

SCREENING OF SOME ROAD SIDE TREES FOR PLANTATION IN POLLUTED URBAN AREAS OF KATHMANDU, NEPAL

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ABSTRACT

Kathmandu valley is facing air pollution problem because of rapid increase in vehicular emissions. Some common road side trees like *Callistemon lanceolatus*, *Cinnamomum camphora*, *Ficus religiosa*, *Grevillea robusta*, *Jacaranda mimosifolia* and *Populus euroamericana* were evaluated for their air pollution tolerance index (APTI) and metal accumulation index (MAI). These two indices were measured during winter season at different polluted sites (Airport, Balaju, Banasthali, Basundhara Dhumbrahi and Gongbu) and less polluted site (Narayanthan) in Kathmandu. Air pollution tolerance Index (APTI) ranged from 12.51 ± 0.74 to 15.67 ± 0.75 . Highest APTI value was observed in *Populus euroamericana* (15.67 ± 0.75) at Dhumbrahi. The mean value of metal accumulation index (MAI) values ranged from 7.34 to 20.44 and highest MAI value was observed in *Populuseuroa mericana* (20.44). Correlation of APTI and heavy metal (Cu, Pb and Zn) accumulation in most of the studied tree leaves were negative showing their inverse relationship, which indicated the heavy metal avoidance mechanism among them for their survival. *Populus euroamericana*, *Jacaranda mimosifolia* and *Callistemon lanceolatus* with high MAI and intermediate APTI range are recommended for plantation on road side of Kathmandu.

KEY WORDS : Metal Accumulation Index (MAI), Urban area, Avoidance mechanism, APTI

INTRODUCTION

Kathmandu valley, ranging from 1200 to 1400 m above sea level in subtropical zone, has bowl shaped topography, which makes it more prone to air pollution (Pradhanet *et al.*, 2012; Shrestha, 2001). Rapid urbanization, industrialization, poor maintenance of road, poorly maintained vehicle deteriorate the air quality in Kathmandu valley (MaYa, 2014) and particulate matter are being one of the major pollutants (Shrestha, 2001). Vehicle numbers in Kathmandu Valley has rapidly increased in the last 15 years. Data have shown that in the fiscal year 2000/2001, number of registered vehicles was 24,003 and by 2015/2016 it has increased to 7,79,822. This shows an increment by more than 32 times in the last one and a half decade (Saud and Paudel, 2018).

Biochemical parameters like leaf pH, relative water content, chlorophyll content, and ascorbic acid content collectively suggested as the best index

of the susceptibility levels of plants known as air pollution tolerance index (APTI) (Kuddus *et al.*, 2011; Rai *et al.*, 2013). The sensitivity or tolerance of a plant depends upon these parameters (Liu nad Ding, 2008; Singh and Verma, 2007). Trees with high air pollution tolerance index (APTI) values can tolerate as well as withstand to air pollution to a greater extent than those with less APTI value. Resistance of heavy metals in a plant is performed either by avoidance or tolerance mechanism. Avoiders group of plants can protect themselves by stopping metal ions through entering their cellular cytoplasm while tolerant plants can detoxify metal ions through crossed organelle biomembranes or internal the plasma membrane (Millaleo *et al.*, 2010). It is not clear if the plants with high APTI value also have high bio-accumulation ability or APTI are inversely related with metal accumulation. Hence to ascertain this, APTI and metal accumulation in six common roadside tree species growing in high vehicle traffic (polluted) and less vehicle traffic (less

polluted) areas were studied in Kathmandu. In the present study, based on the high APTI and MAI value the common road side tree species will be screened for plantation, as the screened plants will be able to accumulate more heavy metals and also will have high tolerance to withstand pollution.

MATERIALS AND METHODS

Leaf samples of six common road side trees like *Callistemon lanceolatus*, *Cinnamomum camphora*, *Ficus religiosa*, *Grevillea robusta*, *Jacaranda mimosifolia* and *Populus euroamericana* were collected from high traffic areas (Airport, Balaju, Banasthali, Basundhara, Gongobu Dhumbarahi) and less traffic area (Narayanthan, a control site) (Fig. 1) on the same day around 9.00 to 10:00 AM for (A) APTI and (B) metal analysis.

