



# Bioaccumulation of cadmium and thallium in Pb-Zn tailing waste water by *Lemna minor* and *Lemna gibba*



Merve Sasmaz<sup>a,1</sup>, Erdal Öbek<sup>b,2</sup>, Ahmet Sasmaz<sup>c,\*,3</sup>

<sup>a</sup> Firat University, Environmental Engineering, 23119, Elazığ, Turkey

<sup>b</sup> Firat University, Bioengineering, 23119, Elazığ, Turkey

<sup>c</sup> Firat University, Geological Engineering, 23119, Elazığ, Turkey

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## ABSTRACT

The present study investigated the removal ability to phytoremediate cadmium and thallium from tailing waste water of *Lemna gibba* and *Lemna minor*. These plants were separately adapted to the reactors, placed in the water and daily collected during the eight days. During the study, the plant and water samples were taken daily and the pH, temperature and electric conductivity of the tailing waste water were daily measured in situ. *L. minor* and *L. gibba* were firstly washed, dried in and then ashed at 300 °C for 24 h in an oven. Both ashed plant and water samples were analyzed by ICP-MS to find out the concentrations of cadmium (Cd) and thallium (Tl). Although Cd and Tl are at low values ( $11.4 \pm 0.5 \mu\text{g L}^{-1}$  for Cd and  $2.85 \pm 0.5 \mu\text{g L}^{-1}$ ) in tailing waste water, the Cd and Tl were accumulated at the highest amounts by *L. minor* ( $31.08 \text{ mg L}^{-1}$  for Cd and  $13.43 \text{ mg L}^{-1}$  for Tl) and *L. gibba* ( $38.9 \text{ mg L}^{-1}$  for Cd and  $17.18 \text{ mg L}^{-1}$  for Tl). Our study on the fourth day showed that *L. minor* accumulated more removal abilities of Cd (94.56 times) and Tl (7.33 times) than in *L. gibba* L. (25.89 times on the third day for Cd and 6.16 times on the fourth day for Tl) but *L. gibba* accumulated higher Cd and Tl concentrations ( $38.9 \text{ mg Cd kg}^{-1}$  and  $17.18 \text{ mg Tl kg}^{-1}$ ) than in *L. minor*. Therefore, these plants can use to remove Cd and Tl in tailing waste water polluted by Cd and Tl.

## 1. Introduction

Toxic effects of heavy metals (HM) such as Cd, As, Hg, Tl, Zn and Pb. have been worked their effects on human health by US EPA (United States Environmental Protection Agency), OECD (The Organization for Economic Cooperation and Development) and WHO (World Health Organization) (Jarup, 2003; Kabata-Pendias and Mukherjee, 2007; Liu et al., 2008). Contamination of aquatic systems/environments by heavy metal contamination is one of the main global problems for all people in the world. Cadmium and thallium occur naturally in ore deposits together with lead, zinc, silver and copper release into the soil and water from different sources such as fuel production, smelting processes, industrial effluents, mining, agricultural chemicals, small-scale industries (OECD, 2003; Babarinde et al., 2016; Chidi and Kelvin, 2018; Siddique et al., 2018; Mehta et al., 2018). Cadmium and thallium accumulate firstly in the kidney and has a long biological half-life in living human and animals. According to WHO (2006), Cd and Tl are among the

highest toxic metals in compared to the other heavy metals. Cadmium toxicity can result in kidney failure and chronic renal failure (Gobe and Crane, 2010; Bawaskar et al., 2010; Płachno et al., 2015). Thallium enters to human body with food, vegetables and water and causes fetal demise, degenerative changes and adverse health effects in living organs (Hoffman, 2000; Cvjetko et al., 2010; Karatepe et al., 2011).

Water resources are rapidly contaminated by human activities over recent years. The HM levels in drinking water of different countries are higher than in WHO (2006)'s levels. The HM contamination in aquatic areas is one of the biggest global problems for some countries (USEPA, 2000; OECD, 2003; Bulut et al., 2016; Demir et al., 2017; Aras et al., 2017; Koç Orhon et al., 2017; Solak et al., 2018; Muhammetoglu et al., 2018).

Phytoremediation is fairly cheap and has eco-friendly technology in compared with the different techniques used to remove the heavy metals (Obek, 2009; Chandra and Yadav, 2011; Sood et al., 2012; Tatar and Obek, 2014; Sasmaz et al., 2015, 2016a, 2016b and 2018). Floating

\* Corresponding author.

