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Leaves of Higher Plants as Indicators of Heavy Metal Pollution along the Urban Roadways

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ABSTRACT

Plants are hitherto regarded as sinks for pollutants and can accumulate higher concentration of heavy metals in urban environment. The concentrations of Pb, Cu, Cd and Zn were determined to assess the impact of automobiles on heavy metal concentrations in plants growing along the roadways. Inductively Coupled Plasma – Optical Emission Spectrometer (ICP – OES) was used to estimate heavy metals concentrations in plants. Concentrations of Pb, Cu, Cd and Zn were significantly higher in plants growing at polluted sites when compared to their control counterparts. The Pb levels of plants at polluted site varied from 46.2 - 84.9 μgg^{-1}, highest being noted in *Ageratum conyzoides*. The species *Ageratum conyzoides* showed higher levels of Cu and Zn at polluted site. The mean concentration of cadmium remained relatively low when compared to other metals, with highest of 10.5 μgg^{-1} in *Cynodon dactylon.* The increase in heavy metal content in plants across polluted sites showed positive correlation with traffic density. With higher levels of Pb, Cu, Cd and Zn *Ageratum conyzoides* can be considered as multi metal indicator.

Keywords: *heavy metals, Ageratum conyzoides, ICP – OES, bioindicator, vehicular traffic, higher plants.*

1. INTRODUCTION

Heavy metal pollution in urban environment is a global phenomenon caused due to variety of anthropogenic activities. A higher level of lead in roadside environment is often associated with the traffic density (Fergusson et al. 1980; Kartal et al. 1992; Arslan 2001). The levels of copper, cadmium and zinc are also reported to correlate with vehicular traffic (Fergusson et al. 1980; Narin 1999). Vehicular emissions along the busy roadways contain high levels of lead which are present in fuel as anti - knock agents (Suzuki et al. 2008; Atayese et al. 2009). Along with lead, other heavy metals such as cadmium, copper and zinc are associated with the vehicular activity; since they are included in petrol, engines, tyres, lubricant oils and galvanized parts of the vehicles (Ardakani 1984).

There are many reports on the interaction of heavy metals and plants on roadside (Othman et al. 1997; Berlizov 2007; Suzuki 2008; Malik 2010; Naveed 2010; Turan 2011; Ogbonna and Ogbonna 2011; Verma et al. 2013). Some plants accumulate heavy metals to concentrations non – toxic to them (Adriano 1986, Brooks 1987) and or may be toxic to other species (Merian et al. 1985). Zhang et al. (2012) reported that higher plants growing along the busy roadways act as sink and effectively decrease the heavy metal concentrations in the atmosphere. Higher plants are also used as bioindicators of heavy metal pollution (Berlizov et al. 2007).

Leaves are the site of major physiological processes and are highly affected by air pollutants (Naveed et al. 2010). Plant leaves respond to subtle changes in the environment and therefore constitutes an excellent material to assess pollution levels. These changes are extensively used for monitoring pollution levels (Turan et al. 2011). In the past few decades, leaves of higher plants have been used for biomonitoring heavy metals (Al - Shayeb et al. 1995; Aksoy and Ozturk 1996; Aksoy and Ozturk 1997; Tiwari 2008).

Today, Indian cities are considered as some of the most polluted in the world and the single most important factor responsible is the ever increasing number of automobiles. Very few studies have been carried out in India and reports on heavy metal pollution in plants are extremely scarce. Bangalore being one of the fastest growing cities in the world harbor phenomenal traffic density. The present work was aimed at assessing heavy metal content in plants alongside the high traffic urban environment in Bangalore.

2. MATERIALS AND METHODS

Study sites

The sites selected for the present study include a control site (Kommasandra - S I), a moderately polluted residential site (Ramamurtynagar - S II) and a highly polluted site (Bangalore city railway station - S III) with an approximate traffic density of < 350, 51 600 and 86 500 vehicles respectively.

