

Phytoextraction of Heavy Metal (Cu, Zn, Pb) from Mining Area by Sunflower (*Helianthus annuus*)

Mustafa AYBAR¹ , Bülent SAĞLAM² , Hatice DAĞHAN³ , Aydın TÜFEKÇIOĞLU² 
Nurcan KÖLELİ⁴ , Fatma Nur YILMAZ² 

¹Artvin Çoruh University, Artvin Vocational High School, Artvin, TÜRKİYE

²Artvin Çoruh University, Faculty of Forestry, Department of Forest Engineering, Artvin, TÜRKİYE

³Eskişehir University, Faculty of Agriculture, Department of Soil Science and Plant Nutrition,
Osmangazi, Eskişehir, TÜRKİYE

⁴Mersin University, Faculty of Engineering, Department of Environmental Engineering, Çiftlikköy
Campus, Mersin, TÜRKİYE

*Corresponding Author: mustafaaybar@artvin.edu.tr

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Abstract

Aim of study: The aim of this study was to determine the phytoremediation capacity of sunflower (*Helianthus annuus* L.), which can also be used as a bioenergy plant, in soils contaminated with copper (Cu), zinc (Zn) and lead (Pb) from the waste dumping sites during the mining operations.

Area of study: Phytoremediation study was carried out in Artvin Coruh University Research greenhouse environment.

Material and Methods: Plants were grown by mixing clean and contaminated soil at the rates of 0%, 50% and 100%. Bioaccumulation (BAF) and translocation (TF) factors were calculated by measuring heavy metal concentrations in soil and plant samples. The bioaccumulation factor is calculated by dividing the metal concentration in the shoots with the metal concentration in the soil. The translocation factor expresses the ratio of the metal concentration in the plant green parts to the root metal concentration.

Main results: The sunflower plant has accumulated the highest Zn in the shoots, while Cu and Pb have accumulated the highest in the roots. The mean BAF values of the plants were determined as 0.72 for Zn, 0.5 for Pb and 0.28 for Cu, while the mean TF values were determined as 1.25 for Zn, 0.97 for Pb and 0.52 for Cu.

Highlights: The findings show that the sunflower plant can be used in the phytostabilization of Cu metal and in the phytoextraction of Zn and Pb metal to reclaim heavy metal contaminated soils.

Keywords: Sunflower, Mine site, Copper, Zinc, Lead, Phytoextraction

Maden Sahasından Ağır Metallerin (Cd, Zn, Pb) Ayçiçeği (*Helianthus annuus*) Bitkisiyle Fitoekstraksiyonu

Öz

Çalışmanın amacı: Bu çalışmanın amacı, maden işletmesindeki pasa döküm sahasından alınan bakır (Cu), çinko (Zn) ve Kurşun (Pb) ile kirlenmiş topraklarda, biyoenerji bitkisi olarak da kullanılabilen ayçiçeği (*Helianthus annuus* L.) bitkisinin fitoremediasyon kapasitesinin belirlenmesidir.

Çalışma alanı: Artvin Çoruh Üniversitesi Araştırma serası ortamında fitoremediasyon çalışması yapılmıştır.

Materyal ve Yöntem: Çalışmada temiz ve kirlili toprak %0, %50 ve %100 oranlarında karıştırılarak bitki yetiştirilmiştir. Araştırmada toprak ve bitki örneklerinin ağır metal konsantrasyonları ölçülerek bitkilerin biyoakümüülasyon (BAF) ve traslokasyon (TF) faktörleri hesaplanmıştır. Biyoakümüülasyon faktörü sürgünlerdeki metal konsantrasyonunun topraktaki metal konsantrasyonuna oranını, traslokasyon faktörü ise bitki yeşil aksamındaki metal derişiminin kök metal derişimine oranını ifade eder.

Sonuçlar: Ayçiçeği bitkisi Zn metalini en fazla yeşil aksamda, Cu ve Pb metallerini ise kökte biriktirmiştir. Bitkinin BAF değerleri ortalamaları Zn için 0.72, Pb için 0.5 ve Cu için 0.28, TF değerleri ortalamaları ise Zn için 1.25, Pb için 0.97 ve Cu için 0.52 olarak belirlenmiştir.

Önemli vurgular: Elde edilen bulgular, ayçiçeği bitkisinin ağır metaller ile kirlenmiş toprakların iyileştirilmesi için Cu metalinin fitostabilizasyonunda, Zn ve Pb metalinin ise fitoekstraksiyonunda kullanılabilceğini göstermektedir.

Anahtar Kelimeler: Ayçiçeği, Maden sahası, Bakır, Çinko, Kurşun, Fitoekstraksiyon



Introduction

Soils are one of the most important resources that people depend on for life, shelter and production. The fulfillment of the growing population's day-by-day needs increases the operation and use of mines, which are one of the raw materials of emerging industries, science, and technology. However, it is necessary to dispose of the wastes generated as a result of mining activities, reorganize the deteriorated landforms in the mining area and improve the ecological conditions again (Sheoran et al., 2010). The presence of one or more heavy metals in the mining area and the surrounding soils and the high concentrations of these metals, have a negative impact on human life, the environment and both plants and animals in the forest ecosystem (Roy et al., 2005; Özel & Özel, 2012; Sağlam et al., 2020).

Heavy metals such as cadmium (Cd), chromium (Cr), copper (Cu), mercury (Hg), lead (Pb), and zinc (Zn) are among the most common pollutants (Jing et al., 2007). On the other hand, heavy metals such as Cd, Pb, arsenic (As), and Hg are metals that can not have a biological function on living organisms. These heavy metals, whether necessary or not, may become toxic when they are taken in excessive amounts by living organisms, may adversely affect the growth and reproduction of organisms, and even cause their death (Prieto et al., 2018). As these pollutants cannot be biodegradable, they are a serious concern for living organisms and the environment as carcinogenic and mutagenic compounds (Wu et al., 2018).