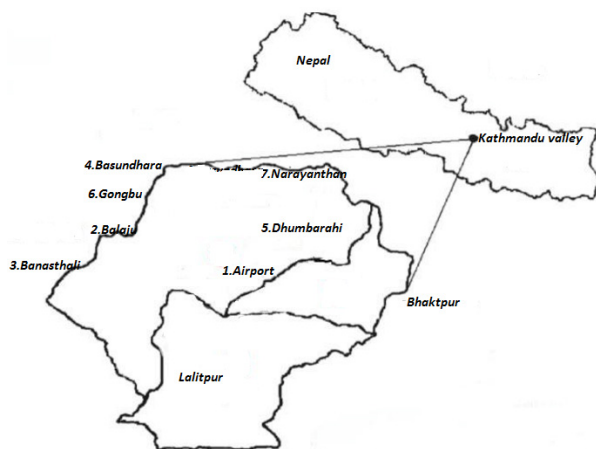


Fig. 1. Map of Study site: Kathmandu showing sampling sites -1. Airport 2. Balaju 3. Banasthali 4. Basundhara 5. Dhumbarahi 6. Gongbu and 7. Narayanthan

(A) APTI : All the collected leaves were cleaned thoroughly in dry condition using brush and cotton before the processing for APTI. APTI was obtained by using the parameters like ascorbic acid content, leaf extract pH, total chlorophyll content and relative water content according to Singh and Rao (1983).

$$\text{APTI} = [A (T + P) + R] / 10$$

(Where A is the ascorbic acid content (mg g^{-1}) of fresh weight; T is the total chlorophyll (mg g^{-1}) of fresh weight; P is the pH of leaf extract and R is the relative content of water (%)).

Ascorbic acid (AA) and total chlorophyll (TC) content of leaf samples was determined using spectrophotometer according to Bajaj and Kaur

(1981) and Barnes *et al.* (1992) respectively. Relative water content (RWC) and leaf extract pH was measured according to Barrs *et al.* (1962) and Apriyantono *et al.* (1989) respectively.

(B) Metal Analysis: For metal analysis, the leaves were washed with running tap water, rinsed with double deionized water and then dried at room temperature (20°C) about 24-48 hours. Again dried at 60°C for 48 hours. The dried leaf samples were grinded using mortar and pestle to prepare representative samples. 1 g DW of the representative sample was dipped in 8 ml concentrated HNO_3 (Merck). These were left overnight at room temperature. On the next day, the mixture was warmed for 2 h at 50°C and subsequently heated at 160°C for 4 h. The cooled extracts were filtered through Ashless filter paper (Whatman 589³) and were diluted to prepare 25 ml with double deionized water (Sawidis *et al.*, 1995c). The filtrates were then analyzed for Cu, Pb and Zn by using Perkin Elmer (2380) Atomic Absorption Spectrometer (AAS) at wavelength 324.7 nm for Cu, 283.3nm for Pb and 213.9nm for Zn (Welz, 1985). Two plant materials of National Bureau of Standard (USA) each with Nos. 1537 (Tomato leaves) and 1575 (Pine needles) were analyzed following the same procedure and the metal recoveries obtained were 95.5%, 94.2% and 97.5% for Cu, Pb and Zn respectively. The relative standard deviation of the measurements was 2.7% for Cu, 7.5% for Pb and 3.9% for Zn. Ultimately metal accumulation index (MAI) was calculated by the formula given by Liu *et al.* (2007).

(C) Statistical Analysis: All the data analysis was conducted using SPSS program version 17.0. To evaluate the significant differences of various biochemical parameters (for the APTI) and heavy metals (Zn, Cu, Pb, Cr and Ni) concentrations among the study sites in the same species, the data obtained were analyzed statistically using one-way ANOVA followed by Duncan's multiple range test. Data of APTI parameters and heavy metals (Zn, Cu and Pb) were further subjected to Pearson's correlations test to assess the relationship between them.

RESULTS

Ascorbic acid, total chlorophyll, relative water content, potentiality of hydrogen pH and Air Pollution Tolerance Index of different plants at different sites of Kathmandu are given in Table 1.

Ascorbic acid (AA) content in the studied plants ranged from 2.96 ± 0.11 to 8.86 ± 0.38 mg g⁻¹. Out of six tree species two species (*Cinnamomum camphora* and *Grevillea robusta*) had highest AA content in Airport site and the rest four species (*Callistemon lanceolatus*, *Ficus religiosa*, *Jacaranda mimosifolia* and *Populus euroamericana*) had highest ascorbic acid content in Narayanthan, a less polluted site. All other polluted sites (Balaju, Banasthali, Basndhara, Gongobu and Dhumbarahi) mostly had less amount of AA content than in control site.

In all the studied tree species, except *Ficus religiosa*, total chlorophyll (TC) was found to be mostly significantly increased in polluted sites than at control site. TC in all the studied plants, except *Grevillea robusta*, were lowest in Airport. Highest TC in *Cinnamomum camphora*, *Grevillea robusta* and *Populus euroamericana* was recorded in Basundhara, a polluted site. Similarly, highest TC content in *Callistemon lanceolatus* and *Jacaranda mimosifolia* was also recorded in polluted sites, Gongobu and Balaju, respectively.

Relative water content (RWC) in the studied plants ranged from 36.93 ± 10.47 to 96.33 ± 2.13 %. Almost all studied plants, except *Grevillea robusta*, recorded more than 80% RWC in some polluted sites (Table 1). In *Jacaranda mimosifolia*, RWC was significantly higher in all polluted sites than in control site, but in other species this trend was not observed.

