

Do *Picea pungens* engelm. organs be a suitable biomonitor of urban atmosphere pollution?

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SILVICULTURE

ABSTRACT

Background: Heavy metal contamination in the air has been gaining importance from scientists due to increasing industrial activities, population growth, and density in urban areas causing many crucial environmental pollution problems. Heavy metals do not quickly disappear and do not decay in nature; they accumulate in plant organs and indirectly affect human and environmental health. On the other hand, some elements are essential for plant growth, but some have poisonous or carcinogenic consequences, even at small concentrations. Thus, determining and observing heavy metal concentrations in the atmosphere is crucial. Plants are the utmost important indicator of heavy metal concentration changes in the air and are called biomonitors. The current study aims to determine the variation in some elements (K, P, Fe, Zn, Cu, Cd, and Cr) concentration in *Picea pungens* Engelm. (valued for its bluish or silvery-gray foliage) grown in the Ankara city center (capital of Türkiye and second crowded city), based on the plant needles and the age of the branch. Thus, this study tried to determine the potential of using needle leaves and branches of *P. pungens* as a biomonitor to determine some heavy metal concentrations.

Results: The heavy metal concentrations significantly differed by organ and organ age. The element levels were higher in the branches in some years and the needles in some years.

Conclusions: This result indicates that *Picea pungens* needles and branches are suitable for observing studied heavy metal concentrations. Using branches and needles does not also harm the plant species.

Keywords: Blue spruce; Cadmium; Chromium; Copper; Heavy metal

HIGHLIGHTS

Heavy metals have poisonous and harmful environmental effects, which are needed to monitor and reduced by using plants as biomonitors.

The metal accumulation in plants is differed by organ and organ age.

The *Picea pungens* Engelm branches and needle leaves are suitable for monitoring atmospheric metal pollutions.

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INTRODUCTION

Production efforts to meet the increasing and diversifying demands and needs of people have resulted in the pollution of water, soil, and air, and as a result, environmental contamination has become one of the utmost critical concerns of the 21st century and today (Leonard et al., 2020; Sevik et al., 2020a; Koç, 2021a; Isinkaralar et al., 2022a, b). Especially in industrialized countries with an increased population in urban areas, air pollution is a much more severe problem and has begun to affect the health of millions of people around the world (Isinkaralar, 2022a,b; Varol et al., 2022; Cetin; Abo Aisha, 2023).

Heavy metals are of specific prominence among the factors of air pollution. In general, heavy metals not only disappear and degrade in nature quickly, but they also bioaccumulate in living things, and some have lethal effects even in a low amount (Turkyilmaz et al., 2020; Koç, 2021a). Some heavy metals (Zinc (Zn), cadmium (Cd), chromium (Cr)) that originate from industrial activities are carcinogenic (Zhang et al., 2018; Aricak et al., 2019; Key et al., 2022), while some (Cd, mercury (Hg), lead (Pb), Cr) are poisonous and toxic for humans and plants with their low amount (Shahid et al., 2017).

The release of toxic pollutants has caused the buildup of heavy metals into the environment, a critical global problem because of industrial advancement, urbanization, and increased agricultural activities. Organic compounds such as solvents, polyatomic hydrocarbons, heavy metals, and pesticides are the primary environmental contaminants that can have lethal effects even in low quantities. Motor vehicles, energy production centers, mines, agriculture and urbanization-industrial activities, volcanic activities, fertilizers (Nitrogen (N), phosphorus (P), potassium (K), Zn, iron (Fe), copper (Cu)), and pesticides are including the primary heavy metal sources (Memon; Schröder, 2009; Aydın; Pakyürek, 2020). In contrast, fertilizers, including plant nutrients (macro and microelements), play an essential role in plant growth, development, and physiological reactions in plants (Koç, 2021b, 2022; Çobanoğlu et al., 2022) not only in their natural growth areas but also in plantation areas (Koç et al., 2022) and marginal lands (Shults et al., 2020). Exceeding the optimum range of fertilizer causes soil contamination, thus accumulating more in plants.

The utmost familiar heavy metal contaminants are Cr, Pb, Cu, Cd, Hg, Ni, and Zn. Traffic and industrial activities are the primary sources of heavy metal contamination in the air. Vehicle traffic significantly affects areas near freeways by raising heavy metal contamination. Heavy metal concentrations such as Cd, Ni, and Pb in areas with heavy traffic increase due to emissions from road traffic (Li et al., 2016; Shahid et al., 2017; Alaçouri et al., 2020a; Aydın; Pakyürek, 2020; Hosseini et al., 2020; Isinkaralar, 2022c). Due to the sink of heavy metal contaminants released into the air from vehicle exhaust smokes, heavy metal accumulation follows in various organs of plants and soil (Cesur et al., 2022). Heavy metals accumulate in the bodies of plants in their biological cycle and can cause significant damage to plants. Moreover, heavy metals can affect human health,

resulting in chronic diseases and cancers, and most of these diseases have limited treatment opportunities and usually end in death (Dabass et al., 2016; Özbolat Tuli, 2016).