Various technologies are available for the remediation of soils contaminated with heavy metals. In general, improvement technologies can be divided into three main groups: physical, chemical and biological (Dellisanti et al., 2009; Ucaroglu & Talinli, 2012; Abdelhafez et al., 2014; Park & Son, 2017). Among these technologies, physical and chemical technologies can cause secondary pollutants as well as being expensive (Marques et al., 2008; Haque et al., 2008) and can cause adverse effects on the biological activities, structure and productivity of the soil (Pulford & Watson, 2003). Therefore, a low-cost and environmentally friendly

cleaning technique such as phytoremediation is often needed.

Phytoremediation is an easy-to-apply, promising, and most importantly inexpensive technique that cleans up heavy metals by using plants from contaminated soil and water, providing an environmentally friendly, aesthetic appearance, and minimal environmental degradation (Abou-Shanab, 2011; Sharma et al., 2015; Aybar et al., 2015; Ogundola et al., 2022). It is also thought that phytoremediation technology has great potential for in situ remediations of heavy metal contaminated soils (Gurajala et al., 2019; He et al., 2019). Plants that are grown in soils contaminated with multiple heavy metal ions, especially originating from mining areas, take metal ions in different amounts. While the plant takes up metal, it is greatly affected by the availability of metals in the soil, which varies depending on both soil properties and plant-related factors (Alaboudi et al., 2018). In addition, this method provides a great advantage since the soil mineral and organic matter do not change negatively in the phytoremediation method and cause minimal environmental degradation (Evangelou et al., 2004). However, the most important disadvantage of the phytoremediation method is that hyperaccumulator plants are defined as secondary pollutants and classified as hazardous waste because they contain excessive amounts of heavy metals in their tissues (Delil et al., 2020). Nevertheless, in this method, the harvested plants can be easily and safely processed by ash, composting or drying, or they can be recycled and removed from the secondary pollutant class (Lasat, 2002; Töre & Özkoç, 2022). Phytoremediation methods can be classified as phytoextraction, phytostabilization, phytodegradation, phytovolatilization and rhizodegradation. The phytoextraction method, which is widely used among these methods, is based on the principle of removing heavy metals from the soil and water environment by taking them with plant roots and accumulating them in the green part (stem+leaf) (Rafati et al., 2011; & Inamullah, 2016; Amin et al., 2018).

Plants differ in their ability to accumulate heavy metals in their bodies (Nouri et al., 2009; Kacálková et al., 2015). Therefore,

when choosing plant species for phytoremediation, the rapid growth of the plant, its ability to produce excess biomass and its high tolerance capacity to metals are taken into account (Dağhan, 2007; Dağhan et al., 2012; Rezanian et al., 2016). Some plant species can accumulate high amount of heavy metals in their bodies but produce little biomass and grow slowly. It is not possible to use the species with this feature in phytoremediation.

The sunflower plant is among the most preferred species in plant breeding studies since it has a high tolerance to heavy metals (Pilon-Smits, 2005; Rahman et al., 2013; Chirakkara & Reddy, 2015; Rizwan et al., 2016; Forte & Mutiti, 2017; Govarthan et al., 2018; Alaboudi et al., 2018; Al-Jobori & Kadhim, 2019). This study was carried out to

determine the heavy metal concentrations accumulated in the roots and shoots of plants using sunflower plants to improve multi-metal (Cu, Zn, Pb) contaminated mine site soil.

Material and Method

Study Area and Soil Material

The Global Positioning System (GPS) was used to determine the coordinates of the study areas where the heavy metal contaminated and uncontaminated soil samples were taken in this study. Contaminated and uncontaminated soils are found in the rust dumping area of the mining factory in Artvin-Murgul ($41^{\circ}13'17''$ N and $41^{\circ}50'39''$ E) and in the Artvin-Sachinka region ($41^{\circ}14'21''$ N and $41^{\circ}33'16''$ E) were taken from different points of forest areas, from a depth of 0-30 cm (Figure 1).