E-mail addresses: [msasmaz91@hotmail.com.tr](mailto:msasmaz91@hotmail.com.tr) (M. Sasmaz), [eobek@firat.edu.tr](mailto:eobek@firat.edu.tr) (E. Öbek), [asasmaz@gmail.com](mailto:asasmaz@gmail.com), [asasmaz@firat.edu.tr](mailto:asasmaz@firat.edu.tr) (A. Sasmaz).

<sup>1</sup> Tel: +904242370000/5979; fax: +904242411226.

<sup>2</sup> Tel: +904242370000/5554; fax: +904242411226.

<sup>3</sup> Tel: +904242370000/5950; fax: +904242411226.

duckweeds rapidly uptake both the contaminants and metals with the rhizofiltration process (Chaudhuri et al., 2014). *Lemna* sp. has been worked often by many scientists for phytoremediation studies due to its easier harvest, faster growth and float, long storage volume, the possibility of transport, low cost, the minimization of chemical or biological sludge capacities (Zayed et al., 1998; Dirilgen, 2011; Khataee et al., 2012). Although duckweeds have the fastest reproduction rates and grow under different climatic conditions (Materazzi et al., 2012). Khellaf and Zerdaoui (2009) detected that the highest growth ratios of *Lemna* sp. were at 21 °C and pH 6 to accumulate Cd and Cu in the aquatic areas.

This study was focused to determine Cd and Tl concentrations in tailing waste water and aquatic plants growing around the Keban village, to detect daily the changes of Cd and Tl levels in both plants, to calculate the accumulation performance of these plants in tailing waste water for Cd and Tl, to find the ideal harvesting time for Cd and Tl, to compare the phytoremediation abilities for Cd and Tl.

## 2. Materials and methods

### 2.1. The study area

Keban mining area is one of the biggest lead and zinc deposits in Turkey (Fig. 1) and related to the syenomonzonitic and syenitic rocks (Akgul, 2015). Cd and Tl contents of these mineralizations contain quite high. The Keban region has generally Pb-Zn-Ag, F-Mo, Fe-Cu, Mn-Ag and W ores. Zn-Pb economic ores including Ag, Cd and Tl, and mined about 6000 years (Seeliger et al., 1985). Therefore, these galleries were used to produce the economic Pb-Zn ores in the historical period and closed nowadays because of unsafe of galleries in this district.

### 2.2. Water and plant samples

The chemical compositions of the tailing waste water depend on ore types and the composition of geologic units. This can also affect the electric conductivity (EC), temperature (T °C) and pH of the tailing waste water. The waters were daily taken with sterilized and clean

plastic bottles from running tailing waste water. Both the plant and water sampling were carried out during eight days. Then, the EC, T and pH of the tailing waste water measured in situ in the field. In the same time, *L. gibba* and *L. minor* daily picked up daily from reactors. It was measured methodically the temperature (a traceable digital thermometer), pH (Gel-filled pH electrode) and EC (an Orion conductivity electrode) of the tailing waste water. The studied plants according to Davis (1984) were identified as *L. gibba* and *L. minor*.

### 2.3. Sample preparation

*L. minor* and *L. gibba* were collected from the pools of the Istanbul University Botanic Park, were grown in natural pools and different reactors during 2 weeks as described by Tatar and Obek (2014) and Sasmaz et al. (2015). The 400 g of *L. minor* and *L. gibba* were put in its place to each reactor, separately. Keban tailing waste water has a sustained flow regime about  $2.85 \text{ L s}^{-1}$ . *L. gibba* and *L. minor* during the experiment were daily collected separately about 50 g from the each reactor. They were washed firstly with distilled water and after that dried in room temperature for 24 h. The dried *L. minor* and *L. gibba* were ashed for 24 h at 300 °C, finally, it was digested in  $\text{HNO}_3$  (Merck, Darmstadt, Germany) for one hour, a mixture of  $\text{HCl}:\text{HNO}_3:\text{H}_2\text{O}$  for one hour at 95 °C. The digests were analyzed for Cd and Tl by the inductively coupled plasma mass spectrometry (ICP-MS; A Perkin-Elmer Elan 9000) (Group SO200 was used for water samples and Group VG104 was used for ashed plant samples).

### 2.4. Accumulation potential (%) of *L. minor* and *L. gibba*

The Cd and Tl accumulation potentials for *L. gibba* and *L. minor* were calculated by the following formula. The accumulation potential of Cd or Tl for the second day in *L. gibba* =  $(\text{LG2}-\text{LG0})/\text{LG0}$ ; the accumulation potential of Cd or Tl for the second day in *Lemna minor* =  $(\text{LM08}-\text{LM0})/\text{LM0}$  (Sasmaz et al., 2018).

