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# Effect of air Pollution on Plants: Epidermal and optical Characteristics of Leaf

Thesis submitted to the Jawaharlal Nehru University  
in partial fulfilment of the requirements for the  
degree of  
**DOCTOR OF PHILOSOPHY**

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December, 1983

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DEDICATED

TO


MY

PARENTS

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## PREFACE

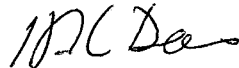
This research work entitled "Effects of Air Pollution on Plants : Epidermal and Optical Characteristics of Leaf" has been carried out at the School of Environmental Sciences, Jawaharlal Nehru University, New Delhi. This work is original and has not been submitted in full or part for any degree or diploma of any other university.

  
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## INTRODUCTION

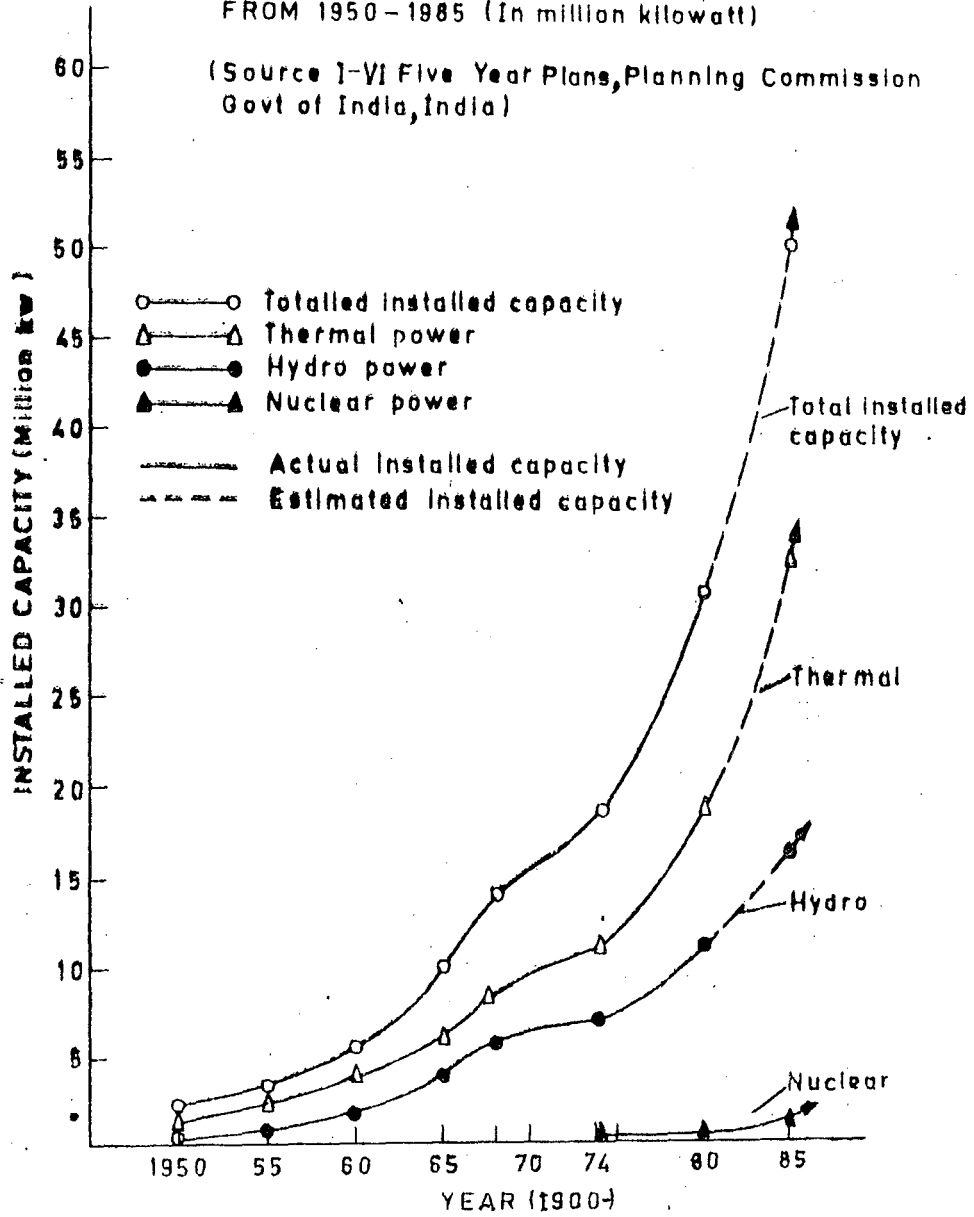
There is a widespread concern towards air pollution problems which are rapidly increasing as a consequence of accelerated growth in thermal power generation, industrialization and transport networks. Over past 30 years, thermal power generation in India has increased considerably. The installed capacity of thermal power generation in 1950 was 1.75 mkw has grown upto 19.00 mkw by March, 1980 (Fig. 1.1). Thermal power constitutes about 65% of the total installed power generation capacity in the country (Table 1.1). It is estimated that installed capacity of thermal power will reach upto 32.84 mkw by 1985, while the total generation capacity will increase upto 50.68 mkw by 1985 (Fig. 1.1).

A significant increase in the installed capacity of thermal power has also enhanced the importance of coal as a primary source of energy. Consumption of coal has increased to 31.2 MT in 1980 as against 2.8 MT in 1950 (Table 1.2). It is anticipated that by the end of the Sixth Five Year Plan (1981-85), the consumption of coal for thermal power generation will

Fig. 1.1

INSTALLED POWER GENERATION CAPACITY IN INDIA FROM 1950-1985 (In million kilowatt)

(Source I-VI Five Year Plans, Planning Commission Govt of India, India)



**Table 1.1: Total installed capacity (in million kw) and the generation type of India during 1950 to 1985**

	1950	1955	1960	1965	68-69	73-74	1980	1985*
Thermal	1.75	2.48	3.73	6.07	8.38	11.32	19.00	32.84
Hydro	0.56	0.94	1.92	4.10	5.91	7.02	11.38	16.51
Nuclear	-	-	-	--	-	0.58	0.64	1.33
Total	2.30	3.42	5.65	10.17	14.29	18.86	31.02	50.68

<sup>1</sup>Estimated figures

From: I-VI Five Year Plans, Planning Commission, Govt. of India, India.

**Table 1.2: Total coal production (Million Tonnes) and its utilization in thermal power plants in India during 1950 to 1985**

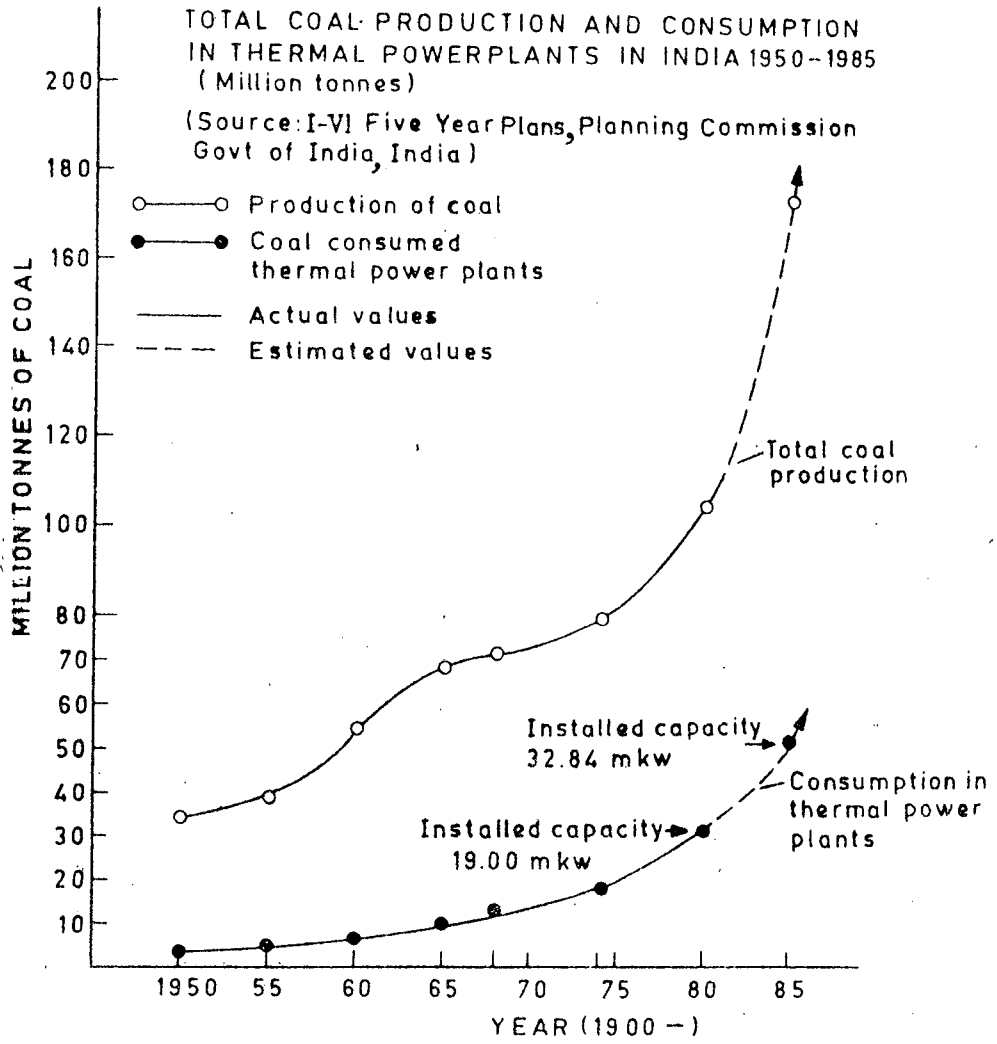
	1950	1955	1960	1965	68-69	73-74	1980	1985*
Coal production	34.4	38.2	54.6	67.7	71.4	79.0	101.0	173.0
Utilization in thermal power plants	2.8	3.82	5.9	9.6	13.1	17.4	31.2	51.2

\*Estimated figures

From: I-VI Five Year Plans, Planning Commission, Govt. of India.



Fig. 1.2



increase to 51.2 MT by 1985, and total coal production may reach upto 173 MT by 1985 (Fig. 1.2).

Combustion of coal results into emission of large amounts of air pollutants, important among them are sulphur dioxide ( $\text{SO}_2$ ) and particulate matter as flyash. Studies have revealed that plants are more sensitive to  $\text{SO}_2$  as compared to other living organisms.\* In view of high sensitivity of plants due consideration has been given in adopting the ambient air quality standards. In USA, under the Clean Air Act, 1971, primary standards are aimed to protect public health and secondary standards are also specified to ensure public welfare (plants, property, aesthetic, etc.). A comparison of primary and secondary air quality standards reveals that the value of the secondary standard for both  $\text{SO}_2$  and particulate matter is much less as compared to the primary standard (Table 1.3). In India, Air (Prevention and Control of Pollution) Act was passed in 1981, and under this Act the Central Board of Prevention and Control of Water Pollution (CBPCWP) adopted air quality standards in November,

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\*Air Quality Criteria for Sulphurdioxide, National Air Pollution Control Administration, Department of Health Education and Welfare, AP-50, 801 N. Randolph Street, Arlington, Virginia, February, 1969.

**Table 1.3: Primary and secondary air quality standards (USA)**

Pollutant	Duration	Primary standard	Secondary standard
Sulphur dioxide	Annual Arithmetic Mean	80 $\text{ug m}^{-3}$	-
	Max. 24 h*	365 $\text{ug m}^{-3}$	
	Max. 3 h*	-	1300 $\text{ug m}^{-3}$
Particulate matter	Annual Geometric Mean	75 $\text{ug m}^{-3}$	60 $\text{ug m}^{-3}$
	Max. 24 h*	260 $\text{ug m}^{-3}$	150 $\text{ug m}^{-3}$

\*Concentration not to exceed in the given time period once a year

From: Environment Protection Agency (EPA), USA.

**Table 1.4: Ambient air quality standards for different categories (India)**

Area	Category	SO <sub>2</sub>	Suspended Particulate matter
A	Industrial and mixed use	120	500
B	Residential and Rural	80	200
C	Sensitive	30	100

Central Board for Prevention and Control of Water Pollution, India.

Three consecutive measurements spaced by at least one week apart or any three out of 10 consecutive measurements spaced by at least one week apart should not exceed the prescribed limits, in sampling done with a frequency not less than once in a week with sampling time of 8 h. Monitoring should be done throughout a year.

1982. The standards have been adopted for three categories (Table 1.4), viz., (a) Industrial and mixed use; (b) residential and rural; (c) sensitive (hill stations, sanctuaries, national parks, and national monuments). It is evident that (Table 1.4) the values for both sulphur dioxide and particulate matter for sensitive category are comparatively less than the other two categories, in order to protect plants and other sensitive organisms and materials.

In spite of growing public concern, the problem of air pollution is progressively increasing. Adverse effects of air pollutants on plants particularly of  $\text{SO}_2$  and/or particulate matter have been widely documented (Thomas, 1956, 1961; Brandt and Heck, 1968; Yeshow, 1970; Arya, 1971; Taylor, 1973; Mudd and Kozlowski, 1975; Lacasse and Trashow, 1978; Hallgren, 1978; Varshney and Garg, 1979; Heath, 1980; Ormrod, 1980; Chaphekar, 1982). A brief review of studies conducted on the effects of  $\text{SO}_2$  and/or particulate matter on plants is given below in two parts.

I. General effect of  $\text{SO}_2$  and/or particulate matter on plants; II. Effects of  $\text{SO}_2$  and/or particulate matter on epidermal and optical characteristics of leaf.

## I. General Effect of $\text{SO}_2$ and Particulate Matter on Plant

In this section studies conducted in field and under artificial exposure have been discussed under three sub-heads, viz. (i) visual injury symptoms; (ii) growth alterations; (iii) physiological and biochemical effects.

### Visual injury symptoms

$\text{SO}_2$ : Usually injury symptoms in the form of chlorosis and necrosis were observed in plants exposed to  $\text{SO}_2$ . Chlorosis often develops in leaves which have been exposed to non-lethal concentrations of  $\text{SO}_2$  for prolonged periods. Chlorosis represents loss or reduction of green pigment chlorophyll, resulting into yellowish colouration, of leaf tissue. Chlorosis caused by  $\text{SO}_2$  has been also referred to as 'sulfate' injury, due to accumulating  $\text{SO}_4^{-2}$  ions, a phytotoxicant.  $\text{SO}_2$  diffuses into leaf tissue through stomata and get dissolved in water in the mesophyll cells to produce  $\text{SO}_3^{-2}$  ions which are relatively more phytotoxic as compared to  $\text{SO}_4^{-2}$  ions. When lethal concentrations of  $\text{SO}_3^{-2}$  ions accumulate in the most susceptible areas of the leaf injury symptoms appear more readily in interveinal areas, tip or margins, as a result of this dull green,

and water-soaked areas appeared initially. The affected area soon become flacid, and in most plants they are bleached to white or ivory upon drying. In some plants the dead tissue may turn red, brown or black.

Visual injury symptoms like chlorosis, tip necrosis, leaf tipburn were very common in Pseudotsuga menziesii, Pinus ponderosa and Abies lasiocarpa, found in the vicinity of a copper smelter (Haywood, 1919). Linzon (1972) observed necrosis in the needles of Pinus strobus growing near nickel and copper smelters. Defoliation and necrotic injury symptoms were observed in plants growing within 100 m of a fertilizer factory in Bombay (Chaphekar, 1972), while plants at a distance of about 500 m were suffering mainly from chlorosis. Chlorosis, necrosis and burning of leaves have been reported in plants growing in industrial areas of Lucknow, Varanasi, Aligarh and Mirzapur (Yunus and Ahmed, 1979). Development of injury symptoms observed in cotton plants revealed that yellowing appears first on the lower surface (abaxial) of the leaf and gradually extends towards the upper (adaxial) surface. In later stages, white or brownish red turgid areas developed

between the veins usually followed by premature leaf fall.

Studies on the effects of SO<sub>2</sub> exposure on plants have shown that SO<sub>2</sub> concentration equal or higher than 140 ppb causes development of necrotic spots on leaves (Hill and Thomas, 1933; Katz, 1949). In an experiment (Markowski et al., 1974) six species of plants fumigated with 0.3 to 0.5 ppm of SO<sub>2</sub> for 14 consecutive days, developed interveinal necrotic areas. Crossandra unduleafolia and Mirabilis jalapa exposed to SO<sub>2</sub> in the range of 50 to 100 ppm for 8 h caused defoliation in less than 48 h (Chaphekar and Karbhari, 1974). Medicago sativa has been considered one of the most sensitive plants and when exposed to SO<sub>2</sub> artificially exhibited bleaching appearance between the veins and on leaf margins. Bleaching extended towards the mid rib with the increase in the dose of SO<sub>2</sub>. The bleached areas become ivory to white on drying as has been observed in other plant species like garden pea and corn. In plants like chrysanthemum, blackberry, the bleached areas turns brown, while red colour was observed in quince plants. Crop plants like wheat, barley, oats, rye have shown necrotic streaks between the veins

near the leaf tip in artificially fumigated to  $\text{SO}_2$ , which extends towards leaf base in conditions of prolonged fumigation (Lacasse and Treshow, 1973).

In some cases, relationship was observed between the development of necrosis and loss in yield while in others loss of yield was observed without accompanying any morphological injury (Bleasdale, 1952; Tingley, 1971; Heggstad, 1972; Taniyama and Sawanaka, 1973; Malhotra, 1977).

**Particulate matter:** It may occur in various forms, viz., cement dust, flyash, soot, fluoride particles of lead, magnesium oxide, iron oxide and sulphuric acid aerosols. Plant responses to flyash exposure have not been studied but studies relating to other forms of particulate matter such as cement dust may be helpful to understand the nature of responses. The size of particle deposited on vegetation vary between 1 to 100  $\mu$ .

As a consequence of cement dust particles, leaf lesions in the form of brown necrotic patches were found (Czaja, 1960). The deposition of particles of road dust on moist leaves resulted into development of annular chlorotic or bleached patches on laminar surfaces (Chaphekar, 1972). It was observed that cement



particles form an impervious coat mostly on the upper surface of the leaf and when the coat is removed it causes severe injury to the leaf surface (Singh and Rao, 1978). Shetye and Chaphekar (1978) in a study concerning with the analysis of leaf washes of mango and Theppesia trees from different locations of Bombay city, concluded that vehicular traffic is the major contributor to the dust load in the city air. Singh and Rao (1980) observed cracking, peeling and withering of leaf surface due to thick hard incrustations of cement dust of varying thickness on their entire surface in Triticum aestivum plants in the vicinity of cement factory.

In the coal unloading areas (Varanasi, U.P.), the dusted leaves of Magnifera indica and Citrus developed lesions initially at leaf tip and later progressed towards lamina, as brown necrotic patches. Considerable amount of dust was entrapped by plants at leaf bases of unfolding buds, especially the apical ones, which invariably appear black and in majority of cases, these buds were dead and branches bearing them were partly crumpled. Thus, due to continued death of terminal buds, latent buds are activated providing bushy

appearance to the plant. The stigmatic surfaces were covered with a thick deposit of coal particles ranging from 10 to 100  $\mu$  in diameter, these particles stuck to the surface so tenaciously that even a vigorous shaking could not dislodge them. Some pollen grains were also found on the stigmatic surface.

An analysis of the deposition of cement dust in the vicinity of a cement factory showed that it varies from  $1.5 \text{ g m}^{-2} \text{ d}^{-1}$  to  $3.8 \text{ g m}^{-2} \text{ d}^{-1}$  (Pajenkamp, 1961; Bohne, 1963). Leaves of bean plants dusted at a rate of  $4.7 \text{ g m}^{-2} \text{ d}^{-1}$  for two days and exposed to naturally occurring dew suffered from visual foliar injury like rolling of leaf margins and death of interveinal tissues (Darley, 1966). Lerman (1972) observed severe damage in bean plants when leaves were dusted with  $6.64 \text{ g m}^{-2} \text{ d}^{-1}$  in the presence of free moisture. Leaves of Hibiscus abelmoschush sprayed with  $2 \text{ g m}^{-2} \text{ d}^{-1}$  cement dust, coal dust and flyash separately for 30 days developed small chlorotic spots due to cement dust, and marginal chlorosis due to coal dust, while leaves sprayed with flyash did not show any visual injury (Pawar et al., 1982).

SO<sub>2</sub> and particulate matter: In the vicinity of coal based power plant where SO<sub>2</sub> and flyash are the main pollutants, visual symptoms in the form of tip necrosis, chlorosis early abscission, random short needles, twisting and elongation of needles, bud failure, adventitious budding and basal spotting of needles were observed in Pinus strobus, Pinus virginia, Pseudotsuga menziesii and Picea abies (Haselhoff and Lindon, 1903; Haywood, 1910; Scheffer and Hedgcock, 1955). Chlorotic spots and tip necrosis were observed in plants growing in the vicinity of IP Power Plant, New Delhi (Garg, 1979). Plants growing near the Satpura Thermal Power Plant (Madhya Pradesh), were reported to suffer from interveinal necrosis, leaf tip burn, marginal necrosis, necrotic spots, malformed leaves with dissected margins and small punctate spots dispersed over the exposed upper leaf surface (Dubey et al., 1982).

In a study of vegetation near Orba Super Thermal Power Plant (Uttar Pradesh) many plants have been reported to suffer from chlorosis. In grasses, bifacial necrotic streaks between larger veins have been observed (Pandey, 1983).

### Growth alteration

SO<sub>2</sub>: Perhaps Pliny (65 A.D.) was the first to observe and describe the apparent SO<sub>2</sub> damage to vegetation surrounding a smelter. Later on, reports from central Europe in middle ages on air pollution by coal burning were also published (Donaubauer, 1980). Since the beginning of 21st century interest in the field had been increasing and a number of workers have reported adverse effects on plants in field and artificial conditions.

Plant growth in German forest had been affected adversely by SO<sub>2</sub> (Haselhoff and Lindon, 1903). Pseudotsuga menziesii was reported to be the most sensitive species in the vicinity of a copper-smelter at Anaconda in Canada (Haywood, 1910). However, Scheffer and Hedgcock (1955) reported that Abies lasiocarpa is the most sensitive species. Linzon (1972) observed that Pinus strobus is the most susceptible species near nickel and copper smelters in Sudbury district in Ontario (Canada). Adverse effects on plants were also reported due to SO<sub>2</sub> from petroleum refineries (Linzon, 1965). Thuja plicata was found to be killed over a considerable area near the copper smelter at Anyox in

British Columbia (Errington and Thirgood, 1971). Taniyama and Sawanaka (1973) reported that there exists a close relationship between levels of SO<sub>2</sub> in ambient air and the amount of rice harvested. Adverse growth effects on Pseudotsuga menziessi and Pinus flexilis, at three plots within 8 km of copper smelters, have been reported by Carlson (1974). Lolium multiflorum cv. S22 and Dactylis glomerata cv. S143 exposed to ambient air polluted with SO<sub>2</sub> (50-90 ug m<sup>-3</sup>) in a sealed glass chamber resulted in 30-40% reduction in dry weight after 8-10 weeks as compared to plants grown under similar conditions receiving filtered air (Crittenden and Read, 1979). It was also noticed that grasses are particularly more susceptible to SO<sub>2</sub> during their early growth phase.

Concentration of SO<sub>2</sub> exceeding 140 ppb has been reported to reduce plant yield under artificial exposures (Hill and Thomas, 1933; Katz, 1949). On the basis of experimental studies, Hill and Thomas (1933) pointed out that the reduction in yield in Medicago sativa was proportional to the degree of leaf area destroyed by SO<sub>2</sub>. Bell and Clough (1973) observed considerable reduction in yield of ryegrass following SO<sub>2</sub> fumigation

at a mean concentration of 1.2 ppm for a period of over 9 weeks. Six species of plants fumigated with  $\text{SO}_2$  at a concentration of 0.3 to 0.5 ppm from 8 to 13 h for 14 consecutive days suffered considerable reduction in height and had reduced stem and leaf biomass (Markowski et al., 1974). Reduction in growth and dry matter production was observed in Sorghum vulgare var. CSH-I exposed to 0.09 ppm of  $\text{SO}_2$  (Boralkar and Chaphekar, 1978). Triticum aestivum plants exposed to 0.8 ppm  $\text{SO}_2$  (coal smoke) for 2 h daily for 60 days adversely affected root and shoot lengths, number and area of leaves, total plant biomass and number and weight of grains per spike (Rao, 1979). Reduction in shoot, root and ear length and the number of tillers and leaves, and in biomass in Oriza sativa was reported (Rao et al., 1981) on exposure to 0.8 ppm of  $\text{SO}_2$  for 1.5 h for one day and 0.25 ppm of  $\text{SO}_2$  for 30 days ( $1.5 \text{ h d}^{-1}$ ). Glycine max plants exposed to 1.0 ppm of  $\text{SO}_2$  for 2 h for 30 days with a gap of unexposed period for 10 day and again exposed to 0.75 ppm  $\text{SO}_2$  for 2 h for 30 days were found to suffer from reduced leaf area and total plant biomass (Rao et al., 1981). On the other hand, slightly higher yield due to exposure to 0.25 ppm for 4 h daily for six weeks was

reported in Arachis hypogaea, however productivity reduced considerably at higher doses (Mishra, 1980).

Few studies have been conducted to evaluate the impact of low SO<sub>2</sub> concentration on plants as Lockyer and Cowling (1981) noticed that Medicago sativa L., exposed to 0 or 96 ug m<sup>-3</sup> SO<sub>2</sub> for 135 d (the plants were harvested four times), had less shoot weight as compared to control.

Based on experiments of O'Gara (1922) and field studies in an area near smelters of Sudbury, Ontario (Canada), a list of sensitive and tolerant species of plants (crops, flowers, trees and garden plants) has been prepared based on their relative sensitivity to SO<sub>2</sub>. It also includes the modifications suggested by Thomas and Hill (1935) and Thomas and Hendrick (1956).

Particulate matter: Vegetation injury in the vicinity of a cement factory was perhaps for the first time reported by Peirce (1910). Reductions in the growth of popular trees (Bohne, 1963), olive trees (Sheikh et al., 1976), spring growth elongation in conifers (Darley, 1966), plant height and number of leaves in cotton plants (Oblisami et al., 1978), phytomass of Triticum aestivum (Singh and Rao, 1980) were reported in plants growing in the vicinity of a cement factory.

Yield of olive (Sheikh et al., 1976) and Triticum aestivum (Singh and Rao, 1980) plants growing near cement factories was found to be reduced. Chaphekar et al. (1980) correlated reduction in yield with dust load in plants kept in different parts of Bombay city. Vyas (1982) has carried out an elaborate study on plant species growing near a cement factory in Udaipur and concluded that nearby sites are favourable for the production of grasses as compared to other species.

A comparative study was made to evaluate the effects of cement dust, coal dust and flyash on Hibiscus abelmoschush. Fifteen days old plants were sprayed with  $2 \text{ g m}^{-2} \text{ d}^{-1}$  for 30 days. Maximum reduction was observed in plants sprayed with cement dust followed by coal dust and flyash (Pawar et al., 1982).

Schonbeck (1960) suggested that deposition of dust on leaf surface may cause an imbalance in the physiology of plants, and may increase the susceptibility of plants towards certain pathogens. For example, sugar beet plants treated with  $2.5 \text{ g m}^{-2}$  of cement dust were found to be heavily infected by leaf spotting fungus Cercospora bellicola as compared to non-dusted plants. Darley (1966) observed that cement dusted plants



of alfalfa were heavily infested with aphids. Some entomologists have speculated that dust may have eliminated aphid predators resulting in high aphids population on the plants exposed to dust.

Reduction in the lateral growth of Acer rubrum and Quercus prinus and Quercus rubra and increase in the lateral growth of Liriodendron tulipifera growing near limestone quarries have been observed (Brandt and Rhoades, 1973). In addition to direct action of lime dust, variations may be due to changes in soil reactions and nutrient availability to the plant.

Under artificial conditions also cement dust has been shown to reduce plant growth (Darley, 1966; Lerman, 1972). Triticum aestivum plants were sprayed with cement dust at a rate of  $7 \text{ g m}^{-2} \text{ d}^{-1}$  for 60 days. It caused reduction in length of root, shoot and spike as well as in number of tiller, leaves and number of grains per spike (Singh and Rao, 1978). Reduction in leaf biomass was reported in Guava leaves coated with cement between  $5.6 \text{ g m}^{-2}$  in May and  $47.5 \text{ g m}^{-2}$  in January (Lal and Ambasht, 1980).

Petrocoke dust (petroleum refineries) was sprayed on Phaseolus aureus at the rate of  $2 \text{ g m}^{-2} \text{ d}^{-1}$  for 40 consecutive days between 25 and 65 days of plant age (Prasad and Rao, 1981). Reduction in phytomass accumulation net primary productivity was observed. It was interesting to find that total chlorophyll content increased in the treated plants initially, but decreased later when the cumulative doses of petrocoke increased progressively.

Particles containing fluoride have been shown to affect plants adversely. Pack et al. (1959) reported that gladiolus leaf was killed when plants were exposed for four weeks to  $0.79 \text{ ug m}^{-3}$  fluoride as HF, but no necrosis developed when exposed to fluoride aerosol averaging  $1.9 \text{ ug m}^{-3}$  fluoride. McCune et al. (1965) observed tip burn upto 4 mm in length on gladiolus exposed to cryolite (sodium aluminium fluoride dust), but the tipburn length increased upto 7 mm when exposed to similar concentration of HF.

Several studies have shown a direct relationship between lead accumulation and the distance from the heavily travelled roads without exhibiting any injury (Cannon and Bowles, 1962; Page et al., 1971).

TH-1

SO<sub>2</sub> and particulate matter: Adverse effects on plants viz., Pinus strobus, Pinus virginia, Pseudotsuga menziessi and Picea abies were reported in the vicinity of a coal based power plant at Mount Storm, West Virginia, USA. Reduction in the number of leaves, leaf area and total plant biomass was observed in Cicer arietenum, Phaseolus aureus, Dolichos lablab, Lens culinaris and Vigna sinensis kept in the vicinity of IP Power Plant, New Delhi (Varshney and Garg, 1980). A study conducted by Dubey et al. (1982) indicated that plant height and leaf biomass was less in plants growing in the vicinity of Satpura Thermal Power Plant (Madhya Pradesh). Bridetha refusa, Magnifera indica, Tectona grandis and Cassia fistula were most affected plants. Photosynthetically active leaf area was reduced in plants growing near Orba Thermal Power Plant (Uttar Pradesh) and Aegle marmelos and Apluda mutica were reported to be sensitive species (Pandey, 1983).