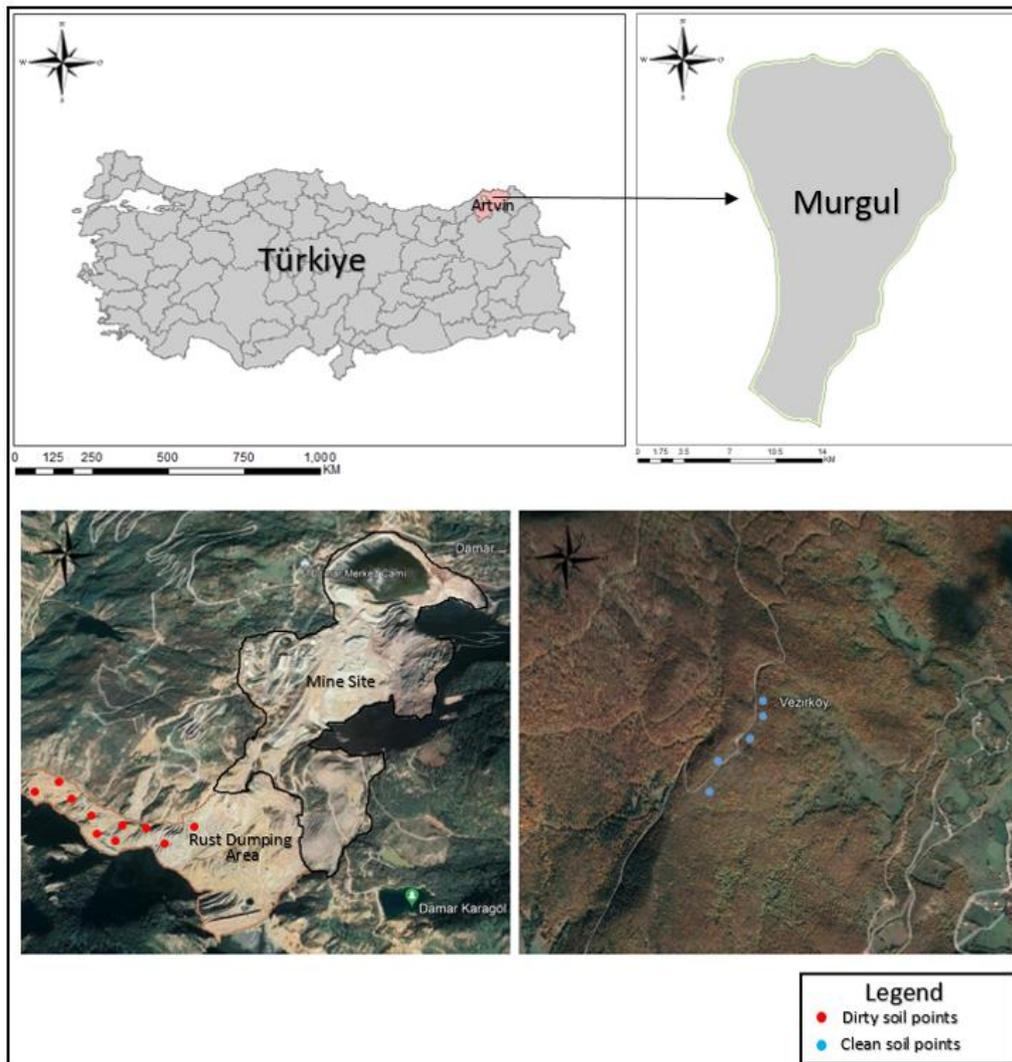


Figure 1. Maps of the studies areas.

The texture of the trial soil was determined according to the Bouyoucos hydrometer method (Bouyoucos, 1951). Soil reaction (pH) was measured in 1:2.5 soil: deionised water suspension (Kacar, 1984). Soil electrical conductivity was determined according to Kacar (1984). Organic matter contents of soil samples were determined according to the modified Walkley-Black method (Nelson & Sommers, 1996). The lime concentration of the soils was determined by Scheibler calcimeter by the volumetric method (Kacar, 1984). At the beginning of the experiment and at the end of the harvest, the total heavy metal (Cu, Zn, and Pb) contents of the soil samples were extracted according to the microwave wet digestion method (USEPA 3052) developed by the United States Environmental Protection Agency (USEPA) (USEPA, 1996). The heavy metal concentration of extracted samples were measured in an Inductively Coupled Plasma Optical Emission Spectrometer (ICP-OES, Perkin-Elmer, Optima 8000 model, USA).

Plant Material

Oliva CL (It is a hybrid sunflower variety suitable for Clearfield technology) type sunflower seeds obtained from the Thrace Agricultural Research Institute were used as plant material. Sunflower; it is a large-flowered, annual plant belonging to the Asteraceae family and can grow in a wide variety of soil types and can grow up to 3 meters. In this study, sunflower was chosen as plant material due to its ability to uptake and accumulate heavy metals from multi-metal polluted soils, high biomass, rapid growth rate, deep root structure, easily harvestable and bioenergy use of the harvested plant at the end of the study (Forte & Mutiti, 2017; Alaboudi et al., 2018; Al-Jobori & Kadhim, 2019).

Pot Trial

For the pot experiment, the polluted soil with high heavy metal concentration was taken from the rust dumping area around the Artvin-Murgul mine site, and the soil with low metal concentration was taken from the forest area within the borders of Artvin Forestry Operations Directorate, Sacinka Forestry Directorate from a depth of 0-30 cm. Soil

samples were air-dried and passed through 1 mm sieve. After that, they were mixed as 0%, 50%, and 100% ratio due to the excessive pollution of the soil taken from the mine site. Before the soils were put into pots, they were incubated for 1 month. At the end of the incubation, 10 kg soil from each mixture was filled in 14 kg pots, and the experiment was carried out for 5 weeks after planting. The completely randomized block design was used in the pot experiment with four replicates.

Ten sunflower seeds were sown in each pot and the pots were irrigated twice a week. After germination, seedlings were thinned to one plant per pot. After the plants were grown under controlled conditions (50-60% humidity, constant $24\pm 2^{\circ}\text{C}$) for 5 weeks in a greenhouse environment, they were harvested and separated into green parts and root parts. Both parts of the plant were washed with deionized water so that the heavy metal amounts in the plant parts are not affected by the soil, dried at 65°C to constant weight and then all samples ground in an Agate mill (Retsch MM301 Mixer Mill, Retsch GmbH, Nordrhein-Westfalen, Germany). The ground plant samples were digested with nitric acid (HNO_3) and hydrogen peroxide (H_2O_2) in a microwave oven (Speedwave MWS-2 Berghof Model, Germany), and the concentrations of heavy metals (Pb, Zn, and Cu) were measured in ICP / OES.

