



Impact of *Peltigera praetextata* on zucchini grown in weathered *p,p'*-DDE-contaminated soil and its responses

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Abstract

Terricolous lichens, located in soils, can change the bioavailability of polluted soil biologically, chemically and physically with their effective enzymatic content. Their effects are unknown especially on weathered *p,p'*-DDE-contaminated soil. However, we obtain new information by increasing our studies on this subject. In the present study, the impact of *Peltigera praetextata* (a terricolous lichen) on zucchini (*Cucurbita pepo* spp. *pepo*) grown in weathered *p,p'*-DDE-contaminated soil was shown via its physiological responses. At the same time, the physiological responses of *Peltigera praetextata* were also given. The results show that the growth and development of the zucchini grown in weathered *p,p'*-DDE-contaminated soil was positively affected by *Peltigera praetextata*. *Peltigera praetextata* was increased the bio-usefulness of weathered *p,p'*-DDE-contaminated soil with its effective enzyme contents. Moreover, *Peltigera praetextata* was able to cope with *p,p'*-DDE contamination with the strong antioxidative defence system.

Keywords Terricolous lichens · *Peltigera praetextata* · Persistent organic pollutants · Weathered *p,p'*-DDE-contamination · Antioxidative defence system · Physiological responses

Introduction

Lichens are known to be an ecological indicator in the identification of environmental pollutions (Conti and Cecchetti 2001; Canha et al. 2014). They are used for the biomonitoring of air pollutants by atmospheric deposition on their thallus surface (Stevens et al. 2012). Moreover, they can take and accumulate heavy metals through not only atmospheric but also the surface they use as a substrate (Pirintsos et al.

2006; Backor and Loppi 2009). Because lichens consist of algae and fungus, lichen hyphae can penetrate within rocks used as a substrate and can also accumulate metals in this way. In fact, there are various studies in the literature which state that lichen-forming hyphae can diffuse and mobilize minerals or metals at wider pH and redox range, more efficiently and quickly than bacteria (Gadd 2007).

Lichens have many interactions with not only the surrounding pollutants but also with living groups. Such allelopathic interaction of lichens were with vascular plants (Favero-Longo and Piervittori 2010; Latkowska et al. 2015), moss species (Lawrey 1977; Glime 2007) and soil microorganisms (Akpınar et al. 2009; Leiva et al. 2016). Secondary metabolite and enzymatic contents of lichens are an important factor in this interaction. For example, usnic acid compounds obtained from *Cladonia* species suppress germination of some of the moss's spore; some *Peltigera* species with their enzymatic activities alter soil mineralization and microorganism content (Akpınar et al. 2009). Furthermore, lecanoric acid and its orsellinate derivatives obtained from *Parmotrema tinctorum* are well-known to have an allelopathic activity on the lettuce (*Lactuca sativa*) and onion (*Allium cepa*) plants (Peres et al. 2009). However, extracted lichen compounds have been used in these studies.

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But the situation in the ecological environment is different and lichens have beneficial effects on lichen and plant interactions (Chen et al. 2000; Escudero et al. 2007; Piervittori et al. 2009). Lichens located in the soil (terricolous lichens) are linked with it (Lawrey 2009). Terricolous lichens with their effective secondary metabolites and enzyme contents could contribute to the viability of plants by increasing the biochemical and biophysical usefulness of soil. For example, an increase in the moisture of the soil containing the plants and in the mobilization of plant nutrients, accumulation of organic layer are contributions of terricolous lichens to plants (Asplund and Wardle 2017). These contributions of lichens have the potential to decrease the negative factors of polluted soils on the growth and development of plants (Valencia-Islas et al. 2007).

DDT (2,2-bis (p-chlorophenyl)-1,1,1-trichloroethane) as well as its derivatives, known as persistent organic pollutants (POPs), are among serious soil contaminants today too although they are banned chemicals. Because of its long half-life, many soils today contain varying levels of DDT and its degradation products. They are exactly binding to the soil organic matter due to their hydrophobic structure ($\log K_{ow} > 3,5$), and immediately decrease soil bioavailability (Alexander 2000). Most in-situ treatment technologies are ineffective against DDTs pollution. Because these approaches require high costs and cause irrecoverable changes in soil properties and deterioration in the soil microflora (Costa et al. 2012; Kumar et al. 2014). Moreover, such chemical methods to be applied can also lead to secondary pollution problems. For this reason, biological solutions are being investigated in the elimination of DDTs pollution. In both laboratory and real-field studies, there are studies conducted to clean the soil from this pollutant via accumulating by various plants, especially *Cucurbita pepo* spp. *pepo*. come forward in these studies. (White 2000, 2001, 2002; White et al. 2005, 2006a; Mattina et al. 2006; Gent et al. 2007). However, it is relevant to develop an approach for the minimization or disposal of these contaminants directly with biological processes rather than spreading into environmental media and accumulating in crops (Balawejder et al. 2014). Moreover, the biological, chemical, and physical responses of an organism that is exposed to any contaminant existent in soil are important in the elimination of contaminants through a biological process (Ehlers and Luthy 2003; Hodson et al. 2011). A reduction in the toxic effect of various contaminants by changing the bioavailability of soils was stated with the addition of compost (Bolan et al. 2003), cyclonic ashes (Ruttens et al. 2006), lime and red mud (Gray et al. 2006) and biochar (Bian et al. 2013; Kim et al. 2015; Hussain et al. 2017). However, effects of terricolous lichens that will be useful to the soil bioavailability were not known on weathered *p,p'*-DDE-contaminated soil. In a previous study, it was revealed that our experiment with *Peltigera*

canina improved the growth and development of zucchini by increasing the bioavailability of soil contaminated by DDTs (Akpınar et al. 2020). Therefore, our study focused on the effect of *Peltigera praetextata*, a terricolous lichen species, on weathered *p,p'*-DDE-contaminated soil via monitoring of zucchini. Since the separation of the lichen secondary metabolite from lichens is not earning efficient results (Kytöviita and Stark 2009), lichen thallus was taken care of conserving its present structure, even in powder form.

Materials and methods

Experimental preparation and design

Soil samples were taken from a special area in Karasu-Sakarya/Turkey where DDTs profiles were researched by Isleyen et al. (2013). Total DDTs (*p,p'*-DDE, *p,p'*-DDD and *p,p'*-DDT) in this area has been reported to range from 504 ng /g to 3557 ng/g dry soil. Samples were taken away from an average 30 cm depth and passed through 2-mm sieves. In this study, an average of 400 ng/g dry soil was arranged as the amount of *p,p'*-DDE with regard to values obtained from historical DDT usage (White et al. 2003, 2007; White 2009).

Peltigera praetextata lichen samples were collected from Bursa province, Turkey. This position is located in the south of Bursa, 39° 38' 36" N, 29° 01' 08" E, 484 m in altitude (Fig. 1). Ten to twelve were the number of lichen thalli used in each treatment. Some of the lichen samples were powdered and others left intact after being isolated from their substrates and cleaned. Lichen samples ready for treatments were immediately treated with soil contaminated by *p,p'*-DDE. While powdered lichen samples were mixed with soil contaminated by *p,p'*-DDE, intact lichen samples were put on soil contaminated with DDTs.

In the present work, zucchini species (*Cucurbita pepo* spp. *pepo* (cv. Raven)) that accumulates the most DDTs in its structure were used (White 2000, 2001, 2002, Wang et al. 2004, White et al. 2005, 2006a, 2006b, Mattina et al. 2006, Gent et al. 2007). Seeds were germinated and grown in weathered soil contaminated with *p,p'*-DDE, simultaneously mixed with powdered and intact *Peltigera praetextata* thallus. On the other way, weathered soil contaminated with *p,p'*-DDE without *P. praetextata* for control group were used.

Zucchini seedlings (Fig. 2), weathered *p,p'*-DDE-contaminated soil and *Peltigera praetextata* lichens (powdered and intact thallus) were used in the experimental setup. Six plants per treatment with three replications were applied. Four groups were established in our study and expressed below and are schematically presented in Fig. 3:

Fig. 1 Photograph of the *Peltigera praetextata* Lichen Species Collected in March 2018 around province of Bursa-Turkey. *P. praetextata* is a terricolous lichen and abundantly in this location



Fig. 2 Photograph of eight-weeks-old zucchini (*Cucurbita pepo* spp. *pepo* (cv. Raven)) seedlings grown to use in experimental design. Zucchini seeds were purchased from Johnny's Selected Seeds (Albion, ME)



1. group: Control – Zucchini grown in weathered soil contaminated with p,p' -DDE (without *P. praetextata*)
2. group: Zucchini grown in weathered soil contaminated with p,p' -DDE and mixed with intact *P. praetextata* thallus
3. group: Zucchini grown in weathered soil contaminated with p,p' -DDE and mixed with powdered *P. praetextata*
4. group: intact *P. praetextata* thallus in weathered soil contaminated with p,p' -DDE (without zucchini)

Growth parameters and chlorophyll analysis in zucchini and intact lichen thallus

Firstly, fresh plant weights (g) that were recorded belonged to parts of zucchini and intact lichen thallus. Plant heights and stem diameter (mm) of zucchini were measured. Chlorophyll content (SPAD value) of zucchini was determined with a portable SPAD chlorophyll meter in fresh plant material (SPAD-502; Konica Minolta Sensing, Inc., Japan) Samples were harvested and separated into plant portions such as leaves and roots. Fresh plant material from the leaves

