

SELECTION OF BIOINDICATORS FOR AIR POLLUTION BASED ON PEROXIDASE ACTIVITY OF PLANT SPECIES FOUND IN THE CHEMBUR INDUSTRIAL AREA OF MUMBAI, INDIA -A PRIMARY STEP TO GREEN ZONE ENRICHMENT

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ABSTRACT

Industrialization and population growth are the important factors for the deterioration of air quality in the current scenario. Air pollution monitoring using plants as bioindicators is the best and cheapest way. Oxidative stress induced on plants due to air pollution can be studied using antioxidant peroxidase enzyme activity. The present study aims at the comparative assessment of peroxidase activity of twelve plant species from the Chembur industrial area and less polluted zone of similar ecological conditions for two years. The peroxidase activity was determined using the spectrophotometric method. The observations based on the current investigation revealed a rise in the peroxidase activity of twelve plant species of the industrial zone as compared to the less polluted zone. It may indicate an increase in the resistant capacity of all experimental samples toward the oxidative stress caused due to air pollution. Plant species like *Pithecellobium dulce* (Roxb.) Benth, *Ricinus communis* L., *Leucaena leucocephala* (Lam.) de Wit., *Albizia saman* (Jacq.) Merr, *Thespesia populnea* (L.) Sol. ex. Correa, *Polyalthia longifolia* (Sonn.) Thwaites, *Prosopis juliflora* (SW.) DC., *Terminalia catappa* L. showed higher peroxidase activity values and hence may be used as bioindicators. *Thespesia populnea* (L.) Sol. ex. Correa, *Polyalthia longifolia* (Sonn.) Thwaites and *Terminalia catappa* L. being evergreen plants may be planted in good numbers in the Chembur industrial area. It may act as a primary step to ameliorate the industrial air pollution in the Chembur industrial zone by enhancing the green zone. This kind of investigation with other factors may help us to sustain our mother nature for future generations.

Keywords: Air Pollution, Peroxidase Activity, Industrial Zone, Bioindicators.

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INTRODUCTION

The major concern of the present world is air pollution. Growth in industrialization and population are the main causes of air pollution. Air pollution monitoring is essential in present conditions as it exhibits various health concerns. Plants can be used as bio-indicators for the assessment of air pollution extent as they incorporate the impact of air pollution for a longer duration and show the effects of various pollutants. Various biochemical variables of plants can be used as indicators to investigate the air pollution effects on plants.¹ Ozone, oxides of nitrogen, and oxides of sulfur affect certain physiological activities in plants.² Plants exhibit physiological and biochemical alterations that are included in various enzymatic as well as non-enzymatic defense mechanisms of plants towards oxidative stress.³ Superoxide dismutase, peroxidase, and catalase enzyme activities rise in response to oxidative stress. Peroxidase activity was considered to be an important bioindicator for air pollution.⁴ Enzyme groups like peroxidase in plants act as catalysts in redox reactions occurring in plants.⁵ Peroxidase being heat-stable enzyme can regain its activity under a certain set of conditions and can also show certain deteriorative changes in the texture, flavor, and color of fruits, and vegetables. Peroxidase enzyme facilitates redox reaction in which hydrogen peroxide accepts hydrogen from the other compounds involved in the reaction.⁶⁻⁹ The current investigation aims at the comparative assessment of peroxidase activities of twelve plant species present in the Chembur industrial area and the same species present in less polluted areas with similar ecological conditions.

