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Physiological responses of some tree species under roadside automobile pollution stress around city of Haridwar, India

P. C. Joshi · Abhishek Swami

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Abstract Studies were carried out to determine the physiological response of few economically important tree species viz., Mango (*Mangifera indica*), *Eucalyptus citriodora*, Sagon (*Tectona grandis*) and Sal (*Shorea robusta*) to roadside automobile pollution during 2004–2005. By determining some physiological parameters, which included chlorophyll *a*, and *b*, total chlorophyll, carotenoids, ascorbic acid, pH and relative water content, impact of automobile exhaust on these species was assessed. The data obtained were further analyzed by using one-way ANOVA and a significant change in all these parameters was found in the leaf samples collected from road side trees, exposed to automobile exhausts in comparison to control. Higher value of air pollution tolerance index (APTI) was recorded for *S. robusta* (9.02) while the minimum value of APTI was recorded for *M. indica* (6.76).

Keywords Roadside pollution · Automobiles · Impact · APTI · Chlorophyll

1 Introduction

With rapid development of human civilization and industrialization the number of automobiles has also increased, which are responsible for almost 65% of air pollution. In recent past, air pollutants, responsible for vegetation injury and crop yield losses, are causing increased concern (Fuji 1973). $SO_{2,}$ which is a major component of air pollutants, affects morphological characteristics of plants such as number of leaves, leaf area, length of stem and roots and number of flowers and fruits (Wali et al. 2004). Air pollutants also affect germination of seeds, length of pedicles, and, number of flowers in inflorescence (Nithyamathi and Indira 2005). Several studies have shown the impact of automobile exhaust on road side vegetation through their visible and

P. C. Joshi (✉) · A. Swami
Department of Zoology and Environmental Sciences, Gurukula Kangri University, Haridwar 249 404, India
e-mail: prakash127@yahoo.com

non visible effects (Farmer and Lyon 1977; Fleckiger et al. 1978; Keller 1974; Sarkar et al. 1986; Leon 1988; Nuhoglu 2005). One of the most common impacts is the gradual disappearance of chlorophyll and concomitant yellowing of leaves, which may be associated with a consequent decrease in the capacity for photosynthesis (Woolhouse 1986). Present study examines the impact of road side air pollution on moisture content, pH, chlorophyll, carotenoids and ascorbic acid in the leaves of selected tree species.

2 Materials and methods

2.1 Study area

Haridwar is one of the most important holy cities of India, located in newly carved state of Uttaranchal. Haridwar lies between latitude 29°58′ in the north to longitude 78°13′ in the east and has subtropical climate. The highest temperature recorded during this study was 40 ± 2.0°C during summer season, whereas lowest temperature recorded was 4 ± 1.0°C during winter. The average annual rainfall recorded was 960 mm, mostly occurring during the monsoon season. Total area of district Haridwar is 2,360 km^2 with a population of 14, 44,213 (according to 2001 census). It receives millions of tourists every month, sometimes just in one day, which increases the number of automobiles of various categories up to 800% per day. The total numbers of vehicles passing through the study area (8-h duration) were counted on each sampling date. Vehicles count helped in understanding the vehicular load on the study area.

2.2 Sampling sites and sampling procedure

Sites selected for sampling were situated on the national highway number 58, which bears high traffic load throughout the day; polluted site was just on the edge of the road, while the control site was 100 m away from the edge of the road. Dominant tree species selected for this study included *Mangifera indiaca*, *Eucalyptus citriodora*, *Tectona grandis* and *Shorea robusta*.

Leaf samples of selected trees were collected fortnightly during the study period from polluted and control site. These were weighed in a single pan electric balance (0.01 mg accuracy) for measuring the dust content and then thoroughly washed with double distilled water for further analysis. Commonly two types of effects were recorded on the selected plant species i.e. visible effects, which were visible with naked eyes and the biochemical effects.

Samples of air pollutants were collected fortnightly with the help of Respiratory Dust Sampler (APM-460) from each site. The apparatus was kept at a height of 2 m from the surface of the ground.

2.3 Analytical methods

The chlorophyll *a*, *b* and total chlorophyll and carotenoids in the leaves of selected plant species were determined as per Arnon (1949). Ascorbic acid was determined

by the method used by Sadasivam and Manikam (1991). Six samples of each of the tree species were analyzed on each sampling date. Similarly replicate samples and analytical blanks were also analyzed to check the reliability of data. Air pollution tolerance index (APTI) was estimated using the method of Singh and Rao (1983).

