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Effect of heavy metals on the growth of gemmae of *Marchantia paleacea* Bertol

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ABSTRACT

The present study has been done to study the effect of heavy metals (Zn, Mn, Cu, Pb and Ni) on the growth of the gemmae of *M. paleacea*. Four different concentrations (10 ppm, 25 ppm, 50 ppm and 100 ppm) of heavy metals (Pb, Cu, Zn, Mn and Ni) and one control were used to carry out experiments. It was found that Cu, Ni, Zn and Pb are potentially toxic to gemmalings of *M. paleacea*, at certain level, while Cu showed considerably greater effects at low levels. Interestingly, in the case of Mn, it has been found that gemmalings of *M. paleacea* tolerate relatively high Mn levels and show better growth as compared to Control.

KEYWORDS

Gemmae;
Marchantia paleacea;
Heavy metals;
Concentrations.

INTRODUCTION

The problem of environmental pollution is receiving much more attention now than in the past years. It is now an established fact that a large number of pollutants including the heavy metals are affecting our environment. The phytotoxicity of these heavy metals generally depends on their bioavailability, which depends on their occurrence in different chemical forms. All plants have the ability to accumulate essential oligoelements such as Cu, Mn and Zn from soil and water. Some plants which also incorporate heavy metals, like Cd, Co, Cr, Hg, Pb, and Se, which have no known biological function as per the print information available, as yet, but infact prove toxic for the majority of plants[1]. Bryophytes possess a counter gradient mechanism to concentrate heavy metals within their tissues and owing to this these are widely used as bioindicators for their unique and very specific responses[2]. Ghate and Chaphekar[3] used *Plagiochasma appendiculatum* as a bio-test for water quality assessment. They exposed the plants in different pollution stresses, especially heavy metals, synthesized in the laboratory, to simulate pollutants reported to occur in industrial areas. The plant was found to be sensitive to Hg and Cu.

EXPERIMENTAL

Preparation of media

Murashige and Skoog media was used to grow *M.*

TABLE 1 : Effect of Zn on gemmae of *M. paleacea* after 15 Days

S.No	Treatment	Gemmae			
		Colour	Size (mm)	Area (mm^2)	Development of gemmae
1	Control	Dark green	3.0-3.5 mm long×1.5-2.0 mm wide	1.51	Well developed, two thalli one from each notch was developed, rhizoids numerous.
2	10 ppm	Green	1.5-2.0 mm long×0.80-0.96 mm wide	0.66	Well developed, two thalli one from each notch was developed, rhizoids numerous.
3	25 ppm	Green	1.5-2.0 mm long×0.48-0.64 mm wide	0.66	Delicate, two thalli one from each notch was developed, rhizoids numerous.
4	50 ppm	Green	1.8-2.0 mm long×0.96-1.12 mm wide	0.56	Delicate, two thalli one from each notch was developed, rhizoids numerous.
5	100 ppm	Yellowish green	1.8-2.0 mm long×0.96-1.12 mm wide	0.37	Very delicate, two thalli one from each notch was developed, rhizoids less in number.

TABLE 2 : Effect of Mn on gemmae of *M. paleacea* after 15 days

S.No	Treatment	Gemmae			
		Colour	Size (mm)	Area (mm^2)	Development of gemmae
1	Control	Dark green	3.0-3.5 mm long×1.5-2.0 mm wide	1.51	Well developed, two thalli one from each notch was developed, rhizoids numerous.
2	10 ppm	Green	2.5-3.0 mm long×1.0-2.0 mm wide	1.30	Well developed, two thalli one from each notch was developed, rhizoids numerous.
3	25 ppm	Green	2.5-3.0 mm long×1.5-2.0 mm wide	2.16	Well developed, two thalli one from each notch was developed, rhizoids numerous.
4	50 ppm	Green	2.5-3.0 mm long×1.0-1.5 mm wide	0.99	Well developed, two thalli one from each notch was developed, rhizoids numerous.
5	100 ppm	Light green	2.5-3.0 mm long×1.0-2.0 mm wide	1.30	Delicate, two thalli one from each notch was developed, rhizoids less in number.

TABLE 3 : Effect of Cu on gemmae of *M. paleacea* after 15 days

S.No	Treatment	Gemmae			
		Colour	Size (mm)	Area (mm^2)	Development of gemmae
1	Control	Dark green	3.0-3.5 mm long×1.5-2.0 mm wide	1.51	Well developed, two thalli one from each notch was developed, rhizoids numerous.
2	10 ppm	Yellowish green	0.80-0.96 mm long×0.40-0.48 mm wide	0.41	Delicate, thalli did not develope, rhizoids less.
3	25 ppm	Yellow	0.56-0.64 mm long×0.40-48 mm wide	0.13	Thalli did not develop, rhizoids absent.
4	50 ppm	Light yellow	0.32-0.40 mm long×0.24-0.32 mm wide	0.07	Thalli did not develop, rhizoids absent.
5	100 ppm	Colourless	0.40-0.48 mm long×0.24-0.32 mm wide	0.067	Thalli did not develop, rhizoids absent.

paleacea in culture. The pH of the media was maintained at 5.8 pH before autoclaving. The media was autoclaved at 15 psi for 15 minutes.

