



## Research article

The potential of *Lemna gibba* L. and *Lemna minor* L. to remove Cu, Pb, Zn, and As in gallery water in a mining area in Keban, TurkeyMerve Sasmaz <sup>a</sup>, Emine Işıl Arslan Topal <sup>a</sup>, Erdal Obek <sup>b</sup>, Ahmet Sasmaz <sup>c,\*</sup><sup>a</sup> Firat University, Dept. of Environmental Engineering, Elazığ 23119, Turkey<sup>b</sup> Firat University, Dept. of Bioengineering, Elazığ 23119, Turkey<sup>c</sup> Firat University, Dept. of Geological Engineering, Elazığ 23119, Turkey

## ARTICLE INFO

## Article history:

Received 26 July 2015

Received in revised form

19 August 2015

Accepted 21 August 2015

Available online xxx

## Keywords:

Accumulation

*Lemna gibba* L.*Lemna minor* L.

Heavy metals

Gallery water

Keban

## ABSTRACT

This study was designed to investigate removal efficiencies of Cu, Pb, Zn, and As in gallery water in a mining area in Keban, Turkey by *Lemna gibba* L. and *Lemna minor* L. These plants were placed in the gallery water of Keban Pb–Zn ore deposits and adapted individually fed to the reactors. During the study period (8 days), the plant and water samples were collected daily and the temperature, pH, and electric conductivity of the gallery water were measured daily. The plants were washed, dried, and burned at 300 °C for 24 h in a drying oven. These ash and water samples were analyzed by ICP-MS to determine the amounts of Cu, Pb, Zn, and As. The Cu, Pb, Zn and As concentrations in the gallery water of the study area detected 67, 7.5, 7230, and 96  $\mu\text{g L}^{-1}$ , respectively. According to the results, the obtained efficiencies in *L. minor* L. and *L. gibba* L. are: 87% at day 2 and 36% at day 3 for Cu; 1259% at day 2 and 1015% at day 2 for Pb; 628% at day 3 and 382% at day 3 for Zn; and 7070% at day 3 and 19,709% at day 2 for As, respectively. The present study revealed that both *L. minor* L. and *L. gibba* L. had very high potential to remove Cu, Pb, Zn, and As in gallery water contaminated by different ores.

© 2015 Published by Elsevier Ltd.

## 1. Introduction

The heavy metal (HM) pollution in aquatic environments has surfaced as a global problem. These toxic metals are the main issue because they are persistent in nature, are non-biodegradable, and bioaccumulation in living animal and plants. These metals enter the environment mainly through various anthropogenic sources such as smelting processes, mining areas, fuel production, industrial effluents, etc. (OECD, 2003) and occur in aquatic systems as a result of the weathering of rocks and soils, from volcanic eruptions, and from different human activities. The most common HM contaminants are As, Cd, Cr, Cu, Ni, Pb, Zn, and Hg, and enter the freshwater from mining areas (US EPA, 2000). Water resources globally have been profoundly influenced over recent years by human activities, which have significantly contributed to the critical water supply and drinking water quality problems that the world is currently facing. In many parts of the world, heavy metal concentrations in drinking water are higher than some international guideline values

(Fernández-Luqueño et al., 2013).

Among the different techniques (precipitation-filtration, ion exchange, reverse osmosis, oxidation reduction, solvent extraction, membrane separation and phytoremediation) used for removal of heavy metals, phytoremediation is a cost-effective and eco-friendly technology that uses living green plants for in-situ removal of contaminants from water and soil (Dabrowski et al., 2004; Obek, 2009; Sasmaz and Obek, 2009, 2012; Sood et al., 2012; Tatar and Obek, 2014; Goswami et al., 2014). The phytoremediation ability depends on the ion uptake mechanism of each species depending on their genetic, physiological, anatomical, and morphological characteristics (Rahman and Hasegawa, 2011). Floating macrophytes usually uptake metal or contaminants through the process of rhizofiltration (Chaudhuri et al., 2014). Zayed et al. (1998) reported that *Lemna* sp. has been studied as another option for phytoremediation by many scientists because of its faster growth and easier harvest.

Among the aquatic macrophytes, *Lemna* sp. can float and has a high growth rate on the water. It has been observed that it is efficient to remove pesticides and metals from water as no long toxicity was seen (Dirilgen, 2011). Nevertheless, Li et al. (2011) stated that *Lemna minor* for pesticide or metal removal is one of

\* Corresponding author.

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

the most advantageous in living plants. Khataee et al. (2012) reports that additional advantages in using *Lemna* sp., such as the low cost, the long storage capacity, the minimization of chemicals or biological sludge volume, the possibility of transport, etc. Although duckweeds grow under different climatic conditions and have the fastest reproduction rates (Materazzi et al., 2012). Khataee et al. (2012) indicated that the tolerable pH range and temperature variations for *Lemna* sp. are 4–9 °C and 5–25 °C, but the best growth of *Lemna* sp. is obtained between pH 6 and 7.5 (Mkandawire and Dudel, 2005; Reema et al., 2011). Khellaf and Zerdaoui (2009) found that the best growth of the plant was at pH 6 and 21 °C to remove Cu and Cd in the aquatic environments by using *Lemna minor*. The present study has been carried out for the following objectives: (1) to determine Cu, Pb, Zn, and As concentrations in gallery water and aquatic plants (*Lemna gibba* L. and *L. minor* L.) growing in mining area, Keban, Turkey; (2) to measure daily the changes of Cu, Pb, Zn, and As concentrations in both *L. minor* L. and *L. gibba* L.; (3) to calculate the phytoremediation potential of *L. gibba* L. and *L. minor* L. in gallery water for Cu, Pb, Zn, and As; (4) to detect the optimal harvesting time for Cu, Pb, Zn, and As of *L. gibba* L. and *L. minor* L.; (5) to compare the phytoremediation potential for Cu, Pb, Zn, and As of *L. gibba* L. and *L. minor* L.

## 2. Materials and methods

### 2.1. Apparatus

A Perkin–Elmer Elan 9000 ICP-MS was used to determine Cu, Pb, Zn, and As in this study. The operation conditions of ICP-MS are given in Sasmaz and Yaman (2008).