Air quality monitoring of gaseous pollutants viz., SO_2 and NO_x was carried out using the method of West and Gaeke (1956) and Jacob and Hochheiser (1958) respectively.

All the data obtained were further analyzed by using one-way ANOVA.

3 Results

The average density of vehicles flowing through the study area was noted as 1,816 vehicles per hour (average of 8 h).

Seasonal variation in the concentration of primary air pollutants has been given in Table 1.

Concentration of RSPM was highest (133.00 μgm^{-3}) during the summer season (April–June) whereas the standard limits as prescribed by Central Pollution Control Board of India is 150, 100, and 75 μgm^{-3} for industrial, residential and sensitive areas, respectively. The SPM was highest during winter (390.85 μgm^{-3}) followed by summer (363.00 μgm^{-3}) at polluted site.

The highest concentration (7.90 μgm^{-3}) of SO_2 was recorded during summer at polluted site, which was 41.89% higher as compared to control site (4.59 μgm^{-3}). Similarly the concentration of NO_x (18.56 μgm^{-3}) was 42.07% higher during summer as compared to concentration of NO_x at control site (10.75 μgm^{-3}).

3.1 Visible effects

The selected plant species showed some of the common visible effects due to auto exhaust pollution. Most frequent effects noted were necrosis and chlorosis. Necrosis started form development of small irregular shaped grayish green areas with water soaked appearances which was due to collapse of mesophyll cells. These areas subsequently became dry to form brownish-red to black colored spots, fruits also developed necrotic patches due to air pollutants. Chlorosis was also noted in the leaves of trees which were exposed to road side pollution.

3.2 Physiological effects

Variations in various physiological characteristics of the selected plant species exposed to automobile emissions are given in Table 2.

3.2.1 Chlorophyll pigments

Chlorophyll *a* and *b* contents in the leaf samples of *M. indica* were reported as 3.04 ± 0.04 and 1.45 ± 0.95 mg gm^{-1} at control site whereas these were 2.50 ± 0.89 and 2.15 ± 0.28 mg gm^{-1} respectively, at polluted sites. A decrease of 17.76% in

Table 1 Primary air pollutants recorded from control and polluted sites (average of 24 readings) during the study period

S.No.	SITE	RSPM (μgm^{-3})			SPM (μgm^{-3})			NO_x (μgm^{-3})			SO_x (μgm^{-3})		
		Winter	Monsoon	Summer	Winter	Monsoon	Summer	Winter	Monsoon	Summer	Winter	Monsoon	Summer
1	Polluted	120.00	122.50	133.00	390.85	350.22	363.12	12.18	10.22	18.56	6.42	5.00	7.90
2	Control	79.00	90.12	77.15	288.48	215.00	297.45	10.16	9.45	10.75	4.00	2.23	4.59
3	%	34.16	26.43	41.99	26.19	38.61	18.08	16.58	7.53	42.07	37.69	55.4	41.89

Where: RSPM = Respirable particulate matter

SPM = Suspended particulate matter

Table 2 Variation in the physiological characteristics of Tree species due to auto exhaust emission

