Effectiveness of *Ludwigia adcendens* and *Ludwigia grandiflora* as Cadmium (Cd) Phytoremediator

Fida Rachmadiarti[1], Marátus Sholikah[2]
{fidarachmadiarti@unesa.ac.id[1]}

Universitas Negeri Surabaya, Surabaya, Indonesia[1,2]

Abstract. Ludwigia adcendens and Ludwigia grandiflora are water weeds rapidly grow and disperse. The root of these plants can take nutrients up from waters, filter and absorb dissolved solids, and as growth base of microbes. The objective of this study was to test the efficiency L. grandiflora and L. adcendens in remediating cadmium (Cd). These plants were acclimatized first in hydroponic for 14 days, then moved to medium with varying treatments; only L. grandiflora, only L. adcendens and combination of both species at different Cd concentration (0, 5, 10, and 15 ppm). Level of Cd was analyzed at 7th and 14th day using Atomic Absorption Spectrophotometer (AAS), while wet biomass of each group was compared. Data collected was analyzed statistically. Efficiency as Cd phytoremediator from low to high was found to be L. grandiflora, L. adcendens > combination of both species, in which bioconcentration factor (BCF) > 1 and translocation factor (TF) < 1. Wet biomass growth also supported these plants as Cd phytoremediator; plants with higher Cd accumulation had yellow leaves compared to plants accumulated lower Cd level. Concentration of Cd was also found to be higher in the roots than in plant leaves.

Keywords: Ludwigia adcendens, Ludwigia grandiflora, BCF, TF, Cadmium

1 Introduction

Ludwigia grandiflora is tropical plant species generally found floating in the water and belongs to family Onagraceae [1]. It grows on the surface of the water body, such as lakes, ponds, water channels, and sewers. This plant was found to have a high tolerance for salinity [2]. In addition, *L. grandiflora* could absorb heavy metals contained in its growth medium and accumulate both cadmium (Cd) and lead (Pb) [3]. The other *Ludwigia* species, *L. adcendens* share the same floating characteristics and are known as vine water flower, locally named *tapak dara air*. This plant is mainly found in ponds, swamps, and water channels. This species can be determined from its characteristics; herbs with greenish-white round stem, spreading leaves, inverted egg-shaped leaf blade, and stipules in every branching petiole [4]. Plant roots were found to be able to remove nutrients from water, filter and absorb suspended solids, and as a growing base of microbes. *L. adcendens* could also take up various heavy metals, such as copper (Cu) in the water body, thus it can be applied for phytoremediation [5].

As both were found to have the ability to absorb inorganic pollutants, especially heavy metals, then phytoremediation of both species needed to be tested as individual species or consortium. This is especially important as in recent years, industrial growth in Indonesia causes waste disposal to contaminate the water body, especially rivers near the industrial area. The

MSCEIS 2019, October 12, Bandung, Indonesia

DOI 10.4108/eai.12-10-2019.2296356

rapid growth of industrial activity also causes a negative effect on humans and their environment, as industrial waste disposed to waters caused the highest danger to organisms [6]. Dangerous industrial waste contains various heavy metals (Hg, Cu, Cd, and Pb) above the allowed standard.

Cadmium (Cd) is one of heavy metals contained in liquid byproduct waste of pulp and paper industry. Cadmium affects humans in the long-term as it accumulates in the body, mainly in the liver and kidney. Based on BKLH, the maximum threshold allowed for Cd is 0.01 ppm. Natural cadmium is silvery-white soft metal (cuticle) easily oxidized in the open air and by ammonia gas [7]. One of the methods can be conducted to reduce the level of this pollutant is via phytoremediation.

Phytoremediation can be applied to stabilize, extract, degrade, or vaporized contaminants to reduce pollution level, either via the plants themselves or related microbe species symbionts. Plants used for efficient pollutant extraction should have various specific characteristics, such as rapid growth, rapid biomass increase, high tolerance and live capacity [8,9]. *L. grandiflora* and *L. adcendens* are plant species with such characteristics. The previous study showed that *L. adcendens* was able to adapt to heavy metals Pb stress at 11 mg/kg with the translocation factor (FT) detected at 1.35 [10].

