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RESEARCH ARTICLE

BIOMONITORING OF AIR QUALITY IN KOLKATA CITY WITH GALL OF (*Alstoniascholaris*)

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ABSTRACT

Air pollution occurs when harmful or excessive quantities of substances including gases, particulates, and biological molecules are introduced into Earth's atmosphere. The concept of monitoring of air quality by plants is a well-established fact. It is known that some plants are very sensitive to air pollutants; they are thus used as indicator species for bio monitoring of air quality. The use of lichens and moss for air pollution level mapping in urban and industrial area are the finest examples of plant bio monitoring of air quality. The use of higher plants for monitoring of air pollution is, however, a recent development. A number of plant parameters either simply or in combination may be used for evaluating the pollution stress. Monitoring and detection by instrument cannot be possible everywhere however, indication from plant species by their alterations in morphological and biochemical parameters may be a suitable and easy screening measurement. The present study aims to detect morphological and biochemical changes in leaves of *Alstonia scholaris* found in Kolkata city area that are exposed to vehicular emission. The results clearly indicate that vehicular load brought significant changes in foliar morphology, numbers of gall formation and biochemical parameters such as pigment content, enzyme activities etc of leaves of *A. scholaris*. It is concluded that increased Air Pollution Index and gall formation in *A. scholaris* can be used as early monitoring parameters for air pollution by vehicular emission.

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INTRODUCTION

Air is a significant and crucial constituent of the environment and is essential for the survival, growth and development of all organisms. So even minimal changes in its composition may have negative effects on all living creatures and life forms. Nowadays release of toxic chemical compounds from nearby industries, automobile exhausts and even domestic furnaces adds to air pollution which not only exhibits devastating impact on human health but also on the large scale global environment. Due to activities such as emissions from the industries and several vehicles, changes in the concentration levels of gases occur causing respiratory diseases in human and a reduction in plant production altogether. So air pollution is a serious issue that needs to be discussed now in all forums as much as possible otherwise in the coming years the society may face a major threat. Different plants react to different concentrations of pollutants. Some might be susceptible while others may show minimal injury. Several plants when exposed to potential pollutants show typical damage in different parts (Claudia et al. 1999). These plants can therefore be used as biological indicators in that particular area.

Increasing attempts are being made by various researchers to use plants to detect the air pollution levels in different cities. Environmental changes can be recorded by observing alterations in the responses of surrounding plant and animal life forms i.e. biomonitoring. Importance of biomonitoring are

- Biomonitoring of plants is cost effective and environment friendly initiative that replaces physico-chemical methods of air pollution monitoring systems which is costly and time consuming.
- In comparison to physico-chemical methods which exhibit low concentrations of air pollutants, biological monitoring allows direct and better assessment of different concentrations of pollutants (Temmerman, Bell, Garrec, Klumpp, Krause, Tonneijck, 2001).
- Biomonitoring does not involve using advanced and high maintenance instruments and also ensures monitoring over a large area.
- Plants accumulate a handful of pollutants within them which can be easily examined with the process of biomonitoring.
- We can use the information collected by biological monitoring to evaluate the environmental impact on all living organisms.

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For several years now, lichens and moss are being used for detecting air quality levels in metropolitan cities and industrial area. Earlier researchers used lichens to detect air pollution in populated cities and towns. Nowadays, monitoring of air quality can also be done by using higher plants and avenue trees. In this study we used *Alstonia scholaris* (Family–Apocynaceae) as a model for biomonitoring. *A. scholaris* (popularly known as Devil tree) is a tall, elegant, evergreen plant found easily on sides of the road and is very common in the Indian subcontinent. The trees require minimal amount of water and thus can effortlessly thrive in polluted areas (Muhammad et al., 2014). The bark of *Alstonia scholaris* shows several medicinal advantages. The milky extract of the bark is used to treat ulcers, diseases like Malaria and epilepsy and also sores. The off-white flowers occur in clusters and give off a sweet fragrance during the blooming season. Gall formation caused by an insect *Pauropsylla tuberculata* Crawf. in the *A. scholaris* trees planted across the city of Kolkata, and also in other parts of the state is spreading at a faster rate. Galls occur either singly or in a group of 3-4 coupled together and are more abundant on the abaxial side of the leaf. Gall formation is induced when the insect deposits the eggs together with some stimulating characteristic fluid. This triggers overgrowth of the cells followed by proliferation of the tissues just beside the area where the eggs were laid. Plants that are sensitive to air pollutants exhibit higher rate of gall formation.

The present study attempts to know the morphological, anatomical, biochemical changes in leaves of *A. scholaris* found in road side with heavy vehicular load and correlation between gall formation and effect of air pollution on *A. scholaris*.

MATERIALS AND METHODS

The study areas were selected as per vehicular loads. The study was carried out at two sampling areas i) medicinal garden of Lady Brabourne College campus, Park Circus area. (low vehicular load) as control area (Sample A), ii) Golpark crossing, Southern avenue (high vehicular load) as experimental area (Sample B).

Morphological and Anatomical study of the leaves of (*Alstonia scholaris* -- few)

Morphological parameters have been taken into account

- Area of the leaf,
- Quantification of galls in the sample B leaf-- by determination of Air Pollution Index (API),
- Anatomy of stomata

Biochemical study of the leaves of (*Alstonia scholaris*)

Estimation of chlorophyll

Chlorophyll content of both the sample leaves were estimated according to Arnon (1949). The chlorophyll content was Determined by using the following formula

$$\text{mg chl a/g tissue} = 12.7 (A_{663}) - 2.69 (A_{645}) \times V / 1000 \times W$$

$$\text{mg chl b/g tissue} = 22.9 (A_{645}) - 4.68 (A_{663}) \times V / 1000 \times W$$

$$\text{mg total chl/g tissue} = 20.2 (A_{645}) + 8.02 (A_{663}) \times V / 1000 \times W$$

Where A= absorbance at specific wavelengths, V= final volume of chlorophyll extract in 80% acetone, W= fresh weight of tissue extracted.

