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Research Article
Determination of Heavy Metal Accumulation in Some Coniferous Species Used in Kastamonu Urban Afforestation

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Abstract
Background and Objective: Determination of heavy metal concentrations in plants and soil is a significant exercise due to its importance in diagnosing the issue and developing environmental and forestry strategies. The main aim is to determine the variation of heavy accumulation depending on plant type in some coniferous species used in afforestation in the city of Kastamonu. **Materials and Methods:** An analytical quantitative technique (ICP-OES: Inductively Coupled Plasma Optical Emission Spectroscopy) is used for the determination of the number of heavy metals in samples obtained from different plant species. Cr, Ni, Fe, Mg, Cu, Ca, Zn, Mn, Pb and Cd concentrations are determined. **Results:** The results of the research demonstrated that most of the species recorded high concentrations of heavy metal, which is attributed to the closeness of the sampling location to a heavy traffic road. Anatolian black pine had the lowest concentrations in five out of the ten heavy metals, followed by *Picea pungens* with three. *Thuja orientalis* had the highest concentrations in four out of the ten heavy metals. **Conclusion:** Out of the several species tested for heavy metal concentrations in this research, certain species showed significantly higher concentrations than others. The overall high concentrations of heavy metals within the tested samples is attributed to the closeness of the sampled plants from roads and highways.

Key words: Heavy metals, forestry, ICP-OES, inductively coupled plasma, ecological elements, domestic effluents, fumes

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Pollution has been one of the most urging subjects worldwide due to the variety of its sources, resulting mainly from human activities and its impacts on the ecological life of different forms. One of the most affected ecological elements are plants, which their pollution can be measured through several test types by understanding the heavy metal content in them. Some heavy metals are essential for the life of the plants, such as Copper (Cu), Zinc (Zn), Iron (Fe), Manganese (Mn), Molybdenum (Mo), Nickel (Ni) and Cobalt (Co). Nonetheless, some other heavy metals are not needed physiologically for the plants and on the contrary, they could impose a toxic effect on the plants themselves as well as animals and humans[1]. Heavy metals exist naturally in the ecological system with a specific balance, however, due to the industrial and agricultural developments of human activities, humans have been exposed more to them. Heavy metals in the environment have several sources[2]: Geogenic: Geological activities, such as volcanic raptures, Industrial: Manufacturing and processing, Agricultural: Fertilizers, Pharmaceutical: Drugs containing heavy metals, Domestic effluents: Liquid waste, Emission and atmospheric: Exhausting smoke and fumes.

The pollution with heavy metals that are found in plants can be caused by several sources, however, the 2 main means are caused by soil pollution or through air pollution as the body of the plants can be exposed to heavy metal ions and the plant can absorb them through the leaves[3]. There are several possible sources for plants heavy metal concentrations, including irrigation water quality, industrial facilities nearby and roads nearby[4]. Other literature sources state that the abnormal heavy metal concentrations in plantations have adverse effects on the plant and the subsequent users of the plants in the food chain. The impacts on the plant include abnormal growth and toxicity. Furthermore, the impacts for the subsequent food chain users, including animals and humans are the impairment of health[5].

The impact of heavy metals extends from the effects on soil and plants to human health. In plants, although the presence of some heavy metals in balanced concentrations is essential for growth and metabolism (e.g., Co, Cu, Fe, Zn), the consumption of these plants by animals and humans could have toxic effects[3]. Studies compared the uptake of heavy metals through several surfaces, routes and pathways into the plant, where the results suggest different effects depending on the source (atmosphere versus soil)[6]. Several species of trees are used for ornamentation purposes, where many of them are located near roads with different traffic volumes. It is expected that fumes and smoke emitted from traffic affect the soil and trees, while the distance from the road and the traffic volume may play a role in the toxicity level.

The significance of this research emerges from the outcomes of the experiments that could clarify the effect of transportation on the ecological system and consequently the impacts on other elements in the system, including animals and humans. Through understanding the correlation between traffic pollution and heavy metal concentrations, solutions can be developed to reduce toxic content, while the study could impact the environmental strategy implemented in Turkey.

The topic addressed in the current research is widely common in the literature due to its importance in diagnosing the issue and developing environmental and forestry strategies. Moreover, the impact of heavy metals on forests and plantations leads to increased interest in the subject, which can be observed through the literature and the methods used for determination.