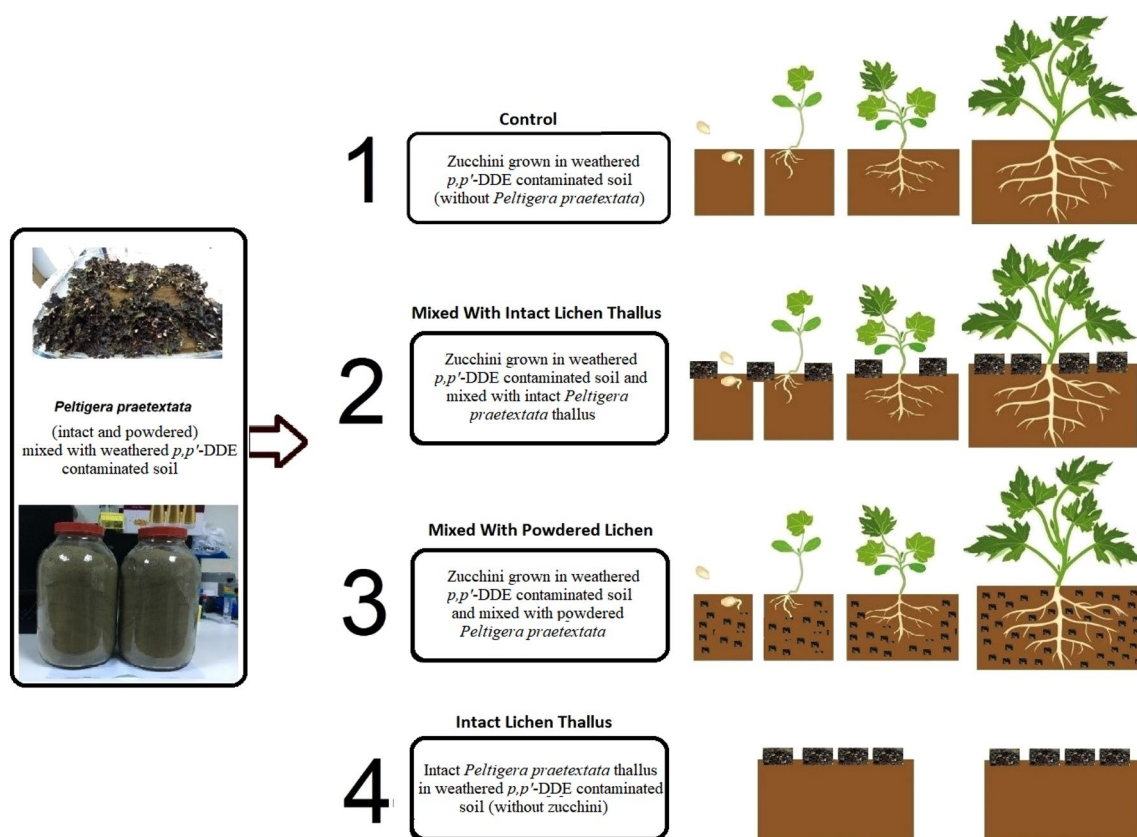


Fig. 3 Experimental design of the study. First of all, soil samples taken from Karasu-Sakarya/Turkey were mixed with *Peltigera praetextata*. *Peltigera praetextata* was used in two different ways as powdered and intact thallus. Then, zucchini seeds were sown to the first three groups as shown in the above figure. The last group was containing only *P. praetextata*. Thus, four groups were established. When

zucchini seedlings were twelve-weeks-old, zucchini was harvested and sampling was done. Also, intact *P. praetextata* samples were taken too. Then, all samples were washed thoroughly with de-ionized water. The zucchini plants were separated into roots, stems and leaves. After physiological and biological determinations using fresh plant material, the remaining samples were stored at $-80\text{ }^{\circ}\text{C}$

of zucchini was taken for determining the percent of ion leakage. And they were frozen in liquid nitrogen and stored at $-80\text{ }^{\circ}\text{C}$.

In the determination of the chlorophyll content for intact lichen thallus (*Peltigera praetextata*), the method described by Barnes et al. (1992) was used. Lichen materials were homogenized with pure DMSO (dimethyl sulphoxide 99%, Merck). Then, it was incubated in the dark ($65\text{ }^{\circ}\text{C}$, 40 min) and filtered with a filter paper (Whatman). The sample absorbance values were obtained at 665 and 648 nm for calculating chlorophyll a (Chl a), chlorophyll b (Chl b), total chlorophyll (a + b), and chlorophyll degradation rate (a/b). Calculations were done according to the following equations:

$$\text{Chlorophyll a} = 14.85A_{665} - 5.14A_{648}$$

$$\text{Chlorophyll b} = 25.48A_{648} - 7.36A_{665}$$

$$\text{Chlorophyll a + b} = \text{Total Chlorophyll} = 7.49A_{665} + 20.34A_{648}$$

Remaining lichen materials were kept at $-80\text{ }^{\circ}\text{C}$ for future analyses.

Ion leakage in leaves of zucchini

The percentage of ion leakage in leaves of zucchini was measured according to a modified method by Gulen and Eris (2003) which was originally published by Arora et al. (1998). Leaf discs at 2 cm diameter were obtained and placed into tubes containing deionized water (20 mL). They were vacuum filtrated to let electrolyte diffusion homogeneously, and then incubated at room temperature for 4 h. The electrical conductivity of each sample (EC_1) was determined using a conductivity meter (WTW Cond 315i, Weilheim, Germany). The samples were then autoclaved and ranged to room temperature, for which electrical conductivity was measured as EC_2 . Percentage of leakage (%) was determined according to the equation; Ion leakage = $(EC_1/EC_2) \times 100$.

Malondialdehyde (MDA) content assay

MDA is a breakdown product of lipid peroxidation. MDA content was analyzed by a slightly modified method originally published by Heath and Packer (1968). Briefly, 0.1 g sample was homogenized in 0.1% (w/v) trichloroacetic acid (TCA) and then centrifuged (15,000 g, 10 min). Supernatant (0.5 mL) was mixed with 20% TCA and 0.5% TBA. After incubation (95 °C, 30 min), the samples were ranged to room temperature and then centrifuged (15,000 g, 4 °C, 5 min). The absorbance values were obtained at 532 nm and 600 nm (Novaspec II, LKB Biochrom). MDA content was communicated as nmol/g fresh weight.

Non-enzymatic antioxidants

Total glutathione (GSH) analysis was performed by following the methodology of Elman (1959). Briefly, 0.2 g frozen sample was homogenized with 5% (w/v) trichloroacetic acid and centrifuged (15,000 g, 15 min). For lichen, samples were powdered and the extracts were prepared by soaking 10 g of material with 250 mL methanol at room temperature. After 3 days, the extracts were filtered using filter paper (Whatman No.1). Supernatants or extracts for lichen were added to the solution containing phosphate-buffered saline (pH 7.7) plus 5,5'-dithiobis (2-nitrobenzoic acid) (DTNB). After incubation (5 min, 30 °C), absorbance value was obtained at 412 nm. Total content of GSH was calculated via a standard curve (Elman 1959).

AsA contents (reduced AsA [AsA] and total ascorbates [AsA + DAsA]) were measured as described by Cakmak and Marschner (1992) with some modifications. 1 g sample was extracted with 5% meta-phosphoric acid (10 mL) and centrifuged (22,000 g, 15 min). For lichens, the same extraction method outlined above was used. 0.2 mL supernatant or lichen extract for total ascorbates was added to 150 mm phosphate buffer (pH 7.4) containing 5 mm EDTA, 10 mM DTT. Samples were incubated at room temperature (10 min), 0.5 N-ethylmaleimide (0.1 mL) was included to expel overabundance DTT. 0.2 mL supernatant or lichen extract for AsA was mixed to the solution containing 0.5 mL 150 mm phosphate buffer (pH 7.4), and 0.2 mL water. Following this, 0.4 mL 44% ortho-phosphoric acid, 0.4 mL 10% TCA, 0.4 mL 4% a'-dipyridyl (with 70% ethyl alcohol), and 0.2 mL 3% FeCl₃ were added, respectively to both reaction mixtures. Then they were incubated (40 °C, 40 min) and measured at 525 nm.

Antioxidative enzyme activity

In this study, superoxide dismutase (SOD), catalase (CAT), ascorbate peroxidase (APX) activities from antioxidative enzymes were determined. Firstly, plant extraction was

performed. 1 g sample was homogenized with solution containing 1 mM EDTA, 2% (w/v) polyvinylpyrrolidone (PVP) and 50 mM Na-phosphate buffer (pH 7.8) in an ice bath. Samples were centrifuged (14,000 g, 40 min, 4 °C) and the supernatant was utilized for the determination of SOD and CAT activities. For the APX activity, 2 mM ascorbate was added to the extraction buffer.

In the determination of the SOD activity, we used the methodology of Beauchamp and Fridovich (1971). The supernatant (0.5 mL) was mixed with buffer containing sodium phosphate (20 mM, pH 7.5), riboflavin (5 µM), methionine (10 mM), *p*-nitroblue tetrazolium (NBT, 0.1 mM) and EDTA (0.1 mM). Samples were then placed under fluorescent light (300 µmol m⁻² s⁻¹, 15 min) and measured spectrophotometrically (Novaspec II, LKB Biochrom) at 560 nm. The kit for SOD assay (Sigma-Aldrich, USA) was used for standards. After the calculation of % inhibition, the enzyme activity was determined according to the linear equation and expressed as U/mg protein.

In the CAT activity assay, the method of Lester et al. (2004) was used with a few adjustments. The supernatant (0.1 mL) was added to 15 mM H₂O₂ and buffered sodium phosphate (20 mM, pH 6.8). Absorbance was obtained at 240 nm for 3 min (Beckman DU-520 UV/Vis Spectrophotometer). The CAT activity was communicated as U/mg protein.

