Sulphur and Some Heavy Metal Contents in Foliage of Corylus avellana and Some Roadside Native Plants in Ordu Province, Turkey

Article *in* Ekoloji · April 2009

DOI: 10.5053/ekoloji.2009.702

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Ekoloji 18, 70, 10-16 (2009)

Sulphur and Some Heavy Metal Contents in Foliage of *Corylus avellana* and Some Roadside Native Plants in Ordu Province, Turkey

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Abstract
The concentration of sulphur and some heavy metals the leaves of *Corylus avellana*, *Alopecurus myosuroides*, *Helleborus orientalis*, *Glechoma hederacea*, *Calamintha nepeta* and *Urtica dioica* were determined around the "polluted (near highways)" and "non-polluted area (far from highways)" sites near Ordu province during summer of the year 2006. Significant differences were found for heavy metals between "polluted" and "non-polluted" areas. Heavy metal contents decreased in the order of Fe>Zn>Cu. The contents of Fe ranged from 188.9 to 519.9 ppm in the foliage of the studied species collected from "polluted area" were higher than its threshold values 13.5 to 115.5 ppm determined for same species in the "non-polluted area". High sulphur contents were found in studied species. The highest sulphur content was found in *Alopecurus myosuroides*. Overall, according to our findings plant species as *Corylus avellana*, *Glechoma hederacea* and *Urtica dioica* are a good bioindicators and can be used in elemental air pollution monitoring studies in urban-industrial conurbations.
Keywords: Automobile emissions, *Corylus avellana*, heavy metal, Ordu, sulphur concentration.

Ordu İlindeki (Türkiye) *Corylus avellana* (Fındık) ve Bazı Yol Kenarlarındaki Doğal Bitkilerin Yapraklarındaki Sülfür ve Ağır Metal İçerikleri

Özet
Ordu ili yakınlarında "kirlenmiş (karayollarına yakın)" ve "kirlenmemiş (karayollarından ve yerleşim alanlarından uzak" alanlarda *Corylus avellana*, *Alopecurus myosuroides*, *Helleborus orientalis*, *Glechoma hederacea*, *Calamintha nepeta* ve *Urtica dioica*'nın yapraklarında 2006 yılının yazında sülfür ve bazı ağır metal konsantrasyonları belirlenmiştir. "Kirlenmiş" ve " kirlenmemiş" alanlar arasında ağır metal içerikleri yönünden istatistiksel olarak önemli farklılıklar tespit edilmiştir. Ağır metal içeriği Fe> Zn>Cu sırası ile azalmaktadır. Fe içeriği "kirlenmiş" alandan toplanmış türlerin yapraklarında "kirlenmemiş" alandaki konsantrasyon değeri olan 13.5-115.5 ppm den fazla bulunmuş olup 188.9 ile 519.9 ppm arasında değişmektedir. Çalışılan türlerde yüsek sülfür içerikleri bulunmuştur. En yüksek sülfür içeriği *Alopecurus myosuroides*'de tespit edilmiştir. Sonuç olarak, bulgularımıza göre *Corylus avellana*, *Glechoma hederacea* ve *Urtica dioica* iyi biyoindikatörler olup endüstrileşmiş şehirlerde hava kirliliğini gözlemede kullanılabilirler.
Anahtar Kelimeler: Ağır metal, egzoz emisyonu, *Corylus avellana*, Ordu, sülfür içeriği.

Hüseyinova R, Kutbay HG, Bilgin A, Kılıç D, Horuz A, Kırmanoğlu C (2009) Sulphur and Some Heavy Metal Contents in Foliage of *Corylus avellana* and Some Roadside Native Plants in Ordu Province, Turkey. Ekoloji 18, 70, 10-16.

INTRODUCTION

The main agent of pollutant discharges to the environment is anthropogenic activity. Increasing industrialisation and human activities intensify the emission of various pollutants into the environment and introduce various harmful substances into the atmosphere. Atmospheric pollution has harmful effects on humanity and plant growth (Dursun et al. 1998).