EXPERIMENTAL

Study Area

The current investigation area of the Chembur suburb is situated in the northeast region of Mumbai (19.051°N 72.894 °E). Ghatkopar, Mahul, Chunabhatti, Deonar, Kurla, and Govandi are the nearby suburbs.¹⁰ The recent Comprehensive Environment Pollution Index (CEPI= 54.67) showed 80th rank of Chembur in among the polluted industrial clusters of India and 1st rank in Mumbai.^{11,12} The industrial units like Hindustan Petroleum Corporation Limited (HPCL), Rashtriya Chemical & Fertilizers (RCF), Tata Power station, Bharat Petroleum Corporation Limited (BPCL), Indian Oil Corporation Limited, Bharat Petroleum Refinery, and Deonar dumping ground are major contributors to the decline of air quality in Chembur area. Deonar dumping ground fumes, the release of nitrogen oxides by the RCF complex, and the inefficient functioning of pollution control devices are the major causes of the elevation in air pollution in this area. An increase in air pollutants of Chembur led to respiratory tract issues and other health concerns.¹³ An elevation in industrial events gave rise to an increase in gaseous air pollutants. The garbage incineration smoke from the Deonar dumping ground added to the gaseous air pollutants. This resulted in earning the name "Gas Chamber of Mumbai" to study area. Maharashtra Pollution Control Board (MPCB) showed the average concentration of 5-6 $\mu\text{g m}^{-3}$ sulfur oxides, 77-83 $\mu\text{g m}^{-3}$ nitrogen oxides, 147-148 $\mu\text{g m}^{-3}$ of Respirable Suspended Particulate matter (RSPM). The elevated average values were observed for nitrogen oxides and RSPM than the normal values.¹⁴

Chemicals and Reagents

Analytical grade reagents like H_2O_2 , Pyrogallol, phosphate buffer (pH 6.8), and H_2SO_4 were used for the analysis of samples. A Milli-Q system of deionized water was used.

Plant Species

The most common plant species from the current investigation area were selected. Three replicates of completely developed leaves of twelve plant species were collected from the Chembur industrial area as experimental samples (ES). All plant species were identified for their botanical name through Blatter Herbarium of St. Xavier's College, Mumbai. The details of plant species are mentioned in Table-1.

Table-1: Common Name and Botanical Name of Plant Species¹⁵

Plant sample	Common name of plant species	Botanical name of the plant species
S1	Ashoka	<i>Polyalthia longifolia</i> (Sonn.) Thwaites
S2	Red date	<i>Ziziphus jujube</i>
S3	Vilayati imli	<i>Pithecellobium dulce</i> (Roxb.) Benth
S4	Katkali, Vilayati Babul	<i>Prosopis juliflora</i> (Sw.) DC.
S5	Silver cock's comb	<i>Celosia argentea</i> L.
S6	Castor oil plant	<i>Ricinus communis</i> L.
S7	Giant milkweed, crown flower	<i>Calotropis gigantea</i> (L.) Dry and
S8	White lead tree, river tamarind	<i>Leucaena leucocephala</i> (Lam.) de wit
S9	Mauritian grass	<i>Apluda mutica</i> L.
S10	Indian almond	<i>Terminalia catappa</i> L.
S11	Rain tree	<i>Albizia saman</i> (Jacq.) Merr
S12	Portia tree	<i>Thespesia populnea</i> (L.) Sol. ex-correa

Sample Collection and Analysis

The commonly found plant species were identified from the present study area. The fully matured leaves of twelve different commonly found plant species in the study area were collected as Experimental Samples (ES). The leaf samples of the same plant species growing in similar ecological conditions from the less polluted area were collected as controlled samples (CS). Peroxidase activity assessment of both ES and CS was done over two years during the summer, rainy, and winter seasons. Cloth/polythene bags were used to collect plant samples from the study area. Then the plant leaf samples were immediately taken to the laboratory and preserved in a refrigerator for peroxidase activity analysis.