In the APX activity, it was measured spectrophotometrically according to the method described by Lester et al. (2004). 1 mL supernatant was added to 1 mM H₂O₂ (33% w/v H₂O₂), 0.25 mM ascorbate and 50 mM potassium phosphate (pH 6.6). Absorbance was obtained at 290 nm for 3 min (Beckman DU-520 UV/Vis Spectrophotometer).

Statistical analysis

Statistical analyses were performed using SPSS Statistics (22.0 version, IBM Corp., Chicago, IL) and covered three replicates of six plants in each treatment. Data were analyzed by One-Way ANOVA followed by post-hoc Tukey's test and significance was set at $p < 0.05$.

Results

Changes in growth parameters and chlorophyll content

Total fresh plant weights (g) of zucchini (*Cucurbita pepo* spp. *pepo*) grown in weathered *p,p'*-DDE-contaminated soil increased notably after mixing with *Peltigera praetextata* (powdered and intact thallus) ($p < 0.05$, Fig. 4). This increase was due to changes of fresh weight in the leaf and stem of the zucchini, no difference was obtained in root weights

Fig. 4 Total fresh plant weights (g) and stem, leaf, root weights of zucchini grown in weathered *p,p'*-DDE contaminated soil (Control) and mixed with *Peltigera praetextata* (powdered and intact thallus). Data are means (\pm SD) of six replicates. Letters show statistical comparisons among the groups of the plant sections, based on One-Way ANOVA (SPSS 22.0) and followed by the LSD, at $p < 0.05$

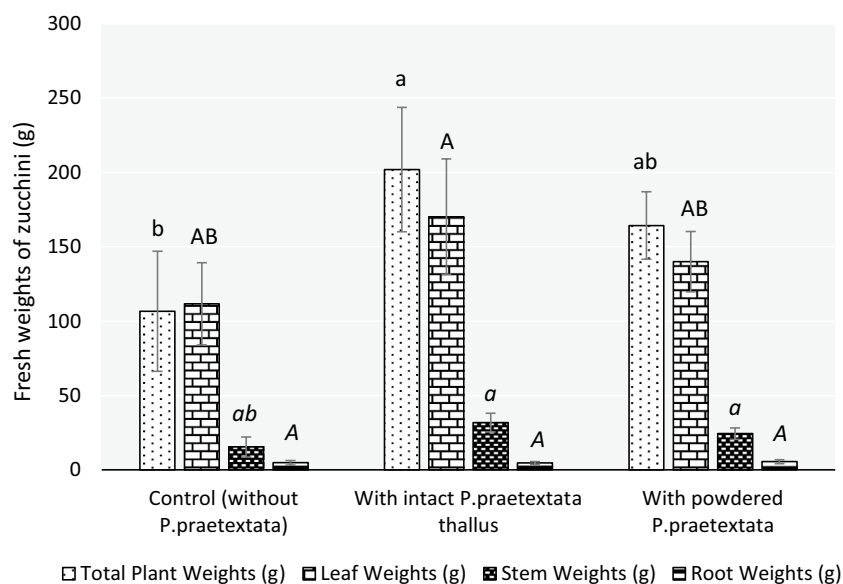


Table 1 Hypocotyl, epicotyl and total plant heights (mm), and stem diameters of zucchini grown in weathered *p,p'*-DDE contaminated soil (Control) and mixed with *Peltigera praetextata* (powdered and intact thallus)

Zucchini (<i>Cucurbita pepo</i> spp. <i>pepo</i>) grown in soils contaminated by <i>p,p'</i> -DDE	Total plant heights mm	Hypocotyl	Epicotyl	Stem Diameter
Control—(without <i>Peltigera praetextata</i>)	52.88 \pm 15.34 ^{ab}	42.25 \pm 9.44 ^a	15.00 \pm 9.68 ^b	9.22 \pm 2.16
With intact <i>Peltigera praetextata</i> thallus	84.67 \pm 19.49 ^a	44.00 \pm 13.19 ^a	44.50 \pm 10.37 ^a	9.01 \pm 0.96
With powdered <i>Peltigera praetextata</i>	59.00 \pm 18.28 ^{ab}	33.33 \pm 10.33 ^{ab}	21.67 \pm 12.11 ^b	8.98 \pm 1.21

Data are means (\pm SD) of six replicates. Different lowercase letters within a column define significant differences based on One-Way ANOVA (SPSS 22.0) and followed by the LSD, at $p < 0.05$

Table 2 Chlorophyll content (SPAD value) in leaves of zucchini grown in weathered *p,p'*-DDE contaminated soil (Control) and mixed with *Peltigera praetextata* (powdered and intact thallus)

Zucchini (<i>Cucurbita pepo</i> spp. <i>pepo</i>) grown in soils contaminated by <i>p,p'</i> -DDE	Chlorophyll Content (SPAD value)
Control—(without <i>Peltigera praetextata</i>)	32.06 \pm 3.16 ^a
With intact <i>Peltigera praetextata</i> thallus	34.80 \pm 2.69 ^a
With powdered <i>Peltigera praetextata</i>	34.53 \pm 1.69 ^a

Data are means (\pm SD) of six replicates. Different lowercase letters within a column define significant differences based on One-Way ANOVA (SPSS 22.0) and followed by the LSD, at $p < 0.05$

(Fig. 4). The highest value in fresh weight for both leaf and stem of zucchini was obtained in the intact *P. praetextata* thallus treatment with approximately two-fold increase than control ($p < 0.05$). Table 1 shows the hypocotyl, epicotyl, total plant heights and stem diameters of zucchini. While *Peltigera praetextata* treatments (powdered or intact thallus) did not change total plant heights, hypocotyl and stem diameters of zucchini, the intact lichen thallus treatment increased epicotyl heights of zucchini ($p < 0.05$). Moreover, *P. praetextata* (powdered and intact thallus) treatments did not show significant changes in chlorophyll content (SPAD value) in leaves of zucchini (Table 2). In Table 3, fresh

Table 3 Fresh weights (g) and chlorophyll degradation rate [(a/b), mg/g FW] of intact *Peltigera praetextata* thallus in weathered *p,p'*-DDE contaminated soil, together with zucchini and without it

Intact <i>Peltigera praetextata</i> thallus in soils contaminated by <i>p,p'</i> -DDE	Fresh weights (g)	Chlorophyll degradation rate (a/b) (mg/g FW)
Control—(without zucchini)	4.54 \pm 0.78 ^b	4.27 \pm 0.075
Intact lichen thallus—(with zucchini)	7.22 \pm 2.36 ^a	3.36 \pm 0.069

Data are means (\pm SD) of six replicates. Different lowercase letters within a column define significant differences based on One-Way ANOVA (SPSS 22.0) and followed by the LSD, at $p < 0.05$

weight (g) of intact *Peltigera praetextata* thallus in weathered *p,p'*-DDE-contaminated soil is given as when together with zucchini and without it. According to this, the fresh weight of the lichen increased in the presence of zucchini ($p < 0.05$). However, chlorophyll content (Chl a, b and total chlorophyll) of intact *Peltigera praetextata* thallus did not change statistically (Fig. 5). But chlorophyll degradation rate (a/b) of intact *P. praetextata* thallus in the presence of zucchini decreased according to control ($p < 0.05$, Table 3).

Oxidative damage to lipids

MDA content in stems and roots of zucchini grown in weathered *p,p'*-DDE-contaminated soil remained unchanged when

mixed with *Peltigera praetextata* (intact or powdered thallus) (Fig. 6). But MDA content in leaves of zucchini increased significantly in both *P. praetextata* treatments ($p < 0.05$). This increase was similar in powdered or intact thallus treatments. When we looked at ion leakage determined in leaves of zucchini, the highest value was obtained in intact thallus treatment (Table 4, $p < 0.05$). MDA content of intact *P. praetextata* thallus decreased when in weathered *p,p'*-DDE-contaminated soil together with zucchini ($p < 0.05$, Table 5).

Responses of non-enzymatic antioxidants

In *Peltigera praetextata* treatments, only intact lichen thallus increased the total GSH content in the leaves of zucchini

Fig. 5 Chlorophyll content (mg/g FW) of intact *Peltigera praetextata* thallus in weathered *p,p'*-DDE contaminated soil, together with zucchini and without it. Data are means (\pm SD) of six replicates. Letters show statistical comparisons among the groups of the plant sections, based on One-Way ANOVA (SPSS 22.0) and followed by the LSD, at $p < 0.05$

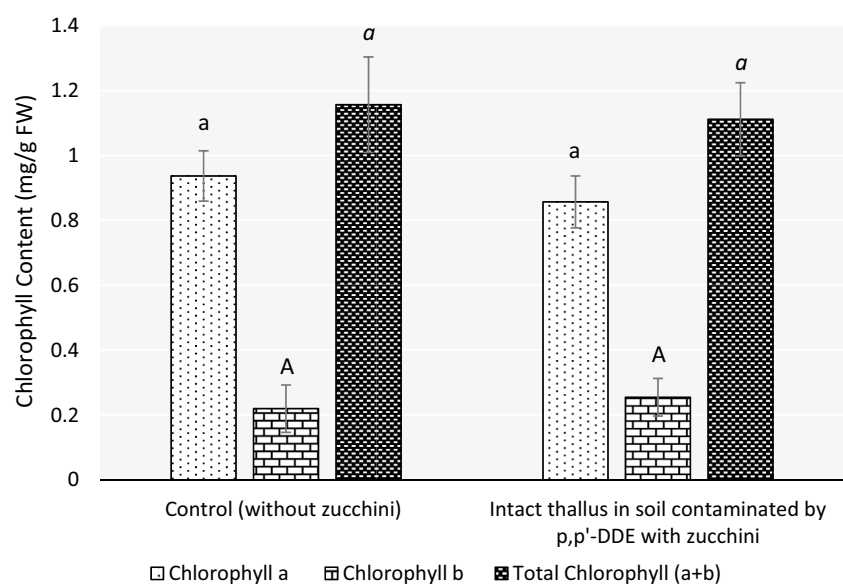


Fig. 6 MDA contents (nmol/g FW) in leaves, stems and roots of zucchini grown in weathered *p,p'*-DDE contaminated soil (Control) and mixed with *Peltigera praetextata* (powdered and intact thallus). Data are means (\pm SD) of six replicates. Letters show statistical comparisons among the groups of the plant sections, based on One-Way ANOVA (SPSS 22.0) and followed by the LSD, at $p < 0.05$.