### 2.2. The study area

This study was carried out in the Keban mining area (Elazig, Turkey) (E38° 40'50" and N38° 47'52"). The Keban region has a very big Pb–Zn–Ag deposit, which is related to the subvolcanics of syenite and syenomonzonite (Akgul, 2015), which outcrop around the town of Keban (Fig. 1). These rocks also contain high amounts of Cu, Pb, Zn, and As. The economic concentrations in Keban mining are Pb–Zn–Ag, Fe–Cu, W, Mn–Ag, and F–Mo ores. Argentiferous Pb–Zn ores used to be the main economic sources of the Keban area and have been mined for 6000 years (Seeliger et al., 1985). A lot of galleries used to exploit the Pb–Zn–Ag ore of the Keban mining district (the last mine closed in 1985). The door of the galleries was closed for security reasons, but there is common water effluent coming from the mining area, which, in the gallery, runs during all year and is finally discharged into the Karakaya Dam Lake (Fig. 1).

### 2.3. Water and plant samples

The composition of the gallery water may vary depending on the geologic units and mineralization type. This can also affect the pH, temperature (T °C) and electric conductivity (EC) of the gallery water. The water samples were collected daily with clean and sterilized plastic bottles from running gallery water in Keban mineralized area and together with plant samples. Both the water and plant samplings were accomplished during eight days. After that, the pH, temperature and electric conductivity of the gallery water in the study area measured in the field when *L. minor* L. and *L. gibba* L. collected daily from reactors (Table 1). Determination of pH was carried out by using either an Orion 4 Star pH meter along with Gel-Filled pH electrode or using an Oaktan pH tester 30. Temperature was recorded using a Traceable digital thermometer. Electric Conductivity was measured by using an Orion 4 Star conductivity meter along with an Orion conductivity electrode. In the

study, the plants are systematically identified as *L. gibba* L. and *L. minor* L., according to the procedure in *Flora of Turkey and the East Aegean Islands*, by Davis (1984).

### 2.4. Preparation of samples

*L. gibba* L. and *L. minor* L. were delivered from the Botanic Garden, Istanbul University, in September 2013. The plants were grown in a natural pool during 2 weeks in the laboratory and were then adapted in reactors, separately. Four hundred grams of the plants were placed in each reactor (Fig. 2) described by Tatar and Obek (2014). One reactor contained *L. gibba* L. and the other *L. minor* L. These reactors were operated under a sustained regime of flow volume (2.85 L s<sup>-1</sup>) of gallery water (Fig. 3). Both *L. gibba* L. and *L. minor* L. were collected daily; about 50 g of plant samples were taken separately from the reactors during the 8-day duration of the experiments. The plant samples were thoroughly washed with tap water, rinsed with distilled water, and dried at 60 °C for 24 h in the laboratory. A chelating EDTA wash was applied, and no differences were observed between EDTA washing and without EDTA washing. The dried plant samples (approximately 50 g) were ashed by heating at 300 °C for 24 h. The ashed samples were digested in HNO<sub>3</sub> (Merck, Darmstadt, Germany) for 1 h, followed by digestion in a mixture of HCl: HNO<sub>3</sub>: H<sub>2</sub>O (1:1:1, v/v; 6 ml per 1.0 g of the ashed sample) for 1 h at 95 °C. The digests were analyzed using ICP-MS techniques (it was used Group SO200 for water samples and Group VG104 for ashed plant samples) for Cu, Pb, Zn, and As (Sasmaz, 2015).

Statistical analysis of data was carried out using Analysis of Variance (ANOVA) and Student Newman Keul's Procedure (SNK) (Sokal and Rohlf, 1995) on SPSS 15.0 software (IBM Corp., Armonk, NY, USA). The metal results (Mn, Fe, Mg, Na, Al, K, P, and S) belong to the *L. gibba* L. and the *L. minor* L. of the study area, and were correlated with Cu, Pb, Zn, and As by using the Spearman Rank correlation (Table 2).

## 3. Results and discussion

### 3.1. Cu, Pb, Zn, and As concentrations in gallery water

Water samples were collected daily during the 8 day experiment in the field. The results of chemical analysis of 8 water samples were too close to each other and not observed significant changes for each metal. The mean of chemical analysis and physicochemical characteristics of gallery waters in the Keban Pb–Zn–Ag mining area are given in Table 1. The mean Cu, Pb, Zn, and As concentrations were detected to be 67.0 ± 2 µg L<sup>-1</sup>, 7.5 ± 1 µg L<sup>-1</sup>, 7230 ± 30 µg L<sup>-1</sup>, and 96.0 ± 5 µg L<sup>-1</sup> in the gallery water (p < 0.5), respectively, as shown in Table 1. Physicochemical characteristics such as pH, T (°C) and EC, together with analytical data of the major ions in the gallery water samples, are also represented in Table 1. The pH values of the gallery water range from 7.17 to 7.52 (mean: 7.36 ± 0.2); the temperature varies within a range of 19.0–21.1 °C (mean: 19.7 ± 1 °C); and the EC values of the gallery water in the study area range from 2.13 to 2.45 mS cm<sup>-1</sup> (mean: 2.29 ± 0.1 mS cm<sup>-1</sup>). These results indicate the close effects of numerous factors; that is, the distance to the feeding area of the gallery water; the residence time to the flow system in the mineralized area of the gallery water; the flow time of the gallery water coming from the feeding area; and the relatively long-term water–rock interaction in the mineralized area. For these reasons, these parameters (pH, T, and EC) of the gallery water were very similar to each other for the 8-day duration of the experiments.