Based on the elaboration, this study was planned to test the efficiency of *L. grandiflora* and *L. adcendens* to remediate Cd at various concentrations as individual species or as combined as a consortium.

2 Methods

Acclimatization. *Ludwigia grandiflora* and *Ludwigia adcendens* plants were collected from wetlands in Surabaya, East Java. Both species were acclimatized first by growing them in a hydroponic system using a batch method in Hoagland's solution medium for seven days.

Cd Exposure. After the acclimatization period, plants were rinsed using distilled water and put 100 gram plants into glass aquarium sized 40x30x35 cm contained 5 L distilled water and Hoagland's solution. Into the medium, the various concentration of $CdCl_2$ was added; 0 ppm as control, 5 ppm, 10 ppm, and 15 ppm. Plants were exposed to Cd either for 7 or 14 days. Treatments in the current study were arranged based on a randomized block design with three replications.

Plant Extraction. Plants were harvested from each aquarium after 14 days. Plants were then separated into two: roots and leaves part. Biomass yield was measured based on the dry weight. The respective part of the plant was oven-dried for 2 hours at 80°C successively, then weighed. The dried plant was then crushed into powder. As much as 0.5 g of each powdered sample was taken and digested with 5 ml HNO3, then diluted to 50 ml with deionized double distilled water. Cadmium analysis was performed using the extraction method. Both medium (50 ml) and digested plant samples were measured of their cadmium level using an atomic absorption spectrophotometer. Total accumulation and partitioning of heavy metals by plants were calculated.

Data Analysis. Bioconcentration factor (BCF) and bioaccumulation factor (BAF) were analyzed from Cd level in plants and medium. Level of BCF for Cd in lower (roots) and upper (leaves) plant systems were calculated according to the formula as proposed by Wang et al. (2007).

$$BCF = C_{roots}/C_{medium} \quad (1)$$

$$BAF = C_{shoots}/C_{medium} \quad (2)$$

in where C_{roots}= Cd concentration in lower system (mg/kg dry weight), C_{shoots} = Cd concentration in upper system (mg/kg dry weight) C_{medium}= Cd concentration in growth medium (mg/L).

Translocation factor (TF) of Cd in plants was calculated to evaluate Cd translocation from roots to shoots using the following equation.

$$TF = BAF_{shoots}/BCF_{roots} \quad (3)$$

Quantitative data was compared statistically using Analysis of variance (ANOVA) and continued to Tukey test if difference was found significant in SPSS ver. 21 (p=0.05).

3 Result and Discussion

Cd level in plants. The pH of the medium was found to be varied, ranged on 6.4–6.7 at the beginning of the experiment, into 6.9–7.2 range at the end. Result of Cd level measurement on plant roots and shoots are presented in Table 1 and Table 2, respectively.

Table 1. Level of Cd in the roots of L. adscendens, L. grandiflora, and combination of both species.

Species	Concentration of Cd (ppm)			
	0	5	10	15
L. adscendens	$0.00{\pm}0.00^{aA}$	$1.68{\pm}0.33^{aB}$	$3.20{\pm}0.39^{aC}$	$3.37{\pm}0.54^{aD}$
L. grandiflora	$0.00{\pm}0.00^{aA}$	$1.56{\pm}0.58^{aB}$	$1.82{\pm}1.00^{aC}$	$3.05{\pm}0.00^{aD}$
L. adscendens +L. grandiflora	$0.00{\pm}0.00^{bA}$	$1.97{\pm}0.00^{bB}$	$4.11{\pm}0.68^{bC}$	$6.29{\pm}2.00^{bD}$

Different small letters indicate significant difference in plant species, while different capital letters indicate significant difference among initial Cd level (p = 0.05).

Table 2. Level of Cd in the shoots of *L. adscendens*, *L. grandiflora*, and combination of both species.

