against Cd stress and wheat cv. 'Sardari39' had the strong self-protection capacity. Although metal-induced proline accumulation in plant tissues has been reported (Li et al., 2013; Tripathi et al., 2013), but reports on the effects of *P. indica* and mycorrhizal symbiosis in proline content are scarce under metal stress conditions. In this study, proline content in leaves and roots of inoculated and control wheats increased in response to Cd addition to the soil suggesting a similar stress response to the excess of this metal in soil. Nevertheless, proline contents in leaves and roots of *P. indica*-inoculated plants showed noticeable increases in response to Cd in soil compared to control and *F. mosseae*-inoculated plants, which indicated the possible role of proline in Cd toxicity alleviation.

In non-inoculated (control) plants, Cd treatment resulted in substantial increase in the activity of GPX in leaves and roots, and that of SOD in roots at 0.3 and 0.6 mM Cd compared to 0 mM Cd. The observed enhancement in the activities of antioxidant enzymes in response to Cd exposure is in agreement with other published reports (Farooq et al., 2013; Han et al., 2013; Xu et al., 2016). The results indicated that the increase in GPX and SOD activities was a response to control a possible excessive H_2O_2 and superoxide radical production under Cd stress. At higher concentrations of Cd treatments, GPX activity in roots, and SOD activity in leaves and roots decreased compared to that of lower concentrations of Cd treatments. Such reduction in the activity of antioxidant enzymes at higher concentrations of Cd in the soil has been reported by Pongrac et al. (2009). Significant reductions in GPX and SOD activities in wheat suggest that detoxification of H_2O_2 by GPX, and superoxide radicals by SOD is not sufficient at higher concentrations of Cd in the soil.

In wheat plants, the presence of *P. indica* increased GPX activity in leaves at 0, 0.3 and 0.6 mM Cd, and GPX activity in roots at 0 and 0.3 mM Cd. Co-inoculated plants produced similar results, but in a lesser capacity than that of *P. indica*, except at 0.3 mM Cd. Also *F. mosseae* increased GPX activity in leaves and roots at 0 and 0.3 mM Cd treatment. *P. indica* inoculation increased SOD activity in leaves and roots of wheat plants. The increase in the antioxidant enzyme activities resulting from AM and *P. indica* inoculation have also been reported in the previous works (Wu et al., 2006; Auge et al., 2007; Kumar et al., 2009; Sun et al., 2010). The higher GPX and SOD activities in *P. indica* and *G. mosseae*-inoculated plants would partly explain the lower H_2O_2 and superoxide concentrations in protecting the plants against oxidative damage, in turn enhancing Cd tolerance. In the leaves of *F. mosseae*-inoculated plants at 0.6 and 0.9, and in the roots of *P. indica*-inoculated plants at 0.9 mM Cd, lower GPX activity than in non-inoculated plants was exhibited, which may indicate that at these Cd concentrations other detoxification mechanisms may be involved in such a process. *F. mosseae* inoculation slightly decreased SOD activity in leaves at 0.6 and 0.9 mM Cd, and SOD activity in roots at 0.3 mM Cd. Similar findings have been reported by Jiang et al. (2016), who revealed improvements of CAT, APX and POD in the leaves of mycorrhizal plants and suggested that colo-

nization helped *S. nigrum* to relieve oxidative damage to biomolecules in Cd-contaminated soil.

CONCLUSIONS

The consistent differences were found between inoculated and control plants of wheat cv. 'Sardari39' in response to the increasing of Cd in the soil. We showed that *P. indica*-colonized plants are more resistant to Cd stress than *F. mosseae*-colonized plants, which include a reduction in MDA and H_2O_2 content, and enhanced antioxidant capacity. *P. indica* unlike *F. mosseae* can be propagated in axenic culture, therefore, rendering it useful as a tool for complement crop-growing strategies in cereals. Further studies are required to understand the modulation of host gene expression by *P. indica* and *F. mosseae* and their effects on the phytoremediation potential of wheat.

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KVIEČIŲ, ESANČIŲ SIMBIOZĖJE SU *FUNNELIFORMIS MOSSEAE* IR *PIRIFORMOSPORA INDICA*, BIOCHEMINIS ATSAKAS Į KADMIO TOKSINĮ POVEIKĮ

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Santrauka

Buvo tiriamas šaknų endofitinių grybų *Piriformospora indica* ir *Funneliformis mosseae* poveikis kviečių *Triticum aestivum* v. ‘Sardari39’ lapų ir šaknų biocheminėms reakcijoms į skirtingas kadmio toksines koncentracijas. Eksperimento metu buvo atliktos trys grybienos inokuliacijos: *F. mosseae*, *P. indica*, *F. mosseae* + *P. indica*, kontroliniai augalai – be inokuliacijos, visais atvejais buvo naudojamos keturios Cd koncentracijos (0; 0,3; 0,6 ir 0,9 mM). Augalai buvo auginami 45 paras. Neinokuliuotų augalų lapai ir šaknys į padidėjusias Cd koncentracijas reagavo padidėjusiu H_2O_2, malondialdehido (MDA) ir prolino kiekiu, tuo tarpu šaknų glutationo peroksidazės (GPX), lapų ir šaknų superoksido dismutazės (SOD) aktyvumas padidėjo žemų Cd koncentracijų ir sumažėjo didelių Cd koncentracijų atveju. Kviečių, inokuliuotų tiek atskirai su *P. indica* ir F. *mosseae,* tiek kartu, lapuose ir šaknyse sumažėjo H_2O_2 ir MDA kiekiai ir padidėjo prolino akumuliacija. Be to, *P. indica* padidino kviečių lapų ir šaknų GPX ir SOD aktyvumą. Trumpai tariant, *P. indica* ir *F. mosseae* simbiozė su kviečiais gali būti naudinga dirvožemyje, turinčiame dideles Cd koncentracijas. Tačiau Cd streso mažinimui *P. indica* poveikis buvo ryškesnis, nei *F. mosseae*.