The leaf extract pH of *Cinnamomum camphora*, *Ficus religiosa*, *Grevillea robusta* and *Populus euroamericana* reduced significantly ($p=0.05$) in most of the polluted sites than at Narayanthan (Control site). In *Callistemon lanceolatus* the highest leaf extract pH was found in Dhumbarahi (6.45 ± 0.21) and lowest in Airport (3.53 ± 0.03). In *Jacaranda mimosifolia* the highest leaf extract pH was recorded in Basundhara (6.48 ± 0.05) and lowest in Airport (3.53 ± 0.03). pH range in *Callistemon lanceolatus* and *Jacaranda mimosifolia* at polluted and control sites were wide ranging from 3.53 to 6.48 in leaf extract, but in *Cinnamomum camphora* it was comparatively moderate and ranged from 4.66 to 6.82. pH range in *Ficus religiosa*, *Grevillea robusta* and *Populus euroamericana* were comparatively narrow and ranged from 5.04 to 6.82 among polluted and less polluted sites (Table 1). From this it is evident that the species like *Ficus religiosa*, *Grevillea robusta* and *Populus euroamericana* are more tolerant to air pollutants.

APTI values obtained by different species at

different sites ranged from 7.09 (in *Grevillea*) to 15.67 (in *Populus*). Based on the maximum APTI value scored, the plant species from lowest to highest were in the order *Cinnamomum camphora* > *Callistemon lanceolatus* > *Grevillea robusta* > *Ficus religiosa* > *Jacaranda mimosifolia* > *Populus euroamericana* and their respective APTI values were 12.51 ± 0.74 > 12.55 ± 0.78 > $13.08 \pm 0.$ > 13.97 ± 0.54 > 14.61 ± 0.30 > 15.67 ± 0.75 (Table 1).

Heavy metal content and Metal Accumulation Index (MAI) in the leaves

Metal (Zn, Cu and Pb) concentrations in different leaf samples at different sites are given in Table 2. All metals accumulation in the polluted sites were mostly significantly ($p=0.05$) higher than the control site (Narayanthan). Zinc accumulation was highest in *Populus euroamericana* in most of the sites, Cu accumulation was highest in *Jacaranda mimosifolia* at most sites and Pb accumulation was recorded highest in *Callistemon lanceolatus* at the Airport. Accumulation of metals in different plants collected from the same area differed (Table 2).

Among six tree species MAI values (40.69) was highest recorded in the leaves of *Populus euroamericana* collected from Airport site. The mean MAI value obtained from low to high were in the order $7.34 > 9.59 > 9.65 > 15.61 > 15.69 > 20.44$ for *Cinnamomum camphora* > *Grevillea robusta* > *Ficus religiosa* > *Callistemon lanceolatus* > *Jacaranda mimosifolia* > *Populus euroamericana* (Table 2)

Correlations of metals with the physiological parameters of APTI

The correlation of different metal (Zn, Cu and Pb) content and different biochemical parameters (RWC, AA, pH, TC) and APTI of the studied six tree species are given in Table 3. RWC and zinc content mostly showed negative correlation in all studied tree species and was significant ($p=0.05$) in *Callistemon lanceolatus*. Similarly Cu was found to be positively significantly ($p=0.05$) correlated with RWC of *Grevillea robusta* and negatively significant ($p<0.01$) with *Callistemon lanceolatus* and *Jacaranda mimosifolia*. Significant negative correlation was observed between Pb content in leaves and RWC in *Grevillea*. Cumulative effect of Pb+Cu+Zn accumulation was found to be negatively correlated with RWC in all plants and was significant ($p<0.05$) in *Grevillea robusta* and *Jacaranda mimosifolia*.

Mostly negative correlation was observed between AA content and Zn accumulation in the

Table 1. The physiological parameters- (A) Ascorbic Acid, (B) Total chlorophyll, (C) Relative water content (D) potentiality of hydrogen pH and (E) Air Pollution Tolerance Index of different plants at different sites of Kathmandu