Species	Concentration of Cd (ppm)			
	0	5	10	15
L. adscendens	$0.00{\pm}0.00^{bA}$	$0.06{\pm}0.02^{bB}$	$0.07{\pm}0.01^{bB}$	$0.11{\pm}0.036^{bC}$
L. grandiflora	$0.00{\pm}0.00^{aA}$	$0.03{\pm}0.00^{aB}$	$0.03{\pm}0.00^{aB}$	$0.046{\pm}0.015^{aC}$

L. adscendens + L. grandiflora	0.00±0.00^{cA}	0.06±0.03^{cB}	0.72±0.01^{cB}	0.19±0.05^{cC}

Different small letters indicate significant difference in plant species, while different capital letters indicate a significant difference among initial Cd level (p = 0.05).

The result showed that there was a significant difference in Cd content between control and exposed plants with the highest Cd level was found in roots. Mechanism of metals absorption may include binding of positively-charged toxic metals ion to negative charges in the cell walls [11]. The higher the Cd concentration given at the initial of the experiment, the higher the Cd level absorbed by the roots. This was in line with a previous study in kale [12] and clover exposed to Pb [13,14]. The same thing also occurred in the shoot of plants, but the accumulation of Cd at the initial Cd level of 5 and 10 ppm were relatively similar. Generally, plants accumulate metals in roots and prevent them to move up into plant shoots. This is the internal mechanism of heavy metals inhibition [15].

A consortium of *L. adscendens + L. grandiflora* had better absorption of Cd metals compared to the non-consortium plant. Plant combination was found to be more effective in remediate liquid waste because of synergy between plant species, resulting in more significant absorption [b]. Plant consortium is a collection of several plant types making up a community in nature, so they produce more optimal uptake. This is caused by each plant roots contained different bacteria that interact to raise plant growth [16].

Non-combined treatment showed no significant difference in Cd absorbance in roots of *L. adscendes* and *L. grandiflora* (Table 1). *L. adscendes* was found to be able to lower Cd in water bodies contaminated by Lapindo muds, while *L. grandiflora* has two types of roots; long and short roots [17]. Bouldin *et al.*, (2006) reported that *L. grandiflora* had potential in waste treatment [18]. This was marked by root's ability in absorbing Cd at various concentrations. Both plants were also able to adapt to medium contaminated by Cd at various levels (**Figure 1** and **Figure 2**).

Bioconcentration factor and translocation factor. Bio-concentration factor (BCF) and translocation factor (TF) were noted in the current study to evaluate the capability of plants to extract metals from the water and the aptitude of plants to transfer metals from roots to shoots [19]. Both BCF and TF of *L. adscendens*, *L. grandiflora*, and a consortium of both were presented in Table 3 and 4.

Table 3. Bioconcentration factor (BCF) of Cd in plants.

Species	BCF at Cd concentration			
	0 ppm	5 ppm	10 ppm	15 ppm
L. adscendens	0.00±0.00^{aA}	52.60±14.57^{aB}	47.93±8.69^{aB}	29.46±6.19^{aB}
L. grandiflora	0.00±0.00^{aA}	45.92±2.48^{aB}	23.66±16.21^{aB}	25.61±0.30^{aB}
L. adscendens + L. grandiflora	0.00±0.00^{bA}	66.38±0.38^{bB}	72.20±19.30^{bB}	81.26±48.82^{bB}

Different small letters indicate a significant difference in plant species, while different capital letters indicate a significant difference among initial Cd level (p = 0.05).

Table 4. Translocation factor (TF) of Cd in plants.

Species	Concentration of Cd (ppm)			
	0	5	10	15
L. adscendens	$0.00{\pm}0.00^{A}$	$0.33{\pm}0.01^{B}$	$0.21{\pm}0.00^{B}$	$0.33{\pm}0.12^{B}$
L. grandiflora	$0.00{\pm}0.00^{A}$	$0.02{\pm}0.00^{B}$	$0.02{\pm}0.01^{B}$	$0.01{\pm}0.00^{B}$
L. adscendens + L. grandiflora	$0.00{\pm}0.00^{A}$	$0.28{\pm}0.01^{B}$	$0.17{\pm}0.00^{B}$	$0.03{\pm}0.01^{B}$

Different small letters indicate significant difference in plant species, while different capital letters indicate significant difference among initial Cd level (p = 0.05).

