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ORIGINAL ARTICLE

Biomonitoring of Pb, Zn, Cu, Ni, and Cr Levels in Barks of Cypress (*Cupressus arizonica* Green.) Tree in the Atmosphere of Tehran City, Iran

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ABSTRACT
The concentrations of heavy metal accumulated by air pollution in the Cypress (Cupressus arizonica Green.) tree barks from the green area of Tehran City were measured by chemical analysis. The bark samples were collected from different pollution regions. The contents of heavy metals (Pb, Zn, Cu, Ni and Cr) were determined using a flame atomic absorption spectrophotometer. Result of this study showed that the lead, nickel, and copper content were found at high concentrations in the highway sites, whereas industrial areas contained high concentrations of zinc and chromium. The variation in metal concentrations between the studied locations is due to heavy traffic volume and industrialized activities. This study demonstrates the suitability of the Cypress tree bark as a suitable bio-indicator of environmental pollution in areas.
***Keywords**: Cypress bark; Biomonitoring; Heavy metals; Atmospheric pollution; Tehran city*

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INTRODUCTION

Trace metals are currently of much environmental concern. They are harmful to human and animals and tend to bioaccumulation in food chain [1]. The increasing industrialization and human activities intensify the emission of various pollutants into the environment and introduce various harmful substances into the atmosphere [2]. Air pollution is aesthetically offensive and can be genuine health hazard to human as well as to plants. The sampling use of plant tissues has long been shown to be an effective indicator of atmospheric pollution[3]. Vegetation is an effective indicator of the impact of a pollution source in its vicinity, because most plants have the ability to accumulate heavy metal so that their metal levels are much higher than those in the air. Further, the effect observed is a time averaged result, which will be more reliable than that obtained from direct determination of the pollutants concentration in air for a short period [4]. Recently, monitoring studies have been carried out to evaluate the degree of inorganic and organic contamination using plant materials [5].

There are many ways to monitor air pollution: (1) measure concentrations of air or rainwater and soil which are expensive and the contamination risk is greater than in analyzing bio indicators[6] (2) The use of bio-indicators, which are easy to collect, cheaper, and have higher concentrations than air and rainwater [7]. Numerous different bio-indicators are used in monitoring air pollution, such as mosses, lichens, vascular plants, woody plants, etc. Both the broad leaved and coniferous tree barks are used in studies of air pollution [8].

Tree barks did not collect sensitively heavy metals and other pollutants unlike mosses because they take their nutrient from rainwater. However, tree bark can be used as an indicator for other various pollutants, such as electrical conductivity, pH, sulphur, nitrogen, and heavy metals. Thus they are suitable indicator in urban and industrial areas, where other bio-indicators are very little [9]. Many different factors have an effect on the collecting of heavy metals in bark surface, such as heavy metal quantities in air, physiological and chemical properties of the bark, soil factors, contamination of other plants, climatic factors, etc[10] .

The aim of this study is to determine the pollution concentrations of Pb, Zn, Cu, Ni, and Cr using cypress tree (*Cupressus arizonica* Green.) barks as a biomonitor for such pollutants in Tehran. The obtained results could serve as a baseline data for future environmental impact assessment.

MATERIALS AND METHODS

Study area

Tehran, the capital city of Iran, which has an elevation of around 1400 m above sea level, at latitude (35°50′N) and longitude (51°37′E). The average annual precipitation in the investigated area is 254 mm/y. Minimum temperature is 5.1 °C in January and maximum temperature is 33.5 °C in August. Relative humidity during daytime is relatively low, ranging from 25% in June to 62% in December. The city suffers from high traffic density caused by vehicles. Therefore, air pollution in Tehran is mainly from vehicular emissions and fossil fuel combustion for heating purposes. The average number of vehicle movements per hour in urban, industrial, highway and control sites of the study area are 420, 310, 1560, and <50, respectively.

Sample collection and analysis

The bark samples from cypress tree (*Cupressus arizonica* Green.) were collected from the sampling point in September 2012. The total number of collected samples was 36, distributed as follow: 10 samples were collected from urban, industrial and highway sites, and 6 samples were collected from control sites. Control samples were taken from an area 20 km away from Tehran and any known source of contamination. Based on documents from the Tehran municipality, cypress trees are widespread in comparison with the other tree species. Cypress trees of about the same age (10-15 years) situated along the sides of main ways were selected, and around 100 g of the outer 5 mm of the bark at 2 m above the ground level were removed using stainless steel knife. The bark used in the study has a rough and hard surface.

