

Anatomical changes of *Ipomoea reptans* due to mercury uptake and accumulation in contaminant soil

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Abstract. Heavy metal contaminants like mercury is a serious problem to human, animals, and some plants' health. Phytoremediation is an alternative technique, which can remediate the contaminants from soil using a hyperaccumulator plant. The goal of this research was to study anatomical changes of plant main organs (roots, stems, and leaves) of *Ipomoea reptans* which are assumed as a hyperaccumulator plant that grows in mercury contaminant. The mercury concentration of the growth medium was 0, 61.871, 92.258, and 107.046 ppb. *Ipomoea reptans* were harvested after 27 days. The anatomical changes of the plant's main organs were observed by preparing the cross-section of roots, stems, and leaves of *I. reptans*. The result showed that mercury treatment has caused anatomical damage at the xylem vessel of the root, and decreased bulliform cell size. The anatomical damage was found only in the root of *I. reptans*. The mercury concentration in media decrease to 0 ppb (P0), 50,420 ppb (P1), 58,583 (P2), and 96,120 (P3).

Keywords: Heavy metal, *Ipomoea reptans*, Leave, Mercury, Root, Stem

INTRODUCTION

Heavy metal pollution can be absorbed by plants from polluted soil and translocated from one organ to another [1,2]. The root was once an organ that absorbed heavy metal ions via the apoplast pathway [2]. Ions of heavy metals will flow through the wall cell [3]. Plant responses to heavy metal toxicity in the environment have been linked to growth, differentiation, and physiology, including photosynthesis, food absorption, and ion transport [4]. Heavy metals are absorbed from the soil by hyperaccumulator plants through their roots. Heavy metal can thus be kept exclusively in the roots or transported to other organs via xylem [5].

Mercury toxicity has a significant impact on plant physiology, such as mineral absorption and distribution [6]. Heavy metals such as Cadmium (Cd), Chromium (Cr), Mercury (Hg), and lead caused cellular damage in the root and stem of *Boerhavia diffusa* (scratches) that grew in media containing heavy metals (Pb). Heavy

metal ions would be immediately exposed to the roots of plants cultivated in the heavy metal-containing mediums. Root growth will be disrupted as a result of cellular damage, cell wall thinning, and brittle root texture [7].

Abdussalam *et al.* (2015) reported that mercury can cause a decrease in size and cells in the roots, as well as damage to the cortex. Plants were grown in a medium containing mercury showed a reduction in sclerenchyma and smaller cells with thick walls. Parenchyma tissue on the stem will be damaged or the cell wall lysis due to the formation of pits. In addition, treatment with other heavy metals such as lead causes damage to the secondary phloem and sclerenchyma tissue.

All the plants are believed to accumulate and translocate metals in various quantities. However, there are some plants that can accumulate high concentrations of metals in the soil, called hyperaccumulator plant [8]. Some plants that can absorb mercury are *Jatropha curcas* [4], *Amaranthus spinosus* [9], *Canna sp.* [10], *Eichhornia crassipes* [11], *Ipomoea cornea* [12], and *Ipomoea Aquatica* [13]. The genus *Ipomoea* generally are plants that are able to absorb and are tolerant of heavy metals [14].

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Ipomoea reptans are able to accumulate heavy metals such as iron and aluminum [15]. *Ipomoea reptans* are suspected able to translocate mercury from the roots to other organs such as stems and leaves. In addition, *I. reptans* roots structure changes caused by the response to mercury [16]. This research was conducted to observe the stems and leaves of *I. reptans* that are caused by mercury toxicity response.

METHODOLOGY

Planting *Ipomoea reptans* in the Greenhouse

Ipomoea reptans seeds soaked in distilled water for 4 hours, then planted in polybags 10x15). Furthermore, *I. reptans* seed watered once a day using distilled water. After a week of *I. reptans* seeds ready to be moved to a treatment medium containing 2 kg of compost soil that has been contaminated with a mercury concentration of 0001 ppb (control), 61 871 ppb (P1), 92 258 ppb (P2), and 107 046 ppb (P3). And the media was sprinkled with 1 g of urea, Triple Super Phosphate (TSP) 0.5 g, and Potassium Chloride (KCl) 0.5 g) and left for 2 days before planting (Ershad *et al.*, 2014). One treatment pot planted five seedlings of *I. reptans*. The treatment was done for 27 days.

Observations cell structure *Ipomoea reptans*

Ipomoea reptans were harvested then cleaned with distilled water. Furthermore, each organ such as primary taproot (the middle part), stems (the middle part), and leaves crosswise. The cross-section of the roots, stems, and leaves is done at the same place on every *Ipomoea reptans*. The slice placed on the object-glass previously been poured safranin % [17]. Then, preparations were closed with a cover glass and observed using a microscope. *Ipomoea reptans* cell observation using Microscope Swift M100 Series at the Laboratory of Plant Anatomy, University of Syiah Kuala.

