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Heavy metal accumulation and impact on protein content of *Brahmina coriaceae* Hope infesting *Populus deltoides* (W. Bartram ex. Marshall)

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Abstract

The present investigation was conducted to study the heavy metal accumulation and impact on protein content of *Brahmina coriaceae* infesting *Populus deltoides* at the Research Farm of Department of Environmental Science, Dr Y S Parmar University of Horticulture and Forestry, Nauni, Solan, H.P during 2014- 2015. The impact of soil application of graded doses of Cd and Hg in the protein content and their accumulation in the body of the white grub larvae was studied. Cd and Hg content in the larval body ranged from 0.032-0.101 ppm and 0.010-0.075 ppm, respectively. There was decrease in protein content from 8.97% (control) to 4.96% (Cd 20 ppm and Hg 20 ppm) in the body of white grub larvae. The observed ability of white grubs to continue growth even at higher doses of Cd and Hg and the ability to accumulate metals in their tissues demonstrated its resistance to moderate to high levels of metals.

Keywords: White grubs, phytoremediation, cadmium, mercury

1. Introduction

Wide range of tree species viz. Poplar, Maple, Robinia, Salix etc. are naturally known to occur in areas where industrial effluents with heavy loads of metals are discarded and therefore recommended for plantations around industrial sites for phytoremediation ^[1]. The genus *Populus* is regarded as model system for forest trees since it offers several advantages including rapid growth, high biomass production and prolific sexual reproduction, combined with a relatively small genome ^[2]. Poplar is also regarded as a suitable candidate for use in phytoremediation of polluted soils due to relevant features such as the ability to withstand environmental stresses, the extensive root system and the high water uptake. It is geographically widespread in various climatic areas, is adapted to contaminated or polluted soils and has the capacity to accumulate heavy metals. Its use is reported for the extraction or immobilization (phytostabilization) of heavy metals present in polluted sites ^[3]. In addition, poplar species are currently grown as cash crops for pulpwood and as a renewable energy source. Therefore, poplar has been proposed as a model for studying the biology of trees ^[4]. Contamination of agricultural soils by spraying of pesticides, use of livestock manure, fertigation and other organic waste has been reported by various workers. Cd and Hg are ubiquitous in sewage, sludges, biosolids and wastewater from industrial (e.g. tanneries), agricultural (liquid manures from industrial farming) and domestic sources that are utilized as low price fertilizers for bioenergy plantations ^[5].

P. deltoides in nurseries is attacked by a wide range of insect pest viz., white grubs, cutworms, termites, nematodes etc. Among the various groups white grubs (*Brahmina coriaceae*) is a serious pest of *P. deltoides* seedlings in nursery ^[6]. Phytophagous insects feeding on hyper-accumulators are reported to accumulate heavy metals in their body. Increased metal availability in the environment can be detrimental for the growth and development of all organisms in a food web due to bio-transfer or bioaccumulation of metals between trophic levels ^[7]. Concentrations of metal found in contaminated habitats have been reported to cause immune stimulation or immune suppression in terrestrial organisms. Low concentration of Ni can stimulate the immune system in *S. litura* but higher concentrations can cause death by inhibiting the immune system. Further, various hyper-accumulators have been shown to be able to defend themselves against infestation by insects and pathogens. Species of bark and wood boring beetles have been shown to accumulate heavy metals in the body tissues according to the degree of industrial pollution of the environment.

The importance of accumulation of heavy metals in phytophagous insects especially phloem feeders such as aphids have been shown in a number of laboratory, green house and field studies. Keeping this in view the present study was taken with the objective to study the accumulation of cadmium and mercury in white grub *Brahmina coriacea* infesting *Populus deltoides* nursey, grown in soils contaminated with the heavy metals and impact of heavy metal accumulation on protein content of grubs of *Brahmina coriacea*.