As shown in Table 1, mean Cu and Pb concentrations in the study area are lower than the limit values (2000 µg L<sup>-1</sup>, 10 µg L<sup>-1</sup>,

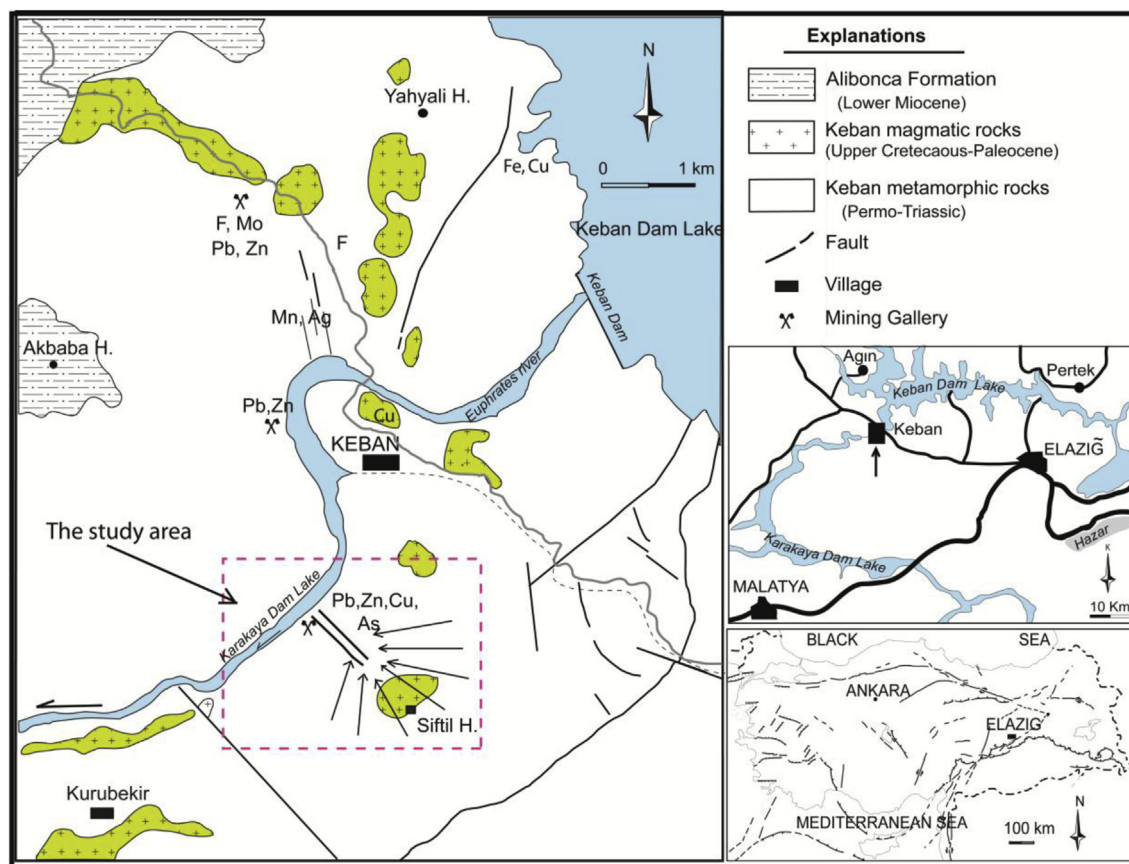


Fig. 1. Geological and location map of the study area (simplified from Akgul, 1987).

Table 1

Physicochemical characteristics of gallery water in the study area and detection limits of ICP-MS for the anion and cations.

| 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> (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> )  |
|---------------|--------------------------|--------------------------|---------------------------|---|--|---------------------------------------|--------------------------------------|--------------------------|--------------------------|--------------------------|
| DL            | —                        | —                        | —                         | —   | —  | —                                     | —                                    | 0.05                     | 0.05                     | 0.05                     |
| Gallery 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                   |
| Parameter     | Na (mg L <sup>-1</sup> ) | 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> ) |
| DL            | 0.05                     | 10                       | 0.05                      | 1   | 1.5  | 5                                     | 0.1                                  | 0.1                      | 0.5                      | 0.5                      |
| Gallery water | 140 ± 5                  | 590 ± 30                 | 820 ± 30                  | 580 ± 15  | 65 ± 3   | 195 ± 7                               | 67 ± 2                               | 7.5 ± 1                  | 7230 ± 30                | 96 ± 5                   |

respectively) and, Zn and As concentrations are higher the limit values (3000 µg L<sup>-1</sup> and 10 µg L<sup>-1</sup>, respectively), established for drinking water by the WHO (2006). This also causes HM pollution in the surrounding soil and water along the Fırat River. Toxic contaminants are not easy to uptake or remove after they have contaminated the surface and ground water. These contaminants directly enter the human body through contaminated surface and ground water. Mining waters cause both soil and water contamination in the environment. Consequently, it is very important to rehabilitate the surface and ground water around the mining areas that has been polluted by HMs (Caussy et al., 2003; Dong et al., 2010). Ning et al. (2011) indicated that the HM concentrations of surface water in the gold mining area were higher than class III or class IV of the national surface water quality standards. Along the flow direction, the concentrations of HMs decreased the further away the water was from the sources of pollution. Ning et al. (2011) observed that there were linear correlations between the correlation coefficients of some metals: Pb–Cr, Hg–Cu, Hg–Cd, and Cu–Cd were 0.960, 0.986, 0.966, and 0.980, respectively. It was ascertained that the metal content of the surface or ground water varies

depending on the possible sources of HMs and their geochemistry.

Three hydrochemical facies have been identified based on the contents of major cations and anions (Ca–Mg–HCO<sub>3</sub>; Ca–Mg–Fe–SO<sub>4</sub>; Na–Cl–NO<sub>3</sub>). Water types in the aquifer were specified by using Piper's (1944) trilinear plotting technique. Ca, Mg, and Na are the dominant cations, and represented more than 90% of the cation in the study area. Sulfate and bicarbonate are the prevailing components for the gallery water in the study area and represent 88–95% of the major anion. The major anion of the gallery water can be described as Ca–Mg–SO<sub>4</sub> bicarbonate water.