Dubey et al. (1983) noticed changes in Cicer arietinum plants when exposed to SO<sub>2</sub> (0.5 ppm d<sup>-1</sup>) and flyash separately and in combination. Leaf area and phyto-mass accumulation increased in plants exposed to flyash

only, but reduced in plants exposed to a combination of SO<sub>2</sub> and flyash and SO<sub>2</sub> alone.

#### Physiological and biochemical effects

SO<sub>2</sub>: Adverse effect of SO<sub>2</sub> on photosynthesis have been observed both in field (Bennett and Hill, 1973), and under artificial conditions (Thomas and Hill, 1937a, 1937b; Katz, 1949; Showman, 1972; Taniyama et al., 1972; Zeigler, 1972; 1973). Sij and Swanson (1974) studied the rate of inhibition and recovery of photosynthesis in Phaseolus vulgaris and Zea mays and concluded that the rates differ not only with plant species but also with age of the leaf. They have reported that the rate of net photosynthesis was reversibly inhibited by 130 to 260 ug m<sup>-3</sup> SO<sub>2</sub> in detached leaves of Pisum sativum. When Vicia faba plants were exposed to air containing SO<sub>2</sub> between 20 and 200 ppb, it inhibited net photosynthesis at concentration exceeding 35 ppb. This inhibitory effect was dependent on SO<sub>2</sub> concentration in conditions of light saturation and not at low light intensities (Black and Unsworth, 1979b).

SO<sub>2</sub> has been shown to increase respiration in plants (Keller and Muller, 1958).

Most of the studies on chlorophyll content and biochemical aspects in relation to air pollutants have been carried out in artificially exposed plants. Rao and LeBlanc (1966) have shown that SO<sub>2</sub> exposure of lichen Xanthoria fallax resulted in the conversion of chlorophyll to photosynthetically inactive phaeophytin by replacing Mg<sup>++</sup> ion of chlorophyll molecule by two H<sup>+</sup> ions, and Syrratt and Wanstall (1969) have measured the destruction of chlorophyll in bryophytes. Aqueous SO<sub>2</sub> concentration ranging from 100 to 500 ppm resulted in a sharp decrease in total chlorophyll content and chl a was more sensitive than chl b, in Pinus contorta (Malhotra, 1977). Choudhary and Rao (1977) have reported that chl a is more sensitive than chl b, to SO<sub>2</sub>. Reduction of chlorophyll was observed in Sorghum vulgare (Boralkar and Chaphekar, 1978) and Triticum aestivum (Singh and Rao, 1978) plants exposed to SO<sub>2</sub>. Reduction in total chlorophyll in Agropyro smithii Rydb. was observed when exposed to low (60 ug m<sup>-3</sup>), medium (105 ug m<sup>-3</sup>) and high (175 ug m<sup>-3</sup>) concentration of SO<sub>2</sub> (Lakenroth and Dodd, 1981). It was also reported that chl a is more sensitive than chl b and sensitivity of chlorophylls to SO<sub>2</sub> changed as the growing season progressed, indicating cumulative effects.

In areas where  $\text{SO}_2$  is the predominant air pollutant Rabe and Kreeb (1979) have reported that total chlorophyll content of the plants growing in the area is adversely affected.

Particulate matter: Reduction in the chlorophyll content was observed in plants sprayed with particulate matter like cement dust, coal dust and petro-coke dust (Lerman, 1972; Auclair, 1976; Fluckinger et al., 1978; Singh and Rao, 1978; Prasad and Rao, 1981).

However, in Triticum aestivum, Singh and Rao (1980) have shown that plants growing in the vicinity of cement factory had less chl a while the chl b was more as compared to control plants. This has been attributed to the shading effect of the cement layer on the leaf surface, leading to enhanced synthesis of chl b.

Pawar et al. (1982) sprayed  $2 \text{ g m}^{-2} \text{ d}^{-1}$  of cement, coal dust and flyash on Hibiscus abelmoschush for 30 days and concluded that a greater reduction in chlorophyll content in plants sprayed with coal dust. However, flyash exposed plants exhibited an increase in the chlorophyll content.

SO<sub>2</sub> and particulate matter: Plants in the vicinity of coal fired power plants, have been shown to have a considerably low amounts of chlorophyll content (Varshney and Garg, 1980; Dubey et al., 1982; Pandey, 1983).

Dubey et al. (1983) have also observed an increase in chlorophyll content in Cicer arietenum plants exposed to flyash (2 g m<sup>-2</sup> d<sup>-1</sup>). The chlorophyll content however, decreased in plants exposed to combination of SO<sub>2</sub> (0.5 ppm m<sup>-2</sup> d<sup>-1</sup>) and flyash (2 g m<sup>-2</sup> d<sup>-1</sup>) and SO<sub>2</sub> alone (0.5 ppm m<sup>-2</sup> d<sup>-1</sup>).

At biochemical level, most of the studies have been conducted on enzymes to evaluate the effect of SO<sub>2</sub> exposure artificially using its hydration products like HSO<sub>3</sub><sup>-1</sup> and SO<sub>3</sub><sup>-2</sup> (Bailey and Cole, 1959; Zeigler, 1972, 1973). Inhibition of regulatory enzyme of CO<sub>2</sub> fixation and of electron transport chain have been documented in detail by Zeigler (1975) and Hallgren (1978). Under field conditions increase in the glutathione was reported (Grill et al., 1979). Some studies have shown that protein synthesis and mineral contents in plants exposed to pollutants are also affected (Arndt, 1970; Ballantype, 1973; Singh and Rao, 1978; Prasad and Rao, 1981; Dubey et al., 1982; Pandey, 1983).

## Epidermal Features

Epidermal features like cuticular (wax) surface, epidermal cells, stomatal morphology (guard cells, subsidiary cell complex, abnormal stomata), stomatal density, stomatal pore size, density and length of trichomes have been shown to be affected by air pollutants. Results of the studies carried out on the effects of air pollutants on the epidermal features of plant leaves are summarized in the table 1.5.

### Cuticular (wax) surface

Cuticle is the outer most surface of the leaf and studies in relation to its morphology, development and influence of certain environmental factors have been summarized by Cuttler et al. (1982). In relation to air pollutants changes in the waxy material of cuticular surface have been observed. Exposure of Beta vulgaris to polluted air containing solar irradiated auto exhaust (smog) promoted excessive extrusion of waxy material forming irregular spots and rodlets on the leaf surface (Bystrom et al., 1968). Scanning electron microscope studies of the leaves of Lolium perenne L. exposed to  $417 \text{ ug m}^{-3}$   $\text{SO}_2$  for 23 days have revealed that leaf surfaces of exposed plants had conspicuously more wax, especially



in white necrotic areas as compared to control plants (Koziol and Cowling, 1981).

#### Epidermal cells

In Calotropis procera and Syzygium cumini the density of epidermal cells on both leaf surfaces was more in plants growing in the vicinity of a cement factory (Mirzapur, UP) as compared to plants from relatively non-polluted areas (Yunus and Ahmed, 1979). Density of epidermal cells was found to increase on both leaf surfaces of Tabernaemontana coronaria and Ipomia fistulosa plants growing near industrial areas (Srivastava et al., 1980; Yunus et al., 1982). Evans et al. (1979) have reported that simulated acid rain treatment (pH 5.7 to 2.3, 10 rainfall, a single 20 min rainfall daily) causes lesions in Tradescantia sp., Pteridium aquilium, Quercus palustris and Glycine max.

On the other hand, the density of epidermal cells was found to be reduced in Psidium guajava plants growing near a cement factory (Yunus and Ahmed, 1979). The authors have not specified the leaf surface. Survival of epidermal cells was reduced on adaxial (13.2%) and abaxial (10.4%) leaf surfaces in Calotropis gigantea

in plants growing in polluted urban areas of Waltair (AP) (Bhiravamurthy and Kumar, 1983).

Some studies have shown changes in the thickness of cell walls of epidermal cells in plants growing in polluted areas (Table 1.5). Scanning electron micrographs of both leaf surfaces of Aesculus hippocastanum have revealed that cell walls of epidermal cells are thin in plants collected from the neighbourhood of a coke plants, Netherlands (SO<sub>2</sub> and particulate matter) as compared to the plants of non-polluted areas. It was also noticed that the normal folds of the epidermal cells on the abaxial leaf surface were replaced by rather much larger folds (Godzik and Sassen, 1978). In Vicia faba plants exposed to SO<sub>2</sub> (50-500 ug m<sup>-3</sup>) epidermal cell walls on both leaf surfaces became corrugated and in some cases epidermal cells collapsed totally (Black and Black, 1980).

#### Stomatal morphology

A stoma consists of two bean shaped guard cells, enclosing stomatal pore. The stoma is surrounded by a group of 2-8 subsidiary cells and their shape and arrangement varies in different plant species.

Guard cells: Extensive guard cell injury was observed on both leaf surfaces in Phaseolus vulgaris after exposure to 1.4 ppm SO<sub>2</sub>, in an experimental set up (Paul and Long, 1975). A recent study on the effect of SO<sub>2</sub> on guard cells in Vicia faba indicated that exposure to 500 ug m<sup>-3</sup> of SO<sub>2</sub> or above causes structural disorganisation or death of one or both guard cells on both leaf surfaces (Black and Black, 1980). Bhairavamurty and Kumar (1983) observed aborted or crippled guard cells. They have also reported that survival of guard cells decreased by 70 and 84% at adaxial and abaxial leaf surfaces in Calotropis gigantea growing in polluted urban areas (Table 1.5).

In some cases abnormal stomata have been reported in leaf samples collected from polluted areas (Table 1.5). In scanning electron micrographs, of both leaf surfaces of Aesculus hippocastanum plants collected from a polluted area (SO<sub>2</sub>, particulate matter) show abnormal stomata, however, dust on or near stomatal pore was not seen (Godzik and Sassen, 1978). Abnormal stomata formed due to the abortion of one or both guard cells in Ricinus cummunis (Yunus and Ahmed, 1979). Distorted stomata were seen in Croton sparsiflorus plants collected from polluted

industrial areas (Srivastava et al., 1982). However, these authors have not specified the leaf surface.

**Subsidiary cells:** The number of subsidiary cells in leaf samples of Acer saccharum collected from polluted area was same as in plants of non-polluted areas (Sharma, 1975). Subsidiary cell complex was not affected in Calotropis procera collected from the area suffering from the area suffering from the pollution by particulate matter, SO<sub>2</sub>, CO and other oxides (Yunus and Ahmed, 1979). A sharp reduction was observed in the proportion of living subsidiary cells on both leaf surfaces in Vicia faba plants exposed to 50 ug m<sup>-3</sup> of SO<sub>2</sub> (Table 1.5). Increase in the concentration from 50 to 500 ug m<sup>-3</sup> further reduced the proportion of the living subsidiary cells (Black and Black, 1979).

#### Stomatal index

Bhiravamurty and Kumar (1983) observed about 10% reduction in stomatal index on both leaf surfaces of Calotropis gigantea collected from polluted urban areas (Table 1.5).

#### Stomatal density

Sharma and Butler (1973) have observed that stomatal density on both adaxial and abaxial leaf surfaces decreased

by 40 and 18% respectively in Trifolium repens plants growing in area of Tennessee, USA, suffering from pollution of SO<sub>2</sub> and particulate matter. The stomatal density reduced by 48% on adaxial and 27% at abaxial leaf surfaces in Trifolium pratense growing in polluted area. Sharma (1975) observed 88% reduction (from 50.3 to 6.3) at abaxial surface in Acer saccharum plants collected from polluted areas in Montreal (Canada). Stomata were however absent on adaxial leaf surface (Table 1.5). Stomatal density was reduced by about 15% on both leaf surfaces of Arenaria patula and on abaxial surface of Lonicera japonica collected from the vicinity of a zinc smelter in Pennsylvania (USA) where Zn, Cd, Pb, Cu, etc. and SO<sub>2</sub> were prominent pollutants (Caiazza and Quinn, 1980). Reduced stomatal density was observed on both leaf surfaces in Achyranthus aspera, Brassica oleracea, Chenopodium album, Ricinus communis, Sonchus asper and Withania somnifera and on adaxial leaf surface in Calotropis procera and Lantana camera growing in the vicinity of a thermal power plant in New Delhi. Stomatal density was less on both leaf surfaces in Cicer arietenum, Dolichus lablab, Lens culinaris, Phaseolus aureus and Vigna sinenses plants kept in the vicinity of a thermal power plant in Delhi

(Garg, 1979). Stomatal density both on adaxial and abaxial leaf surfaces was reduced in Calotropis gigantea growing in polluted areas (Bhairavamurthy and Kumar, 1983).

However, few reports indicate that stomatal density was more in plants growing in polluted areas (Table 1.5). Yunus and Ahmed (1979) have reported that stomatal density was more in Calotropis procera collected from polluted environment. However, leaf surface has not been specified in this study. Examination of eighty five leaf samples of Ricinis communis, collected from the non-polluted and polluted environment (main pollutant, cement dust from Churk Cement Factory, Mirzapur, UP), on examination revealed a significant increase in stomatal density on both adaxial and abaxial leaf surfaces, in samples collected from polluted areas. Stomatal density was higher in leaf samples of Syzygium cummuni and P. guajava collected from the vicinity of a cement factory as compared to the plants from relatively non-polluted areas (Yunus and Ahmed, 1979). The stomatal density was high on both leaf surfaces in Tabernaemontana coronaria and Ipomea fistulosa collected from polluted urban areas (Srivastava et al., 1980; Yunus et al., 1982).

In all 27 plant species have been investigated (Table 1.6) to evaluate the changes in stomatal density under air pollution stress. It was noticed that out of these, 19 plant species had low stomatal density, while in 7 plant species there was an increase in stomatal density. There was no change in the remaining one species.

#### Stomatal pore size

The air pollutants have been shown to affect the size of stomatal pore adversely (Table 1.5). Plants of Trifolium repens collected from areas polluted by SO<sub>2</sub> and particulate matter were found to have reduced stomatal pore, both on adaxial (20%) and abaxial (7%) leaf surfaces (Sharma and Butler, 1973). In leaf samples of Trifolium pratense, stomatal pore size decreased (8%) at the abaxial surface (Sharma and Butler, 1975). Length and breadth of stomatal pore reduced on adaxial surface by 28 and 21% respectively in Withania somifera and 30% and 26% in Brassica oleracea, and on abaxial leaf surface by 27% and 20% in Chenopodium album and 18 and 10% in Dolichus lablab due to pollution stress from power plant (Garg and Varshney, 1980). In addition, reduction in the length and breadth of stomatal pore on both leaf surfaces has been also reported in Achyranthus asper, Lantana camera,

Ricinus communis, Sonchus asper, Cicer arietenum, Lens culinaris, Phaseolus aureus and Vigna sinenses. Such changes were also observed at adaxial leaf surface in Calotropis procera, Chenopodium album and Dolichus lablab and on abaxial leaf surface of Brassica oleracea and Withania somnifera (Garg, 1979). However, there was no change in Cynodon dactylon on any of the leaf surfaces and in Calotropis procera (Garg, 1979). Size of a stomatal pore was less on both leaf surfaces in Tabernaemontana coronaria, but no change was observed in stomatal pore size in Croton sparsiflorus collected polluted urban areas (Srivastava et al., 1980, 1982). On the other hand, the stomatal pore size increased at the adaxial leaf surface in Trifolium pratense, collected from polluted area (Sharma and Butler, 1975).

Data in table 1.6 reveals that stomatal pore size was measured in 18 plant species growing in polluted areas. Out of these 16 have shown reduction in stomatal pore size.

#### Trichome density

Most of the workers have reported an increase in trichome density in plants exposed to polluted environment (Table 1.5). Leaf samples of Trifolium repens, from polluted



sites, have more than twice the trichomes on both adaxial and abaxial leaf surfaces as compared to the leaf samples of comparatively non-polluted areas. It has also been reported that the multicellular type of trichomes were relatively more abundant as compared to unicellular type in plants of polluted areas (Sharma and Butler, 1973). Three times higher trichome density has been observed on abaxial leaf surface in Trifolium pratense (Sharma and Butler, 1975). In Acer saccharum plants (surface not specified) growing in polluted areas in trichome density increased manifold (Sharma, 1975). Trichome density also increased in Calotropis procera and Psidium quajava collected from polluted areas, however, the authors have not specified the leaf surface (Yunus and Ahmed, 1979). Trichome density increased significantly in case of both leaf surfaces in Arenaria patula and adaxial surface of Lonicera japonica (Caiazza and Quinn, 1980). Trichome density increased on adaxial leaf surface of Lantana camera and Sonchus asper by 83% and 50% respectively and on abaxial leaf surface of Cicer arietenum by 31% due to pollution stress in the vicinity of a power plant (Garg and Varshney, 1980). An increase in trichome density on both leaf surfaces <sup>was observed</sup> in Achyrathus aspera, Brassica oleracea, Calotropis procera, Chenopodium album,

Cynodon dactylon, Ricinus communis, and Withania somnifera, Dolichus lablab, Lens culinaris, Phaseolus aureus and Vigna sinenses. Increase was also noticed on adaxial leaf surface of Sonchus asper and abaxial leaf surface of Lantana camera and Cicer arietenum (Garg, 1979). In Ipomia fistulosa trichome density was found to be higher on both leaf surfaces in plants colled from polluted areas (Yunus et al., 1982).

A perusal of data in table 1.6 show that 22 plant species were studied for change in trichome density due to air pollutants stress. All of them have exhibited an increase in the trichome density.

#### Trichome length

Studies on the effect of air pollutants have indicated that trichome length increased under pollution stress (Table 1.5). It increased in leaf samples of Trifolium repens, Trifolium pratense collected from polluted areas by 9 and 40% respectively as compared to plants from relatively non-polluted areas. It is not clear from the results that whether observation relate to adaxial or abaxial surface (Sharma and Butler, 1973, 1975). In Acer saccharum trichome length increased significantly (Sharma, 1975). Trichome length increased on adaxial

leaf surface by 25% and 70% in Lantana camera and Sonechus asper respectively, and 26% on abaxial leaf surface of Cicer arietenum. In addition, <sup>increase at</sup> both leaf surfaces of plants viz., Achyranthus aspera, Brassica oleracea, Calotropis procera, Chenopodium album, Cynodon dactylon, Ricinus communis, Withania somnifera, Dolichus lablab, Lens culinaris, Phaseolus aureus and Vigna sinenses was observed. Such changes were also noticed at adaxial leaf surfaces of Cicer arietenum and abaxial leaf surface of Lantana camera and Sonchus asper due to impact of pollutants from power plants (Garg, 1979). Longer trichome on both leaf surfaces were observed in Ipomea fistulosa plants collected from polluted areas (Yunus et al., 1982).

It is clear from the table 1.6 that trichome length increases in all the 17 plant species in response to air pollutants.

A total evaluation of the studies carried out on epidermal features under pollution stress indicate (Table 1.6) that trichomes (density and length) are more sensitive as compared to stomata (stomatal pore size and density).

Table 1.5: A summary of effects of air pollution on leaf epidermal features

Epidermal Features	Species	Leaf surface	Observation	Reference
1	2	3	4	5
Cuticular (Wax) surface	Beta vulgaris	Ad	Inc.	1
		Ab	Inc.	1
	Lolium perenne	Ad	Inc.	2
		Ab	Inc.	2
<b>Epidermal Cells</b>				
i) Density	Calotropis procera	Not spec.	Inc.	3
	Syzygium cumini	"	Inc.	3
	Tabernaemontana coronaria	Ad	Inc.	4
		Ab	Inc.	4
	Ipomia fistulosa	Ad	Inc.	3a
		Ab	Inc.	3a
	Psidium guajava	NS	Dec.	3
	Calotropis gigantea	Ad	Dec. (% survival)	11
		Ab	Dec. (% survival)	11
	ii) Cell wall	Aesculus hippocastanum	Ad	Dec. (thickness)
Ab			Dec (thickness)	5
Vicia faba		Ad	Corrugation of cell walls	6
		Ab	Corrugation of cell walls	6
<b>Stomatal Morphology</b>				
i) Guard cells	Phaseolus vulgaris	Ad)	Injury to	10
		Ab)	guard cells	
	Vicia faba	Ad)	Structural	6
		Ab)	disorganisa- tion of one or both or "death of guard cells Dec. (% survival)	

Ad - Adaxial; Ab - Abaxial; Inc. - Increase; Dec. - Decrease; NS - Not specified.

Table 1.5 (contd...)

1	2	3	4	5
	Calotropis procera	Ad) Ab)	Aborted or cripped guard cells Dec. (% survival)	11
	Aesculus hippocastanum	Ad) Ad	Abnormal stomata	5
	Ricinus cummunis	NS	Aborted one or both guard cells	3
	Croton sparsiflorus	NS	Distorted stomata	4a
ii) Subsidiary cells	Acer saccharum	NS	No change	7b
	Calotropis procera	NS	No change	3
	Vicia faba	Ad) Ab)	Dec. (% survival)	6
Stomatal Index	Calotropis gigantea	Ad Ab	Dec. Dec.	11 11
Stomatal Density	Trifolium repens	Ad Ab	Dec. Dec.	7 7
	Trifolium pratense	Ad Ab	Dec. Dec.	7a,7b 7a,7b
	Acer saccharum	NS	Dec.	7c
	Arenarea patula	Ad Ab	Dec. Dec.	8 8
	Lonicera japonica	Ab	Dec.	8
	Calotropis procera	Ad Ab	Dec. Dec.	11 11
	Achyranthus aspera	Ad Ab	Dec. Dec.	
	Brassica oleracea	Ad Ab	Dec. Dec.	
	Calotropis procera	Ad Ab	Dec. No change	
	Chenopodium album	Ad Ab	Dec. Dec.	9 and 9a
	Cynodon dactylon	Ad Ab	No change No change	

Table 1.5 (contd...)

1	2	3	4	5
	Lantana camera	Ad Ab	Dec No change	
	Ricinus cumminis	Ad Ab	Dec. Dec.	
	Sonchus asper	Ad Ab	Dec. Dec.	
	Withania somnifera	Ad Ab	Dec. Dec.	9 and
	Cicer arietenum	Ad Ab	Dec. Dec.	9a
	Dolichus lablab	Ad Ab	Dec. Dec.	
	Lens culinaris	Ad Ab.	Dec. Dec.	
	Phaseolus aureus	Ad Ab	Dec. Dec.	
	Vigna sinensis	Ad Ab	Dec. Dec	
	Calotropis procera	NS	Inc.	3
	Ricinus cimmunis	Ad Ab	Inc. Inc.	3 3
	Syzygium cuminii	NS	Inc.	3
	Psidium guajava	NS	Inc.	3
	Tabernaemontana coronaria	Ad Ab	Inc. Inc	4 4
	Ipomia fistulosa	Ad Ab	Inc. Inc.	3a
	Croton sparseflonus	NS	Inc.	4a
Stomatal Pore Size	Trifolium repens	Ad Ab	Dec. Dec.	7
	Trifolium pratense	Ad Ab	Inc. Dec.	7a
	Achyranthus aspera	Ad Ab	Dec. Dec.	9 and
	Brassica oleracea	Ad Ab	Dec. Dec.	9a

Table 1.5 (contd...)

1	2	3	4	5
	Calotropis procera	Ad Ab	Dec. No change	
	Chenopodium album	Ad Ab	Dec. Dec.	
	Cynodon dactylon	Ad Ab	No change No change	
	Lantana camera	Ad Ab	Dec. Dec.	
	Ricinus cumminis	Ad Ab	Dec. Dec.	
	Sonchus asper	Ad Ab	Dec. Dec.	
	Withania somnifera	Ad Ab.	Dec. Dec.	9 and 9a
	Cicer arietenum	Ad Ab	Dec. Dec.	
	Dolichus lablab	Ad Ab	Dec. Dec.	
	Lens culinaris	Ad Ab	Dec. Dec.	
	Phaseolus aureus	Ad Ab	Dec. Dec.	
	Vigna sinensis	Ad Ab	Dec. Dec.	
	Tabernaemontana coronaria	Ad Ab	Dec Dec.	4
	Croton sprasefloxus	NS	No change	4a
Trichome Density	Trifolium repens	Ad Ab	Inc. Inc.	7 7
	Trifolium pratense	Ab	Inc.	7a
	Acer saccharum	NS	Inc.	7b
	Calotropis procera	NS	Inc.	3
	Psidium guajava	NS	Inc.	3
	Arenaria potula	Ad Ab	Inc. Inc.	8 8
	Lonicera japonica	Ad	Inc.	8

Table 1.5 (contd...)

1	2	3	4	5
	Achyranthus aspera	Ad Ab	Inc. Inc.	
	Brassica oleracea	Ad Ab	Inc. Inc.	
	Calotropis procera	Ad Ab	Inc. Inc.	
	Chenopodium album	Ad Ab	Inc. Inc.	
	Cynodon dactylon	Ad Ab	Inc. Inc.	
	Lantana camera	Ad Ab	Inc. Inc.	
	Ricinus cumminis	Ad Ab	Inc. Inc.	
	Sonchus asper	Ad Ab		9 and 9a
	Cicer arietenum	Ad Ab	Inc. Inc.	
	Withania somnifera	Ad Ab	Inc. Inc.	
	Dolichus lablab	Ad Ab	Inc. Inc.	
	Lens culinaris	Ad Ab	Inc. Inc.	
	Phaseolus aureus	Ad Ab	Inc. Inc.	
	Vigna sinensis	Ad Ab	Inc. Inc.	
	Ipomia fishulosa	Ad Ab	Inc. Inc.	3a
Trichome Length	Trifolium repens	NS	Inc.	7
	Trifolium pratense	NS	Inc.	7a
	Acer saccharum	NS	Inc.	7b
	Achyranthus aspera	Ad Ab	Inc. Inc.	9 and 9a
	Brassica oleracea	Ad Ab	Inc. Inc.	



1	2	3	4	5
	Calotropis procera	Ad Ab	Inc. Inc.	
	Chenopodium album	Ad Ab	Inc. Inc.	
	Cynodon dactylon	Ad Ab	Inc. Inc.	
	Lantana camera	Ad Ab	Inc. Inc.	
	Ricinus cumminis	Ad Ab	Inc. Inc.	
	Sonchus asper	Ad Ab	Inc. Inc.	
	Withania somnifera	Ad Ab	Inc. Inc.	9 and 9a
	Cicer arietenum	Ad Ab	Inc. Inc.	
	Dolichus lablab	Ad Ab	Inc. Inc.	
	Lens culinaris	Ad Ab	Inc. Inc.	
	Phaseolus aureus	Ad Ab	Inc. Inc.	
	Vigna sinensis	Ad Ab	Inc. Inc.	

## References:

1. Bystrom et al., 1968
2. Kaziol and Cowling, 1981
3. Yunus and Ahmed, 1979
- 3a. Yunus et al., 1982
4. Srivastava et al., 1980
- 4a. Srivastava et al., 1982
5. Godzik and Sassen, 1978
6. Black and Black, 1979
7. Sharma and Butler, 1973
- 7a. Sharma and Butler, 1975
- 7b. Sharma, 1975
8. Caiazza et al., 1980
9. Garg, 1979
- 9a. Garg and Varshney, 1980
10. Paul and Long, 1975
11. Bhairavamurty and Kumar, 1983.

Table 1.6: Number of Plants studied for their responses to air pollutants in relation to epidermal features

Epidermal features	Total number of plants studied	Increase in parameter	Decrease in parameter	No change in parameter
Cuticular (Wax) surface	2	2	-	-
Epidermal cells				
i) Density	6	4	2	-
ii) Cell wall thickness	2	-	2	-
Stomatal morphology				
i) Guard cells	6	-	6	-
ii) Subsidiary cell	3	-	1	2
Stomatal Index	1	-	1	-
Stomatal density	27	7	19	1
Stomatal pore size	18	1	16	1
Trichome density	22	22	-	-
Trichome length	17	17	-	-

### Leaf Diffusive Resistance

Diffusive resistance of leaf signifies the rate of exchange of gases and water vapour between the leaf and atmosphere. The inverse of the leaf resistance is the leaf conductance, which according to Jarvis (1982) could be defined as the proportionality parameter relating flux of a property in gas phase in or out of a leaf to the driving force existing between the leaf and bulk air outside the leaf boundary layer. The property might be water vapour,  $\text{CO}_2$ , or pollutant such as gaseous  $\text{SO}_2$ .

The flux of water in transpiration,  $F$  for example, is proportionate to the difference in specific humidity,  $\Delta q$  ( $\text{Kg Kg}^{-1}$ ) between leaf and air.

$$F = g \Delta q \rho$$

$\rho$  is density of air ( $\text{kg m}^{-3}$ ),

$g$  is leaf conductance ( $\text{ms}^{-1}$ ).

Leaf conductance can be partitioned into:

Cuticular - Boundary layer cuticle,

Stomatal - Stomatal ante-chamber, stomatal pore, sub-stomatal cavity, etc.

However, for most leaves, the largest part of gaseous flux is through the stomatal pore and consequently the leaf conductance can generally be regarded as

almost synonymous to stomatal conductance, in higher plants. Flower and Unsworth (1974), Fowler (1982) have studied the  $\text{SO}_2$  fluxes in a wheat field and have evaluated relative importance of stomata, the cuticle and soil layer as sinks for  $\text{SO}_2$ . In daytime, before senescence, 60% of  $\text{SO}_2$  flux is absorbed through stomata and most of the remaining 40% is absorbed by cuticle (soil uptake accounts for at most 10% of total flux). When stomatal conductance is infinite, then mesophyll resistance plays an important role. According to the study of Garland and Branson (1977) who used  $^{35}\text{SO}_2$ , there is no mesophyll resistance to  $\text{SO}_2$  comparable to that of  $\text{CO}_2$ .

Studies relating to the effects of air pollutants on leaf resistance have been conducted mainly with  $\text{SO}_2$  exposure in artificial conditions as summarized by Black and Unsworth (1981). Solberg and Adams (1956) have reported that abaxial leaf surface exhibits comparatively more visual injury symptoms as compared to adaxial leaf surface when exposed to 0.5 ppm  $\text{SO}_2$ . The differential injury of the adaxial and abaxial leaf surfaces may be due to quantitative variation in  $\text{SO}_2$  fluxes passing through the two leaf surfaces because the stomatal density is greater on the abaxial leaf surface. Stomatal opening

was observed at 0.5 ppm SO<sub>2</sub> in Vicia faba L. when atmospheric relative humidity was greater than 40% at 18°C (water vapour deficient less than 7 mm Hg), while closure of stomatal was at lower humidities (Allaway and Mansfield, 1969; Majernik and Mansfield, 1970, 1971). Mansfield and Majernik (1970) observed that 0.25 to 1.0 ppm of SO<sub>2</sub> enhanced the rate of opening of stomata in treated plants in light, but in dark the stomata in treated plants took longer time to close fully.