A group of authors researched the concentrations of lead and zinc in plantation and soil near an environmentally compromised site in Pakistan. Two locations were studied, contaminated and reference, with samples collected from the soil and the plants in each one of them. Using an atomic absorption spectrometer, the results show that the contaminated site had higher concentrations in the soil and the plantation of several heavy metals, where most of the tested for heavy metals showed a significant difference between the 2 sites using an ANOVA statistical testing[7].

Another study tested heavy metal concentrations in the soil and vegetation beside traffic roads in Nepal. Four different profiles were considered during sample collection: Location, sample type, distance (0, 10, 30, 50 and 100 m) and plantation type[8].

The main aim is to determine the variation of heavy accumulation depending on plant type in some coniferous species used in afforestation in the city of Kastamonu, Turkey.

MATERIALS AND METHODS

Study area: The samples were collected between October and November, 2020 from afforested areas around Kastamonu city in Turkey. The areas were close to roads and highways. The research duration is from July, 2020-March, 2021.

Sample collection: For this purpose, leaf samples were collected from individual plants that grew in areas with high-density traffic in Kastamonu, which are widely used in afforestation works.

Methodology: A spectrometric method is going to be used for the analysis of the heavy metal concentration for the experimental part as it is considered a reliable method used in several similar applications in the literature[9,10].

The spectrometric method is used in several studies that experimented the heavy metals concentrations in plants. The method requires using analytical grade chemicals including nitric acid, hydrogen peroxide and perchloric acid. Moreover, a flame atomic absorption spectrophotometer is used to analyse the heavy metal content of the samples. Several studies used the same method for heavy metal content analysis. Therefore, the main opportunity is the ability to compare the obtained results with several studies leading to a solid conclusion[11-13].

An analytical quantitative technique (ICP-OES): Inductively Coupled Plasma Optical Emission Spectroscopy) is used for the determination of the number of heavy metals in samples obtained from different plant species. The concept of the method is based on the difference of energy absorption of the atoms of each heavy metal, where electrons from the last level of energy are excited. The wavelengths emitted from the transition from a higher to lower energy level is captured after inducing a 10,000 Kelvin heat from an argon plasma. Thereafter, the relationship between the concentration of an element and the emitted light intensity is described using the Beer-Lambert law[14].

The ICP-OES process starts by feeding the sample into the instrument, followed by exciting the atoms using an argon plasma and controlled by a radio frequency generator. The intensity of the emitted light resulting from the return of electrons of metal atoms to lower energy levels is measured using a polychromator and a detector. The concentration of heavy metal in the sample is then calculated according to the calibration graph fed into the machine at the beginning[14].

In the samples collected, washing was performed and kept in the drying stage. Samples classified and labelled in the laboratory are kept for 15 days until they became air-dry. Samples that become air dry are dried in the oven at 45° for a week. Thereafter, plant samples are grounded, powdered, weighed in 0.5 g and placed in tubes designed for microwaves. About 10 mL of 65% HNO_3 is added to the samples. During these processes, the fume hood is worked on. The prepared samples are burned in the microwave device at 280 psi pressure and 180°C for 20 min. The tubes are removed from the microwave after the processes are completed and left to cool.

Deionized water is added to the cooled samples to be completed to 50 mL. After the prepared samples are filtered from the filter paper, heavy metals are determined by reading in the appropriate wavelengths in the ICP-OES device. Cr, Ni, Fe, Mg, Cu, Ca, Zn, Mn, Pb and Cd concentrations are determined by making heavy metal analysis in prepared samples.

Statistical analysis: Statistical analysis is performed according to the type and traffic density of the determined heavy metal quantities.

RESULTS

An analysis was performed on nine plant species for the concentration of ten heavy metals: Ca, Mg, Cd, Cr, Cu, Fe, Pb, Mn, Ni and Zn. Three readings were taken from each species and the mean was used to perform statistical analysis and comparison between them. Table 1 shows the mean values of the readings obtained for each heavy metal from each species. Ca and Mg are measured in ppm, while the rest of the heavy metals are measured in ppb. Ca has the highest concentration in most species (mean = 35.758 ppm), followed by Mg (mean = 10.996 ppm), Fe (mean = 2.378 ppm), Mn (mean = 0.159 ppm), Zn (mean = 0.101 ppm), Cu (mean = 0.037 ppm), Pb (mean = 0.019 ppm), Cr (mean = 0.012 ppm), Ni (mean = 0.011 ppm), then Cd (mean = 0.002 ppm).