Due to the importance of the adverse heavy metals effect on environmental and human health, various studies have been performed to observe metal contamination in the atmosphere. In addition, direct measurement for heavy metal pollution often requires expensive measuring instruments. Therefore, biomonitors are often preferred to determine heavy metal contamination in the environment. In addition to being simple and inexpensive, this method can offer more trustworthy information about the periodic variation of heavy metal ratios. When exposed to heavy metal pollution, plants collect heavy metals in their body parts, such as leaves, wood, and fruit. Plants also show the sequence of the rise in heavy metal rates in the airborne over time. Hence, instead of directly tracking heavy metal contamination, biomonitors are often used as a marker of contamination (Matin et al., 2016; Ugulu et al., 2016; De Nicola et al., 2017). The most critical problem is determining which plants are more suitable and useful as biomonitors.

Leaves are mostly used to observe the heavy metal concentration change in the atmosphere. Using leaves gives reliable results in detecting heavy metals accumulated during a vegetation period, especially in deciduous species. However, this method does not provide sufficient information about the year heavy metals accumulate in evergreen plants. The needle leaves in evergreen species can remain on the plants for several years. It is possible to know how old the needles are with the help of the nodes formed in the conifers. Therefore, studies on species such as spruce, fir, and pine provide vital evidence about the heavy metal concentrations change in the recent past, but at the same time, the tree is damaged (Turkyilmaz et al., 2018a; Keçeci, 2019).

Determining the heavy metal pollution in the atmosphere is expensive and difficult from various aspects, which is why biomonitors are preferred for observation. Using a single tree for observing heavy metals in plants and organs avoids divergences from the genetic structure. It is well-known that plant development and expansion are formed by the related interaction of environmental features and genetic codes (Koç, 2019; Koç et al., 2022). Thus, similar plants with different genetic codes may have several phenotypic and physiological traits even under similar environmental factors (Koç, 2019; Varol et al., 2022), affecting the possible heavy metal accretion in plants (Karacocuk et al., 2022). Working on the same single tree eliminates the genetic differences.

Although *Picea pungens* Engelm (Blue spruce) is naturally distributed in North America, it has been grown in many countries due to its widespread use in landscaping and as a Christmas tree due to thick branches and a conical crown when free-growing (Černý et al., 2016). They are also preferred species in the landscape, parks, and gardens due to their resistance to summer drought, toxic gases, and acid rain able to grow in dry and poor soils (Çobanoğlu, 2019).

This study aimed to determine the variation of several heavy metal concentrations in needles and branches of different ages of *Picea pungens* grown in a heavily

trafficked area in Ankara, Türkiye ($18.4 \mu\text{g}/\text{m}^3$), one of the most polluted cities in Europe (IQAir, 2019). Thus, it has been tried to determine the potential of using needles and branches of *Picea pungens* as a biomonitor to determine the studied metal element levels in the previous years.

MATERIAL AND METHODS

Description of the study area

The current research was implemented on the samples obtained from the branches of a tree facing the road, approximately 2 m from the road, on the side of the Ulus-Kızılay highway in the Ulus district located at Gençlik Park in Ankara city center (Figure 1). The main road has four lanes in both directions, and there was heavy traffic almost every hour during the day.

Sample collection and preparation for analysis

The branches were obtained by cutting the side branches of the *Picea pungens* tree and transferred to the

laboratory. Afterward, needles and branches were cut and sorted according to their age (Figure 2) in the laboratory, placed on cardboard, and kept in a room ($25 \text{ }^\circ\text{C}$ – without direct sunlight) for a month until they became air-dried.

Samples were placed in a drying oven at $45 \text{ }^\circ\text{C}$ for a month. Oven-dried samples were ground using a steel grinder, then 0.5 g powder of samples was put into flasks, and 10 mL nitric acid (HNO_3) was added to the flasks. Afterward, the specimens were burnt using a microwave device at $180 \text{ }^\circ\text{C}$ for 20 minutes under 280 PSI pressure and then left tubes for cooling. The cooled flasks were filled up to 50ml volume with deionized (DI) water. The preparations were filtered using $45 \mu\text{m}$ filter papers, and the solution was placed in an ICP-OES machine (GBC Scientific Equipment Pty Ltd., Melbourne, Australia) for the element analysis. At the appropriate wavelength, a total of 7 elements (K, P, Zn, Cu, Fe, Cd, and Cr) were determined in the branches and needles of *Picea pungens*. This procedure was previously applied to various metal elements in several studies (Koç, 2021a; Turkyilmaz et al., 2020; Isinkaralar et al., 2022a, b; Karacocuk et al., 2022; Key et al., 2022). The changes in these heavy metals were evaluated based on the organ and year.