The emission of toxic substances in the environment has been spread from industrialised countries. Many industrial plants and also heavy traffic may produce heavy metal and gaseous compounds into the atmosphere. Levels of environmental pollution in some areas from Turkey are alarmingly high and it is a national concern (Karademir and Toker 1998, Onder and Dursun 2006).

Geliş: 14.11.2008 / Kabul: 28.01.2009

Industrialization and urbanization result in the release of gaseous and particulate pollutants, such as sulphur and heavy metals (Kuang et al. 2007) . Point source pollution resulting from pulp and paper mills, mining, stainless steel works and from other kinds of industrial activities, e.g. municipal energy production, chemical works and petrol storage, occur all over the world, often in some very remote regions. These point sources emit a range of gaseous and particle pollutants into the air depending on the process and the type of activities at point source. Air pollution by gaseous compounds e.g. sulphur dioxide (SO_2) and total reduced sulphur compounds (TRS compounds) such as hydrogen sulphyde ($H2_2$), methyl mercoptane (CH_3SH) and methylsulphides [$(CH_3)_2S$ and $(CH_3)_2S_2$] is released into the ambient air from pulp and paper mills (Kuang et al. 2007).

Sulphur, as an essential nutrient is required for plant growth and development. This element, involved protein synthesis and is part of amino acids cystine and thiamine. Sulphur is associated with disease resistance. At the low concentration, plants can utilize atmospheric SO_2 as sulphur nutrient. But when its concentration reaches about a certain level, it becomes toxic to plants and reports to produce various physiological and biochemical changes in plants (Iqbal and Mahmood 1992). High sulphur concentration causes premature aging and dying off of leaves. On the other hand, distruction of the chlorophyll and cells by the effect of SO_2 caused the reduction in the thicknesses of the annual rings of the trees (Kantarci 2003).

The use of plants as a complementary tool to traditional (instrumental) methods of studing atmospheric pollution from anthropogenic and natural sources became an established technique in the past 30-40 years because of the development of powerful analytical techniques. Analysis of anthropogenic sources of atmospheric air pollution in a biomonitoring study requires knowledge of the threshold levels of major and minor elements in the substrate- biomonitor. This is especially important for elements which are characterized by high enrichments (Berlizov et al. 2007). A comprehensive review on the use of plants for air-monitoring purposes was given by Nimis (1990) and Mulgrew and Williams (2000).

More sensitive species are missing from regions with higher levels of substances to which they are sensitive. In many areas of the world, anthropogenic sources of pollutants have resulted in drastic reductions in lichen and bryophyte floras, particularly of sensitive, ecologically important, nitrogen-fixing lichen species (Henriksson and Da Silva 1978, Henriksson and Pearson 1981, Hallingbaeck 1986, Sigal and Johnston 1986, Wetmore 1989). For this reason, the search for alternative biological indicators becomes especially important.

The use of higher plants, especially different parts of trees, for air monitoring purposes is becoming more and more widespread. Uptake of elements into plants can happen via different ways. Plants can absorb trace elements through their roots or, to a lesser extent, their leaves. In addition, some plants even exhibit ion-exchange properties. Once deposited on the leaf surface some elements may also be taken up into the leaf via the stomata (Reimann et al. 2001).

Botanical materials such as fungi, lichens, tree bark, tree rings and leaves of higher plants have been used to detect pollution. The number of air pollution biomonitoring studies have been performed using bark and leaves of different tree species. Deposited materials on the leaves and needles, which is an important part of trees for photosynthesis, had effects on chlorophyll, cell membrane and stomas, and reduced the plants's development. Because of dry and wet deposition, the growth of main and side buds stops, leaving colour to fade and some parts of trees to get dry. These types of changes reduce the resistance of trees to drought, frost, insects and fungi (Shanker et al. 2005).

In this study hazelnuts (*Corylus avellana* L.) and some herbaceous plant leaves for chemical element air pollution monitoring around paper mill in Ordu Province (Turkey) was investigated.

