

A STUDY ON VARIATION IN BIOCHEMICAL ASPECTS OF DIFFERENT TREE SPECIES WITH TOLERANCE AND PERFORMANCE INDEX

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ABSTRACT

The present study was undertaken to evaluate the variation between biochemical characteristics and air pollution tolerance index (APTI) of 6 different plant species. The results of the present study indicate that APTI was significantly correlated with total chlorophyll, ascorbic acid, leaf pH for all species and are the most significant and determining factors on which the tolerance depends. The order of tolerance index of plant species is as follows *Saraca indica* (13.71), *Azadirachta indica* (12.98), *Shorea robusta* (12.64), *Eucalyptus sp.* (12.61), *Ficus religiosa* (12.61) and *Tectona grandis* (13.33). According to anticipated performance index (API) all species were tolerant i.e. *Azadirachta indica*, *Ficus religiosa*, *Saraca indica*, *Tectona grandis* (75%), *Shorea robusta* and *Tectona grandis* (68%). The present study suggests that evaluation of plant tolerance and performance index might be very useful in the selection of appropriate species which can be expected to perform well for the development of green environment.

INTRODUCTION

Plants play an important role in monitoring and maintaining the ecological balance and also provide enormous leaf area for impingement, absorption and accumulation of air pollutants to reduce the pollution level in the environment (Escobedo *et al.*, 2008). Sensitivity and response of plants to air pollutants is variable. The identification and categorization of plants into sensitive and tolerant groups is important because the former can serve as indicators and the latter as sink for the air pollutants in urban and industrial habitats (Kuddus *et al.*, 2011). Most plants experienced physiological changes before exhibiting visible damage to leaves when exposed to air pollutants (Liu and Ding, 2008). To screen plants for their sensitivity/tolerance level to air pollutants, large number of plants parameter have been used including leaf or stomatal conductance, ascorbic acid, relative water content, membrane permeability, peroxidase activity, chlorophyll content and leaf extract pH (Farooq and Beg, 1980, William and Christopher, 1986, Tripathi *et al.*, 1991, Ninave *et al.*, 2001). To indicate the susceptibility level of plant, pollution-induced changes in individual parameters are usually quantified and correlated with the level of plant response.

Tolerance level of plants can be evaluated by calculating an index known as air pollution tolerance index (APTI) based on four biochemical & physiological parameters *viz.* chlorophyll, ascorbic acid, pH, relative water content (Singh and Rao, 1983, Dwivedi *et al.*, 2008). Combining the tolerance index of plants with some relevant biological and socioeconomic characters, the anticipated performance index (API) was determined.

However, the combination of these biochemical and physiological parameters gave a more reliable result than those of individual parameter. Therefore, the aim of the present study is to determine the variation in biochemical parameters to establish the susceptibility level of different tree species with reference to their tolerance and performance index which might be very useful in the selection of appropriate species which can be expected to perform well for the development of green environment.

MATERIALS AND METHODS

The present study was carried out within the different intersection points of Haridwar city, located in the state of Uttarakhand, is one of the important holy cities of India and is extended from latitude 29° 30' in the north to longitude 78° 43' in the east with a subtropical climate during the time period of July 2011 to June 2012. The sites selected for the present study includes Forest Area (S-I), Urban Area (S-II) and Industrial Area (S-III). Rapid industrialization in the city increases the density of vehicles which further increased the load of vehicular concentration on the roads of Haridwar which directly affects the road side plants which remain in direct contact with these types of pollutants. Therefore, road side plant species were selected for the study: *Shorea robusta* (Sal) & *Tectona grandis* (Teak) from S-I, *Azadirachta indica* (Neem) & *Ficus religiosa* (Peepal) from S-II and *Saraca indica* (Ashoka) & *Eucalyptus sp.* (Eucalyptus) from S-III. Five triplicates of fully matured leaves from these tree species were randomly collected from all sampling sites and immediately taken to the

laboratory for physiological and biochemical analysis.

Physiological and biochemical analysis

The following physiological & biochemical parameters were analyzed: leaf extract pH was determined by pH meter after calibrating with buffer solution, Relative Water Content (RWC) by estimating the turgid & dry weight of leaf samples (Weatherly, 1965), Total Chlorophyll (T Chl) by Arnon's equation (1949) and Ascorbic Acid (AA) by 2,6- Dichlorophenol- Indophenol Visual Titration method (Sadasivam & Manickam, 1992).

Air pollution tolerance index (APTI)

The values of air pollution tolerance index (APTI) were determined by the method of Singh and Rao, 1983.