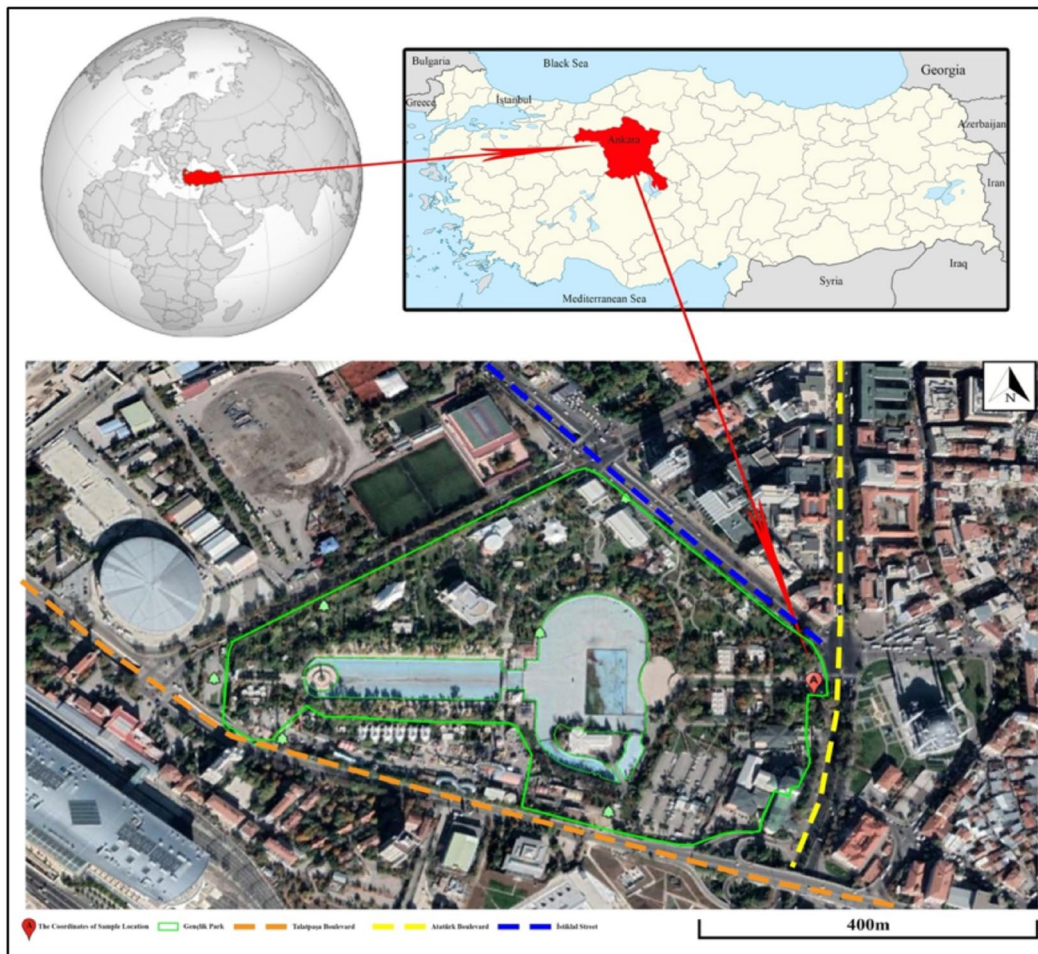


Figure 1: The sample location in Gençlik Park, Ankara.

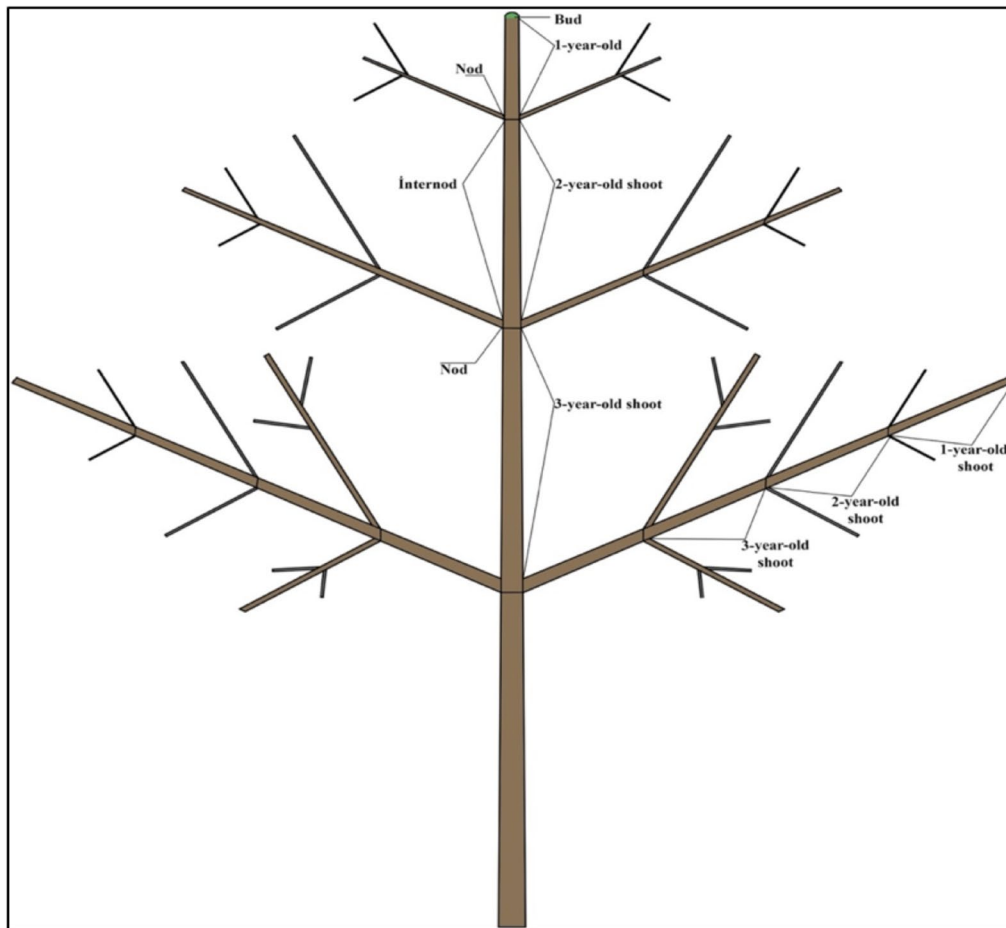


Figure 2: The determination of ages in *Picea pungens* Engelm.