Unsworth et al. (1972) studied the variation in diffusive resistance in Vicia faba and Zea mays plants exposed to 1 to 50 pphm of SO<sub>2</sub> for 30 min with humidity as variable factor. They observed that reduction in stomatal resistance was approximately same in plants exposed to different concentrations of SO<sub>2</sub> at 50-60% humidity, while in dry conditions, increase in resistance was observed. Biscoe et al. (1973) observed that resistance decreased by 20% in Vicia faba plants exposed to 72 to 1430 ug m<sup>-3</sup> of SO<sub>2</sub> for 24 h. He also reported that stomatal opening was more in older leaves in plants exposed to 29 ug m<sup>-3</sup> of SO<sub>2</sub> as compared to younger leaves. Diffusive resistance changed in Raphanus sp. and Perilla sp. when exposed to 2 ppm of SO<sub>2</sub>, but there was no change

in Arachis sp. and Lycopersicum sp. (Kondo and Sugahara, 1978). In Vicia faba plants, exposed to 0.02 to 0.2 ppm of SO<sub>2</sub> for 24 h diffusive resistance decreased by 20-25% (Black and Black, 1979). They observed that reduced sensitivity of SO<sub>2</sub> under low humidity condition is due to decreased SO<sub>2</sub> uptake. Biggs and Davis (1980a) took wide range of SO<sub>2</sub> concentrations, i.e., 0.3, 0.6, 0.9 and 1.2 ppm for 1, 2, 3 and 4 h to study the effect on diffusive resistance in Betula pendula (white birch), B. lutea Michx. (yellow birch) and B. populifolia Marsh. (gray birch). Stomatal resistance in white and yellow birch decreased after exposure to 0.3 ppm for 1 and 2 h and increased in response to higher doses of SO<sub>2</sub>. Stomatal resistance of gray birch increased only after exposure to 0.6 ppm for 1 and 3 h and decreased with respect to other doses. They also observed that exposure to 0.9 ppm for 2 h that relative susceptibility may be significantly correlated with pre-exposure leaf conductance rates of Betula nigra and B. papyrifera but not in B. pubescens (Biggs and Davis, 1980b). Effect of exposure to 0.25 to 9.0 ppm SO<sub>2</sub> have also been studied in relation to the diffusive resistance of Vicia faba L. (Olszyk and Tibbitts, 1981a,b). At 0.25 ppm, opening of stomata was observed during the first half of the photoperiod, while at 1.0 ppm, the stimulating

effect was apparent throughout the photoperiod and in the first two hours of the darkness, higher concentration upto 9.0 ppm causes little increase in the effect beyond that found at 1.0 ppm.

Stomata closure has been reported at higher doses of  $\text{SO}_2$  ( $\geq 500 \text{ ug m}^{-3}$ ) which may partly be due to accumulation of  $\text{SO}_2$  in substomatal cavities following  $\text{SO}_2$  inhibition of photosynthesis (Sij and Swanson, 1974) and partly due to changes in membrane permeability of the guard cells (Black and Black, 1979). Guard cells remain unaffected upto  $200 \text{ ug m}^{-3}$  of  $\text{SO}_2$ . At high concentrations, their disorganization or death of one or both of the guard cells have been frequently observed at or above  $500 \text{ ug m}^{-3}$  of  $\text{SO}_2$  (Black and Black, 1979).

Barton et al. (1980) have shown that higher concentration in the range of 0.5 to 2.0 ppm  $\text{SO}_2$  in Phaseolus vulgaris L. cause increase in mesophyll resistance and not the stomatal resistance.

Except for few exceptions, ozone increases diffusive resistance of Phaseolus vulgaris, soybean and radish leaves (Hill and Littlefield, 1969; Beckerson and Hofstra, 1979a,b). Exposure to  $\text{SO}_2$  and  $\text{O}_3$  also increases diffusive resistance (Beckerson and Hofstra, 1979a,b; Hofstra and Beckerson, 1981; Olszyk and Tibbitts, 1981a,b).

Czaja (1960) observed that stomata of conifers may be plugged by dust particles preventing normal exchange of gases. However, Lerman (1972) observed with the help of scanning electron microscope that only few stomatal pores are clogged on the upper and lower leaf surfaces of bean plants dusted daily with  $6.64 \text{ g m}^{-2}$ . Ricks and Williams (1974) observed in Quercus petraea plant leaves had particles on the lower surface which interfered with normal stomatal behaviour, which led to reduction in maximal diffusive resistance in plants growing in the vicinity of Phurnacite fuel stations (South Wales). He suggested that reduction in diffusive resistance may enhance  $\text{SO}_2$  uptake.

In most of the studies conducted on affect of  $\text{SO}_2$  on plants for short term duration have revealed that diffusive resistance gets decreased thereby allowing the polluted air to enter into the leaf at much faster rate.



### Optical Characteristics of Leaf

Variation in optical characteristics of leaf as a result interaction of air pollutants has not been worked out. This aspect may play a significant role in plant life as revealed by Eller (1977) that in a dusted leaf of Rhododeudron catawbiense growing along the road side, absorptivity of infrared (700-1350 nm) radiation was more than double as compared to leaf from non-polluted area, thereby causing an increase in leaf temperature by 2 to 4°C.

\*\* In view of the lack of comprehensive evaluation mainly on optical characteristics, diffusive resistance (long-term) and epidermal features (laboratory conditions), due to air pollution stress, the present study was carried out. Plant species were exposed both in field (in vicinity of IP Power Plant) and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash for 45 and 90 days and quantitative assessments were made for morphological parameters, epidermal features, diffusive resistance, leaf surface temperature and leaf absorbance.

## MATERIALS AND METHODS

## Plant Material

Three plant species, viz., Medicago sativa Linn. (a sensitive plant), Triticum aestivum Linn. (a less sensitive plant) and Zea mays Linn. (a resistant plant) varying in their sensitivity towards  $SO_2^*$  were selected (O'Gara, 1922; Thomas and Hill, 1935; Thomas and Hendrick, 1956).

Plants were raised from seeds in earthen pots (height 30 cm) filled with sandy loam soil. Five seeds were sown in each pot and 112 pots were prepared for each species. The experiments were started, when seeds were 12 days old. Pots were regularly irrigated during the experimental period. Observations were taken after 45 and 90 days of exposure in field or artificially to  $SO_2$ , flyash and a combination of  $SO_2$  and flyash.

Leaves at specific position of the plants were selected for measurements. Leaves at the 8th node of the main branch in Medicago sativa, at the 4th node of

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\*Medicago sativa: Cultivated during the cold season.  
Triticum aestivum: Cultivated during the cold season, as rabi crop.  
Zea mays: Cultivated during the rainy season, as kharif crop.

the central tillar in Triticum aestivum, and at the 4th node in Zea mays were selected.

#### Plant Performance

Leaves were closely examined to detect morphological foliar injury such as chlorosis, necrosis etc. Leaf area was calculated using centimeter graph paper. Biomass of fruit, leaves, stem and root was determined by drying in an electric oven at 80°C for 24 h and weighed using an electric balance.

#### Chlorophyll Measurement

Total chlorophyll content was estimated according to method given by Arnon (1949). One gram of leaf tissue was ground by pestle and mortar with a small quantity of acid washed sand and 80% acetone. The homogenate was centrifuged. Volume of the supernatant was made upto 100 ml with 80% acetone. The optical density of the chlorophyll extract was measured at 645 nm and 663 nm. The total amount of chlorophyll (mg/g) was calculated according to the following formula

$$20.2 (D_{645}) + 8.02 (D_{663}) \times \frac{V}{100 \times W}$$

where,  $D_{645}$  - OD at 645 nm

$D_{663}$  - OD at 663 nm

V - Total volume of extract

W - Weight of leaf tissue taken.

### Diffusive Resistance

Diffusive resistance of leaves was measured using a battery powered, diffusive resistance meter, with a horizontal sensor style (LI-60 Lambda Instrument Corporation, USA). Diffusive resistance was measured by recording the time ( $\Delta t$ ) required for a given quantity of water vapour to diffuse into the sensor cup and be absorbed by the humidity sensing element. The average time lapse ( $\Delta t$ ) was recorded for each standard set of holes on the horizontal plate, for which the resistance is known. A standard curve was plotted taking diffusive resistance on X-axis (abscissa) and  $\Delta t$  on Y-axis (ordinate)(Fig. 2.1).

$t$  was observed for different plant leaves and with the help of a standard curve the corresponding values of leaf diffusive resistance was calculated (Fig. 2.1).

### Epidermal Characteristics

A scanning electron microscope (SEM) was used to study epidermal features of plant leaves.

### Sample preparation

Collected leaves were washed with double distil water using soft camel hair brush. After gently wiping the leaves with tissue paper, they were placed with their adaxial surfaces facing upward and leaf samples of 2 mm

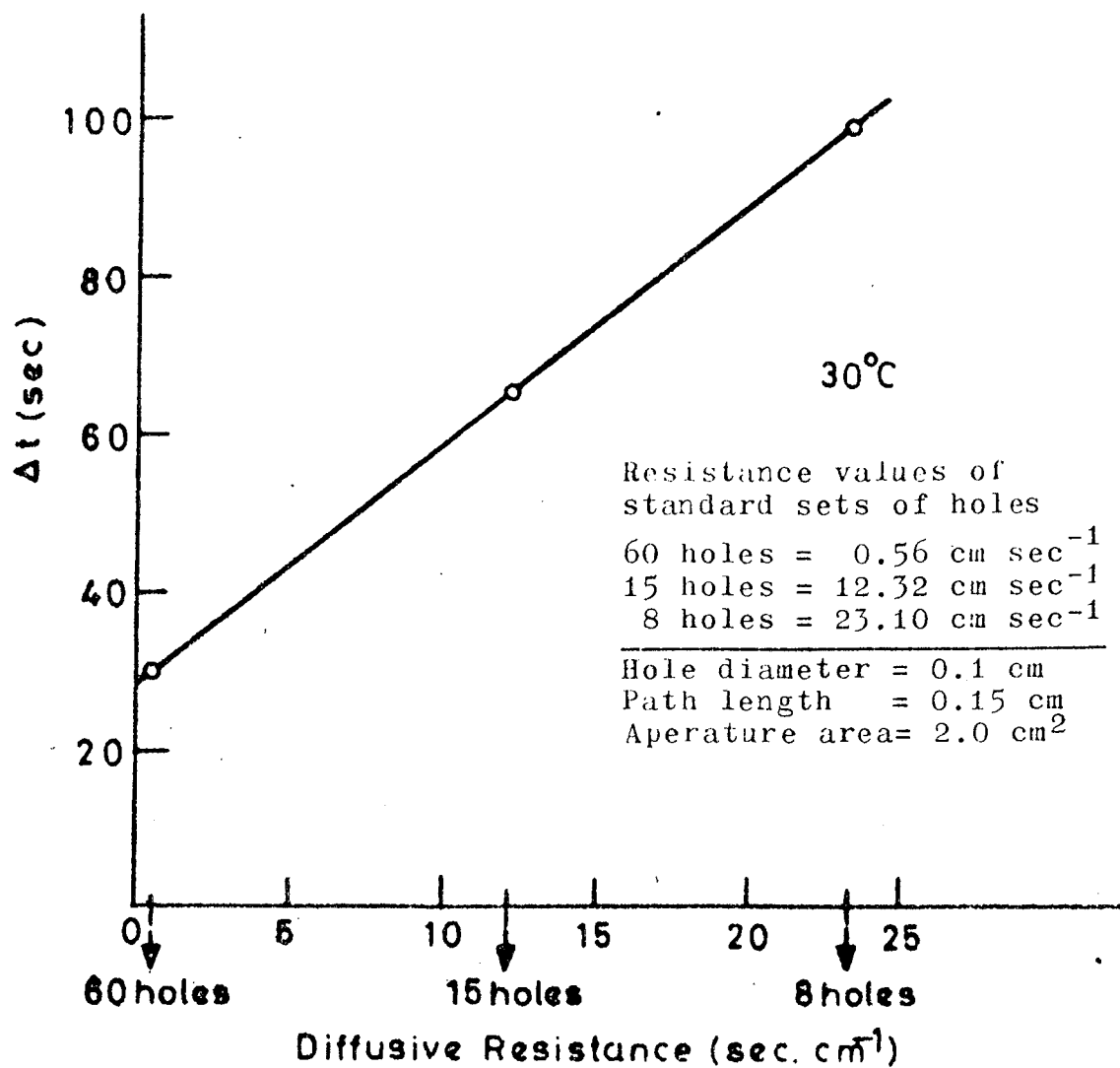


Fig. 2.1 Standard curve for diffusive resistance

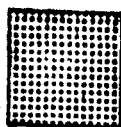
square were obtained. Two notches at the lower right hand side were made as shown in the Fig. 2.2. Thus, triangular gaps on the lower right hand side were helpful in identifying the adaxial and abaxial leaf surfaces during subsequent processing.

#### Fixation

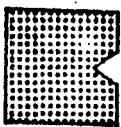
Leaf samples were fixed in 3% glutaraldehyde (45 min) in phosphate buffer (pH 7.2), post-fixed in one percent osmium tetroxide (45 min) (in phosphate buffer, pH 7.2), dehydrated in 35, 50, 85, 95 and 99% ethanol (three changes, five minutes each) placed in amyl acetate for 15 min (Krause, 1976). They were kept on specimen holder with quickfix, followed by a coating of 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 for both abaxial and adaxial leaf surfaces, were taken at magnification between 200 and 240, while photographs for stomata and trichomes were taken at magnifications ranging between 500 to 2000.

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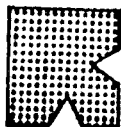
In the scanning electron microscope, used for present study, a negative gets reduced by five times of the magnification (x) indicating by the microscope. Positive prints were made by magnifying them 4.5 times. Thus the magnification of the positive photograph was calculated as  $\frac{9x}{10}$ .



Leaf sample was kept with  
adaxial 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 differentiating 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).

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After Garg (1979)

For calculating the density of stomata and trichomes, the area of the photograph was divided by the square of the magnification which provides the actual area of leaf scanned. The number of stomata and trichomes were counted from the photograph and density was calculated for unit area. Length and breadth of stomata and trichome length was determined by actual measurement from the photograph and dividing it with the magnification factor.

#### Leaf Surface Temperature

Leaf surface temperature was determined electrically based on thermo-couple principle. A surface temperature probe no. 409A (Yellow Spring Co., Ohio, USA) was used. The temperature sensing element housed in a probe is attached to a plastized vinyl jacketed shielded lead wire terminating in a phone plug. The phone plug was inserted into the temperature meter (Aplab Electronics, India) which gives direct reading of the surface temperature of the subject.

Following precautions were taken for the measurement of leaf surface temperature :

1. Leaf was kept with adaxial surface upwards on insulating surface like wood, without detaching it from the mother plant.



2. The knob of the temperature probe was touched with the surface of the leaf gently for about one min, in order to ensure a steady equilibrium state.

#### Absorption Spectra of Leaf

Absorption spectra of intact leaves (without washing etc) were measured both in visible (340-740 nm) and infrared (740-2500 nm) regions, using Shimadzu Seisa Kusho Spectrophotometer, model MPS-500 (Japan).

#### Absorption spectrum

Leaf samples were clipped in a leaf holder with adaxial surface facing the source of light and absorption pattern in visible and infra-red regions was recorded. Percentage absorbance at wave length representing maximum absorbance was calculated on the basis of optical density, using the formula  $(2 - \log (\text{transmission})) = \text{optical density}$ .

### Determination of SO<sub>2</sub>

Concentration of SO<sub>2</sub> was determined spectrophotometrically, using pararosaniline as an indicator (West and Gaeke, 1956). In a 25 ml volumetric flask, 10 ml of unexposed tetrachloromercurate (TCM) solution was added. Sampling was done by aspirating air for 30 min at the rate of 1.0 litre min<sup>-1</sup>, using a small air pump (Fig. 2.3 and 2.4), followed by addition of 1 ml 0.6% sulphamic acid. The solution was kept for 10 min. To this, 2 ml of 0.2% formaldehyde, 5 ml of 0.02% of pararosaniline solution, and distilled water were added to make the volume upto 25 ml. The solution was kept again for 30 min, for the pink colour to develop and optical density (O.D.) was measured at 548 nm.

Concentration of SO<sub>2</sub> (ug m<sup>-3</sup>) was calculated on the basis of standard graph (Fig. 2.10) and the formula as described below (Fig. 2.5)

$$SO_2 \text{ (ug m}^{-3}\text{)} = \frac{(A - A_0) 10^3 \times B_s}{V_r} \times D$$

where, A = O.D. of the sample

A<sub>0</sub> = O.D. of blank

B<sub>s</sub> = Calibration factor - ug SO<sub>2</sub>/unit of absorbance

D = Dilution factor

V<sub>r</sub> = Volume of air passed through the solution.

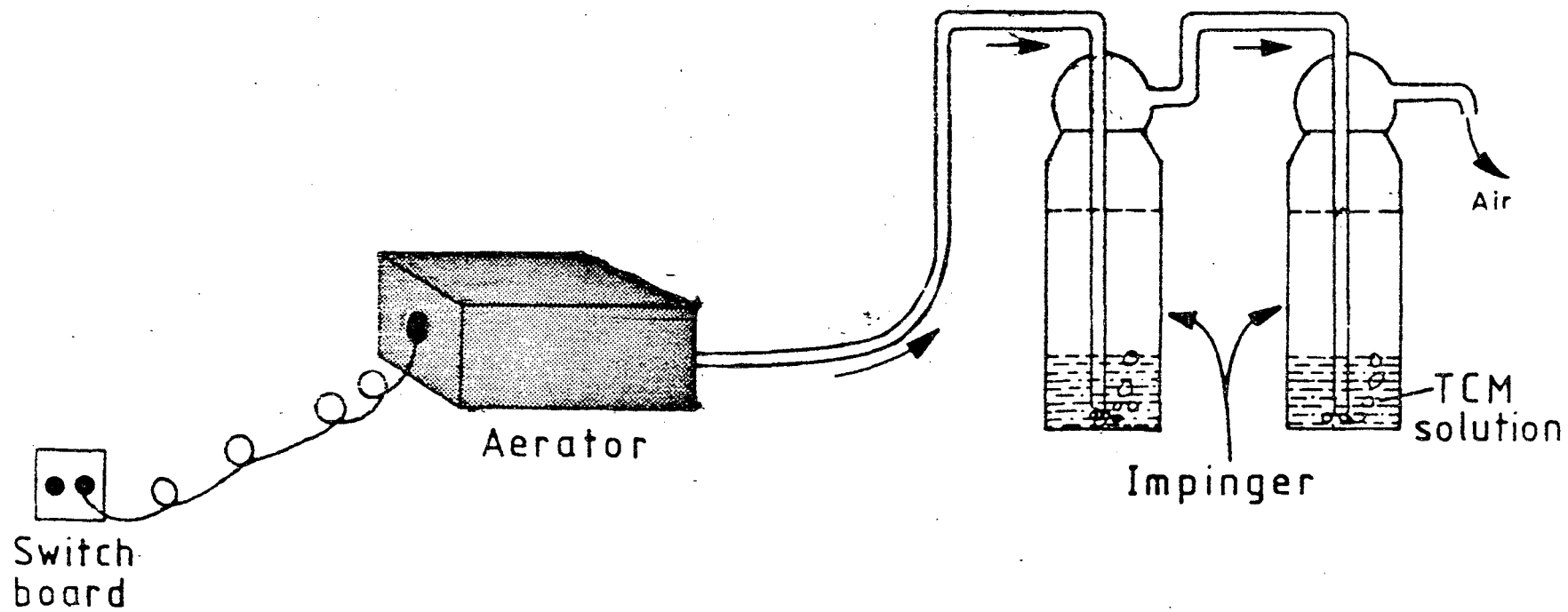


Fig. 2.4 Diagrammatic representation of the assembly used for scrubbing  $\text{SO}_2$  by TCM solution for determining  $\text{SO}_2$  concentration in the air

Fig. 2.3 Photograph showing SO<sub>2</sub> scrubbing set up for determining SO<sub>2</sub> concentration using TCM solution.



Here,

$$B_s = \frac{(A - B) N \times 32,000}{25} \times 0.02$$

where, A = volume of thiosulphate (blank)

B = volume of thiosulphate (sample)

N = normality of sample

32,000 = milliequivalent of  $\text{SO}_2$

0.02 = dilution factor

25 = volume of standard sulphite solution.

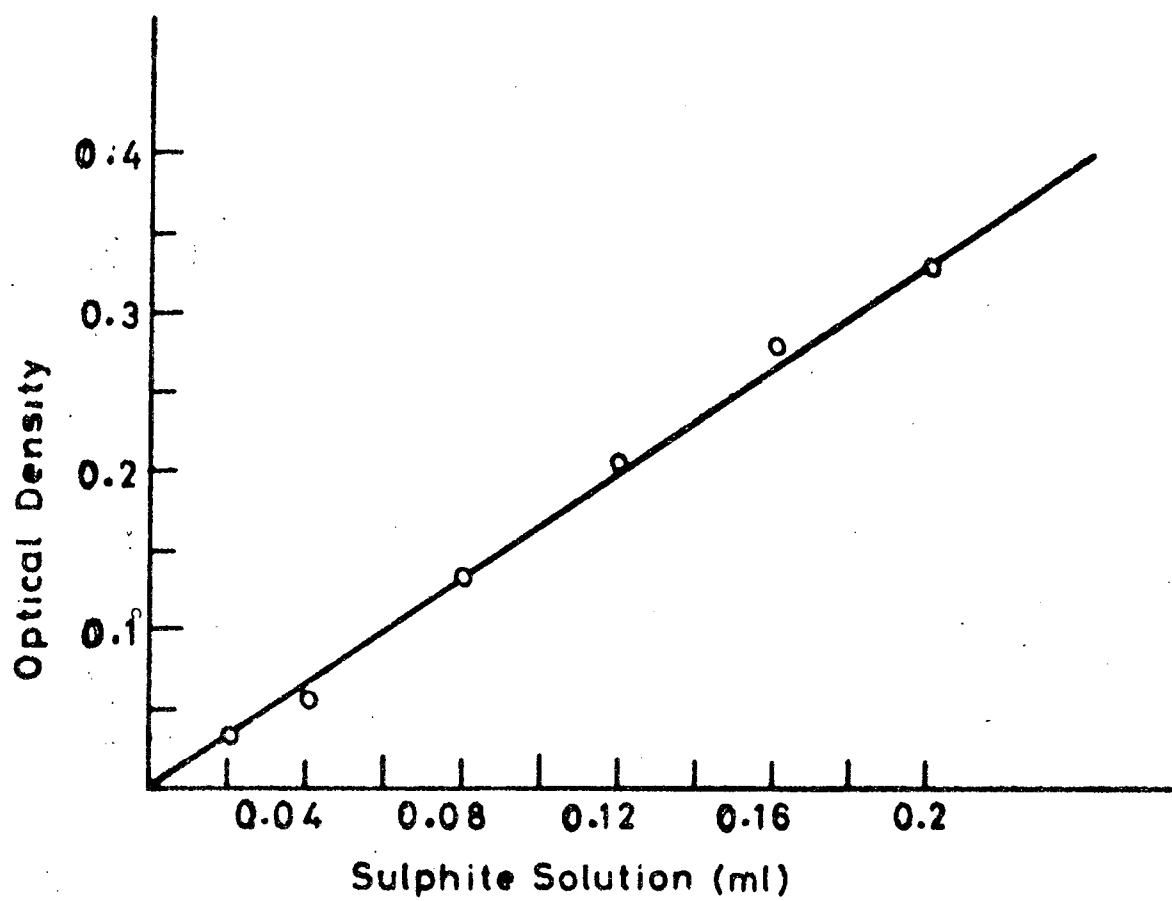


Fig. 2.5 Standard curve for sulphite solution for determining  $\text{SO}_2$  concentration.

### Experimental Study

The twelve days old seedlings were used for field and artificial exposure studies. Plants in field were exposed at a 'site' selected in the vicinity of the IP Power Plant, New Delhi (see Field Site Characteristics). Plants were exposed artificially to SO<sub>2</sub> and flyash separately and in combination. Plants kept in the ecological nursery at Jawaharlal Nehru University (JNU), New Delhi, which is comparatively free from pollution, served as control.

### Exposure Chamber

Plants were exposed artificially to SO<sub>2</sub> in dynamic 2 m<sup>2</sup> exposure chambers constructed in the ecological nursery, JNU. Sixteen pots arranged in 4 x 4 rows containing the experimental plants were buried upto the brim into the soil (Fig. 2.6). An iron rod frame of 2 m<sup>3</sup> was placed on the soil surface. The frame was covered by a transparent polythene cover (gauge 200), before starting the SO<sub>2</sub> exposure. The polythene cover was left free at the bottom to ensure free flow of air (Fig. 2.7).

### Source of SO<sub>2</sub>

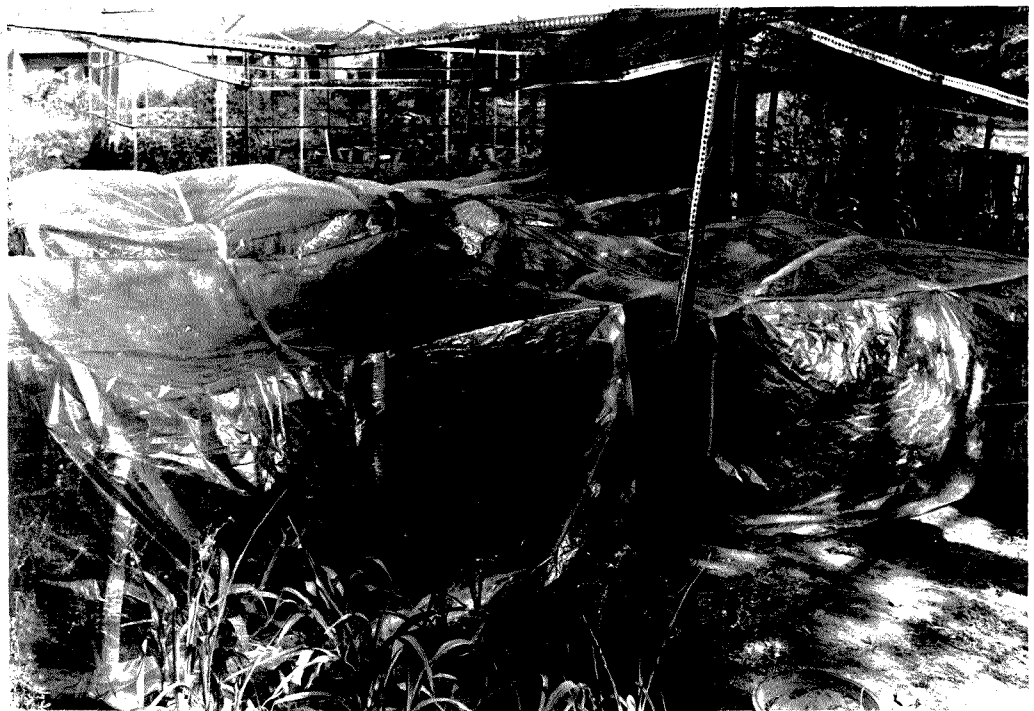
A SO<sub>2</sub> gas cylinder was the source of SO<sub>2</sub>. Gas was allowed to pass through a double stage regulator to ensure a



Fig. 2.6 Photograph showing 16 experimental pots in 4 x 4 rows and perforated PVC pipe network on the soil surface for uniform distribution of SO<sub>2</sub> in the exposure chamber.



Fig. 2.7 Chambers covered with transparent polythene cover for SO<sub>2</sub> exposure.



steady and desired flow of the gas.  $\text{SO}_2$  was diluted by ambient air using air blowers ( $1.5 \text{ m}^3 \text{ min}^{-1}$ ) and passed through a manifold with 4 outlets (Fig. 2.8 and 2.9). From each outlet the  $\text{SO}_2$  containing air was fed to a junction box (the central position of the perforated PVC pipe arrangement).

#### Distribution of $\text{SO}_2$

In order to achieve uniform distribution of  $\text{SO}_2$ , a perforated PVC pipe network was laid at the soil surface of the fumigation chamber. It consisted of 12 pipes each of 90 cm length. These were used with suitable connectors to form a square with a cross connecting each arm of the square measuring 180 cm. A junction box was placed in the center with 4 outlets as shown in Fig. 2.6.

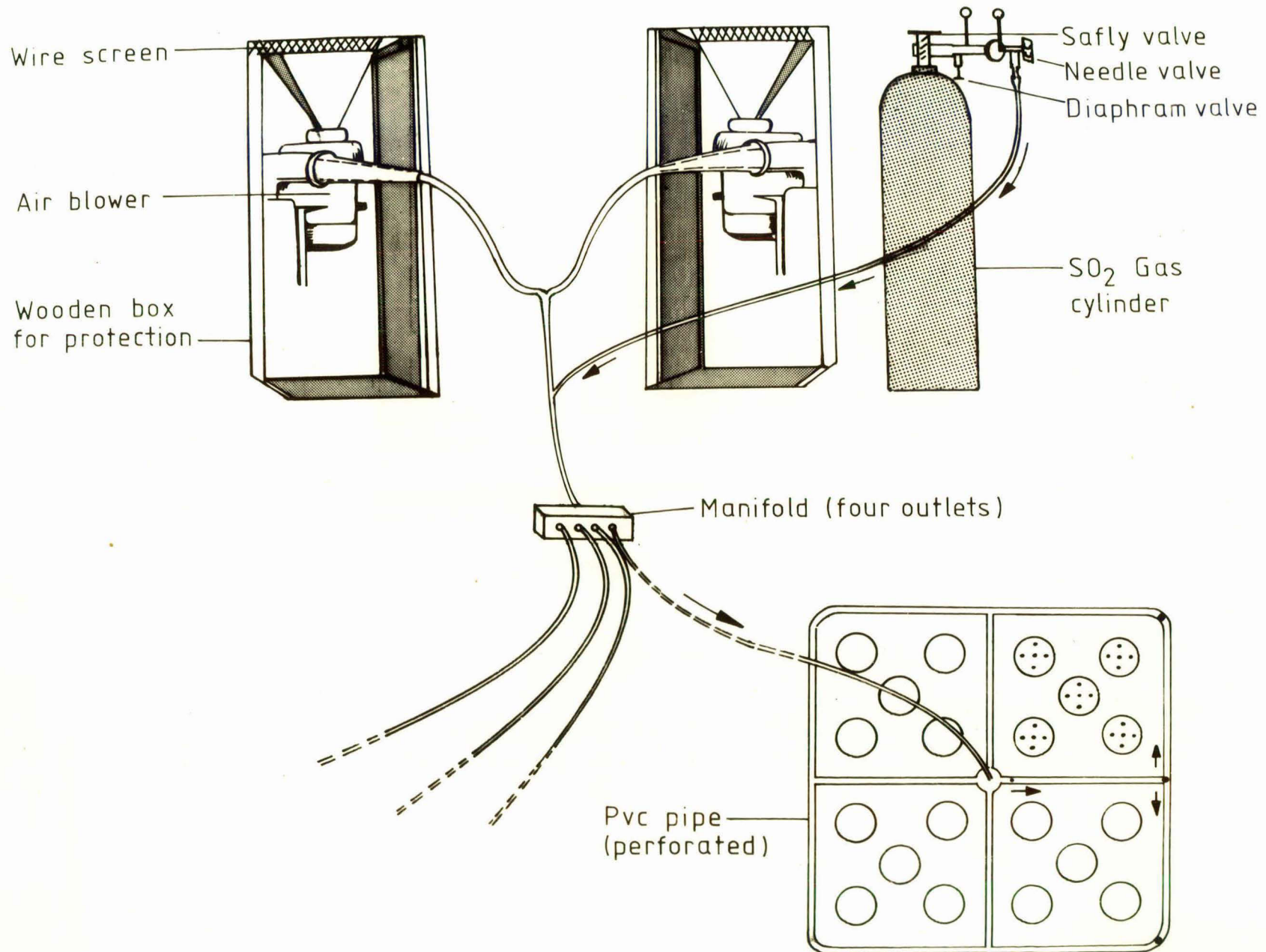
In each of the 4 pipes, forming a cross inside the square, 9 holes, each 10 cm apart were made. The diameter of the first hole starting from the center was 0.15 cm. The diameter of subsequent holes was increased progressively by 0.05 cm. Thus, the diameter of the last hole located at the mid point of each arm of the square was 0.55 cm (Fig. 2.10).

