



## Effects of Industrial Air pollution on Biochemical parameters of *Shorea robusta* and *Acacia auriculiformis*

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### Abstract

Air pollution has become a great concern to the environment due to increasing industrialization and urbanization. The present study examines the effects of air pollution on various biochemical parameters of plants in Barjora forest at Barjora, Bankura, West Bengal. To evaluate the susceptibility of plants to air pollution, four biochemical parameters, namely; chlorophyll, carbohydrate, ascorbic acid and protein, in two different tree species viz., *Shorea robusta* and *Acacia auriculiformis*, were determined. For both the plant species, the chlorophyll content was decreased significantly as compared to the control. The carbohydrate and protein content were also decreased for both the species, whereas, ascorbic acid content was increased in both the species.

**Keywords:** Air pollution, *Shorea robusta*, *Acacia auriculiformis*, Chlorophyll, Carbohydrate, Protein, Ascorbic acid.

### Introduction

In the recent years, various types of anthropogenic activities, such as industrialization, urbanization, increased number of vehicles become a great concern to the whole ecosystem. In urban areas various types of pollutants such as SO<sub>x</sub>, NO<sub>x</sub>, ozone (O<sub>3</sub>), carbon monoxide (CO), volatile organic compounds (VOC<sub>s</sub>), SPM, RSPM are released due to these anthropogenic activities, which are causing an adverse effect on plants.

Trees have a very important role in combating the levels of air pollutants in the atmosphere by taking up gaseous pollutants and particulates and improving the ambient air quality. Leaves are the very good indicator of pollution. Primarily, most of the particulate matters are deposited on the upper surface of the leaves. Due to their deposition, the reduction has been measured in photosynthetic pigments like, chlorophyll, carotenoids. So, the total productivity of the plants gets decreased. Before showing any physiological changes, various visible leaf damages occurred due to the absorption of pollutants<sup>1</sup>. From the previous studies it has been seen that the contents of chlorophyll, ascorbic acid, protein and carbohydrate are affected due to the air pollution<sup>2,3</sup>. It was found that the effects of gaseous as well as particulate pollutants were much more in woodlands than in shorter vegetations<sup>4,5</sup>. Biochemical studies of foliar tissues showed decrease in chlorophyll content and the increase in ascorbic acid content as a result of air pollution<sup>6-9</sup>. Reduction of ascorbic acid, protein, carbohydrate and pigments in *Phaseolus aureus* was observed by Prasad and Rao in petroleum-coke treated plant<sup>10</sup>.

In the present study an attempt has been made to find out the effects of gaseous and particulate air pollutants on the

biochemical parameters of *Shorea robusta* and *Acacia auriculiformis*.

### Materials and Methods

**Study area:** In the present study two different study areas were selected. The first being Barjora forest at Barjora, Bankura, situated in a highly polluted region due to the presence of various small, medium and large scale industries, as well as two operating collieries and thereby releasing different pollutants. And the second site, Ballavpur Wildlife Sanctuary (BWLS) situated at Santiniketan, under Bolpur Range and relatively free from pollution will serve as control site.

**Selection of plants:** Among the various trees in these two forests, two dominant tree species, namely, Sal (*Shorea robusta*), and Sonajhuri (*Acacia auriculiformis*) were selected for measurement of different biochemical parameters.

**Collection of leaf sample:** Five trees of each species were selected from both of the sites. Samplings of leaves were undertaken by collecting the fifth leaf (from the apical bud) in each small lower branch and were taken for the measurement of different biochemical parameters of foliar tissues. The freshly collected leaves were kept in zipper pack and then preserved in an ice can and then brought to the laboratory for analyzing various biochemical parameters.

**Measurement of biochemical parameters: Estimation of chlorophyll content:** Chlorophyll content of the leaves was measured by the method of Arnon<sup>11</sup>. At first 1g leaf sample was homogenized with 20 ml 80% acetone and centrifuged at 5,000rpm for 5 min. Then the supernatant was taken and the

volume was made up to 100ml with 80% acetone. The absorbance of the solution was recorded at 645 and 663 nm in spectrophotometer against 80% acetone as blank. The amount of chlorophyll present in the leaf extract, in mg chlorophyll per gm tissue, was calculated by using the formulae given by Arnon<sup>11</sup>.

**Estimation of carbohydrate content:** The total carbohydrate was determined by anthrone method following by Hedge and Hofreiter<sup>12</sup>. Firstly 100mg of the sample was taken into boiling tube and hydrolysed in boiling water bath for 3 hours with 5ml of 2.5N HCl and then was cooled at room temperature. To neutralize the extraction sodium carbonate was used until the effervescence ceases and volume was made up to 100 ml and centrifuged. 0.5 ml of supernatant was taken and volume was made up to 1 ml. Then 4 ml of anthrone reagent was added to it and was heated for 8 minutes in boiling water bath. Then was cooled rapidly and the absorbance was determined at 630 nm against the blank.