Heavy Metal Accumulations and Translocations

Bioaccumulation factor and translocation factor are commonly used to evaluate the phytoremediation potential of plants. In this study, BAF and TF values of sunflower were calculated for each heavy metal. The bioaccumulation factor refers to the ratio of heavy metal concentration in shoots to the heavy metal concentration in the soil (Li et al., 2007; Rezvani & Zaefarian 2011; Tózsér et al., 2019). The BAF values of metals were calculated as follows (Eq. 1): It is reported that the higher the bioaccumulation factor value, the more suitable a plant for phytoextraction (Blaylock et al., 1997). If the bioaccumulation factor value is greater than two, it is considered a high value (Mellem et al., 2012).

$$\text{BAF} = \frac{\text{Shoot Metal Concentration (mg kg}^{-1}\text{)}}{\text{Soil Metal Concentration (mg kg}^{-1}\text{)}} \quad (1)$$

The translocation factor (Eq. 2) is the ratio of the metal concentration in plant green parts (shoot) to the root metal concentration (Karami & Shamsuddi, 2010).

$$\text{TF} = \frac{\text{Shoot Metal Concentration (mg kg}^{-1}\text{)}}{\text{Root Metal Concentration (mg kg}^{-1}\text{)}} \quad (2)$$

Statistical Analysis

Mean and standard deviation values were calculated in all analyzes for soil and plant samples. Data were checked using statistical

analysis program SPSS 22.0, analysis of variance (ANOVA) and Duncan's multiple comparison test to determine the significance of differences between means ($p \leq 0.05$).

Results and Discussion

Soil taken from the mine dump site (100%), mix of forest and mine soil (50%) and samples taken from the forest area (0%) were determined by analyzing air-dry soil samples that were sieved with a 2 mm sieve for pH, EC and lime analyzes and 0.25 mm sieved for organic matter and heavy metal analyzes. (Table 1).

Table 1. Some physical and chemical properties of soils according to their pollution levels

Soil Properties	0%	50%	100%
pH	7.04	6.42	5.85
EC (dS/m)	0.52	0.84	1.02
CaCO ₃ (%)	3.50	2.76	1.99
Organic Matter (%)	3.89	2.41	0.64
Texture Class	Sandy Loam	Sandy Loam	Loamy Sand

EC: Electrical conductivity

The forest area soil which was taken for control and described as clean has a sandy loam texture, pH (7.04) neutral, unsalted (0.52dS/m), slightly calcareous (3.5%) and moderate organic matter (3.89%) content. On the other hand, the soil taken from the mine dump site has a loamy sandy texture with slightly acidic reaction (pH: 5.85), unsalted

(1.02dS/m), slightly calcareous (1.99%) and low organic matter content (0.64%). The total heavy metal concentrations of the experimental soil were determined after the soils were mixed at 0%, 50% and 100% and filled into the pots, and after the plants were planted and harvested in the pots (Table 2).

Table 2. Total heavy metal concentrations of potting soil at the beginning of the experiment and at the end of the harvest.

Soil Pollution Level	Heavy Metal Concentrations of Soils at the Beginning of the Trial (mg kg ⁻¹)			Heavy Metal Concentrations of Potted Soils at the End of the Harvest (mg kg ⁻¹)		
	Cu	Zn	Pb	Cu	Zn	Pb
0%	76.9±12.7	95.9±10.4	70.6±8.7	61.7±14.6	64.3±14.1	37.6±5.2
50%	493±39.5	233±36.9	115±28.8	395±38.6	102±12.6	62.9±12.4
100%	1102±47.3	324±38.7	230±46.9	900±37.5	213±42.9	77.1±15.8

Various metals are found in the soil under natural conditions. Average Cu>50, Pb>50 and Zn>150 mg kg⁻¹ values in soils with pH less than 6 show toxic effects. In soils with a pH greater than 6, Cu>140, Zn>300 and Pb>300 mg kg⁻¹ values show toxic effects (Anonymous, 2005). Considering these values, it was determined that the total Cu concentration of the trial soil was above the critical limit value at all pollution levels

except 0%, while it was above the limit value only at 100% pollution level in Pb and Zn. In the soil after harvest, Cu concentration was found above the critical value at all doses except 0%, as in the experimental soil. It was observed that the zinc and Pb concentration exceeded the critical limit value only at 100% pollution level. The results showed that with the heavy metals that sunflower absorbs from the soil, some metal concentration in the soil

decreases below the critical metal level. These findings are supported by the heavy metal

concentration values accumulated in the plant, given in Table 3.

Table 3. Heavy metal concentrations in the green parts and roots of the sunflower plants.

Soil Pollution Level	Green Part			Root		
	Cu	Zn	Pb	Cu	Zn	Pb
	(mg kg ⁻¹)					
0%	50.25c	92.50c	49.13b	50.13c	65.50c	50.88b
50%	63.13b	123.60b	55.75b	172.60b	107.80b	58.88ab
100%	79.38a	214.40a	74.00a	450.00a	179.10a	72.63a
F	15.85**	53.73**	6.26**	90.54**	49.41**	5.40**

***p* < 0.01

Along with the increase in soil pollution levels, a statistically significant increase (*p* < 0.01) was observed in the heavy metal concentrations of the plant's green parts and roots (Table 3). The highest Cu (79.38 mg kg⁻¹), Zn (214.4 mg kg⁻¹) and Pb (74.00 mg kg⁻¹) concentrations in green parts were measured in 100% polluted soil. However, green component Pb concentrations were found to be similar at control and 50% pollution levels. With a similar trend in the root, the highest Cu (450 mg kg⁻¹), Zn (179.1 mg kg⁻¹) and Pb

(72.63 mg kg⁻¹) were obtained at 100% pollution level.

According to the limit values (Table 4) reported by Kabata-Pendias (2000) for Cu, Zn and Pb in leaf tissues, the green part and root Zn concentrations are at a sufficient level for plants at 0% dose, while this value is within toxic limits at 50% and 100% doses (100-400 mg kg⁻¹). On the other hand, the concentrations of green parts and root Pb and Cu were found to be between the toxic limit values at all doses.