### 3.2. Cu, Pb, Zn, and As in *Lemna minor* L. and *Lemna gibba* L.

Phytoremediation is an efficient and cost-effective method to decontaminate environments. However, in order to optimize the systems, knowledge of how HMs affect plant physiology must be obtained prior to designing a system of decontamination (Pilon-Smits, 2005). Certain aquatic plants are considered to be HM pollution indicators and are successfully used as a method for monitoring environmental pollution (Cenci, 2000). Albers and





Fig. 2. The plants were placed in each reactor, separately. One reactor contained *L. gibba* L. and the other *L. minor* L..



Fig. 3. These reactors were operated under a sustained regime of flow volume of gallery water.

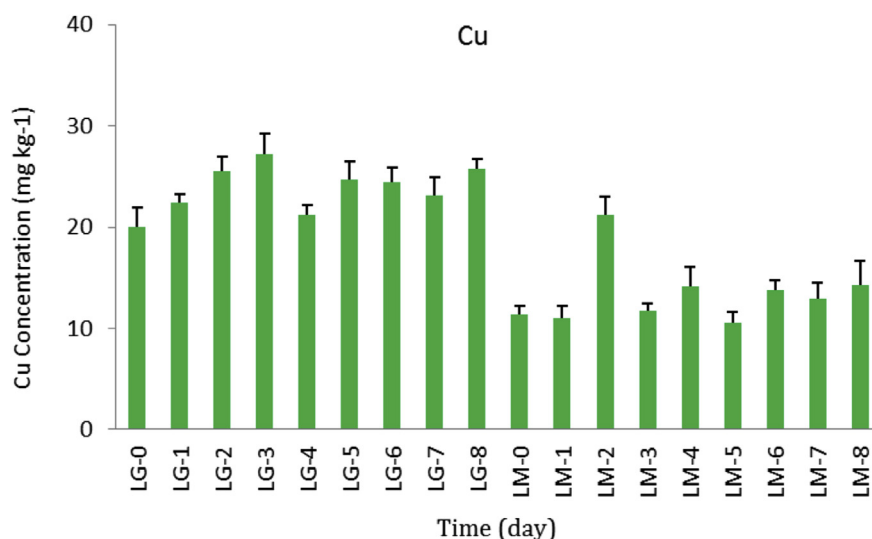
Camardese (1993) stated that metal concentrations in aquatic plants can be 100,000 times more than the values in associated waters. The HMs (Cu, Zn, Ag, Hg, Cd, As, Cr, Tl, and Pb) are toxic, have a high density, and are dangerous because of their abilities to bioaccumulate in a biological organism over time (Baby et al., 2010).

Cu values before the experimental study of *L. gibba* L. (LG-0) and *L. minor* L. (LM-0) are  $19.97 \text{ mg kg}^{-1}$  and  $11.38 \text{ mg kg}^{-1}$ , respectively (Fig. 4). These Cu values before the experimental study can be accepted as control groups for both LG-0 and LM-0. From the first day during the experimental study, on a daily basis, *L. gibba* L. and *L. minor* L. accumulated  $19.97 \text{ mg Cu kg}^{-1}$  and  $11.38 \text{ mg Cu kg}^{-1}$ , respectively. In the study, the amounts of Cu absorbed from low Cu

concentrations in the gallery water by *L. gibba* L. increased from 12.4% on the first day, to 27.5% on the second day, and to 36.2% on the third day. However, the performance of Cu accumulations decreased over the following days because of the saturation to Cu of *L. gibba* L. The amounts of Cu absorbed by *L. minor* L. decreased from 3.25% on the first day and increased to 86.7% on the second day. Small increases and decreases in Cu accumulations were observed over the following days. As presented in Fig. 4 maximum accumulations of Cu were observed on the third day for *L. gibba* L. and the second day for *L. minor* L. during the 8 days. Although very low concentrations (mean:  $67 \mu\text{g L}^{-1}$ ) of Cu were contained in the gallery water, *L. gibba* L. and *L. minor* L. accumulated 108 and 147

**Table 2**  
Spearman's correlation coefficients between some metals with Cu, Pb, Zn and As in *Lemna gibba* L. and *Lemna minor* L.

|    | Fe    | Ca    | Mg    | Na    | Al    | K     | P     | S     | Mn    | Cu   | Pb   | Zn   | As   |
|----|-------|-------|-------|-------|-------|-------|-------|-------|-------|------|------|------|------|
| Fe | 1.00  |       |       |       |       |       |       |       |       |      |      |      |      |
| Ca | 0.37  | 1.00  |       |       |       |       |       |       |       |      |      |      |      |
| Mg | 0.37  | 0.99  | 1.00  |       |       |       |       |       |       |      |      |      |      |
| Na | -0.22 | -0.73 | -0.68 | 1.00  |       |       |       |       |       |      |      |      |      |
| Al | 0.19  | 0.21  | 0.20  | 0.09  | 1.00  |       |       |       |       |      |      |      |      |
| K  | 0.26  | -0.59 | -0.61 | 0.49  | 0.06  | 1.00  |       |       |       |      |      |      |      |
| P  | -0.53 | -0.95 | -0.93 | 0.69  | -0.29 | 0.51  | 1.00  |       |       |      |      |      |      |
| S  | -0.06 | -0.86 | -0.86 | 0.69  | -0.03 | 0.88  | 0.77  | 1.00  |       |      |      |      |      |
| Mn | -0.15 | -0.86 | -0.84 | 0.52  | -0.19 | 0.66  | 0.77  | 0.79  | 1.00  |      |      |      |      |
| Cu | 0.65  | 0.87  | 0.87  | -0.69 | 0.19  | -0.39 | -0.86 | -0.67 | -0.71 | 1.00 |      |      |      |
| Pb | 0.98  | 0.39  | 0.38  | -0.20 | 0.34  | 0.27  | -0.57 | -0.05 | -0.16 | 0.63 | 1.00 |      |      |
| Zn | 0.76  | 0.07  | 0.06  | -0.01 | 0.25  | 0.53  | -0.29 | 0.24  | 0.29  | 0.21 | 0.81 | 1.00 |      |
| As | 1.00  | 0.32  | 0.32  | -0.20 | 0.19  | 0.31  | -0.49 | 0.02  | -0.08 | 0.60 | 0.98 | 0.80 | 1.00 |