Anticipated performance index (API)

By combining the resultant APTI values with some relevant biological and socio-economic characters (plant habit, canopy structure, type of plant, laminar structure & economic values), the API was calculated for different species. Based on these characters, different grades (+ or -) are allotted to plants. Different plants are scored according to their grades (Mondal *et al.*, 2011). The criteria used for calculating the API of different plant species are given in Table 1 and 2.

Statistical analysis

Correlation coefficient was calculated between independent variable viz. pH, T Chl, RWC, Ascorbic acid and dependent variable such as APTI to determine the degree of correlation between the variables by using Excel, 2007. For all primary data, n = 12 and significance was tested at 1% and 5% level of significance (*i.e.* $p = 0.01$ & 0.05).

RESULTS AND DISCUSSION

The analyzed value for 4 biochemical parameters along with tolerance index value has been presented in Table 3.

Physiological and biochemical characteristics

The mean value of leaf extract pH ranged from 6.63 (*Tectona grandis*) to 6.22 (*Eucalyptus* sp.) which were found to be acidic in nature for all plant species across the sampling sites which may be due to the presence of SO_2 and NO_x in the ambient air causing a change in pH of leaf sap (Swami *et al.*, 2004). High pH may increase the efficiency of conversion from hexose sugar to AA, while low leaf extract pH showed good correlation with sensitivity to air pollution (Conklin, 2001). The pH ranged between 4.4 and 8.8 lies in both intermediately tolerant and sensitive plant species (Lakshmi *et al.*, 2009). Thus, all species are intermediately tolerant and sensitive.

RWC is the water content of a given amount of leaf relative to its fully hydrated or turgid state. The large quantity of water (in terms of RWC) in plant body helps in maintaining its physiological balance under stress conditions of pollution (Gonzalez and Reigosa, 2001). The maximum value of relative water content was recorded in *Saraca indica* (84.32%) at industrial area and minimum value was recorded for *Tectona grandis* (70.36 %) at forest area followed by other tree species (Table 3). Maximum value of RWC at polluted site further provide evidence that plants retain more water than those at less polluted site this might be due to adaptive feature which helps in maintaining its physiological balance against pollution stress which may be tolerant to pollutants. Kuddus *et al.*, 2011 also recorded higher values of RWC *i.e.* 89.86 % under polluted conditions.

Table 1: Gradation of plant species on the basis of air pollution tolerance index (APTI) and other biological and socio-economic characters

Grading	Character	Pattern of assessment	Grade allotted
(a) Tolerance	Air pollution tolerance Index (APTI)	12.0-16.0	+
		16.1-20.0	++
		20.1-24.0	+++
		24.1-28.1	++++
		28.1-32.0	+++++
		32.1-36.0	++++++
(b) Biological and Socio-Economic	(i) Plant habit	Small	-
		Medium	+
		Large	++
	(ii) Canopy structure	Sparse/irregular globular	-
		Spreading crown/ open semi dense	+
		Spreading dense	++
	(iii) Type of plant	Deciduous	-
		Evergreen	+
	(iv) Laminar structure size	Small	-
		Medium	+
		Large	++
	Texture	Smooth	-
		Coriaceous	+
	Hardiness	Delineate	-
		Hardy	+
(v) Economic value	Less than three uses	-	
	Three or four uses	+	
	Five or more uses	++	

Maximum grades that can be scored by a plant = 16

Foliar ascorbic acid is generally accepted as a good biomonitoring system. It activates many physiological and defence mechanisms and its reducing power has been known to be directly proportional to its concentration (Lewin, 1976). The minimum value of ascorbic acid recorded in *Shorea robusta* (5.65 mg/g) at S-I followed by *Azadirachta indica* (5.71 mg/g), *Tectona grandis* & *Eucalyptus sp.* (5.83 mg/g), *Ficus religiosa* (5.99 mg/g) and *Saraca indica* (6.49 mg/g) and this increase level of ascorbic acid may be due to the defence mechanism of the species against pollution load at different sites. Similarly, Bhattacharya *et al.*, 2012 also recorded higher values of ascorbic acid in plant leaves at polluted sites. The free radical production under SO₂ exposure would increase the free radical scavengers such as ascorbic acid, superoxide

dismutase and peroxidase (Pieree and Queiroz, 1981).