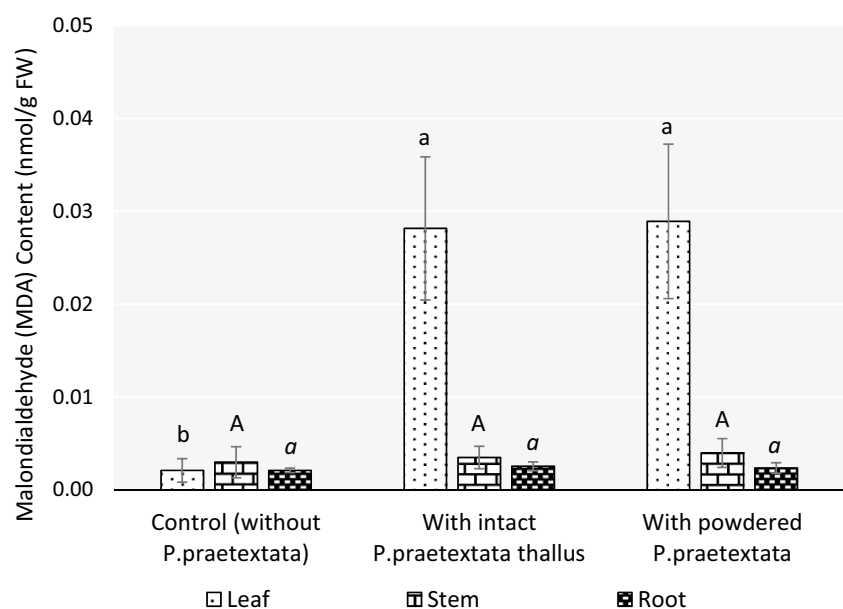


Table 4 Ion leakage (%) in leaves of zucchini grown in weathered *p,p'*-DDE contaminated soil (Control) and mixed with *Peltigera praetextata* (powdered and intact thallus)

Zucchini (<i>Cucurbita pepo</i> spp. <i>pepo</i>) grown in soils contaminated by <i>p,p'</i> -DDE	Ion Leakage (%)
Control—(without <i>Peltigera praetextata</i>)	20.09 ± 2.31 ^b
With intact <i>Peltigera praetextata</i> thallus	27.12 ± 3.52 ^a
With powdered <i>Peltigera praetextata</i>	23.57 ± 3.61 ^{ab}

Data are means (±SD) of six replicates. Different lowercase letters within a column define significant differences based on One-Way ANOVA (SPSS 22.0) and followed by the LSD, at $p < 0.05$

(Fig. 7, $p < 0.05$). Total GSH content decreased in the stem of zucchini grown in weathered *p,p'*-DDE-contaminated soil mixed with intact *P. praetextata* thallus ($p < 0.05$). Lichen treatments (powdered and intact lichen thallus) did not cause a difference in the total GSH content of the root (Fig. 7). Furthermore, the total GSH content of intact *Peltigera praetextata* thallus in soils contaminated by *p,p'*-DDE did not change by the presence of the zucchini (Table 5). Similarly, no change was also observed in total AsA and AsA

content of intact *Peltigera praetextata* thallus in weathered *p,p'*-DDE-contaminated soil due to the presence of zucchini (Fig. 8). Total AsA and AsA content in leaves, stems and roots of zucchini are also given in Fig. 9. According to this, total AsA content in roots of zucchini increased with powdered lichen treatment while it decreased with intact lichen thallus ($p < 0.05$). It was determined that no changes were obtained in the leaves and stems of zucchini (Fig. 9).

Responses of antioxidative enzymes

SOD activity of zucchini in control and treatment groups is given in Fig. 10. SOD activity was intensive in the stem of zucchini belonging to control group, but it decreased with the addition of *Peltigera praetextata* (powdered and intact thallus) ($p < 0.05$). Otherwise, the highest values in CAT activity were determined in the leaves of zucchini belonging to control group (Fig. 11). However, it was found that CAT activity decreased by 66% with the addition of *Peltigera praetextata* (powdered and intact thallus) ($p < 0.05$, Fig. 11). APX activity of zucchini in control and treatment groups is given in Fig. 12. Similarly, the highest APX

Table 5 MDA content (nmol/g FW), Glutathione content ($\text{U}/\mu\text{g cm}^{-2}$) of intact *Peltigera praetextata* thallus in weathered *p,p'*-DDE contaminated soil, together with zucchini and without it

Intact <i>Peltigera praetextata</i> thallus in soils contaminated by <i>p,p'</i> -DDE	Malondialdehyde (MDA) Content nmol/g FW	Glutathione content $\text{U}/\mu\text{g cm}^{-2}$
Control—(without zucchini)	0.0054 ± 0.0013 ^a	42.14 ± 3.47 ^a
Intact lichen thallus—(with zucchini)	0.0030 ± 0.0008 ^b	41.38 ± 2.10 ^a

Data are means (±SD) of six replicates. Different lowercase letters within a column define significant differences based on One-Way ANOVA (SPSS 22.0) and followed by the LSD, at $p < 0.05$

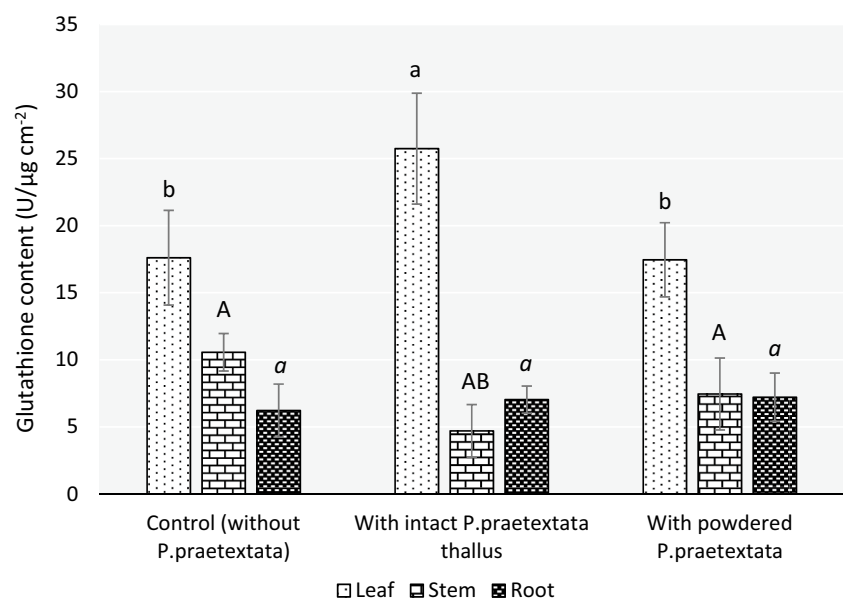
Fig. 7 Glutathione content ($\text{U}/\mu\text{g cm}^{-2}$) in leaves, stems and roots of zucchini grown in weathered *p,p'*-DDE contaminated soil (Control) and mixed with *Peltigera praetextata* (powdered and intact thallus). Data are means (±SD) of six replicates. Letters show statistical comparisons among the groups of the plant sections, based on One-Way ANOVA (SPSS 22.0) and followed by the LSD, at $p < 0.05$.

Fig. 8 Total AsA (AsA + DAsA) and AsA content ($\mu\text{g/g}$ FW) of intact *Peltigera praetextata* thallus in weathered *p,p'*-DDE contaminated soil, together with zucchini and without it. Data are means (\pm SD) of six replicates. Letters show statistical comparisons among the groups of the plant sections, based on One-Way ANOVA (SPSS 22.0) and followed by the LSD, at $p < 0.05$.