**Fig. 4.** Cu accumulations by *Lemna gibba* L. and *Lemna minor* L.

times more copper than in the gallery water, respectively. *L. minor* L. was observed to have the ability or performance to accumulate Cu higher than in *L. gibba* L., compared to copper values before the experimental study of both *L. gibba* L. and *L. minor* L. (Fig. 4). Copper in *L. gibba* L. and *L. minor* L. ( $p < 0.5$ ) showed a high linear Spearman's correlation with the Fe, Ca, and Mg, and strong negative correlations with P, Mn, Na, and S in *L. gibba* L. and *L. minor* L. (Table 2). Cu has been accepted as an essential micronutrient for plant metabolism (Upadhyay et al., 2007). Cu had an important role in the structure of the cell and a vital for energy transfer and structural stability of chromosomes (Pahlsson, 1989). Obek (2009) found that *L. gibba* L. accumulated high levels of Cu, Zn, and Pb in the first few days, but then some decreases in the accumulation levels were recorded, most probably due to its saturation level.

Pb values in *L. gibba* L. (LG-0) and *L. minor* L. (LM-0) before the experimental study were  $2.51 \text{ mg kg}^{-1}$  and  $1.72 \text{ mg kg}^{-1}$ , respectively (Fig. 5). The values of LG-0 and LM-0 for Pb were accepted as control groups for both plants (Fig. 5). From the first day of the experimental study, on a daily basis, *L. gibba* L. and *L. minor* L. accumulated  $5.27 \text{ mg Pb kg}^{-1}$  (110% performance) and  $6.89 \text{ mg Pb kg}^{-1}$  (301% performance), respectively. During the 8-day study, the amount of Pb in the gallery water, which had a low Pb concentration, increased to 1015% on the second day, and to 948%, 557%, 1145%, 1363%, 1530%, 2107% on the following days as *L. gibba* L. accumulated it. This increased to 1259% on the second day, and to 279%, 846%, 465%, 863%, 1077%, and 1617% on the following days as

*L. minor* L. accumulated it. Although very low concentrations (mean:  $7.5 \text{ } \mu\text{g L}^{-1}$ ) of Pb were observed in the gallery water, on the second day and the eighth day *L. gibba* L. accumulated 3397-times and 7051-times more Pb, respectively, than in the gallery water. On the second day and the eighth day, *L. minor* L. accumulated 2888-times and 3708-times more Pb, respectively, than in the gallery water. As presented in Fig. 5, the accumulation of Pb by *L. gibba* L. increased especially during days 2 and 8 of the treatment, and after that, this was observed by variations in increases and decreases. *L. minor* L. also showed important increases during days 2, 4, and 8; and decreases were observed on days 3 and 5. *L. gibba* L. was observed to have the ability to accumulate higher levels of Pb than *L. minor* L., compared to Pb values before the experimental study of both plants (Fig. 5). Pb concentrations in both *L. gibba* L. and *L. minor* L. ( $p < 0.5$ ) showed a high linear with the HMs Fe, Zn, As, and Cu, and a negative Spearman's correlation (Table 2). Pb is toxic to the growth of plants, but extremely low levels of Pb (Pons et al., 2000). Obek (2009) worked on the Pb accumulation of *L. gibba* L. in the secondary treated municipal wastewater and observed faster Pb accumulation until day 5, but then began to decrease.

Zn values before the experimental study of *L. gibba* L. (LG-0) and *L. minor* L. (LM-0) were  $815.4 \text{ mg kg}^{-1}$  and  $421.4 \text{ mg kg}^{-1}$ , respectively (Fig. 6). These values of LG-0 and LM-0 for Zn were accepted as control groups for both plants. From the first day during the experimental study, on a daily basis, *L. gibba* L. and *L. minor* L. accumulated  $815.4 \text{ mg kg}^{-1}$  and  $421.4 \text{ mg Zn kg}^{-1}$ , respectively. The

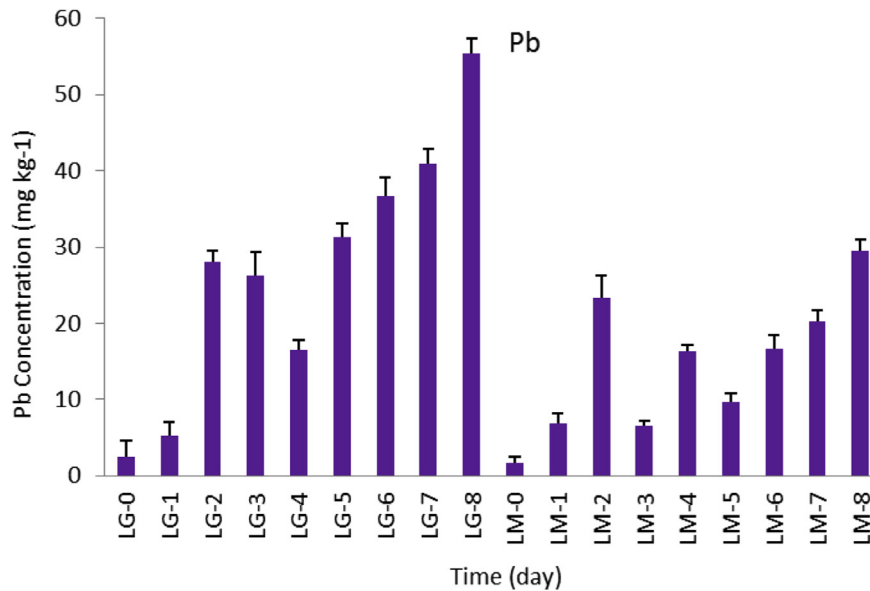


Fig. 5. Pb accumulations by *Lemna gibba* L. and *Lemna minor* L.