2. Materials and methods

The present research experiment was laid to study the accumulation of Cd and Hg in white grub larvae infesting *P. deltoides* seedlings at the Research Farm of Department of Environmental Science Dr Y S Parmar University of Horticulture and Forestry, Nauni, Solan, Himachal Pradesh during the year 2014- 2015. The Research farm is located at an altitude of 1273 m amsl and at latitude of 35.5°N, longitude of 77.8°E which falls in the mid hill zone of Himachal Pradesh having sub-temperate and semi-humid type of climate.

Young cuttings of uniform size from one-year-old cut back stems and branches of *P. deltoides* were planted in pots. The growing media was prepared by using sand, soil and FYM in the ratio 1:1:1. The cuttings sprouted completely after 30-40 days of planting and the selected treatment combinations were applied after complete establishment of the seedlings. In order to study the effect of cadmium and mercury accumulation on white grubs, field experiment with following treatments viz: T₁ (Cd₀Hg₀ ppm), T₂ (Cd₀Hg₁₀ ppm), T₃ (Cd₀Hg₂₀ ppm), T₄ (Cd₁₀Hg₀ ppm), T₅ (Cd₁₀Hg₁₀ppm), T₆ (Cd₂₀Hg₀ ppm), T₇ (Cd₂₀Hg₂₀ ppm) was laid with 4 replications. Cd was applied through CdCl₂.H₂O (M.W. 201.324 g/mol) and Hg through HgCl₂ (M.W. 271.524 g/mol). Ten second instar white grubs were released in each experimental pot. At the end of the experiment the grubs were collected from each experimental pot, brought to the laboratory and processed for metal accumulation and protein content.

2.1 Estimation of Cd and Hg content in white grub (*B. coriacea*).

Cd and Hg content in the body of white grub was estimated as per the method [8]. Cd and Hg concentration in each sample was detected using Inductively Coupled Plasma Atomic Spectrometer (ICP-6300 DUO).

2.2 Estimation of protein content in white grub (*B. coriacea*).

White grub sample for detection of protein were prepared as per A.O.A.C. method [9]. The prepared samples were kept at 37°C for 10 minutes for appearance of purple color. The absorbance was recorded at 520 nm against blank reagent of standard protein solution. The protein content was calculated using a standard graph.

2.3 Statistical analysis

The observations recorded on accumulation of Cadmium (Cd) and Mercury (Hg) in grubs of *Brahmina coriacea* and effect of graded doses of Cd and Hg on protein content in grubs were subjected to statistical analysis under CRD (Completely Randomized Design). Analysis of Variance (ANOVA) was worked out and critical difference at 5 percent level of significance was calculated as suggested by Cochran and Cox [10].

3. Results and Discussion

3.1 Accumulation of Cd and Hg in white grub larvae (*B. coriacea*)

The Cd and Hg content in the white grub body ranged from 0.032-0.101ppm and 0.010-0.075 ppm, respectively (Table 1). There was significant difference in Cd content in the body of white grubs at different treatment. The lowest Cd content (0.032 ppm) was observed in T₂ (Cd₀Hg₁₀), whereas, the highest Cd concentration (0.101ppm) was found in the body of white grubs fed on *P. deltoides* grown at (T₆). Cadmium is a toxic metal because of its relatively high mobility in the soil-plant system. Cadmium is easily translocated from plant roots to above ground tissues. The concentration of Cd in the body of white grubs is supported by the findings of Merrington [11] who reported that soil treated with different concentrations of Zn and Cd (0, 10 and 30 t dry solids/ha) increased the accumulation of heavy metals in arthropods through soil- plant-arthropod system. The similar findings were obtained by Gintenreiter *et al.* [12] who reported increase in cadmium content in the larval body of gypsy moth (*Lymantria dispar*) which reached much higher levels as compared to levels in food. Qiang *et al.* [13] also reported accumulation of zinc in different tissues of *S. litura* larvae which increased with the increasing zinc dose in artificial diet. Present findings are also in confirmation with the findings of Xia *et al.* [14] who reported nickel accumulation in the sixth instar larvae of *S. litura* within a generation which increased with the increase of the nickel dose in the artificial diet. Qiang *et al.* [15] reported accumulation of zinc in the larval hemolymph and fat body of *S. litura* which was a doses-dependent relationship. The Hg concentration recorded in the body of white grub larvae infesting *P. deltoides* seedlings grown in heavy metal contaminated soils had no significant difference among them.