RESULTS AND DISCUSSION

Mercury Levels influence on the Roots *Ipomoea reptans*

Ipomoea reptans root preparation shows the change in the vascular tissue (Figure 1). The root preparation of *I. reptans* control shows the regular vascular tissue, while the incision root of *I. reptans* grown on media treatment content with mercury shows the shape of irregular vascular tissue. The plant treated with a higher concentration of mercury in the soil causes damage to root tissues, cause damage on the vascular of *I. reptans* root. Damaging root tissue is caused by

the distribution of mercury ions from the root to another organ of *I. reptans*. The distribution of mercury ions passing through the vascular tissue, such as the xylem.

Because plants do not require mercury, there is no specialized pathway for mercury transfer across plant cell membranes. Mercury in the growth media, on the other hand, can accumulate in plant cells via important cation transporters. Heavy metal ions are absorbed actively or passively by cell membranes and can bind to micronutrient cations in plant root cell membranes. The high affinity for binding sites and transport mechanisms that accumulate in the plasma membrane allows heavy metal ions to bind to the cell membrane. The symplastic theory, in which ions enter the cell through plasmodesmata, is the most possible pathway for ions to enter the cell from the epidermis to the endodermis and then to the xylem [18].

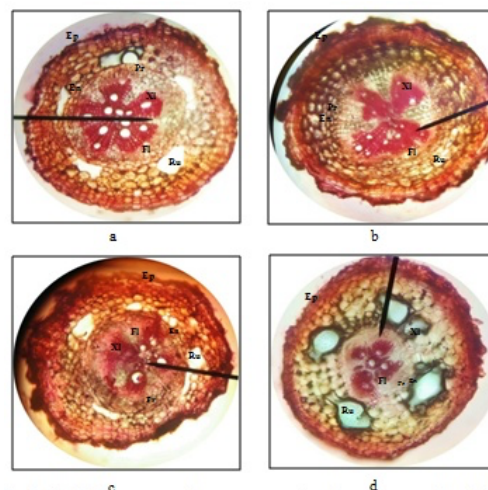


Figure 1. A cross-section incision *I. reptans* root: a. at 0001 ppb treatment (negative control); b. in the treatment of 61 871 ppb (P1); c. in the treatment of 92 258 ppb (P2); d. in the treatment of 107 046 ppb (P3) (Description: Ep: epidermis; Pr: parenchyma; En: endothermic; Fl: phloem; XI: Xylem; Ru: air cavity). (10x10)

Root tissue damage is caused by the distribution of mercury ions from the roots to the other organ. The distribution of mercury ions passing through the vascular tissue, xylem. In this experiment, the mercury concentration in root, stem, and leaf was measured with the AAS method. The highest mercury concentration was detected in the root of the third treatment (P3), which was 2,915 ppb, while the lowest mercury concentration was detected in the control medium (P0) with a mercury level of 0 ppb.

Xylem is a tissue that transports water and nutrients from the soil. In addition, there are substances dissolved in it. Xylem is able to transport the mercury to the stems and leaves, so it caused damage at the xylem tissue of *I. reptans* root that grows in media with mercury (Figure 1: b, c, d). Mercury that is accumulated at the xylem can cause stress for the plant. Heavy metal, like mercury, affects to plant metabolism process. Heavy metal also can damage the essential element at the membrane cell [19].

Mercury in *Boerhavia diffusa* has caused damage to the cortex, the reduction of root cell size, and the cell roots becoming thin and brittle [20]. This is caused by the root is the organ that directly absorbs mercury ions from the soil. But at the root of mercury-contaminated *I. reptans* shows no brittleness of the roots. This is thought to occur because each plant has a different response to metals such as mercury.

Effect of Mercury Levels in Batang *Ipomoea reptans*

The results at stem cross-section *I. reptans* grown on media control and media treatment with different mercury concentrations do not show damage to both the vascular tissue and the other tissue (Figure 2). The stem is an organ that is just passed by mercury ions and only a small amount is accumulated in the tissues of the stem. So that it does not cause changes in stem tissue [21].

