EFFECT OF AIR POLLUTION ON LEAF EPIDERMIS

Dissertation submitted to the Jawaharlal Nehru University in partial fulfilment of the requirements for the Degree of MASTER OF PHILOSOPHY

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SCHOOL OF ENVIRONMENTAL SCIENCES , DIE LILLE . 1979

JAWAHARLAL NEHRU UNIVERSITY

NEW DELHI

OCTOBER 1979

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PREFACE

This dissertation entitled "Effect of Air Pollution on Leaf Epidermis" has been carried out at the School of Environmental Sciences, Jawaharlal Nehru University, New Delhi. The work is original and has not been submitted in full or in part for any degree or diploma of any university.

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ACKNOWLEDGMENTS

enjoyed such extensive and valuable supports as I have in carrying out the present research work. Dr. C.K. Varshney, Associate Professor, School of Environmental Sciences, Jawaharlal Nehru University, New Delhi helped originally to formulate the problem and his constant encouragement and ready store of fresh ideas strongly influenced me throughout the course of study. His wide ranging research effort is reflected in every Chapter.

My thanks are due to Professor B. Bhatia, the former Dean and Prof. J.M. Dave, Dean of School of Environmental Sciences for providing me a cooperative environment.

I am indebted to my colleagues, Mr. S.E.K. Varshney, Kailash Mandhan and J.M. Rao for their valuable suggestions and kind helps.

Efforts of Gajanan Hegde for rapid typing and retyping deserves full appreciation. Indispensable help of Mr. Saini for beautiful photographs is worth regardable.

CHAPTER - I

INTRODUCTION

INTRODUCTION

Growth of thermal power generation and rapid expansion of industries and transport systems have created severe problems of air pollution both in the developed and the developing countries. Air pollution adversly affects vegetation, materials, structures, animals and human health. During the last two decades there has been a lot of interest in the study of biological responses to air pollutants such as particulate matter, oxides of carbon, oxides of sulphur, oxides of nitrogen and photochemical oxidants. A rapid survey of the effects of air pollutants on the living system shows that green plants are relatively more vulnerable to air pollution. A comparative account of plant and animal responses towards the varying concentrations of major air pollutants are given in Table 1. Air pollution injury to plants represented by necrotic patches on leaves, excessive leaf fall, stunted growth occurs at comparatively lower concentrations of air pollutants as compared to their effect on animals. Concentration of air pollutants at which plants are reported to die are not fatal to animals and merely cause eye irritation and some impairment of respiratory process in animals.

Table 1

A Comparision of Plants and Animals Responses
Towards Major Air Pollutants

Pollutant	Conc. ug/m ³	Effects on plants	Effects on animals
Sulphur dioxide ¹	80-100	Chronic injury	No visible effects
	120-140	Chlorosis of leaves; presence of large number of necrotic patches; leaf fall in sensitive plants.	Difficulty in breathing respiratory problems
	140-180	Synergestic reactions with 03 and NO2; adverse effect on photosynthesis and flowering; growth and overall development stunted.	Increased respiratory diseases
	500-600	Death of plants	Increased lung diseases
Nitrogen dioxide ²		Visible leaf damage in sensitive plants	No effect
	2500-4000	Large number of necrotic patches; reduced photosynthesis	Polycythemia; epithelial changes in rat and monkey
Photo-chemical	60-80	Reduction in chlorophyll	No effect
oxides ³	200-250	Serious damage to leaves effect on flowering	Eye irritation
	400-600	Serious injury to plants	Respiratory problems
	1000	Almost death of plants	Impaired infusion capacity

Data from:

- Air Quality Criteria for Sulphur Oxides, National Air Pollution Control Administration, Department of Health Education and Welfare. AP-50, 801 N. Randolph Street, Arlington Wirginia, Pebruary, 1969
- 2. Air Quality Criteria for Nitrogen Oxides, National Air Pollution Control Administration, Department of Health Education and Welfare, AP-84, Washington, D.C., Jan. 1971.
- Air Quality Criteria for Photochemical Oxidants, National Air Pollution Control Administration, Department of Health, Education and Welfare, AP-63, Washington, D.C., March, 1970.

Information on effects of air pollutants on plants have grown considerably over the years. Plants responses to air pollutants are broadly divided into three categories: (a) Biochemical: Effect on enzymes (Baily and Cole, 1959; Ziegler, 1972), amino acid (Arndt, 1970), ATP (Ballantyne, 1973), membrane permeability (Wellburn et al., 1972); (b) Physiology: Effect on rate of respiration (Showman, 1972), CO₂ fixation (Ziegler, 1972, 1973) and loss of chlorophyll (Rao and Le Blane, 1966) and (c) Gross morphology: Growth rate (Bleasdale, 1972; Bell and Clough, 1973), Chlorofis and necrosis (Thomas, 1961; Brandt and Heck, 1968) and epidermal features (Sharma and Butler, 1973, 1974, 1975).

Studies conducted at the physiological and the biochemical levels are mostly based on the effect of a single pollutant or a combination of two pollutants at the most, under laboratory conditions. Thus, in nature where complex interactions are taking place among a wide array of air pollutants, it is not possible to make use of the data from the laboratory experiments reported in literature to interpret the impact of air pollution under field conditions. Field studies automatically represent an integrated impact of all the pollutants present in the atmosphere. Epidermal features, like

blast and frequency of epidermal cells, stomata, idioblast and trichomes, among other morphological parameters
have been shown to have considerable sensitivity towards
air pollutants thus, leaf epidermis could be helpful in
indicating the impact of air pollution. This view point
drives its support from two main considerations, namely,
(i) position and (ii) sensitivity of epidermis towards
environmental factors.

- (1) Position: Epidermis is the outer most layer of the leaf, therefore, it comes in intimate contact with air pollutants contineously.
- (ii) Sensitivity of epidermis towards environmental factors: Salisbury (1927) on the basis of his extensive work suggested that stomatal frequency can be of great significance in understanding environmental conditions. According to him, stomatal frequency augments with increase in the osmotic and suction pressures which could only mean a negative correlation with humidity. Leaves of Nerium oleander normally have sunken stomata, however, Aykin (1952) has shown raised stomata in these leaves by allowing them to develop in an atmosphere saturated with water vapour. Schurmann (1959) observed that by increasing the water content of a plant during leaf formation, the ratio of the stomata to epidermal cells is enhanced

but gets reduced with the reduction in the water content.

Brustrom (1961) allowed the leaves to develop under water

from the bud stage. The stomata were initiated in the

usual pattern but subsequently differentiated into stru
ctures having close resemblance with hydathodes.

Pazourek (1970) showed a close relationship between stomatal frequency and light intensity. In the case of Iris hollandica stomata developed on both the leaf surfaces under high light intensity, in constrast to plants grown under low light intensity had rather low stomatal frequency on the abaxial leaf surface. When plants were transferred from high to low light intensity, they showed a progressive decrease in the formation of stomata. was concluded that initiation of stomata is affected by the environmental conditions prevailing at that time, rather than those experienced previously. Sharma (1972) studied the effect of epidermal features in case of Verbena canadensis subjected to four different environment having varied temperature, humidity and light regimes. The stomatal frequency, epidernal cell frequency, stomatal index and trichome density were found to be affected by the environmental variations. In contrast subsidiary cell complex and trichome type remain unaffected. Sharma (1975) also reported altitudinal variation in leaf epidermis of

Cannabis sativa. Plants growing at low elevations had higher stomatal frequency and numerous long trichomes as compared to plants at high elevations.

During the past few years, there have been some studies pertaining to the effects of air pollution on the epidermal features of leaf surfaces (Table 2).

Sharma and Butler (1973) showed decrease in stomatal frequency and increase in the density and length of trichomes in the population of Trifolium repens growing in polluted area. They have also reported that the multicellular type of trichome were generally more abundant than the unicellular type in polluted areas. further observed similar changes in case of Trifolium pratense. Acer saccharum grown under polluted environment also showed similar changes in theleaf epidermis (Sharma, 1975). Recently Godzik and Sassen (1978) conducted electron microscopic study to determine the changes in the epidermal features of Aesculus hippocastanum grown under polluted environment. The epidermal cells of the adaxial leaf surface without damage in the form of visible spots were differently shaped, had fewer folds and a smooth cell surface compared with control level. On abaxial surface, normal folds had disappeared but the surface was not smooth. Stomata did not have a

normal appearence but dust on or near stomata was not common. It was suggested that alternation in the morphology of folds along with the changes in the ultrastructure of the outer wall of the epidermis may contribute to the loss of elasticity of leaves (Godzik and Sassen, 1978).

Gasalman and Davis (1978) in their study on ozone susceptibility of Azalea cultivars (Rododendron sp.) pointed out that neither rate of gas exchange nor stomatal frequency was correlated with degree of visible injury induced by ozone. They were exposed to 0.30 \pm 0.05 ppm ozone (590 \pm 100 ug/m³) for 8 hours at various times during the summer. A comparative study of the culticular and epidermal features of Calotropis procera (Asclepindaceae) collected from healthy and polluted environments (chief pollutants - carbon particulates sulphur dioxide, carbon monoxide and other oxidents) done by Yunus and Ahmad (1979) revealed that the frequency of epidermal cells, stomata and trichomes was conspicuously higher (2, 2 and 5 times respectively) in plants growing in polluted areas as compared to healthy populations. In contrast to the stomatal size, type of stomata, stomatal index and subsidiary cell complex remained unaffected by air pollution.

Eighty five leaf samples of Ricinus communis encompassing five population, four collected from healthy and one from polluted environment were examined. Stomatal frequency per mm2 and percentage of abhormal stomata (single guard cell or both the guard cell abarted) on both the upper and lower epidermis showed a marked increase in plants growing in polluted areas. One or two per cent of stomata on lower epidermis showed a slight decrease in size. A correlation of idioblast frequency and pollution was also observed (Yunus and Ahmed. 1979). Poliar specimen of Syzygium cuminii collected from the area polluted by cement dust showed higher stomatal and epidermal cell frequency as compared to the populations of healthy areas. The epidermal features of leaves of ten populations of Psidium guajava growing under different environmental conditions (polluted and healthy) have been studied. P. guajava populations collected from polluted environment (Churk cement factory compound) showed high stomatal and trichome density and short epidermal cells and trichomes as compared to healthy populations (Yunus and Ahmed, 1979).

The above studies have shown the importance of epidermal features in environmental studies. They also exhibite appreciable response to air pollutants. The quick responsive nature of the epidermal features offer

an attractive opportunity for employing leaf epidermis as a reliable bioindicator for environmental monitoring. Leaf epidermis could provide a suitable substitute for expensive and sophisticated instruments generally recommended for environmental monitoring. Standardization and development of epidermis as bioindicator would be necessary before employing them in air pollution monitoring. For this purpose, a systematic study of the effect of air pollutants on the epidermis of a wide range of plant species may provide a reliable basis for evaluating their potential as a bioindicator of air pollution.

The present study was undertaken to examine critically the effects of air pollution on the epidermal features of local plants species. This investigation consists of two parts, the first deals with the field study concerned with the comparision of the epidermal features of plants growing in areas having different levels of air pollution. The second part relates to the experimental study designed to confirm field observations by growing plants both in polluted and non-polluted environments.

Table 2 Relationship Between Atmospheric Pollution and Epidermal Features

Sl. No.	Name of plant	Type of pollution	Epidermal features	Polluted	Control	Refe- rence
1	2	3		5	6	77
	Trifolium repens		Stomatal frequency Upper Lower Stomatal size range	96.8±10.3 37.2± 7.8	44.3± 9.6	Sharma and Butler
		other oxidants	Upper Lower	7-17 15-25	10 - 15 $17 - 25$	1973
			Trichome density/cm Upper Lower	U* = 60 M* = 123	U* = 25 M* = 60	
	•		Trichome length	95	82	
2.	Trifolium pratense	particulate, matter, carbon	Stomatal frequency Upper Lover	14.4± 3.4 44.6± 2.5	25.9± 6.0 59.3± 9.5	Sharma a nd
		monoxide, other oxidants	Stomatal size range Upper Lower,	8-15 8-14	8-10 8-16	Butler 1974
	· .		Trichome density/cm'	220	80	
		•	Trichome length	336	283	•
3.	Trifolium pratence	Particulate matter, sulphur dioxide, Carbon	Stomatal frequency Upper Lower	13.9± 4.1 43.3± 7.9	25.9± 610 59.3± 9.5	-do-
: .		monoxide, other oxidants	Stomatal size range Upper	8-15	8-10 8-16	•
	•		Trichome density/cm	196	80	•
			Trichome length	396	283	

Table 2 (Contd...