Parameters	*Mangifera indica*			*Eucalyptus citriodora*			*Tectona grandis*			*Shorea robusta*		
	Polluted	Control	%	Polluted	Control	%	Polluted	Control	%	Polluted	Control	%
Chlorophyll *a* (mg/g) ($p < 0.01\%$)	$2.50 \pm 0.89^{**}$	3.04 ± 0.04	17.76 (−)	1.75 ± 0.48^{ns}	1.83 ± 0.38	4.37 (−)	$1.49 \pm 0.30^{***}$	2.11 ± 0.67	29.38 (−)	$1.95 \pm 0.52^{**}$	2.46 ± 1.46	20.73 (−)
Chlorophyll *b* (mg/g) ns	2.15 ± 0.28^{ns}	1.45 ± 0.95	32.55 (+)	0.80 ± 0.27^{ns}	0.83 ± 0.29	3.61 (−)	$0.83 \pm 0.28^{***}$	1.07 ± 0.49	22.42 (−)	$0.94 \pm 0.79^{***}$	1.15 ± 0.67	18.26 (−)
Total Chlorophyll (mg/g) ($p < 0.01\%$)	4.65 ± 0.91^{ns}	4.49 ± 0.41	3.44 (+)	2.55 ± 0.57^{ns}	2.66 ± 0.65	4.13 (−)	$2.32 \pm 0.75^{**}$	3.18 ± 0.22	27.04 (−)	$2.89 \pm 0.22^{**}$	3.61 ± 1.00	19.94 (−)
Carotenoids (mg/g) ns	2.42 ± 0.60^{ns}	2.97 ± 0.41	18.51 (−)	1.34 ± 0.43^{ns}	1.86 ± 0.56	27.95 (−)	1.66 ± 0.50^{ns}	2.00 ± 0.55	17.00 (−)	2.52 ± 0.27^{ns}	2.65 ± 1.62	4.90 (−)
Ascorbic acid (mg/100 g) ($p < 0.05\%$)	1.66 ± 0.40^{ns}	2.04 ± 0.53	18.62 (−)	1.63 ± 0.25^{ns}	1.67 ± 0.37	2.39 (−)	1.93 ± 0.60^{ns}	2.18 ± 0.56	11.46 (−)	1.66 ± 0.64^{ns}	2.28 ± 0.89	27.19 (−)
Relative moisture content (% $p < 0.01\%$)	$50.01 \pm 0.46^{***}$	41.83 ± 0.54	16.35 (+)	$60.11 \pm 0.23^{**}$	63.22 ± 0.56	4.91 (−)	$58.11 \pm 0.08^{***}$	64.47 ± 0.57	9.86 (−)	75.91 ± 0.67^{ns}	75.66 ± 0.21	0.32 (+)
Dust deposited (mg/leaf) $p < 0.05\%$	0.19 ± 0.05^{ns}	0.038 ± 0.02	80.00 (+)	0.58 ± 0.47^{ns}	0.54 ± 0.40	6.89 (+)	0.86 ± 0.80^{ns}	0.54 ± 0.68	37.20 (+)	$1.94 \pm 0.36^{*}$	0.04 ± 0.03	97.93 (+)
(pH $P < 0.05$)	$5.98 \pm 0.49^{*}$	6.45 ± 0.36	7.28 (−)	5.52 ± 0.68^{ns}	5.76 ± 0.75	4.16 (−)	$5.94 \pm 0.69^{*}$	6.42 ± 0.24	7.47 (−)	$5.76 \pm 0.53^{*}$	6.30 ± 0.23	8.57 (−)
Air pollution tolerance index	6.76 ± 0.40			7.32 ± 0.11			7.40 ± 0.59			9.02 ± 0.20		

Where: % (+) value indicates that value of polluted site is higher than control site, and % (−) values indicates that value of control site is higher than polluted site

*, **, *** indicate least significant difference at 0.05%, 0.01% and 0.001% level

($p < 0.01$) and ($p < 0.05$) are ANOVA values, ns = non significant

chlorophyll *a* in comparison to control, while an increase of 32.55% in chlorophyll *b* was recorded at polluted site in comparison to control site. Total chlorophyll content recorded for *Mangifera indica* was 4.49 ± 0.41 mg gm^{-1}, and it was 4.65 ± 0.91 mg gm^{-1} at control and polluted sites, respectively.

Chlorophyll *a* and *b* contents of *Eucalyptus citriodora* were reported as 1.83 ± 0.38 and 0.83 ± 0.29 mg gm^{-1} at control site and 1.75 ± 0.48 and 0.80 ± 0.27 mg gm^{-1} at polluted sites, respectively. A decrease of 4.37 and 3.61% was thus recorded in chlorophyll *a* and *b* contents, respectively. Total chlorophyll content recorded at polluted and control sites was 2.55 ± 0.57 and 2.66 ± 0.65 mg gm^{-1}, respectively. There was a reduction of 4.13% in the total chlorophyll content in the leaf samples from the polluted site as compared to control.

Chlorophyll *a* and *b* content of *T. grandis* were reported as 2.11 ± 0.67 and 1.07 ± 0.49 mg gm^{-1} at control and 1.49 ± 0.30 and 0.83 ± 0.28 mg gm^{-1} at polluted sites, respectively. A decrease of 29.38 and 22.42% was recorded in chlorophyll *a* and *b* respectively. Total chlorophyll content recorded at polluted site for *T. grandis* was 2.32 ± 0.75 mg gm^{-1} and it was 3.18 ± 0.22 mg gm^{-1} at control site, a reduction of 27.04% in the total chlorophyll content of plant samples from polluted site as compared to control site was recorded.