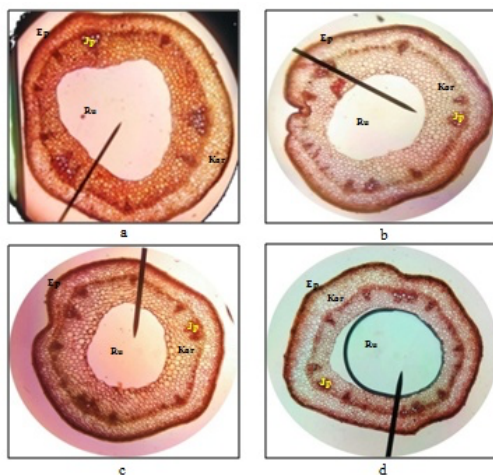


Figure 2. The cross-section incision *I. reptans* rod: a. at 0001 ppb treatment (negative control); b. in the treatment of 61 871 ppb (P1); c. in the treatment of 92 258 ppb (P2); d. in the treatment of 107 046 ppb (P3) (Description: Ep: epidermis; Cr: cortex; Jp: Tissue carrier; Ru: air cavity). (10x10)

Usually, plants store metal ion like mercury in the root cell to avoid the toxic effect. But sometimes mercury ion can be translocated by

xylem to stem. It causes mercury concentration at the stem is less than at the root. The root is an organ that most accumulates mercury [22]. However, Hussain (2010) reported mercury translocated to the stem *B. diffusa* resulting in its reduced size hypodermic cells of the epidermis and cells, as well as the modification of epidermal cells into the form of trichomes.

Effect of Mercury Treatment in Leaves *Ipomoea reptans*

The accumulation of mercury can cause changes in the structure of the leaf tissue. The results showed changes in the bulliform cell on the leaves of *I. reptans*. The results showed changes in bulliform cells of *I. reptans*. Bulliform cells in control leaf sections generally appear to be fewer than in leaves grown on treatment media (Figure 3).

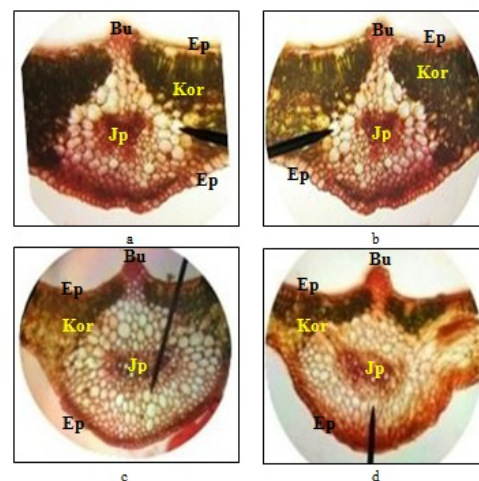


Figure 3. A cross-section incision leaves of *I. reptans*: a. at 0001 ppb treatment (negative control); b. in the treatment of 61 871 ppb (P1); c. in the treatment of 92 258 ppb (P2); d. in the treatment of 107 046 ppb (P3) (Description: Ep: epidermis; Cr: cortex; Jp: Tissue carrier; Bu: Bulliform). (10x10)

The decline of the bulliform size on leaves is suspected to be a strategy to minimize water evaporation during transpiration. It is also related to increases turgor capabilities in leaves that adapt to survive in the polluted area. This is what ultimately makes the bulliform cells in the leaves that planted on media treatment are smaller than the bulliform size at control media. Haryati *et al.* (2012) describe one of the toxic effects of heavy metals in plants as the difficulty of the roots absorbing water from the soil because they influence the osmotic pressure, and

therefore contributes to a decrease in transpiration rate. Melo *et al.* (2007) reported that bulliform cell size difference caused by respon to water deficit and pollution. Decreasing size of the bulliform cause curly leaves. This is can be a strategy for the plant to reduce the surface for transpiration.

The tissue structure in *Brachiaria decumbens* leaves (grass malela) changed as a response to heavy metal toxicity. Changes show in the cells of the endodermis that looks a lot on leaves contaminated by heavy metals. Changes also show in tissue that appears thicker sclerenchyma on leaves contaminated with heavy metals with a higher concentration [23].

CONCLUSION

The plants frown in mercury treatment plots shows vascular tissue damage in roots, causing damage on the vascular of *I. reptans* root. At the stem cross-section, *I. reptans* grown on media control and media treatment with different mercury concentrations do not show damage to both the vascular tissue and the other tissue. Bulliform cells at the control leaves generally seemed to amount more than in leaves grown on media with mercury.