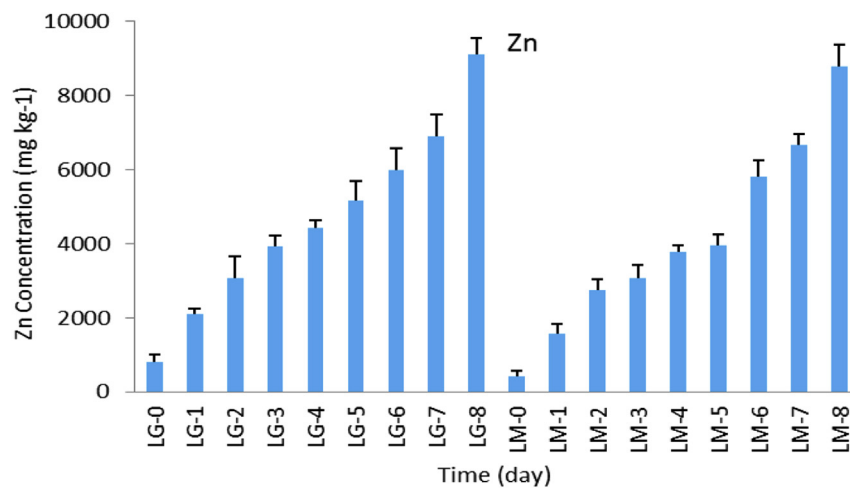


Fig. 6. Zn accumulations by *Lemna gibba* L. and *Lemna minor* L.

amounts of Zn absorbed from Zn concentrations in the gallery water (mean:  $7230 \mu\text{g L}^{-1}$ ) by *L. gibba* L. increased to 158% on the first day, 277% on the second day, 382% on the third day, and 443%, 532%, 634%, 746%, and 1017% on the following days. The Zn amounts absorbed by *L. minor* L. showed a regular increase from the first day until the eighth day (Fig. 6); for example, 270% on the first day, 551% on the second day, 628% on the third day, 795% on the fourth day, and 840%, 1278%, 1485%, and 1985% on the following days. Although low concentrations (mean:  $7230 \mu\text{g L}^{-1}$ ) of Zn were observed in the gallery water (Table 1), *L. gibba* L. and *L. minor* L. accumulated 1146-times and 1156-times more zinc, respectively, than in the gallery water on the eighth day. *L. minor* L. was observed to have the ability or performance to accumulate higher levels of Zn than *L. gibba* L., compared to Zn values before the experimental study of both plants (Fig. 6). Zn in *L. gibba* L. and *L. minor* L. ( $p < 0.5$ ) showed a high linear Spearman's correlation with Pb, As, Fe, and K (Table 2). Zn did not show negative correlations with other metals (Table 2). Zn is also vital for higher plants and is involved in some metabolic processes (Paschke et al., 2000). Obek (2009) studied the

Zn accumulation of *L. gibba* L. in the secondary treated municipal wastewater and stated that it increased linearly during the first 6 days, then began to decrease. Megateli et al. (2009) indicated that the removal of Cu and Zn by *L. gibba* L. was very rapid in the first 2 days, reaching 73% and 69% for Cu and 65% and 70% for Zn, respectively. After 2 days, the uptake of Zn continued at a lower rate from 89 to 100%. Khellaf and Zerdaoui (2009) demonstrated that *L. gibba* L. was very effective in removing Zn. They indicated that *L. gibba* L. was a great choice for phytoaccumulation because of its excellent uptake performance and high Zn accumulation ability.

As values in *L. gibba* L. (LG-0) and *L. minor* L. (LM-0) before the experimental study were  $2.1 \text{ mg kg}^{-1}$ , and  $5.3 \text{ mg kg}^{-1}$ , respectively (Fig. 7). The values of LG-0 and LM-0 for As were accepted as control groups for both plants. From the first day during the experimental study, on a daily basis, *L. gibba* L. and *L. minor* L. accumulated  $43.4 \text{ mg As kg}^{-1}$  (1967% performance) and  $90.8 \text{ mg As kg}^{-1}$  (1613% performance), respectively. As accumulations during the 8-day experiment, the amounts of As in the gallery water, which had a low As concentration ( $96 \mu\text{g L}^{-1}$ ) (Table 1), increased to 19,728% on

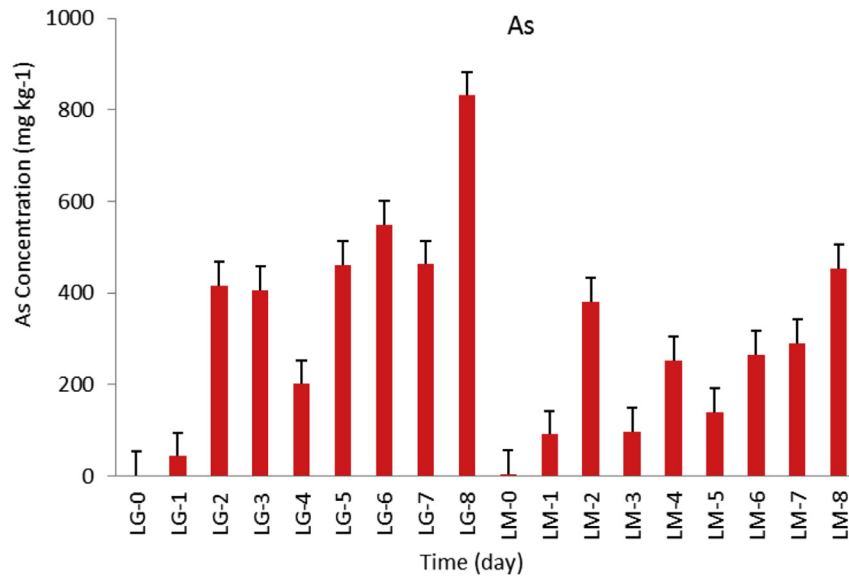


Fig. 7. As accumulations by *Lemna gibba* L. and *Lemna minor* L.

