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SHORT COMMUNICATION

EFFECT OF CADMIUM AND NICKEL TOXICITY ON THE PEROXIDASE ACTIVITY AND CAROTENOIDS CONTENT IN MOSS *THUIDIUM CYMBIFOLIUM*.

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Effects of two heavy metals (nickel and cadmium) with different phytotoxicity were examined on the activity of peroxidase (POX) and carotenoids content in pleurocarpous moss *Thuidium cymbifolium* (Doz and Molk.). Cd treatment for 3 days caused an increase in peroxidase activity only at 0.01 M concentration from 0.464 to 0.514 $\triangle A_{470}$ min^{-1} g^{-1} fw, while for Ni, peroxidase activity was higher for all concentrations in comparison to control. However, carotenoids content after 3 days treatment increased upto 51% for Ni only and decreased upto 5-6% of control for Cd. POX activity after 15 days treatment decreased 74% for Ni and 90% for Cd. Contrastingly carotenoid declined upto 65% for Ni and only upto 26% of control for Cd. Results suggest that both POX and carotenoid content could be used as bio-indicators of metal pollution. Study suggests that Cd is more phytotoxic than Ni.

***Key words*: Cadmium, carotenoids, nickel, peroxidase, *Thuidium cymbifolium*.**

Due to the impact of human activity the issues of metal contamination are becoming increasingly relevant (Fernandes and Henriques 1991). Metal pollutants that are considered to be a potential threat include Cd, Cr, Cu, Hg, Ni, Zn and Pb. Due to their distinct chemistry and characteristics each represents a rather different hazard to the environment. Cd is a non-essential element for which no biological function has been found up till now (Cieslinski *et al*. 1996), Ni is, however, a beneficial micronutrient (Gerendas and Sattelmacher 1997) in low concentrations. Exposure to high levels of cadmium results in reduced rates of photosynthesis (Kahle 1993), oxidaton of sulfhydryl groups and formation of metal thiolate bonds, alteration in protein secondary structures, changes in the redox status of the cell, interference with essential metal uptake and finally death (Sandalio *et al*. 2001). Ni reduces plant growth and disrupts metabolic and physiological processes, especially of photosynthesis (Krupa *et al*. 1993).

Cadmium is found to induce oxidative stress in cells (Somashekaraiah *et al*. 1992). The enhanced production of ROS during stress can pose a threat to cells and therefore, detoxification of excess ROS especially, during stress is essential. Major ROS-scavenging mechanisms of plants include enzymes such as superoxide dismutase (SOD), peroxidases (POX) and catalase (CAT) (Allen 1995) and non-enzymatic scavengers involves ascorbic acid, glutathione, α-tocopherols and carotenoids (Asada and Takahashi 1987). Little information is available on the effect of metal pollutants on POX activity and carotenoids content and their contribution in protection from oxidative stress. The present study was therefore, conducted to study the effects of heavy metals Cd and Ni on POX activity and carotenoids contents in moss *Thuidium cymbifolium*.

Samples of moss *Thuidium cymbifolium* were collected from Artola in December 2002 in polythene bags from a uniform area of 50 cm^2. After collection, adhering litter was removed by hand and finally washed in running tap water. Dilute solutions of heavy metals $CdCl_2$ and $NiSO_4$ were used to determine plant sensitivities to metal toxicity. Moss samples were treated with different concentrations (0.01, 0.1 and 0.2 M) for 3 and 15 days respectively. After treatment periods plant

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tissues were used for physiological analysis. All the treatments had three replications. The activity of POX was measured by spectrophotometric method of Putter (1974). Moss samples (1g) were homogenized in 0.1 M phosphate buffer (pH 7.4) at 0-4°C and centrifuged at 10,000 g for 20 min. For POX, the oxidation of guaiacol was measured by the increase in absorbance at 470 nm for 1 min. The reaction was started with H_2O_2. Carotenoids (xanthophylls and carotenes) were determined spectrophotometrically according to the Arnon (1949) method by extracting 0.5 g of moss samples in 80% Acetone.