Estimation of carotenoid

Carotenoid content of both the sample leaves were estimated according to Kirk and Allen (1965). The carotene content was determined by using the following formula

$$\text{mg carotene/g tissue} = (1000 A_{480} - 1.82 \text{chl}_a - 85.02 \text{chl}_b) / 198$$

Estimation of proline

Proline content of both the sample leaves were estimated according to Bates *et al.* (1973). The proline content was determined from a standard curve prepared with proline and the results were expressed in milligram per gram dry weight.

Estimation of phenol

Phenol content of both the sample leaves were estimated according to Malick C.P. and Singh M.B. (1980). The phenol content was determined from a standard curve prepared with Folin-Ciocalteu reagent and the results were expressed in mg phenol/100g material.

Estimation of total carbohydrate

Total Carbohydrate content of both the sample leaves were estimated according to Hedge, J.E. and Hofreiter, B.T (1962). The total Carbohydrate content was determined from a standard curve prepared with glucose and the results were expressed in mg reducing sugar/gram fresh weight.

Estimation of peroxidase activity

Peroxidase content of both the sample leaves were estimated according to Faug and Kao (2000). The specific activity of the enzyme was calculated using the molar extinction coefficient, $\epsilon = 26.6 \text{mM}^{-1} \text{cm}^{-1}$ for tetraguaiacol and was expressed in $\mu\text{mol tetraguaiacol}/\text{min}/\text{mg protein}$.

$$\mu\text{M}/\text{min}/\text{gram fresh weight} = \Delta A = \epsilon CL$$

Where ϵ = Molar extinction coefficient = $26.6 \text{mM}^{-1} \text{cm}^{-1}$, C = Specific activity of enzyme,

L = Length of cuvette

Estimation of ascorbic acid

Ascorbic acid content of both the sample leaves were estimated according to Harris, L.J. and Ray, S.N. (1935). The ascorbic acid content was measured by the following formula

$$\text{Amount of ascorbic acid mg}/100 \text{ g sample}$$

$$= 0.5 \text{mg}/V_1 \text{ ml} \times V_2 / 5 \text{ ml} \times 100 \text{ ml}/\text{weight of the sample} \times 100$$

Estimation of air pollution tolerance index

APTI of both the sample leaves were estimated according to Singh and Rao (1983). APTI includes four biochemical parameters ascorbic acid, chlorophyll, leaf extract pH and relative water content in leaf samples.

Relative Water Content was determined by the formula

$$RWC = [(FW-DW)/(TW-DW)] \times 100.$$

Where: FW= Fresh weight, DW= Dry weight, TW= Turgid weight.

The air pollution tolerance indices for leaves of *Alstonia scholaris* (both Sample A and Sample B leaves) were determined by following method

The formula is given as: $APTI = [A (T+P) + R] / 10.$

Where: A=Ascorbic acid content (mg/gm), T=Total chlorophyll (mg/gm), P=pH of the leaf extract, R=Relative water content of leaf (%).

RESULTS AND DISCUSSION

Anatomical Study

Sl no	Parameter	Sample A	Sample B
1.	Area of leaf	29.72 cm	27.9 cm
2.	Air pollution Index	00	1.61
3.	Length of stomata	2.16 μm	2 μm
4.	Breadth of stomata	2.66 μm	2 μm
5.	Stomatal frequency	16355 per unit area	14356 per unit area.

Air Pollution Index (API) can be calculated by following formula:
 No. of galls per centimeter² x Whole leaf Length/Breadth ratio

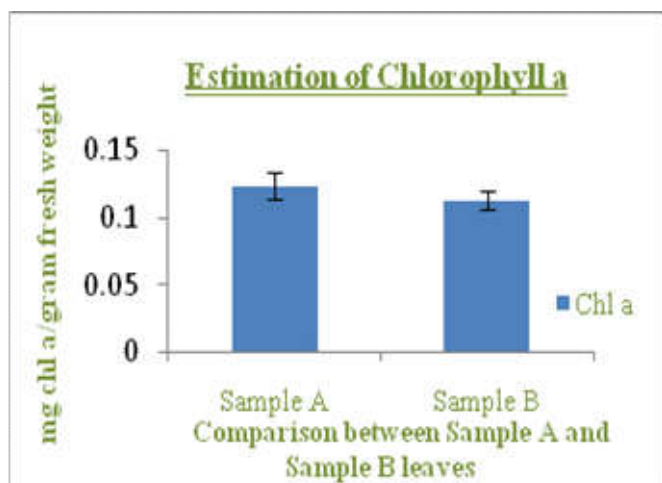
Biochemical study

Estimation of chlorophyll

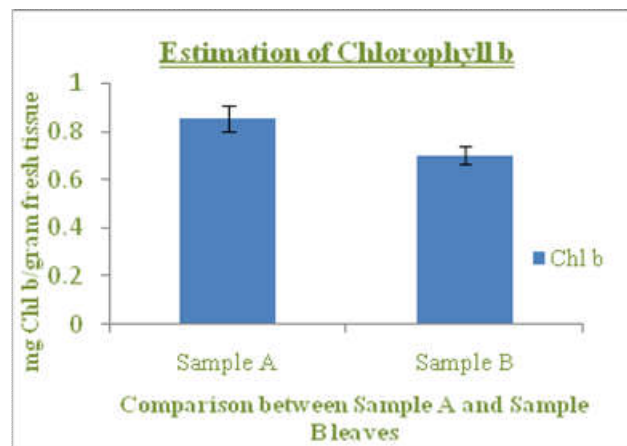
The readings from table 1 show that sample A leaves contain maximum Chlorophyll a, Chlorophyll b, Total Chlorophyll. On the other hand, the quantity of Chlorophyll a, Chlorophyll b and Total Chlorophyll declined in the aphid infested sample B leaves.