Physiological parameters	Plants	Sites							
		Airport	Balaju	Banasthali	Basundhara	Dhumbarahi	Gongbu	Narayanthan	
Ascorbic Acid (mg/g)	<i>Callistemon lanceolatus</i>	7.54±0.79C	5.42±2.12 AB	4.38±2.25 AB	3.78±0.37 A	4.58±1.19 AB	4.28±0.27 AB	7.62±1.69 C	
	<i>Cinnamomum camphora</i>	8.6±0.53 D	3.04±0.32 A	5.28±0.63 C	3.78±0.96 AB	4.56±1.04 BC	2.96±0.11 A	4.5±0.84 BC	
	<i>Ficus religiosa</i>	8.38±0.34 C	3.06±0.23 A	3.32±0.35 A	3.96±0.65 A	3.7±0.80 A	5.22±0.74 B	8.38±0.65 C	
	<i>Grevillea robusta</i> ,	7.28±0.73 C	4.2±1.60 AB	5.34±2.21 B	8.52±0.00 C	5.04±0.74 B	4.94±0.34 AB	5.54±0.32 B	
	<i>Jacaranda mimosifolia</i>	6.28±2.06 BC	6.52±1.68 BC	4.95±1.74 AB	7.86±0.20 CDE	8.36±0.58 DE	3.78±0.38 A	8.64±0.35 E	
Total chlorophyll (mg/g)	<i>Populuseuroa mericana</i>	8.35±0.51 B	8.32±0.3 B	8.6±0.60 B	6.74±2.01 A	8.86±0.38 B	8.35±0.67 B	8.15±0.76 B	
	<i>Callistemon lanceolatus</i>	0.14±0.02 A	0.48±0.05 B	0.61±0.107 C	0.61±0.10 C	0.61±0.08 C	0.74±0.04 D	0.68±0.041 CD	
	<i>Cinnamomum camphora</i>	0.11±0.04 A	0.54±0.06 B	0.66±0.02 CD	0.72±0.11 D	0.57±0.14 BC	0.2±0.05 A	0.67±0.07 CD	
	<i>Ficus religiosa</i>	0.2±0.09 A	0.79±0.37 B	0.4±0.09 A	0.81±0.32 B	0.71±0.02 B	0.8±0.01 B	1.17±0.11 C	
	<i>Grevillea robusta</i> ,	1.12±0.01 E	0.42±0.01 A	1.13±0.02 E	1.14±0.02 E	0.6±0.05 B	0.79±0.03 C	1.03±0.05 D	
Relative Water content (%)	<i>Jacaranda mimosifolia</i>	0.31±0.08 A	1.65±0.00 E	0.93±0.35 BC	1.25±0.21 D	0.93±0.11 BC	1.12±0.059 B	0.79±0.07 A	
	<i>Populuseuroa mericana</i>	0.58±0.07 A	0.71±0.02 AB	0.71±0.02 AB	1.4±0.08 D	0.92±0.04 C	0.69±0.30 AB	0.7±0.11 AB	
	<i>Callistemon lanceolatus</i>	70.83±10.89 BC	57.63±8.52 A	62.92±11.07 AB	62.17±10.18 AB	93.33±4.01E	86.65±3.82 DE	75.4±8.31 C	
	<i>Cinnamomum camphora</i>	60.77±4.64 A	80.85±6.6 B	70.45±4.59 A	83.15±13.60BC	92.37±2.65 C	66±4.66 A	86.08±9.32 BC	
	<i>Ficus religiosa</i>	56.21±6.70 A	96.33±2.13 D	62.92±23.7 AB	75.8±18.13 B	94.35±0.77 CD	69.13±9.53 AB	73.08±6.47 BC	
Potentiality of hydrogen pH	<i>Grevillea robusta</i> ,	69.21±7.08 C	43.95±2.53 AB	69.21±7.08 CD	47.78±3.17 B	75.19±4.06 D	36.93±10.47 A	72.71±13.26 CD	
	<i>Jacaranda mimosifolia</i>	57.25±2.72 B	58.59±14.11 B	86.09±1.52C	58.96±13.09 B	85.76±5.50 C	85.843±3.45C	40.44±2.77 A	
	<i>Populuseuroa mericana</i>	73.9±5.46 BC	74.03±3.05 BC	71.6±17.43ABC	66.32±6.25 AB	89.65±7.04 D	60.03±9.81A	73.39±11.68 BC	
	<i>Callistemon lanceolatus</i>	3.58±0.02 A	5.79±0.01 C	4.57±0.50 B	6.25±0.03 D	6.45±0.21D	6.14±0.03 D	5.57±0.34 C	
	<i>Cinnamomum camphora</i>	5.71±0.02 B	5.98±0.03 CD	5.9±0.02 C	6.62±0.08E	6.62±0.08 E	4.66±0.03 A	6.82±0.13 F	
Air pollution Tolerance Index (APTI)	<i>Ficus religiosa</i>	5.94±0.01 D	6.82±0.02 F	5.04±0.03 B	6.37±0.05 B	6.63±0.03 E	5.63±0.06 C	6.78±0.09 F	
	<i>Grevillea robusta</i> ,	6.24±0.03 C	6.02±0.03 B	6.56±0.04 D	5.78±0.05 A	6.08±0.02 B	6.08±0.01 B	6.76±0.12 E	
	<i>Jacaranda mimosifolia</i>	3.53±0.03 A	5.34±0.13 D	4.9±0.25B	6.48±0.05 F	6.15±0.03 E	4.76±0.03 B	5.14±0.01 C	
	<i>Populuseuroa mericana</i>	6.24±0.03 B	6.32±0.18 BC	6.42±0.05 BC	6.59±0.38B C	6.64±0.15 C	5.18±0.63 A	6.68±0.10 C	
	<i>Callistemon lanceolatus</i>	9.89±1.15 B	9.17±1.19 AB	8.66±2.40 AB	12.08±0.19 C	12.55±0.78 C	11.61±0.50 C	12.26±0.66 C	
Tolerance Index (APTI)	<i>Cinnamomum camphora</i>	11.09±0.60 BC	10.07±0.83 B	10.51±0.84 B	10.87±1.35 BC	12.51±0.74 D	8.03±0.41 A	12.03±1.20 CD	
	<i>Ficus religiosa</i>	10.77±0.65 BC	11.96±0.35 C	8.1±2.35 A	10.43±1.35 A	12.15±0.62 C	10.27±1.26 B	13.97±0.54 D	
	<i>Grevillea robusta</i> ,	13.08±0.61 C	7.1±1.15 A	11.03±2.07281 B	10.67±0.33 B	10.88±0.73 B	7.09±1.02 A	11.59±1.23 BC	
	<i>Jacaranda mimosifolia</i>	8.21±0.82 A	12.32±1.74 BC	8.65±2.57 A	12.01±1.20 BC	14.61±0.30 D	10.79±0.52 B	9.02±0.24 A	
	<i>Populuseuroa americana</i>	13.09±0.77 BC	13.26±0.56 BC	13.54±0.61 C	11.97±1.49 AB	15.67±0.75 D	10.94±1.09 A	13.36±0.97 BC	