Sl. No.	Name of plant	Type of pollution	Epidermal features	Polluted	Control	kefe- rence
4.	Trifolium pratence	Sulphur dioxide particulate matter, carbon	Stomatal frequency Upper Lower	17.1± 3.4 47.2± 9.7	25.9± 6.0 59.3± 9.5	- do -
		monoxide, other oxidants	Stomatal size range Upper Lower Trichome density/cm ²	8-16 8-15	8-10 8-16	
			Trichome density/cm2	488	^	•
			Lower Trichome length	175 366	80 283	
5.	Acar saccharum	High concentra- tion of pollu- tants due to industries, oil	Stomatal frequency (x±0) Trichome density/cm ² Trichome length u(x)	6.3± 1.2 6780 110	50.3 <u>+</u> 4.9	Sharma, 1975
	, ,	refineries, and automobiles.	Subsidiary cells	5-6	5-6	•
6.	Acer saccharum	Less pollutants due to jet airport and automobiles	Stomatal frequency (x±0) Trichome density/cm ² Trichome length u(x)	18.4 <u>+</u> 2.7	50.3 <u>+</u> 4.9	- do -
		A STATE OF THE STA	Subsidiary cells	5-6	5-6	
7.	Acer saccharum	Very less pollutants due	Stomatal frequency (x±0)	33.6± 3.7	50.3± 4.9	- do -
		to automobiles only	Trichome density/cm ² Trichome length u(x) Subsidiary cells	0 0 5-6	0 0 5-6	1

(Contd.....

Table 2 (Contd.....

SI.		Type of pollution	n Rpidermal features	Polluted	Control	Reference
8.	Aesculus tuppocatanum	Pollutants from coke plant	Shape of stomata Normal folds on lower surface Thick epidermal wall	Abnormal Absent Decreased	Normal Present Normal	Godzik and Sassen, 1978.
9.	Azalea cultivars	Ozone	Stomat frequency Rate of gas exchange through stomatal pore	No change	Normal	Gasalmand and Davis, 1978.
10.	Calotropis procera	Carbon particu- tate SO ₂ , CO ₂ and other oxidants	Epidermal cell frequency Stomatal frequency Trichome frequency Stomatal size Type of stomata Stomata Index Subsidiary cell complex	Twice Twice Five times No change No change No change No change	Normal Normal Normal Normal Normal Normal	Yunus and Ahmed, 1979.
11.	Ricinus communis	Polluted environment	Stomatal frequency/mm ² Percent of abnormal stomata Size of stomata on lower opidermis	Increased Increased One or two percent decrease	Normal Normal Normal	- đo -
12.	Syzygium cuminii	Chief pollutant cement dust	Stomatal frequency Epidermal cell frequency	Increased Increased	Normal Normal	- do -
13.	Psidium guajava	Chief pollutant cement dust	Stematal frequency Trichome density Size of epidermal cells	Increased Increased Decreased	Normal Normal Normal	- đo -

Chapter - II

MATERIALS AND METHODS

MATERIALS AND METHODS

Area of Study

Delhi is one of the most polluted city of India (Parikh, 1977). Delhi has been reported to have maximum concentration of sulphur dioxide, hydrogen sulphide and particulate matter as compared to other urban centers in India (Parikh, 1977). The contributing factors are thermal power stations, high density of motor vehicles and growing industrial complexes in and around Delhi. Indraprastha Power Station is a major source of air pollution in Delhi and has received considerable publicity in press. Three areas in the neighbourhood of the Power Station were selected for studying the effect of air pollution on the morphology of leaf epidermis.

Site of Study

A. Source of Air Pollution: Indraprastha Thermal Power Station is situated in the heart of the metropolitan city of Delhi at a distance of about 400-500 meters from the West bank of the river Yamuna. To the North-West and the Southern side of the power station lie the thickly populated residential complexes prominant offices and shopping centres.

The Indraprastha Power Station was commissioned in 1963. The power generation by this Power Station

varies from 100-250 MW depending upon a variety of factors, the maximum capacity is 284 MW. Approximately 3,400 tonnes of coal is burnt everyday. Three stacks are operating at present. The flyash emission of Stack I is almost double as compared with Stack II and Stack III (Table 2). Major pollutants released from the Power Station are flyash, oxides of sulphur, and carbon dioxide (Table 3). It is evident from the table that harmful pollutants such as fly ash and sulphur dioxide are emitted by the Power Station in enormous quantities.

TABLE 3

Consumption of Coal (in tonnes) and Rate of Emission of Fly ash (x 10⁷ u gm/sec) from the Indraprastha Power Station

Stack	No.	Amount	of	coal	burnt/day	Emission rat
I				400		17.65
II		e. '		1500		35.27
III				1500		35.27

Data from: Personal communication with the Indraprastha Thermal Power Station authorities.

TABLE 4

Nature and Quantity of Main Air Pollutants Released From the Indraprastha Power Station

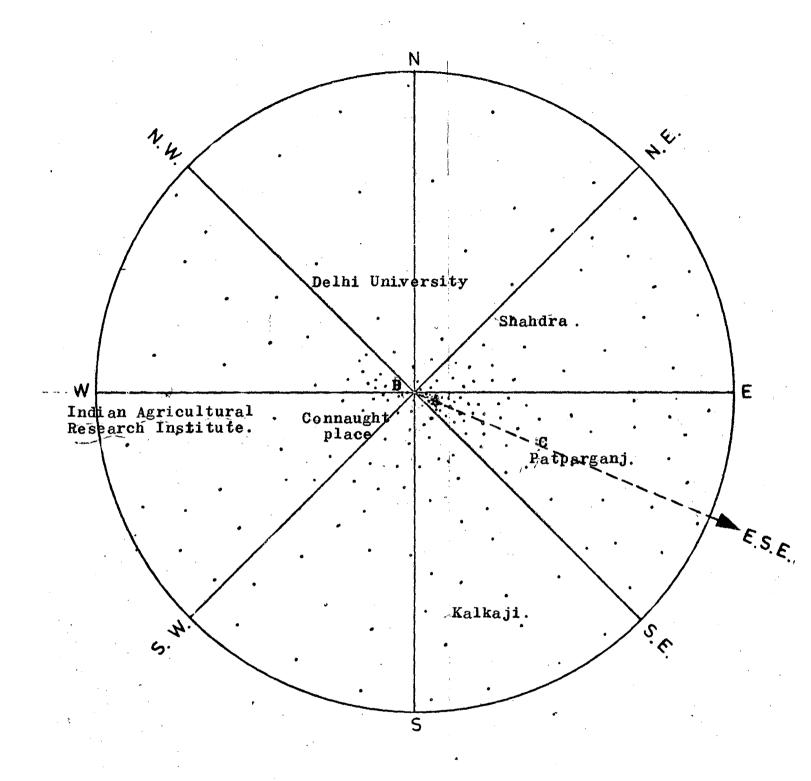
Name of pollutant	Total amount (in metric tonnes)
Fly ash	40-81*
SO ₂ (99 per cent) } SO ₃ (01 per cent) }	18
co ₂	4000

*According to "Indian Journal of Air Pollution Control" (News and Views column (annoymus), January, 1978), the amount of fly ash emitted from the Indraprastha Power Station is about 81 tonns/day but Indraprastha Power Station authorities gave a figure of 40 tonns/day (personal communication). These estimates are based on calculation, which takes into consideration the maximum efficiency of the mechanical and the electrostatic precipitators installed in the Power Station (personal communication). But, in view of the fact that these mechanical and theelectrostatic precipitators meldom work, accordingly it seems reasonable that fly ash released from the Power Station is such more than 40 tonns/day. The daily fly ash emission may well be around 81 tonns as reported in the Indian Journal of Air Pollution. ash contains about 50% SiO2, 24% Al203, 7% Fe203 trace amounts of MgO, SO3, NagO, KoO, PgO, etc.

A wide variety of plants are found in the vicinity of the Power Station, which are continuously exposed to air pollutants. The vegetation near the Power Station provides a good opportunity to study the effect of air pollutants under field conditions.

B. Site Characteristics: According to Padmanabhamurthy and Gupta (1977), the zone of high deposition/concentration is located between 0.8 to 1.6 km from the Power Station. This zone ocillates between the East and the South-East during the greater part of the year except in the monsoon menths. Zones for moderate concentration lie in the West. Three different sites (viz., Site A, Site B and Site C) were selected for detailed study representing progressively decreasing levels of air pollution (Fig. 1.1)).

Site A, located at about one km from the Power Station in the East-South-East (ESE) direction is the zone of heavy air pollution. Site B, located at the same distance in the West represents the zone of moderate level of air pollution. Site C, located at about 6-7 km from the Power Station in the East-South-East (ESE) direction represents low level of pollution. Floristic survey of the vegetation growing in the vicinity of the Power Station was undertaken at three sites by traversing the area twice in two directions. Plants that were growing at Site A, Site B and Site C are given in Table 5. Ten plants species common to three sites were selected (Table 6), for studying changes in epidermal features in response to different levels of air pollution.



Pig. 1.1: Location of three Sites A, B. and C in Field Study.

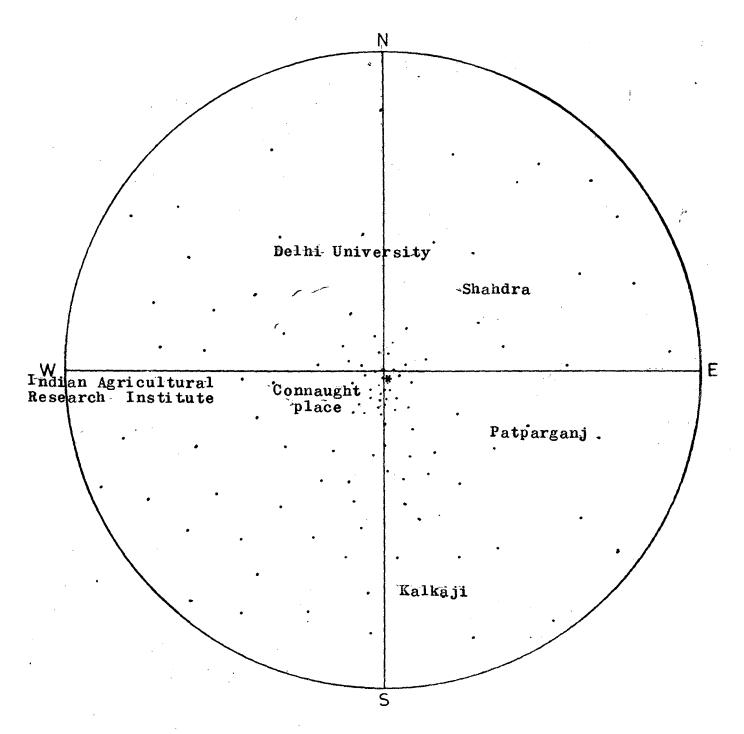
Site A: Zone of heavy pollution, at about 1 km. in the ESE. direction.

Site B: Zone of moderate pollution, at about 1 km. in the Western direction.

Site C: Zone of low pollution, at about 6-7 km. in the SE direction.

Experimental Design

Seedlings for experimental studies were raised in earthernware pots filled with equal quantities of loam soil. Pive plants namely Dolichus lablab, Lens culinaris, Phaseolus aureus, Cicer arietenum and Vigna sinensis were selected for study. Each species was grown in 16 pots, and five seeds were planted in each pot. Bight pots of each species (in all 40 pots) after seven days following germination, were transported from the Javaharlal Nehru University nursery, to a distance approximately 500-500 meters to the South of the Indraprastha Power Station (Fig. 1-11) (zone of maximum air pollution in the months of April and May ocillates between SSE to SW direction according to Padmanabhamurthy and Gupta, 1977). The second set having equal number of pots were maintained at the Jawaharlal Nehru University, The plants at both the locations were carefully watered with equal amount of water every day. Observations were made after 53 days, from the time the pots were transported to the Indraprastha Power Station with respect to grass morphology, growth, biomass and epidermal features, in the two sets of plants grown in polluted and non-polluted environments.



Pig. 1.ii: * Represents location of plants in experimental study.

(Plants were kept at about 500-600 meters in the Southern direction, which is zone of high concentration of pollutants in April-Nay).

Gross Morphology

Number of nodes and number of leaves per node were recorded for each species. Leaf area per node was calculated using graph peper. Leaves were closely examined for chlorosis, necrosis or any other foliar injury symptoms.

Biomass

Leaves for both polluted and non-polluted plants were plucked from every node for each plant. They were dried in an electric oven at 80°C for 24 hours. Electric eal balance was used for weighing and four sets of observations were taken for calculating average values. Similarly biomass of stem, root and fruit was taken for both polluted and non-polluted plants.

Cholorophyll Estimation

The total chlorophyll for both polluted and non-polluted plants was estimated according to the method given by Strain et al. (1966), one gram of leaf tissue was grinded in a mortar and pestle with a small quantity of acid washed sand in 80 per cent acetone. The optical density of the filtered extract was measured at 645 nm for chlorophyll and at 663 nm for chlorophyll b, the total amount of chlorophyll was calculated according to the following formula.