In Ca, six of the nine species demonstrated high concentrations: Scots pine, *Abies bornmuelleriana*, *Cedrus libani*, *Cupressus arizonica*, *Picea pungens* and *Picea abies*. The lowest Ca concentrations were found in *Cryptomeria Japonica*, Anatolian black pine and *Thuja orientalis*, respectively. In Mg, the highest mean value was demonstrated by *Cryptomeria Japonica*, followed by *Abies bornmuelleriana* with more than 5 ppm. Most of the species averaged between 10 and 12 ppm. The lowest Mg concentrations were demonstrated by *Picea pungens* and *Thuja orientalis*, respectively, with Mg concentrations around 5 ppm. In Cd, the highest mean value was demonstrated by *Thuja orientalis* (approx. 2.5 ppb). The remaining species averaged between 2.15 and 2.35 ppb. The lowest Cd concentration was demonstrated by Anatolian black pine (2.15 ppb).

In Cr, the highest concentrations were demonstrated by *Thuja orientalis* (approx. 22 ppb), followed by *Cedrus libani* (approx. 17.5 ppb). The lowest concentrations were demonstrated by *Picea pungens* and Anatolian black pine (approx. 6 ppb). In Cu, the highest concentrations were

Table 1: Descriptive statistics for heavy metal concentrations in different species

Species	Ca	Mg	Cd	Cr	Cu	Fe	Pb	Mn	Ni	Zn
Scots pine										
Mean	40.85	11.67	2.353	13.436	39.957	2735.51	20.058	249.042	11.026	137.685
SD	0.008	0.077	0.045	0.041	1.033	9.845	0.353	0.844	0.171	0.502
Anatolian black pine										
Mean	26.545	9.939	2.156	6.311	24.288	931.822	10.399	150.938	5.563	65.143
SD	0.29	0.056	0.02	0.162	0.859	6.629	0.449	1.391	0.159	0.739
Abies bornmuelleriana										
Mean	41.003	14.926	2.234	10.708	29.656	1983.82	16.282	93.37	8.118	143.948
SD	0.006	0.169	0.01	0.091	0.797	21.453	0.204	0.362	0.078	0.77
Cedrus libani										
Mean	40.919	10.654	2.337	17.719	62.723	3736.53	19.92	130.833	16.028	89.769
SD	0.018	0.059	0.003	0.197	0.841	8.89	0.233	0.603	0.111	0.323
Cupressus arizonica										
Mean	40.613	10.367	2.301	10.532	26.4	2023.53	17.877	188.577	10.211	74.432
SD	0.012	0.057	0.006	0.06	0.767	5.912	0.494	0.197	0.081	0.189
Picea pungens										
Mean	40.516	4.289	2.227	5.966	29.539	647.778	13.661	270.499	9.299	124.59
SD	0.12	0.008	0.029	0.041	0.255	2.049	0.329	0.334	0.117	0.149
Picea abies										
Mean	40.931	11.096	2.294	12.705	44.738	2366.5	18.215	79.528	13.677	141.092
SD	0.042	0.182	0.02	0.077	0.342	19.789	0.358	0.576	0.04	1.454
Thuja orientalis										
Mean	30.906	5.091	2.516	21.348	45.009	4854.21	27.655	165.299	15.197	77.258
SD	0.598	0.091	0.02	0.181	0.567	9.646	0.631	0.297	0.164	0.12
Cryptomeria japonica										
Mean	19.539	20.935	2.265	12.249	32.882	2120.28	22.52	106.552	10.33	58.977
SD	0.006	0.238	0.023	0.129	0.602	6.89	0.489	0.108	0.095	0.165

demonstrated by *Cedrus libani* (approx. 63 ppb), which was much higher than the second-highest *Picea abies* and *Thuja orientalis* (approx. 45 ppb). The lowest concentrations were demonstrated by Anatolian black pine (24 ppb), followed by *Cupressus arizonica* (28 ppb). In Fe, the highest concentrations were demonstrated by *Thuja orientalis* (4900 ppb), followed by *Cedrus libani* (3800 ppb). The lowest concentrations were demonstrated by *Picea pungens* (800 ppb), followed by Anatolian black pine (1000 ppb). The remaining species had Fe concentrations between 2000 ppb and 2800 ppb.