Result showed that TF of the threes plant treatment was less than 1.0, indicating that most of the metal ions were concentrated in roots [20] and that L. adscendens and L. grandiflora functioned as accumulator plants. Plants can be categorized as excluder and accumulator if TF<1 and BCF>1. Proper disposal of harvested plant parts is the final and most important step in any kind of plant-based remediation technology. Higher BCF and TF of tested species enable them to accumulate large amounts of hazardous metals in the harvested parts and if not disposed of properly the accumulated heavy metals may circle back to the ecosystem or enter the food chain.

Fig. 1. Morphology of a) L. adscendens; b) L. grandiflora; c) Combination (L. adscendens and L. grandiflora) at 0th day of Cd exposure.

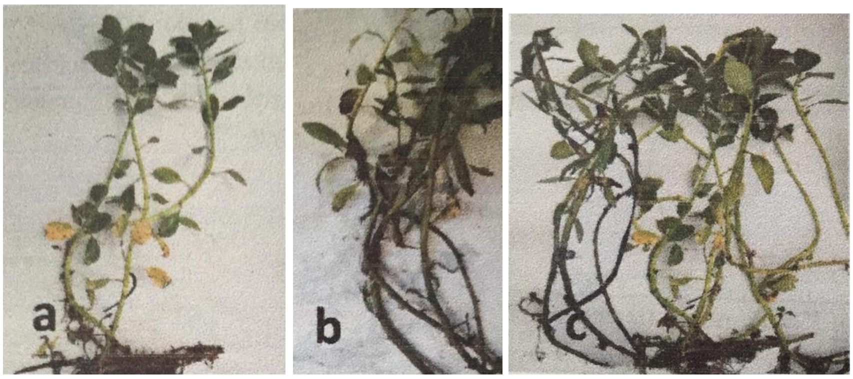

Fig. 2. Morphology of a) L. adscendens; b) L. grandiflora; c) Combination (L. adscendens and L. grandiflora) at 14th day of Cd exposure.

The ability of L. adscendes and L. grandiflora or consortium of both to accumulate Cd affected on their morphology (**Figure 1** and **Figure 2**). Control plants showed normal growth until the end of study. Cd level at 5 and 10 ppm had not yet affected the morphology of L. adscendes and L. grandiflora significantly. Cd concentration of 15 ppm caused slight change of morphology, in which some leaves was found to have chlorosys. L. grandiflora was found to have a faster rate of chlorosys compared to L. adscendens. Heavy metals Cd can induce yellow color to leaves, as a low level of it caused toxicity to plants. Cd can also replace Mg in chlorophyll, resulting in a change of color in leaf and stem of plants [20].

4 Conclusion

Efficiency as Cd phytoremediator from low to high was found to be *L. grandiflora, L. adcendens* > combination of both species, in which bioconcentration factor (BCF) > 1 and translocation factor (TF) < 1. Wet biomass growth also supported these plants as Cd phytoremediator; plants with higher Cd accumulation had yellow leaves compared to plants accumulated lower Cd level. The concentration of Cd was also found to be higher in the roots than in plant leaves.

Acknowledgement. Thank to Firas K,M.Si., Dini Dwi Nilamsari, S.Si. Nur Khalifah, S,Si. for helping the research.