Chlorophyll *a* and *b* contents of *S. robusta* were reported as 2.46 ± 1.46 and 1.15 ± 0.67 mg gm^{-1} at control site and 1.95 ± 0.52 and 0.0.94 ± 0.79 mg gm^{-1} at polluted sites, respectively, with a decrease of 20.73 and 18.26% in chlorophyll *a* and *b*, respectively. Total chlorophyll content recorded at polluted site was 2.89 ± 0.22 mg gm^{-1} which was 3.61 ± 1.00 mg gm^{-1} at control site, thus in this case there was a reduction of 19.94% at the polluted site as compared to control.

3.2.2 Carotenoids and ascorbic acid

Carotenoids contents of *M. indica* recorded at control and polluted sites were 2.97 ± 0.41 and 2.42 ± 0.60 mg gm^{-1} respectively. A decrease of 18.51% was thus recorded in the amount of caratenoids in the polluted site as compared to control. Ascorbic acid content recorded was 2.04 ± 0.53 and 1.66 ± 0.40 mg per 100 gm at control and polluted sites respectively. In this case there was a reduction of 18.62% in ascorbic acid content of samples collected from polluted site as compared to control.

Carotenoids contents of *E. citriodora* recorded at control and polluted sites were 1.86 ± 0.56 and 1.34 ± 0.43 mg gm^{-1}, respectively. A decrease of 27.95% was thus recorded in the amount of caratenoids in the polluted site. Ascorbic acid content recorded was 1.67 ± 0.37 and 1.63 ± 0.25 mg per 100 gm at control and polluted sites respectively. In this case there was a reduction of 2.39% in ascorbic acid content of samples collected from polluted site as compared to control.

Carotenoids content of *T. grandis* recorded at control site were 2.00 ± 0.55 and 1.66 ± 0.50 mg gm^{-1} at polluted site. A decrease of 17.00% was thus recorded in the amount of caratenoids in the polluted site. Ascorbic acid content recorded was 2.18 ± 0.56 and 1.93 ± 0.60 mg per 100 gm at control and polluted sites, respectively. In this case there was a reduction of 11.46% in ascorbic acid content of samples collected from polluted site.

In *S. robusta*, carotenoids content recorded at control and polluted sites were 2.65 ± 1.62 and 2.52 ± 0.27 mg gm^{-1}, respectively. A decrease of 4.90% was thus recorded in the amount of caratenoids in the polluted site. Ascorbic acid content recorded was 2.28 ± 0.89 and 1.66 ± 0.64 mg per 100 gm at control and polluted sites respectively. In this case there was a reduction of 27.19% in ascorbic acid content of samples collected from polluted site as compared to control.

3.2.3 Relative moisture content, dust content, pH and APTI

Relative moisture content of *M. indica* was higher by 16.35% in the leaf samples collected from polluted site. Dust deposited on leaves was also higher (0.19 ± 0.05 mg per leaf) in polluted site in comparison to control site (0.038 ± 0.02 mg per leaf). pH of leaf samples from polluted site was recorded as 5.98 ± 0.49, where as it was 6.45 ± 0.36 for the samples from control site . Thus a change of 7.28% towards the acidic side was recorded in the samples collected from polluted site. The air pollution tolerance index (APTI) of *M. indica* was determined as 6.76 ± 0.40 at polluted site.

Relative moisture recorded for *E. citriodora* at control site was 63.22 ± 0.56% whereas it was 60.11 ± 0.23% at polluted site. Thus there was a reduction of 4.91% in relative moisture content of samples collected from polluted. Amount of dust deposited on leaves was higher in polluted site in comparison to control site. pH of leaves collected from polluted site was recorded as 5.52 ± 0.68 , for control site it was recorded as 5.76 ± 0.75. Thus a change of 4.16% toward the acidic side was recorded in the samples collected from polluted site. Air pollution tolerance index of *E. citriodora* was determined as 7.32 ± 0.11 at polluted site.