Fig. 2.8 The set up used for exposing plants to  $\text{SO}_2$ .



Fig. 2.9 A diagrammatic representation of the set up used for exposing plants with  $\text{SO}_2$ .





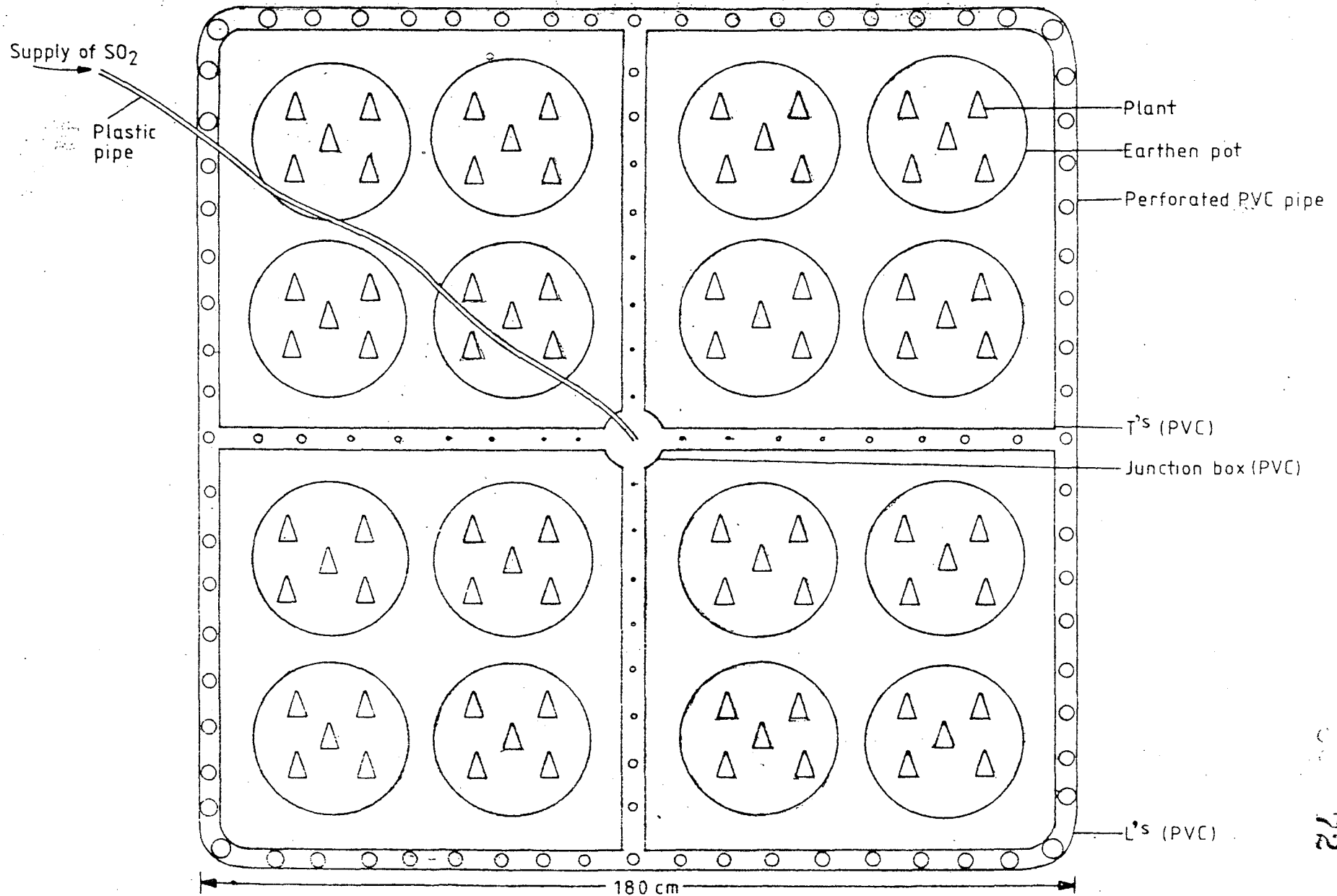


Fig. 2.10

A diagrammatic representation of perforated PVC pipe network on the ground for achieving uniform distribution of  $\text{SO}_2$  in the exposure chamber. (The diameter of holes in PVC pipes increases from the center of square to mid point of each arm of square and further upto each corner of square).

In addition, 9 holes, each 10 cm apart, were made from the mid point of each arm of the square upto the corner of the square. Here also, the diameter of the holes was progressively increased by 0.05 cm, starting from 0.6 cm. Thus, the diameter of the hole at the corners was 1.0 cm (Fig. 2.10).

#### Concentration of SO<sub>2</sub> in chamber

Plants were exposed in chambers each day for one hour. The SO<sub>2</sub> concentration in the exposure chamber varied from 183.6 to 258.02 ug m<sup>-3</sup>. There was only minor variation in SO<sub>2</sub> concentration with height inside the exposure chamber (Table 2.1).

#### Flyash treatment

16 pots in 4 x 4 rows were burried in the soil upto the ground level, in a plot of 2 m<sup>2</sup>. Flyash was sprayed with the help of a manually operated dust rotator (Fig. 2.11). Its capacity was determined on the basis of the amount of flyash released per rotation. This information was used to determine the number of rotation needed to spray 1.6 to 2.1 g m<sup>-2</sup> d<sup>-1</sup> flyash.

Table 2.1: Concentration of SO<sub>2</sub> (ug m<sup>-3</sup>) in different fumigation chambers

Month	Chamber I (SO <sub>2</sub> )	Chamber III (SO <sub>2</sub> +Flyash)	Chamber I (SO <sub>2</sub> )	Chamber III (SO <sub>2</sub> +Flyash)
Nov. 80	258.02-I	244.72-II	231.42-II	228.76-I
Dec. 80	215.46-III at 60 cm 226.1-III at 90 cm	236.74-I at 60 cm	223.44-I at 60 cm	247.38-IV at 60 cm
Jan. 81	225.36-I at 90 cm	220.78-IV at 90 cm	191.52-III at 60 cm 204.82-III at 90 cm	215.46-III at 90 cm
May 81	234.08-III	183.54-I		
June 81	244.72-I at 30 cm 226.1 -I at 60 cm	215.46-IV at 60 cm		
July 81	223.44-III at 90 cm	234.08-III at 90 cm		
Nov. 81	242.06-I at 30 cm	250.04-IV at 30 cm	228.76-III at 30 cm	258.02-IV at 30 cm
Dec. 81	242.06-IV at 60 cm	228.76-III at 30 cm 212.8-III at 60 cm	244.72-IV	242.06-I at 30 cm
Jan. 82	223.44-I at 90 cm	183.54-II at 90 cm	202.16-I at 90 cm	228.76-III at 90 cm
May 82	215.46-II	250.04-III		
June 82	215.46-I at 30 cm	199.50-IV at 60 cm		
July 82	218.12-IV at 90 cm	202.16-II at 90 cm		

I, II, III and IV represent the square.

Medicago sativa and Triticum aestivum plants exposed from Nov. to Jan, and Zea mays from May to July.

**Fig. 2.11** Spraying of flyash by a manually operated dust sprayer.



### Exposure Schedule

Plants were exposed for both long and short term, as described below.

#### Long term exposure

Plants in 112 pots for each species were grouped into five sets of 16 pots each, and four sets of 8 pots each. Among the five sets (16 pots) one set was kept at a site for field exposure and three sets for artificial exposure to  $\text{SO}_2$ , flyash and a combination of  $\text{SO}_2$ , and flyash and one set was kept as control. Plants were exposed for 45 days in field (F) and artificially (A). After 45 days, 8 pots out of 16 pots (field and artificial exposed) were removed and maintained at control site (without any exposure) for next 45 days (FC and AC). At their place, 8 pots which have not received any exposure for 45 days were kept. Now, 16 pots in total representing two sets of pots (8 pots exposed for 45 days and another 8 pots without any previous exposure) were exposed for another 45 days. This provided two more sets of plants, viz. (i) Exposed for 90 days (FF and AA) and (ii) control for 45 days followed by exposure for 45 days (CF and CA). Details of different sets exposed in field and artificially are given below.

- F = Exposed in field for 45 days  
A = Exposed artificially for 45 days  
C = Kept as control for 45 days  
FF = Exposed in field for 90 days  
AA = Exposed artificially for 90 days  
CF = Maintained as control for 45 days followed by field exposure for 45 days  
CA = Maintained as control for 45 days followed by artificial exposure for 45 days  
FC = Exposed in field for 45 days and kept at control site without any exposure for 45 days  
AC = Exposed artificially for 45 days and kept at control site without any exposure for 45 days  
CC = Kept as control for 90 days.

#### Short term exposure

Plant species were exposed on 45th day and on 90th day in field (24 h) and artificially to  $\text{SO}_2$  ( $218.3 \mu\text{g m}^{-3}$  for 1 h) and once to flyash ( $1.7 \text{ g m}^{-2}$ ) separately and in combination.



### Field Site Characteristics

A field site was selected in the vicinity of IP Power Plant which is a major source of air pollution in Delhi and has received a considerable publicity in the press. It is situated in the heart of the metropolitan city of Delhi at a distance of about 400-500 m from the West bank of the river Yamuna (Fig. 2.12). To the North-West and the Southern side of the power station lie prominent office complexes, shopping centers and residential areas.

The IP Power Plant was commissioned in 1963. The power generation by the Power Plant varies from 100-250 mw depending upon a variety of factors, the maximum capacity is 284 mw. In all, there are 5 units operating with total coal consumption of about 3500-4000  $\text{td}^{-1}$ . Unit one is consuming about 400  $\text{td}^{-1}$  while the consumption rate of each of other 4 units is around 750  $\text{td}^{-1}$ . Three stacks are operating at present. The calculated flyash emission (without control devices) of Stack I is almost half as compared with the emissions of Stack II and Stack III (Table 2.2). Major pollutants released from the Power Plant are flyash, oxides of sulphur and carbon dioxide (Table 2.3). It is evident from the table that harmful pollutants such as flyash and  $\text{SO}_2$  are emitted by the IP Power Plant in enormous quantities.

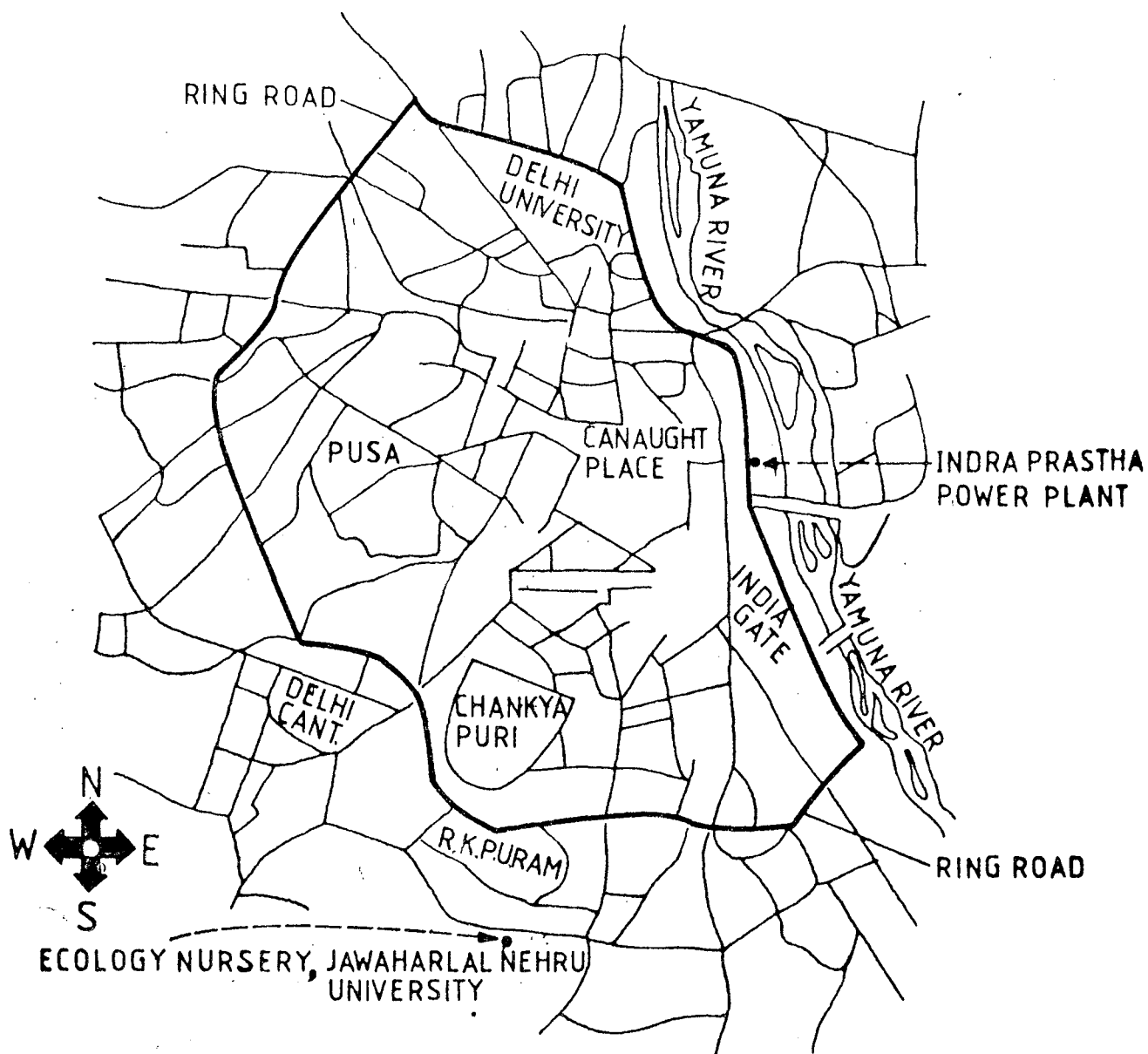


Fig. 2.12 A Delhi map showing locations (with dotted line) of IP Power Plant (the Field Site) and Ecological Nursery, Jawaharlal Nehru University (for Artificial exposure and for keeping plants as Control).

Table 2.2: Consumption of coal ( $\text{td}^{-1}$ ) and rate of emission of flyash ( $\times 10^7 \text{ u g sec}^{-1}$ ) from the IP Power Plant (New Delhi)

Stack No.	Stack height (m)	Amount of coal	Calculated emission rate
I	61.0	400	17.65
II	62.53	1500	35.27
III	62.53	1500	35.27

Table 2.3: Nature and quantity of main air pollutants released from the IP Power Plant (New Delhi)

Pollutant	Total amount ( $\text{td}^{-1}$ )
Flyash	40-81*
SO <sub>2</sub> (99%)	18
SO <sub>3</sub> (01%)	
CO <sub>2</sub>	4000

\*According to 'Indian Journal of Air Pollution Control' (News and Views column, Anonymous, 1978), the amount of flyash emitted from the IP Power Plant is about 81  $\text{td}^{-1}$  but IP Power Plant authorities gave a figure of 40  $\text{td}^{-1}$  (personal communication). These estimates are based on calculations, which take into consideration the maximum efficiency of the mechanical and the electrostatic precipitators installed in the Power Plant (personal communication). But in view of the fact that these mechanical and the electrostatic precipitators seldom work at their rated capacity, accordingly it seems reasonable that flyash released from the Power Plant is much more than 40  $\text{td}^{-1}$ . The daily flyash emission may well be around 81 t as reported in the Indian Journal of Air Pollution.

Ash content of the coal supplied to power plants is approximately around 30 to 35%, out of this 67% is estimated as the flyash. Out of 5 units, unit nos. 1, 4 and 5 have electrostatic precipitators which the power plant authorities claim work at an efficiency of 98%, and the remaining two units have only mechanical precipitators.

According to Padmanabhamurthy and Gupta (1977), the zone of high deposition of flyash is located between 0.8 to 1.6 km from the IP Power Plant. This zone oscillates between the East and the South-East for the greater part of the year except in the monsoon months. The zone of moderate deposition lies in the West. Flyash was collected from the hopper of the electrostatic precipitator of unit no. 4, as it is the same which is being released unchecked by the two units not equipped with electrostatic precipitators.

#### Methods used for flyash analysis

**Loss on ignition:** A 5 g sample of flyash was taken in a previously weighed platinum crucible and ignited for an hour on a bunsen burner. After combustion was complete, the crucible was cooled and weighed. The difference in weight represent loss on ignition and which is expressed as % of flyash (Table 2.4).

**Sieve analysis:** Particle size analysis of ash was carried out by sieve method. Seven sieves of mesh size (φ) 2.0, 2.5, 3.0, 3.5, 4.0, 4.5 and 4.5, were arranged in the descending order from bottom to top (sieve with 2.0 at the top). 100 g flyash was kept in the top most sieve and covered with a lid. The whole assembly was

placed in a sieve shaker (Scientific Instrument Services, India), which was connected to a 2 HP motor of 1450 rpm. Sieving was carried out for 10 min. Seven fractions were carefully collected and weighed separately and results have been expressed as % of flyash (Table 2.5).

The average particle size and shape in each fraction was examined using Transmission Electron Microscope (AEI 802, U.K.). Flyash particles were placed on a copper grid of 3.02 mm diameter having 400 holes. The grid was placed in the specimen holder and kept in the specimen stage of the microscope. A high vacuum was created and an electron beam was passed and photographs were taken at a magnification of 80 units. Mean particle size of each sieve fraction of flyash was determined by measuring the maximum diameter length of individual particles from the (Fig. 2.13) photographs and results have been expressed in mm (Table 2.5).

**Water holding capacity:** Water holding capacity was determined following the method outlined by Piper (1966).

Brass cups with perforated bottom and a split brass-o-rings were used for this purpose. Weight of each brass cup along with the ring was recorded ( $W_1$ ). Whatman no. 1 filter paper was placed at the bottom of the cup with

the help of brass-o-ring. Dry flash was filled in the cup by gradual tapping. Flyash packed cup was kept in a petridish containing distil water. The cups were allowed to stand in distil water for 24 h, to allow water absorption. Thereafter, it was removed, excess water was wiped out by a tissue paper, and weighed ( $W_2$ ), and kept in an electric oven at  $105^\circ\text{C}$  for 24 h and dry weight was recorded ( $W_3$ ).  $W_4$  was determined by finding out the difference between the weight of distil water soaked filter paper and dry filter paper.

The water holding capacity (W.H.C.) of flyash was determined (Table 2.4) using the following formula (Piper, 1966)

$$\text{W.H.C.} = \frac{W_2 - (W_3 + W_4)}{W_3 - W_1} \times 100$$

pH: pH was measured electrometrically by means of a calomel glass electrode. A 1:5 flyash suspension was made in distil water and stirred for one hour. The electrodes were immersed in a freshly shaken suspension and pH value was determined (Table 2.4) with the help of a Philips electronic pH meter.

**Electrical conductivity:** A 1:20 flyash suspension was made in distil water. Conductivity was determined using a conductivity meter (Sambros and Co., Model 303, India). The meter was standardized with the help of 0.1 N KCl solution at 25°C and conductivity values have been expressed in umho (Table 2.4).

**Organic matter:** Organic matter was determined using Walkley-Black method (1947). 5 g of flyash was taken in an flask to which 10 ml normal potassium dichromate solution and 200 ml of concentrated sulphuric acid was added. The mixture was allowed to stand for 30 min. 200 ml of distil water was added along with 10 ml phosphoric acid and 1 ml of 0.42% diphenyl-amine (acidic) indicator. The mixture was titrated against 1 N ferrous ammonium sulphate solution till green tinge appeared. Again 0.5 ml dichromate solution was added and sulphate solution was added drop by drop till green tinge appeared again. Percentage organic carbon was determined using the formula given below (Walkley-Black, 1947)

$$\frac{V_1 - V_2}{W} \times 100 \times 0.003$$

where,  $V_1$  - Volume of normal potassium dichromate (10.5 ml)  
 $V_2$  - Volume of normal ferrous ammonium sulphate in ml  
 $W$  - Weight of the soil.

Table 2.4: Physico-chemical properties of flyash, collected from the hopper of electrostatic precipitator of unit no. 4 of IP Power Plant (New Delhi)

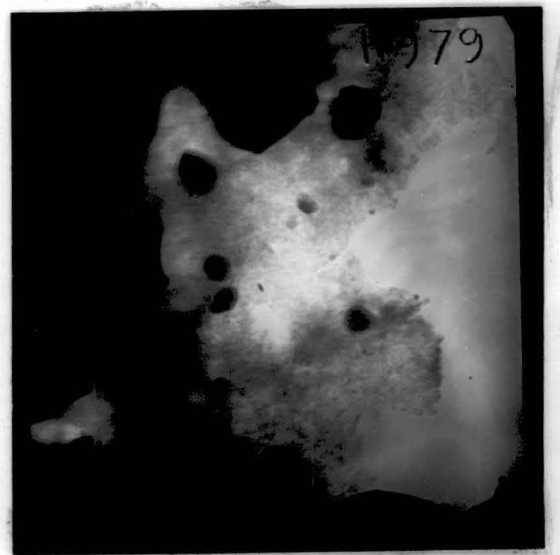
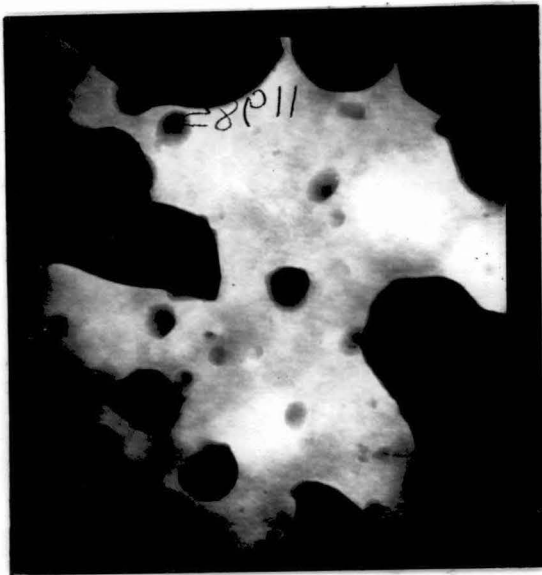
Properties	Value
Loss on ignition (%)	45.116
Water holding capacity	110.141
pH	8.650
Electrical Conductivity (umho)	184.000
Organic matter (%)	2.172

Table 2.5: Particle size distribution by sieve method (%) and average diameter of particles (mm) using transmission electron microscopy (TEM) of fly-ash collected from electrostatic precipitator of unit no. 4 of IP Power Plant (New Delhi)

Mesh size (mm)	Distribution (%)	Average Particle size (mm)
2.0 = 0.25	0.67 ± 0.05	0.2625
2.5 = 0.18	3.04 ± 0.09	0.1625
3.0 = 0.125	79.48 ± 2.41	0.1125
3.5 = 0.090	8.57 ± 0.43	0.0875
4.0 = 0.063	2.18 ± 0.28	0.0750
4.5 = 0.045	9.68 ± 0.33	0.0500
4.5 = 0.032	0.93 ± 0.08	0.0375
Loss	0.65 ± 0.12	



Fig. 2.13 Transmission electron micrographs of flyash particles representing seven different sieve fractions; mesh size ( ) range from 2.0 to 4.5, (a) 2.0 (X80); (b) 2.5 (X80); (c) 3.0 (X80); (d) 3.5 (X80); (e) 4.0 (X80); (f) 4.5 (X100); (g) 4.5 (X80).



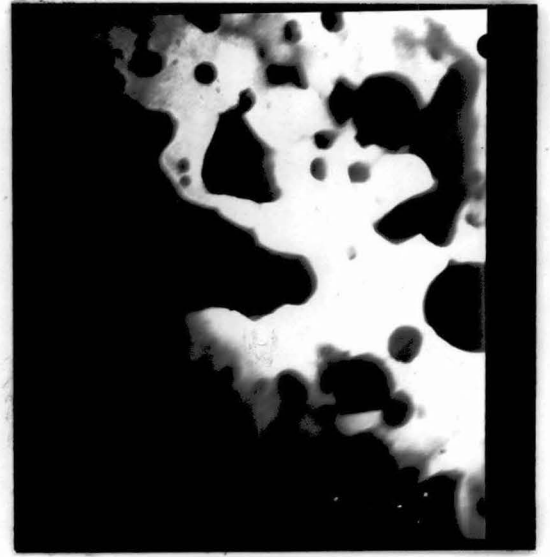


Table 2.6: Chemical composition of flyash, collected from electrostatic precipitator of unit no. 4 of IP Power Plant (New Delhi)

Major constituents	%
Silica	55.0 - 60.0
Al <sub>2</sub> O <sub>3</sub>	18.0 - 31.0
Fe <sub>2</sub> O <sub>3</sub>	5.0 - 8.0
TiO <sub>2</sub>	2.4 - 2.8
CaO	2.0 - 9.0
P <sub>2</sub> O <sub>5</sub>	1.9 - 2.1
MgO	0.6 - 3.6
Alkalies (Na <sub>2</sub> O + K <sub>2</sub> O)	2.2 - 3.0
Sulphur	0.16- 0.30
SO <sub>3</sub>	0.09- 0.11
Cd	2.7 (ppm) <sup>a</sup>
As	1.1 (ppm) <sup>b</sup>
Titanium	Trace amounts

Source: Personal communication with IP Power Plant Authorities.

a. After Naiak (1980)

b. After Phulekar (1980).

Percentage carbon value was multiplied by a factor of 1.734 to express the results in % organic matter (2.4).

Major chemical constituents of flyash as referred by IP Power Plant authorities are given in Table 2.6.

The field site for the present study was selected at about a distance of one km from the IP Power Plant in the direction ESE, thus indicating highest level of flyash deposition during the experiment (Fig. 3).

Concentration of  $\text{SO}_2$  at field site: The concentration of  $\text{SO}_2$  was measured at regular intervals at the selected site during the growth period of plants. It ranged from 26.6 to 119.7  $\mu\text{g m}^{-3}$  (Table 2.7).

Concentration of flyash at field site: It can be calculated from the data computed from Padmanabhamurthy and Gupta (1977) on a long term basis, that the average deposition of particulate matter from the IP Power Plant in the South-South East (zone of maximum deposition) ranges from 0.0744 to 0.185  $\text{g m}^{-2} \text{d}^{-1}$ . Deposition of particulate matter, on monthly basis revealed that in August, 0.185  $\text{g m}^{-2} \text{d}^{-1}$  has been recorded as maximum deposition, where 0.0744  $\text{g m}^{-2} \text{d}^{-1}$  as lowest in the month of October (Table 2.8).

Table 2.7: Concentration of  $\text{SO}_2$  ( $\mu\text{g m}^{-3}$ ) measured during the study period at the field site (approximately 1 km in South-South East direction from the IP Power Plant, New Delhi)

Month	Concentration of $\text{SO}_2$
Nov. 80	93.10
Dec. 80	89.16
Jan. 81	103.84
May 81	77.14
June 81	69.06
July 81	85.12
Nov. 81	111.72
Dec. 81	95.76
Jan. 82	119.70
May 82	58.52
June 82	66.50
July 82	26.60

Table 2.8: Rate of deposition of flyash ( $\text{g m}^{-2} \text{d}^{-1}$ )  
 in different months (approximately 1 km  
 in the South-South East direction of IP  
 Power Plant, New Delhi)  
 (calculated on the basis of 1977 data)

Month	Concentration of flyash ( $\text{g m}^{-2} \text{d}^{-1}$ )
January	0.0982
February	0.0940
March	0.1000
April	0.1400
May	0.1060
June	0.1100
July	0.0964
August	0.1850
September	0.0762
October	0.0744
November	0.0935
December	0.0994

After Padmanabhamurthy and Gupta (1977).

### Statistical Analysis

The internal variability or dispersion of the data on total biomass, biomass of fruit, stem, leaf and root, chlorophyll content, diffusive resistance and leaf temperature was subjected to statistical analysis and standard deviation ( $\sigma$ ) was calculated. The sum of squares of the deviations ( $\sum (x - \bar{x})^2$ ) from the mean ( $\bar{x}$ ) divided by the number of observations (N). The square root of the resultant represents the value of standard deviation ( $\sigma$ ).

$$\sigma = \sqrt{\frac{\sum (x - \bar{x})^2}{N}}$$

where,  $\sigma$  - Standard deviation  
 $\sum$  - Sign of algebraic sum  
 $x$  - Observed value  
 $\bar{x}$  - Mean of observed values  
 $N$  - Number of observations.