Total amount of chlorophyll

= 20.2
$$(D_{645}) + 8.02 (D_{663}) \times \frac{V}{1000 \times V}$$

where, D₆₄₅ - OD at 645 nm

D₆₆₃ - OD at 663 nm

V - Total volume of extract

W - Weight of leaf tissue taken.

Preparation of Epidermal Peels

Leaves for preparing epidermal peels were collected in the morning and brought to the laboratory. were thoroughly washed with distilled water using a soft camel hair brush. Epidermal peels of Brassica oleracea, Chenopodium album, Calotropis procera, Cynodon dactylon, Ricinus communis, Withania somnifera, Vigna sinensis and Lens culinaris were removed manually, for both abatial and adaxial surfaces. In case of Achyranthus aspera. Lantana camera, Sonchus asper, Dolichus lablab, Cicer arietenum, Phaseolus aureus, manual peeling of epidermis was found to be extremely difficult. To overcome this difficulty epidermal preparations were made following the technique described by Mohan Ram and Nayyar (1974). The leaf pieces were boiled in 2 ml. of 5 to 10 per cent aqueous cupric sulphate (CuSO, 5H, 0) solution, for 1 to to 2 minutes. It faciliates the seperation of epidermal layers. 4 ml. of concentrated hydrochloric acid was

added to the cupric sulphate solution and the material was again boiled for 1 to 2 minutes. This treatment dissolves out the unwanted tissues between the upper and the lower epidermis. After the separation of the epidermal layers the entire contents were poured into a petridish, and the fluid was decanted. Epidermal fragments were separated out and were washed 4 - 5 times with water to remove adhering debris. Temporary slides of epidermal peels were prepared using 20 per cent glycerine.

Light Microscopic Study

The epidermal preparations were examined under the light microscope (10 X x 40 X). Number of stomata per field view were counted. Similarly trichomes were also enumerated and their density was determined. The size of stomatal pore and trichome length were measured using occular micrometer which was caliberated with the help of a stage micrometer. At random six different microscopic fields were examined to determine the average density of stomata and trichomes.

Scanning Electron Microscopic Study

A. Sample preparation: Collected leaves were washed with double distilled water using soft camel hair brush After wiping the leaves with tissue paper, they were



Leaf sample was kept with adamial surface upward



A notch was made at the right hand side.



Another notch was made at the lower side keeping leaf in the same position. (Adaxial surface upward)

Fig. 2: A simple process for differenciating the adaxial leaf surface from abaxial, since it is necessary for scanning electron microscopic study. (Leaf sample with notches in the lower right hand side will represent the adaxial surface).

placed with their adaxial surface upward and leaf samples of 2 mm square were obtained. Two notches at the lower right hand side were made as shown in the Fig. 2. Thus, triangular gaps on the lower right hand side were helpful in identifying the adaxial leaf surface during subsequent processing.

B. Fixation: Leaf samples were fixed in 3 per cent glutalaidehyde (in phosphate buffer pH 7.2) post fixed in one per cent osmium tetraoxide (in phosphate buffer pH 7.2) Dehydrated in 35, 50, 85, 95, 99 per cent ethnol (three changes, five minutes each) placed in amyl acetate (15 min). They were kept on specimen holder with quickfix, followed by coating approximately 200 Å thick layer of silver by vacuum evaporation method under vacuum of 10⁻⁵ Torr. The preparations were scanned using a Cambridge Stereoscann Model S4-10 electron microscope. Scanning electron micrographs of stomata and trichomes were taken for both abaxial and adaxial leaf surfaces.

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CHAPTER - III

OBSERVATIONS

OBSERVATIONS

Field Study

Site A. B and C represent decreasing levels of air pollution (Fig. 1(11)). The number of species recorded at Site A. B and C were 19, 16 and 16 respectively (Table 5). Ten species, namely Achyranthus aspera, Brassica oleracea, Calotropis procera, Chenopodium album, Cynodon dactylon, Lantana Camera, Nerium indicum, Ricinus communis. Sonchus asper and Withania somnifera. common to the three sites were selected for a comparative study of their epidermal features (Table 6). In general, plants of Site A and B were stunted as compared to the plants of Site C. Plants at Site A and B had visual pollution injury symptoms (Table 7). The leaves of Sonchus asper and Ricinus communis were crumpled and necrotic with enrolled margins. In Achyranthus aspera and Brassica oleracea either leaves tips or leaf margins were yellow. Cynodon dactylon and Nerium indicum appear comparatively resistant as they were free from any visual injuries.

Epidermal Characteristics

Plants at Site C were regarded as control because this site was comparatively free from air pollution.

Morphology of stomata and trichome of Site C plants was studied for comparing the effect of air pollution of leaf

epidermis of Site A and B plants.

The number of stomata on the adaxial leaf surfaces varied from 34 in Calotropis process to 14 in Cynodon dactylon (Table 9). On the adaxial leaf surface, the stomata were less (11-43 per cent) as compared to the abaxial leaf surface.

On the basis of the arrangement of subsidiary cells six types of stomata, viz., anomocytic, anisocytic, diacytic, paracytic cyclocytic and tetracytic have been described in literature. The stomatal pattern of the selected plant is recorded in Table (8).

Achyranthus aspera, Chenopodium album, Sonchus asper and Withania sommifera have anomocytic type of stomata. Paracytic type of stomata are found in Calotropis procera, Nerium indicum and Ricinus communis, The stomata in Cynodon dactylon and Lantana camara are diacytic type, while in Brassica oleracea are anisocytic type.

The abaxial leaf surface in <u>Sonchus asper</u> was villose while the adaxial surface was pilose in nature (Table 8). The leaves of <u>Achyranthus aspera</u>, <u>Brassica oleracea</u> and <u>Lantana camara</u> were pilose on the abaxial surface and pubescent on the adaxial surface. Both the leaf surfaces in <u>Withania somnifera</u> and abaxial leaf

surface of Nerium indicum and Chenodpodium album were pubescent. Leaf surfaces of Calotropis procera and Ricinus communis, adaxial surface in Nerium indicum, and Chenopodium album and abaxial leaf surface of Cynodon dactylon were glabrescent.in nature. Adaxial leaf surface and in Cynoden dactylon was glabrous. Stomatal number was found to decrease from Site C to A (Table 9). The stomatal reduction on both leaf surfaces in Site A plants was 0-11 per cent more as compared to Site B The actual reduction varied from species to plants. species. Maximum stomatal reduction was observed in the adaxial leaf surface of Sonchus asper (36 per cent) followed by Brassica oleracea (28 per cent), Lantana camara (20 per cent). Ricinus communis (19 per cent), Achyranthus aspera (18 per cent), Chenopodium album (15 per cent), Winthania somnifera (10 per cent) and Calotropis procera (7 per cent) at Site A.

The abaxial leaf surface seems to be relatively resistant to air pollution in comparision to adaxial leaf surface (Table 9). Stomatal number was reduced in the following sequence: <u>Brassica oleracea</u> (18 per cent), <u>Sonchus asper</u> (15 per cent), <u>Lantana camara</u> (13 per cent).

A significant variation in the size of stomatal pore was observed. The length of stomatal pore in Site A plants was 0-13 per cent more as compared to Site B plants. The length of the stomatal pore on the adaxial leaf surface in Sonohus asper was 32 per cent less followed by Brassica oleracea (30 per cent), Calotropis procera (28 per cent), Chenopodium album (26 per cent), Withania somnifera (24 per cent), Ricinus communis (16 per cent). In Lantana camara, Achyranthus aspera and the reduction was about 10 per cent (Table 10).

Reduction in the length of stomatal pore at abaxial leaf surface was observed in the following sequence: Calotropis process (36 per cent), Chenopodium album (27 per cent), Lantana camara (23 per cent)

Brassica oleracea (22 per cent), Withania somnifera (21 per cent), Ricinus communis (20 per cent), Sonchus asper (18 per cent) and Achyranthus aspera (6 per cent) (Table 10).

The breadth of stomatal pore was significantly reduced on the adamial leaf surfaces in Site A plants (Table 11). Forty per cent reduction was observed in Achyranthus aspers and Brassica oleraces followed by Sonchus asper (38 per cent), Chenopodium album (28 per cent) and Ricinus communis (20 per cent). In Calotropis

procera, Ricinus communis and Withania somnifera only 0-14 per cent reduction was observed. The breadth of stomatal pore on the abaxial leaf surface seems to be less affected as compared to the adaxial leaf surface (2-14 per cent). There was no significant change in the breadth of stomatal pore on either leaf surfaces in the plants of Site A and B. In Cynodon dactylon, there was practically no variation in the stomatal characteristics at Site A and B.

the length of trichomes and their density on the adaxial leaf surface were found to be affected. Trichomes in most of the plants at Site A were denser (0-8 per cent) as compared to Site B plants except Ricinus communis. In general trichomes in Sonchus asper were most sensitive (Table 12). The density of trichomes in Sonchus asper was 50 per cent higher at Site A. The trichome density increased in Withania somnifera, Nerium indicum and Achyranthus asper at Site A by 35, 30 and 27 per cent respectively. In Calotropis procera, Chenopodium album, Brassica oleracea, Lantana camara and Ricinus communic increased trichome the density of trichomes was upto 25 per cent more at Site A.

The trichome density on the abaxial leaf surface did not vary much and in no case it was more than 22 per cent in comparison to Site C plants. Trichome density in Site A plants was 0-10 per cent higher than Site B plants except Calotropis process.

Maximum increase (61 per cent) in the trichome length was observed on the adaxial leaf surface in Sonchus asper at Site A. The length of trichomes in Nerium indicum, Ricinus communis, Withania somnifera and Calotropis procera at Site A increased by 44, 40, 35 and 30 per cent respectively. In Lantana camara, Achyranthus aspera, Brassica oleracea and Chenopodium album at Site A the increase in the trichome length was below 16 per cent. The data in Table—shows that trichome length at the abaxial leaf surface increased marginally (below 19 per cent). The trichome length increases more (0-14 per cent) in length on the adaxial leaf surface in Site A plants as compared to Site B plants (Table 13).

Chlorophyll Content

Plants at Site A and B varied in their chlorophyll content. Site A plants had comparatively less chlorophyll. Data on chlorophyll content are summerized in Table 14. Chlorophyll reduction varied from 32.3 per cent in Sonchus asper to 7.2 per cent in Nerium indicum.

Scanning Electron Microscopic Study

Figs. 3 and 4 represent scanned micrographs of adaxial leaf surface of <u>Brassica oleracea</u> growing at Site C and A respectively. The size of the stomatal pore in plant growing at Site A is small (Fig. 4) in comparison with the plants of Site C (Fig. 3). In the later case, the boundary of stomatal pore is comparitively smooth (Fig. 3). The subsidiary cell complex in plants of Site A show numerous folds which are absent in Site B plants.

Figs. 5 and 6 show scanned micrographs of stomata of <u>Withania somnifera</u> leaves from Site C and A respectively. The size of the stomatal pore is small in Site A plants (Fig. 6) in comparison with Site C plants (Fig. 5). In the later case, the stomata appears to be in a groove with comparitively smooth pore boundary. Rugged surface of subsidiary cells in Site A plants (Fig. 6) is more prominant.

Pig. 7 and 8 represent the scanned micrographs of trichomes at adamial leaf surface in <u>Lantana camara</u> from Site C and Site A. Trichomes in Site A plants are much longer (Fig. 8) in comparison with plants of Site C and at the same time trichomes at the former Site were broken.