Data analysis

SPSS 21.0 statistical package software was used to complete all analyses. The obtained data from *Picea pungens* branches and needle leaves were subjected to ANOVA (analysis of variance) and then the Duncan test for studied elements. Alpha=0.05 was used for statistical significance. The results were simplified and interpreted.

RESULTS

Element concentrations change in the tree branch and needle

The ANOVA determined whether there were significant ($p < 0.05$) changes between the mean concentrations of the elements in branches and needles and the element concentrations based on organs (Table 1). The variation of all elements concentrations between branches and needles was significant ($p < 0.05$) except Cu and Cr ($p > 0.05$). Among the elements with a statistically significant difference, the K and Zn concentrations in the branches had higher mean values than the needles. In contrast, the P, Fe, and Cd concentrations in the needles had higher values than in the branches.

Potassium (K) concentration changes by plant organs and year

The variation of K (ppm) concentration by organ and year is given in Table 2. The variations in K concentration determined in branches and needles had a significant difference ($p \leq 0.001$) in all years except 2014. While the K concentration was higher in the branches than in the needles in 2013, 2015, 2018, and 2019, the needles had a higher K concentration than in the branches within 2016 and 2017 (Table 2). Another significant outcome is that there are no significant K concentration changes between the branches and needles. For instance, the most significant K concentration difference was detected in 2019 in that the branches were approximately 1.93 times higher mean value than the needles. When the changes based on years are examined, according to the analysis of variance, the K concentration varies in both branches and needles yearly was significant ($p \leq 0.001$). The K concentration in the branches varies between 4785.5 ppm (in 2017) and 9247.0 ppm (in 2019), and in the needles, it varies between 4787.7 ppm (in 2019) and 7954.0 ppm (in 2016). It is tough to say that the K concentration change based on year is at a meaningful level.

Table 1: Change of element concentration by plant organs.

Element	Branches	Needle	F-value
K (ppm)	6770.6 b	5725.1 a	7.526**
P (ppm)	809.0 a	955.6 b	5.657*
Fe (ppm)	276.2 a	413.4 b	7.176*
Zn (ppm)	69.6 b	41.0 a	36.219***
Cu (ppb)	9562.0	8984.8	0.332ns
Cd (ppb)	48.4 a	62.0 b	7.447**
Cr (ppb)	1975.7	2048.0	0.107ns

Lower case number indicates a significant difference within plant organs in each element. Different letters indicate significant differences within plant organs for each element. ns: not significant. * p<0.05, ** p<0.01, *** p<0.001.

Table 2: Variation of K element concentration (ppm) by plant organ and year.

Year	Branch	Needle	F-value
2013	7319.0 Be	5584.3 Ac	22281.633***
2014	6113.2 c	6152.5 d	2.349ns
2015	5440.4 Bb	4839.2 Ab	857.388***
2016	7397.6 Af	7954.0 Be	218.207***
2017	4785.5 Aa	6060.2 Bd	8112.081***
2018	7091.5 Bd	4864.7 Ab	4505.259***
2019	9247.0 Bg	4787.7 Aa	27262.614***
F-value	4333.883***	9452.425***	

Upper letters refer horizontal direction, whereas lower cases indicate vertical directions. Different letters indicate significant differences within plant organs and year. ns: not significant. *** p<0.001.

Phosphorus (P) Concentration Changes by Plant Organs and Year

The variation of P (ppm) concentration by organ and year is given in Table 3. The changes in P concentration determined in branches and needles had a significant difference (p<0.001) in all years except in 2018. The P concentration determined in the needles was higher than in the branches in all years except 2019. The highest significant P concentration difference was observed in 2017, that the needles were approximately 1.98 times higher mean value than the branches. Based on years, the P concentration change in both branches and needles was determined to be significant (p<0.001). Whereas the lowest P concentration in the branches was determined in 2017 (603.7 ppm), the lowest P concentration in the needles was determined in 2019 (752.3 ppm). In contrast, the highest P concentrations in branches and needles were determined in 2019 (1114.2 ppm) and 2016 (1366.0 ppm), respectively (Table 3).

Iron (Fe) concentration changes by plant organs and year

The variation of Fe (ppm) concentration by organ and year is given in Table 4. The changes in Fe concentration

determined in branches and needles significantly differed in all years (p<0.001). The Fe concentration determined in the needles was higher than in the branches in all years except 2016. The highest significant Fe concentration difference was observed in 2015; the needles were 4.47 times higher in mean value than the branches. Based on years, The Fe concentration change in both branches and needles was determined to be statistically meaningful (p<0.001). Whereas the minimum Fe concentrations were determined in 2019 (110.5 ppm) and 2016 (153.2 ppm), the highest Fe concentrations were observed in 2014 (508.2 ppm) and 2014 (708.5 ppm) in the branches and needles, respectively (Table 4).