Different capital letter along the row after mean ± sd indicates significant difference at p=0.05 obtained from Duncan's multiple range test after one way ANOVA

Table 2. Metal content in the leaves and the Metal Accumulation Index (MAI) in different plants at different sites

Plants	Sites	Zn mg/kg	Cu mg/kg	Pb mg/kg	MAI
<i>Callistemon lanceolatus</i>				Mean	15.61
	Airport	15.54±0.42 C	5.41±0.62 EF	40.33±1.84 E	20.09
	Balaju	38.46±0.91 F	5.87±0.30 F	32.41±0.93 C	32.09
	Banasthali	43.76±3.23 G	7.56±0.38 G	0.83±0.67 A	11.65
	Bsundhara	12.15±0.68 B	3.63±0.25 CD	0.73±0.49 A	11.25
	Dhumbarahi	20.83±1.13 DE	2.39±0.53 B	17.10±1.04 B	13.09
	Gongbu	21.53±0.84 E	0.93±0.42 A	37.56±1.49 D	17.61
	Narayanthan	4.22±1.51 A	4.81±0.78 E	0.61±0.37 A	3.54
<i>Cinnamomum camphora</i>				Mean	7.34
	Airport	23.23±2.85 D	8.79± 0.90 CD	13.53±1.60 B	8.78
	Balaju	15.69±1.38 B	5.23±1.04 AB	32.06±2.16 D	10.42
	Banasthali	1.40±0.59 A	7.99±0.88 CD	0.83±0.58 A	4.29
	Basundhara	18.07± 2.25 BC	3.39±2.25 A	24.99±4.28 C	5.11
	Dhumbarahi	20.11± 2.13 C	9.29±1.54 D	2.86±1.62 A	5.74
	Gongbu	20.03±0.99 C	6.51±0.91 BC	21.87±1.85 C	13.05
	Narayanthan	1.27± 0.64 A	13.93±1.79 E	0.84± 0.38 A	3.99
<i>Ficus religiosa</i>				Mean	9.65
	Airport	16.02±1.71 D	9.13±0.78 D	21.42±1.47 C	11.90
	Balaju	22.20±2.42 E	7.41±1.40 C	39.61±2.39 E	10.33
	Bansthali	22.37±2.27 E	11.09±1.67 E	36.65±2.73 D	9.98
	Basundhara	8.79±1.06 B	0.90±0.09 A	0.053±0.01 A	7.06
	Dhumbarahi	12.71±1.12 C	7.91±0.96 CD	3.82±0.76 B	8.17
	Gongbu	13.13± 1.62 C	3.93±0.23 B	35.83±2.06 D	14.18
	Narayanthan	1.54±0.51 A	15.28±1.49 F	0.50±0.10 A	5.95
<i>Grevillea robusta</i>				Mean	9.59
	Airport	22.093±2.86 C	9.29±1.01 D	19.83±1.51 D	10.03
	Balaju	15.06±3.42 AB	6.20±0.72 BC	37.8±1.56 F	12.40
	Banasthali	13.49±0.90 AB	7.58±1.39 CD	16.92±1.41 C	10.80
	Basundhara	14.03±0.91 AB	2.96± 0.50 A	12.84± 2.12 B	9.11
	Dhumbarahi	15.2±2.43 AB	5.14±0.69 B	24.34±2.99 E	7.28
	Gongbu	12.01±2.59 A	2.97±0.89 A	36.67±1.15 F	13.23
	Narayanthan	12.45±2.21 A	8.16±1.61 CD	4.97± 2.25 A	4.31
<i>Jacaranda mimosifolia</i>				Mean	15.69
	Airport	34.98±1.56 E	19.93±1.68 E	25.06±1.38 E	17.47
	Balaju	31.18±1.28 D	20.27±0.64 E	3.70±1.37 A	19.52
	Banasthali	22.68±1.33 B	5.72± 0.43 B	29.37±1.67 F	15.97
	Basundhara	23.34±2.00 BC	13.83±1.35 D	19.42±0.64 D	17.37
	Dhumbarahi	25.54±0.84 C	11.72±0.71 C	11.58±1.21 B	18.78
	Gongbu	22.54±0.84 B	11.61±1.12 C	15.52±1.04 C	17.44
	Narayanthan	0.96± 0.45 A	2.45± 0.47 A	1.40±0.56 A	3.28
<i>Populus euroamericana</i>				Mean	20.44
	Airport	104.27±1.02 E	11.31±0.63 D	0.93±0.59 A	40.69
	Balaju	69.06±3.32 D	8.38±0.53 C	27.51±1.74B	17.37
	Banasthali	132.38±3.71 F	11.93±0.60 D	0.83±0.58 A	18.96
	Basundhara	57.89±1.01 BC	0.89±0.10 A	1.66±1.52 A	25.71
	Dhumbarahi	57.09±2.71 BC	1.86±1.06 AB	1.54±0.99 A	8.14
	Gongbu	65.61±1.30 CD	1.26±1.07 AB	2.32±0.84 A	18.12
	Narayanthan	14.46±0.47 A	2.41± 0.86 B	0.59±0.06A	14.13