References

[1] Rao D. L.: Heavy metals causing toxicity in humans, animals and environment. Journal of Chemical and Pharmaceutical Sciences, Vol 3. pp. 172–174 (2013)
[2] Hakim L., Sudrajat, Budiman.: Efektivitas Kombinasi Tumbuhan Eceng Gondok dan Ganggang dalam Remediasi Air Limbah Migas. In Seminar SAINS dan Teknologi FMIPA Universitas Mulawarman. 1(1) ISBN 9786027265813 (2015)
[3] Visitacion, R. S., Jesus, J. D., Su, S., & Ragragio, E. L. E. N. A.: Germination and Seedling Growth Responses of Water Spinach (Ipomoea aquatica) and Spinach (Spinacia Oleraceae) at Varying Lead Concentrations. Journal of Applied Technology in Environmental Sanitation.Vol 1(2). pp. 139 – 142 (2011)
[4] Nirmal, K. P., Subhasish, G., & Indrani, C.: Jussiaea Repens L. is a Nontoxic Antigonadal Herb – A Dose-Dependent Study On Male Rats. International Journal Of Pharma And BioSciences. Vol. 4(2). pp. 131 – 143 (2013)
[5] Cheng, S.: Heavy Metals in Plants and Phytoremediation Environmental Science and Pollution Research, Vol. 10(5). pp. 335-340 (2003)
[6] Mahida, U.N.: Pencemaran Air dan Pemanfaatan Limbah Industri. Jakarta: Rajawali Press. (1984)
[7] Palar H.: Pencemaran dan Toksikologi Logam Berat. Bandung: Rineka Cipta (2008)
[8] Morel, J. L., Echevarria, G., & Goncharova, N.: Phytoremediation of Metal-Contaminated Soils. Vol. 86. Netherland: Springer (2006)
[9] Nur, F.: Fitoremediasi Logam Berat Kadmium (Cd). Jurnal Biogenesis. ISSN 2302-1616 Vol 1, No. 1. pp.74-83 (2013)
[10] ripathi, P., Dwivedi, S., Mishra, A., Kumar, A., Dave, R., Srivastava, S., & Tripathi, R. D.: Arsenic Accumulation in Native Plants of West Bengal, India: Prospect For Phytoremediation but Concern with The Use of Medical Plants. Environ Monit Assess. Vol. 184. pp.2617-2631 (2012)
[11] Grill, E., Winnacker, E. L., & Zenk, M. H.: Phytochelatins, The Principal Heavy Metal Complexing Peptides of Higher Plants. Science (Washington, DC). Vol. 230(4726). pp. 674-676 (1985)
[12] Rachmadiarti, F., Soehono, L. A., Utomo, W. H., Yanuwiyadi, B., & Fallowfield, H.: Resistance of Yellow Velvetleaf (Limnocharis flava (L.) Buch.) Exposed to Lead, J. Appl. Environ. Biol. Sci., Vol. 2(6). pp. 210-215 (2012)
[13] Rachmadiarti, F, and Trimulyono, G.: The efficacy of salvinia molesta mitch. and marsilea crenata presl. as phytoremediators of lead pollution. Journal of Applied Horticulture Vol. 20(1) (2018)
[14] Rachmadiarti, F, and Trimulyono, G.: Phytoremediation capability of water clover (Marsilea crenata (L). presl.) in synthetic PB solution.Applied Ecology and Environmental Research vol. 17(4). pp. 9609-9619 (2019)
[15] Göthberg, A., Greger, M., Holm, K., & Bengtsson, B. E.: Influence of Nutrient Levels on Uptake and Effect of Mercury, Cadmium and Lead in Water Spinach. Journal of Environmental Quality. Vol, 33(4). pp. 1247-1255 (2004)
[16] Aiman, U., Tantriati, T., & Sriwijaya, B.: Pemberian Macam Konsorsium Bakteri Hasil Isolasi Tumbuhan Pantai pada Kangkung (Ipomea reptans Poirs.). Journal Agro Science. Vol. 5(1). pp. 1-6 (2017)
[17] Hathaway, S.: Ludwigia grandiflora (Michx) Greuter & Burdet A Guide to Identification, Risk Assessment and Management. Plant Protection Service, Wageningen, NI. Centre for Ecology and Hydrology-Wallingford, UK (2011)
[18] Bouldin, J. L., Farris, J. L., Moore, M. T., Smith Jr, S., & Cooper, C. M.: Hydroponic Uptake of Atrazine and Lambda-Cyhalothrin in Juncus effuses and Ludwigia peploides. Chemosphore. Vol. 65(6). pp. 1049-1057 (2006)
[19] Yoon, J., Cao, X., Zhou, Q., & Ma, L. Q.: Accumulation of Pb, Cu, and Zn in Native Plants Growing on A Contaminated Florida Site. Sci. Total Environ. Vol. 368(2-3). pp. 456–464 (2006)
[20] Rosidah, S.: Uji Toleransi Tanaman Tembakau (Nicotiana tabacum L.) terhadap Cekaman Kadmium (Cd), Timbal (Pb) dan Tembaga (Cu) pada Kultur Cair. Universitas Negeri Semarang (2014)