General Procedure

The leaf samples were homogenized with 10 mL of phosphate buffer (pH 6.8) and centrifuged. The clear upper solution was used for the enzyme source. This enzyme source was diluted 20 times and 1 mL of the diluted enzyme extract was mixed with 125 μ moles of phosphate buffer (pH 6.8), 50 μ moles of pyrogallol, and 50 μ moles of H_2O_2 . It was incubated for 5 min at 25°C after which the reaction was stopped by adding 0.5 mL of 5% (v/v) H_2SO_4 . The absorbance of the solution was measured at 420 nm to determine the amount of purpurogallin formed during the reaction. The time required for the mixture optical density to be increased by 0.1 at 420 nm was recorded and used for calculation.¹⁶

RESULTS AND DISCUSSION

The peroxidase activity values of all plant species from the experimental zone (ES) and controlled zone (CS) are shown in Table-2. From the values shown in Table-2, it is clear that all the experimental samples showed higher peroxidase activity. The controlled samples showed lower peroxidase activity. The peroxidase activities of all experimental samples from the industrial zone were higher in all seasons as compared to controlled samples from less polluted zones. The S8 sample exhibited the highest peroxidase activity (10.000 $\text{min}^{-1} \text{gm}^{-1}$) in winter 2019 and the S6 sample exhibited the lowest peroxidase activity (1.177 $\text{min}^{-1} \text{gm}^{-1}$) among experimental samples in rainy 2019. In controlled samples, the highest peroxidase activity was exhibited by S7 in summer 2018 and the least peroxidase activity was exhibited by S11 in winter 2018. The S8 sample showed the highest mean peroxidase activity of 4.644 $\text{min}^{-1} \text{gm}^{-1}$ whereas the S2 sample showed the least mean peroxidase activity of 1.954 $\text{min}^{-1} \text{gm}^{-1}$ in an industrial area. S7 controlled sample showed the highest mean peroxidase activity of 1.939 $\text{min}^{-1} \text{gm}^{-1}$ and S2 showed the least mean peroxidase activity of 1.299 $\text{min}^{-1} \text{gm}^{-1}$. The rise in the peroxidase activity of experimental plant samples in comparison with controlled area plant samples is mentioned in Table-3.