## RESULTS

### Plant Performance

Plant performance was evaluated in terms of visual injury symptoms, leaf area, leaf biomass, total plant biomass and chlorophyll content. For field exposure, one set of plants (1st set) was kept in the vicinity (about 1 km of east-south east direction) of IP Power Plant. At the field site, the concentration of  $\text{SO}_2$  varied from 46.6 to 119.7  $\mu\text{g m}^{-3}$  and flyash concentration as calculated by Padmanabhamurthy and Gupta (1977) varies from 0.0744 to 0.185  $\text{g m}^{-2} \text{d}^{-1}$ . Three sets of plants (2nd, 3rd and 4th) were exposed artificially to  $\text{SO}_2$  and flyash, individually and in combination. The 2nd set was exposed to 183.6 to 258.02  $\mu\text{g m}^{-3}$   $\text{SO}_2$  for one hour daily, the 3rd set of plants was sprayed once with 1.6 to 2.1  $\text{g m}^{-2} \text{d}^{-1}$  flyash and the 4th set was exposed to a combination of 183.6 to 258.02  $\mu\text{g m}^{-3}$   $\text{SO}_2$  for one hour daily and 1.6 to 2.1  $\text{g m}^{-2} \text{d}^{-1}$  flyash. The 5th set of plants was kept in a relatively pollution-free environment at the ecological nursery of Jawaharlal Nehru University, which served as control. The schedule followed for exposing the plants in field and under experimental conditions, can

be broadly classified as follows:

- F = Exposed in the field for 45 days in the vicinity of IP Power Plant.
- A = Exposed artificially for 45 days to SO<sub>2</sub> and/or flyash.
- C = Kept as control for 45 days.
- FF = Exposed in field for 90 days in the vicinity of IP Power Plant.
- AA = Exposed artificially for 90 days to SO<sub>2</sub> and/or flyash.
- CF = Maintained in pollution-free environment for 45 days followed by field exposure for 45 days in the vicinity of IP Power Plant.
- CA = Maintained as control for 45 days followed by artificial exposure for 45 days to SO<sub>2</sub> and/or flyash.
- FC = Exposed in field for 45 days, in the vicinity of IP Power Plant, and thereafter maintained for 45 days in pollution-free environment.
- AC = Exposed artificially for 45 days to SO<sub>2</sub> and/or flyash and thereafter maintained for 45 days in pollution-free environment.
- CC = Kept as control for 90 days.

#### Visual injury symptoms

Chlorotic spots were observed in Medicago sativa exposed in field, and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash in F and A, and FF and AA exposure pattern.

In field exposed plants small circular and elliptical chlorotic spots mainly in the interveinal regions of the leaf were observed (Figs. 3.1, 3.2). Irregular chlorotic leaf margins were observed in plants exposed to SO<sub>2</sub> artificially (Figs. 3.3, 3.4). Chlorotic spots of different shapes varying from about 1.0 to 3.0 mm developed in interveinal areas of the leaf surfaces in plants exposed to a combination of SO<sub>2</sub> and flyash (Figs. 3.5, 3.6). Mature leaves suffered more as compared to young leaves. The intensity of chlorotic spots increased by prolonging the exposure period from 45 to 90 days. Exposed plants of Triticum aestivum and Zea mays did not suffer from any visual injury.

#### Leaf area

F and A exposure: Leaf area in control plants of Medicago sativa was  $815.48 \pm 84.52$  cm<sup>2</sup> (Table 3.1). In plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash, it reduced by 9.5, 10.2, 3.8 and 9.7% respectively (Table 3.2). Maximum reduction in leaf area was observed in plants exposed to SO<sub>2</sub> followed by plants exposed to a combination of SO<sub>2</sub> and flyash, in field and to flyash. Leaf area in control plants of Triticum aestivum was  $565.21 \pm 50.12$  cm<sup>2</sup> (Table 3.1). In plants exposed in field, and

artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash it reduced by 8.2, 7.4, 3.3 and 6.8% respectively (Table 3.2). Maximum reduction was observed in plants exposed in field followed by plants exposed to SO<sub>2</sub>, a combination of SO<sub>2</sub> and flyash and to flyash. Control plants of Zea mays had leaf area of 119.27 ± 123.24 cm<sup>2</sup> (Table 3.1). In plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash it reduced by 5.0, 5.6, 0.4 and 5.3% respectively (Table 3.2). Maximum reduction was observed in plants exposed to SO<sub>2</sub>, followed by plants exposed to a combination of SO<sub>2</sub> and flyash, in field and to flyash.

FF and AA exposure: The leaf area of control plants of Medicago sativa was 1403.29 ± 134.68 cm<sup>2</sup> (Table 3.1). In plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash, it reduced by 18.9, 20.2, 7.1 and 22.6% respectively (Table 3.2). Maximum reduction was observed in plants exposed to SO<sub>2</sub> and flyash in combination, followed by exposure to SO<sub>2</sub>, in field and to flyash. Leaf area of control plants of Triticum aestivum was 667.99 ± 61.72 cm<sup>2</sup> (Table 3.1). In plants exposed in field and artificially to SO<sub>2</sub>, flyash and combination of SO<sub>2</sub> and flyash, it reduced by

15.8, 14.4, 5.4 and 16.9% respectively (Table 3.2). The maximum reduction was observed in plants exposed to a combination of SO<sub>2</sub> and flyash followed by plants exposed in field, to SO<sub>2</sub> and to flyash. Leaf area of control plants of Zea mays was 1911.68 ± 166.23 cm<sup>2</sup> (Table 3.1). In plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash, it reduced by 7.2, 8.7, 2.3 and 8.4% respectively (Table 3.2).

CF and CA exposure: Leaf area in control plants of Medicago sativa, Triticum aestivum and Zea mays was same as mentioned in FF and AA exposure pattern (Table 3.1). In Medicago sativa plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash, it reduced by 4.9, 5.71, 4.52 and 9.4% respectively (Table 3.2). Maximum reduction was observed in plants exposed to a combination of SO<sub>2</sub> and flyash followed by plants exposed to SO<sub>2</sub>, in field and to flyash. Triticum aestivum plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash, the leaf area reduction was less than 5% (Table 3.2). However, maximum reduction was observed in plants exposed to a combination of SO<sub>2</sub> and flyash,

followed by plants exposed in field, to SO<sub>2</sub> and to flyash. In Zea mays plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash the reduction in leaf area was less than 2% (Table 3.2).

FC and AC exposure: In control plants of Medicago sativa, Triticum aestivum and Zea mays, leaf area was same as mentioned in FF and AA exposure pattern (Table 3.1), and in exposed plants it reduced by less than 5, 3, 2% respectively (Table 3.2).

A perusal of the data reveals that (i) Maximum reduction was in Medicago sativa, followed by Triticum aestivum and Zea mays, (ii) Maximum reduction (10.2%) was observed in plants exposed to SO<sub>2</sub> (F and A exposure) followed by a combination of SO<sub>2</sub> and flyash, in field and to flyash. While in 90 days exposure (FF and AA), maximum reduction (22.6%) was observed in plants exposed to a combination of SO<sub>2</sub> and flyash followed by plants exposed to SO<sub>2</sub>, in field and to flyash (Table 3.2).

### Leaf biomass

F and A exposure: Leaf biomass in control plants of Medicago sativa was  $1.564 \pm 0.12$  g (Table 3.3). In plants exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash, it reduced by 16.3, 18.8 and 7.1% respectively (Table 3.4). In flyash exposed plants it increased by 2.7%. Maximum reduction in leaf biomass was observed in plants exposed to SO<sub>2</sub> followed by exposure in field and artificially to a combination of SO<sub>2</sub> and flyash. In control plants of Triticum aestivum leaf biomass was  $1.452 \pm 0.23$  g (Table 3.3). In plants exposed in field, and artificially to SO<sub>2</sub>, and a combination of SO<sub>2</sub> and flyash, it reduced by 11.7, 12.8 and 11.5% respectively (Table 3.4). In plants exposed to flyash, leaf biomass increased by 3.8%. Leaf biomass of control plants of Zea mays was  $6.146 \pm 0.93$  g (Table 3.3). In plants exposed in field and artificially to SO<sub>2</sub>, and a combination of SO<sub>2</sub> and flyash, it reduced by 6.4, 6.8 and 6.2% respectively (Table 3.4). The leaf biomass increased by 0.8% in flyash exposed plants.

FF and AA exposure: Leaf biomass in control plants of Medicago sativa was  $2.720 \pm 0.32$  g (Table 3.3). In

plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash it reduced by 28.7, 25.8, 10.7 and 31.2% respectively (Table 3.4). Maximum reduction in leaf biomass was observed in plants exposed to a combination of SO<sub>2</sub> and flyash followed by plants exposed in field, to SO<sub>2</sub> and to flyash. In control plants of Triticum aestivum leaf biomass was 1.734 ± 0.28 g (Table 3.3). In plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash it reduced by 21.7, 20.4, 8.4 and 23.9% respectively (Table 3.4). Leaf biomass in control plants of Zea mays was 7.597 ± 0.14 g (Table 3.3). In plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash it reduced by 12.1, 10.1, 5.2 and 13.1% respectively (Table 3.4).

CF and CA exposure: Leaf biomass in control plants of Medicago sativa, Triticum aestivum and Zea mays was same as described under FF and AA exposure (Table 3.3). The leaf biomass of Medicago sativa plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash was reduced by 9.6, 7.4, 5.6 and 11.4% respectively (Table 3.4). The maximum reduction was observed in plants exposed to a combination of SO<sub>2</sub> and



flyash followed by plants exposed in field, to SO<sub>2</sub> and to flyash. In Triticum aestivum and Zea mays plants exposed in field and artificially, the reduction in leaf biomass was less than 0.1% and 4.1% respectively. The sequence of leaf biomass reduction was similar to the pattern observed in Medicago sativa (Table 3.4).

FC and AC exposure: Leaf biomass in control plants of Medicago sativa, Triticum aestivum and Zea mays was same as mentioned in FF and AA exposure pattern (Table 3.3). In Medicago sativa plants exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash, leaf biomass reduced by 5.4, 7.1 and 6.1% respectively (Table 3.4). Flyash exposed plants exhibited an increase of 3.4%. In Triticum aestivum and Zea mays plants exposed in field and artificially to SO<sub>2</sub>, and a combination of SO<sub>2</sub> and flyash, leaf biomass reduction was less by 5 and 4% respectively (Table 3.4). Flyash exposed plants of Triticum aestivum and Zea mays exhibited an increase in leaf biomass by 2.5 and 0.6% respectively.

It can be seen from the table 3.4 that (i) maximum reduction in leaf biomass was in Medicago sativa plants followed by Triticum aestivum and Zea mays; (ii) in general, in F and A exposure (45 days) maximum reduction

(18.8%) was observed in plants exposed to SO<sub>2</sub> followed by plants exposed in field, and to a combination of SO<sub>2</sub> and flyash. However, in flyash exposed plants, it increased by 3.8%. In FF and AA exposure, maximum reduction (31.2%) was observed in plants exposed to a combination of SO<sub>2</sub> and flyash followed by plants exposed in field, to SO<sub>2</sub> and to flyash (Table 3.4).

#### Total plant biomass

F and A exposure: Total plant biomass in control plants of Medicago sativa was 6.367 ± 0.65 g (Table 3.5). In plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash it reduced by 13.7, 16.7 and 15.8% respectively (Table 3.9). In flyash exposed plants it increased by 4.1%. Maximum reduction was observed in plants exposed artificially to SO<sub>2</sub>, followed by a combination of SO<sub>2</sub> and flyash and in field. In control plants of Triticum aestivum total plant biomass was 6.883 ± 0.71 g (Table 3.5). In plants exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash it reduced by 8.7, 11.8 and 10.6% respectively (Table 3.9). Flyash exposed plants exhibited an increase of 3.2%. In Zea mays plants kept under controlled conditions total plant biomass was

32.583  $\pm$  5.12 g (Table 3.5). In plants exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash it reduced by 5.1, 6.1 and 5.4% respectively (Table 3.9). In flyash total plant biomass increased by 0.7%.

FF and AA exposure: In control plants of Medicago sativa total plant biomass was 10.226  $\pm$  1.31 g (Table 3.6). In plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash it reduced by 20.4, 18.7, 9.4 and 26.6% respectively (Table 3.9). Maximum decrease was observed in plants exposed artificially to a combination of SO<sub>2</sub> and flyash followed by plant exposed in field, to SO<sub>2</sub> and to flyash. Total plant biomass in control plants of Triticum aestivum was 11.13  $\pm$  1.39 g (Table 3.6). In plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash, it reduced by 18.2, 15.6, 7.2 and 19.7% respectively (Table 3.9). In control plants of Zea mays total plant biomass was 45.532  $\pm$  5.47 g (Table 3.6). In plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash it reduced by 8.4, 7.3, 5.7 and 9.7% respectively (Table 3.9).

CF and CA exposure: Total plant biomass in control plants of Medicago sativa, Triticum aestivum and Zea mays was same as mentioned under FF and AA exposure (Table 3.6), 3:7). In Medicago sativa plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash total plant biomass reduced by 9.0, 8.1, 4.3 and 10.2% respectively (Table 3.9). Reduction in total plant biomass was less than 7.1 and 3.1% in exposed plants of Triticum aestivum and Zea mays respectively (Table 3.9).

FC and AC exposure: Total plant biomass of Medicago sativa, Triticum aestivum and Zea mays plants was same as mentioned under FF and AA exposure pattern (Tables 3.8, 3.9). It reduced by less than 5.7% in Medicago sativa, 4.2% in Triticum aestivum and 2.7% in Zea mays plants due to exposure in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash. However, in flyash exposed plants it increased by 3.2% (Table 3.9).

An examination of the data shows: (i) Maximum reduction in total plants biomass was in Medicago sativa plants followed by Triticum aestivum and Zea mays; (ii) in general, in F and A exposure (45 days) maximum reduction (16.7%) was in plants exposed to SO<sub>2</sub> followed by plants exposed to a combination of SO<sub>2</sub> and flyash, and

in field. It however, increased by 4.1% in flyash exposed plants. In FF and AA exposure maximum reduction (26.6%) was in plants exposed to a combination of SO<sub>2</sub> and flyash followed by plants exposed in field, to SO<sub>2</sub> and to flyash (Table 3.9).

#### Total chlorophyll content

F and A exposure: The total chlorophyll content in control plants of Medicago sativa was  $3.952 \pm 0.34 \text{ mg g}^{-1}$  (Table 3.10). In plants exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash it reduced by 10.2, 14.2 and 11.1% respectively (Table 3.11). In flyash exposed plants it increased by 3.1% (Table 3.11). Maximum reduction in total chlorophyll content was in plants exposed to SO<sub>2</sub> followed by plants exposed to a combination of SO<sub>2</sub> and flyash and in field. In control plants of Triticum aestivum total chlorophyll content was  $3.516 \pm 0.31 \text{ mg g}^{-1}$  (Table 3.10). In plants exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash it reduced by 5.8, 8.5 and 6.2% respectively. In flyash exposed plants it increased by 2.1% (Table 3.11). Total chlorophyll content in control plants of Zea mays was  $3.767 \pm 0.33 \text{ mg g}^{-1}$  (Table 3.10).

In plants exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash, it reduced by less than 4.2%. However, in flyash exposed plants it increased by 1.3% (Table 3.11).

FF and AA exposure: In control Medicago sativa plants total chlorophyll content was  $3.968 \pm 0.38 \text{ mg g}^{-1}$  (Table 3.10). In plants exposed in field and artificially SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash it reduced by 14.4, 18.9, 7.2 and 15.7% respectively (Table 3.11). Maximum reduction in total chlorophyll content was observed in plants exposed to SO<sub>2</sub> followed by plants exposed to a combination of SO<sub>2</sub> and flyash, in field and flyash. The total chlorophyll content in control plants of Triticum aestivum was  $3.528 \pm 0.32 \text{ mg g}^{-1}$  (Table 3.10). In plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash it reduced by 11.8, 14.1, 5.3 and 12.3% respectively (Table 3.11). In control of Zea mays the total chlorophyll content was  $3.772 \pm 0.38 \text{ mg g}^{-1}$  (Table 3.10). In plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash it reduced by 5.6, 8.7, 3.2 and 6.1% respectively (Table 3.11).

CF and CA exposure: The total chlorophyll content of control plants of Medicago sativa, Triticum aestivum and Zea mays was same as described under FF and AA exposure (Table 3.10). In Medicago sativa plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash the total chlorophyll content was reduced by 8.9, 12.1, 3.7 and 9.6% respectively (Table 3.11). Triticum aestivum and Zea mays plants exposed in field and artificially showed a reduction of less than 6.4% and 3.4% respectively (Table 3.11).

FC and AC exposure: In control plants of Medicago sativa, Triticum aestivum and Zea mays the total chlorophyll content reduced by less than 4.3, 2.5 and 1.9% respectively (Table 3.11). When exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash. However, it increased marginally by less than 2.1% (Table 3.11).

Data in table 3.11 indicate: (i) Maximum reduction in total chlorophyll content was in Medicago sativa followed by Triticum aestivum and Zea mays; (ii) in general, in F and A exposure (45 days) maximum reduction (14.2%) was in plants exposed to SO<sub>2</sub> followed by plants exposed to a combination of SO<sub>2</sub> and flyash,

and in field. However, in flyash exposed plants, a marginal increase of 3.1% was observed. In FF and AA exposure (90 days) maximum reduction (18.9%) was observed in plants exposed to SO<sub>2</sub> followed by plants exposed to a combination of SO<sub>2</sub> and flyash, in field and to flyash.



### Epidermal Features

Medicago sativa has anomocytic type of stomata (Table 3.12) and both adaxial and abaxial leaf surfaces are glabrous (free from trichomes). Paracytic type of stomata are found in Triticum aestivum and Zea mays.

In Triticum aestivum both the leaf surfaces are glabrescent. The adaxial leaf surface in Zea mays is pubescent and the abaxial surface is glabrescent (Table 3.12), but trichomes were limited to veinal regions.

Stomatal density, length and breadth of stomatal pore were measured in Medicago sativa, Triticum aestivum and Zea mays plants exposed in field and artificially to SO<sub>2</sub> and flyash individually and in combination under FF and AA schedule and compared with control.

#### Stomatal density.

Stomatal density of the adaxial and abaxial leaf surfaces in control plants of Medicago sativa was 171 and 156 mm<sup>-2</sup> (Table 3.13). Stomatal density was more in case of adaxial than abaxial surface. In plants exposed in field and artificially to SO<sub>2</sub>, flyash and in combination, it decreased by 23.39 and 35.09% in case of adaxial surface and 8.77 and 16.08% in case of abaxial surface respectively (Table 3.14, Fig. 3.7-3.12). Stomatal density of the adaxial surface in control plant of

Triticum aestivum was  $46 \text{ mm}^{-2}$  (Table 3.13). In plants exposed in field and artificially to  $\text{SO}_2$ , flyash and a combination of  $\text{SO}_2$  and flyash, it decreased by 8.9, 15.22, 6.52 and 15.22% respectively (Table 3.14, Fig. 3.13-3.17). Stomatal density of the adaxial and abaxial leaf surfaces in control plants of Zea mays was 64 and  $110 \text{ mm}^{-2}$  (Table 3.13). It was more in case of abaxial than adaxial leaf surface. In plants exposed in field and artificially to  $\text{SO}_2$ , flyash and a combination of  $\text{SO}_2$  and flyash it decreased by 28.12, 17.18 and 23.43% in case of adaxial surface and 8.18, 16.36, 5.45 and 13.64% in case of abaxial surface respectively (Table 3.14, Fig. 3.18-3.26).

A perusal of the data reveals that (i) reduction in stomatal density was more in Medicago sativa as compared to Zea mays and Triticum aestivum; (ii) reduction in stomatal density was more in case of adaxial than abaxial surface.

#### Length and breadth of stomatal pore

Length and breadth of stomatal pore of adaxial leaf surface in control plants of Medicago sativa was 10.6 and  $2.0 \mu$  respectively (Table 3.15). Plants exposed in field and artificially to  $\text{SO}_2$ , flyash and a

combination of SO<sub>2</sub> and flyash had length of stomatal pore reduced by 43.4, 40.57, 42.45 and 43.4% respectively. Breadth of stomatal pore reduced by 75.0, 25.0 and 50.0% in plants exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash (Table 3.16, Fig. 3.27 - 3.31). In plants exposed to flyash breadth of stomatal pore increased by 35.0%. Length and breadth of stomatal pore of the abaxial surface in control plants of Medicago sativa was 8.5 μ and 2.0 μ (Table 3.15). Plants exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash it reduced by 10.59, 28.24, 35.29% respectively. Plants exposed to flyash showed an increase of 3.53%. The breadth of stomatal pore was reduced by 75.0, 20.0, 20.0 and 75.0% in plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash respectively (Table 3.16, Fig. 3.32-3.36).

Data in tables 3.16 and Figs. 3.29-3.36 reveal that (i) in Medicago sativa reduction in length and breadth of stomatal pore was more in case of adaxial as compared to abaxial leaf surface; (ii) reduction in length and breadth of stomatal pore in field exposed plants and in plants exposed artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash was more or less the same.

In flyash exposed plants the length and breadth of the stomatal pore reduced marginally.

Length and breadth of stomatal pore of adaxial leaf surface in control plants of Triticum aestivum was 29.5 and 1.5  $\mu$  (Table 3.15). The length of stomatal pore reduced by 16.46, 29.11, 26.58 and 38.24% in plants exposed in field and artificially to  $\text{SO}_2$ , flyash and a combination of  $\text{SO}_2$  and flyash respectively. Breadth of stomatal pore did not change in plants which were exposed either in field or artificially to a combination of  $\text{SO}_2$  and flyash. However, in plants exposed to  $\text{SO}_2$  and flyash it reduced by 66.7 and 33.3% respectively (Table 3.16, Figs. 3.37-3.41). Length and breadth of stomatal pore of abaxial leaf surface was 42.0 and 1.0  $\mu$  respectively (Table 3.15). The length of a stomatal pore reduced by 21.43, 50.0 and 28.7% and the breadth of a stomatal pore reduced by 50.0, 10.0 and 10.0% in plants exposed in field and artificially to  $\text{SO}_2$  and a combination of  $\text{SO}_2$  and flyash respectively (Table 3.16, Figs. 3.42-3.45).

Length and breadth of stomatal pore of adaxial leaf surface in control plants of Zea mays was 30.0 and 50.0  $\mu$  respectively (Table 3.15). The length of stomatal pore decreased by 16.67, 7.33, 8.33 and 13.33% and the

breadth was reduced by 30.0, 38.0, 28.0 and 56.0% in plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash respectively (Table 3.16, Fig. 3.46-3.50). Length and breadth of a stomatal pore of abaxial surface was 30.  $\mu$  and 4.4  $\mu$  (Table 3.15). In plants exposed in field and to SO<sub>2</sub> and flyash individually the length of stomatal pore decreased by 13.53, 11.76 and 3.8% respectively. The length of stomatal pore increased by 7.35% when exposed artificially to a combination of SO<sub>2</sub> and flyash. The breadth of stomatal pore in plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash reduced by 29.55, 18.18, 29.55 and 88.64% respectively (Table 3.16, Fig. 3.51-3.55). In Zea mays the effect on the length and breadth of stomatal pore in case of adaxial leaf surface was more or less similar to the changes observed on abaxial surface. The effect on length and breadth of stomatal pore due to exposure in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash was more or less the same.

An examination of the table 3.15 and 3.16 shows:  
(i) reduction in length and breadth of stomatal pore was more in Medicago sativa as compared to Zea mays and

Triticum aestivum; (ii) in general, adaxial leaf surface has shown comparatively much more reduction in length and breadth of stomatal pore than abaxial surface; (iii) reduction in length and breadth of stomatal pore in field exposed plants and plants exposed artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash was more or less comparable. The flyash treatment does not affect much the stomatal pore.

Measurements were also made for trichome density and trichome length in Triticum aestivum plants exposed under FF and AA schedule in field and artificially and compared with control.

Trichome density (no. mm<sup>-2</sup>)

Trichome density of the adaxial and abaxial leaf surfaces in control plants of Triticum aestivum was 60 and 49 respectively (Table 3.17). In plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash the trichome density increased by 76.66, 21.66, 30.0 and 58.33% respectively in case of adaxial surface and 24.49, 15.96, 20.41 and 36.73% respectively in case of abaxial leaf surface (Table 3.18, Fig. 3.18-3.21; 3.56-3.60). The trichome density of the adaxial surface in Triticum aestivum increased more as compared to the abaxial surface in plants exposed in field and

artificially to a combination of SO<sub>2</sub> and flyash.

Increase in trichome density treated plants to SO<sub>2</sub> and to flyash was relatively less as compared to other treatments.

#### Trichome length ( $\mu$ )

The length of trichomes of adaxial and abaxial leaf surfaces in control plants of Triticum aestivum was  $36.4 \pm 7.3$  and  $45.5 \pm 10.7 \mu$  respectively (Table 3.19). In plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash it increased by 13.89, 5.56, 25.0 and 27.78% respectively in case of adaxial surface and 11.11, 6.67, 15.56 and 15.56% respectively in case of abaxial leaf surface (Table 3.20, Fig. 3.61 - 3.70). In Triticum aestivum trichome length increased more in case of adaxial surface as compared to abaxial surface. The length of trichome increased more in plants exposed to a combination of SO<sub>2</sub> and flyash as compared to plants exposed in field and artificially to SO<sub>2</sub> and to flyash.

### Diffusive Resistance

Leaf diffusive resistance for both adaxial and abaxial leaf surfaces was measured in Medicago sativa, Triticum aestivum and Zea mays plants exposed in field and artificially for short term (after exposure) and long term exposure (between 11.00 and 12.00 hrs for three consecutive sunny days).

#### Effect of short term exposure

Plants of 45 and 90 days old (after seedling stage) were exposed in field (24 h) and artificially to SO<sub>2</sub> (1 h) and flyash separately and in combination.

Exposure on 45th day: Diffusive resistance of adaxial and abaxial leaf surfaces in control plants of Medicago sativa was  $0.76 \pm 0.11$  and  $0.83 \pm 0.13$  cm sec<sup>-1</sup> (Table 3.21). The diffusive resistance of abaxial leaf surface was more as compared to adaxial surface. In plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash it reduced by 3.8, 12.8, 1.2 and 13.1% at the adaxial surface and 2.9, 8.4, 0 and 10.1% at the abaxial surface respectively (Table 3.22). Data in table 3.22 indicates that diffusive resistance of adaxial surface was affected more as compared to abaxial surface. Maximum reduction of diffusive resistance was at the adaxial leaf surface in plants exposed



to a combination of SO<sub>2</sub> and flyash followed by SO<sub>2</sub>, field and flyash exposures. The diffusive resistance of the abaxial leaf surface also decreased in the same sequence.

In control plants of Triticum aestivum, the diffusive resistance of adaxial and abaxial leaf surfaces was  $5.30 \pm 1.05 \text{ sec cm}^{-1}$  and  $8.14 \pm 1.51 \text{ sec cm}^{-1}$  (Table 3.21). The diffusive resistance of abaxial leaf surface was more as compared to the adaxial leaf surface. In field exposed plants and plants exposed to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash artificially, it reduced by 2.6, 8.6, 8.8 and 0.8% in case of the adaxial and 1.2, 5.2, 0 and 4.9 at the abaxial leaf surfaces respectively (Table 3.22).

Table 3.22 indicates that diffusive resistance of adaxial leaf surface changed more as compared to abaxial leaf surface. Maximum reduction was at the adaxial leaf surface in plants exposed to a combination of SO<sub>2</sub> and flyash followed by exposure to SO<sub>2</sub>, in field and flyash. The diffusive resistance of the abaxial leaf surface also decreased in the same sequence.

In control plants of Zea mays, diffusive resistance on adaxial and abaxial leaf surface was  $2.72 \pm 0.33$  and

$1.91 \pm 0.31 \text{ cm sec}^{-1}$  respectively (Table 3.21). The diffusive resistance of adaxial leaf surface was more as compared to the abaxial leaf surface. In plants exposed in field and artificially to  $\text{SO}_2$ , flyash and a combination of  $\text{SO}_2$  and flyash. It reduced by 1.4, 6.8, 0.6 and 6.8% at adaxial leaf surface and 1.4, 5.4, 0 and 6.8% at abaxial leaf surface (Table 3.22). The data in table 3.22 show that the change in the diffusive resistance of adaxial leaf surface was close to abaxial leaf surface. The maximum reduction was observed in plants exposed to a combination of  $\text{SO}_2$  and flyash followed by plants exposed to  $\text{SO}_2$ , in field and to flyash.

An examination of the data in table 3.22 reveals that (i) decrease in diffusive resistance was more in case of adaxial leaf surface than at abaxial leaf surface; (ii) in Medicago sativa the decrease in diffusive resistance was more, followed by Triticum aestivum and Zea mays; (iii) plants exposed to a combination of  $\text{SO}_2$  and flyash exhibited maximum reduction in diffusive resistance followed closely by exposure to  $\text{SO}_2$ . In plants exposed in field and to flyash the reduction was comparatively less.

Exposure on 90th day: Diffusive resistance of adaxial leaf surface in control plants of Medicago sativa was  $0.81 \pm 0.13$  and  $0.900 \pm 0.15$  cm sec<sup>-1</sup> (Table 3.23). The diffusive resistance of abaxial leaf surface was more as compared to adaxial surface. In plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash it reduced by 4.9, 17.4, 1.6 and 17.7% at adaxial surface respectively. It reduced by 3.1, 11.9 and 12.4% at abaxial surface when exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash respectively (3.24). There was no change in the diffusive resistance on abaxial surface in plants exposed to flyash. Data in table 3.24 reveal that change in diffusive resistance was more in case of adaxial surface as compared to abaxial surface. Maximum reduction was observed on the adaxial surface in plants exposed to a combination of SO<sub>2</sub> and flyash followed by exposure to SO<sub>2</sub>, in field and to flyash. The diffusive resistance of abaxial leaf surface also decreased in the same sequence.

In control plants of Triticum aestivum diffusive resistance of adaxial and abaxial leaf surfaces was  $6.20 \pm 1.28$  and  $8.77 \pm 1.63$  cm sec<sup>-1</sup> respectively (Table

3.23). The diffusive resistance of abaxial surface was more as compared to adaxial leaf surface. In plants exposed in field and artificially to  $\text{SO}_2$ , flyash and to a combination of  $\text{SO}_2$  and flyash it reduced by 3.1, 11.1, 1.1 and 12.1% at adaxial surface respectively. Plants exposed in field and artificially to  $\text{SO}_2$  and to a combination of  $\text{SO}_2$  and flyash diffusive resistance decreased by 1.6, 5.7 and 5.4% respectively at adaxial surface (Table 3.24). There was no change at adaxial leaf surface in flyash exposed plants. Diffusive resistance of adaxial and abaxial leaf surfaces in control plants of Zea mays was  $2.70 \pm 0.33$  and  $2.32 \pm 0.39$   $\text{cm sec}^{-1}$  respectively (Table 3.23). Diffusive resistance of adaxial leaf surface was more as compared to abaxial surface. In plants exposed in field and artificially to  $\text{SO}_2$ , flyash and a combination of  $\text{SO}_2$  and flyash it reduced by 1.8, 8.2, 0.9 and 8.3% respectively at adaxial leaf surface. At abaxial surface it reduced by 1.9, 8.3 and 8.4% in plants exposed in field and artificially to  $\text{SO}_2$  and a combination of  $\text{SO}_2$  and flyash whereas no change at adaxial surface was observed in plants exposed to flyash (Table 3.24).