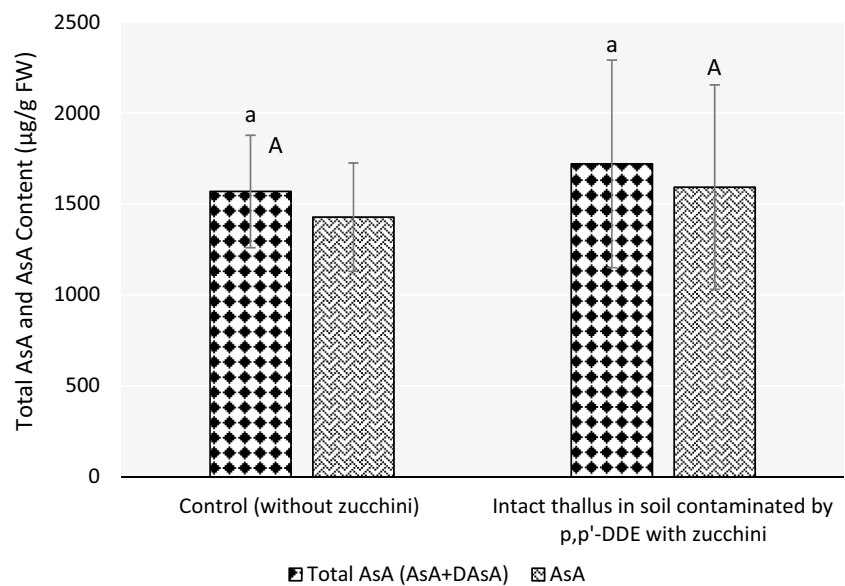
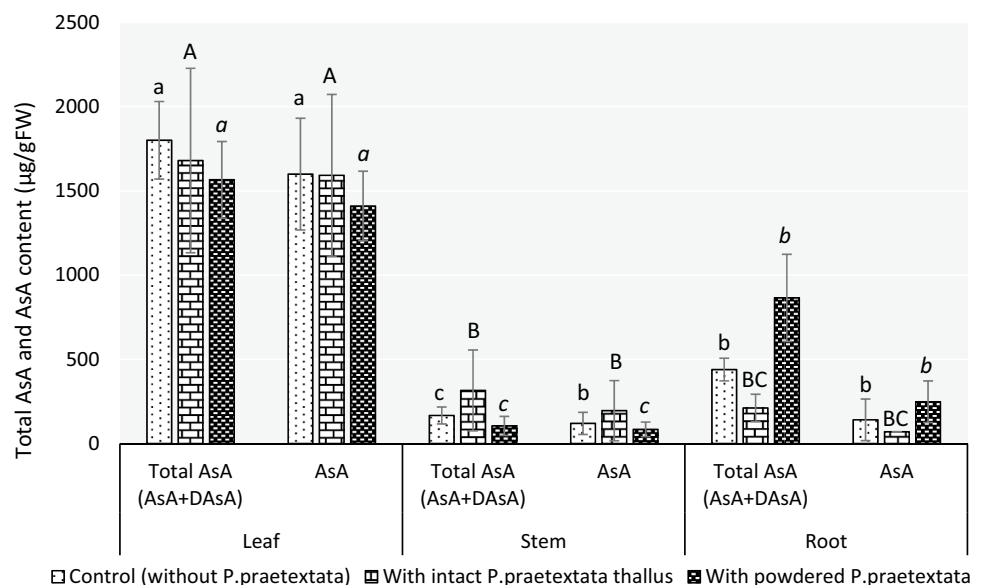


Fig. 9 Total AsA (AsA + DAsA) and AsA content ($\mu\text{g/g}$ FW) in leaves, stems and roots of zucchini grown in weathered *p,p'*-DDE contaminated soil (Control) and mixed with *Peltigera praetextata* (powdered and intact thallus). Data are means (\pm SD) of six replicates. Letters show statistical comparisons among the groups of the plant sections, based on One-Way ANOVA (SPSS 22.0) and followed by the LSD, at $p < 0.05$.



activity was determined in leaves of zucchini belonging to control group ($p < 0.05$). Accordingly, APX activity in leaves of zucchini decreased with the addition of *Peltigera praetextata* (powdered and intact thallus) ($p < 0.05$). While the decrease in APX activity with the addition of intact lichen thallus treatment was not statistically significant, powdered *P. praetextata* treatment was noteworthy at the $p < 0.05$ level (Fig. 12). Antioxidative enzyme activities of intact *Peltigera praetextata* thallus in weathered *p,p'*-DDE-contaminated soil are given in Table 6. SOD, CAT and APX activity of intact *Peltigera praetextata* thallus in weathered *p,p'*-DDE-contaminated soil increased by the presence of the zucchini ($p < 0.05$). While the rate of increase in SOD and APX activity was 70% according to control, there was a twofold increase in CAT activity ($p < 0.05$).

Discussion

In the bioavailability of contaminated soil, research about the physiological changes of the plants that are grown in the contaminated soil is as important as those investigating the soil properties (Ehlers and Luthy 2003; Hodson et al. 2011). Also, knowledge obtained about biochemical, physiological and metabolic processes of plant species and other organisms exposed to various pollutants are very important in terms of developing phytoremediation techniques (Yang et al. 2005; Bhargava et al. 2012). Therefore, the impact of *P. praetextata* (powdered and intact thallus) on zucchini plants grown in weathered soil contaminated with *p,p'*-DDE was investigated in this study.

Fig. 10 SOD Activity (U/mg protein) in leaves, stems and roots of zucchini grown in weathered *p,p'*-DDE contaminated soil (Control) and mixed with *Peltigera praetextata* (powdered and intact thallus). Data are means (\pm SD) of six replicates. Letters show statistical comparisons among the groups of the plant sections, based on One-Way ANOVA (SPSS 22.0) and followed by the LSD, at $p < 0.05$.

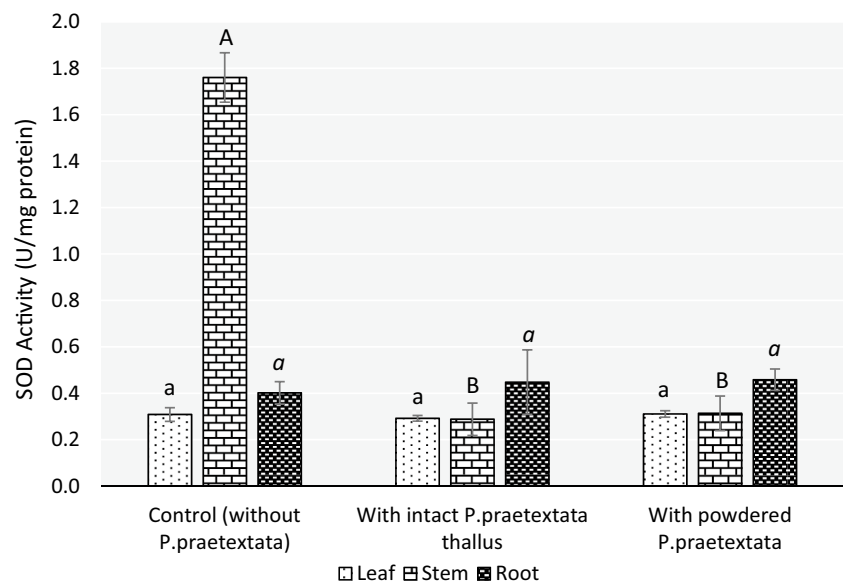
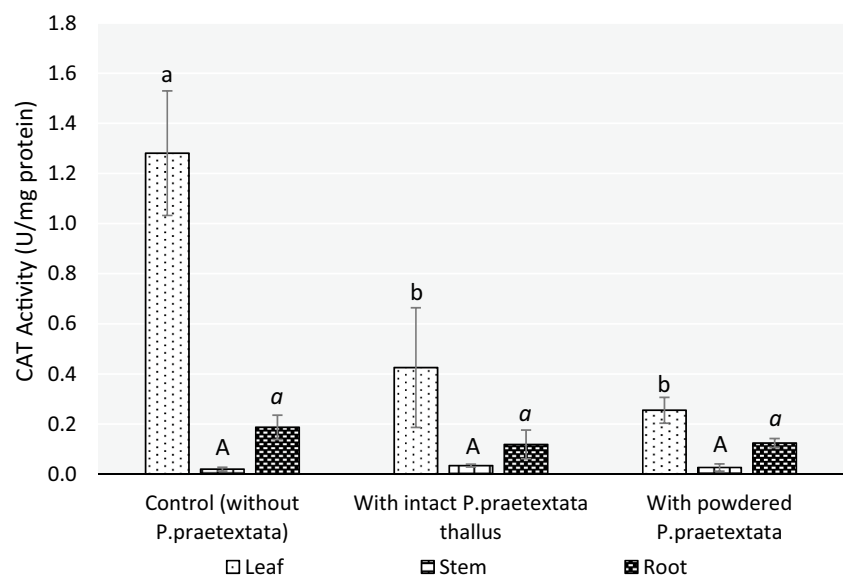


Fig. 11 CAT Activity (U/mg protein) in leaves, stems and roots of zucchini grown in weathered *p,p'*-DDE contaminated soil (Control) and mixed with *Peltigera praetextata* (powdered and intact thallus). Data are means (\pm SD) of six replicates. Letters show statistical comparisons among the groups of the plant sections, based on One-Way ANOVA (SPSS 22.0) and followed by the LSD, at $p < 0.05$.



Physiological responses of *P. praetextata* (intact lichen thallus) and zucchini were obtained.

According to this, while the growth of the above-ground organs of the zucchini has increased in the presence of intact lichen thallus, there has been no change in its roots. The fresh weight of the stem and leaves, and epicotyl length of zucchini increased when it was treated with intact *Peltigera praetextata* thallus (Table 1, Fig. 4). In the first stage of seedling development, the epicotyl produces the stem, branches, buds, leaves and other above-ground organs. Changes in epicotyl lengths occur in the control of light, soil moisture and temperature, and some growth hormones (Potter et al. 1999; Haga and Lino 2006; Tiansawat and Dalling 2013; Hao et al. 2014). In our results, the increase of fresh weight in the above-ground organs of zucchini was due to the protection

of the temperature and moisture of the soil. Since zucchini plants were covered by intact *P. praetextata* thallus, maintenance of water availability and temperature of soil also led to an increase in epicotyl length. It is also stated in various studies that the epicotyl length increases depending on the soil temperature and humidity (Oliveira and Macedo, 2015). At the same time, this increase leads us to think that the soil nutrients required for plant growth can be made more available from *P. praetextata*, because DDTs, which bind tightly to the organic matter in the soil, prevent from being in a usable form of nutrients by reducing the bioavailability of the soil (Alexander 2000). In our study, an increase in the growth of the above-ground organs of zucchini has resulted from the change that occurred of the lichen enzymatic contents in weathered *p,p'*-DDE-contaminated soil. Moreover,

Fig. 12 APX Activity (U/mg protein) in leaves, stems and roots of zucchini grown in weathered *p,p'*-DDE contaminated soil (Control) and mixed with *Peltigera praetextata* (powdered and intact thallus). Data are means (\pm SD) of six replicates. Letters show statistical comparisons among the groups of the plant sections, based on One-Way ANOVA (SPSS 22.0) and followed by the LSD, at $p < 0.05$.