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REFERENCE

- [1] Baker, A. J. M.; McGrath, S. P.; Sidoli, C. D. M; Reeves, R. D. 1994. *Resour. Conserv. Recycl.* **11** 41-49.
- [2] Bell, P. F.; Chaney, R. L.; Angle, J. S. 1991. Free metal activity and total metal concentrations as inducers of micro nutrient availability to barley (*Hordeum vulgare* L.) klages. *Plant. Soil.* **130** 51-62.
- [3] Haryati. M.; Purnomo, T.; Kuntjoro, S. 2012. Kemampuan tanaman genjer (*Limnocharis Flava* (L.) Buch.) menyerap logam berat timbal (Pb) limbah cair kertas pada biomassa dan waktu pemaparan yang berbeda. *LenteraBio.* **1** 131-138.
- [4] Orcutt, D. M.; Nilson, E. T. 2000. *Physiology of plants under stress: soil and biotic factors.* (New York: John Wiley and Sons Inc).
- [5] Prasad, M.N.V. 2003. Phytoremediation of metal-polluted ecosystems: hype commercialization. *Russ. J. Plant Physiol.* **5** 686-700.
- [6] Beauford, W.; Barber, J.; Barringer, A. R. 1977. Uptake and distribution of mercury within higher plants. *Physiol. Plant.* **39** 261-265.
- [7] Abdussalam, A. K.; Ravindran, C. P.; Ratheesh, C. P.; Azeez, K.; Nabeesa, S. 2015. Physiological effects of heavy metal toxicity and associated histological changes in *Boerhavia diffusa* L. *J. Glob. Biosci.* **4** 1221-1234.
- [8] Wise, D. L.; Trantolo, D. J.; Cichon, E. J.; Inyang, H. I.; and Stottmeister, U. 2000. *Bioremediation of contaminated soils.* (New York: Marcel Dekker Inc).
- [9] Kardievel, K.; Jegadeesan, M. 2014. Mercury and cadmium accumulation in selected weed plants: implication for phytoremediation. *AJPSKY.* **5** 1-4.
- [10] Melo, H. C.; Castro, E. M.; Soares, A. A.; Melo, L. A.; Alves, J. D. 2007. Anatomical and physiological alteration in *Setaria anceps* stapf ex massey and *Paspalum paniculatum* L. under water conditions. *Hoehnea.* **34** 145-153.
- [11] Bhardwaj, R.; Thukral, A. K.; Garg, S. K. 2011. Phytoremediation of mercury using *Eichhornia crassipes* (Mart.) Solms. *Int. J. Environ. Waste Manag.* **8** 92-105.
- [12] Komal, T.; Mustafa, M.; Ali, Z.; Kazi, A. G. 2014. Heavy metal adaption strategies and repair mechanism in plants. *Endocytobiosis Cell Res.* **25** 33-41.
- [13] Wise, D. L.; Trantolo, D. J.; Cichon, E. J.; Inyang, H. I.; Stottmeister, U. 2000. *Bioremediation of contaminated soils.* (New York: Marcel Dekker Inc).
- [14] Suhendrayatna; Marlina, H.; Zaki, M.; Elvitriana. 2015. Uptake of mercury ion in a waterland plant *Canna* sp.. The 5th AIC Conference; Banda Aceh: Syiah Kuala University; p. 1-5.
- [15] Elvira, T. H.; Isa. S.; Suleman, N. 2012. Fitoremediasi pada media tanah yang mengandung Cu dengan tanaman kangkung darat. *Sainstek.* **6** .
- [16] Setia, R. C.; Bala, R. 1994. Anatomical changes in root and stem of wheat (*Triticum aestivum* L.) in response to different heavy metals, *Int. J. Plant Morphol.* **44** 95-104.
- [17] Hussain, K. 2010. *Bacopa monnieri* L. Pennell- a good biomarker of water

- pollution/contamination. *J. stress Physio. Biochem.* **6** 91-101.
- [18] Jean-Philippe, S. R. 2010. The effect of mercury contamination on tree, fungal, and soil composition along east folk poplar greek, anderson and roane countries, tennessee. *Doctoral Dissertation.* (Knoxville: University of Tennessee).
- [19] Tchounwou, P. B.; Yedjou, C. G.; Patlolla, A. K.; Sutton, D. J. Heavy metals toxicity and the environment. *PMC.* **101**: 133–164.
- [20] Abdussalam, A. K.; Ravindran, C. P.; Ratheesh, C. P.; Azeez, K.; Nabeesa, S. 2015. Physiological effects of heavy metal toxicity and associated histological changes in *Boerhavia diffusa* L, Journal of Global Biosciences. **4** 1221-1234.
- [21] Nasir, M.; Tauhida, N.; Harnelly, E.; Bahi, M. 2017. Mercury uptake and effect on growth of *Ipomoea reptans*, The 7th AIC-ICMR on Helath and Life Sciences Conference; Banda Aceh: Syiah Kuala University; p. 369-374.
- [22] Gomes, M. P. 2011. Ecophysiological and anatomical changes due to uptake and accumulation of heavy metal in *Brachiaria decumbens*. *Sci. Agric.* **68** 566-573.