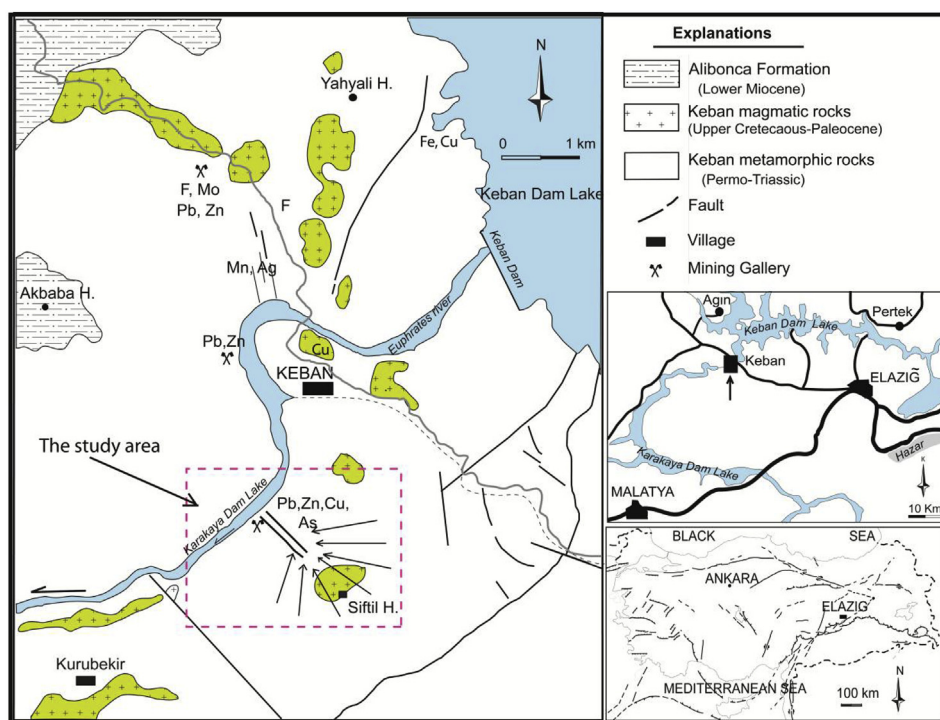


Fig. 1. The geology map of the investigated area (Akgul, 2015).

**Table 1**  
The detection limits and results ( $p < 0.5$ ) of the anion and cations analyzed in ICP-MS and physicochemical characteristics of tailing waste water.

Parameter	T (°C)	pH	EC (mS cm <sup>-1</sup> )	HCO <sub>3</sub> <sup>-</sup> (mg L <sup>-1</sup> )	NO <sub>3</sub> <sup>-</sup> (mg L <sup>-1</sup> )	SO <sub>4</sub> <sup>-</sup> (mg L <sup>-1</sup> )	F <sup>-</sup> (mg L <sup>-1</sup> )	Ca (mg L <sup>-1</sup> )	Mg (mg L <sup>-1</sup> )	K (mg L <sup>-1</sup> )	Na (mg L <sup>-1</sup> )
DL	–	–	–	–	–	–	–	0,05	0,05	0,05	0,05
Tailing waste water	19.7 ± 1	7.36 ± 0.2	2.29 ± 0.1	370 ± 10	9 ± 1	1280 ± 20	0.26 ± 0.1	780 ± 10	260 ± 10	16 ± 1	140 ± 5
	Fe (µg L <sup>-1</sup> )	Mn (µg L <sup>-1</sup> )	S (µg L <sup>-1</sup> )	P (µg L <sup>-1</sup> )	B (µg L <sup>-1</sup> )	Cu (µg L <sup>-1</sup> )	Pb (µg L <sup>-1</sup> )	Zn (µg L <sup>-1</sup> )	As (µg L <sup>-1</sup> )	Cd (µg L <sup>-1</sup> )	Tl (µg L <sup>-1</sup> )
DL	10	0,05	1	1,5	5	0,1	0,1	0,5	0,5	0,5	0,01
Tailing waste water	590 ± 30	820 ± 30	580 ± 15	65 ± 3	195 ± 7	67 ± 2	7.5 ± 1	7230 ± 30	96 ± 5	11.4 ± 0.5	2.85 ± 0.5

## 2.5. The removal of heavy metals in the water

This value was calculated by the following formula described by Sasmaz et al. (2018), the removal ratio for second day of Cd ( $Cd = LG2-LG0/Cd_{water} = 13430-620/11.4 = 1124$  Lt) and Tl ( $Tl = LG2-LG0/Tl_{water} = 4910-860/2.85 = 1421$  Lt) concentration in the gallery water ( $\mu g L^{-1}$ ) (Sasmaz et al., 2018).