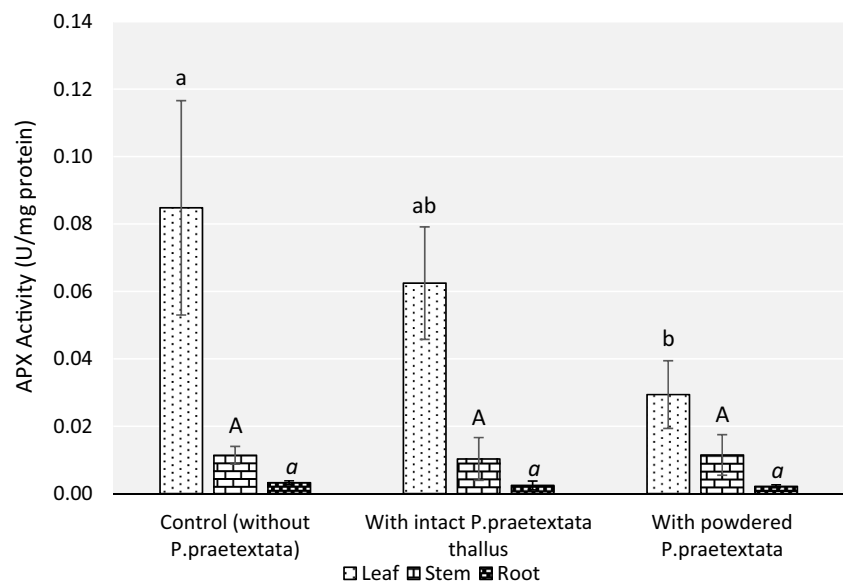


Table 6 SOD, CAT and APX Activity (U/mg protein) of intact *Peltigera praetextata* thallus in weathered *p,p'*-DDE contaminated soil, together with zucchini and without it

Intact <i>Peltigera praetextata</i> thallus in soils contaminated by <i>p,p'</i> -DDE	SOD Activity U/mg protein	CAT Activity	APX Activity
Control—(without zucchini)	1.08 \pm 0.058 ^b	0.745 \pm 0.155 ^b	0.088 \pm 0.039 ^b
Intact lichen thallus—(with zucchini)	1.85 \pm 0.056 ^a	1.567 \pm 0.217 ^a	0.148 \pm 0.014 ^a

Data are means (\pm SD) of six replicates. Different lowercase letters within a column define significant differences based on One-Way ANOVA (SPSS 22.0) and followed by the LSD, at $p < 0.05$

there was no negative change in the chlorophyll content of the zucchini when treated with intact *Peltigera praetextata* thallus (Table 2). In our previous paper, the chlorophyll content of zucchini in weathered *p,p'*-DDE-contaminated soil was also increased when it is treated with intact *Peltigera canina* thallus (Akpınar et al. 2020).

When we look at the situation of intact lichen thallus in weathered *p,p'*-DDE-contaminated soil, there was no negative effect (Table 3). The chlorophyll degradation rate (*a/b*) ratio in a healthy lichen must be between 2 and 4 (Chettri et al. 1998; Bačkor et al. 2003). Our results suggest that the (*a/b*) rate of *Peltigera praetextata* is in the range (Table 3), like that was also observed in *Peltigera canina* (Akpınar et al. 2020). The presence of the zucchini or absence did not also change this situation in *P. praetextata*. However, the fresh weight of *P. praetextata* in weathered *p,p'*-DDE-contaminated soil increased in presence of the zucchini (Table 3). It is observed that the biological responses of the *P. praetextata* exposed to *p,p'*-DDE contamination are more positive in the presence of zucchini. It is also seen from the decrease in MDA content of *P. praetextata* on weathered *p,p'*-DDE-contaminated soil in the presence of zucchini (Table 5). MDA, which originated from membrane

lipid peroxidation and markers of oxidative damage in the cell membranes, are necessary molecules for assessing the physiological performance of plants and organisms under various stresses (Mishra et al. 2006; Gajewska and Skłodowska 2010; Sytar et al. 2013). Thus, oxidative damage was not detected in *P. praetextata* in the presence of zucchini in weathered *p,p'*-DDE-contaminated soil. But MDA content in leaves of zucchini increased when treated with both powdered and intact *Peltigera praetextata* thallus (Fig. 6). It was shown that oxidative damage occurred in the leaves of zucchini. Percentages of ion leakage detected in leaves of zucchini also support this state (Table 4).

It is known that hydroxyl radical (OH^\cdot)-mediated reactions are effective in the degradation of pollutants such as DDTs in the organism (Purnomo et al. 2011; Balawejder et al. 2014). The heterogeneous degradation of some pesticides like it by organisms is performed by OH^\cdot radicals (Liu et al. 2005; Al Rashidi et al. 2011, 2014; Lester et al. 2017; Mattei et al. 2018). The oxidative damage occurred in the leaves of zucchini may have been caused by the OH^\cdot ions needed for the degrading of the *p,p'*-DDE in the zucchini, which can survive in the soil contaminated by DDTs (White 2000, 2001, 2002; Wang et al. 2004; White et al.

2005, 2006a, b; Parrish et al. 2006; Mattina et al. 2006; Gent et al. 2007).

Oxidative stress occurred by the formation of reactive oxygen species (ROS) such as O_2^- , $^1O_2^-$, H_2O , and OH^- can be eliminated by several antioxidants (non-enzymatic or enzymatic) in the tissues (Sharma et al. 2012) and cell redox homeostasis is provided (Foyer and Noctor 2005). GSH and AsA are key molecules that are effective in defending against oxidative stress. Exposure to various stresses results in changes in levels of GSH, particularly (Noctor and Foyer 2000). Our results show that GSH activity is more effective than ascorbic acid content in zucchini. This may be because GSH is involved in protection from toxic pollutants by forming conjugation bridges between the thiol groups of proteins with pesticides such as DDTs, in addition to scavenging free radicals (Smirnoff 2005). *p,p'*-DDE reacts with glutathione (GSH) has been demonstrated in the Ph.D. thesis of Cantillana (2009). It is also known that GSH levels vary in plants exposed to various stresses (Noctor et al. 2002). In our results, the total GSH level was increased in leaves of zucchini treated with intact *P. praetextata* thallus (Fig. 7). The increase in GSH content could positively affect the cell redox homeostasis (Szalai et al. 2009; Noctor et al. 2012). In the study, we carried out with *P. canina*, total GSH activity was decreased in both the root and leaves of zucchini (Akpınar et al. 2020). Zhang et al. (2005) explain this situation as follows; GSH level increases as a result of adaptation mechanisms that come into play when level of oxidative stress is low, while GSH level decreases when oxidative stress is strong. If so, in presence of *P. praetextata*, the oxidative stress of zucchini grown in weathered soil contaminated with *p,p'*-DDE is lower than *P. canina*.

Enzymatic antioxidants analyzed in this study are SOD, CAT and APX. SOD activity is included in the primary step for decreasing the level of ROS radicals in the cell. The generated H_2O_2 resulting from SOD activity is eliminated by CAT and APX enzymes. If it cannot be detoxified to water (H_2O), H_2O_2 with Haber–Weiss or Fenton reactions catalyzed by Fe is transformed into OH^- radical and its derivatives, which is a very short-lived radical that initiates lipid peroxidation. It was determined in our study that CAT and APX activities in leaves of zucchini decreased with the *P. praetextata* treatments (powdered and intact lichen thallus) and the MDA content increased accordingly (Figs. 11, 12). These short-term radicals are also involved as signal molecules in growth and development and other metabolic processes of plants (Noctor et al. 2014). Therefore, although oxidative damage was detected in the leaves of zucchini, the growth and development of the zucchini were positively affected by *Peltigera praetextata* when other parameters were taken into account.

Conclusion

Our study contributes to the understanding of the effects of *Peltigera praetextata* on the growth and development of zucchini grown in weathered soil contaminated with *p,p'*-DDE, in addition to following the physiology and defence system of *P. praetextata*. *Peltigera praetextata* treatments increased the growth of above-ground organs of zucchini grown in weathered *p,p'*-DDE-contaminated soil. While oxidative damage occurred in the leaves of the zucchini, it did not occur in intact *Peltigera praetextata* thallus in weathered *p,p'*-DDE-contaminated soil. These findings show that *P. praetextata* has a strong antioxidative defence system and was not adversely affected by the weathered *p,p'*-DDE-contaminated soil with and without zucchini. Moreover, *P. praetextata* has affected the life of plants by increasing the biochemical, and biophysical utility of soil through active secondary metabolites and enzyme contents. In zucchini, oxidative damage was localized in the leaf parts. In addition, our results show that the damage that occurred is at the level of the cell membrane. This situation may concern the increase in *p,p'*-DDE uptake of zucchini in the presence of *Peltigera praetextata*. It is suggested to carry out detailed studies about the effect and mechanisms of different lichen species on the degradation of DDTs in future studies.