Plant sample

The plant species selected for the present study were

Species	Family
Ageratum conyzoides L.	Asteraceae
Bambusa bambos L. Voss	Poaceae
Bougainvillea spectabilis Comm. Ex. Juss.	Nyctaginaceae
Cynodon dactylon L. Pers	Poaceae
Ficus religiosa L.	Moraceae
Mangifera indica L.	Anacardiaceae
Peltophorum pterocarpum DC. K. Heyne	Fabaceae
Portulaca oleraceae L.	Portulacaeae
Ricinus communis L.	Euphorbiaceae
Terminalia catappa L.	Combretaceae

The leaf samples of the 10 selected plants were collected in three different seasons for two years from all the study sites. Samples were washed with running tap water to remove the adhering dust particles. After blotting, the samples were air dried in shade, ground to fine powder and used for further analysis.

Sample Digestion

The sample preparation was performed according to the guidelines of US - EPA 3050b. About 0.3 g of plant material was weighed in a 250 mL Erlenmeyer flask and 5 mL of 1 : 1 HNO_3 was added. The solution was heated on a hot plate to 95 ºC, refluxed for 15 minutes without boiling. After cooling, 2.5 mL of concentrated HNO_3 was added and the sample was refluxed for 30 minutes at 95 ºC without boiling. This step was repeated. Thereafter, the sample was evaporated to 5 mL without boiling. After cooling, 2 mL of double distilled water was added to the sample followed by the slow addition of 3 mL of 30 % H_2O_2. The solution was then heated until effervescence subsided. Later, 6 mL of 30 % H_2O_2 in 1 mL aliquots was added and the solution was refluxed. After cooling, 2.5 mL of concentrated HCl was added and the sample was refluxed for 15 minutes without boiling. After cooling to room temperature, the sample was filtered using Whatman No. 1 filter paper and diluted to 50 mL with double distilled water.

All samples were analyzed in triplicates by Inductively Coupled Plasma – Optical Emission Spectrometer. The measurements were performed using the Perkin Elmer Optima ICP - OES instrument, ICP version 4.0 software for simultaneous measurement of all analyte wavelengths of interest.

Table 1: ICP – OES parameters used

ICP – OES parameters		Value
Wavelengths	Lead	220.353 nm
Wavelengths	Copper	327.393 nm
Wavelengths	Cadmium	226.502 nm
Wavelengths	Zinc	213.857 nm
Power		1450 W
Plasma Gas Flow		15 $Lmin^{-1}$
Auxiliary Gas Flow		0.2 $Lmin^{-1}$
Nebulizer Pressure		140.00 kPa
Replicate Time		5 s
Stab Time		10 s
Sample Uptake		22 s
Rinse Time		10 s
Pump Rate		25 rpm

Statistical analysis was performed using Pearson’s correlation co – efficient.

3. RESULTS

The mean concentration of the elements in the samples collected from the polluted site was significantly higher than their levels at control site (table 2).

Table 2: Mean concentrations of lead, copper, cadmium and zinc of selected plant species at different study sites