In the laboratory the bark samples were carefully washed three times with distilled water to remove adhering particles [11] and dried in an oven at 105 °C for 48h. The dried samples were ground, then homogenized by sieving them through a 2-mm plastic sieve to remove large particles.

Analysis of heavy metals was carried out by digesting 1 g from each pre-washed and dried bark sample with 10 ml of 50% HNO_3 solution, then leaving the mixture overnight. The digested samples were ultrasonicated for 1 h and heated in a test tube heater for another 1 h at 90 °C [12]. The final extracts were filtered into 25-ml polyethylene volumetric flasks through 45-μm filters, and then diluted to the mark with 1% (v/v) HNO_3 solution. Heavy metals concentrations were measured by flame atomic absorption spectrophotometer, Perkin-Elmer AAS Analysis 300 model, with three replicates. Used metal standards were from Merck, Germany.

Electrical conductivity and pH values of bark samples were determined according to an earlier developed method [13]: 1.5 g of bark was mixed with 15-ml deionized water and left for 24 h. Electrical conductivity and pH of the extracted bark samples were determined using a WTW conductivity meter (LF 320) and WTW 525 pH-meter, respectively.

Analysis of variance (ANOVA) was used to compare the significant difference in the mean concentration of heavy metals between the sampling sites. F is a parameter in the level of 5%. Pearson's correlation coefficient was used to measure the degree of correlation between logarithms of the metal concentrations. The ANOVA test and Pearson's correlation coefficient were performed using the SPSS statistical program. Also, three replications were considered for all tests.

RESULTS AND DISCUSSION

Heavy metal concentrations in tree bark samples from the sampling sites are presented in Table 1. The pH values show relatively equal distribution in the area. The lowest values were found in the samples collected from the control site (5.39) and the highest value (6.21) was found in samples from the highway site. High electrical conductivity (EC) values (1575 μS cm^{-1}) were found in industrial sites, while the lowest value of EC was found in the control site (993 μS cm^{-1}). The results indicate that the highest and the lowest metal concentrations were found in the heavy traffic sites and the control site, respectively. The mean metal concentration values are arranged in the following order: Pb > Zn > Ni > Cu > Cr., Where higher Zn, and Cr characterize the industrial sites, whereas areas of high traffic highway are dominated by elevated concentration of Pb, Ni, and Cu.

Table 1: pH, EC, and metals content values for 36 cypress tree bark samples collected from different sites in Tehran city (mean value ± S.D.)

Parameters	Urban	Highway	Industrial	control
pH	5.52±0.3	6.21±0.5	5.67±0.4	5.39±0.3
EC (µS cm^{-1})	1200±39.3	1365±43.9	1575±49.8	993±26.1
Pb (ppm)	49.57±2.1	72.68±3.2	53.25±2.4	16.45±0.7
Zn (ppm)	16.86±0.7	21.56±0.8	28.18±1.1	1.78±0.2
Cu (ppm)	9.25±0.4	18.03±1.1	12.36±0.6	2.91±0.09
Ni (ppm)	11.58±0.3	19.48±0.9	15.7±0.8	2.16±0.07
Cr (ppm)	2.05±0.2	3.65±0.3	4.62±0.5	0.45±0.01

There are many high significant correlation coefficients between heavy metals in all sampling sites, such as Pb vs. Cu, Ni, and Zn (r = 0.84, 0.80 and 0.78, respectively), Zn vs. Ni, and Cu (r = 0.78, and 0.77, respectively), and also Cu vs. Ni (r = 0.76) (Table 2). This indicates that these sites were influenced by a different source of pollution, most probably vehicular emission and motor vehicle tires wheel for Pb, and Zn, whereas Ni, and Cr are major components of the industry dumping areas and automobile parts corrosion. A meaningful correlation was not found between Cr vs. Pb, Cu, Zn, and Ni because of low Cr concentration.

Table 2: Correlation coefficients between metals in cypress tree bark

	Pb	Zn	Cu	Ni
Pb				
Zn	0.78			
Cu	0.84	0.77		
Ni	0.80	0.78	0.76	
Cr	0.27	0.12	0.18	0.10

Analysis of variance (ANOVA) of heavy metal concentrations between the sampling sites is shown in Table 3. The result indicated that there are significant difference in Pb, Cu and Ni concentrations in cypress tree samples collected from different sites, whereas no significant differences were found for the rest of elements. This can be attributed to different anthropogenic activities between the sites.