A perusal of data (Table 3.24) reveals that (i) Decrease in the diffusive resistance in case of adaxial leaf surface was more than the abaxial leaf surface; (ii) maximum decrease in diffusive resistance was in Medicago sativa followed by Triticum aestivum and Zea mays. Maximum reduction in diffusive resistance (17.7%) was observed in plants exposed artificially to a combination of SO<sub>2</sub> and flyash, followed closely by exposure to SO<sub>2</sub> (17.4) while plants exposed in field and to flyash exhibited marginal reduction.

A comparison of data from Table 3.22 and 3.24 reveals that decrease in diffusive resistance was more at both adaxial and abaxial leaf surface in plants exposed on 90th day than on 45th day to the same dose of pollutant.

#### Effect of long term exposure

Plants were exposed in field and artificially to SO<sub>2</sub> and flyash separately and in combination for 45 (F and A) and 90 days (FF and AA, CF and CA, FC and AC).

F and A exposure: Diffusive resistance of adaxial and abaxial leaf surfaces in control plants of Medicago sativa was  $0.76 \pm 0.11$  and  $0.83 \pm 0.13$  cm sec<sup>-1</sup> respectively (Table 3.25). Diffusive resistance of abaxial

leaf surface was more than adaxial surface. In plants exposed in field and artificially to  $\text{SO}_2$ , flyash and a combination of  $\text{SO}_2$  and flyash it increased by 14.47, 19.72, 5.26 and 17.10% at adaxial surface and 10.34, 15.13, 2.74 and 12.39% in case of abaxial surface respectively (Table 3.26). Data in table 3.26 reveal that change in adaxial leaf surface was more as compared to abaxial leaf surface. Maximum increase was observed in plants exposed to  $\text{SO}_2$ , followed by exposure to a combination of  $\text{SO}_2$  and flyash, in field and to flyash. The same sequence of increase was observed in case of abaxial leaf surface.

Diffusive resistance of adaxial and abaxial leaf surfaces in control plants of Triticum aestivum was  $5.3 \pm 1.05$  and  $8.14 \pm 1.51$   $\text{cm sec}^{-1}$  respectively (Table 3.25). In plants exposed in field and artificially to  $\text{SO}_2$ , flyash and a combination of  $\text{SO}_2$  and flyash it increased by 11.32, 15.09, 7.74 and 13.96% at adaxial surface and 7.32, 10.12, 0.89 and 9.3% at abaxial surface respectively (Table 3.26). In control plants of Zea mays diffusive resistance of adaxial and abaxial leaf surfaces was  $2.7 \pm 0.33$  and  $1.91 \pm 0.31$   $\text{cm sec}^{-1}$  respectively (Table 3.25). Diffusive resistance of adaxial leaf

surface was more as compared to abaxial surface. In plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash, it increased by 9.26, 13.33, 5.19 and 10.0% in case of adaxial and 10.12, 13.24, 3.21 and 12.41% in case of abaxial surfaces respectively (Table 3.24). An examination of the data (Table 3.26) reveal that (i) diffusive resistance of adaxial leaf surface was more than abaxial surface; (ii) maximum increase in diffusive resistance was in Medicago sativa followed by Triticum aestivum and Zea mays; (iii) diffusive resistance increased more in plants exposed to SO<sub>2</sub> followed closely by exposure to a combination of SO<sub>2</sub> and flyash and in field. In flyash exposed plants, it increased marginally.

FF and AA exposure: Diffusive resistance of adaxial and abaxial leaf surfaces in control plants of Medicago sativa was  $0.81 \pm 0.13$  and  $0.9 \pm 0.15$  cm sec<sup>-1</sup> respectively (Table 3.27). Diffusive resistance of abaxial leaf surface was more than adaxial surface. In plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash it increased by 27.17, 24.69, 18.52 and 32.81% in case of adaxial and 16.21, 19.32, 9.34 and 20.47% in case of abaxial surfaces

respectively (Table 3.28). The diffusive resistance of the adaxial leaf surface increased more as compared to abaxial leaf surface. Maximum increase was in case of adaxial leaf surface in plants exposed to a combination of SO<sub>2</sub> and flyash followed by exposure in field, to flyash and to SO<sub>2</sub>. Sequence of increase in diffusive resistance of abaxial leaf surface was same as for the adaxial surface.

In control plants of Triticum aestivum diffusive resistance of adaxial and abaxial leaf surfaces was  $6.22 \pm 1.28$  and  $8.77 \pm 1.63$  cm sec<sup>-1</sup> respectively (Table 3.27). Diffusive resistance of abaxial leaf surface was more than adaxial surface. In plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash it increased by 14.52, 17.72, 9.68 and 20.58% in case of adaxial and 8.65, 13.74, 4.12 and 16.23% in case of abaxial surfaces respectively (Table 3.28). Diffusive resistance of adaxial and abaxial leaf surfaces in control plants of Zea mays was  $3.4 \pm 0.51$  and  $2.32 \pm 0.39$  cm sec<sup>-1</sup> respectively (Table 3.27). Diffusive resistance of adaxial leaf surface was more than abaxial leaf surface. In plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of



SO<sub>2</sub> and flyash it increased by 12.94, 12.06, 9.12 and 16.47% at adaxial and 13.13, 15.46, 7.25 and 18.92% at abaxial surfaces respectively (Table 3.28).

A perusal of the data (Table 3.28) reveals that (i) diffusive resistance in case adaxial leaf surface increased more than abaxial surface; (ii) change in diffusive resistance in Medicago sativa was more as compared to Triticum aestivum and Zea mays; (iii) the maximum increase in diffusive resistance was observed in plants exposed to a combination of SO<sub>2</sub> and flyash, followed by field exposed plants, SO<sub>2</sub> and flyash treated plants.

CF and CA exposure: Diffusive resistance of adaxial and abaxial leaf surfaces in control plants of Medicago sativa, Triticum aestivum and Zea mays was same as described under FF and AA exposures (Table 3.27). In Medicago sativa plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash, diffusive resistance of adaxial surface increased by 17.28, 13.58, 11.11 and 24.61% and 11.94, 11.44, 2.90 and 13.29% for abaxial surface respectively (Table 3.28). In exposed plants of Triticum aestivum it increased by 10.16, 9.35, 6.13 and 13.54% in case of adaxial surface

and 6.48, 7.42, 1.28 and 8.38% in case of abaxial surface respectively. In Zea mays it increased by 9.41, 8.24, 6.47 and 11.96% at adaxial surface and 10.14, 10.17, 2.73 and 12.21% for the abaxial surface respectively (Table 3.28).

FC and AC exposure: Diffusive resistance of adaxial and abaxial leaf surfaces in control plants of Medicago sativa, Triticum aestivum and Zea mays was same as described under FF and AA exposures (Table 3.27).

Increase in the diffusive resistance of less than 5, 4, and 3% was observed in case of adaxial surface in Medicago sativa, Triticum aestivum and Zea mays plants respectively, when exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash. However, increase in diffusive resistance of adaxial leaf surface increased more as compared to abaxial surface.

A perusal of the data (Table 3.28) reveals that (i) increase in the diffusive resistance was more in adaxial leaf surface as compared to abaxial leaf surface; (ii) The diffusive resistance of both adaxial and abaxial leaf surfaces in Medicago sativa increased more as compared to Triticum aestivum and Zea mays; (iii) plants exposed to a combination of SO<sub>2</sub> and flyash exhibited

maximum increase (32.81%) followed by field exposure, SO<sub>2</sub> and flyash exposures.

A comparison of F and A (45 days) and FF and AA (90 days) exposures reveals that increase in diffusive resistance was more in FF and AA exposures as compared to F and AA exposures (Tables 3.26 and 3.28).

### Leaf Surface Temperature

Adaxial leaf surface temperature was measured between 11.00 and 12.00 hrs for three consecutive sunny days in Medicago sativa, Triticum aestivum and Zea mays plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash for 45 (F and A exposure) and 90 days (FF and AA exposure) and compared with controls.

#### F and A exposure

Leaf surface temperature of Medicago sativa in control plants was 30.38°C (Table 3.29). It decreased by 0.45, 1.03 and 0.34°C in plants exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash. However, leaf temperature of flyash exposed plants was higher by 0.61°C (Table 3.30). Leaf surface temperature of control plants of Triticum aestivum was 28.96°C (Table 3.29). It decreased by 0.35, 0.64 and 0.51°C in plants exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash. However, in flyash exposed plants leaf surface temperature increased by 0.42°C (Table 3.30). Leaf surface temperature of control Zea mays plants was 29.75°C (Table 3.29). It decreased by 0.23, 0.22 and 0.41°C in plants exposed in field and

artificially to  $\text{SO}_2$  and a combination of  $\text{SO}_2$  and flyash, however, it increased by  $0.41^\circ\text{C}$  in flyash exposed plants (Table 3.30).

#### FF and AA exposure

Leaf surface temperature in control plants of Medicago sativa was  $30.46^\circ\text{C}$  (Table 3.29). It decreased by 0.45, 1.41 and  $0.93^\circ\text{C}$  in plants exposed in field and artificially to  $\text{SO}_2$  and a combination of  $\text{SO}_2$  and flyash. However, an increase of  $0.82^\circ\text{C}$  was observed in plants exposed to flyash (Table 3.30). Leaf surface temperature in Triticum aestivum plants was  $28.99^\circ\text{C}$  (Table 3.29), in plants exposed in field,  $\text{SO}_2$  and a combination of  $\text{SO}_2$  and flyash, it decreased by 0.52, 0.95 and  $0.73^\circ\text{C}$  respectively. In flyash exposed plants temperature increase was  $0.60^\circ\text{C}$  (Table 3.30). In control Zea mays plants, the leaf surface temperature was  $29.78^\circ\text{C}$  (Table 3.29). In field and artificially exposed plants to  $\text{SO}_2$  and a combination of  $\text{SO}_2$  and flyash, it decreased by 0.33, 0.44 and  $0.41^\circ\text{C}$  respectively. An increase of  $0.50^\circ\text{C}$  was observed in plants exposed to flyash alone (Table 3.30).

A perusal of the data in table 3.30 indicate that plants exposed under FF and AA exposure had maximum reduction in leaf surface temperature in plants exposed

to SO<sub>2</sub> followed by plants exposed to a combination of SO<sub>2</sub> and flyash and in field. Leaf surface temperature increased in flyash exposed plants. Maximum variation in the leaf surface temperature was in exposed plants of Medicago sativa.

#### Absorption Spectra

Absorption spectra in the visible and infra-red regions for adaxial leaf surface were recorded for Medicago sativa, Triticum aestivum and Zea mays, exposed both in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash. Absorption spectra of intact leaf of plants subjected to F and A, and FF and AA exposures were recorded (for details see Chapter II).

In all, four absorption peaks were observed in visible and infra-red regions, two in the visible range (between 420 and 450, and 675 nm) and two in the infra-red region (1450 and 1930 nm). The characteristic features of absorption spectra in the visible and infra-red regions are briefly described below.

In the visible region (380 nm to 740 nm) absorbance gradually increased from 380 nm and reached the maximum between 420-450 nm. It rapidly declines from 450 to 490 nm followed by slow decline from 500 upto a

minimum at 550 nm (Fig. 3.71). The absorption at 550 nm represents the least absorption point of the spectra. From 550 nm onwards, a gradual increase in the absorbance was recorded upto a maximum at 675 nm (Fig. 3.71). It represents second highest peak point of the spectra. There was a steep decline from 675 nm onwards.

The decline continued in the infra-red region (740-2500 nm) from 740 nm to 775 nm followed by a slow increase in absorbance upto 900 nm (Fig. 3.72). From this point gradual decrease was observed till 1250 nm which represents the minimum absorption. Absorption again increased rapidly upto 1450 nm which represents the second highest peak of leaf absorption. A gradual decrease was observed followed by a rather quick increase with maximum absorption at 1930 nm representing the highest peak of the spectra. Thereafter, the absorbance decreases upto 2150 nm followed by a marginal increase upto 2200 nm. It was followed by a decrease in the absorption upto 2250 nm with sudden increase upto 2260 nm which was followed by a gradual increase in absorbance upto 2500 nm (Fig. 3.72).

### Wave lengths for comparison

Variations were observed in the absorption spectra of leaf in exposed plants as compared to their control, in visible and infra-red regions. It has been observed that there was no qualitative shift in the peak of absorption spectrum in any exposed plant. However, in quantitative terms, the absorptivity was found to be different at all wavelengths. For comparison, the wavelengths representing the peak points in visible and infra-red regions were selected.

The wavelengths representing the peaks in absorption leaf spectra in visible as well as in infra-red regions have exhibited similar pattern, irrespective of the plant species (Fig. 3.71-3.72). Two absorption peaks, one at 667 nm in visible region, and the other at 1930 nm in infrared region have been chosen for evaluating the effects of different exposure treatments.

Quantitative changes observed in terms of percent absorbance of leaf have been described here with respect to the exposure pattern, plant species, and nature of pollutants.



### Leaf absorbance in F and A exposure

Visible region: The leaf absorbance in control plants of Medicago sativa was 84.22% (Table 3.31). It decreased by 1.20, 6.10 and 6.0% in plants exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash respectively (Table 3.32, Fig. 3.73). However, in flyash exposed plants, it increased by 2.84%. In control plants of Triticum aestivum, leaf absorbance was 78.22% and it decreased by 3.22%, 8.30 and 6.66% in plants exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash respectively. In flyash exposed plants, it increased by 0.98% (Table 3.32, Fig. 3.74). The leaf absorbance in control plants of Zea mays was 82.01%. It decreased by 0.21%, 6.28% and 5.45% in plants exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash respectively (Fig. 3.75). However, in flyash exposed plants it increased by 1.62%.

Infra-red region: Leaf absorbance in control plants of Medicago sativa was 88.52% (Table 3.31, Fig. 3.76). It decreased by 2.01, 12.51 and 3.73% in plants exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash respectively (Table 3.32). However, in flyash exposed plants absorption increased by 1.48%.

### Leaf absorbance in F and A exposure

Visible region: The leaf absorbance in control plants of Medicago sativa was 84.22% (Table 3.31). It decreased by 1.20, 6.10 and 6.0% in plants exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash respectively (Table 3.32, Fig. 3.73). However, in flyash exposed plants, it increased by 2.84%. In control plants of Triticum aestivum, leaf absorbance was 78.22% and it decreased by 3.22%, 8.30 and 6.66% in plants exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash respectively. In flyash exposed plants, it increased by 0.98% (Table 3.32, Fig. 3.74). The leaf absorbance in control plants of Zea mays was 82.01%. It decreased by 0.21%, 6.28% and 5.45% in plants exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash respectively (Fig. 3.75). However, in flyash exposed plants it increased by 1.62%.

Infra-red region: Leaf absorbance in control plants of Medicago sativa was 88.52% (Table 3.31, Fig. 3.76). It decreased by 2.01, 12.51 and 3.73% in plants exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash respectively (Table 3.32). However, in flyash exposed plants absorption increased by 1.48%.

In control plants of Triticum aestivum, leaf absorbance was 79.96% and it decreased by 0.85, 5.08 and 8.14% in plants exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash respectively (Fig. 3.77). In flyash exposed plants it increased by 0.09%. The leaf absorbance in control plants of Zea mays was 82.83% (Fig. 3.78). It decreased by 0.20, 3.62 and 2.68% in plants exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash respectively. However, in flyash exposed plants it increased by 0.89% (Table 3.32).

A perusal of data in table 3.32 reveals that maximum reduction in leaf absorbance both in visible and in infra-red regions was in SO<sub>2</sub> exposed plants followed by plants exposed to a combination of SO<sub>2</sub> and flyash artificially. Field exposed plants, exhibited the least reduction. However, in flyash treated plants, absorbance increased both in visible and infrared regions.

#### Leaf absorbance in FF and AA exposure

**Visible region:** The leaf absorbance in control plants of Medicago sativa was 87.12% (Table 3.31, Fig. 3.79). In plants exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash, it decreased by 3.44%, 11.39% and 6.62% respectively. In flyash exposed plants,

it increased by 1.4% (Table 3.32). Leaf absorbance in control plants of Triticum aestivum was 79.58%. It decreased by 3.35, 15.06 and 9.57% in plants exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash respectively. However, in flyash exposed plants, it increased by 2.26% (Fig. 3.80). The leaf absorbance in control plants of Zea mays was 85.21% (Fig. 3.81). In plants exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash it decreased by 2.0, 8.38 and 3.2% respectively. In flyash exposed plants, it increased by 1.55% (Table 3.32).

**Infra-red region:** Leaf absorbance in control plants of Medicago sativa was 87.0% (Table 3.31, Fig. 3.82). In plants exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash it decreased by 0.34%, 15.51% and 2.49% respectively. However, in flyash exposed plants it increased by 1.17% (Table 3.32). In control plants of Triticum aestivum, the leaf absorbance was 82.62% (Fig. 3.83). In plants exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash it decreased by 0.4%, 14.34% and 0.73% respectively. However, it increased by 3.73% in flyash exposed plants (Table 3.84). It decreased by 3.66%, 6.85% and 3.9%

in plants exposed in field and artificially to  $\text{SO}_2$  and a combination of  $\text{SO}_2$  and flyash respectively. In flyash exposed plants, it increased by 1.21% (Table 3.32).

The data in table 3.32 show that maximum reduction in visible and infrared regions was in  $\text{SO}_2$  exposed plants followed by plants exposed to a combination of  $\text{SO}_2$  and flyash artificially. Field exposed plants have shown least reduction. In flyash treated plants, leaf absorbance increased both in visible and infra-red regions.

Table 3.1: Leaf area ( $\text{cm}^2$ ) of Medicago sativa, Triticum aestivum and Zea mays plants exposed for 45 days (F and A) and for 90 days (FF and AA, CF and CA and FC and AC) in field and artificially to  $\text{SO}_2$ , flyash and a combination of  $\text{SO}_2$  and flyash

Exposure pattern	Species	Field Exposure	Artificial Exposure			Control
			$\text{SO}_2$	Flyash	$\text{SO}_2$ + flyash	
F and A	Medicago sativa	738.01 +70.24	732.31 +79.41	846.47 +81.23	736.38 +76.38	815.48 +84.52
	Triticum aestivum	518.85 +39.41	523.37 +44.54	583.87 +48.32	526.77 +42.32	565.21 +50.12
	Zea mays	1132.66 +101.41	1125.51 +100.52	1201.81 +126.12	1129.08 +114.31	1192.27 +123.24
FF and AA	Medicago sativa	1138.07 +100.42	1119.83 +103.84	1303.66 +121.23	1086.14 +107.32	1403.29 +134.68
	Triticum aestivum	562.47 +50.23	571.82 +49.23	611.89 +54.12	555.12 +47.42	667.99 +61.72
	Zea mays	1138.43 +161.23	1871.41 +151.31	1853.72 +154.42	1830.79 +149.25	1911.68 +166.23
CF and CA	Medicago sativa	1334.53 +101.74	1323.16 +144.74	1339.86 +109.82	1272.1 +112.29	1403.29 +134.68
	Triticum aestivum	641.94 +56.41	644.62 +54.38	649.16 +58.52	637.27 +51.24	667.99 +61.92
	Zea mays	1865.18 +161.23	1849.85 +151.24	1870.92 +162.23	1859.45 +157.41	1911.05 +166.23
FC and AC	Medicago sativa	1349.96 +111.92	1334.53 +121.52	1445.39 +127.32	1344.35 +117.32	1403.29 +132.68
	Triticum aestivum	651.96 +61.24	646.62 +54.48	682.01 +62.39	649.96 +56.83	667.99 +61.92
	Zea mays	1880.45 +159.92	1870.92 +154.24	1935.65 +161.29	1878.55 +161.42	1911.05 +160.23

Table 3.2: Change in leaf area in Medicago sativa, Triticum aestivum and Zea mays plants exposed for 45 days (F and A) and for 90 days (FF and AA, CF and CA, FC and AC) in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash (Expressed as % change over control)

Exposure pattern	Species	Field Exposure	Artificial Exposure		
			SO <sub>2</sub>	Flyash	SO <sub>2</sub> + flyash
F and A	Medicago sativa	-9.5	-10.2	-3.8	-9.7
	Triticum aestivum	-8.2	-7.4	-3.3	-6.8
	Zea mays	-5.0	-5.6	-0.4	-5.3
FF and AA	Medicago sativa	-18.9	-20.2	-7.1	-22.6
	Triticum aestivum	-15.8	-14.4	-5.4	-16.9
	Zea mays	-7.2	-8.7	-2.3	-8.4
CF and CA	Medicago sativa	-4.9	-5.71	-4.52	-9.4
	Triticum aestivum	-3.9	-3.5	-2.82	-4.6
	Zea mays	-2.1	-3.6	-1.49	-3.8
FC and AC	Medicago sativa	-3.8	-4.9	-5.0	-4.2
	Triticum aestivum	-2.4	-3.2	-2.1	-2.7
	Zea mays	-1.6	-2.1	-0.24	-1.7

Table 3.3: Leaf biomass (g) of Medicago sativa, Triticum aestivum and Zea mays plants exposed for 45 days (F and A) and for 90 days (FF and AA, CF and CA, FC and AC) in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash

Exposure pattern	Species	Field Exposure	Artificial Exposure			Control
			SO <sub>2</sub>	Flyash	SO <sub>2</sub> +Flyash	
F and A	Medicago sativa	1.309±0.14	1.270±0.10	1.638±0.13	1.293±0.09	1.564±0.12
	Triticum aestivum	1.282±0.18	1.266±0.21	1.507±0.20	1.291±0.26	1.452±0.23
	Zea mays	5.753±0.62	5.728±0.63	6.195±0.94	5.765±0.71	6.146±0.93
FF and AA	Medicago sativa	1.939±0.20	2.018±0.31	2.429±0.31	1.880±0.31	2.720±0.33
	Triticum aestivum	1.358±0.20	1.380±0.15	1.588±0.1	1.320±0.29	1.734±0.28
	Zea mays	6.678±0.82	6.830±0.81	7.202±0.62	6.601±0.81	7.597±0.14
CF and CA	Medicago sativa	2.459±0.21	2.508±0.29	2.568±0.24	2.401±0.17	2.720±0.33
	Triticum aestivum	1.621±0.19	1.633±0.30	1.652±0.19	1.594±0.21	1.734±0.28
	Zea mays	7.384±0.82	7.293±0.92	7.458±0.81	7.286±0.77	7.597±0.14
FC and AC	Medicago sativa	2.573±0.30	2.527±0.21	2.812±0.35	2.554±0.34	2.720±0.33
	Triticum aestivum	1.668±0.20	1.664±0.21	1.777±0.21	1.661±0.19	1.734±0.28
	Zea mays	7.392±0.88	7.316±0.89	7.643±0.64	7.384±0.45	7.587±0.14



Table 3.4: Change in leaf biomass of Medicago sativa, Triticum aestivum and Zea mays plants exposed for 45 days (F and A) and for 90 days (FF and AA, CF and CA, FC and AC) in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash (Expressed as % change over Control)

Exposure pattern	Species	Field Exposure	Artificial Exposure		
			SO <sub>2</sub>	Flyash	SO <sub>2</sub> + flyash
F and A	Medicago sativa	-16.3	-18.8	+2.7	-7.1
	Triticum aestivum	-11.7	-12.8	+3.8	-11.1
	Zea mays	-6.4	-6.8	+0.8	-6.2
FF and AA	Medicago sativa	-28.7	-25.8	-10.7	-31.2
	Triticum aestivum	-21.7	-20.4	-8.4	-23.9
	Zea mays	-12.1	-10.1	-5.2	-13.1
CF and CA	Medicago sativa	-9.6	-7.4	-5.6	-11.4
	Triticum aestivum	-6.5	-5.8	-4.7	-8.1
	Zea mays	-2.8	-4.0	-1.8	-4.1
FC and AC	Medicago sativa	-5.4	-7.1	+3.4	-6.1
	Triticum aestivum	-3.8	-5.2	+2.5	-4.2
	Zea mays	-2.7	-3.7	+0.6	-2.8

Table 3.5: Total plant biomass (g) of Medicago sativa, Triticum aestivum and Zea mays plants exposed for 45 days (F and A) in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash

Species	Plant part	Field Exposure	Artificial Exposure			Control
			SO <sub>2</sub>	Flyash	SO <sub>2</sub> + flyash	
Medicago sativa	Stem	1.638±0.12	1.584±0.13	1.938±0.16	1.608±0.13	1.854±0.15
	Leaf	1.309±0.10	1.270±0.10	1.638±0.13	1.293±0.09	1.564±0.12
	Root	1.548±0.17	2.450±0.13	3.045±0.26	2.466±0.31	2.949±0.28
	Total	5.495±0.35	5.367±0.439	6.621±0.53	5.367±0.44	6.367±0.57
Triticum aestivum	Stem	3.258±0.29	3.201±0.31	3.614±0.27	3.229±0.21	3.508±0.38
	Leaf	1.282±0.18	1.266±0.21	1.507±0.20	1.291±0.20	1.452±0.23
	Root	1.745±0.13	1.604±0.16	1.982±0.16	1.630±0.10	1.923±0.20
	Total	6.285±0.68	6.071±0.73	7.103±0.79	6.150±0.54	6.883±0.71
Zea mays	Stem	17.638±1.32	17.481±1.92	18.478±2.10	17.567±2.12	18.335±2.42
	Leaf	5.753±0.62	5.728±0.64	6.195±0.94	5.765±0.71	6.146±0.93
	Root	7.529±0.57	7.384±0.81	8.135±0.74	7.488±0.82	8.102±0.91
	Total	30.920±4.91	30.593±5.02	32.808±5.67	30.820±4.16	32.583±5.12

Table 3.6: Total plant biomass (g) of Medicago sativa, Triticum aestivum and Zea mays plants exposed for 90 days (FF and AA) in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash

Species	Plant part	Field Exposure	Artificial Exposure			Control
			SO <sub>2</sub>	Flyash	SO <sub>2</sub> + flyash	
Medicago sativa	Fruit	0.306±0.04	0.314±0.03	0.334±0.04	0.300±0.05	0.367±0.05
	Stem	2.438±0.32	3.512±0.42	3.873±0.53	2.367±0.34	3.192±0.40
	Leaf	1.939±0.26	2.018±0.31	2.429±0.31	1.880±0.31	2.720±0.33
	Root	3.289±0.41	2.465±0.33	2.624±0.41	2.952±0.41	3.947±0.44
	Total	7.972±1.23	8.309±0.98	9.260±1.28	7.501±1.10	10.226±1.31
Triticum aestivum	Fruit	1.649±0.20	1.709±0.20	1.812±0.17	1.601±0.09	1.874±0.21
	Stem	4.046±0.39	4.121±0.52	4.531±0.44	3.972±0.51	4.938±0.42
	Leaf	1.358±0.20	1.380±0.15	1.588±0.11	1.320±0.18	1.734±0.29
	Root	2.052±0.22	2.184±0.18	2.398±0.20	2.045±0.18	2.584±0.26
	Total	9.105±0.92	9.394±1.10	10.329±1.01	8.938±0.92	11.130±1.39
Zea mays	Fruit	7.924±0.81	8.017±0.92	8.236±0.91	7.879±0.91	8.432±0.92
	Stem	18.874±2.11	19.124±2.31	19.934±2.14	18.739±1.60	20.369±3.12
	Leaf	6.678±0.78	6.830±0.81	7.202±0.62	6.601±0.81	7.597±1.14
	Root	8.234±1.12	8.239±0.71	7.563±0.61	8.084±0.92	9.132±1.22
	Total	41.710±5.01	42.210±4.81	42.935±4.67	41.300±4.01	45.532±5.47

Table 3.7: Total plant biomass (g) of Medicago sativa, Triticum aestivum and Zea mays plants exposed for 90 days (CF and CA) in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash

Species	Plant part	Field Exposure	Artificial Exposure			Control
			SO <sub>2</sub>	Flyash	SO <sub>2</sub> + flyash	
Medicago sativa	Fruit	1.328±0.04	0.334±0.03	0.351±0.04	0.316±0.04	0.367±0.05
	Stem	2.871±0.32	2.914±0.26	3.016±0.33	2.826±0.38	3.192±0.40
	Leaf	2.459±0.21	2.508±0.29	2.568±0.29	2.410±0.17	2.720±0.33
	Root	3.650±0.38	3.637±0.40	3.846±0.42	3.628±0.39	3.947±0.44
	Total	9.308±1.23	9.393±1.08	9.781±1.18	9.178±1.02	10.226±1.31
Triticum aestivum	Fruit	1.719±0.21	1.742±0.02	1.792±0.01	1.701±0.19	1.874±0.23
	Stem	4.614±0.51	4.641±0.52	4.774±0.41	4.582±0.55	4.938±0.42
	Leaf	1.621±0.19	1.633±0.30	1.652±0.19	1.594±0.21	1.734±0.29
	Root	2.498±0.28	2.350±0.28	2.522±0.21	2.430±0.29	2.584±0.26
	Total	10.452±1.32	10.366±1.12	10.745±1.01	10.307±1.22	11.150±1.59
Zea mays	Fruit	8.362±0.91	8.329±0.73	8.391±0.82	8.342±0.67	8.432±0.92
	Stem	19.846±2.52	19.784±1.83	20.294±2.11	19.781±2.12	20.369±3.12
	Leaf	7.384±0.82	7.293±0.92	7.458±0.81	7.286±0.77	7.597±1.14
	Root	8.938±0.71	8.814±1.19	9.235±1.12	8.713±0.76	9.132±1.22
	Total	44.530±5.13	44.220±5.02	45.376±5.07	44.120±4.89	45.532±5.47

Table 3.8: Total plant biomass (g) of Medicago sativa, Triticum aestivum and Zea mays plants exposed for 90 days (FC and AC) in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash

Species	Plant part	Field Exposure	Artificial Exposure			Control
			SO <sub>2</sub>	Flyash	SO <sub>2</sub> + flyash	
Medicago sativa	Fruit	0.360±0.04	0.356±0.02	0.377±0.03	0.360±0.03	0.367±0.105
	Stem	3.014±0.29	2.942±0.27	3.274±0.34	3.002±0.27	3.192±0.40
	Leaf	2.573±0.30	2.527±0.21	2.812±0.35	2.554±0.34	2.720±0.33
	Root	3.783±0.42	3.813±0.45	4.084±0.51	3.773±0.31	3.947±0.44
	Total	9.730±1.08	9.638±0.91	10.547±1.19	9.689±1.64	10.226±1.31
Triticum aestivum	Fruit	1.864±0.16	1.861±0.17	1.882±0.26	1.864±0.21	1.874±0.21
	Stem	4.816±0.52	4.800±0.51	5.037±0.48	4.801±0.50	4.938±0.42
	Leaf	1.668±0.20	1.644±0.21	1.777±0.21	1.661±0.19	1.734±0.29
	Root	2.437±0.29	2.358±0.31	2.678±0.30	2.393±0.34	2.584±0.26
	Total	10.785±1.23	10.663±1.31	11.374±1.27	10.719±1.19	11.130±1.39
Zea mays	Fruit	8.354±0.92	8.300±1.10	8.462±1.21	8.326±0.97	8.432±0.92
	Stem	20.062±2.31	20.000±2.31	20.432±2.47	20.041±2.41	20.369±3.12
	Leaf	7.392±0.88	7.316±0.89	7.643±0.64	7.384±0.95	7.597±1.14
	Root	8.724±1.23	8.684±1.12	9.223±0.83	8.791±0.89	9.132±1.22
	Total	44.532±5.92	44.300±5.32	45.760±5.07	44.542±5.61	45.532±5.47

Table 3.9: Change in total plant biomass of Medicago sativa, Triticum aestivum and Zea mays plants exposed for 45 days (F and A) and 90 days (FF and AA, CF and CA, FC and AC) in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash. (Expressed as % change over control)

Exposure pattern	Species	Field Exposure	Artificial Exposure		
			SO <sub>2</sub>	Flyash	SO <sub>2</sub> + flyash
F and A	Medicago sativa	-13.7	-16.7	+ 4.1	-15.8
	Triticum aestivum	- 8.7	-11.8	+ 3.2	-10.6
	Zea mays	- 5.1	- 6.1	+ 0.7	- 5.4
FF and AA	Medicago sativa	-22.4	-18.7	- 9.4	-26.6
	Triticum aestivum	-18.2	-15.6	- 7.2	-19.7
	Zea mays	- 8.4	- 7.3	- 5.7	- 9.7
CF and CA	Medicago sativa	- 9.0	- 8.1	- 4.3	-10.2
	Triticum aestivum	- 6.1	- 6.87	- 3.46	- 7.4
	Zea mays	- 2.2	- 2.89	- 0.37	- 3.1
FC and AC	Medicago sativa	- 4.8	- 5.7	+ 3.2	- 5.2
	Triticum aestivum	- 3.1	- 4.2	+ 2.2	- 3.7
	Zea mays	- 2.2	- 2.7	+ 0.51	- 2.4

Table 3.10: Chlorophyll content ( $\text{mg g}^{-1}$ ) of Medicago sativa, Triticum aestivum and Zea mays plants exposed for 45 days (F and A) and 90 days (FF and AA, CF and CA, FC and AC) in field and artificially to  $\text{SO}_2$ , flyash and a combination of  $\text{SO}_2$  and flyash

Exposure pattern	Species	Field Exposure	Artificial Exposure			Control
			$\text{SO}_2$	Flyash	$\text{SO}_2$ + Flyash	
F and A	Medicago sativa	3.549 $\pm$ 0.27	3.391 $\pm$ 0.29	4.075 $\pm$ 0.35	3.513 $\pm$ 0.26	3.952 $\pm$ 0.34
	Triticum aestivum	3.312 $\pm$ 0.27	3.213 $\pm$ 0.29	3.590 $\pm$ 0.29	3.298 $\pm$ 0.30	3.516 $\pm$ 0.31
	Zea mays	3.650 $\pm$ 0.31	3.600 $\pm$ 0.33	3.805 $\pm$ 0.34	3.640 $\pm$ 0.32	3.767 $\pm$ 0.33
FF and AA	Medicago sativa	3.383 $\pm$ 0.22	3.205 $\pm$ 0.28	3.667 $\pm$ 0.29	3.330 $\pm$ 0.24	3.968 $\pm$ 0.38
	Triticum aestivum	3.100 $\pm$ 0.29	3.020 $\pm$ 0.28	3.330 $\pm$ 0.31	3.083 $\pm$ 0.29	3.528 $\pm$ 0.33
	Zea mays	3.557 $\pm$ 0.30	3.439 $\pm$ 0.31	3.646 $\pm$ 0.29	3.535 $\pm$ 0.30	3.779 $\pm$ 0.38
CF and CA	Medicago sativa	3.600 $\pm$ 0.29	3.474 $\pm$ 0.31	3.808 $\pm$ 0.28	3.573 $\pm$ 0.29	3.968 $\pm$ 0.38
	Triticum aestivum	3.354 $\pm$ 0.31	3.290 $\pm$ 0.29	3.450 $\pm$ 0.31	3.330 $\pm$ 0.29	3.528 $\pm$ 0.33
	Zea mays	3.695 $\pm$ 0.29	3.638 $\pm$ 0.34	3.726 $\pm$ 0.32	3.684 $\pm$ 0.32	3.779 $\pm$ 0.38
FC and AC	Medicago sativa	3.807 $\pm$ 0.28	3.785 $\pm$ 0.28	4.035 $\pm$ 0.32	3.790 $\pm$ 0.29	3.968 $\pm$ 0.38
	Triticum aestivum	3.449 $\pm$ 0.30	3.430 $\pm$ 0.28	3.560 $\pm$ 0.32	3.445 $\pm$ 0.29	3.528 $\pm$ 0.33
	Zea mays	3.725 $\pm$ 0.31	3.695 $\pm$ 0.29	3.790 $\pm$ 0.30	3.714 $\pm$ 0.32	3.779 $\pm$ 0.38

Table 3.11: Change in chlorophyll content of Medicago sativa, Triticum aestivum and Zea mays plants exposed for 45 days (F and A) and 90 days (FF and AA, CF and CA, FC and AC) in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash (Expressed as % change over Control)

Exposure pattern	Species	Field Exposure	Artificial Exposure		
			SO <sub>2</sub>	Flyash	SO <sub>2</sub> + flyash
F and A	Medicago sativa	-10.2	-14.2	+ 3.1	-11.1
	Triticum aestivum	- 5.8	- 8.5	+ 2.1	- 6.2
	Zea mays	- 3.1	- 4.2	+ 1.3	- 3.3
FF and AA	Medicago sativa	-14.4	-18.9	- 7.2	-15.7
	Triticum aestivum	-11.8	-14.1	- 5.3	-12.3
	Zea mays	- 5.6	- 8.7	- 3.2	- 6.1
CF and CA	Medicago sativa	- 8.9	-12.1	- 3.7	- 9.6
	Triticum aestivum	- 4.6	- 6.4	- 2.0	- 5.3
	Zea mays	- 1.9	- 3.4	- 1.1	- 2.2
FC and AC	Medicago sativa	- 3.7	- 4.3	+ 2.1	- 4.1
	Triticum aestivum	- 1.9	- 2.5	+ 1.3	- 2.0
	Zea mays	- 1.1	- 1.9	+ 0.62	- 1.4



Table 3.12: Epidermal features of Medicago sativa,  
Triticum aestivum and Zea mays plants

Species	Stomatal type	Leaf surface	Trichome type
Medicago sativa	Anomocytic	Adaxial	Glabrous - smooth surface free from hairs
		Abaxial	Glabrous - smooth surface free from hairs
Triticum aestivum	Paracytic	Adaxial	Glabrescent - short hairs
		Abaxial	Glabrescent - short hairs
Zea mays	Paracytic	Adaxial	Pubescent - short soft and straight hairs
		Abaxial	Glabrescent - short hairs

Table 3.13: Stomatal density (no. mm<sup>-2</sup>) of adaxial and abaxial leaf surfaces of Medicago sativa, Triticum aestivum (adaxial leaf surface) and Zea mays plants exposed for 90 days (FF and AA) in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash

Species	Leaf Surface	Field Exposure	Artificial Exposure			Control
			SO <sub>2</sub>	Flyash	SO <sub>2</sub> + flyash	
Medicago sativa	Adaxial	131	-	-	111	171
	Abaxial	142	-	-	131	156
Triticum aestivum	Adaxial	42	39	43	39	46
	Abaxial	-	-	-	-	-
Zea mays	Adaxial	46	53	-	49	64
	Abaxial	101	92	104	95	110

Table 3.14: Change in stomatal density of adaxial and abaxial leaf surfaces of Medicago sativa, Triticum aestivum (adaxial leaf surface) and Zea mays exposed for 90 days (FF and AA) in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash  
(Expressed as % change over control)

Species	Leaf surface	Field Exposure	Artificial Exposure		
			SO <sub>2</sub>	Flyash	SO <sub>2</sub> + flyash
Medicago sativa	Adaxial	-23.39	-	-	-35.09
	Abaxial	- 8.77	-	-	-16.58
Triticum aestivum	Adaxial	- 8.69	-15.22	- 6.52	-15.22
	Abaxial	- -	-	-	-
Zea mays	Adaxial	-28.12	-17.18	-	-23.43
	Abaxial	- 8.18	-16.36	- 5.45	-13.64

Table 3.15: Length ( $\mu$ ) and breadth ( $\mu$ ) of stomatal pore of adaxial and abaxial leaf surface of Medicago sativa, Triticum aestivum and Zea mays plants exposed for 90 days (FF and AA) in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash.

Species	Leaf surface	Stomatal pore	Field Exposure	Artificial Exposure			Control
				SO <sub>2</sub>	Flyash	SO <sub>2</sub> + Flyash	
Medicago sativa	Adaxial	Length	6.0	6.3	6.1	6.0	10.6
		Breadth	0.5	1.5	2.7	1.0	2.0
	Abaxial	Length	7.6	6.1	8.8	5.5	8.5
		Breadth	0.5	1.6	1.6	0.5	2.0
Triticum aestivum	Adaxial	Length	33.0	28.0	29.0	24.0	39.5
		Breadth	1.5	0.5	2.0	1.5	1.5
	Abaxial	Length	33.0	21.0	-	30.0	42.0
		Breadth	1.5	0.9	-	0.9	1.0
Zea mays	Adaxial	Length	25.0	32.2	27.5	26.0	30.0
		Breadth	3.3	3.1	3.1	2.2	5.0
	Abaxial	Length	29.6	32.7	30.0	37.5	34.0
		Breadth	2.1	3.6	2.1	0.5	4.4

Table 3.16: Change in length and breadth of stomatal pore of adaxial and abaxial leaf surfaces of Medicago sativa, Triticum aestivum and Zea mays plants exposed for 90 days (FF and AA) in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash. (Expressed as % change over Control)

Species	Leaf Surface	Stomatal pore	Field Exposure	Artificial Exposure		
				SO <sub>2</sub>	Flyash	SO <sub>2</sub> + Flyash
Medicago sativa	Adaxial	Length	-43.40	-40.57	-42.45	-43.40
		Breadth	-75.00	-25.00	+35.00	-50.00
	Abaxial	Length	-10.59	-28.24	+ 3.53	-35.29
		Breadth	-75.00	-20.00	-20.00	-75.00
Triticum aestivum	Adaxial	Length	-16.46	-29.11	-26.58	-39.24
		Breadth	0	-66.67	+33.33	0
	Abaxial	Length	-21.43	-50.00	-	-28.57
		Breadth	+50.00	-10.00	-	-10.00
Zea mays	Adaxial	Length	-16.67	+ 7.33	- 8.33	-13.33
		Breadth	-34.00	-38.00	-38.00	-56.00
	Abaxial	Length	-13.53	- 3.82	-11.76	+ 7.35
		Breadth	-29.55	-18.18	-29.55	-88.64

Table 3.17: Trichome density (no. mm<sup>-2</sup>) of adaxial and abaxial leaf surfaces of Triticum aestivum plants exposed for 90 days (FF and AA) in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash

Species	Leaf Surface	Field Exposure	Artificial Exposure			Control
			SO <sub>2</sub>	Flyash	SO <sub>2</sub> + flyash	
Triticum aestivum	Adaxial	106	73	78	98	60
	Abaxial	61	57	59	67	49

Table 3.18: Change in trichome density of adaxial and abaxial leaf surfaces of Triticum aestivum plants exposed for 90 days (FF and AA) in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash (Expressed as % change over control)

Species	Leaf Surface	Field Exposure	Artificial Exposure		
			SO <sub>2</sub>	Flyash	SO <sub>2</sub> + Flyash
Triticum aestivum	Adaxial	+76.66	+21.66	+30.00	+58.33
	Abaxial	+24.49	+15.96	+20.41	+36.73

Table 3.19: Trichome length ( $\mu$ ) of adaxial and abaxial leaf surfaces of Triticum aestivum plants exposed for 90 days (FF and AA) in field and artificially to  $\text{SO}_2$ , flyash and a combination of  $\text{SO}_2$  and flyash

Leaf surface	Field Exposure	Artificial Exposure			Control
		$\text{SO}_2$	Flyash	$\text{SO}_2$ + flyash	
Adaxial	41.3 $\pm$ 9.7	38.7 $\pm$ 7.4	44.6 $\pm$ 11.8	46.8 $\pm$ 11.9	36.4 $\pm$ 7.3
Abaxial	50.6 $\pm$ 11.2	48.3 $\pm$ 8.6	52.4 $\pm$ 13.1	52.1 $\pm$ 12.6	45.5 $\pm$ 10.7

Table 3.20: Change in trichome length of adaxial and abaxial leaf surfaces of Triticum aestivum plants exposed for 90 days (FF and AA) in field and artificially to  $\text{SO}_2$ , flyash and a combination of  $\text{SO}_2$  and flyash (Expressed as % change over Control)

Leaf surface	Field Exposure	Artificial Exposure		
		$\text{SO}_2$	Flyash	$\text{SO}_2$ + flyash
Adaxial	+13.89	+5.56	+25.00	+27.78
Abaxial	+11.11	+6.67	+15.56	+15.56

Table 3.21: Diffusive resistance ( $\text{sec cm}^{-1}$ ) of adaxial and abaxial surfaces of Medicago sativa, Triticum aestivum and Zea mays plants exposed on 45th day in field (24 h) and artificially to  $\text{SO}_2$  ( $1 \text{ h d}^{-1}$ ) and flyash separately and in combination

Species	Leaf Surface	Field Exposure	Artificial Exposure			Control
			$\text{SO}_2$	Flyash	$\text{SO}_2 + \text{Flyash}$	
Medicago sativa	Adaxial	0.731 $\pm$ 0.092	0.663 $\pm$ 0.084	0.751 $\pm$ 0.130	0.660 $\pm$ 0.072	0.760 $\pm$ 0.110
	Abaxial	0.806 $\pm$ 0.121	0.756 $\pm$ 0.084	0.830 $\pm$ 0.131	0.746 $\pm$ 0.098	0.830 $\pm$ 0.131
Triticum aestivum	Adaxial	5.160 $\pm$ 0.91	4.840 $\pm$ 1.01	5.250 $\pm$ 1.11	4.830 $\pm$ 0.72	5.300 $\pm$ 1.05
	Abaxial	8.040 $\pm$ 1.32	7.710 $\pm$ 1.23	8.140 $\pm$ 1.51	7.740 $\pm$ 1.12	8.140 $\pm$ 1.51
Zea mays	Adaxial	2.660 $\pm$ 0.27	2.520 $\pm$ 0.23	2.680 $\pm$ 0.31	2.520 $\pm$ 0.31	2.700 $\pm$ 0.33
	Abaxial	1.880 $\pm$ 0.26	1.700 $\pm$ 0.29	1.910 $\pm$ 0.31	1.760 $\pm$ 0.22	1.910 $\pm$ 0.31



Table 3.22: Change in diffusive resistance of adaxial and abaxial leaf surfaces of Medicago sativa, Triticum aestivum and Zea mays plants exposed on 45th day in field (24 h) and artificially to SO<sub>2</sub> (1 h d<sup>-1</sup>) and to flyash separately and in combination (Expressed as % change over control)

Species	Leaf surface	Field Exposure	Artificial Exposure		
			SO <sub>2</sub>	Flyash	SO <sub>2</sub> + flyash
Medicago sativa	Adaxial	-3.8	-12.8	-1.2	-13.1
	Abaxial	-2.9	- 8.4	0	-10.1
Triticum aestivum	Adaxial	-2.6	- 8.6	-0.8	- 8.8
	Abaxial	-1.2	- 5.2	0	- 4.9
Zea mays	Adaxial	-1.4	- 6.8	-0.6	- 6.8
	Abaxial	-1.4	- 5.4	0	- 6.8

Table 3.23: Diffusive resistance ( $\text{sec cm}^{-1}$ ) of adaxial and abaxial leaf surfaces of Medicago sativa, Triticum aestivum and Zea mays plants exposed on 90th day in field (24 h) and artificially to  $\text{SO}_2$  ( $1 \text{ h d}^{-1}$ ) and to flyash separately and in combination

Species	Leaf Surface	Field Exposure	Artificial Exposure			Control
			$\text{SO}_2$	Flyash	$\text{SO}_2 + \text{Flyash}$	
Medicago sativa	Adaxial	0.77±0.098	0.669±0.092	0.797±0.151	0.666±0.075	0.81±0.131
	Abaxial	0.87±0.112	0.793±0.107	0.900±0.154	0.788±0.134	0.90±0.154
Triticum aestivum	Adaxial	6.00±1.72	5.460±0.93	6.130±1.21	5.450±0.960	6.20±1.280
	Abaxial	8.62±1.39	8.270±1.01	8.770±1.63	8.290±1.470	8.77±1.630
Zea mays	Adaxial	3.33±0.41	2.520±0.23	2.680±0.31	2.520±0.31	2.70±0.330
	Abaxial	2.77±0.41	2.130±0.34	2.320±0.39	2.120±0.36	2.32±0.390

Table 3.24: Change in diffusive resistance of adaxial and abaxial leaf surfaces of Medicago sativa, Triticum aestivum and Zea mays plants exposed on 90th day in field (24 h) and artificially to SO<sub>2</sub> (1 h d<sup>-1</sup>) and to flyash, separately and in combination (Expressed as % change over Control)

Species	Leaf surface	Field Exposure	Artificial Exposure		
			SO <sub>2</sub>	Flyash	SO <sub>2</sub> + flyash
Medicago sativa	Adaxial	-4.9	-17.4	-1.6	-17.7
	Abaxial	-3.1	-11.9	zero	-12.4
Triticum aestivum	Adaxial	-3.1	-11.1	-1.11	-12.1
	Abaxial	-1.6	- 5.7	zero	- 5.4
Zea mays	Adaxial	-1.8	- 8.2	0.9	- 8.3
	Abaxial	-1.9	- 8.3	zero	- 8.4

Table 3.25: Diffusive resistance ( $\text{cm sec}^{-1}$ ) of adaxial and abaxial leaf surfaces of Medicago sativa, Triticum aestivum and Zea mays plants exposed for 45 days (F and A) in field and artificially to  $\text{SO}_2$ , flyash and a combination of  $\text{SO}_2$  and flyash.

Species	Leaf Surface	Field Exposure	Artificial Exposure			Control
			$\text{SO}_2$	Flyash	$\text{SO}_2$ + Flyash	
Medicago sativa	Adaxial	0.869±0.120	0.909±0.134	0.799±0.110	0.889±0.123	0.76±0.112
	Abaxial	0.916±0.130	0.956±0.101	0.856±0.121	0.933±0.149	0.83±0.131
Triticum aestivum	Adaxial	5.900±1.12	6.100±1.23	5.710±1.27	6.040±1.19	5.30±1.05
	Abaxial	8.700±1.38	8.970±1.21	8.220±1.08	9.930±1.69	8.14±1.51
Zea mays	Adaxial	2.950±0.38	3.060±0.46	2.860±0.35	2.970±0.41	2.70±0.33
	Abaxial	2.100±0.36	2.150±0.42	1.970±0.37	2.150±0.39	1.91±0.31

Table 3.26: Change in diffusive resistance of adaxial and abaxial leaf surfaces of Medicago sativa, Triticum aestivum and Zea mays plants exposed for 45 days (F and A) in field and artificially to SO<sub>2</sub> and flyash and a combination of SO<sub>2</sub> and flyash (Expressed as % change over Control)

Species	Leaf Surface	Field Exposure	Artificially Exposure		
			SO <sub>2</sub>	Flyash	SO <sub>2</sub> + Flyash
Medicago sativa	Adaxial	+14.47	+19.72	+5.26	+17.10
	Abaxial	+10.34	+15.13	+2.72	+12.39
Triticum aestivum	Adaxial	+11.32	-15.09	+7.74	+13.96
	Abaxial	+ 7.30	+10.12	+0.89	+ 9.34
Zea mays	Adaxial	+ 9.26	+13.33	+5.19	+10.00
	Abaxial	+10.12	+13.24	+3.21	+12.41

Table 3.27: Diffusive resistance ( $\text{cm sec}^{-1}$ ) of adaxial and abaxial leaf surface of Medicago sativa, Triticum aestivum and Zea mays plants exposed for 90 days (FF and AA; CF and CA; FC and AC) in field and artificially to  $\text{SO}_2$ , flyash and a combination of  $\text{SO}_2$  and flyash.

Exposure pattern	Species	Leaf Surface	Field Exposure	Artificial Exposure			Control
				$\text{SO}_2$	Flyash	$\text{SO}_2$ + Flyash	
FF and AA	Medicago sativa	Adaxial	1.03 $\pm$ 0.14	1.01 $\pm$ 0.12	0.96 $\pm$ 0.12	1.00 $\pm$ 0.15	0.81 $\pm$ 0.13
		Abaxial	1.046 $\pm$ 0.174	1.074 $\pm$ 0.193	0.985 $\pm$ 0.164	1.084 $\pm$ 0.207	0.900 $\pm$ 0.154
	Triticum aestivum	Adaxial	7.13 $\pm$ 1.41	7.34 $\pm$ 1.48	6.86 $\pm$ 1.33	7.62 $\pm$ 1.36	6.22 $\pm$ 1.28
		Abaxial	9.54 $\pm$ 1.83	9.97 $\pm$ 2.16	9.13 $\pm$ 1.50	10.20 $\pm$ 2.10	8.77 $\pm$ 1.63
	Zea mays	Adaxial	3.84 $\pm$ 0.58	3.81 $\pm$ 0.67	3.71 $\pm$ 0.64	3.96 $\pm$ 0.56	3.40 $\pm$ 0.51
		Abaxial	2.62 $\pm$ 0.43	2.67 $\pm$ 0.37	2.49 $\pm$ 0.35	2.75 $\pm$ 0.49	2.32 $\pm$ 0.39
CF and FF	Medicago sativa	Adaxial	0.95 $\pm$ 0.16	0.919 $\pm$ 0.14	0.899 $\pm$ 0.15	1.01 $\pm$ 0.17	0.81 $\pm$ 0.13
		Abaxial	1.007 $\pm$ 0.133	1.003 $\pm$ 0.162	0.926 $\pm$ 0.121	1.02 $\pm$ 0.213	0.900 $\pm$ 0.154
	Triticum aestivum	Adaxial	6.83 $\pm$ 1.36	6.78 $\pm$ 1.51	6.58 $\pm$ 1.63	7.04 $\pm$ 1.49	6.2 $\pm$ 1.28
		Abaxial	9.35 $\pm$ 1.42	9.43 $\pm$ 1.68	8.93 $\pm$ 2.01	9.52 $\pm$ 2.23	8.77 $\pm$ 1.63
	Zea mays	Adaxial	3.72 $\pm$ 0.62	3.68 $\pm$ 0.73	3.62 $\pm$ 0.59	3.80 $\pm$ 0.69	3.4 $\pm$ 0.51
		Abaxial	2.55 $\pm$ 0.34	2.56 $\pm$ 0.36	2.38 $\pm$ 0.36	2.62 $\pm$ 0.42	2.32 $\pm$ 0.39
FC and AC	Medicago sativa	Adaxial	0.844 $\pm$ 0.15	0.840 $\pm$ 0.14	0.828 $\pm$ 0.16	0.849 $\pm$ 0.19	0.81 $\pm$ 0.13
		Abaxial	0.919 $\pm$ 0.173	0.922 $\pm$ 0.187	0.917 $\pm$ 0.134	0.926 $\pm$ 0.218	0.900 $\pm$ 0.154
	Triticum aestivum	Adaxial	6.31 $\pm$ 1.39	6.34 $\pm$ 1.51	6.29 $\pm$ 1.46	6.44 $\pm$ 1.46	6.2 $\pm$ 1.28
		Abaxial	8.83 $\pm$ 1.74	8.80 $\pm$ 1.94	8.87 $\pm$ 2.34	8.81 $\pm$ 1.49	8.77 $\pm$ 1.63
	Zea mays	Adaxial	3.45 $\pm$ 0.62	3.45 $\pm$ 0.59	3.48 $\pm$ 0.67	3.47 $\pm$ 0.57	3.40 $\pm$ 0.51
		Abaxial	2.38 $\pm$ 0.35	2.36 $\pm$ 0.46	2.35 $\pm$ 0.41	2.36 $\pm$ 0.46	2.32 $\pm$ 0.39

Table 3.28: Change in diffusive resistance of adaxial and abaxial leaf surfaces of Medicago sativa, Triticum aestivum and Zea mays plants exposed for 90 days (FF and AA; CF and CA; FC and AC) in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash (Expressed as % change over Control)

Exposure pattern	Species	Leaf Surface	Field Exposure	Artificial Exposure		
				SO <sub>2</sub>	Flyash	SO <sub>2</sub> + flyash
FF and AA	Medicago sativa	Adaxial	+27.17	+24.69	+18.52	+32.81
		Abaxial	+16.21	+19.32	+ 9.34	+20.47
	Triticum aestivum	Adaxial	+14.52	+17.72	+ 9.68	+22.58
		Abaxial	+ 8.65	+13.74	+ 4.12	+16.23
	Zea mays	Adaxial	+12.94	+12.06	+ 9.12	+16.47
		Abaxial	+13.13	+15.46	+ 7.25	+18.92
CF and CA	Medicago sativa	Adaxial	+17.28	+13.58	+11.11	+24.69
		Abaxial	+11.94	+11.44	+ 2.90	+13.29
	Triticum aestivum	Adaxial	+10.16	+ 9.35	+ 6.13	+13.54
		Abaxial	+ 6.48	+ 7.42	+ 1.8	+ 8.38
	Zea mays	Adaxial	+ 9.41	+ 8.24	+ 6.47	+11.76
		Abaxial	+10.14	+10.17	+ 2.73	+12.21
FC and AC	Medicago sativa	Adaxial	+ 4.17	+ 3.76	+ 2.2	+ 4.88
		Abaxial	+ 2.1	+ 2.40	+ 1.9	+ 2.9
	Triticum aestivum	Adaxial	+ 1.78	+ 2.26	+ 1.45	+ 3.55
		Abaxial	+ 0.70	+ 1.10	+ 0.40	+ 1.30
	Zea mays	Adaxial	+ 1.5	+ 1.6	+ 1.35	+ 1.9
		Abaxial	+ 1.91	+ 1.82	+ 1.43	+ 2.44

Table 3.29: Leaf surface temperature of Medicago sativa, Triticum aestivum and Zea mays plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash for 45 days (F and A) and 90 days (FF and AA)

Exposure pattern	Species	Field Exposure	Artificial Exposure			Control
			SO <sub>2</sub>	Flyash	SO <sub>2</sub> + flyash	
F and A	Medicago sativa	29.92	29.35	30.99	29.56	30.38
	Triticum aestivum	28.61	28.34	29.38	28.45	28.96
	Zea mays	29.52	29.53	30.16	29.34	29.75
FF and AA	Medicago sativa	29.72	29.05	31.48	29.53	30.46
	Triticum aestivum	28.47	28.04	29.59	28.26	28.99
	Zea mays	29.45	29.34	30.28	29.37	29.78



Table 3.30: Change in leaf surface temperature of Medicago sativa, Triticum aestivum and Zea mays plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash for 45 days (F and A) and 90 days (FF and AA) (Expressed as % change over Control)

Exposure pattern	Species	Field Exposure	Artificial Exposure		
			SO <sub>2</sub>	Flyash	SO <sub>2</sub> + flyash
F and A	Medicago sativa	-0.45	-1.03	+0.61	-0.82
	Triticum aestivum	-0.35	-0.62	+0.42	-0.51
	Zea mays	-0.23	-0.22	+0.41	-0.41
FF and AA	Medicago sativa	-0.74	-1.41	+0.82	-0.93
	Triticum aestivum	-0.52	-0.95	+0.60	-0.73
	Zea mays	-0.33	-0.44	+0.50	-0.41

Table 3.31: Percentage absorbance at wavelengths representing peak points of absorption spectra in the visible and infrared regions of intact leaf of Medicago sativa, Triticum aestivum and Zea mays plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash, for 45 days (F and A) and 90 days (FF and AA)

Species	F and A									
	Visible					Infrared				
	Field	SO <sub>2</sub>	Flyash	SO <sub>2</sub> + flyash	Control	Field	SO <sub>2</sub>	Flyash	SO <sub>2</sub> + flyash	Control
Medicago sativa	83.02	78.12	87.06	78.22	84.22	86.51	76.01	90.00	48.79	88.52
Triticum aestivum	75.00	69.92	79.20	71.56	78.22	79.96	79.11	74.88	80.05	79.96
Zea mays	82.01	81.80	75.73	83.63	82.01	82.82	82.62	79.20	83.71	82.82
	FF and AA									
Medicago sativa	83.78	75.73	88.52	80.50	87.12	86.88	71.49	88.17	84.51	87.00
Triticum aestivum	76.23	64.52	81.84	70.01	79.58	82.22	68.38	86.35	81.89	82.62
Zea mays	83.21	76.83	86.76	82.01	85.21	79.82	76.67	84.69	79.58	83.48

Table 3.32: Change in percentage absorbance at wavelengths representing peak points of absorption spectra in visible and infrared regions of intact leaf of Medicago sativa, Triticum aestivum and Zea mays plants exposed for 45 days (F and A) and 90 days (FF and AA) in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash (Expressed as % change over Control)

Species	F and A							
	Visible				Infrared			
	Field	SO <sub>2</sub>	Flyash	SO <sub>2</sub> + flyash	Field	SO <sub>2</sub>	Flyash	SO <sub>2</sub> + flyash
Medicago sativa	-1.20	-6.10	+2.84	-6.00	-2.01	-12.51	+1.48	-3.73
Triticum aestivum	-3.22	-8.30	+0.98	-6.66	-0.85	- 5.08	+0.09	-8.14
Zea mays	-0.21	-6.28	+1.62	-5.45	-0.20	- 3.62	+0.89	-2.68
	FF and AA							
Medicago sativa	-3.44	-11.39	+1.40	-6.62	-0.34	-15.51	+1.17	-2.49
Triticum aestivum	-3.35	-15.06	+2.26	-9.51	-0.40	-14.34	+3.73	-0.73
Zea mays	-2.00	-8.38	+1.55	-3.20	-3.66	- 6.85	+1.21	-3.90