Table-2: Peroxidase Activity Values of ES and CS Plant Species

Parameter	Sample	ES								CS							
	Season	Summer		Rainy		Winter		Mean	Range	Summer		Rainy		Winter		Mean	Range
	Year	2018	2019	2018	2019	2018	2019			2018	2019	2018	2019	2018	2019		
Peroxidase activity (min ⁻¹ gm ⁻¹)	S1	2.611 (±0.024)	4.000 (±0.018)	2.500 (±0.015)	3.534 (±0.012)	3.003 (±0.029)	2.308 (±0.016)	2.993	2.308-4.000	1.764 (±0.010)	2.222 (±0.026)	1.017 (±0.024)	2.857 (±0.021)	0.758 (±0.019)	0.667 (±0.014)	1.548	0.667-2.857
	S2	2.141 (±0.011)	1.934 (±0.020)	2.611 (±0.015)	1.934 (±0.012)	1.876 (±0.009)	1.225 (±0.017)	1.954	1.225-2.611	1.464 (±0.023)	1.395 (±0.028)	1.667 (±0.016)	1.250 (±0.018)	0.962 (±0.011)	1.053 (±0.014)	1.299	0.962-1.667
	S3	5.000 (±0.021)	2.309 (±0.013)	1.464 (±0.018)	3.155 (±0.030)	1.499 (±0.028)	2.069 (±0.007)	2.583	1.464-5.000	1.072 (±0.012)	1.499 (±0.026)	1.304 (±0.010)	2.500 (±0.019)	0.645 (±0.015)	1.580 (±0.024)	1.433	0.645-2.500
	S4	3.003 (±0.012)	4.000 (±0.029)	3.534 (±0.008)	1.395 (±0.005)	4.000 (±0.017)	3.000 (±0.020)	3.155	1.395-4.000	1.715 (±0.015)	1.091 (±0.028)	0.926 (±0.023)	1.201 (±0.018)	2.326 (±0.016)	1.091 (±0.025)	1.392	0.926-2.326
	S5	3.003 (±0.026)	2.857 (±0.019)	3.333 (±0.022)	3.155 (±0.017)	2.141 (±0.030)	1.818 (±0.011)	2.718	1.818-3.333	1.153 (±0.013)	2.000 (±0.027)	2.725 (±0.018)	1.539 (±0.023)	0.794 (±0.020)	1.305 (±0.008)	1.586	1.153-2.725
	S6	3.155 (±0.017)	5.456 (±0.027)	6.667 (±0.014)	1.177 (±0.010)	2.632 (±0.028)	3.751 (±0.032)	4.139	1.177-6.667	1.250 (±0.029)	2.500 (±0.015)	1.200 (±0.012)	1.133 (±0.016)	1.621 (±0.020)	0.952 (±0.025)	1.443	0.952-2.500
	S7	5.988 (±0.033)	4.292 (±0.009)	1.876 (±0.027)	2.309 (±0.013)	1.364 (±0.023)	1.667 (±0.010)	2.916	1.364-5.988	3.745 (±0.018)	2.143 (±0.025)	1.715 (±0.015)	2.141 (±0.029)	0.855 (±0.038)	1.034 (±0.022)	1.939	0.855-3.745
	S8	4.608 (±0.026)	2.609 (±0.017)	3.155 (±0.014)	3.745 (±0.023)	3.745 (±0.035)	10.000 (±0.030)	4.644	2.609-10.000	1.667 (±0.015)	1.304 (±0.028)	1.000 (±0.020)	1.580 (±0.007)	2.000 (±0.019)	0.926 (±0.012)	1.413	0.926-2.000
	S9	2.725 (±0.021)	1.622 (±0.025)	1.395 (±0.019)	2.000 (±0.022)	2.857 (±0.015)	2.500 (±0.011)	2.183	1.395-2.857	1.364 (±0.013)	1.034 (±0.018)	0.500 (±0.027)	1.304 (±0.028)	2.611 (±0.009)	1.818 (±0.016)	1.439	0.500-2.611
	S10	1.499 (±0.014)	2.000 (±0.017)	5.000 (±0.010)	4.608 (±0.015)	1.539 (±0.031)	1.579 (±0.029)	2.704	1.499-5.000	0.769 (±0.034)	1.818 (±0.012)	2.070 (±0.020)	2.725 (±0.028)	0.719 (±0.019)	1.429 (±0.024)	1.588	0.719-2.070
	S11	1.667 (±0.015)	5.456 (±0.008)	5.000 (±0.025)	1.621 (±0.033)	1.304 (±0.026)	6.002 (±0.018)	3.508	1.304-6.002	1.034 (±0.013)	2.857 (±0.011)	1.364 (±0.023)	1.111 (±0.028)	0.498 (±0.016)	1.200 (±0.022)	1.344	0.498-2.857
	S12	3.333 (±0.017)	3.750 (±0.015)	1.580 (±0.024)	4.608 (±0.020)	1.934 (±0.008)	3.159 (±0.014)	3.061	1.580-4.608	0.714 (±0.019)	1.580 (±0.013)	0.833 (±0.029)	1.153 (±0.030)	1.852 (±0.027)	3.003 (±0.025)	1.523	0.714-3.003

Table-3: An Increase in Peroxidase Activity Values of Experimental Samples Compared with Controlled Samples

Plant sample	Rise in peroxidase activity of ES in comparison to CS ($\text{min}^{-1} \text{gm}^{-1}$)					
	Summer		Rainy		Winter	
	2018	2019	2018	2019	2018	2019
S1	0.847	1.778	1.483	0.677	2.245	1.641
S2	0.677	0.539	0.944	0.684	0.914	0.172
S3	3.928	0.810	0.160	0.655	0.854	0.489