	Lead (μgg^{-1})			Copper (μgg^{-1})			Cadmium (μgg^{-1})			Zinc (μgg^{-1})		
Species	**S I**	**S II**	**S III**	**S I**	**S II**	**S III**	**S I**	**S II**	**S III**	**S I**	**S II**	**S III**
A. conyzoides	1.17±0.06	27.56±0.14	84.98±0.11	4.57±0.17	13.71±0.31	55.64±0.17	0.12±0.16	2.89±0.27	9.56±0.25	9.34±0.26	30.42±0.30	80.34±0.26
B. bambos	1.14±0.21	17.89±0.28	56.87±0.14	1.46±0.28	10.41±0.14	22.78±0.28	0.24±0.12	2.00±0.24	6.96±0.16	3.92±0.34	28.39±0.14	67.76±0.17
B. spectabilis	2.28±0.15	20.24±0.32	74.45±0.30	1.78±0.14	12.30±0.27	33.65±0.16	0.56±0.28	1.87±0.18	8.24±0.17	8.65±0.13	28.65±0.32	45.97±0.14
C. dactylon	1.13±0.18	18.56±0.27	46.76±0.26	3.12±0.16	17.76±0.26	41.36±0.14	0.32±0.32	2.97±0.14	10.56±0.28	5.89±0.35	25.67±0.26	60.24±0.28
F. religiosa	0.87±0.20	19.68±0.31	62.66±0.18	0.47±0.18	7.27±0.16	44.87±0.22	0.76±0.36	1.11±0.13	8.65±0.36	2.76±0.13	20.35±0.15	59.26±0.14
M. indica	1.17±0.27	15.67±0.16	76.34±0.13	1.37±0.38	9.18±0.17	36.89±0.15	0.65±0.18	1.32±0.25	5.89±0.16	8.35±0.27	28.35±0.18	50.43±0.25
P. pterocarpum	1.08±0.30	9.37±0.31	84.34±0.27	2.34±0.27	4.27±0.25	42.64±0.30	0.23±0.17	1.23±0.31	4.67±0.19	1.60±0.12	11.60±0.14	43.78±0.18
P. oleraceae	1.35±0.17	16.67±0.16	65.34±0.22	3.54±0.29	6.24±0.12	20.45±0.25	0.45±0.28	1.43±0.27	6.35±0.21	2.56±0.16	25.97±0.28	56.98±0.15
R. communis	1.24±0.29	12.78±0.05	46.23±0.12	1.84±0.15	7.76±0.26	17.87±0.13	0.39±0.14	1.56±0.28	5.35±0.30	6.42±0.19	28.71±0.24	67.67±0.19
T. catappa	1.16±0.13	15.71±0.11	49.98±0.14	1.44±0.09	11.31±0.30	49.76±0.25	0.29±0.19	1.97±0.18	9.12±0.29	5.89±0.24	22.67±0.31	70.54±0.32

n = 18

Lead

The concentration of lead in plants increased with increase in traffic density. All the species showed manifold increase in the lead concentration in relation to the control site. The lead concentration in plants at control site was significantly lower and ranged from 0.87 – 2.28 μgg^{-1} respectively in *Ficus religiosa* and *Bougainvillea spectabilis.* Amongst polluted sites *Ageratum conyzoides* and *Peltophorum pterocarpum* accumulated maximum levels of lead in highly polluted site with a concentration of 84.98 and 84.34 μgg^{-1} respectively. Followed by *Mangifera indica* (76.34 μgg^{-1}), *Bougainvillea spectabilis* (74.45 μgg^{-1}), *Portulaca oleraceae* (65.34 μgg^{-1}), *Ficus religiosa* (62.66 μgg^{-1}), *Bambusa bambos* (56.87 μgg^{-1}), *Terminalia catappa* (49.98 μgg^{-1}), *Cynodon dactylon* (46.76 μgg^{-1}) and *Ricinus communis* (46.23 μgg^{-1}). It is interesting to note that all these maximum levels were recorded at S III, a site with maximum traffic density (86 500 per day). This indicated significant lead accumulation in plants growing along the roadside. The increase in lead content at polluted sites showed significant positive correlation with traffic density ($r = 0.930$, $p \leq 0.001$).