Different capital letter after mean±sd along the column indicates significant difference at p=0.05 obtained from Duncan's multiple range test after one way ANOVA

studied tree leaves and was significant (p<0.05) in *Jacaranda mimosifolia*. Significant positive correlation was observed between Cu accumulation and AA in

Ficus religiosa. Pb accumulation mostly negative correlation with AA and was significant in *Cinnamomum camphora* but in *Jacaranda mimosifolia*

it was positively significant ($p < 0.01$). Cumulative effect of Pb+Cu+Zn accumulation was found to be significantly ($p < 0.05$) negatively correlated with AA in *Grevillia robusta*.

Mostly negative correlation was observed between leaf extract pH and Zn content, and was significant ($*p < 0.05$) in *Cinnamomum camphora* and ($p < 0.01$) *Jacaranda mimosifolia*. Cu content in leaf was mostly found to be positive correlation with pH in most of the tree species and was significant ($p < 0.01$) in *Cinnamomum camphora* but in *Callistemon lanceolatus* and *Jacaranda mimosifolia* it was found to be significantly ($p < 0.01$) negatively correlated. Pb content in leaf showed negative correlation with pH in most trees and was significant ($p < 0.01$) in *Grevillea robusta*, *Cinnamomum camphora* and *Jacaranda mimosifolia*. Cumulative effect of Pb + Cu + Zn accumulation was found to be mostly negatively correlated with pH in most trees and was significant ($p < 0.05$) in *Grevillea robusta* and *Cinnamomum camphora*.

Zinc accumulation in leaves showed negative correlation with total chlorophyll in most of the trees and was significant ($p < 0.01$) in *Ficus religiosa*, *Jacaranda mimosifolia* and *Populus euroamericana*. Cu content in leaves was significantly ($p < 0.01$) negatively correlated with TC in *Jacaranda*

mimosifolia whereas Pb content in leaf showed significant ($p < 0.05$) negatively correlated with TC in *Ficus religiosa*. Cumulative effect of Pb+Cu+Zn content in leaves were negatively correlation in most of the studied plants and was significant in *Ficus religiosa* and *Jacaranda mimosifolia*.

APTI was found to be negatively correlated with Zinc accumulation in most trees and was significant ($p < 0.01$) in *Callistemon lanceolatus* and *Jacaranda mimosifolia*. Cu content in leaves showed negative correlation with APTI in some studied tree species and was significant ($p < 0.05$) in *Ficus religiosa*, *Callistemon lanceolatus* and *Jacaranda mimosifolia* but in *Cinnamomum camphora* it was significantly ($p < 0.01$) positive. Pb content showed negative correlation with APTI in most studied tree species and was significant ($p < 0.01$) in *Ficus religiosa*, *Populus euroamericana*, *Cinnamomum camphora* and *Grevillea robusta*. Cumulative effect of Pb+Cu+Zn accumulation was found to be mostly significant negatively correlated in all studied plants and was significant ($p < 0.05$) in *Grevillea robusta*, *Cinnamomum camphora*, *Callistemon lanceolatus* and *Jacaranda mimosifolia*.