Author contribution statement The authors contributed jointly to the study design. Aysegul Akpınar conducted experiments, analyzed the data, and wrote the manuscript. Asuman Cansev also conducted experiments and contributed to the preparation of manuscript. Mehmet Isleyen found the funding of study and made the last checks in the manuscript.

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Declarations

Competing interests The authors declare that there are no competing interests.

References

- Akpınar A, Ozturk S, Sinirtaş M (2009) Effects of some terricolous lichens [*Cladonia rangiformis* Hoffm., *Peltigera neckerii* Hepp ex Müll. Arg., *Peltigera rufescens* (Weiss) Humb.] on soil bacteria in natural conditions. *Plant Soil Environ* 4:154–158

- Akpınar A, Cansev A, Isleyen M (2020) Effects of the lichen *Peltigera canina* on *Cucurbita pepo* spp. *pepo* grown in soil contaminated by DDTs. *Environm Sci Pollut Res*. <https://doi.org/10.1007/s11356-020-11665-4>
- Al Rashidi M, El Mouden O, Chakir A, Roth E, Salghi R (2011) The heterogeneous photo-oxidation of difenoconazole in the atmosphere. *Atmos Environ* 45:5997–6003. <https://doi.org/10.1016/j.atmosenv.2011.07.062>
- Al Rashidi M, Chakir A, Roth E (2014) Heterogeneous oxidation of folpet and dimethomorph by OH radical: A kinetic and mechanistic study. *Atmos Environ* 82:164–171. <https://doi.org/10.1016/j.atmosenv.2013.10.031>
- Alexander M (2000) Aging, bioavailability, and overestimation of risk from environmental pollutants. *Environm Sci Technol* 34(20):4259–4265
- Arora R, Pitchay DS, Bearce BC (1998) Water-stress-induced heat tolerance in geranium leaf tissues: A possible linkage through stress proteins? *Physiol Plant* 103(1):24–34
- Asplund J, Wardle DA (2017) How lichens impact on terrestrial community and ecosystem properties. *Biol Rev* 92:1720–1738
- Bačkor M, Loppi S (2009) Interactions of lichens with heavy metals. *Biol Plant* 53(2):214–222
- Bačkor M, Kováčik J, Dzubaj A, Bačkorová M (2003) Physiological comparison of copper toxicity in the lichens *Peltigera rufescens* (Weis) Humb and *Cladonia arbuscula* subsp. *Mitis* (Sandst.) Ruoss. *Plant Growth Regul* 58:279–286
- Balawejder M, Antos P, Czyjt-Kuryło S, Józefczyk R, Pieniazek M (2014) A novel method for degradation of DDT in contaminated soil. *Ozone Sci Eng* 36:166–173
- Barnes JD, Balaguer L, Manrique E, Elvira S, Davison AW (1992) A reappraisal of the use of DMSO for the extraction and determination of chlorophyll a and b in lichens and higher plants. *Environ Exp Bot* 32:85–100
- Beuchamp C, Fridovich I (1971) Superoxide dismutase; improved assays and an assay applicable to acrylamide gels. *Anal Biochem* 44:276–287
- Bhargava A, Carmona FF, Bhargava M, Srivastava S (2012) Approaches for enhanced phytoextraction of heavy metals. *J Environ Manage* 105:103–120
- Bian R, Chen D, Liu X, Cui L, Li L, Pan G, Xie D, Zheng J, Zhang X, Zheng J, Chang A (2013) Biochar soil amendment as a solution to prevent Cd-tainted rice from China: Results from a cross-site field experiment. *Ecol Eng* 58:378–383
- Bolan NS, Adriano DC, Curtin D (2003) Soil acidification and liming interactions with nutrient and heavy metal transformation and bioavailability. *Adv Agron* 78:215–272
- Cakmak I, Marschner H (1992) Magnesium deficiency and high-light intensity enhance activities of superoxide dismutase, ascorbate peroxidase and glutathione reductase in bean leaves. *Plant Physiol* 98:1222–1227
- Canha N, Almeida-Silva M, Freitas MC, Almeida SM, Wolterbeek HT (2014) Lichens as biomonitors at indoor environments of primary schools. *J Radioanal Nucl Chem* 291(1):123–128
- Cantillana T (2009) Toxicologically important DDT metabolites: Synthesis, enantioselective analysis and kinetics. Department of Environmental Chemistry Stockholm University, Stockholm
- Chen J, Blume HP, Beyer L (2000) Weathering of rocks induced by lichen colonization: a review. *CATENA* 39:121–146
- Chettri MK, Cook CM, Vardaka E, Sawidis T, Lanaras T (1998) The effect of Cu, Zn and Pb on the chlorophyll content of the lichens *Cladonia convolute* and *Cladonia rangiformis*. *Environ Exp Bot* 39:1–10
- Conti ME, Cecchetti G (2001) Biological monitoring: lichens as bioindicators of air pollution assessment – a review. *Environ Pollut* 114:471–492
- Costa ET, Guimarães Guilherme LR, Chaves de Melo ÉE, Teixeira Ribeiro B, dos Santos B, Inácio E, da Costa Severiano E, Faquin V, Hale BA (2012) Assessing the tolerance of castor bean to Cd and Pb for phytoremediation purposes. *Biol Trace Elem Res* 145:93–100
- Ehlers LJ, Luthy RG (2003) Contaminant bioavailability in soil and sediment. *Environ Sci Technol* 37:295A–302A
- Elman GL (1959) Tissue sulphhydryl groups. *Arch Biochem Biophys* 82:70–77
- Escudero A, Martinez I, de la Cruz A, Ojalora MAG, Maestre FT (2007) Soil lichens have species-specific effects on the seedling emergence of three gypophile plant species. *J Arid Environ* 70(1):18–28
- Favero-Longo SE, Piervittori R (2010) Lichen-plant interactions. *J Plant Interact* 5(3):163–177
- Foyer CH, Noctor G (2005) Redox homeostasis and antioxidant signaling: a metabolic interface between stress perception and physiological responses. *Plant Cell* 17:1866–1875
- Gadd GM (2007) Geomycology: biogeochemical transformations of rocks, minerals, metals and radionuclides by fungi, bioweathering and bioremediation. *Mycol Res* 111:3–49
- Gajewska E, Sklodowska M (2010) Differential effect of equal copper, cadmium and nickel concentration on biochemical reactions in wheat seedlings. *Ecotoxicol Environ Saf* 73:996–1003
- Gent MPN, White JC, Parrish ZD, Isleyen M, Eitzer BD, Mattina MI (2007) Uptake and translocation of *p, p'*-dichlorodiphenyl-dichloroethylene supplied in hydroponics solution to *Cucurbita*. *Environ Toxicol Chem* 26(12):2467–2475
- Glime JM (2007) Bryophyte ecology, Chapter 5–3, *Ecophysiology of Development: PROTONEMA* vol 1, *Physiological Ecology*
- Gray CW, Dunham SJ, Dennis PG, Zhao FJ, McGrath SP (2006) Field evaluation of in situ remediation of a heavy metal contaminated soil using lime and red-mud. *Environ Pollut* 142(3):530–539
- Gulen H, Eris A (2003) Some Physiological Changes in Strawberry (*Fragaria × ananassa* cv. *camarosa*) Plants Under Heat Stress. *J Hort Sci Biotech* 78:894–898
- Haga K, Lino M (2006) Asymmetric distribution of auxin correlates with gravitropism and phototropism but not with autostraightening (autotropism) in pea epicotyls. *J Exp Bot* 57(4):837–847
- Hao H, He Z, Li H, Shi L, Tang Y (2014) Effect of root length on epicotyl dormancy release in seeds of *Paeonia ludlowii*, Tibetan Peony. *Ann Bot* 113(3):443–452. <https://doi.org/10.1093/aob/mct273>
- Heath RL, Packer L (1968) Photoperoxidation in isolated chloroplasts: I. Kinetics and stoichiometry of fatty acid peroxidation. *Arch Biochem Biophys* 125:189–198
- Hodson ME, Vijver MG, Peijnenburg WJGM (2011) Bioavailability in soils. *Dealing with Contaminated Sites: From Theory towards Practical Application*, 721–746
- Hussain M, Farooq M, Nawaz A, Al-Sadi AM, Solaiman ZM, Alghamdi SS, Ammara U, Ok YS, Siddique KHM (2017) Biochar for crop production: potential benefits and risks. *J Soils Sediments* 17:685–716
- Isleyen M, Sevim P, Hawthorne J, Berger W, White JC (2013) Inheritance profile of weathered chlordane and *p, p'*-DDTs accumulation by *Cucurbita pepo* hybrids. *Int J Phytoremediat* 15(9):861–876
- Kim HS, Kim KR, Kim HJ, Yoon JH, Yang J, Ok Y, Owens G, Kim KH (2015) Effect of biochar on heavy metal immobilization and uptake by lettuce (*Lactuca sativa* L.) in agricultural soil. *Environ Earth Sci* 74:1249–1259
- Kumar D, Kumar Tripathi D, Kumar CD (2014) Phytoremediation potential and nutrient status of *Barringtonia acutangula* Gaertn. Tree seedlings grown under different chromium (CrVI) treatments. *Biol Trace Element Res* 157(2):164–174