Table 1. Estimation of Chlorophyll a, Chlorophyll b and Total Chlorophyll in Sample A and B leaves

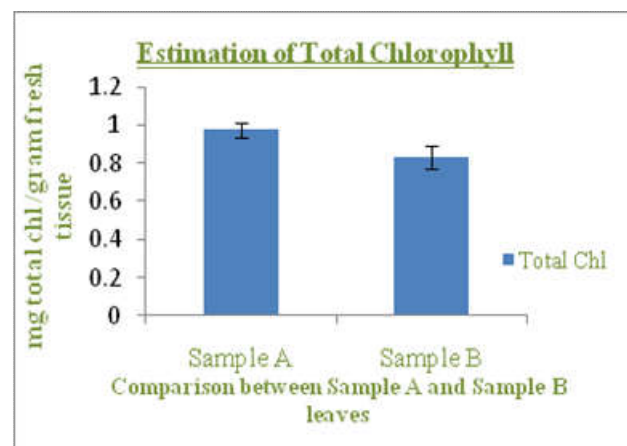
Parameters	Sample A Leaf (mg/g tissue)	Sample B Leaf (mg/g tissue)
Chlorophyll a	0.124 ± 0.01	0.113 ± 0.0072
Chlorophyll b	0.852 ± 0.055	0.701 ± 0.036
Total Chlorophyll	0.976 ± 0.039	0.831 ± 0.061



Graph 1. Graph showing the estimation of Chlorophyll a in leaves of Sample A and Sample B of *Alstonia scholaris*



Graph 2. Graph showing the estimation of Chlorophyll b in both Sample A and Sample B leaves of *Alstonia scholaris*



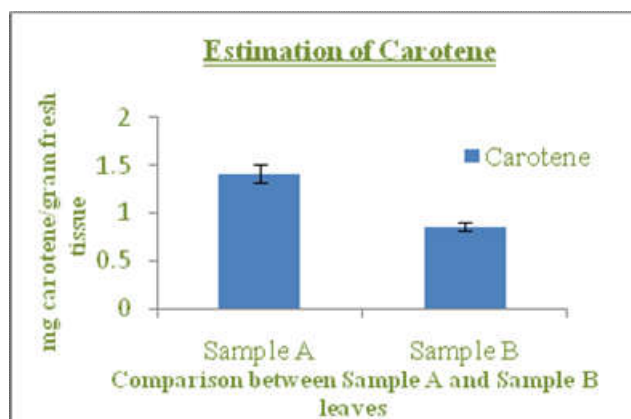
Graph 3. Graph showing the estimation of Total Chlorophyll in both Sample A and Sample B leaves of *Alstonia scholaris*

Estimation of carotene

The quantity of carotene is given below in Table 2. It is evident from the data from table 2 that Sample A leaves contain maximum carotene i.e. 1.42 mg/gram fresh tissue and Sample B leaves contain 0.86 mg/gram fresh tissue of carotene.

Table 2. Estimation of Carotene in Sample A and Sample B leaves

Parameter	Sample A leaf (mg/g tissue)	Sample B leaf (mg/g tissue)
Carotene	1.42 ± 0.098	0.86 ± 0.047



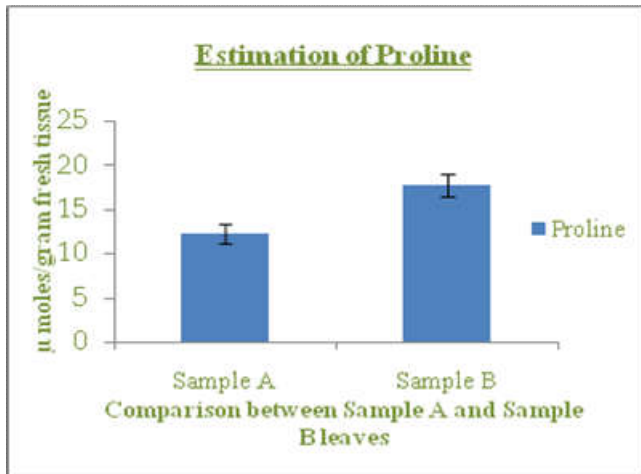
Graph 4. Graph showing the estimation of Carotene in both Sample A and Sample B leaves of *Alstonia scholaris*

Estimation of proline

The quantity of proline in Sample A and Sample B leaves are given below in Table 3. The readings above show proline content in the Sample B leaves are as high as 17.83 μ moles/gram tissue whereas in the Sample A leaves it is 12.29 μ moles/gram tissue which decreased.