In Pb, the highest concentrations were demonstrated by *Thuja orientalis* (28 ppb), followed by *Cryptomeria japonica* (23 ppb). The lowest concentrations were demonstrated by Anatolian black pine (11 ppb), followed by *Picea abies* (14 ppb). The remaining species averaged Pb concentrations between 16 and 21 ppb. In Mn, the highest concentrations were demonstrated by *Picea abies* (270 ppb), followed by Scots pine (250 ppb). The lowest concentrations were demonstrated by *Picea abies* (80 ppb), followed by *Abies bornmuelleriana* (100 ppb). The remaining species had Mn concentrations exceeding 100 ppb and lower than 200 ppb. In Ni, the highest concentrations were demonstrated by *Cedrus libani* (16 ppb), followed by *Thuja orientalis* (15 ppb) and *Picea abies* (13.8 ppb). The lowest concentrations were demonstrated by Anatolian black pine (5.8 ppb), followed by *Abies bornmuelleriana* (8.1 ppb). The other 4 species had Ni concentrations between 9.5 ppb and 11 ppb. In Zn, 4 species demonstrated the highest concentrations: *Abies bornmuelleriana* (145 ppb), *Picea abies* (140 ppb), Scots pine (137 ppb) and *Picea pungens* (125 ppb), respectively. The lowest concentrations were demonstrated by *Cryptomeria japonica* (59 ppb), followed by Anatolian black pine (64 ppb). The remaining 3 species had Zn concentrations of approximately 80 ppb.

The one-way ANOVA testing of the heavy metal concentrations showed significant differences at the 0.05 level for all tested plant species: Ca: $F(8, 18) = 3860.141$, $p = 0.000$, Mg: $F(8, 18) = 466.633$, $p = 0.000$, Cd: $F(8, 18) = 57.984$, $p = 0.000$, Cr: $F(8, 18) = 4818.247$, $p = 0.000$, Cu: $F(8, 18) = 875.573$, $p = 0.000$, Fe: $F(8, 18) = 36509.857$, $p = 0.000$, Pb: $F(8, 18) = 436.035$, $p = 0.000$, Mn: $F(8, 18) = 32339.336$, $p = 0.000$, Ni: $F(8, 18) = 2372.898$, $p = 0.000$ and Zn: $F(8, 18) = 8920.111$, $p = 0.000$.

A summary of the ANOVA and posthoc testing is shown in Table 2 for the differences between each species and other species for each heavy metal. Anatolian black pine had the lowest concentrations in five out of the ten heavy metals, followed by *Picea pungens* with three. *Thuja orientalis* had the highest concentrations in four out of the ten heavy metals.

Table 2: Summary of ANOVA and posthoc testing for differences in heavy metal concentrations between plant species

Species	Ca	Mg	Cd	Cr	Cu	Fe	Pb	Mn	Ni	Zn
Scots pine	*	**	*	**	**	**	**	**	**	**
Anatolian black pine	**L	**	**L	**	**L	**	**L	**	**L	**
Abies bornmüelleriana	*	**	*	**	**	**	**	**	**L	**H
Cedrus libani	*	**	*	**	**H	**	**	**	**H	**
Cupressus arizonica	*	**	*	**	**	**	**	**	**	**
Picea pungens	*	**L	*	**L	**	**L	**	**H	**	**
Picea abies	*	**	*	**	**	**	**	**L	**	**
Thuja orientalis	**	**	**H	**H	**	**H	**H	**	**	**
Cryptomeria japonica	**L	**	*	**	**	**	**	**	**	**L

*Significant differences with some species, ** Significant differences with all or most species, L: Lowest significant concentration and H: Highest significant concentration

Most heavy metals concentrations showed significant differences between all species, which are marked in the table with 2 stars (**). Ca and Cd showed that Anatolian black pine and *Thuja orientalis* had significant differences with all the species, while the rest of the species had few significant differences as marked with 1 star (*). Anatolian Black pine recorded the lowest concentrations in five heavy metals: Ca, Cd, Cu, Pb and Ni as marked with the Letter (L). Similarly, *Abies bornmuelleriana* had one of the lowest concentrations of Ni, *Picea abies* had the lowest concentrations of Mn and *Cryptomeria japonica* had one of the lowest concentrations of Ca. *Picea pungens* had the lowest concentrations in several heavy metals: Mg, Cr, Fe. The species that recorded the highest concentrations in every heavy metal are marked with the letter (H).