Table 3: Correlation coefficients (r) between heavy metals in plants and traffic density

Species	Lead	Copper	Cadmium	Zinc
A. conyzoides	0.951	0.907	0.944	0.945
B. bambos	0.946	0.981	0.932	0.972
B. spectabilis	0.927	0.956	0.894	0.998
C. dactylon	0.972	0.965	0.931	0.967
F. religiosa	0.948	0.886	0.833	0.950
M. indica	0.904	0.915	0.874	0.991
P. pterocarpum	0.860	0.835	0.917	0.922
P. oleraceae	0.923	0.889	0.892	0.983
R. communis	0.930	0.968	0.922	0.962
T. catappa	0.946	0.908	0.902	0.936

Significance level: $p \leq 0.001$

Copper

The copper levels in plants collected from the busy roadway were higher when compared to their control counterparts. The mean copper concentration in controls ranged from 0.47 – 4.57 μgg^{-1}. The species *Ageratum conyzoides* accumulated more copper at S III followed by *Terminalia catappa*, while *Ricinus communis* and *Portulaca oleraceae* exhibited lower levels of copper at S III. Moderate levels were noted in *Bougainvillea spectabilis* and *Mangifera indica*. A significant positive correlation ($r = 0.921$, $p \leq 0.001$) was observed between increased copper content in plants and vehicular traffic.

Cadmium

The mean cadmium content in plants generally remained low. Plants growing in control site showed very low levels of cadmium ranging between 0.12 and 0.76 μgg^{-1}. At polluted site, the mean cadmium in plants was higher indicating cadmium enrichment in the roadside plants attributable to vehicular activity along the roadways. Of the ten species investigated, highest level of cadmium was seen in *Cynodon dactylon* (10.56 μgg^{-1}) followed by *Ageratum conyzoides* (9.56 μgg^{-1}), while *Peltophorum pterocarpum* (4.67 μgg^{-1}) and *Ricinus communis* (5.35 μgg^{-1}) showed lower levels of cadmium at S III. Moderate levels of cadmium were observed in *Portulaca oleraceae* and *Bambusa bambos*. The increase in cadmium content was positively correlated to traffic density ($r = 0.904$, $p \leq 0.001$).

Zinc

Zinc level in plants correlated with average daily traffic on the roads. At control site, plants showed 1.60 – 9.34 μgg^{-1} of zinc and subsequently its concentration increased at highly polluted site (43.78 - 80.34 μgg^{-1}). *Ageratum conyzoides* and *Terminalia catappa* at site III showed higher levels of zinc content with 80.34 μgg^{-1} and 70.54 μgg^{-1} respectively. This is followed by *Bambusa bambos* (67.76 μgg^{-1}), *Ricinus communis* (67.67 μgg^{-1}), *Cynodon dactylon* (60.24 μgg^{-1}), *Ficus religiosa* (59.26 μgg^{-1}), *Portulaca oleraceae* (56.98 μgg^{-1}), *Mangifera indica* (50.43 μgg^{-1}), *Bougainvillea spectabilis* (45.97 μgg^{-1}) and *Peltophorum pterocarpum* (43.78 μgg^{-1}). A significant positive correlation was noted between traffic density and zinc content ($r = 0.962$, $p \leq 0.001$) in plants.

4. DISCUSSION

Lead

Lead is a major contaminant posing significant environmental problems (Shen et al. 2002) and is an accumulative protoplasmic poison (Verma et al. 2013). There are reports of direct relationship between levels of lead in plants and traffic density (Atayese et al. 2009; Naveed et al. 2010; Turan 2011; Shafiq 2012; Verma et al. 2013). The higher mean concentration of lead in plants at site III is because of increased traffic density. Species such as *Ageratum conyzoides, Peltophorum pterocarpum* and *Mangifera indica* exhibited higher levels of lead suggesting their ability to accumulate lead in comparison to other plant species and are considered as tolerant species. Dada et al. (2012) reported heavy metal accumulation in *A. conyzoides*, especially higher levels of lead, while Sahar (2011) showed that *Mangifera indica* accumulate higher levels of lead. Shafiq et al. (2012) reported 7 – 81 μgg^{-1} of lead in *Peltophorum pterocarpum* which is in agreement with the present values. Lead accumulation in *Cynodon dactylon* in the present study concur with that reported by Othman et al. (1997) from roadside. Therefore it is evident that *Ageratum conyzoides* is an indicator of lead pollution along with *Peltophorum pterocarpum* and *Mangifera indica*.