Table 3. Estimation of proline in Sample A and Sample B leaves

Parameter	Sample A Leaf (μ moles/g tissue)	Sample B Leaf (μ moles/g tissue)
Proline	12.29 \pm 1.09	17.83 \pm 1.34



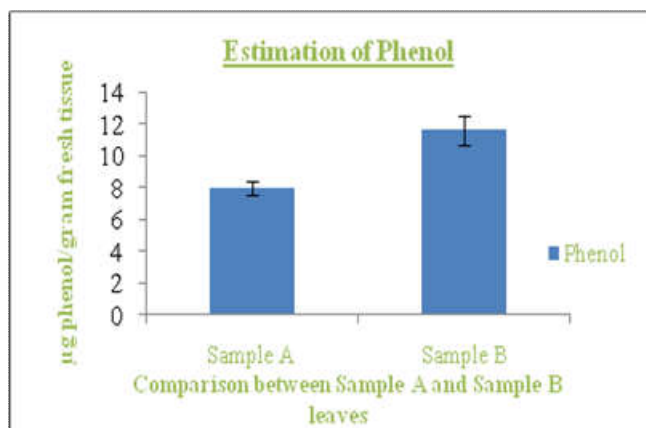
Graph 5. Graph showing the estimation of Proline in both Sample A and Sample B leaves of *Alstonia scholaris*

Estimation of phenol

The quantity of phenol in Sample A and Sample B leaves are given below in Table 4. The estimation of phenol was determined with Folin-Ciocalteu reagent. It is evident from the above data from table 4 that Sample A leaves have phenol content as low as 8 μ g phenol/gram fresh tissue. On the other hand, the quantity of phenol in Sample B leaves was 11.6 μ g phenol/gram fresh tissue i.e. the phenol content increased.

Table 4. Estimation of Phenol in Sample A and Sample B leaves

Parameter	Sample A leaf (μ g /g tissue)	Sample B leaf (μ g /g tissue)
Phenol	8 \pm 0.46	11.6 \pm 0.96



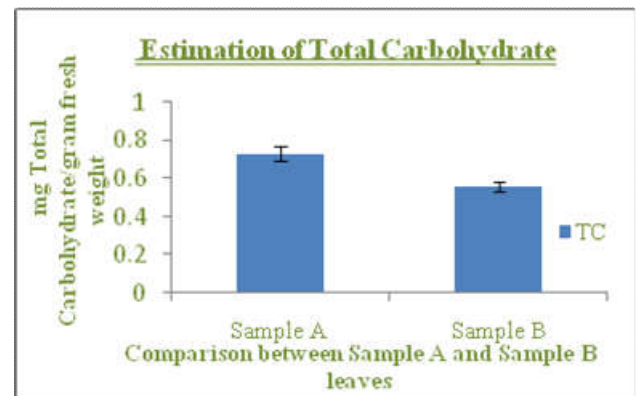
Graph 6. Graph showing the estimation of Phenol in both Sample A and Sample B leaves of *Alstonia scholaris*

Estimation of total carbohydrate

The quantity of Total Carbohydrate in Sample A and Sample B leaves are given below in Table 5. It is evident from the above data from table 5 that Sample A leaves have higher Total Carbohydrate content which is 0.732 mg/gram fresh tissue. On the other hand, the quantity of Total Carbohydrate in Sample B leaves was 0.56 mg/gram fresh tissue.

Table 5. Estimation of Total Carbohydrate in Sample A and Sample B leaves

Parameter	Sample A leaf (mg/g tissue)	Sample B leaf (mg/g tissue)
Total Carbohydrate	0.732 \pm 0.04	0.56 \pm 0.025



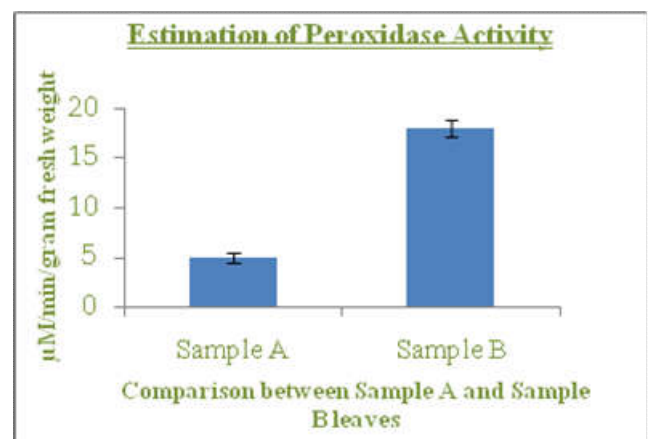
Graph 7. Graph showing the estimation of Total Carbohydrate in both Sample A and Sample B leaves of *Alstonia scholaris*

Estimation of peroxidase activity

The readings of Peroxidase activity in Sample A and Sample B leaves are given below in Table 6. The readings above in table 6 show Peroxidase activity in Sample B leaves are as high as 18 μ M/min/g fresh weight whereas the Sample A leaves show comparatively less peroxidase activity i.e. 5 μ M/min/g fresh weight.

Table 6. Estimation of Peroxidase in Sample A and Sample B leaves

Parameter	Sample A leaf (μ M/min/g tissue)	Sample B leaf (μ M/min/g tissue)
Peroxidase activity	5 \pm 0.5	18 \pm 0.9



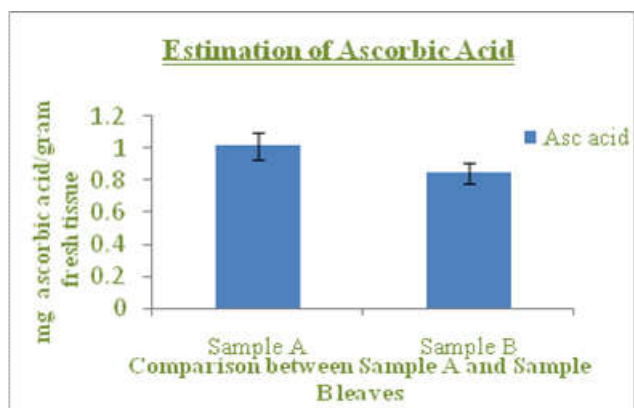
Graph 8. Graph showing the estimation of Peroxidase activity in both Sample A and Sample B leaves of *Alstonia scholaris*

Estimation of ascorbic acid

The quantity of Ascorbic acid in Sample A and Sample B leaves are given below in Table 7. The Sample B leaves of *Alstonia scholaris* show Ascorbic acid content of 0.8475 mg/gram fresh tissue whereas the Sample A leaves show ascorbic acid content of 1.016 mg/gram fresh tissue.