Fig. 3: Scanned electron micrograph of adaxial leaf surface of <u>Brassica oleracea</u> growing at control site. (x2200)

Fig. 4: Scanned electron micrograph of adaxial leaf surface of <u>Brassica oleracea</u> growing at polluted site. (x2200)

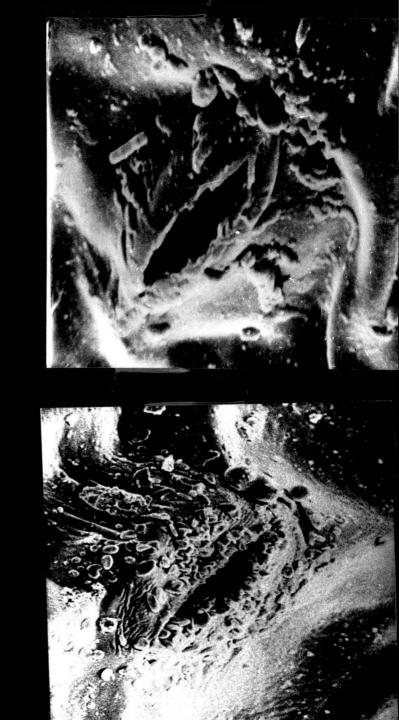


Fig. 5: Scanned electron micrograph of adaxial leaf surface of <u>Withania somnifera</u> growing at control site. (x2100)

Fig. 6: Scanned electron micrograph of adaxial leaf surface of <u>Withania somnifera</u> growing at polluted site. (x2100)

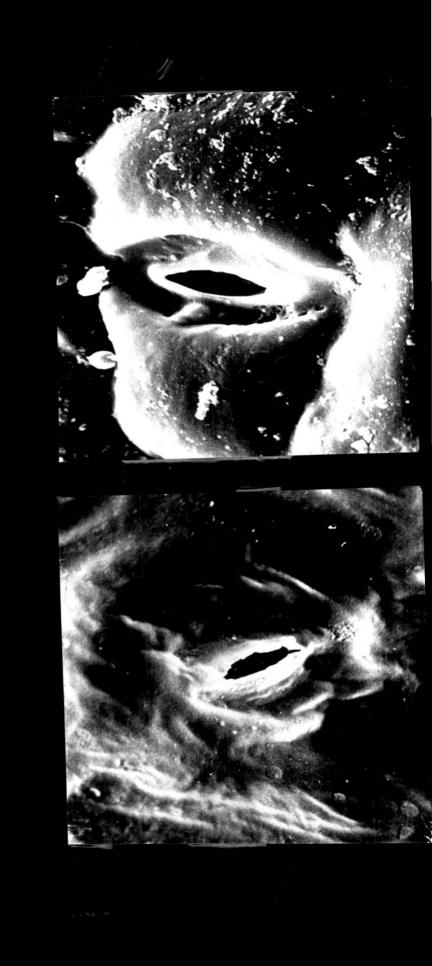


Fig. 7: Scanned electron micrograph of adaxial leaf surface of Lantana camara growing at control site. (\times 56)

Fig. 8: Scanned electron micrographs of adaxial leaf surface of Lantana camara growing at polluted site (x 52)

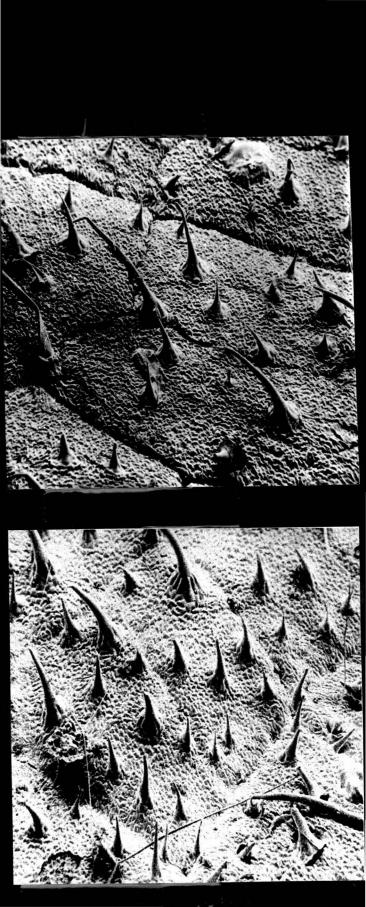


Fig. 9: Scanned electron micrograph of adaxial leaf surface of Sonchus asper growing at control site. (x120)

Fig. 10: Scanned electron micrograph of adaxial leaf surface of Sonchus asper growing at polluted site. (×2/2)



Fig. 9 and 10 show the scanned micrographs of Sonchus asper leaf surface from Site C and A respectively. A significant increase in the size of the trichomes in plant growing at Site A is quite prominant.

Experimental Study

Experiments were designed to compare the effects of air pollution observed in the field observations. Two sets of plants were grown in identical soil for 53 days following germination at polluted and non-polluted Sites. At the conclusion of the experiment, the gross morphology of the plants including number of leaves at each mode (Table 14) leaf area and leaf biomass (Tables 15, 17, 19, 21 and 23) and total plant biomass (Tables 16, 18, 20, 22 and 24) as well as leaf injury and their epidermal characteristics were observed.

Table 15 gives the number of leaves at each of the first three nodes, the number of leaves were same in both sets of plants. The leaf number, however, decreased gradually, beyond third node as a result the total number of leaves at the conclusion of the experiment were less in plants grown at polluted site. In <u>Dolichos lablab</u>, reduction in number of leaves was 33 per cent, followed by <u>Phaseolus aureus</u> (29 per cent), <u>Lens culanaris</u> (27 cent) and <u>Cicer arietenum</u> (25 per cent). <u>Vigna sinensis</u>

seems to be comparitively resistant to air pollution since it showed only 9 per cent reduction.

Data regarding leaf area and leaf biomass for the two sets of plants are given in Tables (16, 18, 20, 22 and 24). Reduction in the leaf area was in the following sequence: Cicer arietenum (54 per cent), Dolichos lablab (44 per cent), Phaseolus aureus (40 per cent), Lens culinaris (39 per cent) and Vigna sinensis (35 per cent). The changes in leaf biomass also followed more or less the same pattern - Dolichos lablab (59 per cent), Cicer arietenum (50 per cent), Phaseolus aureus (49 per cent), Lens culanaris (42 per cent) and Vigna sinensis (30 per cent).

The total plant biomass was significantly affected by air pollution (Tables 17, 19, 21, 23 and 25).

In <u>Dolichos lablab</u> it was reduced by about 52 per cent followed by <u>Cicer arietenum</u> (51 per cent), <u>Lens culinaris</u> (49 per cent), <u>Phaseolus aureus</u> (46 per cent) and <u>Vigna sinensis</u> (36 per cent).

Visual Injuries

Visual injuries in the leaves of plants grown at polluted sites were observed (Table 26). Leaf injuries such as necrotic patches, chlorosis, dead intervenial tissue and enrolled leaf margins were observed in <u>Dolichos</u>

<u>lablab</u>. In <u>Cicer arietenum</u>, <u>Lens culinaris</u> and <u>Phaseolus</u>

<u>aureus</u> injury symptoms appear at later stages suggesting

that they are comparatively more resistant. <u>Vigna</u>

<u>sinensis</u> was free from any injury.

Epidermal Characteristics

Epidermal features including type of stomata and trichome morphology (Table 27) in plant grown at non-polluted site were studied for evaluating the effect of air pollution.

Cicer arietenum has pilose leaf surface while in Dolichos lablab, Lens culinaris and Phaseolus aureus leaf surfaces are pubescent. The leaf surfaces of Vigna einensis are glabrescent. Number of stomata in plants grown under experimental study in control set of plants varies from 18 in Phaseolus aureus to 7 in Lens culinaris on adaxial leaf surface. Abaxial leaf surface in these plants have more stomata (25-43 per cent)(Table 28). Plants species used in the experimental study have paracytic type of stomata in Cicer arietenum, Dolichos lablab, Phaseolus aureus and Vigna simensis while anisocytic type in Lens culinaris.

The number of stomata on adaxial leaf surface in Cicer arietenum was reduced by 34 per cent (Table 28).

Phaseolus aureus (29 per cent) and Dolichus lablab (27 per

cent) also exhibited considerable reduction. In <u>Lens</u> <u>culinaris</u> and <u>Vigna sinensis</u>, they were reduced by 19 and 15 per cent respectively.

The abaxial leaf surface do not respondequally to air pollution as in no case the reduction in the number of stomata was more than ten per cent.

The length of stomatal pore was also found to be affected by air pollution (Table 29). In <u>Cicer arietinum</u> 42 per cent reduction was observed followed by <u>Dolichus lablab</u> (36 per cent) and <u>Lens cullinaris</u> (28 per cent). In <u>Vigna sinensis</u>, it was reduced by 10 per cent only. It is interesting to note that reduction in the length of stomatal pore on abaxial leaf surface in most of the plants was almost half of the reduction on the adaxial surface.

The breadth of stomatal pore on both leaf surfaces also appear to be influenced by air pollution. The breadth of stomatal pore on the adaxial surface was reduced by 44 per cent in <u>Dolichos lablab</u> and <u>Phaseolus aureus</u> (Table 30). Lens culinaris and <u>Cicer arietenum</u> it was 31 and 30 per cent respectively. 13 per cent decrease was observed in <u>Vigna sinensis</u>. Breadth of stomatal pore on the abaxial leaf surface was found to decrease in the following sequence: <u>Dolichos lablab</u> (37 per cent), <u>Phaseolus aurous</u>

arietenum (14 per cent) and Vigna sinensis (12 per cent).

Appreciable differences were also observed in the length and density of trichomes particularly at the ada-

On the adaxial leaf surface, the trichome density
was 46 per cent higher in <u>Cicer arietenum</u> followed by

<u>Phaseolus aureus</u> and <u>Dolichos lablab</u> (44 per cent),

<u>Lens culinaris</u> (33 per cent) and <u>Vigna sinensis</u> (33 per cent)(Table 31). On the abaxial leaf surface the increase in the density of trichomes was below 18 per cent.

Clearly visible effects were observed so far as length of trichomes of abaxial leaf surface are concerned (Table 31). The length of trichome on the adaxial leaf surface in <u>Dolichos lablab</u> increase considerably (50 per cent), followed by <u>Phaseolus aureus</u> (per cent), <u>Cicer arietenum</u> (31 per cent), <u>Lens culinaris</u> (28 per cent) and in <u>Vigna sinensis</u> (12.5 per cent). The length of trichome did not change much on the abaxial leaf surface in all species (Table).

Chlorophyll Content

Data on the amount of total chlorophyll in plants grown in polluted and control environments are given in Table 33. It was 37.8 per cent less in <u>Cicer arietenum</u>

followed by <u>Phaseolus aureus</u> (31.4 per cent) and <u>Vigna</u>
sinensis (11.1 per cent).

Electron Microscopic Study

The scanned micrographs (Figs. 11-12) of the stomata at the abaxial leaf surface of Chenopodium album show that the size of the stomatal pore was less in plants of polluted environment (Fig. 12). The edge of stomatal pore in plants of polluted environment (Fig. 14) appear uneven.

Figs. 13 and 14 are the scanned micrographs of stomata of abaxial leaf surface in <u>Dolichus lablab</u> from non-polluted and polluted environment. In polluted environment, stomatal pore size becomes considerably small (Fig. 14).

Figs. 15 and 16 represent scanned micrographs of adaxial leaf surface of <u>Gicer arietenum</u> grown in non-polluted and polluted environment. The density of trichomes has considerably increased in plants grown in polluted environment (Fig. 16).

A comparative study of the scanned micrographs of plants grown in polluted and non-polluted environment reveals that the air pollution reduces the size of stomatal pore, promote folding of the subsidiary cell surface and increase the density of trichomes.

Fig. 11: Scanned electron micrographs of abaxial leaf surface of Chenopodium alba growing at control site. (x2200)

Fig. 12: Scanned electron micgograph of abaxial leaf surface of Chenopodium alba growing at polluted site. (×2200)





Fig. 13: Scanned electron micrograph of abaxial leaf surface of <u>Dolichus lablab</u> grown in control stte. (x2100)

Fig. 14: Scanned electron micrograph of abaxial leaf surface of <u>Dolichus lablab</u> grown in polluted Site. (x 2/00

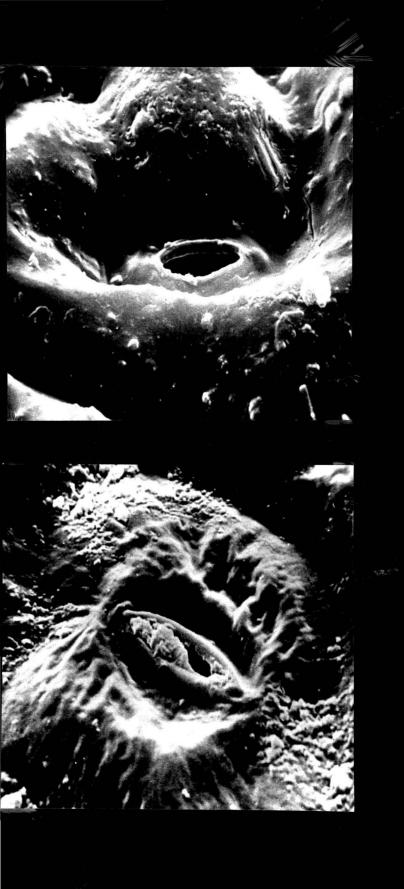


Fig. 15: Scanned electron migrograph of adaxial leaf surface of <u>Cicer arietenum</u> grown in control xxxxxxxx site. (x53)

Fig. 16: Scanned electron micrograph of adaxial leaf surface of <u>Cicer arietenum</u> grown in polluted site. (×53)

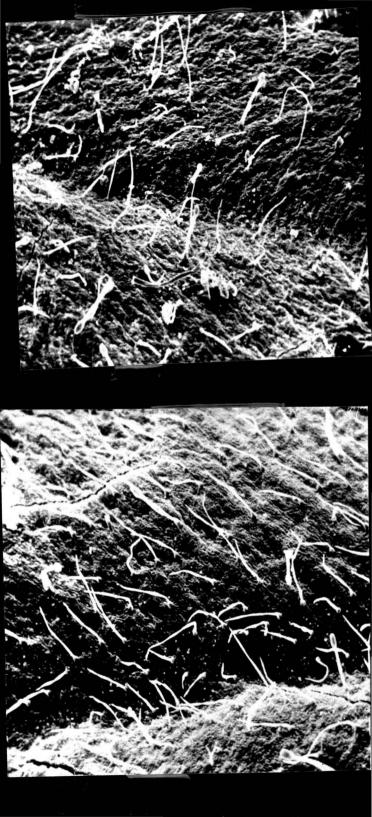


Table 5
List of Plants of Site A, B and C.