Copper

The increase in copper content in plants grown in highly polluted site is attributed to increased traffic density. Increased copper levels in plant leaves subjected to vehicular pollution is well reported (Berlizov et al. 2007; Sahar 2011; Adamsab et al. 2012; Hassan 2013). The species *Ageratum conyzoides* and *Terminalia catappa* showed higher levels of copper. The copper levels in *Ageratum conyzoides* (55.64 μgg^{-1}) in S III was higher than the value reported by Mkumbo (2012). However in *Portulaca oleraceae* copper concentration was lower than that reported by Malik et al. (2010). Studies indicate that the normal limits of copper concentrations in plants were in the range of 4 - 15 μgg^{-1} (Bowen 1979) and between 20 - 100 μgg^{-1} was considered as toxic values (Pendias and Pendias 2001). Based on these values, the copper concentrations in plants growing at less polluted S I were within normal limits (0.47 - 4.57 μgg^{-1}) and have reached toxic levels at S III (17.87 - 55.64 μgg^{-1}). By virtue of high copper content, *Ageratum conyzoides* and *Terminalia catappa* were regarded as copper pollution indicators.

Cadmium

The increased cadmium content in plants along the busy roadways is attributed to increased traffic density (Sharma and Prasad 2010; Sahar 2011; Shafiq et al. 2012; Christiana and Samuel 2013; Verma et al. 2013). The range of cadmium between 4.67 – 10.56 μgg^{-1} at polluted site in the present study was higher than the values reported by Ogbonna and Ogbonna (2011) which was in the range of 0.02 – 1.44 μgg^{-1}, indicating the plants ability to accumulate more cadmium. Being a trace metal, cadmium is found in very low levels (0.2 – 0.8 μgg^{-1}) and toxic concentrations of cadmium are defined as 5 - 30 μgg^{-1} (Kloke et al. 1984; Pendias and Pendias 2001). At control site, the concentration was in the normal range, while at the polluted site the concentrations were nearing toxic levels. It is apparent that various species growing in same conditions exhibited differential heavy metal absorption. *Cynodon dactylon* and *Ageratum conyzoides* with higher levels of cadmium can be considered as indicators of cadmium pollution along the busy roadways.

Zinc

Zinc has been considered as an essential element as it is a component of various enzymes (Woolhouse 1983). The control plants revealed 1.60 – 9.34 μgg^{-1} of zinc which was lesser than the values obtained at S III (43.78 - 80.34 μgg^{-1}). The increase in levels of zinc in plants grown at S III is because of the increased traffic density (Sheng and Peart 2006; Ogbonna and Ogbonna 2011; Sahar 2011; Christiana and Samuel 2013; Hassan 2013). The zinc levels observed in *Peltophorum pterocarpum*, *Ficus religiosa* and *Mangifera indica* were significantly higher than those reported by Adamsab et al. (2012). Similarly, the level of zinc in *Ageratum conyzoides* was higher than the level observed by Mkumbo (2012). Plants can tolerate zinc levels of up to 387 – 1221 μgg^{-1} (Li et al. 2005), which is a lot higher than the levels seen in the present study. However, with an increasing gradient in relation to vehicular density, the plants tend to accumulate substantial quantity of zinc. The species *Ageratum conyzoides* and *Ricinus communis* with higher levels of zinc accumulation were considered as zinc pollution indicators along busy roadways.

5. CONCLUSION

Concentrations of lead, copper, cadmium and zinc in plants across the polluted sites remained significantly higher over the control samples. Their magnitude showed direct relationship with traffic density indicating automobiles as the main source of these heavy metals in plants. *Ageratum conyzoides* with higher levels of lead, copper, cadmium and zinc was considered as a multi metal indicator and can be used to monitor and ameliorate heavy metal pollution in the urban localities.

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