Table 7. Estimation of Ascorbic acid in Sample A and Sample B leaves

Parameter	Sample A leaf (mg/g tissue)	Sample B leaf (mg/g tissue)
Ascorbic acid	1.016 ± 0.082	0.8475 ± 0.066



Graph 9. Graph showing the estimation of Ascorbic acid in both Sample A and Sample B leaves of *Alstonia scholaris*

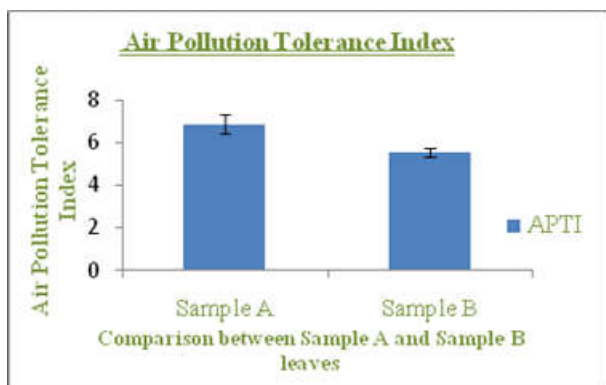
Estimation of air pollution tolerance index

Impact of air pollution is increasing day by day and a single parameter is not enough to know the rate of air pollution in the city. To simplify the problem, APTI acts as a significant tool that serves the purpose of knowing how much a plant is tolerant to environmental pollution (Nayak et al. 2014). APTI combines four biochemical parameters i.e. pH of leaf extract (P), total chlorophyll content (TC), ascorbic acid content (AA) and relative water content (RWC).

Table 8. Estimation of Air Pollution Tolerance Index in Sample A and Sample B leaves

Parameter	Sample A leaf	Sample B leaf
pH	6.03	7.14
Relative water content	61.40	48.38
Total Chlorophyll (mg/g tissue)	0.976 ± 0.039	0.831 ± 0.061
Ascorbic acid (mg/g tissue)	1.016 ± 0.082	0.8475 ± 0.066

Air Pollution Tolerance Index : Sample A = 6.851, Sample B = 5.513



Graph 10. Graph showing the estimation of Air Pollution Tolerance Index in both Sample A and Sample B leaves of *Alstonia scholaris*.

DISCUSSION

Air pollution is the presence of any abnormal particulate matters or harmful gas in the atmosphere which is not naturally occurring. In this era of industrialization and development, higher levels of pollution is mainly caused due to burning of fossil fuels, increasing number of motor vehicles and industrial emissions. In order to adapt to the pollution, many plants accumulate these pollutants within their body or just simply eliminate them. But those which cannot adapt, face injury, wounding in their cells and eventually die. Release of pollutants directly into the atmosphere forms primary pollutants and when these primary pollutants mix with other toxic compounds, they form secondary pollutants. Sulfur dioxide (SO₂), nitrogen oxides (NO_x, NO), carbon monoxide (CO), heavy metals and suspended particulate matters are some of the most potential and hazardous primary air pollutants found in the industrial area. The biggest sources of particulate matter pollution in metropolitan cities are motor vehicles and industrial emissions. Further reports by Tripathi and Gautam (2007) suggest that production of SO₂ occurs both naturally by sulphur bacteria activities and by man-made sources like burning of fossil fuel, automobile emissions. Air pollution is an unhealthy activity and it puts the life of plants as well as humans beings at risk (Agbaire and Akporhonor, 2014). Several primary and secondary air pollutants have been recognized as a hazardous element for plants. As reported by Seyyednejad, Niknejad, and Yusefi in 2009, plants in response to short duration exposure of high concentrations of SO₂ show symptoms of leaf necrosis. Plants undergo a number of functional and structural changes when exposed to air pollutants at first but later on they display morphological symptoms of injury (Liu and Ding, 2014). Plants show characteristic symptoms when exposed to air pollutants. These symptoms may occur depending upon the type of pollutant and the time of exposure to that specific pollutant. It mainly includes browning on the edges and spotting on the leaf surface, induction of galls, buds, flowers, leaves, fruits falling off, reduction in the girth and height of the plant. In this project work, we try to observe the Biochemical, Morphological and Anatomical changes of *Alstonia scholaris* leaves which are affected due to air pollution and show gall formation and we try to compare them with the sample A Leaves of the same plant. *Pauropsylla tuberculata* (Psyllidae), is a gall inducer on leaves of *Alstonia scholaris*. About 350 species of gall inducers are present in the psyllidae family that attack the leaves of dicot plants (Hodkinson 1984). The present study of gall formation on the roadside located *Alstonia scholaris* showed leaves closer to the ground had maximum numbers of galls while leaves towards the top of the canopy, exhibited no gall formation. Young plant cells show much more resistance against gall inducing insects in comparison to modified tissues (Rohfritsch, 1992).

Morphological and anatomical results in the present study showed variations between the sample A and the sample B leaves. Firstly, the area of the leaves of sample B leaves (27.92 sq cm) seemed to be smaller in size than the sample A (29.72 sq cm) ones. Studies conducted by earlier researchers showed pollution by suspended particulate matters along with other air pollutants such as CO, SO₂ and NO_x are the reasons for deformities in the leaf (Joshi and Swami, 2007; Deepalakshmi, 2013; Talukdar et al. 2016). Secondly, it was also noted that the Air Pollution Index (1.61) value was significantly higher in

the experimental area compared to the control area as represented by bar diagram. Since sample A leaves showed no sign of galls, so Air Pollution Index is zero. So it can be said that API is an useful parameter that helps in early detection of air pollutants affecting *Alstonia scholaris*. Apart from this, changes in the number and shape and size of stomata were also noticed. The stomatal frequency in the sample A leaves was 16355 per unit area whereas stomatal frequency in the sample B leaves was 14356 per unit area. The length and breadth of the stomata also reduced in case of the sample B leaves (L=2 μm , B=2 μm) compared to the sample A leaves (L=2.16 μm B=2.66 μm).