Table 4. Concentrations of heavy metals in mature leaf tissue for various species (mg kg⁻¹) dry weight

Heavy Metal	Deficiency	Sufficient	Toxic	Tolerable value for agricultural products	Threshold for hyperaccumulators
	(mg kg ⁻¹)				
Cu	2-5	5-30	20-100	5-20	>1000 ^a
Pb	-	5-10	30-300	0.5-10	>1000 ^a
Zn	10-20	27-150	100-400	50-100	>10000 ^b

a: Baker et al., (2000); b: Lasat, (2002)

Sunflower accumulated Cu and Pb metals most in their roots, while Zn metal accumulated more in green parts. The results showed that the sunflower plant could grow in this soil and accumulate high levels of heavy metals in its tissues, despite the heavy metal content exceeding the toxic level in the 100 % polluted soil. Although these amounts are not as high as 1000 mg kg⁻¹ for Cu and Pb and 10000 mg kg⁻¹ for Zn, it has been determined that this plant is important in terms of being able to be grown in toxic soil with multi-metal (Cu, Zn and Pb).

According to the heavy metal accumulation status of the leaves and root parts of the plants, it was observed that Cu was accumulated in the root parts compared to the

leaves. It was determined that the concentrations of zinc in the leaves were higher than in the roots, and the concentrations of Pb were approximately the same in both parts. Metal uptake and accumulation in plants may vary depending on the presence of one or more metals in the soil, the concentration of the metal, soil properties and the type of plant. Therefore, different results can be obtained in studies conducted by researchers. Indeed, Angelova et al. (2016), Eckhardt & Khanal (1999), and Lombi et al. (1998) stated in their studies that Pb, Zn and Cd heavy metals mainly accumulate in the leaves of sunflower plants. The findings obtained in this study were also compatible with the results of these studies.

Jadia & Fulekar (2008) determined that sunflower shoots were the main organ in heavy metal accumulation. Hamvumba et al., (2014) mixed Pb in different proportions with a good soil located near the mine and it was observed at the end of the study that it accumulated lead in its tissues.

Bioaccumulation and Translocation Factors

In the phytoremediation method, the ability of the plant to remove pollutants from

the soil and carry them to the above-ground parts by taking them with their roots is determined by two parameters defined as BAF and TF (Grzegórska et al., 2020). It has been reported that plants with BAF and TF values higher than 1 can be used for phytoextraction of metals, and plants with BAF and TF values lower than 1 can be used for phytostabilization (Mendez & Maier, 2008). BAF and TF values of sunflower plant are given in Table 5.

Table 5. BAF and TF values of the sunflower plants according to the pollution rates at increasing doses

Soil Pollution Level	BAF			TF		
	Cu	Zn	Pb	Cu	Zn	Pb
0%	0.654	0.965	0.696	1.004	1.412	0.965
50%	0.128	0.531	0.485	0.366	1.147	0.947
100%	0.072	0.661	0.322	0.176	1.197	1.019
Avg.	0.284	0.719	0.501	0.515	1.252	0.980

The findings showed that the average of BAF values was lower than 1 for all metals (Cu, Zn and Pb) investigated. The highest BAF value (average 0.965) was obtained at 0% dose of Zn metal and the lowest average (0.072) at 100% pollution dose of Cu metal. Similar results were also found in TF values. However, TF value was higher than 1 for Zn at all application doses, higher than 1 for Pb metal at 100% dose, and close to 1 at other doses. While this situation was calculated as ≥ 1 at 0% dose in Cu metal, it was found to be lower than 1 at 50% and 100% doses. As reported by Šijakova-Ivanova, et al., (2017), the fact that $TF > 1$ indicates that metals can be effectively transported from root to shoots. Our results showed that Zn and Pb can be transported from the roots to the green parts, but Cu accumulates in the roots. In addition, TF values indicate that sunflower can be used in the phytostabilization of Cu metal and in the phytoextraction of Zn and Pb metal. On the other hand, one of the reasons for the $BAF < 1$ may be that as a result of the toxic levels of these heavy metals in the soil, which is the plant growing environment, the plant roots are damaged by blocking the metals they take up in their tissues and limiting their transport to the above-ground parts.

Since metal uptake in plants depends on the plant species, age, metal type, concentration and soil properties, researchers have obtained different BAF and TF values in their studies (Yazdanbakhsh, et al., 2020). Kötschau et al., (2014) reported $BAF < 1$ for Cu, Pb and Zn (0.156, 0.013, and 0.783, respectively) in sunflowers in a study they conducted in the mining area. Similarly, many researchers [(Al-Jobori & Kadhim, 2019; Alaboudi et al., 2018; Forte & Mutiti, 2017; Lee et al., 2013] found that reported that the BAF and TF values of the *H. annuus* plant were lower than 1. They also reported that sunflower is not suitable for Pb accumulation in their tissues compared to other heavy metals. Although the findings are similar to the literature in terms of BAF values, it has been revealed that the TF values are higher than the values in the literature. In order to fully determine the effect of multi-metals in the mine sites, the growing period of plants should be extended. In addition, these studies should be tested in field conditions other than controlled environmental conditions.

Conclusions

This study was carried out to determine the phytoremediation capacity of the sunflower plant, which was grown in contaminated soils

with Cu, Zn and Pb and could also be used as a bioenergy raw material. The results showed that sunflower plant grown in soils contaminated with Cu, Zn and Pb has the ability to accumulate different concentrations of Cu, Zn and Pb in its tissues (roots and leaves) with the increasing in pollution rates.

When heavy metal concentrations were compared, the metals with the highest bioaccumulation factor were determined as Zn>Pb>Cu, respectively. Translocation factor was found to be above 1 for Zn and Pb and less than 1 for Cu at 100% contamination level. The results show that sunflower was suitable for phytostabilization of Cu metal and phytoextraction of Zn and Pb metals in soils contaminated with these metals. Such annual plants can contribute to the restoration of mining degraded areas and pre-afforestation studies. In order to maximize the removal efficiency of heavy metals, there is a need to use chelates and fertilizers to accelerate plant growth and to support this work with on-site treatment.