The main aim of this study was to investigate the levels of sulphur and some heavy metals (Fe, Zn and Cu) in the hazelnuts tree and some herbaceous species leaves grown near the main traffic roads in Ordu province.

MATERIAL AND METHOD

The plant species were collected during summer of the year 2006. The concentration of sulphur and some heavy metals in the leaves of *Corylus avellana* L. (Corylaceae), *Alopecurus myosuroides* Hudson (Poaceae), *Helleborus orientalis* Lam.

(Ranunculaceae), *Glechoma hederacea* L. (Lamiaceae), *Calamintha nepeta* (L.) Savi (Lamiaceae) and *Urtica dioica* L. (Urticaceae) were determined around the "polluted" and "non-polluted area" sites near Ordu province. Two areas were taken into account in this study. The first one, named "non-polluted" area because it is far from the main traffic roads with relatively low anthropogenic impact and hardly any industry and 500 m a.s.l. (mean altitude from the sea level). The second one, named "polluted area", is located near the main traffic roads and there were some local timber factories 5 m a.s.l. Plant samples which collected from the non-polluted area are named " control group" whereas samples from polluted area are named "exposured to pollution".

At least five different plant specimens were used for each species in both "polluted" and "non-polluted" areas. Same species were selected from "polluted" and "non-polluted" areas. Plant species were chosen due to widespread distribution and co-occurred with *Corylus avellana*.

Leaf samples of *C. avellana* from throughout the midcrown per individual were taken with a tree-pruner and consisted of the leaves with no evidence of insect attack. Individuals were selected ≥2.5 m. from the stems of neighboring canopy trees to avoid potential microsite variation (Boerner and Koslowsky, 1989). About 1 g leaf samples of herb species co-occurred with *Corylus avellana* were taken.

In the laboratory, leaf samples were dried to a constant weight at 60ºC with a microwave oven. 0.5 g of the dried leaf samples were then powdered and they were extracted with a mixture of concentrated HNO_3 and $HClO_4$ (3:1). This digest was filtered through a Whatman filter paper No.42. Sulphur content of the leaves were determined by a Jenway U.V. spectrophotometer, using turbidimetric calcium-sulphate method (Bayrakli 1987). Concentrations of heavy metals (ppm gr^{-1} dry wt) were determined by a Perkin Emler 2280 atomic absorption spectrophotometer, using the air/acetylene flame (Allen et al. 1986). The following heavy metals were analysed: Zn, Cu and Fe. In AAS analyses, calibration graphs were obtained by a linear calibration model using eight working elemental standard solutions (WAKO Pure Chemical Industries Ltd., Japan). The obtained calibration graphs were all of excellent quality, always presenting the correlation coefficient $r^2 > 0.999$.

One-way analysis of variance (ANOVA) test were carried out by using SPSS 10.0 version. Tukey's honestly significant difference (HSD) test was used to rank means following the analysis of variance by using SPSS 10.0 version (Anonymous 1999).

RESULTS

Mean ratios of the elemental concentrations in the leaves of studied in both "polluted" and " non-polluted" areas are shown in Table 1. As seen in Table 1, sulphur concentrations was highest in all the plant species collected from the " polluted area", which is located near the main traffic highway. Heavy metal concentrations were usually ranked in the order of Fe> Zn>Cu in the studied species. Nevertheless, there was a minor change in the order of Fe and Zn in *Corylus avellana* and *Alopecurus myosuroides*. Statistically significant differences were found between "polluted" and "non-polluted" areas with respect to sulphur and heavy metals (Table 2).

The present results and threshold levels for the studied species which determined in the "non-polluted" area indicated that all element concentrations were higher in plant species collected from the "polluted area" (Figs. 1-4). Elevated concentrations of heavy metals in plant tissues generally indicate contamination associated with these elements (Guderian 1977). Significant differences were found for heavy metals between the study areas. The concentrations of Fe ranged from 188.9 to 519.9 ppm in the foliage of the studied species collected from "polluted area" were higher than its threshold values 13.5 to 115.5 ppm determined for same species in the "non-polluted area". The highest Fe concentration was observed in *Urtica dioica* as 519.9 ppm.