Biochemical results in the present study showed variations between the sample A and sample B leaves. The Chlorophyll content (Chlorophyll a, Chlorophyll b, Total Chlorophyll) decreased in the infested leaves with the increase in the number of galls. According to Speeding and Thomas (1973), plant cells under stressful conditions show a decrease in their total chlorophyll content. A decrease in the chlorophyll content of the infested leaves results in reduction of plant productivity. Colouration of the area of the leaf changes where the eggs are first laid (Datta and Dhiman, 2016). This might be due to the loss of chlorophyll in those leaves. Another reason for the decline in chlorophyll level in the damaged leaves could be chloroplast damage. Higher the number of galls, higher will be the chlorophyll degradation. As a result discolouration of these leaves occur and they turn yellow, crumpled and finally fall off. Carotene content decreased in the sample B leaves in comparison to the sample A leaves. Since Chlorophyll and carotene pigments go hand in hand, so the degradation of Chlorophyll also results in the degradation of the carotene pigment in the leaves showing gall formation. Increased proline content in the sample B leaves of *Alstonia scholaris* was observed in comparison to the ungalloled leaves. Studies on the galloled leaves of *Populus* showed increased proline level (el-Akkad 2004). Increased proline production in plants in a stressful environment is quite common (Gibon et al. 2008). Enhanced proline levels increase reactive oxygen species (ROS) production in the mitochondria when the sample B leaves exhibit symptoms like gall formation, chlorosis, necrosis of leaves. Hypersensitive response of plants has shown that higher amount of ROS is produced through the metabolism of proline. Increased proline level occurs due to environmental pollution which makes the plant more tolerant to stress by maintaining cell turgor pressure. So, proline can be used as a stress indicator for gall infested plants. Higher phenol concentration was observed in the sample B leaves in comparison to the sample A ones. Studies in the leaf galls of some species of *Ficus* and *Quercus* showed changes in their phenol levels. Balakrishna and Raman (1992) reported that the formation of galls increases the phenol content in the leaves. High phenol content in turn increases production of secondary phenols and other compounds which acts as a defense mechanism against the gall inducing insects (Kar, Jena, Srivastava, Giri, Sinha, 2013). The quantity of Total Carbohydrate decreased in the infested leaves in comparison to the sample A leaves. The damage to the Sample B leaves causes disintegration of sugars, be it reducing or non reducing. As a result, the infected leaves receive decreased food supply. Depletion in sugar levels force the cells to undergo necrosis, chlorosis and finally result in premature death. The peroxidase activity increased in the Sample B leaves whereas decreased in the sample A ones. Pollution stress stimulates production of high levels of peroxidase in the plant cell. The plant's response

to stress is evident from the increased levels of peroxidase in the galloled leaves. Enhanced level of secondary metabolites in the leaves is another indication of stress. Alteration in the biochemical pathways occur due to the increased gall formation on the leaves. High level of peroxidase in the damaged leaves suggest that it also can be used as an indicator of stress (Chauhan 2015).

The quantity of Ascorbic acid decreased in the gall infested leaves whereas it increased in the Sample A leaves. Being a strong reducer, ascorbic acid defends the plants by initializing a number of physiological methods and thus plays a significant part in photosynthesis. Higher the concentration of ascorbic acid, higher is it's reducing power (Agbaire and Esiefarienrhe, 2009; Raza and Murthy, 1988). Increased conversion of hexose sugar to ascorbic acid is mainly due to high pH which makes the plant tolerant to pollutants. Being a natural detoxicant, ascorbic acid protects the plant cells from injury caused due to pollution (Singh, Rao, Agrawal, Pandey, and Naryan, 1991). High ROS production is due to depletion in the ascorbic acid levels. How the plant responds to the surrounding air pollutants depends on a number of factors. It mainly varies from one species to another and mostly depends on the kind of pollutant, it's concentration, how it is reacting with the plant cells and the time of exposure to the pollutants. The plant cells incorporate these pollutants into the system and cause some changes in the chemical composition, structure and may alter some of their functions before being removed or absorbed. Some plants show least changes while others are susceptible to it. Since the application of a single parameter is not enough to understand the alterations instigated by several pollutants, so the concept of APTI is put to use (Farooq and Beg, 1980). Air Pollution tolerance index is a combination of a number of parameters like ascorbic acid, relative water content, total chlorophyll and pH of leaf extract required for the assessment of plant tolerance to pollutants.

APTI value	Response
<1	Very sensitive
1 to 16	Sensitive
17 to 29	Intermediate
30 to 100	Tolerant

APTI of both sample A and sample B (1 to 16 range) shows *Alstonia scholaris* is sensitive to air pollution. Species having lower APTI value may act as bio-indicators of pollution. APTI is considered as an authentic method for checking the sensitivity of a large number of trees to air pollutants. APTI (Air Pollution Tolerance Index) was calculated by taking into account the total chlorophyll, ascorbic acid, pH of the leaf extract and relative water content of the leaves to determine their susceptibility to air pollution (Singh and Rao 1983; Choudhury and Banerjee, 2009; Tripathi et al., 2009). Morphological and biochemical changes occurred along with physiological alterations when plants were exposed to air pollutants (Prajapati and Tripathi, 2008; Hamid and Jawaid, 2009; Assadi, Pirbalouti, Malekpoor, Teimori, and Assadi, 2011; Taneer and Albert, 2013). Prior reports by Ranft (1968) and Pfeffer (1978) suggest that heavy air pollution zones show higher infestation by gall-forming aphids such as *Sacchiphantes* sp. This study on the morphological, biochemical and physiological changes in leaves of *Alstonia scholaris* caused due to gall formation contributes to the existing knowledge of gall study done in other species of plants and gives us information on how the plants adapt to environmental pollution.