Table 3: Variation of P element concentration (ppm) by plant organ and year.

Year	Branch	Needle	F-value
2013	694.7 Ac	818.4 Bc	3561.216***
2014	784.6 Ad	857.8 Bd	1985.524***
2015	641.3 Ab	765.2 Bb	4941.377***
2016	889.6 Ae	1366.0 Bg	24477.084***
2017	603.7 Aa	1194.7 Bf	200052.186***
2018	935.2 f	935.0 Be	0.012ns
2019	1114.2 Bg	752.3 Aa	784.369***
F-value	1310.626***	28216.844***	

Upper letters refer horizontal direction, whereas lower cases indicate vertical directions. Different letters indicate significant differences within plant organs and year. ns: not significant. *** p<0.001.

Table 4: Change of Fe Element Concentration (ppm) By Plant Organ and Year.

Year	Branch	Needle	F-value
2013	434.1 Af	480.9 Be	314.282***
2014	508.2 Ag	708.5 Bg	3849.936***
2015	116.8 Ab	523.0 Bf	119448.290***
2016	401.7 Be	153.2 Aa	52417.091***
2017	132.3 Ac	351.6 Bc	56049.332***
2018	230.1 Ad	411.2 Bd	17835.341***
2019	110.5 Aa	265.6 Bb	8229.375***
F-value	28667.804***	12313.426***	

Upper letters refer horizontal direction, whereas lower cases indicate vertical directions. Different letters indicate significant differences within plant organs and year. *** p<0.001.

Zinc (Zn) concentration changes by plant organs and year

The variation of Zn (ppm) concentration by organ and year is given in Table 5. The alterations in Zn concentration determined in branches and needles had statistically different in all years (p<0.001). The Zn concentration determined in the branches was higher than in the needles in all years except 2019. The highest

significant Zn concentration difference was observed in 2016 when the branches were 3.16 times higher in mean value than the needles. Based on years, the Zn concentration change in both branches and needles was determined to be statistically meaningful ($p \leq 0.001$). Whereas the lowest Zn concentrations were determined in 2019 (37.7 ppm) and 2016 (28.6 ppm), the highest Zn concentrations were observed in 2014 (93.1 ppm) and 2013 (51.9 ppm) in the branches and needles, respectively (Table 5). Overall, it can be stated that the Zn concentration was found to be higher in the older branches.

Table 5: Change of Zn element concentration (ppm) by plant year and organ.

Year	Branch	Needle	F-value
2013	87.3 Be	51.9 Af	6408.205***
2014	93.1 Bg	43.3 Ad	31928.929***
2015	71.7 Bd	38.0 Ab	255025.000***
2016	90.5 Bf	28.6 Aa	67616.647***
2017	52.4 Bb	37.8 Ab	11935.563***
2018	54.6 Bc	45.7 Ae	9045.125***
2019	37.7 Aa	41.6 Bc	69.970***
F-value	6268.120***	7135.567***	

Upper letters refer horizontal direction, whereas lower cases indicate vertical directions. Different letters indicate significant differences within plant organs and year. *** $p \leq 0.001$.

Copper (Cu) concentration changes by plant organs and year

The variation of Cu (ppb) concentration by organ and year is given in Table 6. The alterations in Cu concentration determined in branches and needles had a statistically meaningful difference in all years ($p < 0.001$). The mean Cu concentration determined in the branches was higher than in the needles in 2013, 2014, 2016, and 2019. The highest significant Cu concentration difference was observed in 2015 when the needles were 1.64 times higher in mean value than the branches.

Table 6: Change of Cu element concentration (ppb) by plant organ and year.

Year	Branch	Needle	F-value
2013	10599.3 Bf	8027.1 Ac	1839.209***
2014	18270.1 Bg	12658.6 Ag	19338.134***
2015	6618.3 Ac	10866.6 Bf	111752.000***
2016	10009.2 Be	6838.1 Ab	2795.352***
2017	6067.2 Aa	9410.3 Be	7082.423***
2018	8879.7 Ad	9217.9 Bd	123.990***
2019	6490.5 Bb	5875.2 Aa	419.305***
F-value	19028.060***	6561.458***	

Upper letters refer horizontal direction, whereas lower cases indicate vertical directions. Different letters indicate significant differences within plant organs and year. *** $p \leq 0.001$.

Based on years, the Cu concentration change in both branches and needles was determined to be significant ($p \leq 0.001$). Whereas the lowest branches and needles Cu concentrations were determined in 2017 (6067.2 ppb) and 2019 (5875.2 ppb), the highest Cu concentrations were observed in 2014 (18270.1 ppb) and (12658.6 ppb) in the branches and needles, respectively (Table 6). Overall, it can be said that the Cu concentration was found to be higher on the older branches.