DISCUSSION

Depending on the APTI value, Padmavathi *et al.*

Table 3. Correlations of metals with RWC, AA, pH, TC AND APTI

Parameters	<i>Ficus religiosa</i> (n=35)	<i>Grevillia robusta</i> (n=50)	<i>Cinnamomum-camphora</i> (n=35)	<i>Callistemon lanceolatus</i> (n=50)	<i>Jacaranda mimosifolia</i> (n=40)	<i>Populus euroamericana</i> (n=45)
RWC-Zn	-0.309	-0.110	-0.180	-0.404**	-0.159	-0.065
RWC-CU	-0.232	0.344*	0.166	-0.643**	-0.0552**	0.126
RWC-Pb	-0.200	-0.389**	-0.125	0.089	-0.158	-0.087
RWC-Zn+Cu+Pb	-0.293	-0.305*	-0.158	-0.208	-0.387*	-0.064
AA-Zn	-0.307	-0.243	0.111	-0.160	-0.369*	0.204
AA-Cu	0.508**	-0.034	0.276	0.199	-0.118	0.046
AA-Pb	0.245	-0.261	-0.376*	-0.023	0.472**	-0.219
AA-Zn+Cu+Pb	0.303	-0.338*	-0.162	-0.083	0.219	0.137
pH-Zn	-0.152	0.049	-0.411*	-0.172	-0.667**	-0.145
pH-Cu	0.732	0.095	0.530**	-0.513**	-0.580**	0.184
pH-Pb	0.427	-0.409**	-0.497**	-0.010	-0.457**	0.071
pH-Zn+Cu+Pb	0.304	-0.330*	-0.486**	-0.140	-0.089	-0.090
Tchl-Zn	-0.754**	0.047	-0.240	0.070	-0.780**	-0.337*
Tchl-Cu	0.097	0.173	0.008	-0.155	-0.827**	-0.030
Tchl-Pb	-0.346*	-0.137	-0.011	0.001	0.031	0.264
Tchl-Zn+Cu+Pb	-0.346*	-0.060	-0.137	0.025	-0.529**	-0.245
APTI-Zn	0.329	-0.212	-0.144	-0.436**	-0.530**	0.078
APTI-Cu	-0.953*	0.243	0.438**	-0.514**	-0.665**	-0.127
APTI-Pb	-0.983**	-0.475**	-0.469**	-0.018	0.061	-0.775**
APTI-Zn+Cu+Pb	-0.482	-0.451**	-0.338*	-0.286*	-0.368*	-0.135

Significance * $p < 0.05$ ** $p < 0.01$ *** $p < 0.00$ significant ($p = 0.05$)

(2013) grouped the plants as sensitive (with APTI value <11), intermediate (with APTI value 12 to 16) and tolerant (with APTI value \geq 17). In the present study all the studied tree species scored APTI value between 12 and 16, hence they were in the intermediate categories of APTI.

Metal Accumulation Index (MAI) depends on various factors like, local atmospheric chemistry, meteorology, sampling height (tree), time of sampling and plant characteristics (Hu *et al.*, 2014). Plants with higher MAI values have good accumulation capacities and also regarded as tolerant species. Hu *et al.*, (2014). From the present study plants like *Populus euroamericana*, *Jacaranda* and *Callistemon* with high MAI value are suitable tree species that can be suggested to be planted for greenbelt area along the road side.

The significant correlation between RWC and all metal accumulation was mostly negative, except Cu accumulation and RWC in *Grevillea robusta*. The excess concentrations of the heavy metals significantly affected plant water status, causing water deficit and subsequent changes in the plants. The most intensive effect on the plants was observed with Cd, less intensive by Cu and Zn and the least intensive by Pb in sunflower (Kastori *et al.* 1992). This might be the reason for showing insignificant relation of Pb accumulation in most of the trees except *Grevillea robusta*. Lead accumulation in root of *Lupinus* has been reported to increase the degree of vacuolization in the meristem cells (Przymusiński and Wozny 1985) and cortex parenchyma (Gzyl *et al.*, 1997), which may suggest that the water status of these cells was not affected by the metal. Thus, Pb-induced vacuolization in *Lupinus luteus* was correlated with high values of RWC, suggesting that water might be stored in the vacuoles in response to metal stress (Rucińska-Sobkowiak *et al.*, 2013). The response of RWC in *Grevillea robusta* to Pb accumulation in the present study was significantly negative, which indicates that the Pb toxicity prevailed in the leaves. From this it can be suggested that the Pb localization in the cell and its toxicity might not be same in all plants. The correlation of RWC and Cu accumulation was found to be positively significant in *Grevillea robusta*. This might be due to low concentration of Cu (below 10 mg/kg), which is an essential element and mostly not toxic below 15-20 mg/kg (Pahlsson, 1989).

Correlation between AA and Zn accumulation in *Jacaranda mimosifolia* was significantly negative.