## 2.6. Statistical analysis

The statistical analyses of the studied samples were carried through by using Statistica version 6.0. The Cd and Tl values of *L. minor* and *L. gibba* were correlated with Mn, Mg, Al, Fe, Na, K, P and S by using the Spearman Rank correlation (Table 2).

## 3. Results

### 3.1. Cd and Tl concentrations in tailing waste water

Water samples during the 8 day were collected daily in the field. The chemical results of 8 water samples had close concentrations for each metal. The physicochemical characteristics and results of average chemical analysis of tailing waste waters are shown in Table 1. The mean Cd and Tl values were observed to be  $11.4 \pm 0.5 \mu g L^{-1}$  and  $2.85 \pm 0.5 \mu g L^{-1}$ , respectively, in the tailing waste water ( $p < 0.5$ ), as given in Table 1. T (°C), pH and EC values with major ions of the tailing waste waters are shown in Table 1. The pHs of the tailing waste water vary to 7.52 from 7.17; the temperature varies between 19.0 and 21.1 °C; and the ECs range between 2.45 and 2.13 mS cm<sup>-1</sup>. These results are related to the flow and residence time in the mineralized area, the rock– water relationship and the distance to the feeding area. For these reasons, the pH, T, and EC parameters in the Keban tailing waste water were found close values to each other during the all experiments.

According to Piper's (1944), the contents of anion and cation in the Keban tailing waste water pointed three hydrochemical facies. The Ca, Mg, and Na in the study area are dominant cations, and occurred more than 90% of the cation. The major components (88–95%) of this waters are bicarbonate and sulfate. Therefore, the Keban tailing waste water was described as the Ca- Mg- SO<sub>4</sub> bicarbonate waters.

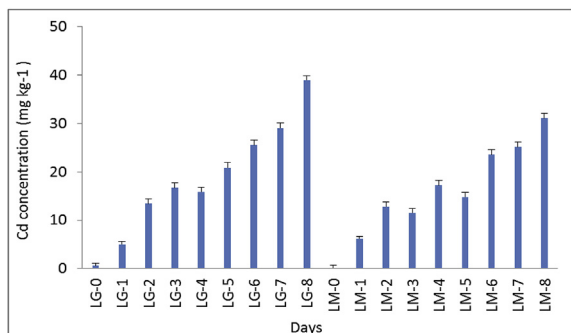
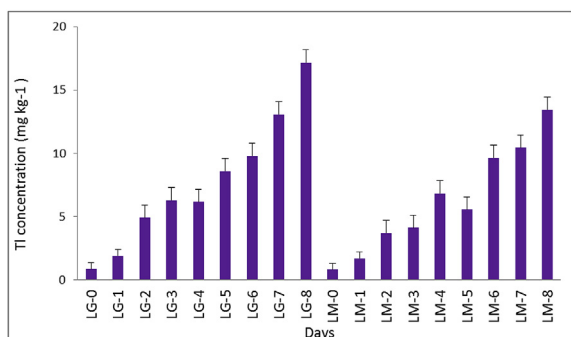
### 3.2. Cd and Tl concentrations in the plants

Cd concentration of (LG-0) *L. minor* and *L. gibba* (LM-0) before the experiment are respectively,  $0.18 mg kg^{-1}$  and  $0.62 mg kg^{-1}$  (Fig. 2). These Cd concentrations have been defined as control samples for these plants. *L. minor* and *L. gibba* on the first day of the experimental study accumulated  $6.19 mg Cd kg^{-1}$  and  $5.05 mg Cd kg^{-1}$ , respectively, on a daily basis. The Cd values accumulated by *L. gibba* increased from 715% on the first day of the environment, to 2066%, 2589%, 2456%, 3268%, 4016%, 4584%, 6174% on the following days. The Cd values accumulated by *L. minor* increased from 3339% on the first day of the environment and to 7022%, 6294%, 9456%, 8106%, 13333%, 13861% and 17167% on the following days. Cd concentrations in both plants were showed regular increases until the eighth day from the first day. As presented in Fig. 2, the highest levels of Cd were seen on the eighth day for both plants. Although this water contains very low levels of Cd (mean:  $11.4 \mu g L^{-1}$ ), *L. gibba* and *L. minor* accumulated, 3357 and 2710 times respectively, higher Cd than in the tailing waste water. *L. gibba* was seen to have the accumulation ability higher Cd than in *L. minor* (Fig. 2). Cadmium in *L. minor* and *L. gibba* ( $p < 0.5$ ) had a high linear in correlated with the Fe, K, Pb, Zn and As in *L. gibba* and *L. minor* (Table 2). This shows that both Cd and other related metals are enriched together within the *L. minor* and *L. gibba* during the experiment.