Cadmium (Cd) concentration changes by plant organs and year

The variation of Cd (ppb) concentration by organ and year is given in Table 7. The alterations in Cd concentration determined in branches and needles had a statistically substantial difference in all years ($p < 0.05$) except in 2014. The mean Cd concentration determined in the needles was higher than in the branches in all years except 2013 and 2016. The highest significant Cd concentration difference was observed in 2017 when the needles were 2.55 times higher in mean value than the branches. Based on years, the Cd concentration change in both branches and needles was determined to be significant ($p \leq 0.001$). Whereas the lowest branches and needles Cd concentrations were determined in 2017 (27.4 ppb) and 2016 (48.6 ppb), the highest Cd concentrations were observed in 2013 (77.6 ppb) and 2014 (77.9 ppb) in the branches and needles, respectively (Table 7).

Table 7: Change of Cd element concentration (ppb) by plant organ and year.

Year	Branch	Needle	F-value
2013	77.6 Bd	59.4 Ab	165.436***
2014	75.2 Ad	77.9 Bd	3.583ns
2015	29.4 Aa	60.7 Bb	68.356***
2016	56.9 Bc	48.6 Aa	8.693*
2017	27.4 Aa	69.8 Bc	885.959***
2018	39.9 Ab	67.3 Bc	436.771***
2019	32.1 Aa	50.5 Ba	113.284***
F-value	197.229***	47.076***	

Upper letters refer horizontal direction, whereas lower cases indicate vertical directions. Different letters indicate significant differences within plant organs and year. * $p < 0.05$. *** $p \leq 0.001$. ns: not significant.

Chromium (Cr) concentration changes by plant organs and year

The variation of Cr (ppb) concentration by organ and year is given in Table 8. The alterations in Cr concentration determined in branches and needles had a statistically substantial difference in all years ($p < 0.05$). The mean Cr amount determined in the needles was higher than in the branches in all years except 2013, 2016, and 2018. The maximum significant Cr concentration difference was

observed in 2015 when the needles were 1.81 times higher in mean value than the branches. Based on years, the Cr concentration change in both branches and needles was determined to be significant ($p \leq 0.001$). Whereas the lowest branches and needles Cr concentrations were determined in 2019 (1101.8 ppb) and 2016 (1196.7 ppb), the highest Cr concentrations were observed in 2014 (3153.0 ppb) and (3302.9 ppb) in the branches and needles, respectively (Table 8). The relationship between metal elements can deliver information on heavy metal sources. This can be done with correlation analysis.

Table 8: Change of Cr element concentration (ppb) by plant organ and year.

Year	Branch	Needle	F-value
2013	2122.4 Bd	2052.5 Ac	16.048*
2014	3153.0 Af	3302.9 Bf	20.666*
2015	1291.3 Ac	2335.5 Be	7644.786***
2016	2379.8 Be	1196.7 Aa	13878.535***
2017	1168.8 Ab	2111.1 Bd	1239.490***
2018	2612.6 Bf	2054.0 Ac	124.103***
2019	1101.8 Aa	1283.3 Bb	96.643***
F-value	1187.975***	2444.600***	

Upper letters refer horizontal direction, whereas lower cases indicate vertical directions. Different letters indicate significant differences within plant organs and year. * $p < 0.05$. *** $p \leq 0.001$. ns: not significant.

Correlation analysis between elements

The analysis of correlation was performed on the data to find out whether the relationship between the elements is statistically substantial ($p < 0.05$), and the direction and severity of the relationship and the results are presented in Table 9. As shown in Table 9, within the statistically significant relationship between elements, Cu – Cr (0.863) and Fe – Cd (0.863) elements had a robust positive correlation followed by Fe – Cr (0.844), Fe – Cu (0.727), Cd – Cr (0.721), Cu – Cd (0.720), K – P (0.579), and Zn – Cr (0.508) elements. There was a positive correlation between Zn and Cr (0.365), while a relatively strong negative relationship between P and Zn (-0.517).

DISCUSSION

In this study, the change of a total of 7 elements (K, P, Fe, Cu, Zn, Cr, and Cd) in branches and needles of *Picea pungens* by year and organ were evaluated. It was found that the element concentrations were higher in the branches in some years and the needles in some years. Similar results have been observed in several studies. For instance, in a study on *Picea pungens*, different results were obtained regarding differences between organs. The lowest Cu values were generally obtained in unwashed barks, while the lowest K values were in the washed barks (Cobanoğlu, 2019). A previous study determined that the maximum concentrations

of Zn, Cr, and Cd elements were observed in different organs of *Picea pungens* (Keçeci, 2019). As a result, the highest Zn levels were found in the branch, Ni in the needle and branch, and Cr and Cd in the needle of *Picea pungens*.

Table 9: Correlation analysis of elements.

	K	P	Fe	Zn	Cu	Cd	Cr
K	-	0.579*	-0.266	0.077	-0.030	-0.112	-0.083
P		-	-0.183	-0.517**	-0.123	0.056	-0.117
Fe			-	0.189	0.727**	0.863**	0.844**
Zn				-	0.508**	0.219	0.365*
Cu					-	0.720**	0.863**
Cd						-	0.721**
Cr							-

* $p < 0.05$. ** $p \leq 0.01$.