Relative moisture content of *T. grandis* leaves collected from control site was 64.47 ± 0.57%, while it was 58.11 ± 0.08% at polluted site. Thus there was a reduction of 9.86% in relative moisture content of samples collected from polluted site as compared to control. Similarly amount of dust deposited on leaves was higher in polluted site. pH of leaves from polluted site was recorded as 5.94 ± 0.69, where as pH of leaves sampled form control site was recorded as 6.42 ± 0.24. Thus a change of 7.47% towards the acidic side was recorded in the samples collected from polluted site. Air pollution tolerance index of *E. citriodora* was determined as 7.40 ± 0.59 at polluted site.

Relative moisture content and amount of dust deposited in the leaves of *S. robusta* collected from polluted site was higher as compared to samples from control site. pH of leaf samples from polluted and control sites was recorded as 5.76 ± 0.53 and 6.30 ± 0.23, respectively with a change of 8.57% towards the acidic in the samples collected from polluted site. Air pollution tolerance index of *S. robusta* was determined as 9.02 ± 0.20 at polluted site.

4 Discussion

The chlorosis in plants studied was due to the effect of Sulphur dioxide, which enters the leaves through stomata attacking the spongy parenchymatus tissues and

then spreads to the plastid layer. Since the cells near the vein are rarely affected, damaged leaves often show a network of green veins on discolored background.

Highest decrease in total chlorophyll content of the samples from polluted site in comparison to control was recorded in *T. grandis* (27.04%) whereas lowest was for *M. indiaca* (3.44%). The analysis of data using one-way ANOVA shows that the reduction in chlorophyll contents of *T. grandis* and *S. roubsta* were significant at 0.01% level. Reduction of chlorophyll may be due to the increase of chlorophyllase enzyme activities, which in turn affects the chlorophyll concentration in plants (Mandal and Mukherji 2000). SO_2 plays an important role in the reduction of chlorophyll content (Mandloi and Dubey 1988; Rao and Dubey 1985). Rao and Leblance (1966) have also reported reduction of chlorophyll content brought by acidic pollutants like SO_2 which causes phaeophythin formation by acidification of chlorophyll. Reductions in chlorophyll contents of a variety of crop plants due to SO_2 and O_3 exposure have also been reported by Agrawal (1985). Chlorophyll contents are essentials for the photosynthetic activity and reduction in chlorophyll content has been used as an indicator of air pollution (Pawar and Dubey 1985).

SO_2 get dissolved in water in the leaf tissues, and causes local injury by germinating toxic ions (Malhotra and Hocking 1976; Manninen et al. 1996). These injuries further retard net assimilation rate of the plants (Weinstein and McCune 1970).

Highest decrease in carotenoid contents was reported for *E. citriodora* among all the plant species studied in the present study, whereas lowest decrease of carotenoid content was recorded in *S. robusta*. Carotanoids protect chlorophyll from photoxidative destruction. (Siefermann-Harms 1987). However, one-way ANOVA shows that caratenoids pigments in all the species were not significantly different at the polluted site as compared to the control.

Ascorbic acid, a natural antioxidant in plants has been shown to play an important role in pollution tolerance (Chen et al. 1990). Present study shows the decline in ascorbic acid content in all four plant species exposed to auto exhaust. Maximum decrease in ascorbic acid was noted in *S. robusta* and minimum in *E. citriodora.* However, no significant change was recorded in all the four selected species.

The change in pH in the leaf samples collected from polluted site was significant at 0.05% level in the plant species *M. Indica, T. grandis* and *S. robusta*.

Highest changes in dust content were found in *S. robusta* which was significant at 0.05% level. High traffic load contributes high dust fall on the plant leaves this also depends upon the condition of roads (Lone et al. 2005) and size and structure of leaves. The change in the value of relative water contents recorded for *M. indica* and *T. grandis* were significant at 0.001 and 0.01% level, respectively.

Air pollution tolerant index is an index, which determines capability of a plant to combat against air pollution. Plants which have higher index value are tolerant to air pollution and can be used as sink to mitigate pollution, while plants having low index value show less tolerance and can be used to indicate levels of air pollution (Sing and Rao 1983). In the present study higher APTI values was recorded for *S. robust* (9.02) and lowest for *M. indica* (6.76).

It may thus be concluded that the plant species, which are grown along the road sides and are economically important, may act as an absorbent of the various pollutants and their analysis may give the level of pollutants in any particular area of interest Similarly, species with higher APTI may be planted in the areas with higher concentration of air pollutants.

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