S.N.	Site A	Site B	Site C
1.	Achyranthus aspera	Acacia` arabica	Achyranthus aspera
2.	Altheca rosea	Achyranthus aspera	Brassica oleracea
3.	Amaranthus spinosus	Amaranthus spinosus	Calotropis procera
4.	Amerenthus viridis	Brassica oleracea	Chenopodium labum
5.	Brassica oleracea	Calotropis procera	Croton bonplandianu
6.	Calotropis procera	Chenopodium album	Cynodon dectylon
7.	Cassia fistula	Croton bonpladianum	Eucalyptus globulus
8.	Chenopodium album	Cynodon dactylon	Lantana camara
9 •	Croton bonplandianum	Eucalyptus globulus	Nerium indicum
10.	Cynodon dactylon	Lantana camara	Parkinsonia aculeat
11.	Eucalyptus globulus	Nerium indicum	Ricinus communis
12.	Lantana Camara	Ricinus communis	Sida alba
13.	Lathyrus aphaca	Sida alba	Sonchus asper
14.	Nerium indicum	Sonchus asper	Vernonia cinerea
15.	Parkinsonia aculeata	Vernonia cinerca	Withania somnifera
16.	Ricinus communis	Withania somnifera	Xanthium strumarium
17.	Sonchus asper		
18.	Vernonia cinerea	•	
19.	Withania somnifera	•	
		•	

Table 6
List of Plants Selected For Detailed Study

1.	Achyranthus aspera
2.	Brassica oleracea
3.	Calotropis procera
4.	Chenopodium album
5.	Cynadon dactylon
6.	Lantana camara
7.	Nerium indicum
8.	Ricinus communis
9.	Sonchus asper
10.	Withania somnifera

Table 7

Visual Symptoms Observed in Plants Common to Site A, B and C

Achyranthus asper: Yellowing of leaf margin gradually extending towards the center and ultimately ending in a crumpled leaf.

Young leaf develop bright red color along Veins and Veinlets.

Brassica oleracea: Chlorosis with lesions of brown color.

Crumpled leaves deformattes can be seen
in young leaves.

Calotropis process: Yellow strips on the leaf, tip of the leaf turning brown from yellow and at last becoming brittle.

Chenopodium album : Chlorosis of leaf.

Cynodon dactylon: No visual symptoms were observed.

Lantana camara : Rolling of leaf margin, crumpled leaves.

Nerium indicum : No visual symptoms were observed.

Ricinus communis : Necrosis, Inteveinal tissues killed.

Sonchus asper : Necrosis, crumpled leaves followed by death of most of the leafy tissue at later stages.

Withania somnifera: Chlorosis, brown patches were also observed.

Table & Bridermal Morphology of Plants Common to Site A, B and C

Species	Stomata type		Trichome Characteristics
Achyranthus aspera	Anomocytic	Adaxial: Abaxial:	and straight hairs.
			with long soft hairs.
Brassica oleracea	Ani socy tic	Adaxial	Pubescent: Short, soft and straight hairs.
		Abaxial	Pilose: Thinly covered with Long soft shirs.
Calotropis procera	Paracytic	Adaxial Adaxial	Glabrescent: Short hairs Glabrescent: Short hairs
Chenopodium album	Anomocy tic	Adaxial	Glabrescent: Smooth and shining surface.
		Abaxial	Pubescent: Short soft and straight hairs.
Cynodon dactylon	Diacytic	Adexial	Glabrous: Smooth surface free from heir.
		Abaxial	Glabrescent: Short hairs.
Lantana camara	Diacytic	Adaxial	Pubescent: Short, soft and straight hairs.
	e e	Abaxi el	Pilose: Thinly covered with comparitively long soft hairs.
Nerium i indicum	Paracytic	Adaxial Abaxial	Glabrescent: Short hairs. Pubescent: Short, soft, and straight hairs.
Ricinus communis	Paracytic	Adaxial Abaxial	Glabrescent: Short hairs. Glabrescent: Short hairs
Sonchus asper	Anomocy tic	Adaxial	Pilose: Thinly covered with long soft hairs.
.		Abaxial	Villose: Thickly covered with long soft hairs.
Withania somnifera	Anomocy tic	Adexial	Pubescent: Short, soft and straight hairs
		Abaxial	Pubescent: Short, soft and straight bairs.

Table 9 Effect of Air Pollution on Stomatal Density (Number/field view)* $(\bar{x}_{\pm}\sigma)$ **

Species	Surface	Site A	Site B	Site C
			1.9	and the second second
Achyranthus aspera	Abaxial	,	14 ± 1.0 16 ± 2.4	
		******	11 ± 1.7	
Brassica oleracea			12 ± 2.0	
Calaturate amount	Adexial	27 ± 2.4	26 ± 2.6	29 ± 4.1
Calotropis procera	Abexial	34 ± 4.1	33 ± 3.6	34 ± 4.4
Chenopodium album	Adaxial	16 ± 2.4	16 ± 3.1	19 ± 2.6
	Abaxial	21 ± 3.7	23 ± 4.1	25 ± 3.1
Cynodon dactylon	Afordal	8 ± 0.2	8 ± 0.7	8 ± 1.2
	Abaxi al	14 ± 1,2	14 ± 1.3	14 ± 1.8
Lantana camera			16 ± 1.3	
	Abaxi al		28 ± 2.9	
Ricinus communis	Adaxial Abaxial		17 ± 1.9	
•			32 ± 3.8	
Sonchus asper			10 ± 1.2 17 ± 2.1	103102
·			17 ± 2.5	
Withania somnifera		The state of the s	24 ± 3.8	

^{* 10}xXX 40x

^{** 🕏 -} Mean

^{~ -} Standard deviation.

Table 10 Bifect of Air Pollution on Length (u) of Stomatal Poza (\$ ± ~)*

Species	Surface	Site A	Site B	Site C
Achyranthus aspera	Adaxial	21 ± 1.9	21 ± 2.1	24 ± 2.4
	Abaxi al	17 ± 1.6	17 ± 1.9	18 ± 2.5
Para and an analysis of the second	Adexial	9 ± 0.5	9 ± 0.6	13 £ 0.9
Brassica oleracea	Abaxial	7 ± 0.3	7 ± 0.4	9 ± 0.6
	Adaxial	12 ± 2.1	13 ± 1.5	17. ± 2.2
Calotropis procera	Abaxial	9 ± 1.3	10 ± 1.2	14 ± 2.9
Abanan Alam Abana	Adaxial	11 ± 0.5	11 ± 0.6	15 ± 0.9
Chenopodium album	Abaxial	8 ± 0.4	8 ± 0.3	11 ± 0.7
Commendate distriction	Adamial	12 ± 0.7	12 ± 0.9	12 ± 1.2
Cynodon dactylon	Abaxial	7 ± 0.2	6 ± 0.5	7 ± 1.2
	Adaxial	15 ± 2.2	16 ± 1.9	19 ± 2.7
Lantana camara	Abaxial	10 ± 1.9	11 ± 0.9	13 ± 2.9
	Adaxial	21 ± 3.1	23 ± 2.8	25 ± 3.7
Ricinus communis	Abaxial	12 ± 1.9	13 ± 2.1	15 ± 2.7
On the state of th	Adaxial	15 ± 1.7	16 ± 2.1	22 ± 2.3
Sonchus asper	Abaxial	9 ± 0.8	9 ± 1.2	11 ± 1.7
5/4 Ab., and	Adaxial	14 ± 1.1	13 ± 2.1	15 ± 2.7
Withania somnifera	Abaxial	11 ± 1.9	12 ± 3,4	14 ± 3.2

Mean Standard deviation.

Table 11 Effect of Air Pollution on Breadth (u) of Stomatal Pore $(\bar{x}_{\pm} -)^*$

Species	Surface	Site A	Site B	Site C
	Aderial	1.5± 0.3	1.5± 0.2	2.5± 0.4
Achyranthus aspera	Abaxial	1.0± 0.1	1.0 ± 0.2	1.540.3
.	Adaxial	1.6± 0.2	1.5± 0.25	2.5±0.3
Brassica oleracea	Abaxial	1.0± 0.1	1.0± 9.1	1.5± 0.2
	Adexial	3.0± 0.4	3.0± 0.2	3.5± 0.5
Calotropia procera	Abaxial	2,52 0.3	2.5± 0.3	2.5± 0.4
	Ada×1 al	2.5± 0.2	2.5± 0.3	3.5± 0.2
Chenopodium album	Abaxial	2.0 ± 0.1	2.0± 0.2	2.5± 0.1
	Adaxi al	3.0± 0.1	3.0± 0.2	3.0± 0.4
Cynodon dactylon	Abaxial	2.0± 0.2	2.04 0.2	2.0± 0.4
	Adexial	3.5± 0.7	3.5± 0.6	4.5 <u>+</u> 0.8
Lantana camara	A Abarial	2.0± 0.5	2.010.4	2.5± 0.6
	Adaxi al	4.3± 0.5	4.5± 0.4	5.0± 0.6
Ricinus communis	Abaxial	4.0± 0.3	4.0± 0.4	4.5± 0.2
	Adaxial	2.5± 0.3	2.5± 0.4	4.0 <u>+</u> 0.6
Sonchus esper	Abaxial	1.5± 0.2	1.5± 0.3	2.0± 0.5
	la ixabA	4.4± 0.3	4.5± 0.5	5.0± 0.7
Withania somnifera		<u>-</u> .	3.5± 0.3	

^{*} Ī - Mean

⁻ Standard deviation.

Table 12

Effect of Air Pollution on the Density (Number/u²)

of Trichome ($\bar{x} \pm \sigma$)*

Species	Surface	Site A	Site B	Site C
A set to a construct the base of the set of the set	Adaxial	140± 23.2	140± 18.7	110± 12.2
Achyranthus asper	Abexial	162± 27.2	162± 29.2	142± 17.1
Brassica oleracea	Adexial	140± 29.2	132± 26.7	115± 19.7
	Abaxial	180± 31.8	170± 21.9	158± 21.8
6-1-4	Adaxial	70± 6.9	70± 5.2	60± 4.6
Calotropis procera	Abexial	80± 8.8	85± 8.1	75± 5.9
Chenopodium album	Adaxial	30± 6.7	26± 2.4	24± 7.7
	Abaxial	110± 11.9	100± 13.8	92± 11.2
One and a second and a second	Adoxial			
Cynodon dactylon	Abaxial	31± 9.2	23± 7.8	28± 8.1
Lantana camara	Adaxial	220± 31.4	220± 26.4	190± 21.2
nantana Camara	Abaxi al	245± 34.4	245± 31.2	220± 21.7
Nerium indicum	Adexial	85± 26.7	64± 22.4	65± 17.2
MCLIAM LAGICAM	Abexial	145± 25.4	145± 23.8	135± 21.2
Ricinus communis	Adaxi al	70± 6.6	75± 8.2	60± 5.2
	Abaxial	80± 8.1	80± 7.0	75± 6.0
Sonchus asper	•		210± 16.8	
A STATE OF THE STA	Abaxial	240± 27.3	240± 12.2	210± 16.2
Vithania somnifera	Adaxi al	100± 7.9	105± 9.2	75± 5.9
er um at entretanten für philat mentem en 15 de 15	Abaxial	105± 8.2	100± 9.0	85± 7.2

^{*} x - Mean

^{- -} Standard deviation

Table 13 Bffect of Air Pollution on the Length (u) of Trichomes $(\bar{x} \pm \sigma)^*$

Species	Surface	Site A	Site B	Site C
	Adaxial	125 <u>±</u> 27.8	122±25.2	110± 9.6
Achyranthus asper	Abexial	152±16.8	252±21.2	146±11.4
Dronnino e lavores	Adexial	110±11.8	100± 9.7	95± 8.9
Brassica oleracea	Abaxial	162± 9.6	157±17.4	150±12.8
Calotropis procera	Adaxial	60± 8.4	65± 4.9	45± 3.2
cerottobre brocere	Abaxial	50±5.9	50± 3.7	50± 4.0
Chenopodium album	Adaxial	13± 3.4	12 <u>±</u> 8.2	112± 1.9
CHANG PAGITUR ST DAW	Abaxial	100±16.2	95±11.2	87± 7.2
Cynodon da oty lon	Adexial	*	***	-
cymodon daetyron	Abaxi al	42± 9.1	39± 7.8	39± 8.4
Lanthana camara	Ağaxi al	130±22.6	150424.2	110 <u>+</u> 16.1
	Abaxial	182±29.9	182±23.8	170±11.8
Nerium indicum	Adaxial	72 <u>±</u> 21.2	69±19.7	50± 8.9
and the file and anti-man. We start the first the start things	Abaxial	112224.1	104±22.1	94±13.8
Ricinus communis	Adoxial	70± 6.1	65± 5.9	50± 4.6
THE WASHINGTON WITH STATE STATE OF	Abaxial	70± 6.8	75生 7.4	654 6.8
Sonchus asper	Adaxial	245±39.6	238±34.7	152±12.4
onwhites stakes.	Abdrial	200±29.6	193 <u>±</u> 35.8	179±18.7
Withania somnifera	Adaxial	100± 9.4	90± 7.8	75± 6.9
ne anomer commerce	Abmial	95±13.9	90± 9.6	80± 7.2