Thus we can say that other than proline, peroxidase and phenol content, the other biochemical parameters reduced in case of sample B leaves and increased in case of sample A leaves.

Conclusion

From the above study, it can be interpreted that due to air pollution the leaves of *Alstonia scholaris* get infested with gall inducing insects and with increase of gall formation, the biochemical, morphological and anatomical characters are altered in the *Alstonia scholaris* leaf. This study is an assessment of host-plant herbivory and the gall formation onto leaf, which can be a suitable indicator to know API index as a primary test of air pollution. Bioindicator plants are very useful to draw public awareness to air pollution problems, since they can demonstrate the visibility of otherwise invisible air pollutants, especially in city environments and in developing countries where industrialisation and urbanisation are increasing. Studies like this should be taken to evaluate the present status of the environment of Kolkata city so that the proper measures by the concerned authorities should be taken and policy making process should be in line with recommendation of such studies like planted trees as *Alstonia scholaris* should be a part of landscape avenues of any developing city as it prove its greater strength of biomonitoring air quality in a city.

REFERENCES

- Agbaire, P.O and Esiefarienrhe, E. 2009. Air Pollution tolerance indices (apti) of some plants around Otorogun Gas Plant in Delta State, Nigeria. *Journal of Applied Sciences and Environmental Management*, 13(1): 11 - 14
- Agbaire, P.O. and Akporhonor, E.E. 2014. The effects of air pollution on plants around the vicinity of the Delta Steel Company, Ovwian-Aladja, Delta State, Nigeria. *IOSR J. Environ. Sci. Toxicol. Food Technol.*, 8 (7): 61–65.
- Arnon, D. I. 1949. Copper Enzymes in Isolated Chloroplasts Polyphenol Oxidase in Beta Vulgaris. *Plant Physiol.*, 24 (1): 1-15.
- Assadi Aghil, Abdollah Ghasemi Pirbalouti, Fatemeh Malekpoor, Nasrin Teimori and Leila Assadi, 2011. Impact of air pollution on physiological and morphological characteristics of *Eucalyptus camaldulensis* Den. *Journal of Food, Agriculture and Environment*, 9 (2): 676-679.
- Badamasi, H. 2017. Biomonitoring of air pollution using plants. *Journal of Environmental Science*, 2:27 – 39.
- Balakrishna, P. and Raman, A. 1992. Cecidogenesis of leaf galls of *Strychnos nux-vomica* (Loganiaceae) induced by the jumping plant louse species, *Diaphorina truncate* (Homoptera: Psylloidea: Psylloidea). *Entomologia Generalis*. 17:285-292.
- Bates, L. S., Waldren, R. P. and Teare, I. D. 1973. Rapid determination of free proline for water-stress studies, *Plant and Soil.*, 39(1): 205–207.
- Chauhan Aparajita, 2015. Peroxidase Activity as an Indicator of SO₂ Tolerance in Crop Plants. *International Journal of Innovative Research and Development*, 4 (13).
- Chauhan. A. and Joshi, P.C. 2010. Effect Of Ambient Air Pollutants On Wheat and Mustard Crops Growing In The Vicinity Of Urban and Industrial Areas. *New York Science Journal*, 3(2):52 -60.
- Choudhury, P. and Banerjee, D. 2009. Biomonitoring of Air Quality in the Industrial town of Asansol using Air Pollution Tolerance Index approach. *Research Journal of Chemistry and Environment*, 13(1): 46-51.
- Datta Om and Dhiman, S.C. 2016. Studies on the histomorphology of leaf galls on *Alstonia scholaris* L. (Apocynaceae) induced by *Pauropsylla tuberculata* Crawford. *International Journal of Agricultural Invention*, 1(1):65 - 70.
- Deepalakshmi, A. P., Ramakrishnaiah, H., Ramachandra, Y. L. and Naveen Kumar, N. 2014. Leaves of Higher Plants as Indicators of Heavy Metal Pollution along the Urban Roadways. *International Journal of Science and Technology*, 3(9):340 -346.
- El-Akkad, S.S. 2004. Biochemical changes induced in *Populus nigra* leaves by galling aphids *Pemphigus populi*. *International Journal of Agricultural and Biology*, 6:659-664.
- Farooq, M. and Beg, M.U. 1980. Effect of aqueous SO₂ on the membrane permeability of common Indian tree leaves, *New Botanisz*, 7: 213-217.
- Furlan, C.M., Salatino, A. and Domingos, M. 1999. Leaf contents of nitrogen and phenolic compounds and their bearing with the herbivore damage to *Tibouchina pulchra* Cogn. (Melastomataceae), under the influence of air pollutants from industries of Cubatão, *São Paulo Revista Brasil. Bot.*, São Paulo. 22(2):317-323.
- Gibon, Y., Sulpice, R. and Larher, F. 2008. Proline accumulation in canola leaf discs subjected to osmotic stress is related to the loss of chlorophylls and to the decrease of mitochondrial activity. *Physiologia Plantarum*, 110(4):469 - 476.
- Hamid, N. and Jawaid, F. 2009. Effect of short-term exposure of two different concentrations of sulphur dioxide and nitrogen dioxide mixture on some biochemical parameter of soybean (*Glycine max*), *Pakistan Journal of Botany*, 41:2223-2228
- Harris, L.J. and Ray, S. N. 1935. Diagnosis of vitamin-C subnutrition by urine analysis with a note on the antiscorbutic value of human milk. *Lancet Journal*, 228:71-77.
- Hedge, J.E. and Hofreiter, B.T. 1962. Carbohydrates chemistry. In: Whistler RL, Be Miller JB (eds) 17, Academic Press, New York.
- Hodkinson, I.D. 1984. The biology and ecology of the gall forming Psylloidea (homoptera), *Biology of gall insects*. pp 59 – 77.
- Holopainen, J.K. and Oksanen, J. 1995. Arboreal insects as indicators of air pollution effects on woody plants.
- Holopainen, J.K. and Oksanen, J. 1995. Arboreal insects as indicators of air pollution effects on woody plants.
- Holopainen, J.K. and Oksanen, J. 1995. Arboreal insects as indicators of air pollution effects on woody plants.
- Joshi, P. C. and Swami Abhishek, 2007. Physiological responses of some tree species under roadside automobile pollution stress around city of Haridwar, India. *The environmentalist*, 27(3): 365–374.
- Kar, PK., Jena, KB., Srivastava, AK., Giri, S. and Sinha, MK. 2013. Gall induced stress in the leaves of *Terminalia arjuna*, food plant of tropical tasar silkworm, *Antheraea mylitta*. *Emir. I. Food Agric.*, 25(3):201-210.
- Kirk and Allen, R.L. Dependence of chloroplast pigment synthesis on protein synthesis: effect of actidione. *Biochem. biophys. Res. Commun.*, 21: 523–530.
- Liu, Y.J. and Ding, H. 2008. Variation in air pollution tolerance index of plants near a steel factory: implications