DISCUSSION

High iron contents were found in studied species which collected from "polluted" area. Enhanced iron, can indicate crustal sources, but the element is also enriched in anthropogenic sources. Especially, roadside environments may be enhanced in iron, because of the abrasion of the metals from automotive engines that end up in exhaust (Rhoades 1995).

Our results demonstrated that the Zn concentrations were 106.8-703.6 ppm in the studied species in the "polluted-area" which exceeded the level 15.6-81.9 ppm from unpolluted natural stands. Zinc is an essential element in all organisms and plays an important role in the biosynthesis of enzymes, auxins and some proteins (Onder and

Table 1. Mean values of element concentrations (ppm) in the studied species in the studied areas.

Elements	Species	Polluted area	Non-polluted area
S	*Corylus avellana*	2631.6 ± 352.9	1111.2 ± 236.4
	Alopecurus myosuroides	3387. ± 1195.0	657.1 ± 238.1
	Helleborus orientalis	1794.3 ± 238.0	71.1 ± 27.6
	Glechoma hederacea	2153.8 ± 241.9	1006.4 ± 83.9
	Calamintha nepeta	1454.4 ± 209.8	204.1 ± 72.5
	Urtica dioica	1416.6 ±199.8	615.8 ± 93.3
Fe	*Corylus avellana*	326.1 ± 67.6	110.2 ± 34.9
	Alopecurus myosuroides	229.9 ± 83.1	50.4 ± 12.6
	Helleborus orientalis	188.9 ± 43.5	13.5 ± 2.7
	Glechoma hederacea	235.8 ± 63.2	17.6 ± 2.5
	Calamintha nepeta	221.0 ± 50.4	30.9 ± 15.8
	Urtica dioica	519.9 ± 49.6	115.5 ± 6.5
Zn	*Corylus avellana*	703.6 ± 150.6	30.3 ± 7.4
	Alopecurus myosuroides	498.7 ± 85.4	20.8 ± 7.6
	Helleborus orientalis	190.2 ± 20.6	37.3 ± 15.0
	Glechoma hederacea	143.7 ± 20.6	15.8 ± 9.9
	Calamintha nepeta	106.8 ± 7.3	15.6 ± 1.7
	Urtica dioica	423.2 ± 40.3	81.9 ± 10.3
Cu	*Corylus avellana*	10.6 ± 2.9	9.1 ± 1.7
	Alopecurus myosuroides	6.1 ± 0.6	1.8 ± 0.1
	Helleborus orientalis	6.3 ± 1.5	3.4 ± 0.4
	Glechoma hedereacea	33.8 ± 2.6	6.9 ± 1.3
	Calamintha nepeta	2.9 ± 0.3	3.6 ± 0.2
	Urtica dioica	12.9 ± 1.5	6.9 ± 2.5

Table 2. Comparison of studied species for element concentrations between "polluted" and "non-polluted" areas by one-way ANOVA test.

Elements	F-Value
Fe	50.763**
Zn	40.494**
Cu	10.179*
S	35.919**

*$P< 0.05$; ** $P< 0.01$

Dursun 2006). A critical toxic level of Zn in the leaves is about 100 ppm in dry plant matter (Allen et al. 1974, Yılmaz and Zengin 2004). High level of Zn in plants may cause a loss of production (Bucher and Schenk 2000). Zinc levels can be enhanced in automobile exhaust, also may be elevated near roadways due to tire wear. According to the results Zn, is at harmful levels as a major threat to the environment in our study. The highest Zn concentrations were found in the leaves of *Corylus*