the second day, and to 19,271%, 9509%, 21,833%, 26,019%, 21,943%, and 39,490% on the following days, as *L. gibba* L. accumulated it, and increased to 7077% the second day, and, 1749%, 4672%, 2534%, 4911%, 5387% and 8454% the following days, as *L. minor* L. accumulated it. Although very low concentrations (mean: 96  $\mu\text{g L}^{-1}$ ) of As were observed in the gallery water, *L. gibba* L. and *L. minor* L. on the second day accumulated 4316-times and 3941-times more As than in the gallery water, respectively. As presented in Fig. 7, the accumulation of As by *L. gibba* L. and *L. minor* L. increased especially during days 2 and 8 of the treatment; on other days, variations in increases and decreases were observed. *L. minor* L. also showed important increases during days 2, 4, and 8 and decreases were observed on days 3 and 5. *L. gibba* L. was observed to have the ability to accumulate higher levels of As than *L. minor* L., compared to the As values before the experimental study of both (Fig. 7). As in *L. gibba* L. and *L. minor* L. ( $p < 0.5$ ) showed a high linear correlation with Pb, Zn, and Cu, and a low linear correlation with Ca, Mg, and K in *L. gibba* L. and *L. minor* L. (Table 2). Sasmaz and Obek (2009) demonstrate that As in secondary effluents was quickly absorbed by *L. gibba* L. in the first 2 days of the experimental study. However, after day 2, variations in the accumulation levels of these elements were reported, which was probably due to the saturation level of the plant material. Mkandawire and Dudel (2005) demonstrated that *L. gibba* L. has a very good ability to accumulate As in the waters around the mining area. Favas et al. (2014) indicated that although they detected low As concentrations in the surface waters around the uranium mining area in the center of Portugal, they found that *L. minor* L. had the ability to accumulate As in higher concentrations in the surrounding environment. The relative growth factor of *L. minor* L. was studied by Goswami et al. (2014) and it was found that the maximum removal of more than 70% As occurred on the fifteenth day of their experimental period. They suggested that *L. minor* L. was suitable for the remediation of As from aqueous areas at low concentrations.

#### 4. Conclusion

In this study, it was proved to be a cheap, green and effective method for the treatment of contaminated water with the mines by using of *Lemna gibba* L. and *Lemna minor* L. among phytoremediation plants for Cu, Pb, Zn, and As. The results of our study

demonstrate that *Lemna gibba* L. accumulated more Cu, Pb, Zn and As than *Lemna minor* L. but it showed less accumulation performance than *Lemna minor* L. in compared with the control group samples (LG-0 and LM-0) of *Lemna gibba* L. and *Lemna minor* L., except for As. The sequence of the HMs accumulated by *L. gibba* L. and *L. minor* L. was determined to be As > Pb > Zn > Cu and optimal harvesting times of *L. gibba* L. and *L. minor* L. for Cu, Pb, Zn and As. *L. gibba* L. and *L. minor* L. accumulated 4316-times and 3941-times more As, 2888-times and 3708-times more Pb, 1146-times and 1156-times more Zn and 108-times and 147-times more Cu than in the gallery water, respectively. The removal of the HMs in contaminated waters by *L. gibba* L. and *L. minor* L. is environmentally and nondestructively cost-effective. Therefore, the harvesting of *L. gibba* L. and *L. minor* L. in mineralized waters should be avoided so that they can help control HM pollution in the aquatic environment and reduce health risks to humans and animals, which are caused by HM pollution.

#### Appendix A. Supplementary data

Supplementary data related to this article can be found at <http://dx.doi.org/10.1016/j.jenvman.2015.08.029>

#### References

- Akgul, B., 1987. Petrography of Metamorphic Rocks in the Vicinity of Keban-Elazig. Master's thesis. F.U. Fen Bilimleri Ens. (in Turkish with English abstract), Elazig, Turkey.
- Akgul, B., 2015. Geochemical associations between fluorite mineralization and A-type shoshonitic magmatism in the Keban-Elazig area, East Anatolia, Turkey. J. Afr. Earth Sci. 111, 222–230.
- Albers, P.H., Camardese, M.B., 1993. Effect of acidification on metal accumulation by aquatic plants and invertebrates: I. constructed wetlands. Environ. Toxicol. Chem. 12, 959–967.
- Baby, J., Raj, J., Biby, E.T., Sankarganesh, P., Jeevitha, M.V., Ajisha, S.U., Rajan, S.S., 2010. Toxic effect of heavy metals on aquatic environment. Int. J. Biol. Chem. Sci. 4, 939–952.
- Caussy, D., Gochfeld, M., Gurzau, E., 2003. Lessons from case studies of metals: investigating exposure bioavailability and risk. Ecotoxicol. Environ. Saf. 56, 45–51.
- Cenci, R.M., 2000. The use of aquatic moss (*Fontinalis antipyretica*) as monitor of contamination in standing and running waters: limits and advantages. J. Limnol. 60, 53–61.
- Chaudhuri, D., Majumder, A., Misra, A.M., Bandyopadhyay, K., 2014. Cadmium removal by *Lemna minor* and *Spirodela polyrrhiza*. Int. J. Phytorem. 16, 1119–1132.
- Dabrowski, A., Hubicki, Z., Podkościelny, P., Robens, E., 2004. Selective removal of