DISCUSSION

For Scots pine, a study measured the accumulation of Cd, Cr, Ni and Pb concentrations in the uncontaminated forest in Poland. The concentrations in the leaves (needles) of the trees averaged 0.10 ppm for Cd, 1.10 ppm for Cr, 2.89 ppm for Ni and 2.10 ppm for Pb. The results of the current study showed Cd concentration in Scots pine of 2.353 ppm, Cr concentration of 13.436 ppm, Ni concentration of 11.026 ppm and Pb concentration of 20.058, which are higher than those recorded by the compared study[15]. Another study investigated the accumulation of heavy metals of Scots pine in Erzincan. Seven heavy metals were tested: Pb, Co, Cu, Zn, Cr, Ni and Fe[16]. The investigation was carried out in 5 locations inside and outside the city through three samples: Soil, trunk and branch. Ni in branches was around 7 ppm, Fe in similar specimens ranged between 500 and 800 ppm, Cu for the control sample was 45 ppm and for other samples ranged between 2 and 9 ppm, Zn for control was 27 ppm and for other samples ranged between 6 and 12 ppm, Pb for control was 2.5 ppm and for other samples ranged between 0.8 and 1.0 ppm, Co ranged between 0.35 and 0.55 ppm and Cr ranged between 3 and 4 ppm for all samples. Cu concentration in the current research was recorded as 39.957 ppm for Scots pine, Fe concentration as 2735.51 ppm and Zn concentration as 137.685. Cu had a similar level of concentration as the compared study[16], while Fe and Zn showed higher concentrations in the current study.

In Anatolian black pine, a study found the concentrations of different heavy metals in the needles including ones that are included in the current study. Ca concentration was found as 2312 ppm, Cd concentration as 0.024 ppm, Cr concentration as 0.99 ppm, Cu concentration as 6.37 ppm, Fe concentration as 43.5 ppm, Mg concentration as 1543 ppm, Mn concentration as 22.8 ppm, Ni concentration as 0.53 ppm, Pb concentration as 0.14 ppm and Zn concentration as 15.6 ppm[17]. In the current research, Ca concentration was recorded as 26.545 ppm, Cd concentration as 2.156 ppm, Cr concentration as 6.311 ppm, Cu concentration as 24.288 ppm, Fe concentration as 931.822 ppm, Mg concentration as 9.939 ppm, Mn concentration as 150.938 ppm, Ni concentration as 5.563 ppm, Pb concentration as 10.399 ppm and Zn concentration as 65.143 ppm. All concentrations in the current study were higher than the ones recorded by the compared study[17], except for Mg concentration, which was lower.

For Turkish fir (*Abies bornmuelleriana*), concentrations of heavy metals in roots were studied by a Turkish study[18]. Pb concentration ranged between 3.6 and 19.8 ppm, Ni concentration ranged between 7.8 and 35.1 ppm, Fe concentration ranged between 1827.3 and 5315.5 ppm, Zn concentration ranged between 14.9 and 38.3 ppm, Cu concentrations ranged between 4.3 and 19.7 ppm and Mn concentrations ranged between 275.9 and 585.5 ppm. The current study showed Pb concentration of 16.282 ppm, Ni concentration of 8.118 ppm, Fe concentration of 1983.82 ppm, Zn concentration of 143.948 ppm, Cu concentration of 29.656 ppm and Mn concentration of 93.37 ppm. All results were similar to the counterparts of the compared study[18], except for Zn, Cu and Mn, which were higher in the results of this research.

For *Cedrus libani*, a research studied specimens from the city centre of Konya in Turkey[19]. Samples were collected from the tree needles to measure the accumulations of heavy metals in a polluted environment: Two locations in parks and gardens beside industrial facilities and other 6 locations near highways. The mean Pb concentration was found as 1.55 ppm, mean Cu concentration was found as 2.72 ppm, mean Zn concentration was found as 14.16 ppm, mean Cr concentration was found as 7.85 ppm and mean Cd concentration was found as 0.15 ppm. In the current study, the concentration of Pb was 19.92 ppm, the concentration of Cu was 62.723 ppm, the concentration of Zn was 89.769 ppm, the concentration of Cr was 17.719 ppm and the concentration of Cd was 2.337 ppm. In comparison with the results of the literature[19], concentrations of heavy metals were higher in the current study.

For *Cupressus arizonica*, the research studied the difference in Zn, Ni and Cu concentrations in 3 areas of Isfahan with heavy and moderate traffic volumes[20]. Leaf concentrations were found as follows: Zn at heavy traffic was 1.11 ppm and for moderate traffic 3.16 ppm, Ni at heavy traffic was 1.96 ppm and for moderate traffic was 1.36 ppm and Cu at heavy traffic was 2.58 ppm and for moderate traffic was 1.74 ppm. In the current research, Zn concentration was recorded as 74.432 ppm, Ni concentration as 10.211 ppm and Cu concentration as 26.4 ppm. The concentration of heavy metals in the current study exceeded the higher limits that were presented in the compared study[20].