Chlorophyll pigments exist in highly organized state, and under stress they may undergo several photochemical reactions such as oxidation, reduction, pheophytinisation and reversible bleaching (Puckett *et al.*, 1973). Hence, any alteration in chlorophyll concentration may change the morphological, physiological and biochemical behaviour of the plant. Reduction in total chlorophyll content of *Saraca indica* (1.80 mg/g) and *Eucalyptus* (1.85 mg/g) species at S-III, *Azadirachta indica* (1.89 mg/g) and *Ficus religiosa* (2.19 mg/g) at S-II compared to *Shorea robusta* (2.58 mg/g) & *Tectona grandis* (2.54 mg/g) at S-I, showed that S-III and S-II was exposed to higher level of pollutants concentration which can be used as an indicator of air pollution. The plants having chlorophyll content between 4 to 16 mg/gm and 0.90 to 9.38 mg/gm are categorized as intermediately tolerant and sensitive plant species respectively (Lakshmi *et al.*, 2009). The reduction in chlorophyll content in the polluted leaves could be due to chloroplast damage (Pandey *et al.*, 1991), inhibition chlorophyll biosynthesis (Esamt, 1993) or enhanced chlorophyll degradation.

Air pollution tolerance index (APTI)

It is evident from Table 3 that different plants respond differently

Table 2: Anticipated Performance Index (API) of plant species

Grade	Score (%)	Assessment category
0	Up to 30	Not recommended
1	31-40	Very poor
2	41-50	Poor
3	51-60	Moderate
4	61-70	Good
5	71-80	Very good
6	81-90	Excellent
7	91-100	Best

Table 3: Mean value of biochemical parameters \pm S.E with tolerance index of leaf samples

Plant species	pH	Relative water content (%)	Chlorophyll content (mg/g)	Ascorbic acid (mg/g)	APTI
<i>Shorea robusta</i>	6.57 \pm 0.021	72.31 \pm 0.070	2.58 \pm 0.012	5.65 \pm 0.020	12.64
<i>Tectona grandis</i>	6.63 \pm 0.021	70.36 \pm 0.069	2.54 \pm 0.013	5.83 \pm 0.018	12.43
<i>Azadirachta indica</i>	6.29 \pm 0.021	83.07 \pm 0.075	1.89 \pm 0.011	5.71 \pm 0.020	12.98
<i>Ficus religiosa</i>	6.45 \pm 0.020	75.35 \pm 0.072	2.17 \pm 0.011	5.99 \pm 0.020	12.61
<i>Saraca indica</i>	6.31 \pm 0.021	84.32 \pm 0.074	1.80 \pm 0.011	6.49 \pm 0.021	13.71
<i>Eucalyptus sp.</i>	6.22 \pm 0.020	79.00 \pm 0.072	1.85 \pm 0.011	5.83 \pm 0.020	12.61

Table 4: Correlation between the biochemical parameters and APTI values of *Shorea robusta*

	pH	RWC	T Chl	AA	APTI
pH	1				
RWC	-0.6144*	1			
T Chl	0.82162**	-0.6687*	1		
AA	0.89377**	-0.4612	0.716952*	1	
APTI	0.83059**	-0.4721	0.535136	0.91189**	1

**significant at 0.01%, *significant at 0.5%

Table 6: Correlation between the biochemical parameters and APTI values of *Azadirachta indica*

	pH	RWC	T Chl	AA	APTI
pH	1				
RWC	-0.78378**	1			
T Chl	0.55847	-0.5351	1		
AA	0.742801**	-0.6584*	0.006667	1	
APTI	0.914634**	-0.7374**	0.383875	0.91465**	1

**significant at 0.01%, *significant at 0.5%

Table 8: Correlation between the biochemical parameters and APTI values of *Saraca indica*

	pH	RWC	T Chl	AA	APTI
pH	1				
RWC	-0.8893**	1			
T Chl	0.903298**	-0.9944**	1		
AA	0.814182**	-0.6548*	0.650294*	1	
APTI	0.837559**	-0.6331*	0.63955*	0.991525**	1

**significant at 0.01%, *significant at 0.5%

Table 5: Correlation between the biochemical parameters and APTI values of *Tectona grandis*

	pH	RWC	T Chl	AA	APTI
pH	1				
RWC	-0.68906*	1			
T Chl	0.792551**	-0.5065	1		
AA	0.911336**	-0.7117*	0.729678**	1	
APTI	0.920984**	-0.6289*	0.843598**	0.976849**	1

**significant at 0.01%, *significant at 0.5%

Table 7: Correlation between the biochemical parameters and APTI values of *Ficus religiosa*

	pH	RWC	T Chl	AA	APTI
pH	1				
RWC	-0.69991*	1			
T Chl	0.468837	-0.4267	1		
AA	0.948357**	-0.7703**	0.4586	1	
APTI	0.742223**	-0.1318	0.427468	0.719722*	1

**significant at 0.01%, *significant at 0.5%

Table 9: Correlation between the biochemical parameters and APTI values of *Eucalyptus sp.*

	pH	RWC	T Chl	AA	APTI
pH	1				
RWC	-0.81489**	1			
T Chl	0.803817**	-0.76791**	1		
AA	-0.29261	0.137244	-0.16537	1	
APTI	0.457377	-0.32369	0.712167*	0.462203	1