- the heavy metal ions from waters and industrial wastewaters by ion-exchange method. *Chemosphere* 56, 91–106.
- Davis, P.H., 1984. Flora of Turkey and the East Aegean Island. Edinb. Univ. Press.
- Dirilgen, N., 2011. Mercury and lead: assessing the toxic effects on growth and metal accumulation by *Lemna minor*. *Ecotoxicol. Environ. Saf.* 74, 48–54.
- Dong, D., Li, H., Zhang, J., Sun, L., 2010. Removal of heavy metals from mine water by cyanobacterial calcification. *Min. Sci. Technol.* 20, 566–570.
- Favas, P.J.C., Pratas, J., Varun, M., D'Souza, R., Paul, M.S., 2014. Accumulation of uranium by aquatic plants in field conditions: prospects for phytoremediation. *Sci. Total Environ.* 470–471, 993–1002.
- Fernández-Luqueño, F., López-Valdez, F., Gamero-Melo, P., Luna-Suárez, S., Aguilera-González, E.N., Martínez, A.I., Pérez-Velázquez, I.R., 2013. Heavy metal pollution in drinking water a global risk for human health: a review. *Afr. J. Environ. Sci. Technol.* 7, 567–584.
- Goswami, C., Majumder, A., Misra, A.K., Bandyopadhyay, K., 2014. Arsenic uptake by *Lemna minor* in hydroponic system. *Int. J. Phytorem.* 16, 1221–1227.
- Khellaf, N., Zerdaoui, M., 2009. Phytoaccumulation of zinc by the aquatic plant *Lemna gibba* L. *Bioresour. Technol.* 100, 6137–6140.
- Khataee, A., Movafeghi, A., Torbati, S., Salehi Lisar, S., Zarei, M., 2012. Phytoremediation potential of duckweed (*Lemna minor* L.) in degradation of Cl acid blue 92: artificial neural network modeling. *Ecotoxicol. Environ. Saf.* 80, 291–298.
- Li, S.X., Feng-Ying, F., Yang, H., Jian-Cong, N., 2011. Thorough removal of inorganic and organic mercury from aqueous solutions by adsorption on *Lemna minor* powder. *J. Hazard. Mater.* 186, 423–429.
- Materazzi, S., Canepari, S., Aquili, S., 2012. Monitoring heavy metal pollution by aquatic plants. *Environ. Sci. Pollut. Res.* 19, 3292–3298.
- Megateli, S., Semsari, S., Couderchet, M., 2009. Toxicity and removal of heavy metals (cadmium, copper, and zinc) by *Lemna gibba*. *Ecotox. Environ. Saf.* 72, 1774–1780.
- Mkandawire, M., Dudel, E.G., 2005. Accumulation of arsenic in *Lemna gibba* L. (duckweed) in tailing water of two abandoned uranium mining sites in Saxony, Germany. *Sci. Total Environ.* 336, 81–88.
- Ning, N., Liyuan, Y., Jirui, D., Xugui, P., 2011. Heavy metal pollution in surface water of linglong gold mining area, China. *Proc. Environ. Sci.* 10, 914–917.
- Obek, E., 2009. Bioaccumulation of heavy metals from the secondary treated municipal waste water by *Lemna gibba*. *Fres. Environ. Bull.* 18, 2159–2164.
- OECD, 2003. Technical Guidance for the Environmentally Sound Management of Specific Waste Streams: Used and Scrap Personal Computers. Organization for Economic Cooperation and Development Working Group on Waste Prevention and Recycling.
- Pahlsson, A.M.B., 1989. Toxicity of heavy-metals (Zn, Cu, Cd, Pb) to vascular plants—a literature review. *Water Air Soil Pollut.* 47, 287–319.
- Paschke, M.W., Redente, E.F., Levy, D.B., 2000. Zinc toxicity thresholds for important reclamation grass species of the Western United States. *Environ. Toxicol. Chem.* 19, 2751–2756.
- Pilon-Smits, E.A., 2005. Phytoremediation. *Ann. Rev. Plant Biol.* 56, 15–39.
- Piper, A.M., 1944. A graphic procedure in the geochemical interpretation of water analyses. *Trans. Am. Geophys. Union* 25, 914–923.
- Pons, T.L., Lambers, H., Chapin, F.S., 2000. *Plant Physiological Ecology*. Springer-Verlag, New York.
- Rahman, M.A., Hasegawa, H., 2011. Aquatic arsenic: phytoremediation using floating macrophytes. *Chemosphere* 83, 633–646.
- Reema, R.M., Saravanan, P., Dharmendra Kumar, M., Renganathan, S., 2011. Accumulation of methylene blue dye by growing *Lemna minor*. *Sep. Sci. Technol.* 46, 1052–1058.
- Sasmaz, M., 2015. *Lemna gibba* L. ve *Lemna minor* L.' ün galeri suyundaki bazı ağır metalleri alım kapasitelerinin karşılaştırılması. Master Tezi. 43 s. Fırat Üniversitesi, Fen Bilimleri Enstitüsü, Elazığ, Turkey (Abstract with English) (unpublished).
- Sasmaz, A., Yaman, M., 2008. Determination of uranium and thorium in soil and plant parts around abandoned Pb-Zn-Cu mining area. *Commun. Soil Sci. Plant Anal.* 39, 2568–2583.
- Sasmaz, A., Obek, E., 2009. The accumulation of arsenic, uranium, and boron in *Lemna gibba* L. exposed to secondary effluents. *Ecol. Eng.* 35, 1564–1567.
- Sasmaz, A., Obek, E., 2012. The accumulation of silver and gold in *Lemna gibba* L. exposed to secondary effluents. *Chem. Erde Geochem.* 72, 149–152.
- Seeliger, T.C., Pernicka, E., Wagner, G.A., Begemann, F., Schmitt-Strecker, S., Eibner, C., Öztunalı, Ö., Baranyi, I., 1985. Archäometallurgische untersuchungen in nord- und ostanatolien. *Jahrbuch des Römisch-Germanisches Zentralmuseum* 32, 597–659.
- Sokal, R.R., Rohlf, F.J., 1995. *Biometry: the Principles and Practice of Statistics in Biological Research*, third ed. W.H. Freeman and Co, New York. p. 887.
- Sood, A., Uniyal, P.L., Prasanna, R., Ahluwalia, A.S., 2012. Phytoremediation potential of aquatic macrophyte, *Azolla*. *Ambio* 41, 122–137.
- Tatar, S.Y., Obek, E., 2014. Potential of *Lemna gibba* L. and *Lemna minor* L. for accumulation of boron from secondary effluents. *Ecol. Eng.* 70, 332–336.
- Upadhyay, A.R., Mishra, V.K., Pandey Sudhir, K., Tripathi, B.D., 2007. Biofiltration of secondary treated municipal wastewater in a tropical city. *Ecol. Eng.* 30, 9–15.
- US EPA, 2000. Abandoned Mine Site Characterization and Clean up Handbook. Seattle WA.
- WHO, 2006. World Health Organization. Guidelines for Drinking-water Quality. Geneva, Switzerland, 553pp.
- Zayed, A., Gowthaman, S., Terry, N., 1998. Phytoaccumulation of trace elements by wetland plants: L. Duckweed. *J. Environ. Qual.* 27, 715–721.