The changes in specific activity of peroxidase and carotenoids content on treatment with Cd and Ni are shown in Figures 1 and 2. Peroxidase activity after 3 days of Cd treatment increased from 0.464 in control to 0.514 $\triangle A_{470}$ min^{-1} g^{-1} fr. wt. in 0.01 M concentration,

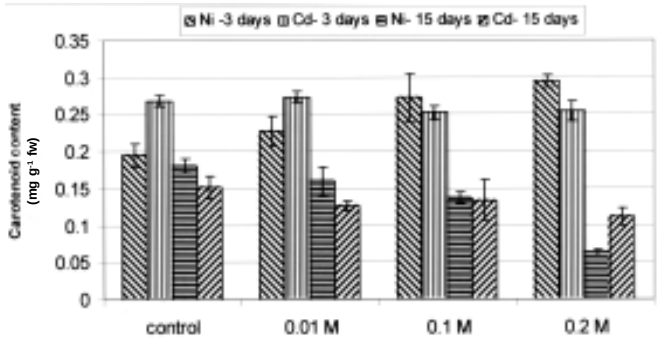

Fig. 1. Effect of different concentrations of Ni and Cd on carotenoids content (mg g^{-1} fw). Each value is the mean of 3 replicates ± S.E.

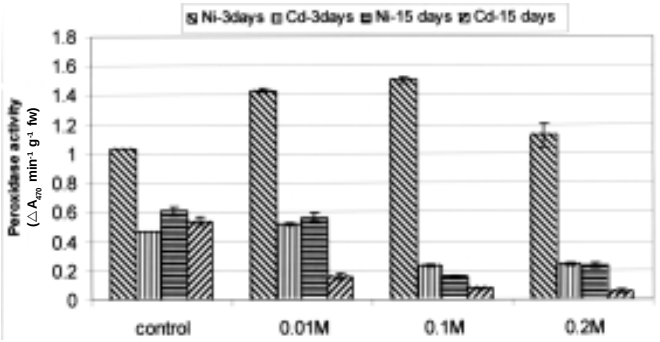

Fig. 2. Effect of different concentrations of Ni and Cd on peroxidase acticity ($\triangle A_{470}$ min^{-1} g^{-1} fw). Each value is the mean of 3 replicates ± S.E.

decreasing to 0.242 in 0.2 M concentration. After 15 days a gradual decrease ranging from 71% - 90% of control was observed, indicating toxic effect of Cd. Similar increase in activity of guaiacol-peroxidase to moderate Cd stress (0.125 -0.5 mM) was noticed by Leon *et al.* (2002) in different cultivars of *Capsicum annum* L. On the other hand in case of Ni a total increase of 45% over to control at 0.01 M concentration was noticed, which, however, decreased remarkably at 0.2 M Ni, though it was still 9% higher compared to control. Saxena and Saxena (2002) reported increase in POX activity by Ni at 0.001 M over control, which decreased on increasing concentration at 0.01 and 0.1 M following 48 h treatment of moss *Sphagnum cuspidatum*. Carotenoids content also increased 51% over control following 3 day of Ni treatment but decreased significantly to 65% of control after 15 days. Phytotoxicity of Cd was apparent by decrease in carotenoids from 0.268 in control to 0.254 mg g^{-1} fr. wt. at 0.2 M concentration after 3 days, while treatment period of 15 days caused a 26% decrease in carotenoids over to control.

Carotenoids degradation is a commonly observed phytotoxic effect of heavy metals (Moustakas *et al.* 1997). It could probably be due to carotenoids degradation as well as inhibition of carotenoids biosynthesis (Dubey 1997) by Cd. However, contrasting results have been reported for Ni. While short duration exposure of 72 h increased carotenoid content by 51% over control longer duration of exposure decreased carotenoids content by 24% in 0.1 M concentration and 65% in 0.2 M. Severe stress intensity (i.e. longer exposure time) caused a significant decrease in POX and carotenoids content indicating phytotoxic effect of Cd and Ni. Cadmium proved to be more phytotoxic than Ni, and physiological response of a test plant depends not only on metal concentrations but is also dependent on duration of exposure.

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