**significant at 0.01%, *significant at 0.5%

Table 10: Evaluation of plant species on the basis of APTI value and some biological and socioeconomic characters

Common name	Scientific name	Assessment parameters				Laminar structure			Grade allotted		% Scoring	API grade
		APTI	Tree habit	Canopy structure	Tree type	Size	Texture	Hardiness	Economic importance	Total plus		
Sal	<i>Shorea robusta</i>	+	++	++	-	++	+	+	++	11	68	Good
Teak	<i>Tectona grandis</i>	+	++	++	-	++	+	+	++	11	68	Good
Neem	<i>Azadirachta indica</i>	+	++	++	+	++	+	+	++	12	75	Very Good
Peepal	<i>Ficus religiosa</i>	+	++	++	+	++	+	+	++	12	75	Very Good
Ashoka	<i>Saraca indica</i>	+	++	++	+	++	+	+	++	12	75	Very Good
Eucalyptus	<i>Eucalyptus sp.</i>	+	++	++	+	++	+	+	++	12	75	Very Good

Table 11: Anticipated Performance Index (API) of Plant Species

S. No.	Local name	Scientific name	Grade allotted			Assessment
			Total	%	API value	
1	Sal	<i>Shorea robusta</i>	11	68	4	Good
2	Teak	<i>Tectona grandis</i>	11	68	4	Good
3	Neem	<i>Azadirachta indica</i>	12	75	5	Very good
4	Peepal	<i>Ficus religiosa</i>	12	75	5	Very good
5	Ashoka	<i>Saraca indica</i>	12	75	5	Very good
6	Eucalyptus	<i>Eucalyptus sp.</i>	12	75	5	Very good

to air pollutants. Variation in 4 physiological & biochemical (pH, relative water content, ascorbic acid and total chlorophyll) aspects of plant species results in the variation in APTI values. Thus, it can be stated that each parameter plays a significant role in the determination of the susceptibility level of the plant which governs the computation of the index. The maximum APTI values were recorded for *Saraca indica* (13.71 %) followed by other species *Azadirachta indica* (12.98 %), *Shorea robusta* (12.64 %), *Ficus religiosa* & *Eucalyptus sp.* (12.61 %) and *Tectona grandis* (12.43 %). Mishra *et al.*, 2012 also recorded the higher values of APTI in *Saraca indica*, *Ficus religiosa*, *Mangifera indica*, *Azadirachta indica* and *Alstonia scholaris* in the high pollution zone.

The species having APTI value <10 is considered as 'sensitive'; value within 10-16 is considered as 'intermediate' and > 17 is 'tolerant' (Chaudhary and Banerjee, 2009). In the present study all species can be used for biomonitoring of polluted area to indicate the level of air pollution because all species are intermediately sensitive and tolerant.

Correlation matrix interpretation

The correlation coefficient values of physiological and biochemical parameters *viz.* pH, relative water content, total chlorophyll and ascorbic acid with air pollution tolerance index (APTI) of different tree species are presented in Table 4-9. The results show significant positive correlation between APTI and different biochemical parameters except relative water content for all species at different sites. It indicates that ascorbic acid & total chlorophyll content of leaf are the most significant and determining factors on which the tolerance depends. Thambavani Sabitha (2011) also studied high positive correlation between APTI with chlorophyll and ascorbic acid.

Anticipated performance index (API)

Plant species were evaluated for various biological and socio-economic as well as a few biochemical characteristics, *viz.* APTI, plant habitat, canopy structure, type of plant, laminar structure and economic values. These parameters were subjected to a grading scale (Table 1) to determine the anticipated performance index (API) of plant species as advocated in reference. The grading pattern of 6 plant species

evaluated in Table 10, with respect to their anticipated performance index.

A comparison of the assessment parameters *w.r.t.* grading characters using a multiplication or summation of the anticipated performance of plant species found those parameters to be quite similar. Table 11, showed that all species are tolerant and good performer. Among which *Azadirachta indica*, *Ficus religiosa*, *Saraca indica* and *Eucalyptus sp.* were categorized as very good performer having spreading dense canopy of evergreen foliage, providing protection from pollution stress. These species has well known economic and aesthetic values and thus recommended for extensive planting. Thus, anticipated performance index might be very useful in the selection of appropriate species.

The results of the study concluded that each physiological & biochemical parameter plays a significant role in the determination of the susceptibility level of the plant species with reference to their tolerance and performance index. Estimation of these indices will be a reliable method for the selection of appropriate species which can be used as bioindicators and mitigators of pollutants in an urban and industrial region.

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