Smith *et al.* (1989) reported the decrease in ascorbic acid in roots and shoots of pigeon pea with increasing concentrations of Zn. Correlation of AA content with Cu accumulation in *Ficus religiosa* and Pb accumulation in *Jacaranda mimosifolia* were significantly positive. Copper is an essential element and its maximum accumulation in *Ficus* was 15.28 mg/kg in control site, indicating nontoxic level of Cu accumulation in most of the sites, which might have resulted in positive correlation. Though Pb is not an essential element, but plant absorb Pb and mostly accumulate in the cell wall as granules (Peterson, 1978; Kabata-Pandias and Pandis, 1985; Chettri *et al.*, 2000). In the present study Pb accumulation in *Jacaranda mimosifolia* leaves reached up to 29.79 mg/kg, and showed positive correlation with Ascorbic acid, which might be due to accumulation of Pb on cell wall and not interfering with the metabolic functions. But in Camphor Pb accumulation and AA showed negative correlation. Variation in Ascorbic acid content is one of the factors, which may be held responsible for differential tolerance capacity of plants against various air pollutants at different sites (Aglar-Silva *et al.*, 2016; Aghajanzadeh *et al.*, 2016). In the present study Ascorbic acid content mostly increased with increase in heavy metal content and also in accordance with other findings of Rai and Panda (2014) and Govindaraju *et al.* (2012).

Correlations of pH with most of the metal content in leaf were negatively significant, except Cu content in *Cinnamomum camphora*. This must be due to reduction of leaf pH value in the presence of heavy metals and an acidic pollutant (Scholz and Reck, 1977), and also suggested that the leaf pH is reduced and the reducing rate is more in sensitive plants compared to that in tolerant plant species. The positive correlation of pH with Cu content in Camphor is mainly due to the fact that both Cu content and leaf pH in Narayanthan is significantly high in this site than in other studied sites. Besides, the Cu is an essential elements and its content in *Cinnamomum camphora* is below the toxic level as suggested by Pahlsson, (1989).

Correlations between total chlorophyll and heavy metal content were negatively significantly in most cases. Heavy metals such as (Hg, Cu, Cr, Cd, and Zn) have been reported to decrease the chlorophyll content in various plants in most cases (Aggarwal *et al.*, 2012). This decline in photosynthetic pigments is most probably due to the inhibition of the reductive steps in the biosynthetic pathways of

photosynthetic pigments due to the high redox potential of many heavy metals. Among the studied plants, *Grevillea*, *Camphora* and *Callistemon* showed insignificant correlation of total chlorophyll with all metals. In the case of *Grevillea robusta* and *Cinnamomum camphora* this might be due to less metal accumulation as indicated by mean metal accumulation index (MAI) in these species. In the case of *Callistemon lanceolatus* though the MAI value is 15.61, the accumulation of Zn and Cu is within the threshold limit and below the toxic level. Besides this Pb accumulation in leaves are up to 40 mg/kg but negative correlation with total chlorophyll was not observed, indicating no effect of Pb on chlorophyll content. The possible reasons for this may be their localization in the cell.

Ultrastructural studies have revealed that Pb are deposited mainly in the intercellular space, cell wall and vacuoles (Wierzbicka and Antosiewicz, 1993; Chettri *et al.*, (2000). A small deposits of Pb have also been reported to be seen in the ER, dictyosomes and dictyosome derived vesicles (Wierzbicka and Antosiewicz, 1993). (*Callistemon lanceolatus*, *Ficus religiosa*, *Populus euroamericana*, *Cinnamomum camphora*, *Grevillea robusta*, *Jacaranda mimosifolia*)

Significant Correlation of APTI with all metal content are mostly negative in all cases except with Cu content in *Cinnamomum camphora*. APTI is predicted on the basis of four major biochemical properties of leaves that are ascorbic acid, relative water content, total chlorophyll and leaf pH (Sing and Rao, 1983). APT varies with these parameters because all these parameters are influenced by the increase or decrease in metal content. From MAI it is evident that *Populus euroamericana*, *Jacaranda mimosifolia* and *callistemon lanceolatus* scored high MAI value including that the heavy metals accumulation might have affected the physiological parameters and this might have resulted in to negative correlation. Though *Cinnamomum camphora*, *Grevillea robusta* and *Ficus religiosa* scored less MAI values, out of these avoidance mechanism in *Cinnamomum camphora* might be high and resulted in to positive response to APTI with Cu accumulation.

CONCLUSION

The APTI values of all studied tree species were within intermediate range. Because of high MAI value, *Populus euroamericana*, *Jacaranda mimosifolia*

and *Callistemon lanceolatus* have been identified as the suitable species for the plantation near the road side. Though *Cinnamomum camphora*, *Grevillea robusta* and *Ficus religiosa*, growing in the same polluted environment with others scored less MAI value, indicating the metal avoidance mechanism in them. From above findings it can be concluded that Air pollution tolerance index (APTI) value of plants could not only be sufficient for selection of plants to mitigate air pollution. For selection of more suitable plants, Metal Accumulation Index (MAI) value of plants should also be considered. Hence *Populus euroamericana*, *Jacaranda mimosifolia* and *Callistemon lanceolatus* with high MAI and intermediate APTI range are recommended for plantation on road side of Kathmandu.

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