Especially, the high concentrations of Ni, Cr and Cd in the needles are significant because these elements are highly harmful to human and environmental health and can show lethal effects on living organisms even at little concentrations (Ghoma et al., 2023; Istanbulu et al., 2023). Due to their potential harm to humans and the environment, these elements are included in the Priority Pollutant Metals list of both ATSDR (Agency for Toxic Substances and Disease Registry) and the US EPA (United States Environmental Protection Agency) (Hsieh et al., 2004; Savas et al., 2021). For this intention, it is of great importance to monitor the variations in the concentrations of these elements in the air, and the study results show that *Picea pungens* needles are suitable biomonitors that can be used for this purpose.

In studies on monitoring heavy metals using biomonitors, leaves are the most frequently used organs (Saleh, 2018; Turkyilmaz et al., 2018b; 2020). Because since the leaves' age in deciduous plants is known, it is also necessary to know how long they are exposed to polluted air. Another reason is that removing leaves from the branches does not harm the plant's vitality and does not affect plant growth significantly, sometimes even not at all (when collected at the end of the growing season), so it is a sustainable method.

As a result of a study conducted on seven different species determined that the Cu and Cd concentrations in the branches were higher than in the leaves (Mossi, 2018). In another study conducted in four different species, the minimum K values were found in the branches, the Cd value in the seed, and the highest concentration for K and Cd were found in the seed and leaves, respectively (Erdem, 2018). However, other organs are also used extensively in monitoring element concentrations, and the variation of heavy metal concentrations based on the organ is one of the most frequently discussed issues. In studies on this subject, leaves and branches (Lei et al., 2019; Sevik et al., 2019a), bark and wood (Turkyilmaz et al., 2018a, b), leaves, branches, and seeds (Sevik et al., 2019a, b), needle, bark

and branch (Cobanoglu, 2019; Keçeci, 2019; Cetin et al., 2020), branch and leaf (Elfantazi et al., 2018), branch, leaf and fruit (Sevik et al., 2020a) determined the variances between organs in the outer bark, inner bark, and wood (Akarsu, 2019; Sevik et al., 2020b; Koç, 2021a). Studies have also determined the effect on leaf morphological and micromorphological characters (Cetin et al., 2017; Muro-González et al., 2020).

As a result, it was observed that while there could be a significant difference in element concentrations between organs, some element concentrations in needles and branches were quite close to each other. In many studies on different species and elements, element concentration changes depending on the organ have been evaluated, and similar results have been obtained. The difference between organs is about 1.8 times in Zn, 2 times in Cr, and 4.3 times in Cu in the study where the leaf, seed, and branch specimens were compared (Pinar, 2019). The Cu and Cd concentrations detected in the branches were 1.43 and 1.79 times higher than in the leaves, respectively (Mossi, 2018). Çömeten (2019) said that the concentration of Cr in *Pinus sylvestris* individuals varies significantly based on the organ, the Cr concentration in wood specimens is quite low, and the difference between wood, bark, and needle can exceed 5 times in the same branch samples. In addition, the same study revealed that the bark concentration levels were considerably higher than in the branches (Çömeten, 2019).

Studies have generally shown that heavy metal concentrations in the barks are much higher in areas with high levels of heavy metal pollution in the air (Sulhan et al., 2022; Isinkaralar et al., 2023). This situation is related to the rough bark structure and the adhesion of particulate matter infected with metals in the air to the bark surface. Therefore, the heavy metal concentrations in the barks do not correlate with the heavy metal concentrations in the air, and it is recommended that the barks should not be used as a biomonitor for monitoring the change of heavy metal concentrations in the air (Cesur et al., 2022; Cobanoglu et al., 2023).

Studies have generally indicated that the element concentration varies significantly according to the species, so some heavy metals are much more in some plant species (Sevik et al., 2019a). Furthermore, heavy metal concentrations differ in the same plants' diverse parts (Saleh, 2018). This situation is strictly linked to the plant anatomy and organ. Entering heavy metal from plant leaves differs depending on the chemical and physical characteristics of metals, as well as the forms of metals, surface texture, surface area, exposure period to heavy metals, environmental situations, plant habitus, leaf morphology, and leaf gas exchange (Shahid et al., 2017; Mossi, 2018; Hmeer, 2020). Another factor affecting the heavy metal amount in plant organs is the intensity of heavy metal contamination in the airborne and its continuity. It is claimed that traffic density and industrial activities are the primary sources of heavy metals (Shahid et al., 2017; Hong et al., 2018; Sevik, 2020).

In a study on *Pinus sylvestris* needles, the Cr concentration was 419.3 ppb in washed needles at the side of the highway, 508.9 ppb in unwashed needles, 1355.8 ppb in washed bark, 1443.7 ppb in unwashed bark, and 414.2 ppb

in woods at 100 m distance from the road (Çömeten, 2019). In individuals, it was determined that Cr concentrations were 525.5, 932.5, 274.9, 395.7, 274.7 ppb in washed bark, unwashed bark, washed needles, unwashed needles, and woods, respectively (Arıcak et al., 2020). Numerous studies demonstrated that heavy metal concentrations in plants that can be used as biomonitors vary depending on traffic density. While the Cr concentration is 16.595 ppm and 23.716 ppm in regions with no traffic and heavy traffic, respectively (Turkylmaz et al., 2018c). Similar results were observed in *Sophora japonica* leaves (Li et al., 2007), *Platanus orientalis* and *Pinus nigra* leaves (Sawidis et al., 2011), *Rosmarinus officinalis* leaves (Bozdogan Sert et al., 2019), *Nerium oleander*, *Acer negundo*, *Robinia pseudoacacia*, *Ulmus minor*, and *Platanus orientalis* leaves (Karacocuk et al., 2022).