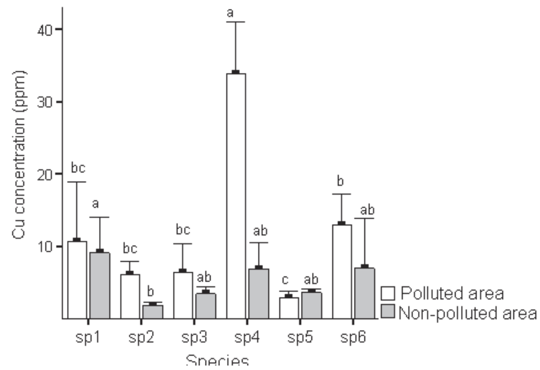

Fig 1: Cu concentrations (ppm) in studied species in "polluted" and "non-polluted" areas. (sp1. *Corylus avellana*;sp2. *Alopecurus myosuroides* sp3 *Helleborus orientalis* sp4. *Glechoma hederacea* sp5.*Calamintha nepeta* sp6. *Urtica dioica*). Means followed by the same letter are not significantly different at the 0.05 level using Tukey's HSD test.

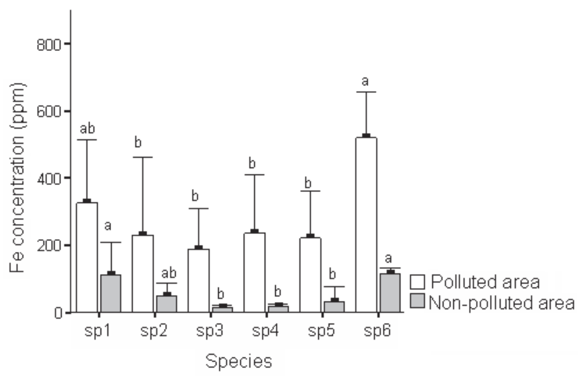

Fig 2: Fe concentrations (ppm) in studied species in "polluted" and "non-polluted" areas. Means followed by the same letter are not significantly different at the 0.05 level using Tukey's HSD test.

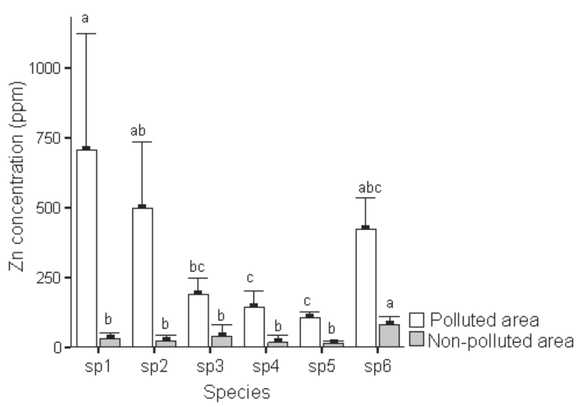

Fig 3: Zn concentrations (ppm) in studied species in "polluted" and "non-polluted" areas. Means followed by the same letter are not significantly different at the 0.05 level using Tukey's HSD test.

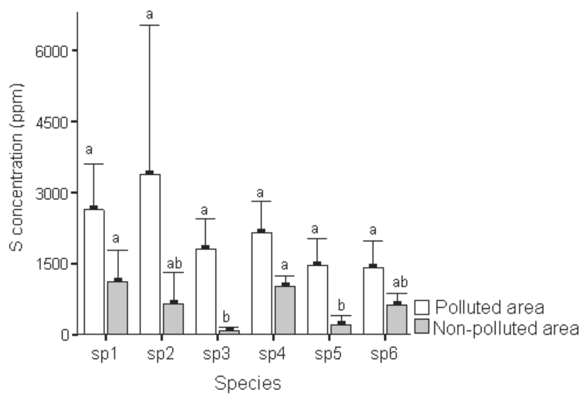

Fig 4: S concentrations (ppm) in studied species in "polluted" and "non-polluted" areas. Means followed by the same letter are not significantly different at the 0.05 level using Tukey's HSD test.

avellana. The mean Zn concentration was recorded 703.6 ppm in the leaf tissues of hazelnuts. Results of this investigation show that this level may cause loss of hazelnuts production around the study area.