Culturing of gemmae

The gemmae were obtained from aseptic cultures of *M. paleacea*. The gemmae were surface sterilized with 3% sodium hypochlorite solution and washed repeatedly with sterile water. The gemmae were picked up by a sterile forcep and transferred to the prepared medium.

The experiment was carried out in laboratory under controlled temperature (20°-23°C) and relative humidity of 50-60%. Artificial lighting from Day light fluorescent tubes was used.

Four different concentrations (10 ppm, 25 ppm, 50 ppm and 100 ppm) of heavy metals (Pb, Cu, Zn, Mn and Ni) and one control were used to carry out experiments. For each treatment 20-25 gemmae were inoculated in each set of three petridishes. The growth of gemmae were observed on 15^{th} day. Thalli area was

TABLE 4 : Effect of Pb on gemmae of *M. paleacea* after 15 days

S.No	Treatment	Gemmae			
		Colour	Size (mm)	Area(mm^2)	Development of gemmae
1	Control	Dark green	3.0-3.5 mm long×1.5-2.0 mm wide	1.51	Well developed, two thalli one from each notch was developed, rhizoids numerous.
2	10 ppm	Green	3.0-3.5 mm long×1.0-1.5 mm wide	1.59	Well developed, two thalli one from each notch was developed, rhizoids numerous.
3	25 ppm	Green	2.0-3.0 mm long×0.96-1.12 mm wide	1.04	Developed, two thalli one from each notch was developed, rhizoids numerous.
4	50 ppm	Green	1.8-2.0 mm long×0.96-1.12 mm wide	0.80	Developed, two thalli one from each notch was developed, rhizoids less than 25 ppm.
5	100 ppm	Green	1.8-2.0 mm long×0.96-1.12 mm wide	0.75	Developed, two thalli one from each notch was developed, but one side thalli is larger than other, rhizoids numerous.

TABLE 5 : Effect of Ni on gemmae of *M. paleacea* after 15 days

S.No	Treatment	Gemmae			
		Colour	Size (mm)	Area (mm^2)	Development of gemmae
1	Control	Dark Green	3.0-3.5 mm long ×1.5-2.0 mm wide	1.51	Well developed, two thalli one from each notch was developed, rhizoids numerous.
2	10 ppm	Green	2.5-3.0 mm long ×0.80-1.0 mm wide	1.27	Delicate, two thalli one from each notch was developed, rhizoids numerous.
3	25 ppm	Light green	1.44-1.6 mm long 0.40 × 0.48 mm wide	0.37	Delicate, thalli did not well develop, rhizoids less.
4	50 ppm	Light yellow	0.80-0.88 mm long × 0.40-0.48- mm wide	0.16	Thalli did not develop, rhizoids absent.
5	100 ppm	Colourless	0.48-0.64 mm long × 0.32-0.40 mm wide	0.08	Thalli did not develop, rhizoids absent.

measured on Area Measurement System of Delta Devices Ltd., England.

RESULTS AND DISCUSSION

In this study, the effects of different concentrations of heavy metals (Pb, Cu, Zn, Mn and Ni) on the growth of gemmalings in *M. paleacea* were investigated. It has been observed that some morphological changes occurred in response to heavy metal in gemmalings.

In the case of Zn, at 10 ppm, 25 ppm, and 50 ppm concentration, there was not any major changes observed but at 100 ppm gemmae become yellowish green, very delicate, rhizoids less in number, size and area of gemmalings were also found to be decreased (TABLE 1). Relatively low concentrations of Zn may interfere with biological process related to the general metabolism of plants, including photosynthesis[4].

No noticeable changes occur in gemmalings, grown in Mn containing media. Gemmalings have shown better growth as compared to control at 25 ppm concentration (TABLE 2).

At 10 ppm concentration of Cu, gemmae become yellowish green, thalli not developed, rhizoids less in number, while progressive reduction in chlorophyll could be observed with increasing Cu concentration. Cu, which is relatively mild in character, is highly toxic to plants even at a micro molar range of exposure[5] but it is also one of the micronutrients essential for plant growth. Cu block the photosynthetic electron transport chain and degrade chlorophyll[6,7]. Gemmalings appeared colourless at 100 ppm. Gemmalings size and area was also found decreasing with increasing Cu concentration (TABLE 3). Cu is a co-factor of various oxidase enzymes, it exerts toxic effects by stimulating the activity of peroxidase enzymes with a subsequent disruptive effect on cellular. Mukherji and Das Gupta[8] have demonstrated such increases in peroxidase activities in lettuce seedlings.

In the case of Pb, gemmaling size and area was found decreasing with increasing Pb concentration. Gemmalings growth was restricted at one side at 100 ppm concentration of Pb (TABLE 4). Lead released into the cytoplasm can distrupt the normal cell function[9].

Progressive reduction in chlorophyll, gemmaling size and area was noticed with increasing Ni concentration. At 100 ppm, gemmalings were colourless, thalli were also not developed (TABLE 5).

From the above results, it become evident that Cu was more toxic to gemmalings, followed by Ni, Zn and Pb. In the case of Mn, It has been found that gemmalings of *M. paleacea* tolerate relatively high Mn levels and show better growth as compared to Control, Cu, Ni, Zn and Pb are potentially toxic to gemmalings of *M. paleacea*, at certain level, while Cu showed considerably greater effects at low levels. The results from the above experiments also demonstrate the problem of establishment of bryophyte populations in polluted localities.

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