- Kytöviita M-M, Stark S (2009) No allelopathic effect of the dominant forest-floor lichen *Cladonia stellaris* on pine seedlings. *Funct Ecol* 23:435–441
- Latkowska E, Chrapusta E, Bober B, Kaminski A, Adamski M, Bialczyk J (2015) Allelopathic effects of epiphytic lichen *Hypogymnia physodes* (L.) Nyl. colonization on the spruce (*Picea abies* (L.) Karst.) bark. *Allelopathy J* 35(1):129–138
- Lawrey JD (1977) Inhibition of moss spore germination by acetone extracts of terricolous *Cladonia* species. In *Bull Torrey Botanical Club* 104(1):49. <https://doi.org/10.2307/2484664>
- Lawrey JD (2009) Diversity of defensive mutualisms. Chapter 11. *Chemical defense in lichen symbiosis*. London: Taylor and Francis Group. pp: 167–181
- Leiva D, Clavero-León C, Carú M, Orlando J (2016) Intrinsic factors of *Peltigera* lichens influence the structure of the associated soil bacterial microbiota. *FEMS Microbiol Ecol* 92(11):fiw18
- Lester C, Moller N, Hammerum A (2004) Conjugal Transfer of Aminoglycoside and Macrolide Resistance between *Enterococcus faecium* Isolates in The Intestine of Streptomycin-Treated Mice. *Feems Microbiol Lett* 235:385–391
- Lester Y, Sabach S, Zivan O, Dubowski Y (2017) Key environmental processes affecting the fate of the insecticide chlorpyrifos applied to leaves. *Chemosphere* 171:74–80. <https://doi.org/10.1016/j.chemosphere.2016.12.013>
- Liu Q, Krüger H, Zetzsch C (2005) Degradation study of the aerosol-borne insecticides Dicolof and DDT in an aerosol smog chamber facility by OH radical in relation to the POPs convention. *Proc Eur Geosci Union Vienna Austria* 7:05760
- Mattei C, Wortham H, Quivet E (2018) Heterogeneous atmospheric degradation of pesticides by ozone: Influence of relative humidity and particle type. *Sci Total Environ* 625:1544–1553. <https://doi.org/10.1016/j.scitotenv.2018.01.049>
- Mattina MI, Isleyen M, Eitzer BD, Iannucci-Berger W, White JC (2006) Uptake by Cucurbitaceae of soil-borne contaminants depends upon plant genotype and pollutant properties. *Environ Sci Technol* 40(6):1814–1821
- Mishra S, Srivastava S, Tripathi RD, Govindarajan R, Kuriakose SV, Prasad MNV (2006) Phytochelatin Synthesis and Response of Antioxidants During Cadmium Stress in *Bacopa monnieri* L. *Plant Physiol Biochem* 44:25–37
- Noctor G, Foyer CH (2000) Homeostasis of adenylate status during photosynthesis in a fluctuating environment. *J Exp Bot* 51:347–356
- Noctor G, Gomez L, Vanacker H, Foyer CH (2002) Interactions between biosynthesis, compartmentation and transport in the control of glutathione homeostasis and signalling. *J Exp Bot* 53(372):1283–1304
- Noctor G, Mhamdi A, Chaouch S, Han Y, Neukermans J, Marquez-Garcia B, Queval G, Foyer C (2012) Glutathione in plants: An integrated overview. *Plant Cell Environ* 35:454–484
- Noctor G, Mhamdi A, Foyer CH (2014) The roles of reactive oxygen metabolism in drought: not so cut and dried. *Plant Physiol* 164:1636–1648
- Oliveira LL, Macedo AF (2015) The effect of light quality, temperature and substrate on seed germination and epicotyl development of *Carapa guianensis*, a multi-use neotropical tree. *J Med Plants Res* 9(18):582–593
- Parrish ZD, White JC, Isleyen M, Gent MPN, Iannucci-Berger W, Eitzer BD, Kelsey JW, Mattina MI (2006) Accumulation of weathered polycyclic aromatic hydrocarbons (PAHs) by plant and earthworm species. *Chemosphere* 64(4):609–618
- Peres MTL, Mapeli AM, Faccenda O, Gomes AT, Honda NK (2009) Allelopathic potential of orsellinic acid derivatives. *Braz Arch Biol Technol*. <https://doi.org/10.1590/S1516-89132009000400027>
- Piervittori R, Favero-Longo SE, Gazzano C (2009) Lichens and biodegradation of stonework: a review. *Chem Today* 27(6):8–10
- Pirintsos SA, Matsi T, Vokou D, Gaggi C, Loppi S (2006) Vertical distribution patterns of trace elements in an Urban environment as reflected by their accumulation in lichen transplants. *J Atmos Chem* 54:121–131
- Potter TI, Rood SB, Zanewich KP (1999) Light intensity, gibberellin content and the resolution of shoot growth in Brassica. *Planta* 207(4):505–511. <https://doi.org/10.1007/s004250050510>
- Purnomo AS, Mori T, Kamei I, Kondo R (2011) Basic studies and applications on bioremediation of DDT: a review. *Int Biodeter Biodegrad* 65(7):921–930
- Ruttens A, Colpaert JV, Mench M, Boisson J, Carleer R, Vangronsveld J (2006) Phytostabilization of a metal contaminated sandy soil. II: Influence of compost and/or inorganic metal immobilizing soil amendments on metal leaching. *Environ Pollut* 144:533–539
- Sharma P, Jha AB, Shanker Dubey R, Pessarakli M (2012) Reactive oxygen species, oxidative damage, and antioxidative defense mechanism in plants under stressful conditions, review article. *J Botany Article ID* 217037, 26 pages
- Smirnoff N (2005) *Antioxidants and reactive oxygen species in plants*. Blackwell Publishing Ltd
- Stevens CJ, Smart SM, Henrys PA, Maskell LC, Crowe A, Simkin J, Cheffings CM, Whitfield C, Gowing DJG, Rowe EC, Dore AJ, Emmett BA (2012) Terricolous lichens as indicators of nitrogen deposition: evidence from national records. *Ecol Ind* 20:196–203
- Sytar O, Kumar A, Latowski D, Kuczynska P, Strzałka K, Prasad MNV (2013) Heavy metal-induced oxidative damage, defense reactions, and detoxification mechanisms in plants. *Acta Physiol Plant* 35:985–999
- Szalai G, Kellős T, Galiba G, Kocsy G (2009) Glutathione as an antioxidant and regulatory molecule in plants under abiotic stress conditions. *Plant Growth Regul* 28:66–80
- Tiansawat P, Dalling JW (2013) Differential seed germination responses to the ratio of red to far-red light in temperate and tropical species. *Plant Ecol* 214:751–764
- Valencia-Islas N, Zambrano A, Rojas JL (2007) Ozone reactivity and free radical scavenging behavior of phenolic secondary metabolites in lichens exposed to chronic oxidant air pollution from Mexico City. *J Chem Ecol* 33:1619–1634
- Wang XP, White JC, Gent MPN, Iannucci-Berger W, Eitzer BD, Mattina MI (2004) Phytoextraction of weathered *p,p'*-DDE by zucchini (*Cucurbita pepo*) and cucumber (*Cucumis sativus*) under different cultivation conditions. *Int J Phytoremed* 6(4):363–385
- White JC (2000) Phytoremediation of Weathered *p,p'*-DDE Residues in Soil. *Int J Phytoremed* 2(2):133–144
- White JC (2001) Plant-facilitated mobilization and translocation of weathered 2,2-bis(p-chlorophenyl)-1,1-dichloroethylene (*p,p'*-DDE) from an agricultural soil. *Environ Toxicol Chem* 20(9):2047–2052
- White JC (2002) Differential bioavailability of field-weathered *p,p'*-DDE to plants of the Cucurbita and Cucumis genera. *Chemosphere* 49(2):143–152
- White JC (2009) Optimizing planting density for *p,p'*-DDE phytoextraction by *Cucurbita pepo*. *Environ Eng Sci* 26(2):369–375
- White JC, Wang XP, Gent MPN, Iannucci-Berger W, Eitzer BD, Schultes NP, Arienzo M, Mattina MI (2003) Subspecies-level variation in the phytoextraction of weathered *p,p'*-DDE by *Cucurbita pepo*. *Environ Sci Technol* 37(19):4368–4373
- White JC, Parrish ZD, Isleyen M, Gent MPN, Iannucci-Berger W, Eitzer BD, Mattina MI (2005) Uptake of weathered *p,p'*-DDE by plant species effective at accumulating soil elements. *Microchem J* 81(1):148–155
- White JC, Parrish ZD, Isleyen M, Gent MPN, Iannucci-Berger W, Eitzer BD, Kelsey JW, Mattina MI (2006a) Influence of citric acid amendments on the availability of weathered PCBs to plant and earthworm species. *Int J Phytoremed* 8(1):63–79

- White JC, Parrish ZD, Gent MPN, Iannucci-Berger W, Eitzer BD, Isleyen M, Mattina MI (2006b) Soil amendments, plant age, and intercropping impact *p, p'*-DDE bioavailability to *Cucurbita pepo*. *J Environ Qual* 35(4):992–1000
- White JC, Peters R, Kelsey JW (2007) Surfactants differentially impact *p, p'*-DDE accumulation by plant and earthworm species. *Environ Sci Technol* 41(8):2922–2929
- Yang X, Feng Y, He Z, Stoffella PJ (2005) Molecular mechanisms of heavy metal hyperaccumulation and phytoremediation. *J Trace Elem Med Biol* 18(4):339–353
- Zhang JF, Liub H, Sun YY, Wang XR, Wu JC, Xue YQ (2005) Responses of the antioxidant defenses of the Goldfish *Carassius*

auratus, exposed to 2,4-dichlorophenol. *Environ Toxicol Pharmacol* 19:185–190

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