- for landscape-plant species selection for industrial areas. *WSEAS Trans. Environ. Dev.*, 1 (4), 24–32.
- Malick, CP. and Singh, MB. 1980. In: plant Enzymology and Histo Enzymology. Kalyani Publishers. New Delhi.pp:286.
- Muhammad, S., Khan, Z.U.D. and Saddiqui, M.F. 2014. *Alstonia scholaris* (L.) r.br. planted bioindicator along different road-sides of lahore city. *Pakistan Journal of Botany*, 46(3): 869-873.
- Nayak. D., Patel, D.P., Thakare, H.S., Satashiya, K. and Shrivastava, P.K. 2015. Evaluation of air pollution tolerance index of trees. *Res. Environ. Life Sci.*, 8 (1):7-10.
- Pfeffer. A. 1978. Wirkungen von Luftverunreinigungen auf die freilebende Tierwelt. Schweiz. Z. Forstwes. 129:362-368.
- Prajapati, S.K. and Tripathi, B.D. 2008. Seasonal Variation of Leaf Dust Accumulation and Pigment Content in Plant Species Exposed to Urban Particulates Pollution. *Journal of Environmental Quality*, 37(3): 865-870.
- Ranft, H. and Daessler, H. G. 1969. Behavior of lichens and mosses as affected by sulfur dioxide gas. *Flora, Abt. B; Journal*, 158.
- Raza, S.H. and Murthy, M.S.R. 1988. Air Pollution Tolerance index of certain plants of Nacharam Industrial Area, Hyderabad. *Indian J. Bot.*, 11(1): 91-95.
- Rohfritsch, O. 1992. Patterns in gall development *In* Biology of insect induced galls (J.D. Shorthouse and O. Rohfritsch, eds.). Oxford University Press, New York, p.60-86.
- Seyyednejad, S.M., Niknejad, M. and Yusefi, M. 2009. The Effect of Air Pollution on Some Morphological and Biochemical Factors of *Callistemon citrinus* in Petrochemical Zone in South of Iran. *Asian Journal of Plant Sciences*, 8 (8): 562-565.
- Singh, S.K., Rao, D.N., Agrawal, M., Pandey, J. and Narayan, D. 1991. Air Pollution Tolerance index of plants. *Journal of Environmental Management*, 32: 45-55.
- Singh, S.K., Rao, D.N., Agrawal, M., Pandey, J. and Naryan, D. 1991. Air pollution tolerance index of plants. *J. Environ. Manag.*, 32 (5): 45–55.
- Speeding, D.J. and Thomas, W.J. 1973. Effect of sulphur dioxide on the metabolism of glycollic acid by barley (*Hordeum vulgare*) leaves. *Aust. J. Biol. Sci.*, 6: 281-286.
- Susy Albert., Padhiar Ameer, Gandhi Dhara and Nityanand Priyanka, 2011. Morphological, anatomical and biochemical studies on the foliar galls of *Alstonia scholaris* (Apocynaceae)., *Revista Brasil. Bot.*, 34(3):343-358.
- Talukdar Partha, Das Kaushiki, Dhar Shrinjana, Talapatra, S.N and Swarnakar Snehasikta, 2016. Galls on *Alstonia scholaris* leaves as air pollution indicator. *World Scientific News*, 52: 181-194.
- Tanee, F.B.G. and Albert, E. 2013. Air pollution tolerance indices of plants growing around Umuebulu Gas Flare Station in Rivers State, Nigeria. *African Journal of Environmental Science and Technology*, 2013. 7(1): 1-8.
- Temmerman, Bell, Garrec, Klumpp, Krause and Tonnnejck, 2001. Biomonitoring of air pollutants with plants- Considerations for the future. pp:337 – 373.
- Tripathi A. K. and Gautam Mukesh, 2006. Biochemical parameters of plants as indicators of air pollution, *Journal of Environmental Biology*, 2006. 28(1):127-132
- Tripathi, A., Tiwari, P.B., Mahima and Singh. D. Assessment of air pollution tolerance index of some trees in Moradabad city, India. *Journal of Environmental Biology*, 30(4) :545-550.