Tl concentrations of *L. gibba* (LG-0) and *L. minor* (LM-0) before the experiment were respectively,  $0.86$  and  $0.82 mg kg^{-1}$ . These concentration values of LM-0 and LG-0 for Tl values of both plants were

**Table 2**Correlation coefficients among some metals with Cd and Tl in *Lemna minor* and *Lemna gibba* ( $p < 0.5$ ).

	K	Na	Ca	Fe	Al	Mg	P	Mn	S	As	Pb	Cu	Zn
Cd	0,55	−0,02	0,08	0,84	0,28	0,07	−0,30	0,25	0,26	0,87	0,87	0,27	0,98
Tl	0,52	0,02	0,11	0,81	0,33	0,11	−0,33	0,21	0,22	0,84	0,86	0,26	0,98

**Fig. 2.** Cd accumulation in *Lemna minor* and *Lemna gibba* collected daily during the eight days. The bars with different letters among the different stages are significantly different at  $p < 0.5$  by Student's t-test.**Fig. 3.** Tl accumulation changes ( $p < 0.5$ ) in *Lemna gibba* and *Lemna minor* collected daily during the eight days. The bars with different letters among the different stages are significantly different at  $p < 0.5$  by Student's t-test.

defined as control values. *L. gibba* and *L. minor* accumulated  $1.88 \text{ mg kg}^{-1}$  and  $1.68 \text{ mg kg}^{-1}$ , respectively, on a daily basis, from the first day of the experiment. The Tl levels accumulated from this water (average:  $2.85 \mu\text{g L}^{-1}$ ) by *L. gibba* rose to 119% on the first day, 471%, 630%, 616%, 899%, 1037%, 1420%, and 1898% on the following days. The Tl levels accumulated by *L. minor* observed a linear enhancement until the eighth day from the first day, except for the fifth day (Fig. 3); 105% on the first day, 349%, 401%, 733%, 578%, 1076%, 1174%, and 1538% on the following days. Although this water contains very low levels of Tl (mean:  $2.85 \mu\text{g L}^{-1}$ ), *L. gibba* and *L. minor* on the eighth day accumulated 5726 and 4424 times respectively, more thallium than in the tailing waste water. *L. gibba* was showed high performances to accumulate higher Tl than in *L. minor* in compared to the control values of Tl (Fig. 3). Tl had positive correlations with Fe, K, As, Zn, and Pb (Table 2) in *L. minor* and *L. gibba* ( $p < 0.5$ ), but no negative correlations among Tl and other elements, except for phosphorus (P). This shows that both Tl and other related metals are enriched together within the *L. minor* and *L. gibba* during the experiment.

#### 4. Discussion

The average Cd ( $11.4 \mu\text{g L}^{-1}$ ) and Tl ( $2.85 \mu\text{g L}^{-1}$ ) levels of the tailing waste water in the study area are higher than the values (Cd:  $5 \mu\text{g L}^{-1}$ ; Tl:  $2 \mu\text{g L}^{-1}$ ), established for drinking water by the WHO (2011) and USEPA (2000). This also causes environmental

contamination in the soil and water areas along the Firat River. After they have polluted the ground and surface waters around the mining areas, toxic metals are very hard to remove or rehabilitate. These enter the human body with polluted ground and surface waters (Dong et al., 2010). Ning et al. (2011) reported that the heavy metal contents of surface water around the gold mining were higher than in class III-IV quality standards of the national surface water. Heavy metal levels decrease as they move away from sources of pollution in the direction of water flow. Ning et al. (2011) pointed that positive correlations among the Cu-Hg, Cd-Hg, Cd-Cu and Cr-Pb were high values respectively, such as 0.99, 0.97, 0.98 and 0.96. It was determined that the metal concentrations of the ground or surface water changes due to their geochemistry and the possible sources of heavy metals.