**Estimation of ascorbic acid:** Ascorbic acid content of the leaves was measured by using the method of Harris and Ray<sup>13</sup>. At first, 0.5 g sample was extracted with 4% oxalic acid and the volume was made up to 100ml with 4% oxalic acid and centrifuged. Then 5ml of supernatant was taken and 10ml 4% oxalic acid was added to it and titrated against the dye (V<sub>2</sub> ml). The amount of ascorbic acid was measured against the standard solution.

**Estimation of Protein content:** The protein content of leaves was measured by the method of Lowry et al.<sup>14</sup> by using bovine serum albumin (BSA) as a standard. Firstly 0.5g of leaf samples were homogenized with pre-cooled phosphate buffer (pH-7) and centrifuged at 10000 rpm for 15 min. The supernatants were used as a crude extract for protein assay by using 0.2ml of the extract. In case of blank, 0.2ml of distilled water was added instead of tissue extract. 2ml of alkaline solution (2% Na<sub>2</sub>CO<sub>3</sub> + 1% NaOH + 1.56% CuSO<sub>4</sub> + 2.37% NaK tartareate) was added to each test tube and was mixed well and was allowed to stand for 10 min at room temperature. After 10 min, 0.2ml 1N folin was added to each test tube and mixed well. After 30 min, the absorbance was recorded at 660 nm against the blank. The total protein content of each foliar tissue was calculated from the standard curve of BSA (1mg/ml) and was expressed as g protein/100g of leaf tissue weight.

**Statistical analyses:** The data were subjected to one-way analysis of variance (ANOVA) of variance at 95% confidence level, using Microsoft excel, 2013.

## Results and Discussion

**Chlorophyll content:** In the present study, the mean chlorophyll content in leaf of two species significantly decreases by 63.9% and 39.3% as compared to the control of Sal (*Shorea robusta*) and Sonajhuri (*Acacia auriculiformis*) respectively (Table-1, Figure-1). Chlorophyll content of foliar tissues varies

from species to species and also depends on the leaf age, level of pollution and also the biotic and abiotic condition<sup>15</sup>. For photosynthetic activity, Chlorophyll content is highly essential for plants and the reduction of chlorophyll content is an indication of air pollution<sup>16,17</sup>. From the various previous studies it has been shown that the chlorophyll content of leaves varies with the degree of pollution. In another study, the chlorophyll content was high in *P. juliflora* leaves, which was exposed to pollution. The particulate matter deposited on the surface of the leaf and might have led to decrease in the level of total chlorophyll content of leaves<sup>18</sup>.

**Carbohydrate content:** The mean carbohydrate contents in leaf of two species of plants studied in this case were decreased by 17.46% and 14.45% as compared to the control of *Shorea robusta* and *Acacia auriculiformis* respectively (Table-1, Figure-1). Carbohydrate is the important part in plant structure and all organisms they are the source of energy. The carbohydrate concentrations, indicating the various physiological activities of a plant and it can also determine the plant sensitivity to air pollution<sup>19</sup>. A study carried out by Thambavani et al. showed that the concentration of total carbohydrate was significantly decreased in foliar tissues from the polluted site as compared to the control site<sup>20</sup>. The possible causes behind it may be the deterioration of the chlorophyll content of the leaf so that the respiration rate has increased and the CO<sub>2</sub> fixation has decreased<sup>20</sup>. In another report the increased level of carbohydrate in polluted site has been found. The total carbohydrate content in the plant has increased in the polluted site is an indication that the plant has become tolerant and resistant to that environmental condition<sup>21</sup>.

**Ascorbic acid content:** Ascorbic acid is an antioxidant, which is present in huge amount in all the growing plant parts and control resistance of the plant to adverse environmental condition<sup>22,23</sup>. In the present study, the mean ascorbic acid contents in leaf of two species, *Shorea robusta* and *Acacia auriculiformis* were found to be increased by 88.24% and 92.31% respectively as compared to the control (Table-1, Figure-1). Ascorbic acid is a stress-reducing factor, which is present in tolerant plant species generally in higher levels<sup>24</sup>. It has been reported that in the polluted condition, the ascorbic acid content of all the plant species increases, which might be due to increasing the production rate of reactive oxygen species (ROS) during the photo-oxidation process<sup>19</sup>. There is another opinion of the increased level of ascorbic acid is an indicator of the tolerance level of the plants against the sulfur dioxide pollution<sup>25</sup>.

**Protein content:** Plant protein is an essential component for the plant growth and development<sup>20</sup>. In the present study, the mean protein content in leaf of two species, *Shorea robusta* and *Acacia auriculiformis* decreased by 5.0% and 20.08% respectively as compared to the control (Table-1, Figure-1). The similar result was also was also reported in the leaves of *Madhuca indica* and *Shorea robusta* around stone crushing

industry in the Lalpahari forest area and a significant reduction in the protein content in foliar tissue as compared to the control site was observed<sup>26</sup>.