^{*} x - Mean

⁻ Standard deviation

Table 14

Effect of Air Pollution on the Total Chlorophyll Content (mg/gm)

Species	Site A	Site B	Site C
Achyranthus aspera	3.037	3.625	3.835
Brassica oleracea	1.774	2.024	2.429
Calotropis procera	2.914	3.214	3.312
Chenopodium album	2.542	2.812	2.872
Cynodin dactylon	1.701	1.818	1.852
Lantana camara	3.671	3.979	4.507
Nerium indicum	3.364	3.582	3.625
Ricinus communis	1.692	1.812	1.972
Sonchus asper	2.163	2.752	3.404
Withania somnifera	2.831	3.101	3.246
			•

Table 15

The Growth of Leaves in Plants Grown in the Polluted and Control Environment

(Observations were made after 53 days, values represent average of five plants)

No. of			Number of leaves							
Nodes	-Dol	ichos C	Lei	ns C	Phase	olus C	P	Cer	- <u>Vi</u>	gna C
							- X			
I	2	2	2	. 2	2	2	1	1	2	2
II	2	2	1-2	2	1	1	1	1	2	2
III	2	2-4	1	2	1	1	1	1	1	2
IV	2-4	4	1	1	1	1	•	1	1	2
V	***	2	. 1	1	***	3	•		1	1
VI .			•	1,	*	1		٠.	1	1
VII			ş ,				•		i	1
VIII				g	à				1	. 1
IX.	•	•			••	y.				1
No.of	8-10	12-14	6-7	9	5	7	3	4	10	11

Table 16

Development of Leaf Area (cm²) and Leaf Biomass (gm) in

<u>Dolichos lablab</u> Grown in Polluted and Control Area

No. of	Les	f area	Leaf biomass		
Nodes	Polluted	Control	Polluted	Control	
1	39 ± 2.9	52 ± 3.6	0.130±0.012	0.220±0.022	
11	33 ± 2.8	61 ± 4.2	0.120±0.009	0.245±0.018	
111	62 ± 4.2	124 ± 9.7	0.239±0.018	0.502±0.103	
IV	113 ±11.7	203±16.4	0.41240.113	0.807±0.970	
₩ :	27 ± 4.9	46 ± 7.8	0.086±0.008	0.181±0.012	
Total	274_+16.2	486 ±24.7	0.88740.160	1.955±0.252	

Table 17

Effect of Air Pollution on the Biomass (gm)

Dolichos lablab

Plant organ	Polluted	Control		
Leaf	0.887 ± 0.160	1.955 ± 0.252		
Stem	0.720 ± 0.013	1.132 ± 0.021		
Root	0.113 ± 0.016	0.490 ± 0.023		
Total	1.710 ± 0.189	3.577 ± 0.296		

Table 18

Development of Leaf Area (cm²) and Leaf Biomass (gm) in
Phaseolus aureus Grown in Polluted and Control Areas

No. of Nodes	Leaf a	Leaf area Leaf area		Leaf biomass	
	Polluted	Control	Polluted	Control	
I	56 ±3.2	75 ± 4.60	0.180 <u>+</u> 0.008	0.350±0.012	
II	14 ±2.9	28 ±3.30	0.078±0.004	0.130±0.012	
III.	7.5±0.7	22 ± 3.90	0.055±0.003	0.092±0.007	
IV	4.0±0.35	14 ± 2.60	0.043±0.003	0.070±0.007	
V	**	8.0± 0.90	•	.0.070±0.007	
VI :	***	3.2± 0.4		0.010±0.002	
Total	81.5 <u>±</u> 1.01	150.2±15.70	0.356±0.018	0.687±0.049	

Table 19

Effect of Air Pollution on the Biomass (gm)

of Phaseolum aureus

Plant organ	Polluted	Control
Leaf	0.356 ± 0.018	0.687 ± 0.049
Stem	0.315 ± 0.007	0.775 ± 0.026
Root	0.080 + 0.004	0.192 ± 0.008
Fruit	***	0.034 ± 0.003
Total	0.751 ± 0.029	1.388 ± 0.086

Table 20

Development of Leaf Area (sq.cm) and Leaf Biomass (gm) in
Lens culinaris Grown in Polluted and Control Areas

No. of	Leaf area		Leaf biomass	
Nodes	Polluted	Control	Polluted	Control
I	38.0± 3.6	47.0± 6.2	0.100±0.006	0.140±0.009
11	29.0± 2.8	49.04 4.1	0.075±0.006	0.145±0.011
III	23.0± 2.9	62.0± 4.6	0.055±0.007	0.170±0.009
IV	20.0+ 2.1	24.0± 1.9	0.045±0.006	0.06540.011
V	16.5± 0.6	16.5± 1.2	0.015±0.002	0.042±0.005
VI	***	8.2± 1.2		0.021±0.003
Total	126.5 <u>±</u> 11.9	206.7±19.2	0.290 <u>±</u> 0.027	0.583±0.048

Table 21

Effect of Air Pollution on the Biomass (gm)

of Lens culinaris

Plant organ	Polluted	Control
Leaf	0.290 ± 0.027	0.583 ± 0.048
Stem	0.340 ± 0.012	0.600 ± 0.022
Root	0.135 ± 0.008	0.195 ± 0.011
Fruit		0.055 ± 0.013
Total	0.765 ± 0.047	1.433 ± 0.094

Table 22

Development of Leaf Area (sq.cm) and Leaf Biomass (gm) in

<u>Cicer arietenum</u> Grown in Polluted and Control Areas

No. of	Leaf	area	Leaf	Biomass
Nodes	Polluted	Control	Polluted	Control
1	21.5± 3.5	34.5± 4.2	0.130±0.012	0.225±0.017
II	16.5± 2.3	29.0± 3.7	0.11040.012	0.205±0.008
III	10.0± 1.3	24.5± 1.6	0.070±0.007	0.170±0.012
IV	•	15.0± 1.2	₩	0.110±0.012
Total	47 ± 7.1	103 ±10.7	0.310±0.031	0.710±0.049

Table 23

Effect of Air Pollution on the Biomass (gm)

of <u>Cicer arietenum</u>

Plant organ	Polluted	Control
Leaf	0.310 ± 0.031	0.710 ± 0.049
Stem	0.020 ± 0.003	0.035 ± 0.006
Root	0.060 ± 0.009	0.085 ± 0.006
Total	0.390 ± 0.043	0.820 ± 0.061

3

Table 24

Development of Leaf Area (cm²) and Leaf Biomass (gm)
in <u>Vigna Sinensis</u> Grown in Polluted And Control Areas

No. of	Lea	I area	Leaf	biomass _
Nodes	Polluted	Control	Polluted	Control
1	57 ± 3.4	55 ± 4.7	0.470±0.021	0.340±0.031
II	59 ± 2.3	58 ± 4.1	0.525±0.019	0.350±0.014
III	82 ± 3.2	135 ± 5.6	0.560+0.019	0.780+0.032
IV	86 ± 2.9	144 ± 3.9	0.590±0.015	0.820±0.018
V	100 ± 2.7	127 ± 5.6	0.670±0.012	0.730±0.019
VI	97 ± 2.4	134 ± 3.1	0.660±0.012	0.810+0.016
VII	85 ± 2.9	142 ± 3.9	0.570±0.018	0.815±0.013
VIII	68 ± 2.4	120 ± 3.1	0.560±0.015	0.690±0.028
IX	•	42 ± 1.9	•	0.250±0.194
Total	637 <u>+</u> 22,2	957 ±35.5	4.755±0.127	6.585±0.194

Table 25

Effect of Air Pollution on the Biomass (gm)
of <u>Vigna Binensis</u>

Plant organ	Polluted	Control
Leaf	4.755 ± 0.127	6.585 ± 0.194
Stem	7.110 ± 0.212	11.780 ± 0.310
Root	0.525 ± 0.014	0.940 ± 0.047
Total	12.390 ± 0.353	19.305 ± 0.551

Table 26
Visual Injuries in Plants Grown for Experimental Study
Near Indraprastha Power Station

Cicer arietenum:	Chrosis, yellowing of leaf tip.
Dolichos lablab:	Necrotic patches, intervenial tissues killed, rolling of margins.
Lens culinaris:	Light green patches, chloses in later stages.
	Yellowing of leaf margin, extending towards the center, necrosis.
Vigna sinensis:	No visual symptoms on leaf.

Table 27
Epidermal Morphology of Plants Grown for Experimental Study
Near Indraprastha Power Station

Species .		Trichome characteristic	Stomata type
	Adaxi al	Pilose: Thinly covered with long soft hairs.	Paracytic
Cicer arietenum	Abaxial	Pilose: Thinly covered with long soft ahirs.	
B. 14	Adaxi al	Pubescent: Short, soft and straight hairs.	*
Dolichos lablab	Abaxiel	Pubescent: Short, soft and straight hairs.	Paracytic
	Adaxi al	Pubescent: Short, soft and straight haris.	
Lens culinaris	Abaxial	Pubescent: Short, soft and straight hairs.	Ani socy ti
	Adaxial	Pubescent: Short, soft and straight hairs	
Phaseolus aureus	Abaxial	Pubescent: Short, soft and straight hairs.	Paracytic
. 7	Adexial	Glabrescent: Smooth and shining surface	
Vigna sinensis	Abexial	Glabrescent: Smooth and shining surface.	Paracytic

Table 28

Density of Stomata in Plants Grown in Polluted and Control Environments (Number/field view*)($\bar{x} \pm \sigma$)**

Spacies		Polluted	Control
	Adaxial	8 ± 1.1	12 ± 2.6
Cicer Arietenum	Abaxial	13 ± 2.2	14 ± 1.6
	Adexial	8 ± 0.9	11 ± 2.1
Dolichos lablab	Abaxi al	14 ± 1.7	16 ± 1.9
	Adaxial	13 ± 1.5	16 ± 2.4
Lens culinaris	Abaxi al	23 ± 2.6	26 ± 2.7
	Adaxial	10 ± 3.2	14 ± 3.8
Phaseolus aureus	Abaxial	17 ± 1.6	19 ± 2.1
	Adaxial	18 ± 1.8	21 ± 2.6
Vigna sinensis	Abaxial	27 ± 3.1	29 ± 3.4

^{* 10}x X 40x

^{**} x - Mean

Standard deviation.

Table 29

Length of Stomatal Pere (u) in Plants Grown in Polluted and Control Environments ($\bar{x} \pm \sigma$)*

Species	Mary St. Annual Million and Additional Conference on the Conferenc	Polluted	Control
	Adexial	7 ± 0.4	12 ± 0.5
Cicer arietenum	Abaxial	7 ± 0.3	9 ± 0.4
	Adaxial	20 ± 0.7	27 ± 1.1
Dolichos lablab	Abaxial	14 ± 0.8	17 ± 1.2
	Adaxial	13 4 0.12	17 ± 0.7
Lens culinaris	Abaxial	8 ± 0.05	9 ± 0.0
	Adaxial	10 ± 0.9	14 ± 1.8
Phaseolus aureus	Abaxial	7 ± 0.7	8 ± 0.7
	Adaxial	19 ± 2.1	. 21 ± 2.2
Vigna sinensis	Abaxial	12 ± 1.6	13 ± 1.6

^{*} x - Mean

⁻ Standard deviation.