Phytoremediation is a very efficient, cost-effective and green method to rehabilitate polluted environments. Different aquatic plants are defined as the indicators plants of the heavy metal contaminations and used as a monitoring of the environmental contamination (Cenci, 2000). Albers and Camardese (1993) detected that some aquatic plants can accumulate the hundred thousand times more than in the concentrations in their associated waters. Over time, the heavy metals (Zn, Cu, Hg, Ag, As, Cr, Pb, Cd, and Tl) are toxic and dangerous for the plant and living animals on the biological environment (Baby et al., 2010).

Saygideger et al. (2005) demonstrated that the dried *Lemna minor* was effectively used in the treatment of waste water containing Cd. However, *Lemna minor* grow in natural environment and it may be another option to more cheap materials. Hou et al. (2007) found that cadmium was more toxic metal than copper on plants and a good candidate for the phytoremediation of waters contaminated with low level metals by *Lemna minor* with advisable harvesting. According to obtained bioconcentration factor values (Uysal and Taner, 2010), *L. minor* can be a good bioaccumulator plant of Cd and a useful plant in treatment systems because it has high uptake ability to heavy metals from water. Uysal and Taner (2010) detected that the plants accumulated the maximum Cd removal at pH 6 and  $25^\circ\text{C}$ , in treatment systems. Furthermore, it showed to have the maximum bioconcentration factor at  $35^\circ\text{C}$  and additional advantageous in tropical regions. Megateli et al. (2009) detected that *L. gibba* has high phytoremediation potential for Cd. Obek (2009) concluded that *L. gibba* had the high removal percent of Cd from contaminated waste waters and indicated that the maximum removal of Cd by *L. gibba* was in the second days and sixth days, reaching 30% and 52%, respectively, for Cd. Duman et al. (2010) pointed that *L. gibba* was a good phytoremediator for the waters contaminated by Cd. Parlak and Yilmaz (2013) showed that the *L. gibba* could be used for the stabilization and rehabilitation of water in wetlands abandoned and polluted by low-level Cd. Chaudhuri et al. (2014) showed that *L. minor* was capable of removing 42–78% and potential cadmium accumulator.

According to Peter and Viraraghavan (2005), there is no more information about the behavior of Tl in fresh water aquatic environments (Kwan and Smith, 1991; Lan and Lin, 2005; Babic et al., 2009; Plachno et al., 2015). Cd and Tl metals were accumulated high performances by *Lemna minor* but the distribution and the absorption of both metals in the plant are very different; Cd absorption is more slowly than that in Tl and under metabolic control. The experiments of desorption and depuration prove that to be within the plasma membrane of a greater accumulation of Tl (80%) (Kwan and Smith, 1991). Lan and Lin (2005) indicated that Tl toxicity was reduced significantly by the formation of Tl due to ultra-trace level applied in the testing solutions. Studies on the



toxicity of Tl in aquatic environments can take more attention in the forthcoming period due to the dominant species ( $\text{Tl}^{+3}$ ) of Tl in surface and ground waters.

## 5. Conclusion

This study was demonstrated to be an effective, green and cheap method for the rehabilitation of waters polluted by Cd and Tl by using *L. gibba* and *L. minor*. Our study proved that *Lemna minor* (94.56 times on the third day for Cd and 7.33 times on the fourth day for Tl) accumulated more Cd and Tl concentrations than *L. gibba* (25.89 times on the third day for Cd and 6.16 times on the fourth day for Tl) with the control samples of *L. minor* and *L. gibba* but *Lemna gibba* accumulated higher Cd and Tl ( $38.9 \text{ mg Cd kg}^{-1}$  and  $17.18 \text{ mg Tl kg}^{-1}$ ) concentrations than *Lemna minor*. Third day for *L. gibba* and fourth day for *L. minor* during the experiment were found as the optimal harvesting times for both Cd and Tl. Although the tailing waste water contained very low Cd and Tl levels, *L. gibba* and *L. minor* accumulated 3357 and 2710 times more cadmium and 5726 and 4424 times more thallium than in the tailing waste water, respectively. It means that LG and LM plants in 8. day of the experiment removed to Cd in 3357 and 2710 Lt water and Tl in 5726 and 4424 Lt water. The accumulations of Cd and Tl in polluted waters by *L. gibba* and *L. minor* are useful, cost-effective and environmentally nondestructively. The harvesting of these plants grown in tailing waste water can help the control of the environmental pollution and reduce health problems led of heavy metal pollution to animals and humans.

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