Table 3.The result of statistical analysis (ANOVA)

Parameter	Sum of squares between groups	df	Mean square between groups	Sum of squares within groups	df	Mean square within groups	F	Observed α
pH	1.88	3	0.63	7.01	32	0.22	2.96	0.23
EC	593578.1	3	197859.36	4356686.8	32	136146.5	1.49	0.47
Pb	17203.06	3	5734.34	24547.78	32	767.12	7.62	0.015*
Zn	840.73	3	280.25	3196.74	32	99.88	2.85	0.73
Cu	463.18	3	154.38	756.12	32	23.62	6.66	0.033*
Ni	329.03	3	109.67	736.08	32	23.01	4.85	0.024*
Cr	111.42	3	37.13	471.13	32	14.72	2.57	0.67

α, significance level.

*Significant difference between the samples (P-value<0.05).

All the urban, industrial and highway sites are polluted in heavy metals compared with the control site as it can be deduced from Table 1. Even though the concentrations at urban site are slightly lower, it is still higher than control site values. It is clear that these is a slight difference in concentration between highway and industrial sites, which could be due to industrial activities it showed higher concentration with respect to Zn, and Cr. On the other hand, highway site is relatively enriched in Pb, Ni, and Cu, which confirms the automobile emission source. The industry and urbanization as a major source of heavy metal pollution are known by many [14].

The Pb levels were the lowest at the control site (16.45 ppm) and the highest at highway sites (72.68 ppm), which have higher traffic density. The chemical form of lead is of critical importance, since this is a factor in movements into plant, translocation and the toxic effectiveness of lead within the plant. Lead pollution on a local scale is caused by emissions from motor vehicle using leaded gasoline [15]. Normal content of Pb in plants is less than 10 ppm [16]. Allen (1989) considered a much lower value of 3 ppm as a normal natural level for plants. The close relationship between lead concentrations and traffic intensity has been demonstrated in detail by many authors [17]. In this research, there was a linear correlation between high Pb level and heavy traffic at Tehran city.

The degree of metals content in the pine needles was found to be proportional to industrial, human activities and urbanization. High metal concentrations in plants are contented in industrial sites and urban and highway roadsides due to the anthropogenic activities in addition to the traffic density [18].

Zinc is an essential element in all organisms and considered an important factor in the biosynthesis of enzymes, auxins and some proteins. Plants with symptoms of Zn deficiency experience a retarded elongation of cells. A critical toxic level of Zn in the leaves is about 100 ppm in dry plant matter [19]. The high contents of zinc in leaves and plant roots may cause the loss of food production and the low levels in plants may cause deformation of leaves (Bucher and Schenk, 2000; Celik *et al.,* 2005). The level of zinc in plant samples decreases with decreased traffic density (Table 1).

Copper is minor trace metal, with 70% Copper in leaves contained in the chloroplast of land plants. It is an important constituent of many enzymes of oxidation-reduction reactions . reported the normal content of Cu in plants ranges to be 2-20 ppm, but in most plants, the normal Cu contents are in a lower range of 4-12 ppm. Results indicated that the lowest mean value of copper (2.91 ppm) was found in samples collected from the control site, but the highest mean copper value (18.03 ppm) was found in sample collected from a highway that has heavy traffic[16].

The highest mean value of nickel was found in samples collected from the highway sites (19.48 ppm), whereas the lowest mean value was determined in a control site (2.16 ppm). This high concentration is attributed to emissions from motor-vehicle that use nickel gasoline and by abrasion and corrosion of nickel from vehicle parts [20].

The level of chromium in the study area was generally low (Table 1). Cr is a toxic, non-essential element for plants[21]. Effects of Cr on plants are symptoms of chlorosis on leaves and decrease of root growth. In the study area, chromium pollution caused by engine and body erosion of automobiles and extensive road marking by yellow lead chromate paint and some industrial activities [21]

CONCLUSION

The study demonstrates the suitability of the cypress (*Cupressus arizonica* Green.) tree bark as a suitable indicator of atmospheric pollution. The results of this work show that the highest and lowest metal concentrations were found in the heavy traffic sites and control site, respectively. The mean concentrations (C) of the studied metals were ordered as follows: $C_{Pb}>C_{Zn}>C_{Ni}>C_{Cu}>C_{Cr}$. The mean values of metal contents are lower at the control site compared to all other sites. The variation in heavy metal concentrations between the studied sites is due to heavy traffic volume and industrial activities. However, traffic and industrial emission were found to be the main source of metal pollution in the atmosphere of Tehran. No significant variations were found in pH values between the sites, which were attributed to a buffering effect of carbonate in the atmosphere. Electrical conductivity was high in all sites. The trend of increasing industrial and traffic activities in the city indicates the need for pollution control in the city environment. The present results showed that *Cupressus arizonica* Green. barks can be used as a simple way to monitor polluted sites.

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