Energy crops, including sunflowers, are likely to be compatible with both the management of soil pollutants and the provision of other environmental services, including waste recycling. Based on responses to heavy metals under controlled conditions, plants such as sunflower appear promising for phytoremediation. However, due to the scarcity of field trials, the true potential of these plants cannot be fully elucidated.

Sunflower plant biomass, which is metal accumulator and characterized as a phytostabilizer, can be investigated in terms of bioenergy potential after this process. There is also a need for studies that will increase the phytoextraction performance of sunflower by supplementing chelators such as ethylenediamine disuccinic acid (EDDS) and fertilization to increase plant biomass in order to increase the phytoextraction potential of Cu, Zn and Pb from the soil in mine sites.

Ethics Committee Approval

N/A

Peer-review

Externally peer-reviewed.

Author Contributions

Conceptualization: M.A.; Investigation: M.A., B.S., F.N.Y.; Material and Methodology: M.A., F.N.Y., H.D.; Supervision: B.S., A.T.; Visualization: M.A., F.N.Y.; Writing-Original Draft: M.A., B.S.; Writing-review & Editing: B.S., A.T., H.D., N.K.; Other: Author has read and agreed to the published version of manuscript.

Conflict of Interest

The authors have no conflicts of interest to declare.

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References

- Abdelhafez, A.A., Li, J. & Abbas, M.H. (2014). Feasibility of biochar manufactured from organic wastes on the stabilization of heavy metals in a metal smelter contaminated soil. *Chemosphere*, 117, 66-71.
- Abou-Shanab, R.A.E.A. (2011). Bioremediation: new approaches and trends. In *Biomangement of metal-contaminated soils*, Springer, Dordrecht, 65-94.
- Alaboudi, K.A., Ahmed, B. & Brodie, G. (2018). Phytoremediation of Pb and Cd contaminated soils by using sunflower (*Helianthus annuus*) plant. *Annals of Agricultural Sciences*, 63(1), 123-127.
- Al-Jobori, K.M. & Kadhim, A.K. (2019). Evaluation of sunflower (*Helianthus annuus* L.) for phytoremediation of lead contaminated soil. *Journal of Pharmaceutical Sciences and Research*, 11(3), 847-854.
- Amanullah, I., & Inamullah, X. (2016). Dry matter partitioning and harvest index differ in rice genotypes with variable rates of phosphorus and zinc nutrition. *Rice Science*, 23(2), 78-87.
- Amin, H., Arain, B.A., Abbasi, M.S., Jahangir, T.M., & Amin, F. (2018). Potential for phytoextraction of Cu by *Sesamum indicum* L. and *Cyamopsis tetragonoloba* L.: a green solution to decontaminate soil. *Earth Systems and Environment*, 2(1), 133-143.
- Angelova, V., Ivanova, R., Todorov, Z. & Ivanov, K. (2016). Potential of Sunflower (*Helianthus Annuus* L.) for Phytoremediation of Soils Contaminated with Heavy Metals. *Agricultural Sciences/Agrarni Nauki*, 8(20).
- Anonymous, (2005). Official Gazette. Soil Pollution Control Regulation. Issue: 25831, Date: 31.05.2005.