S4	1.288	2.909	2.608	0.194	1.674	1.909
S5	1.850	0.857	0.608	1.616	1.347	0.513
S6	1.905	2.956	5.467	0.044	1.011	2.799
S7	2.243	2.149	0.161	0.168	0.509	0.633
S8	2.941	1.305	2.155	2.165	1.745	9.074
S9	1.361	0.588	0.895	0.696	0.246	0.682
S10	0.730	0.182	2.930	1.883	0.820	0.150
S11	0.633	2.599	3.636	0.510	0.806	4.802
S12	2.619	2.170	0.747	3.455	0.082	0.156

As shown in Table-3, all the twelve experimental plant samples showed a rise in peroxidase activity in all seasons. Experimental samples S3, S4, S6, S8, S10, S11, and S12 showed a greater rise in peroxidase activity compared to controlled area samples. Experimental samples S1, S5, and S7 also exhibited a comparatively higher rise in peroxidase activity. The highest rise of $9.074 \text{ min}^{-1} \text{ gm}^{-1}$ in peroxidase activity was exhibited by the S8 experimental sample in the winter 2019 season. The raised peroxidase activity may be due to an increase in plant stress under air pollution. The lower peroxidase activity was observed in controlled samples as there is less stress on plants. Plants have self-defense mechanism which helps them to survive even under pollution stress. The air pollution load increases the metabolic activities in plants where the peroxidase enzyme breaks down the hydrogen peroxide into water and oxygen. The rise in peroxidase activity may indicate an elevation in the tolerance level of plant species to withstand the air pollution stress. Different plant species show variation in the rise of peroxidase activity.¹⁷ Many researchers proposed that the peroxidase activity of plants may be useful as a general indicator of oxidative stress due to air pollution and cannot be utilized as an explicit indicator for a particular air pollutant.¹ Various studies reported a strong relation between the rise in peroxidase enzyme activity of plant leaves with their increasing resistance towards air pollution-induced oxidative stress.^{18,19,20,21} The young leaves show higher antioxidant concentrations than the aged leaves. All the experimental samples exhibited an appreciable rise in peroxidase activity indicating their resistance against industrial air pollution. The rise in peroxidase activity in plant leaf cells may be due to some physical damage, microbial invasion, or pollution stress. Elevation in peroxidase activity differs from species to species in plants. It also depends on the nature and the concentration of pollutants present in the environment.²² A greater rise in the peroxidase activity may be due to a greater resistance of plants towards air pollution.²³ There is very little research on the comparative study of peroxidase activity of plant species from polluted and controlled zones. The present study exhibited the comparison between the peroxidase activity levels of industrial and less polluted zone plant leaves that may be useful in selecting suitable air pollution-resistant plant species for enhancing the green zone in the Chembur industrial area. The information about the concentration of air pollutants cannot serve as a reliable tool to study the impact of air pollution on plants or the environment. Plants may act as bioindicators for the investigation as well as mitigation of air pollution effects.

CONCLUSION

The investigations of present study may be used to detect the primary effects of industrial air pollution before any visible injury occurs to the plants. Plant species like *Pithecellobium dulce* (Roxb.) Benth, *Prosopis juliflora* (SW.) DC., *Ricinus communis* L., *Leucaena leucocephala* (Lam.) de Wit., *Terminalia catappa* L., *Albizia saman* (Jacq.) Merr, *Thespesia populnea* (L.) Sol. ex. Correa may be used as a bioindicator. *Thespesia populnea* (L.) Sol. ex. correa and *Terminalia catappa* L. being evergreen plants may be planted in good numbers in the Chembur industrial area to enrich the green zone around it. It may act as a primary step to mitigate the industrial air pollution in the Chembur industrial zone. The present investigation may help us to preserve our environment.

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CONFLICT OF INTERESTS

The authors have declared that no conflict of interest exists.

AUTHOR CONTRIBUTIONS

All the authors contributed significantly to this manuscript, participated in reviewing/editing, and approved the final draft for publication. The research profile of the authors can be verified from their ORCID IDs, given below:

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