Copper is a minor trace metal, with 70% copper in leaves contained in the chloroplast of land plants (Wilkinson 1994) 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 narrower range of 4-12 ppm. An excessive supply of copper can cause symptoms of chlorosis that are similar to the symptoms of Fe deficiency (Bergman 1983). Disturbances in Cu supply can cause significant modifications of biochemical processes in plants, leading to lower yields and quality of agricultural crops. On the other hand, excessive Cu may destroy sub-cellular structure of plants (Sresty and Madhava 1999). Levels of copper in all studied species in "polluted area" were generally within the threshold range except for *Glechoma hederacea*. In the present study, the highest mean value of Cu was 33.8 ppm in the leaves of *G. hederacea*. This value was higher than its threshold level of 6.9 ppm in the same species collected from the "non-polluted area". These results show that the current Cu level exceeded the level 1.5-10.1 ppm from unpolluted natural stands for the studied species.

Sulphur as an essential nutrient is required for plant growth and development. But when its concentration reaches about a certain level, it becomes toxic to plants and reports to produce various physiological and biochemical changes in plants (Iqbal and Mahmood 1992). Statistical differences among the species for sulphur accumulation were not found. However, the concentration of total sulphur (S) significantly differed between the "polluted" and "non-polluted area". The main reason of high sulphur contents in the leaves of studied species are automobile emissions.

Phytoremediation, the use of plants to restore polluted sites, has recently become a tangible alternative to traditional methodologies (Glass 2000). It has been established that certain wild and crop plant species have the ability to accumulate elevated concentrations of toxic heavy metals (Blaylock and Huang 2000, Reeves and Baker 2000, Ernst et al. 2000). However, researchers, all over the world are researching new plant species susceptible to be used in phytoremediation (Onder and Dursun 2006). For example, field bindweed, *Convolvulus arvensis* L., a prostrate perennial plant, drought resistant, with a deeply penetrating taproot that grows mostly in dry soils (Hickey and King 1988) has been identified among the plants that grow in the multi-metal contaminated soil of Spain and Poland (Del Rio et al. 2002).

According to the results of the present study sulphur and Zn and Cu concentrations were far above the threshold levels, implying that these elements could be potentially harmful on local vegetation and environments in the future if the emission of pollutants is not controlled effectively. Baslar et al. (2003) were found high Cu contents as compared to present study in needles of *Pinus brutia* Ten. which occurred around highways. Similarly, Fe contents were also higher than that of studied species except for *U. dioica* which has high Fe contents. Baslar et al. (2005) were also found rather high Cu contents in leaves of *Populus nigra* L. which occurred in a urban roadside as compared to the present study. However, Zn contents of all speices in the present study were rather high than that of Zn contents of *P. nigra* leaves. Fe contents were higher in *U. dioica* than that of *P. nigra*. However, the other species had lower Fe contents as compared to *P. nigra*. Such differences may be originated the differences between studied life forms (i.e herb species were usually used in the present study except for *C. avellana*).

However, Dogan et al. (2007) have been reported rather low Cu contents in the bark of *P. brutia* in Western Anatolia, whilst Fe contents were usually higher than that of studied plants except for

U. dioica. According to these findings *U. dioica* may be treated as a very useful plant for biomonitoring purposes. It has been reported that plants with spiny and hairy leaves accumulated more heavy metals as compared to smooth-leaved plants (Kutbay and Kılınc 1991).

As C. avellana has an economic value for Turkey economy, high element concentrations may cause loss of hazelnuts production in the future in the study area. These results might be explained by the fact that sulphur emissions and heavy traffics, the use of low quality fuels have spread sulphur and some metals and accumulated directly in plant leaves.

Overall, the results reported here demonstrate that plant species as *C. avellana*, *G. hederacea* and *U. dioica* are a good bioindicators and can be used in elemental air pollution monitoring studies in urban-industrial conurbations.

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