The present results on significant difference in concentration of cadmium among treatments and non-significant difference in uptake of mercury at soil-plant-herbivorous insect (white grub) interaction find support from the studies of Zhang *et al.* [16] who reported the concentration factors of Hg and Cd in soil plant- the herbivorous insect, carnivorous insect food chain as 0.18, 6.57 and 7.88 for Hg and 6.82, 2.01 and 0.48 for Cd i.e. movement of Cd from soil plant was more as compared to Hg and hence more accumulation in the body. As compared to Cd, Hg has more tendencies of bio-magnifications among the first and second consumer level.

3.2 Protein content in white grub (*B. coriacea*).

The protein content in the body of *B. coriacea* had decreasing trend with increasing doses of Cd and Hg in interaction (Table 2). Statistically, highest protein content of 8.97 per cent was recorded in T₁(Cd₀Hg₀) i.e. when white grubs fed on *P. deltoides* seedlings grown in uncontaminated soil (control) as compared to other treatments. Lowest protein content of 4.96 per cent was recorded in T₇ (Cd₂₀Hg₂₀). Since protein is an important component of an organism's body, in the present study protein content of 8.97 per cent was reported at treatment T₁ i.e. in the body of the white grubs which infested the *P. deltoides* seedlings grown in soils to which heavy metals were not applied. The observations are close to the findings of Alhassan [17] who reported 12.75 percent protein in the body of white grubs under normal conditions while studying proximate composition of *B. coriacea*. El-Sheikh *et al.* [18] reported a significant decrease in total carbohydrate content and total lipid content in the whole body of males and females of *Culex pipens* when exposed to

cadmium and copper and they further reported that presence of heavy metals in environment system of mosquito would contribute to reduction of mosquito breeding. Similarly, in the present studies, when exposed to high levels of heavy metals there is reduction in protein content of white grubs, which may contribute in the management of pest. Similarly, Hg caused significant mortality of hemocyte cells of Pacific Oyster (*Crassostrea gigas*) after 24 hours of invitro incubation and also inhibited the phenol oxidase like activity under Hg stress Gagnaire *et al.* [19].

Table 1: Accumulation of Cadmium (Cd) and Mercury (Hg) in grubs of *Brahmina coriaceae*

Treatments	Cd(ppm)	Hg(ppm)
T ₁ Cd ₀ Hg ₀	0.020	0.010
T ₂ Cd ₀ Hg ₁₀	0.032	0.041
T ₃ Cd ₀ Hg ₂₀	0.047	0.066
T ₄ Cd ₁₀ Hg ₀	0.052	0.025
T ₅ Cd ₁₀ Hg ₁₀	0.066	0.057
T ₆ Cd ₂₀ Hg ₀	0.101	0.033
T ₇ Cd ₂₀ Hg ₂₀	0.079	0.075
C.D _{0.05}	0.024	NS

Table 2: Effect of graded doses of Cd and Hg on protein content in grubs of *Brahmina coriaceae*

Treatments	Protein content (%)
T ₁ Cd ₀ Hg ₀	8.97
T ₂ Cd ₀ Hg ₁₀	6.44
T ₃ Cd ₀ Hg ₂₀	5.45
T ₄ Cd ₁₀ Hg ₀	6.47
T ₅ Cd ₁₀ Hg ₁₀	6.28
T ₆ Cd ₂₀ Hg ₀	5.72
T ₇ Cd ₂₀ Hg ₂₀	4.96
C.D _{0.05}	1.22

4. Conclusion

The present study confirms the earlier findings about the accumulation of heavy metals in insects. Such accumulation may hinder the growth by way of suppressing the feeding indices or accumulation of heavy metals may enhance growth. In the present study, the protein content in the body of *B. coriaceae* had decreasing trend with increasing doses of Cd and Hg. The actual effect of heavy metals may be considered as various physiological processes working together.

5. Acknowledgement

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