- Aybar, M., Bilgin, A. & Sağlam, B. (2015). Removal heavy metals from the soil with phytoremediation. *Journal of Natural Hazards and Environment*, 1(1-2), 59-65.
- Baker, A.J., McGrath, S.P., Reeves, R.D. & Smith, J.A.C. (2020). Metal hyperaccumulator plants: A review of the ecology and physiology of a biological resource for phytoremediation of metal polluted soils. *Phytoremediation of Contaminated Soil and Water*, 85-107.
- Blaylock, M.J., Salt, D.E., Dushenkov, S., Zakharova, O., Gussman, C., et al. (1997). Enhanced accumulation of Pb in Indian mustard by soil-applied chelating agents. *Environmental Science & Technology*, 31(3), 860-865.
- Bouyoucos, G.I. (1951). A Calibration of the Hydrometer Method for Making Mechanical Analysis of the Soils. *Agronomy Journal*, 4, 9-434.
- Chirakkara, R.A. & Reddy, K.R. (2015). Biomass and chemical amendments for enhanced phytoremediation of mixed contaminated soils. *Ecological Engineering*, 85, 265-274.
- Dağhan, H. (2007). Phytoremediation: Cleaning contaminated areas using plants. GAP V. *Agriculture Congress Proceedings*, 362-367.
- Dağhan, H., Köleli, N., Uygur, V., Arslan, M., Önder, D., et al. (2012). Investigation of the use of transgenic tobacco plant in phytoextraction treatment of cadmium contaminated soils. *Soil Water Journal*, 1(1), 1-6.
- Delil, A.D., Köleli, N., Dağhan, H. & Bahçeci, G. (2020). Recovery of heavy metals from canola (*Brassica napus*) and soybean (*Glycine max*) biomasses using electrochemical process. *Environmental Technology & Innovation*, 17, 100559.
- Dellisanti, F., Rossi, P.L. & Valdrè, G. (2009). In-field remediation of tons of heavy metal-rich waste by Joule heating vitrification. *International Journal of Mineral Processing*, 93(3-4), 239-245.
- Eckhardt, H. & Khanal, S.K. (1999). Suitability of Bangkok sewage and nightsoil sludges for agricultural use with emphasis on potentially toxic elements. *Journal of Environmental Science & Health Part A*, 34(10), 2007-2021.
- Evangelou, M.W., Daghan, H. & Schaeffer, A. (2004). The influence of humic acids on the phytoextraction of cadmium from soil. *Chemosphere*, 57(3), 207-213.
- Forte, J. & Mutiti, S. (2017). Phytoremediation potential of *Helianthus annuus* and *Hydrangea paniculata* in copper and lead-contaminated soil. *Water, Air, & Soil Pollution*, 228(2), 1-11.
- Govarthanan, M., Mythili, R., Selvankumar, T., Kamala-Kannan, S. & Kim, H. (2018). Myco-phytoremediation of arsenic- and lead-contaminated soils by *Helianthus annuus* and wood rot fungi, *Trichoderma* spp. Isolated from decayed wood. *Ecotoxicology and Environmental Safety*, 151, 279-284.
- Grzegórska, A., Rybarczyk, P., Rogala, A. & Zabrocki, D. (2020). Phytoremediation-From Environment Cleaning to Energy Generation-Current Status and Future Perspectives. *Energies*, 13(11), 2905.
- Gurajala, H.K., Cao, X., Tang, L., Ramesh, T.M., Lu, M. & Yang, X. (2019). Comparative assessment of Indian mustard (*Brassica juncea* L.) genotypes for phytoremediation of Cd and Pb contaminated soils. *Environmental Pollution*, 254, 113085.
- Haque, N., Peralta-Videa, J.R., Jones, G.L., Gill, T.E. & Gardea-Torresdey, J.L. (2008). Screening the phytoremediation potential of desert broom (*Baccharis sarothroides* Gray) growing on mine tailings in Arizona, USA. *Environmental Pollution*, 153(2), 362-368.
- Hamvumba, R., Mataa, M. & Mweetwa, AM (2014). Evaluation of sunflower (*Helianthus annuus* L.), sorghum (*Sorghum bicolor* L.) and chinese cabbage (*Brassica chinensis*) for phytoremediation of lead contaminated soils. *Environment and Pollution*, 3(2), 65.
- He, J., Strezov, V., Kumar, R., Weldekidan, H., Jahan, S., et al. (2019). Pyrolysis of heavy metal contaminated *Avicennia marina* biomass from phytoremediation: Characterisation of biomass and pyrolysis products. *Journal of Cleaner Production*, 234, 1235-1245.
- Jadia, C.D. & Fulekar, M.H. (2008). Phytoremediation: The application of vermicompost to remove zinc, cadmium, copper, nickel and lead by sunflower plant. *Environmental Engineering & Management Journal*, 7(5).
- Jing, Y.D., He, Z.L. & Yang, X.E. (2007). Role of soil rhizobacteria in phytoremediation of heavy metal contaminated soils. *Journal of Zhejiang University Science B*, 8(3), 192-207.
- Kacar, B. (1984). *Plant Nutrition*. Ankara University. Faculty of Agriculture Arrow. No: 899, 169-175.
- Kabata-Pendias, A. (2000). *Trace Elements in Soils and Plants*, third ed. CRC Press, FL USA.
- Kacálková, L., Tlustoš, P. & Száková, J. (2015). Phytoextraction of risk elements by willow and poplar trees. *International Journal of Phytoremediation*, 17(5), 414-421.
- Karami, A. & Shamsuddin, Z.H. (2010). Phytoremediation of heavy metals with several efficiency enhancer methods. *African Journal of Biotechnology*, 9(25), 3689-3698.