Heavy metals can penetrate the tree body via the roots from the soil, the leaves from the air, or the stem parts (Cobanoglu et al., 2023). However, it is challenging to know how much metals detected in the tree originated from the air and the soil. In addition, weather conditions can seriously impact heavy metal entrance into the tree (Sulhan et al., 2022; Isinkaralar et al., 2023; Key et al., 2022). In addition, the metal accumulation possibility of trees cultivated under identical conditions can be diverse. Because, like all phenotypic characters, metal accumulation in trees is under the influence of genetic design as well as environmental conditions (Kuzmina et al., 2023; Erdem et al., 2023). Therefore, it is not possible to know whether the heavy metals in the tree initiate from the air or the soil and to determine the influence of the genetic design on this accumulation. In order to reach accurate results, the equalization of soil and weather conditions in biomonitors is to be used in observing the transition of heavy metal concentration in the air during the process, elimination of genetic structure-related differences, and comparison. For these reasons, it is recommended to use the organs of the same tree to determine the alteration in metal pollution in the air (Yayla et al., 2022; Cobanoglu et al., 2023).

Although the usage of annual rings is an effective technique for monitoring the variation of heavy metal concentration in the air in the recent past, this method is not seen as a sustainable method. The used method is considered a technique that can be used effectively in determining the recent variation in the heavy metal levels in the environment and could not harm the tree's vitality. Studies have been performed to monitor the variation of heavy metal pollution on *Picea* and *Abies* genera's needles in the recent past using this method (Alkharam, 2019; Cobanoglu, 2019; Keçeci, 2019).

In the current study, the change of elements based on years was significant. The Zn and Cu concentrations are higher in the older branches than young ones. However, based on the year, it is tough to say that the variations in the concentrations of other elements are consistent. The changes in some elements based on a year do not positively correlate with age (Cobanoglu, 2019). There was not a linear relationship between organ age and heavy metal concentrations in his study on *Abies nordmanniana* subsp. *equi-trojani* (Alkharam, 2019). It is stated that some metal

concentrations are at higher levels in the medium ages, and the lowest and highest concentrations can be determined in two consecutive years (Alkharam, 2019).

This situation is because heavy metal concentrations in plants are shaped by several independent parameters that affect each other (Mossi, 2018; Alkharam, 2019). Heavy metal accumulation potential in plant organs is closely associated with plant anatomy and, therefore, the plant species (Sevik, 2020). Therefore, it is essential to determine which heavy metal concentration is more intense in which organ of which plant. For example, the species that can be used to watch heavy metal contamination in the recent past include *Picea*, *Abies*, *Pinus*, and other genera. However, a study conducted on some species of these genera found that the most suitable species was *Picea pungens* (Turkyilmaz et al., 2018c).

Environmental conditions can also directly link to the heavy metal ratios in the air. Plant heavy metal accumulation potential emerges from the interaction of environmental situations and genetic codes (Hrivnák et al., 2017; Varol et al., 2022). Therefore, it is typical for each genetic structure to react adversely to similar environmental conditions (Yigit et al., 2019). For example, studies show that different varieties of similar species have diverse water or drought stress (Koç, 2021b; 2022) and cold stress resistance (Yildiz et al., 2014). Therefore, different individuals of same species have diverse genetic structures, so their morphological and phenotypic traits differ (Cetin et al., 2017).

CONCLUSION

This study revealed that some element concentrations considerably differed by organ and organ age. This result indicates that the needles and branches of the *P. pungens* are suitable for tracking heavy metal concentrations. However, due to the rough bark structure of the branches, particulate matter infected with metals in the ambience can easily stick to the bark. In this case, the results obtained in the branches can be misleading. It could be said that the most suitable biomonitors among the organs are the needles. It is known that different origins of the same species have different morphological, phenological, and anatomical features. Therefore, there could be differences in heavy metal accumulation between different origins of the same species. This issue should be considered in future studies, and studies should be continued on the metal accumulation in the same plant species from different origins.

Heavy metal pollution is a critical issue for human health and tree species. Therefore, it is crucial to determine and use the best appropriate method for observing heavy metal contamination in the environment. Recent studies have emphasized that biomonitors are the most appropriate technique for observing metal pollution in the air. However, some problems are encountered in the use of biomonitors. It is unknown how long the organs are exposed to heavy metal pollution in the air in living things such as moss and lichen or in perennial evergreen plants. The leaves of non-evergreen plants give only one-year data and cannot be interpreted because the data obtained cannot be compared. The supply of the material

needed for annual rings causes vital damage to the tree, and the results may not be reliable as heavy metals can be transferred to the wood. The method used in this study eliminates all the disadvantages listed and stands out as a reliable and sustainable monitoring method. As explained in the discussion section, the usage of biomonitors is an indirect method, and various factors affect the heavy metals accumulation in the plant. The number of reports on the entrance and stack of heavy metals into the tree body and the process within the plant is quite limited. Therefore, there needs to be more information about the active factors in this process. Therefore, the research should be focused on this subject and be continued by varying and increasing.

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