Table 30 Breadth of Stomatal Pore (u) in Plants Grown in Polluted and Control Environments ($\tilde{x} \pm \sigma$)*

Species		Polluted	Control
	Adexiel	1.75 ± 0.29	2.50 ± 0.35
Cicer arietenum	Abaxial	1.80 ± 0.12	2.1 ± 0.18
· ·	Adaxi al	2.8 ± 0.2	5.0 ± 0.34
Boliches lablab	Abaxial	2.0 ± 0.18	3.2 ± 0.26
	Adexial	2.4 ± 0.18	3.5 ± 0.21
Làns culinaris	Abaxial	1.6 ± 0.12	1.9 ± 0.34
	Adaxial	2.0 ± 0.32	3.6 ± 0.41
Phaseolus aureus	Abaxial	1.4 ± 0.26	2.2 ± 0.34
	Adaxi al	4.0 ± 0.11	4.6 ± 0.23
Vigna sinensis	Abaxial	2.9 ± 0.16	3.3 ± 0.19

^{*} X - Mean

⁻ Standard deviation

Table 31 Density of Trichomes (Number/ u^2) in Plants Grown in . Polluted and Control Environments ($\bar{x} \pm \sigma$)*

Species		Polluted	Control
,	Adaxi al	190 ± 19.7	130 ± 14.7
Cicer arietenum	Abaxial	170 ± 17.4	160 ± 16.2
	Adaxi al	115 ± 12.1	80 ± 8.4
Dolichos lablab	Abaxial	130 ± 14.1	110 ± 11.8
	Adaxial	60 ± 7.0	45 ± 5.3
Lens culinaris	Abaxial	70 ± 7.1	65 ± 8.2
	Adaxial	130 ± 14.1	90 ± 10.2
Phaseolus aureus	Abexial	120 ± 15,2	115 ± 10.4
	Adexial	40 ± 6.0	30 ± 5.6
Vigna sinensis	Abaxial	40 ± 5.0	35 ± 5.8

^{*} T - Mean

⁻ Standard deviation.

Table 32 Length of Trichomes (u) in Plants Grown in Polluted and Control Environments ($\bar{x} \pm \sigma$)*

Species		Polluted	Cont rol
	Adaxial	120 ± 17.8	130 ± 12.3
Cicer arietenum	Abaxial	150 ± 14.2	145 ± 12.8
	Adexila	120 ± 16.2	80 ± 9.2
Dolichos lablab	Muxial	100 ± 11.8	95 ± 7.4
	Adaxi al	90 ± 18.3	70 ± 7.4
Lens culinaris	Abaxial	90 ± 10.4	80 ± 7.0
	Adexiel	110 ± 13.4	.75 ± 4.9
Phaseolus aureus	Abaxial	95 ± 10.1	85 ± 6.2
	Adaxial	45 ± 6.2	40 ± 4.2
Vigna sinensis	Abaxial	50 ± 5.5	50 ± 3.2

^{*} x - Mean

⁻ Standard deviation

Table 33

Total Chlorophyll Content (mg/gm) in Plants Grown in Polluted and Control Environments

Species	Polluted	Control	Percent Reduction
Dolichus lablab	3.301	4.283	20.9
Lens culinaris	3.615	4.732	22.7
Phaseolus aureus	2.621	3.929	37. 8
Cicer ariecatinum	2.021	3.237	18.2
Vigna sinensis	3.121	3.512	11.2

Table 34

Plants Selected for Scanning Electron Microscopic Study

1.	Brassica oleracea
2.	Chenopodium album
3.	Cicer arietenum
4.	Dolichus lablab
5.	Lantana camara
6.	Sonchus asper
7.	Withania somnifera

CHAPTER - IV

DI SCUSSION

DI SCUSSION

During the last decade there has been a growing interest in the study of the effect of air pollution on plants (Mudd and Kozlowski', 1975; Mansfield, 1971). Most of the information available on this subject is from the studies made in Northern America and Europe. Air pollution responses on tropical plants particularly under Indian conditions have not been studied, though problems of air pollution are increasing at an alarming rate (Parikh, 1977; Varshney and Garg, 1978).

Most studies on the effect of air pollution on plants are either confined to morphological affects or field observations or are concerned with metabolic disorders studied under laboratory conditions. Field experiments for studying the effect on air pollution have not been widely utilized. Also the impact of air pollution on epidermal features such as stomatal number, size of stomatal pore, length and density of trichomes have not been systematically studied.

The present study was carried out to evaluate the effect of air pollution on plants in Delhi. Gross morphology, including number of leaves per node, leaf biomass, total plant biomass, total chlorophyll content and epidermal features were studied to determine plant responses to air pollution. Rpidermal features of

seven species (Table 34) were analysed in detail, with the help of a scanning electron microscope for evaluating the effect of air pollution on leaf epidermis.

Severe leaf injury symptoms such as necrotic patches, death of interveinal tissues and yellowing of leaf margins were observed in <u>Sonchus asper</u>, <u>Achyrenthus aspera</u> and <u>Dolichus lablab</u>, while the other plants did not have equal amount of injuries. These injury symptoms appear to be due to both particulate matter and sulphur dioxide because Darley (1966) reported death of interveinal tissue due to particulate matter pollution and bleaching of leaf margins and formation of necrotic patches in alfalfa has been ascribed to sulphur dioxide pollution by Thomas (1961).

Data in Tables (7, 8, 26 and 27) show that leaves having pilose surface were more injured in comparison to plants having glabrescent leaf surface. Plants with glabrous leaf surface such as Cynodon dactylon were least effected. It appears that plants having glabrous leaf surfaces are relatively resistant to air pollution. In literature, information on the relationship between leaf vesture and plant succeptibility to air pollution is lacking. Hence, it has not been possible to compare these observations. In future, however, it is important to examine the relationship between nature of leaf

vesture and succeptibility of plants to air pollution.

The effects of air pollution on the gross morphology of plants are fairly significant (Tables 15-25).

Leaf area was found to be greatly reduced in plants grown in polluted environment as compared to control plants. Leaf biomass and total plant biomass were also greatly reduced in plants grown in polluted environment.

Loss of yield as a result of air pollution has been a common experience. Considerable reduction in the growth and yield in several crops were found to be due to sulphur dioxide and ozone acting alone or in mixture (Reinert, et al., 1970). Reduction in the growth of shoot and root has been recorded in <u>Ricinus</u> sativus due to sulphur dioxide and ozone, either alone or in combination (Tingley, et al., 1971a).

Reduction in leaf area, leaf biomass and total plant biomass was maximum in experimental plants having pilose and pubescent leaf surfaces while <u>Vigna sinensis</u>, which has glabrescent leaf surface was least affected. Again, information on the relationship between leaf vesture and its relation to the impact of air pollution in terms of leaf area, leaf biomass or total plant biomass is completely lacking. Leaf vestures appear to play an important role in determining plants succeptability to air pollution. Ecological significance and

consistancy of such relationship need to be examined in future studies.

The present study has shown that chlorophyll content in plants grown in the polluted environment was much less in comparison to plants from non-polluted areas (Tables 14, 33). Rao and Le Blanc (1966) were the first to report decrease in chlorophyll content in Lichen alga due to sulphur dioxide pollution. Malhotra (1977) has also reported appreciable decrease in chlorophyll content in Pinus countria due to aqueous sulphur dioxide pollution. The likely expalnation for chlorophyll reduction in plants exposed to sulfur dioxide is the degradation of the photosynthetically active chlorophyll into photosynthetically inactive brownish pigment called phaeophytin. It results from the displacement of the magnesium ion from the center of porphyrin ring of the chlorophyll molecule by hydrogen ion (Rao and Le Blanc, 1966). It may be observed that the reduction in the amount of chlorophyll is related to the reduction in leaf area, leaf biomass and total plant biomass (Tables 14-24 and 32).

Salisbury (1927) after a detailed study on stomatal behaviour, has suggested that numerical variation in stomata could be of great importance in

mental stress such as humidity (Salisbury, 1927; Aykin, 1952; Schurmann, 1959), light intensity (Pazourek, 1970) and altitudinal variation (Sharma, 1975) have been shown to be quite substantial. Recently, Sharma and Butler (1973, 1974, 1975) have shown that the effect of air pollution on epidermal features are quite significant. In the present study the effects of air pollution on the number of stomata, size of stomatal pore, density and length of trichomes have been examined.

The number of stomata on the adaxial leaf surface was less in the polluted area as compared to the plants of non-polluted sites. High percentage reduction in the number of the stomata on adaxial leaf surface was observed in plants having mostly pilose and pubescent type of surface pattern (Sonchus asper, Brassica oleracea) (Tables 8 and 9), where as significantly low values were observed for glabrous and glabrescent surface nature (Cynodon dactylon, Calotropis procera). In experimental study maximum reduction in the number of stomata was observed in pilose leaf of Cicer arietenum and minimum in glabrescent leaf of Vigna sinensis (Tables 27 and 28).

These observations are in agreement with the studies of Sharma and Butler (1973, 1974, 1975) who have

observed appreciable reduction in the number of stomata in Trifolium répens. Trifolium pratense and Acer saccharum grown in an environment, polluted with sulphur dioxide, particulate matter and exides of carbon (Table 2). Contrary to this in Calotropis procera, growing in an area having particulate matter and sulphur dioxide as major pollutants, an increase in the number of stomata has been recorded by Yunus and Ahmed (1979). In addition, few studies have been conducted on the effect of gaseous pollutants on stomatal behaviour. Some ozone sensitive tobacco varieties were found to have higher stomatal frequency (Dean. 1972: McKee. 1973). In other study related to the effect of sulphur dioxide no relation was found between the number of stomata per unit leaf area and relative succeptibility of plants (Zimmermann and Hitchocock. 1956).

The ecological significance of reduction or increase in stomatal frequency in response to air pollution cannot be specified as there is a lack of consistency between numerical variation in stomata and succeptibility of plants to air pollution. Future work on the effect of air pollution on the stomatal frequency might be able to clarify the situation.

Leaf vesture and reduction in number of stomata, however, appear to have some relationship. Leaves with pilose vesture were found to show substantial reduction. Further work on these lines will evaluate the validity of the relationship between leaf vesture and variation in stomatal number due to air pollution.

In polluted environment the length of stomatal pore on the adaxial leaf surface was found to be reduced (Tables 8, 10, 27 and 29). Plants like Sonchus asper, Brassica oleracea and Cicer arietenum having pilose and pubescent leaf vesture showed maximum reduction. On the other hand, length of stomatal pore in plants such as Cynodon dactylon, Calotropis process and Vigna sinensis having glabrous and glabrescent leaf surface did not change much. Electron microscopic study also support the reduction in size of stomatal pore in plants from polluted areas (Figs. 3-6 and 11-16).

The air pollution was also found to have considerable effects on the breadth of stomatal pore. The breadth of stomatal pore was reduced more in plants with pubescent and pilose leaf vesture as compared to those plants having glabrous and glabrescent leaf surfaces (Table 8 and 11). However, it is interesting to note that breadth of stomatal pore in pubescent leaf of

Dolichus lablab and Phaseolus aureus and Achyranthus

aspera showed maximum reduction while Souchus asper

and Cicer arietenum having pilose leaf vesture exhibited

moderate reduction (Tables 27 and 30).

It may be observed that leaves with pilose leaf vesture exhibited higher reduction in the length and pubesent leaf in breadth of stomatal pore as compared to glabrous and glabres t leaf surfaces.

Specific information on the effect of air pollution on the length and the breadth of stomatal pore is all together lacking, however, Sharma and Butler (1973, 1974, 1975) have reported little variation in the stomatal size range in <u>Trifolium repens</u>, <u>Trifolium pratense</u> and <u>Acer saccharum growing in polluted areas</u>. The available information shows that the sulphur dioxide promotes stomatal opening as reported by Majernik and Mansfield (1970) in <u>Vicia faba</u> plant treated by sulphur dioxide.

Like stomata, trichomes also show considerable response towards air pollution. Increase in the density and length of trichomes were observed in the plants of polluted areas.

Adamial leaf surface having pilose vesture in Sonchus asper and Cicer arietenum showed appreciable increase in the density of trichomes. Changes in leaves having glabrescent surface in plants like <u>Ricinus communis</u> and <u>Vigna sinesis</u> were not significant (Tables 8, 12, 27 and 31).

Considerable increase in the length of trichomes was observed on adaxial leaf surfaces in Sonchus asper and asper Dolichos lablab having pilose and pubescent leaf vesture respectively, whereas glabrescent leaf vesture at adaxial surface in Chenopodium album and Vigna sinensis were not prominent (Tables 8, 13, 27 and 32).

Studies done so far on the effect of air pollution on trichome characteristics also support the above observations. Increase in the density and the length of trichomes were reported in <u>Trifolium repens</u>, <u>Trifolium pratense</u> and Acer saccharum growing in areas polluted by sulphur dioxide, particulate matter and carbon monoxide. (Sharma and Butler, 1973, 1974, 1975). Similarly higher trichome density in <u>Calotropis procera</u> has been reported in plants polluted with particulate matter and sulphur dioxide (Yunus and Ahmed, 1979).

Increase in density and length of trichomes also appear to have some relationship with leaf vesture since

pilose and pubescent leaf were generally found to show greater increase in the length and density of trichomes.

Further studies on these aspects are required to draw reliable conclusions and to elucidate the mechanism of action of the air pollution on leaf trichomes.