- Kötschau, A., Büchel, G., Einax, J.W., von Tümpling, W. & Merten, D. (2014). Sunflower (*Helianthus annuus*): Phytoextraction Capacity for heavy metals on a mining-influenced area in Thuringia, Germany. *Environmental Earth Sciences*, 72(6), 2023-2031.
- Lasat, M.M. (2002). Phytoextraction of toxic metals: A review of biological mechanisms. *Journal of Environmental Quality*, 31(1), 109-120.
- Lee, K.K., Cho, H. S., Moon, Y.C., Ban, S.J. & Kim, J.Y. (2013). Cadmium and lead uptake capacity of energy crops and distribution of metals within the plant structures. *KSCE Journal of Civil Engineering*, 17(1), 44-50.
- Marques, A.P., Oliveira, R.S., Rangel, A.O. & Castro, P.M. (2008). Application of manure and compost to contaminated soils and its effect on zinc accumulation by *Solanum nigrum* inoculated with arbuscular mycorrhizal fungi. *Environmental Pollution*, 151(3), 608-620.
- Mellem, J.J., Baijnath, H. & Odhav, B. (2012). Bioaccumulation of Cr, Hg, As, Pb, Cu and Ni with the ability for hyperaccumulation by *Amaranthus dubius*. *African Journal of Agricultural Research*, 7(4), 591-596.
- Mendez, M.O. & Maier, R.M. (2008). Phytostabilization of mine tailings in arid and semiarid environments—an emerging remediation technology. *Environmental Health Perspectives*, 116(3), 278-283.
- Nelson, D.W. & Sommers, L.E. (1996). Total carbon, organic carbon, and organic matter. *Methods of Soil Analysis: Part 3 Chemical Methods*, 5, 961-1010.
- Nouri, J., Khorasani, N., Lorestani, B., Karami, M., Hassani, A. H., et al. (2009). Accumulation of heavy metals in soil and uptake by plant species with phytoremediation potential. *Environmental Earth Sciences*, 59(2), 315-323.
- Li, M.S., Luo, Y.P. & Su, Z.Y. (2007). Heavy metal concentrations in soils and plant accumulation in a restored manganese mineland in Guangxi, South China. *Environmental Pollution*, 147(1), 168-175.
- Lombi, E., Gerzabek, M.H. & Horak, O. (1998). Mobility of heavy metals in soil and their uptake by sunflowers grown at different contamination levels. *Agronomie*, 18(5-6), 361-371.
- Ogundola, A.F., Adebayo, E.A. & Ajao, S.O. (2022). Phytoremediation: The ultimate technique for reinstating soil contaminated with heavy metals and other pollutants. In: *Phytoremediation Technology for the Removal of Heavy Metals and Other Contaminants from Soil and Water*, Elsevier, 19-49.
- Özel, H. U. & Özel, H. B. Investigation on Heavy Metal Pollution in Uludag Fir Forests (*Abies nordmanniana* subsp. *bornmülleriana* MATTF) in the Bartın Region. *Kastamonu University Journal of Forestry Faculty*, 12(3), 155-160.
- Park, B. & Son, Y. (2017). Ultrasonic and mechanical soil washing processes for the removal of heavy metals from soils. *Ultrasonics Sonochemistry*, 35, 640-645.
- Pilon-Smits, E. (2005). Phytoremediation. *Annual Review of Plant Biology*, 56, 15-39.
- Prieto, M.J., Acevedo, S.O.A., Prieto, G.F. & González, N.T. (2018). Phytoremediation of soils contaminated with heavy metals. *Biodiversity International Journal*, 2, 362-376.
- Pulford, I. D., & Watson, C. (2003). Phytoremediation of heavy metal-contaminated land by trees a review. *Environment International*, 29(4), 529-540.
- Rahman, M., Azirun, S. & Boyce, A. (2013). Enhanced accumulation of copper and lead in amaranth (*Amaranthus paniculatus*), Indian mustard (*Brassica juncea*), and sunflower (*Helianthus annuus*). *Institute of Biological Sciences*, 8(5), 1-9.
- Rafati, M., Khorasani, N., Moattar, F., Shirvany, A., Moraghebi, F., et al. (2011). Phytoremediation potential of *Populus alba* and *Morus alba* for cadmium, chromium and nickel absorption from polluted soil. *International Journal of Environmental Research*, 5(4), 961-970.
- Rezania, S., Taib, S.M., Din, M.F.M., Dahalan, F. A. & Kamyab, H. (2016). Comprehensive review on phytotechnology: Heavy metals removal by diverse aquatic plants species from wastewater. *Journal of Hazardous Materials*, 318, 587-599.
- Rezvani, M. & Zaefarian, F. (2011). Bioaccumulation and translocation factors of cadmium and lead in *Aeluropus litoralis*. *Australian Journal of Agricultural Engineering*, 2(4), 114-119.
- Rizwan, M., Ali, S., Rizvi, H., Rinklebe, J., Tsang, D. C., et al. (2016). Phytomanagement of heavy metals in contaminated soils using sunflower: A review. *Critical Reviews in Environmental Science and Technology*, 46(18), 1498-1528.
- Roy, S., Labelle, S., Mehta, P., Mihoc, A., Fortin, N., et al. (2005). Phytoremediation of heavy metal and PAH-contaminated brownfield sites. *Plant and Soil*, 272(1/2), 277-290.
- Sağlam, B., Bilgin, A. & Aybar, M. (2020). Assessment of heavy metal pollution in soil and sediments of Murgul copper mine and its surroundings. *Kastamonu University Journal of Forestry Faculty*, 20(1), 25-37.

- Sharma, S., Singh, B. & Manchanda, V.K. (2015). Phytoremediation: Role of terrestrial plants and aquatic macrophytes in the remediation of radionuclides and heavy metal contaminated soil and water. *Environmental Science and Pollution Research*, 22(2), 946-962.
- Šijakova-Ivanova, T., Boev, B., Zajkova-Paneva, V., Boev, I. & Karakaševa, E. (2017). Bioaccumulation and translocation factor of heavy metals in the plants *Linaria* sp., *Moricandia* sp. and *Viola lutea* Huds from the Alšar locality–Republic of Macedonia. *Geologica Macedonica*, 31(2), 143-156.
- Sheoran, V., Sheoran, A.S. & Poonia, P. (2010). Soil reclamation of abandoned mine land by revegetation: A review. *International Journal of Soil, Sediment and Water*, 3(2), 13.
- Tözsér, D., Tóthmérész, B., Harangi, S., Baranyai, E., Lakatos, G., et al. (2019). Remediation potential of early successional pioneer species *Chenopodium album* and *Tripleurospermum inodorum*. *Nature Conservation*, 36, 47-69.
- Töre, G.Y. & Özkoç, Ö.B. (2022). Recent developments in aquatic macrophytes for environmental pollution control: A case study on heavy metal removal from lake water and agricultural return wastewater with the use of duckweed (Lemnaceae). In: *Phytoremediation Technology for the Removal of Heavy Metals and Other Contaminants from Soil and Water*. Elsevier, 75-127.
- Ucaroglu, S. & Talinli, I. (2012). Recovery and safer disposal of phosphate coating sludge by solidification/stabilization. *Journal of Environmental Management*, 105, 131-137.
- USEPA, (1996). *US Environmental Protection Agency, EPA-Method 3052, Microwave assisted acid digestion of siliceous and organically based matrices*. US Government Printing Office, Washington, DC.
- Wu, W., Wu, P., Yang, F., Sun, D.L., Zhang, D. X., et al. (2018). Assessment of heavy metal pollution and human health risks in urban soils around an electronics manufacturing facility. *Science of the Total Environment*, 630, 53-61.
- Yazdanbakhsh, A., Alavi, SN, Valadabadi, SA, Karimi, F. & Karimi, Z. (2020). Heavy metals uptake of salty soils by ornamental sunflower, using cow manure and biosolids: A case study in Alborz city, Iran. *Air, Soil and Water Research*, 13, 1-13.