For blue spruce (*Picea pungens*), few studies addressed using it as a bio-monitor for heavy metal accumulations. A study measured the accumulation of Mn in the species at different ages[21]. Trees at 1 year old had an average of 43.46 ppm and reached 88.14 ppm at 7 years old. The increase was not linear, while some intermediate ages had lower Mn concentrations than the average of the 1st year. Mn concentration for blue spruce was recorded as 270.499 ppm, which is higher than the compared study[21].

Moreover, the research investigated the concentrations of Zn, Ni, Co, Cr, Cd and Pb in *Picea pungens*[22], where Zn concentrations ranged between 88 and 170 ppm, Ni concentrations ranged between 2.814 and 12.261 ppm, Co concentrations ranged between 0.557 and 43.742 ppm, concentrations of Cr ranged between 2.130 and 24.736 ppm, concentrations of Cd ranged between 0.243 and 1.037 ppm and concentrations of Pb ranged between 1.14 and 59.46 ppm in leave samples. In comparison with the results of the current research, Zn fell within the range with an average of 124.59 ppm, Ni fell within the range with an average of 9.299 ppm, Co was not included, Cr fell within the range closer to the lower bound with an average of 5.966 ppm, Cd was higher than the range with 2.227 ppm and Pb fell within the range closer to the lower bound with 13.661 ppm. They compared study[22], indicated that the species can be used as a biomonitor for heavy metals in the Kastamonu region.

For *Picea abies*, the research studied the concentration of Cd, Pb, Zn and Cu in several exotic and natural species in the city of Samsun, Turkey[23]. The concentration of Cu in *Picea abies* leaves were averaged at 13 ppm, the concentration of Zn was averaged at 28 ppm, the concentrations of Cd were averaged at 0.08 ppm and the concentrations of Pb were averaged at 3.5 ppm. In the current research, Cu concentration was recorded as 44.738 ppm, Zn concentration as 141.092 ppm, Cd concentration as 2.294 ppm and Pb concentration as 18.215 ppm. Concentrations of heavy metals in the current study are higher than those recorded by the compared study[23].

For *Thuja orientalis*, the research evaluated the concentrations of Fe, Zn, Cu, Ni, Pb and Cd in species[24]. The assessment was carried in 2 locations: Polluted and non-polluted. Fe non-polluted concentration was recorded as 400 ppm and polluted as 480 ppm. Zn non-polluted concentration was recoded as 12 ppm and polluted as 13 ppm. Cu non-polluted concentration was recorded as 4 ppm and polluted as 4.5 ppm. Ni non-polluted concentration was recorded as 3.2 ppm and polluted as 3.4 ppm. In the current research, Fe concentration was recorded as 4854.21 ppm, Zn concentration as 77.258 ppm, Cu concentration as 45.009 ppm and Ni concentration as 15.197 ppm. All concentrations were deemed higher in the current study.

Overall, the results of the research demonstrated that most of the species recorded high concentrations of heavy metal, which is attributed to the closeness of the sampling location to a heavy traffic road. Anatolian black pine had the lowest concentrations in five out of the ten heavy metals, followed by *Picea pungens* with three. *Thuja orientalis* had the highest concentrations in four out of the ten heavy metals. The species used in the study can also be used as biomonitors for heavy metal concentrations when compared with the concentrations indicated for the same species in other experiments of the literature.

CONCLUSION

In one-way ANOVA testing, all plants showed significant differences between them at the 0.05 level. Therefore, a posthoc test was carried out to understand the nature of these differences. The results of the Tukey HSD test confirmed the findings of the descriptive and graphic statistics. Finally, Anatolian black pine had the lowest concentrations in five out of the ten heavy metals, followed by *Picea pungens* with three. *Thuja orientalis* had the highest concentrations in four out of the ten heavy metals. The overall results of the research demonstrated that most of the species recorded high concentrations of heavy metal, which is attributed to the closeness of the sampling location to a heavy traffic road.

SIGNIFICANCE STATEMENT

The current research discovered that plant species that are used as biomarkers in afforestation accumulate heavy metals with different extents. Nonetheless, the closeness of heavy metal sources, including highways, roads, industrial zones, etc. contribute significantly to increasing these concentrations. The results of the study help researchers understand the ability of side number of species in accumulating different heavy metals, which contributes to afforestation planning and strategies according to land use. Moreover, the findings of the experiment can be used to develop policies and government rules that can minimize the accumulation of heavy metals in afforestation.

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