The probable reason behind the reduction of protein content in the polluted plant might be the enhanced rate of protein denaturation and also the breakdown of existing protein to amino acid<sup>20</sup>. Another reason may be an enhance in the activity of the degradative enzyme like proteases which catalyses the breakdown of polypeptides into amino acids to resist the stress induced by pollution<sup>27</sup>.

## Conclusion

On the basis of the study, it could be concluded that the various biochemical parameters like chlorophyll, carbohydrate, ascorbic acid and protein of the two tree species, *Shorea robusta* (Sal)

and *Acacia auriculiformis* (Sonajhuri) were found to be highly affected. Increasing and decreasing levels of various plant parameters at selected sites can be considered as an adaptation of the plant that environmental condition to protect plants against air pollution stress. Chlorophyll content, protein and carbohydrate content of both species were decreased compared to the control. On the other hand the ascorbic acid contents of the both plants were increased compared to the control, indicating the tolerance level of the plant species against the various air pollutants. The result also indicated that the Sonajhuri was more tolerant than Sal.

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**Table-1**  
**Effects of air pollution on biochemical parameters of two plant species (*Shorea robusta* and *Acacia auriculiformis*)**

Name of the plant	Parameters	Mean $\pm$ SD (Control)	Mean $\pm$ SD (Industrial)
<i>Shorea robusta</i>	Ch-a	1.76 $\pm$ 0.30	0.64 $\pm$ 0.03
	Ch-b	0.65 $\pm$ 0.11	0.24 $\pm$ 0.04
	Ch-T	2.41 $\pm$ 0.41*	0.87 $\pm$ 0.07*
	Carbohydrate( $\mu$ g/100mg)	81.92 $\pm$ 8.50	67.62 $\pm$ 4.4
	AA	0.17 $\pm$ 0.05	0.32 $\pm$ 0.08
	Protein	33.80 $\pm$ 1.70	32.11 $\pm$ 0.87
<i>Acacia auriculiformis</i>	Ch-a	1.02 $\pm$ 0.11	0.63 $\pm$ 0.25
	Ch-b	0.33 $\pm$ 0.03	0.20 $\pm$ 0.07
	Ch-T	1.36 $\pm$ 0.14	0.83 $\pm$ 0.32
	Carbohydrate( $\mu$ g/100mg)	91.32 $\pm$ 11.7	78.12 $\pm$ 12.3
	AA	0.13 $\pm$ 0.03	0.25 $\pm$ 0.05
	Protein	38.34 $\pm$ 1.19	30.64 $\pm$ 0.49

SD: standard deviation, \*indicate that there is a significant difference (p<0.05)

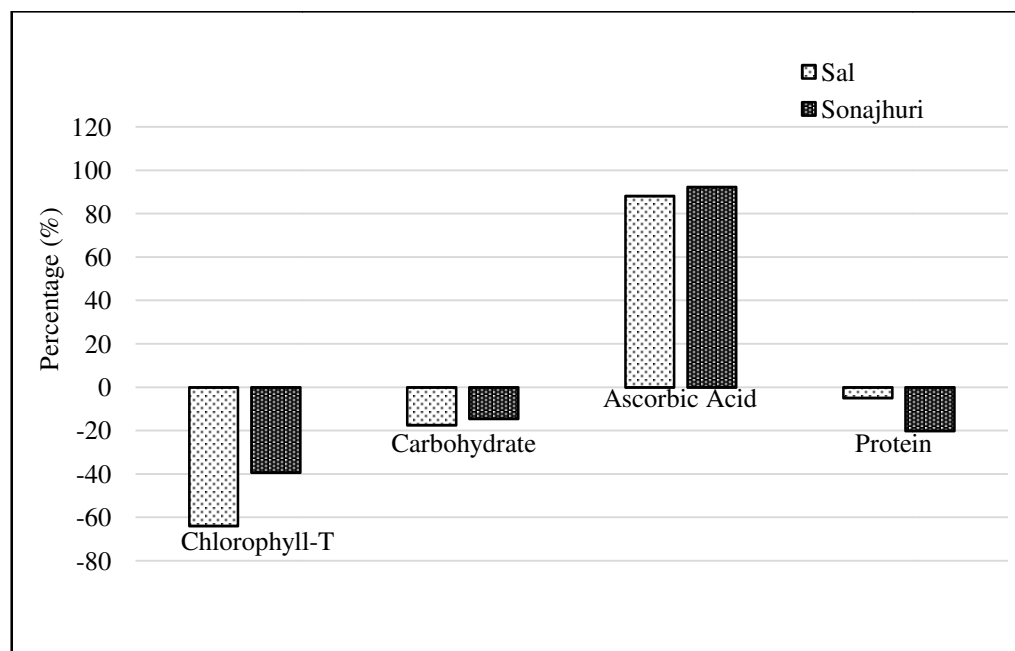


Figure-1

Percentage of increase or decrease of some biochemical parameters of *Shorea robusta* and *Acacia auriculiformis* due to air pollution

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