The abaxial leaf surface appears to be relatively resistant to air pollution as compared to adaxial leaf surface. Though the number of stomata also decrease slightly on the abaxial surface, they do not reduce more than 20 per cent (Tables 9 and 28). Similarly, the size of stomatal pore remains more or less unaffected, on abaxial leaf surface except Calotropis procera, Achyranthus aspera, Dolichus lablab and Phaseolus aureus where decrease in either length or breadth of stomatal pore was around 30-36 per cent (Tables 10, 11, 29 and 30). As expected, length of trichome and their density were not as much affected as on adaxial leaf surface (Tables 12, 13, 31 and 32).

Very few studies have attempted to show the effect of air pollution separately on the two leaf surfaces. Reduction in the number of stomata was found to be more at the adaxial leaf surface in <u>Trifolium repens</u> and <u>Trifolium pratense</u> (Sharma and Butler, 1973, 1974). They have also reported that stomatal size range at abaxial

and adamial surfaces remains more or less the same. No study has been done to compare the response of trichomes to air pollution on the two leaf surfaces.

One of the likely explanation for comparatively resistant nature of abaxial leaf surface, as observed in present study could be that the abaxial surface is not equally exposed to air pollution when compared to adaxial leaf surface.

Stomatal reduction in the plants of polluted area appears to have some adaptive significance. Sharma and Butler (1974) have strongly argued that reduction in the number of stomata help the plant in reducing the diffusion of gaseous pollutants into the leaf. It seems that reduction in number of stomata could be due to interference of pollutants in the normal development of stomata at a fairly early stage of their formation.

Sharma and Butler (1974) have also suggested that changes in the length of trichome and their density has adaptive significance for plants as it helps to reduce the temperature of leaf surface as well as they may also protect the leaf surface and stomata from particulate matter. The long and denser trichomes may also have opposite effects since long and dense trichomes would trap particulate matter more effectively. It seems that

with time the deposition of particulate matter will increase and it may thereby increase the leaf temperature (Etler, 1977), with its obvious effects on biochemical reactions in the leaf.

The studies made so far on the effect of air pollution on leaf epidermis indicate that the epidermal features are very sensitive to air pollution. It appears likely that adaxial leaf surface having long dense trichomes are more responsive as compaired to glabrous leaves. Epidermis on adaxial leaf surface may serve as a bioindicator for monitoring air pollution. The need for a relaiable bioindicator of air pollution cannot be overemphasized for an effective environmental monitoring. Air monitoring using a simple and inexpensive bioindicator is of great practical help for improving and conserving air quality. Concept of biomonitoring of the environment with the help of an indicator organisms is receiving considerable attention (Puckett et al.. 1971: Rao, 1977). Recently introduced concept of "Mussel Watch", i.e., utilization of bivalves as sentinel organisms for indicating pollution by heavy metals. artificial nucleides, petroleum components and halogenated hydrocarbons in the coastal areas of the marine environment is now gaining much importance (Goldberg

et al., 1978). Similarly, leaf epidermis could be developed as a convinent system for monitoring air pollution. For this purpose systematic work on a wide variety of plants is needed for selecting and standardizing appropriate plants for their leaf epidermis. Studies on the effect of individual air pollutants on the leaf epidermis and detailed analysis of the epidermal surface using scanning electron microscope will be of great practical value.

CHAPTER - V

SUMMARY

SUMMARY

Steadily increasing problems of air pollution have promoted the studies related to the effect of air pollutants, viz. sulphur dioxide, particulate matter, carbon monoxide and hydrocarbons on plants, animals and materials. Plants seem to be more vulnerable to air pollution than animals. Position and structure of leaves have made them to suffer most due to air pollutiants. Leaf epidermis, being the outermost layer to come in continuous centact with air pollutants is likely to bave more severe effects. Ecophysiological studies have shown that leaf epidermis is sensitive to humidity (Salisbury, 1927: Aykin, 1959), light intensity (Pazourek. 1970) and altitudinal variation (Sharma, 1975). Most of these studies have been done on plants under temperate regions. Comparable information on the effects of air pollution on tropical plants is fragmantory or lacking.

The present study was designed to examine the effect of air pollution caused by Indraprastha Thermal Power Station (New Delhi) on leaf epidermis. For this purpose, following ten plants viz., Achyranthus aspera Brassica oleracea, Calotropis procesa, Chenopodium album, Cynodon daetylon, Lantana camara, Nerium indicum

Ricinus communis, Sonchus asper and Withania sonnifera, common to three Sites A, B and C, representing decreasing levels of air pollution, were selected. Observations regarding visual injury symptoms, number of stomata, size of stomatal pore, length and density of trichomes and chlorophyll content were made for plants grown both in the polluted and non-polluted environment.

The effects of air pollution was clearly visible in plants growing near pollution source as their leaves exhibited typical visual injury symptoms such as necrotic patches, chlorosis and death of intervenial tissues. Air pollution seems to reduce the number of stomata in plants. Length and breadth of stomatal pore were also found to be sufficiently less in plants from polluted area. The density and length of trichomes were found to be higher in plants from the polluted environment.

An experimental study was designed to confirm the above results. Five plants, viz., <u>Cicer arietenum</u>, <u>Dolichos lablab</u>, <u>Phaseolus aureus</u>, <u>Lens culinaris</u> and <u>Vigna sinensis</u> were grown in polluted and non-polluted areas to observe the changes in gross morphology (number of leaves per node, leaf area, leaf biomass and total plant biomass), epidermal features (number of

stomata, size of stomatal pore, trichome length and their density) and chlorophyll content.

The effect of air pollution on experimental plants was found to be more or less same as observed during the field study.

exhibited that the adaxial leaf surface is more succeptable than the abaxial leaf surface. An apparent relationship was found between leaf vesture and changes in epidermal features, as pilose leaf vesture show considerable changes in number of stomata, length and breadth of stomatal pore, trichome length and density, under the influence of air pollution.

The results of the present study reveal that adaxial pilose leaf surface is extremely sensitive to air pollution and it has the potential for being developed as a suitable bioindicator for monitoring air pollution.

REFERENCES

REFERENCES

- Arndt, U. (1970). Konzentrationsanderungen bei freien aminosauren in Pflanzen unter dem einfluss von fluorwasserstoff und schwiefedioxd, Staub 30, 2560.
- Ballantype, D. (1973). Sulphite inhibition of ATP formation in plants mitochondria. Phytochemistry 12, 1207.
- Bailey, J.L. and Cole, R.D. (1959). Studies on the relation of sulfite with proteins. <u>J. Biol. Chem.</u> 234, 1733.
- Bell, J.N.B. and Clough, W.S. (1973). Depression of yeild in ryegrass exposed to SO₂. Nature (London) 241. 43.
- Brandt, C.S. and Heck, W.W. (1968). Effect of air pollution on vegetation. In Air Pollution (Ed. by A.C. Stern), vol. 1, p. 401. Academic Press, N.Y.
- Burstorm, H. (1961). Development of stomata on submerged leaves. Kungl. Fsysiografiska Sallekapets I. Lund Forhandlingar 31, 25-30.
- Eller, B.M. (1977). Road dust induced increase of leaf temperature. Env. Poll. 13(2), 99-107
- Gesalman, C.M. and Davis, D.D. (1978). Ozone susceptibility of <u>Ten Aslea</u> Cultivars as related to stomatal frequency or conductance. <u>J. Amer. Soc.</u>

 <u>Hort. Sci.</u> 103(4), 489-491.
- Godzik, S. and Sassen, M.M.A. (1978). A scanning electron microscope examination of <u>Aesculus hippocastanum L.</u> leaves from control and air-polluted areas. <u>Env. Poll.</u> 17, 13-18.
- Goldberg, E.D. et al. (1978). The mussel watch. <u>Environmental Conservation</u> 5(2), 101-125.
- Khush, G.S. and Stebbins, G.L. (1959). Variation in the organization of theguard cells complex in the leaf epidermis of the monocotyfedons and its hearing of their phylogeny. Paper presented at IX Int. Bot. Cong., Montreal, Candda.

- Krause, C.R. (1976). Surface characteristics of American Elm clones for identification by scanning electron microscopy. <u>Hort. Science</u> 11(4), 386_388.
- Majernik, O. and Mansfield, T.A. (1971). Effects of SO₂ pollution on stomatal movements in <u>Vicia faba</u>
 Phytopath. 2. 71, 123-218.
- Malhotra, S. (1977). Effect of aqueous sulphur dioxide on chlorophyll destruction in <u>Pinus countrata</u>.

 New Phytologist. 78(1), 101.
- Manser, H.A., Hodges, G.H. and McKee, C.G. (1973).

 Effect of air pollution on Maryland (type 32).

 tobaccoo. Journal of Environmental Quality 2,
 253-258.
- Mohan Ram, H.Y. and Nayyar, V.L. (1977). A leaf clearing technique with a wide range of applications. <u>Proc. Indian Acad. Sci. 87B(5)</u>, 125-127.
- Mudd, J.B., Kozlawski, T.T. (Eds.) (1975). Résponses of Plants to Air Pollution, Academic Press, N.Y.
- Padmanabhamurthy, B. and Gupta and R.N. (1977). Particulate follution in Delhi due to Indraprastha Power Station. Ind. J. Met. Hydrol. Geophys. 28(3).
- Parikh, K.J. (1977). Environmental problems of India and their possible trends in future. Environmental Conservations 4(3), 189-198.
- Pazourek, J. (1970). The effect of light intensity on stomatal frequency in leaves of <u>Iris fiollindica</u>
 <u>Host. Bio. Plant. 12</u>, 208-215.
- Rao, D.M. and Leblanc, F. (1966). Effects of sulphur dioxide on the lichen alga with special reference to chlorophyll. <u>Bryologist</u>. 69, 69-75.
- Salisbury, E.J. (1928). On the causes and ecological significance of stomatal frequency with special reference to the woodland flora. Phil. Trans. Roy. Soc. London 216, 1-65.
- Reinert, R.A., Heagle, A.S., Miller, J.R. and Greekeler, W.R. (1970). Field studies of air pollution injury to vegetation in Cinicinanti, Ohio. Plant Dis. Rep. 54, 8-11.

- Schurmann, B. (1959). uber den einfluss der hydralur und des linntes auf die ausbildung der stomata Initialen Flora 147, 471-520.
- Sharma, G.K. (1972). Environmental modification of leaf epidermis and morphological features in <u>Verbena</u> canadensis <u>S.West. Nat.</u> 17, 221-218.
- Sharma, G.K. (1975). Leaf surface effects of environmental pollution on sugar maple (Acer saccharum) in Montreal. Can. J. Bot. 53, 2312-2314.
- Sharma, G.K. (1975). Altitudingl variation of the leaf epidermis in the taxonomy of the Combretaceae III. The genus Combretum in America. Brittionia 21, 130-143.
- Sharma, G.K. and Butler, J. (1973). Leaf cuticular variations in <u>Trifolium repens</u> L. as indicators of environmental pollution. <u>Environ. Pollut.</u> 5, 287-293.
- Sharma, G.K. and Butler, J. (1974). Environmental pollution: Leaf cuticular patterns in <u>Trifolium pratense</u> L. <u>Ann. Bot.</u>, 1087-90.
 - Stunny Exax \$2969\$
 - Showman, R.E. (1972). Residual effects of SO, on the net photosynthesis and respiratory rates on lichen thalli and cultime lichen symbionts. Bryologist 75, 335.
 - Stace, C.A. (1969). The significance of the leaf epidermis in the taxonomy of the combretaceae III. The Combretum in America. Brittonia 21, 130-143.
 - Thomas, M.D. (1961). Effects of Air pollution on Plants. World Health Organization, Genova, Monograph No. 46, 233-278.
 - Tingrey, D.T., Heck, W.W., Reinert, R.A. (1971). Effect of low concentration of ozone and sulphur dioxide on foliage, growth and yield of radish. J. Amer. Soc. Hort. Sci. 96, 369-371.
 - Varshney, C.K. and Garg, J.K. (1978). A quantitative assessment of sulphur dioxide emission from fossil fuels in India. <u>J. Air. Pollut. Cont. Assoc. 28(II)</u>, ||4|-42
 - Wellburn, A.R., Majermk, O. and Wellburn, F.A.M. (1972). Stomatal responses to SO₂. Nature (Lond.) 239, 458.

- Yunus, M. and Ahmed, K.J. (1979). Use of epidermal traits of plants in pollution monitoring. Reprinted from the Proceedings of National Seminar on Environmental Pollution and its Control A Status Review, Session V, National Productivity Council, Bombay.
- Ziegler, I. (1972). The effect of SO₃ on the activity of ribulose 1, 5 diphosphate carboxylase in isolated spinach chloreplasts. Planta 103, 155.
- Ziegler, I. (1973). Effect of sulphite on phosphoenol pyruvale carboxylase and malate formation in extracts of Zea mays. Phytochemistry 12, 1027.
- Zimmermann, P.W. and Hitchock, A.E. (1956). Succeptability of plants to hydrofloric acid and sulphur dioxide gases. Contrib. Boyce, Thompson Inst. 18, 263-279.