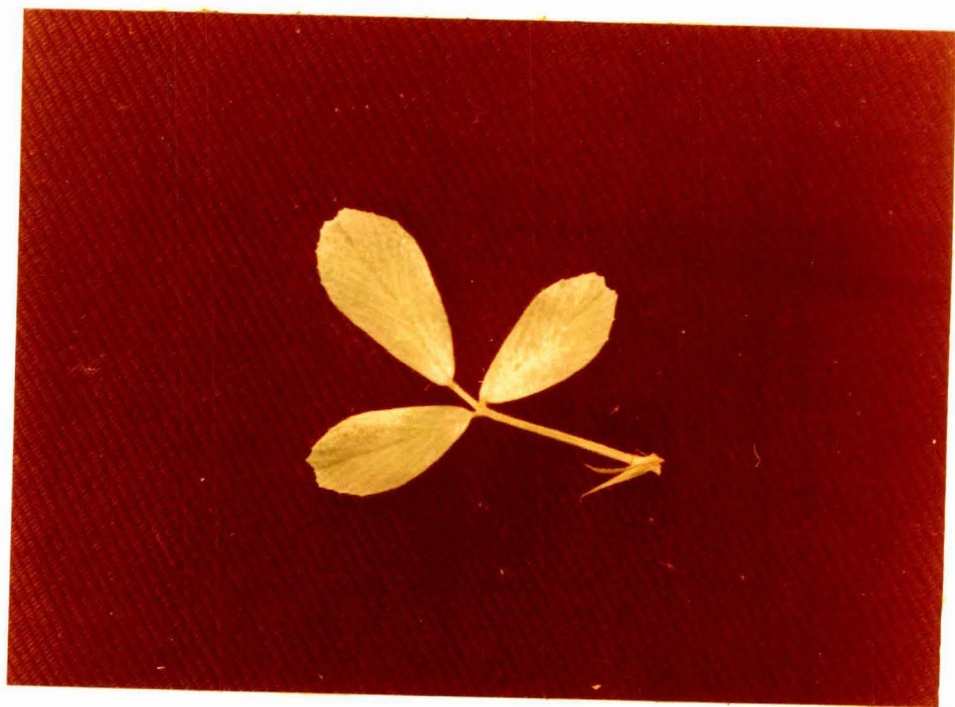
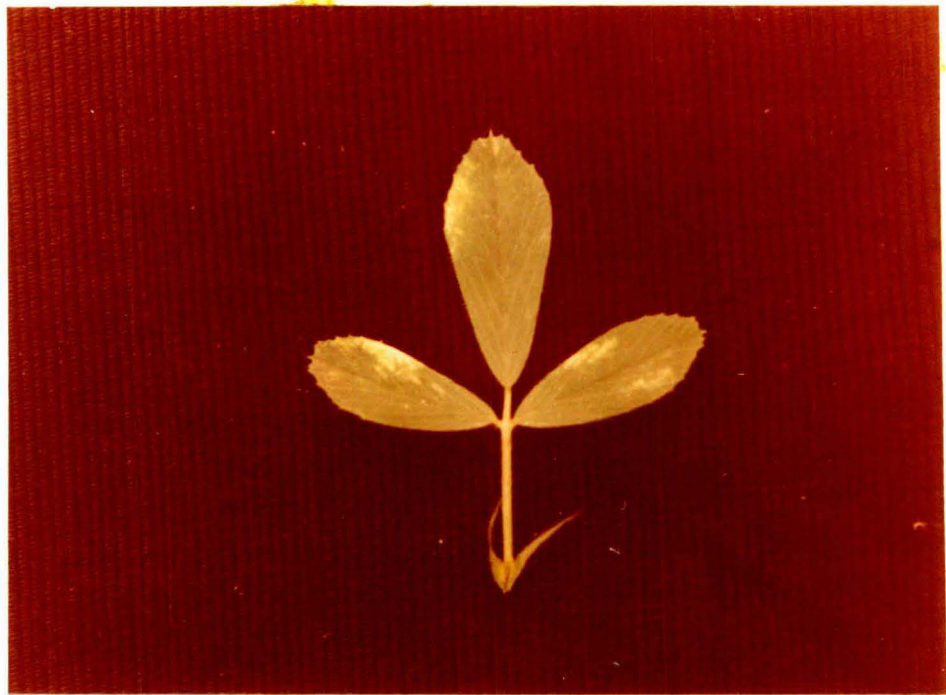
Fig. 3.1 Chlorotic spots on the leaf of Medicago sativa exposed for 45 days near IP Power Plant.

Fig. 3.2 Chlorotic spots on the leaves of Medicago sativa exposed for 90 days near IP Power plant.



Fig. 3.3 Chlorotic regions on margin and tip of leaf of Medicago sativa exposed for 45 days to SO<sub>2</sub> artificially.

Fig. 4.4 Chlorotic regions on margin and tip of leaf of Medicago sativa exposed for 90 days to SO<sub>2</sub> artificially.



**Fig. 3.5** Irregular chlorotic spots on the leaf of Medicago sativa exposed for 45 days to SO<sub>2</sub> and flyash artificially.

**Fig. 3.6** Irregular chlorotic spots on the leaf of Medicago sativa exposed for 90 days to SO<sub>2</sub> and flyash artificially.





Fig. 3.7 Scanning electron micrograph of the adaxial leaf surface of Medicago sativa kept as control for 90 days (X450)

Fig. 3.8 Scanning electron micrograph of the adaxial leaf surface of Medicago sativa exposed in field for 90 days near IP Power Plant (X450).

Fig. 3.9 Scanning electron micrograph of the adaxial leaf surface of Medicago sativa exposed artificially to a combination of SO<sub>2</sub> and flyash for 90 days (X450).

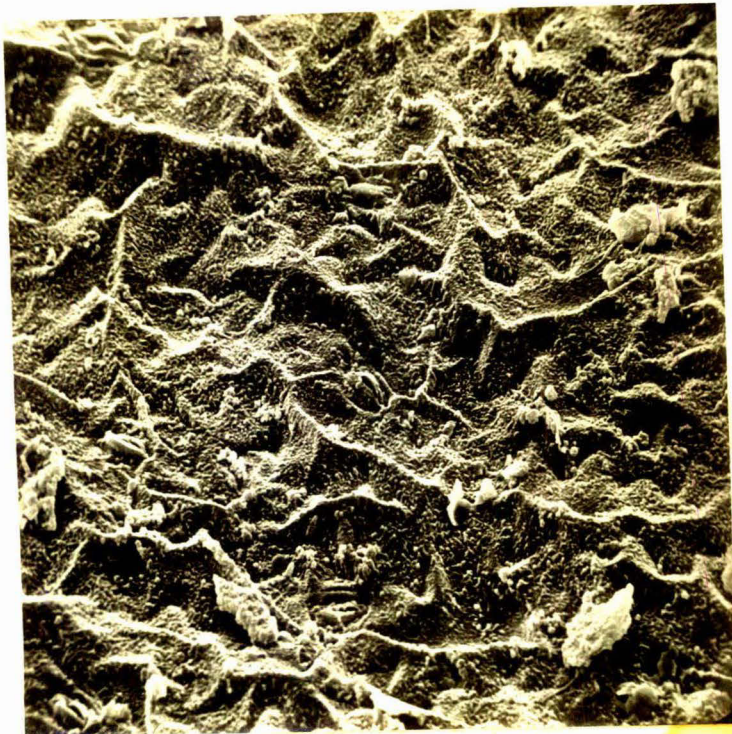
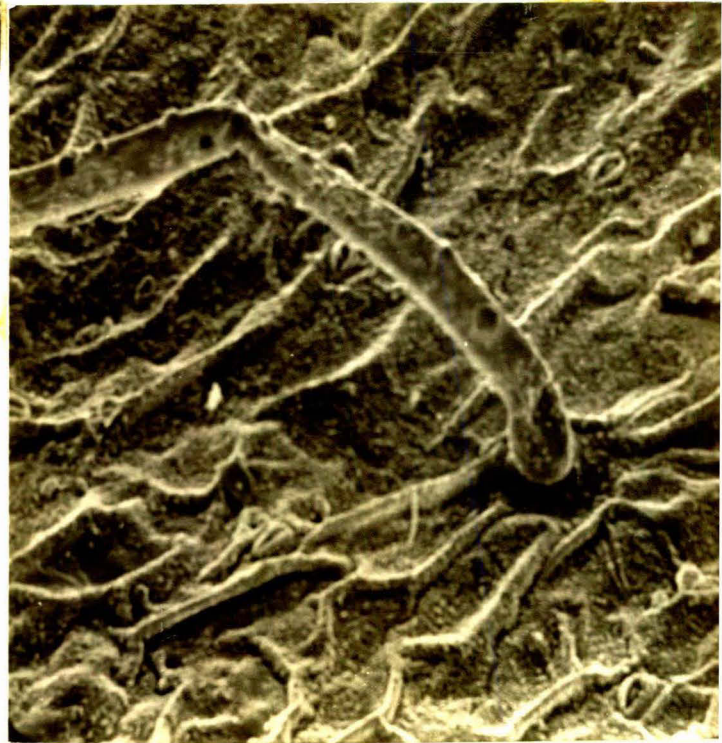
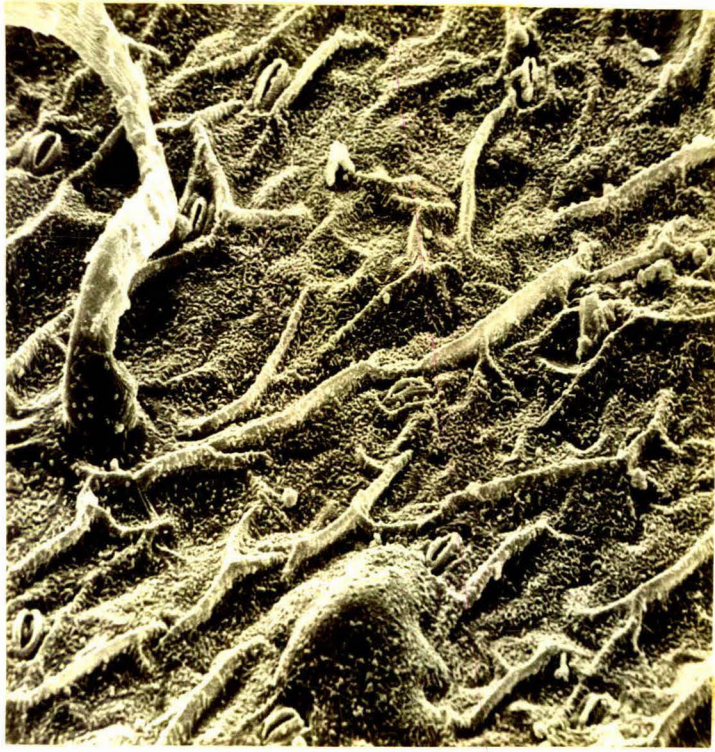


Fig. 3.10 Scanning electron micrograph of the abaxial leaf surface of Medicago sativa kept as control for 90 days (X450)

Fig. 3.11 Scanning electron micrograph of the abaxial leaf surface of Medicago sativa exposed in field near IP Power Plant for 90 days (X450)

Fig. 3.12 Scanning electron micrograph of the abaxial leaf surface of Medicago sativa exposed artificially to a combination of SO<sub>2</sub> and flyash for 90 days (X200).

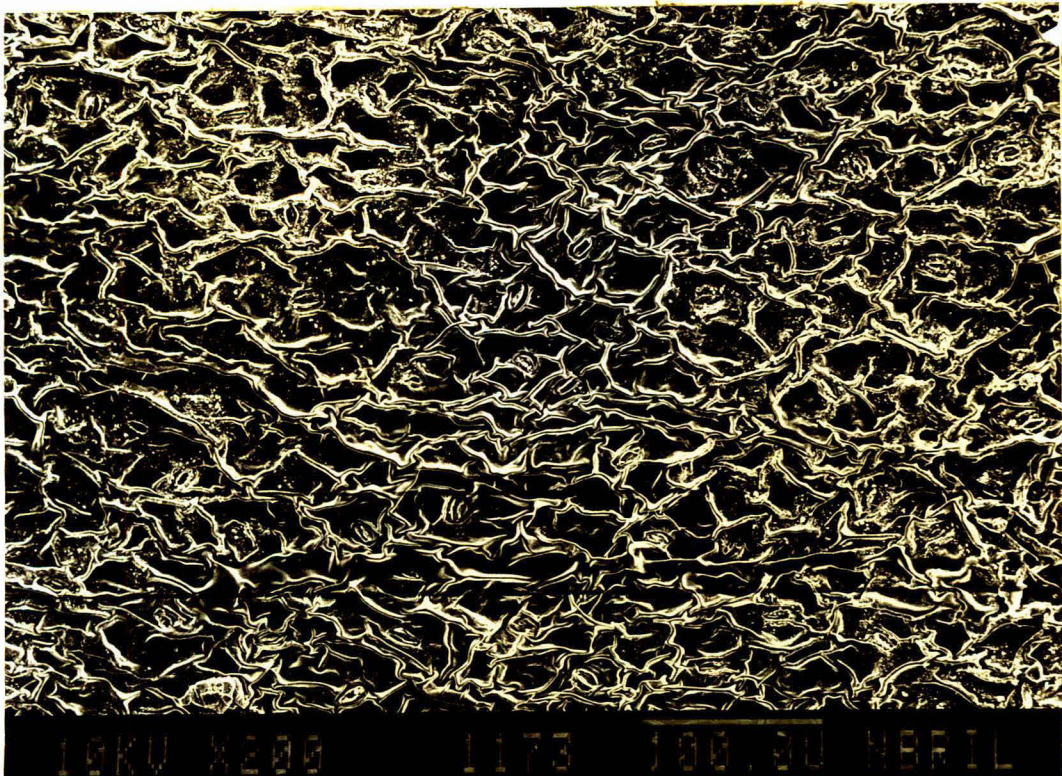
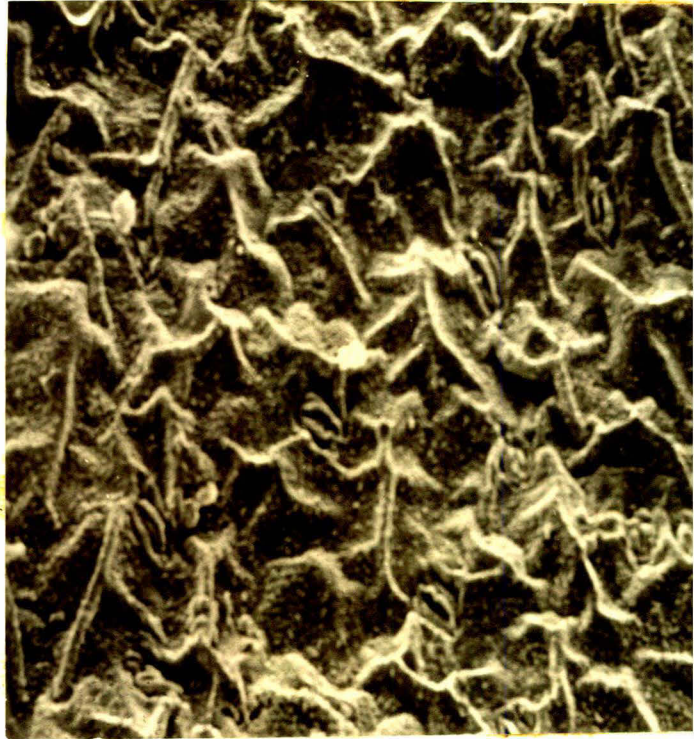
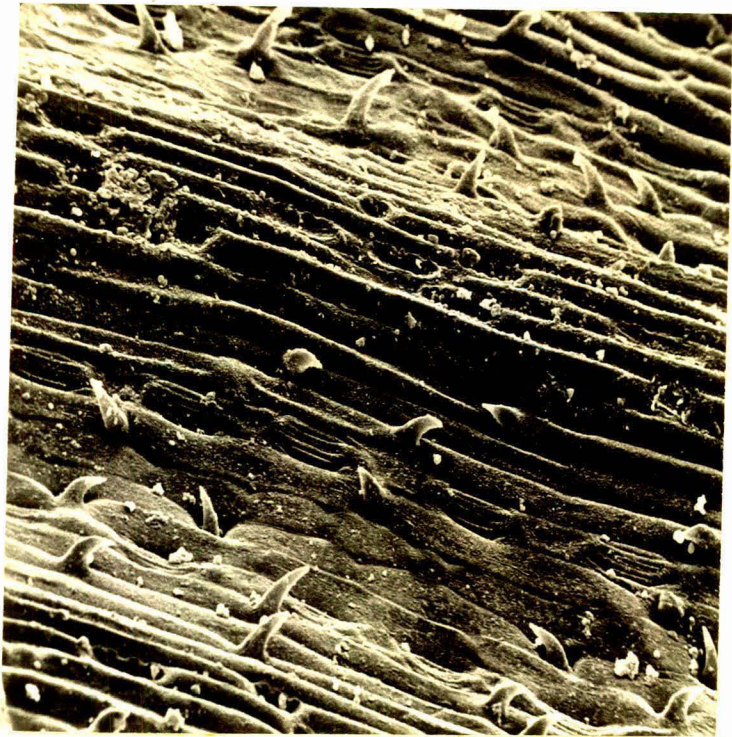


Fig. 3.13 Scanning electron micrograph of the adaxial leaf surface of Triticum aestivum kept as control for 90 days (X189).

Fig. 3.14 Scanning electron micrograph of the adaxial leaf surface of Triticum aestivum exposed in field near IP Power Plant for 90 days (X189)



**Fig. 3.15** Scanning electron micrograph of the adaxial leaf surface of Triticum aestivum exposed artificially to  $\text{SO}_2$  for 90 days (X216).

**Fig. 3.16** Scanning electron micrograph of the adaxial leaf surface of Triticum aestivum exposed artificially to flyash for 90 days (X216).

**Fig. 3.17** Scanning electron micrograph of the adaxial leaf surface of Triticum aestivum exposed artificially to a combination of  $\text{SO}_2$  and flyash for 90 days (X189).



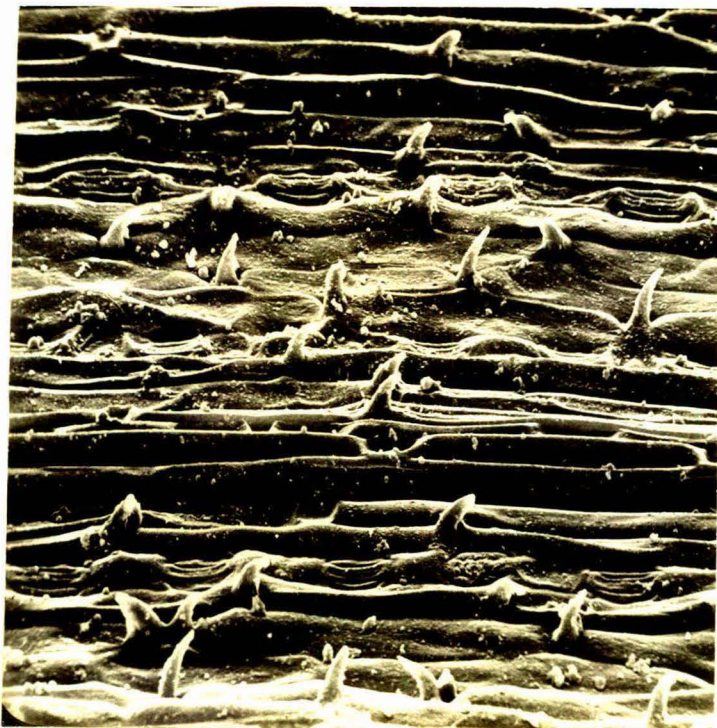
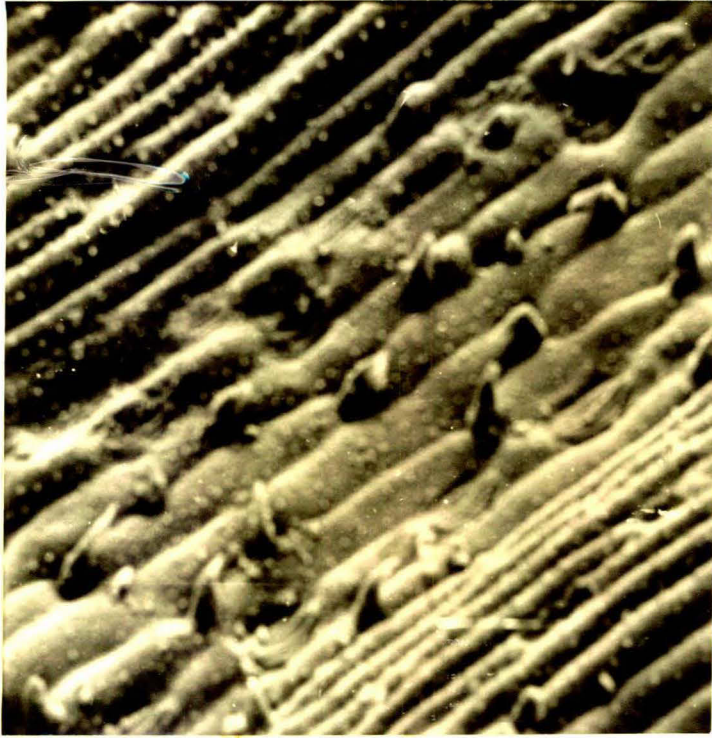


Fig. 3.18 Scanning electron micrograph of the adaxial leaf surface of Zea mays kept as control for 90 days (X180)

Fig. 3.19 Scanning electron micrograph of the adaxial leaf surface of Zea mays exposed in field near IP Power Plant for 90 days (X198)

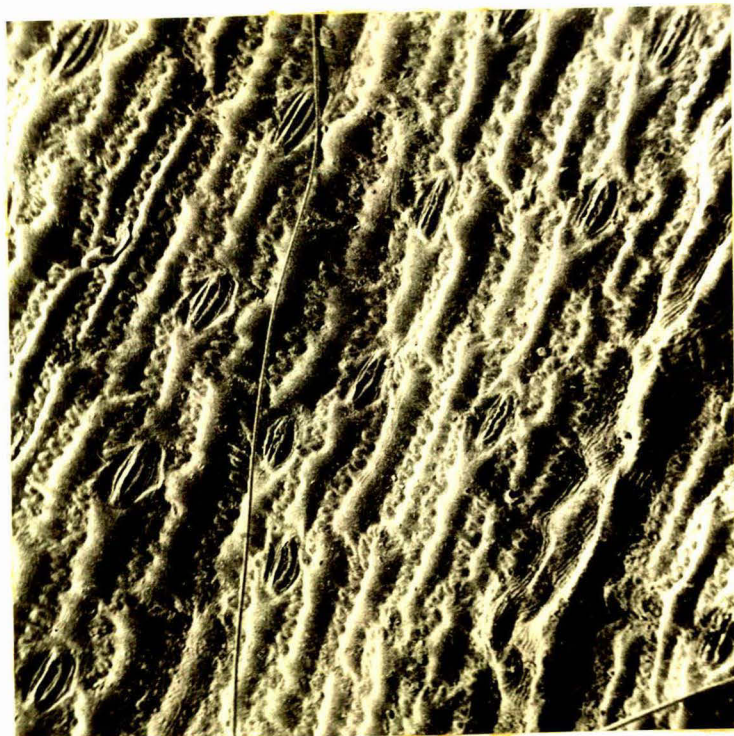
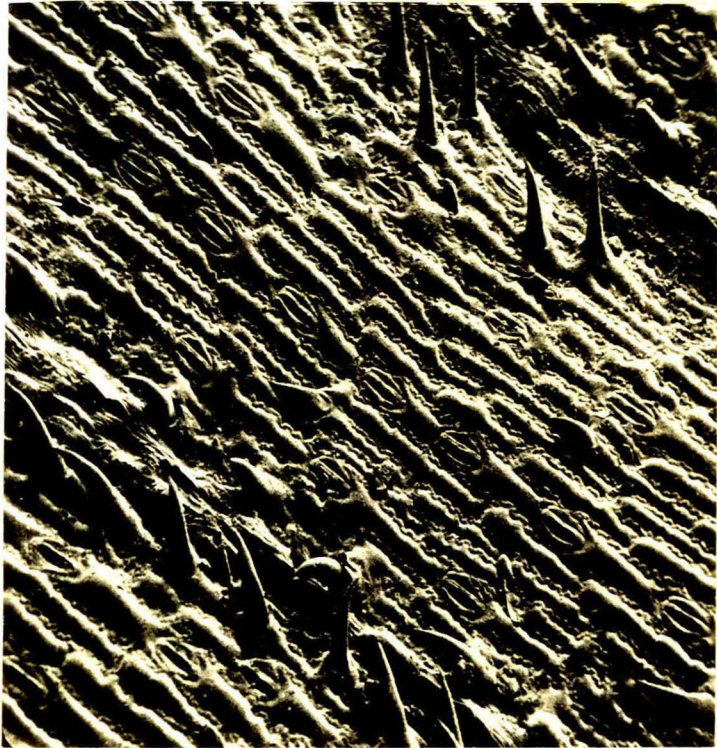


Fig. 3.20 Scanning electron micrograph of the adaxial leaf surface of Zea mays exposed artificially to  $\text{SO}_2$  for 90 days (X180)

Fig. 3.21 Scanning electron micrograph of the adaxial leaf surface of Zea mays exposed artificially to a combination of  $\text{SO}_2$  and flyash for 90 days (X198)

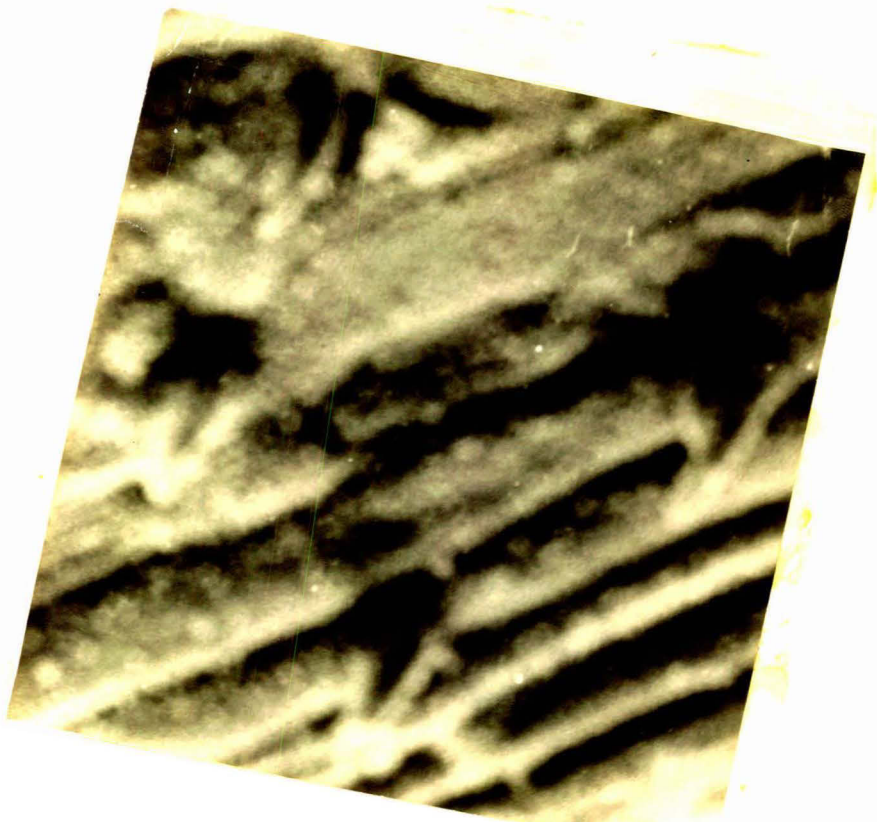


Fig. 3.22 Scanning electron micrograph of the abaxial leaf surface of Zea mays kept as control for 90 days (X198)

Fig. 3.23 Scanning electron micrograph of the abaxial leaf surface of Zea mays exposed in field near IP Power Plant for 90 days (X200)

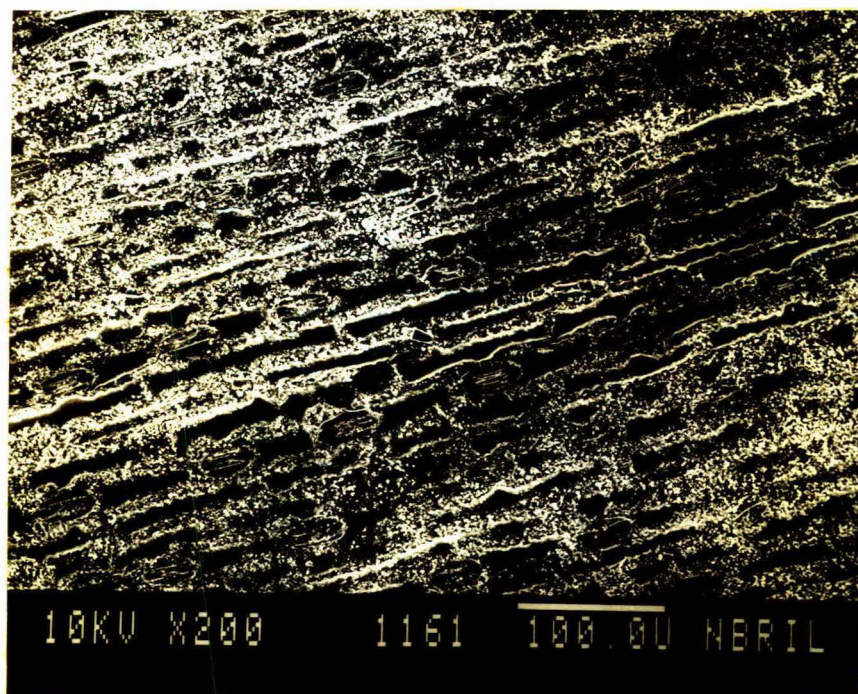


Fig. 3.24 Scanning electron micrograph of the abaxial leaf surface of Zea mays exposed artificially to SO<sub>2</sub> for 90 days (X180)

Fig. 3.25 Scanning electron micrograph of the abaxial leaf surface of Zea mays exposed artificially to flyash for 90 days (X200)

Fig. 3.26 Scanning electron micrograph of the abaxial leaf surface of Zea mays exposed artificially to a combination of SO<sub>2</sub> and flyash for 90 days (X189)



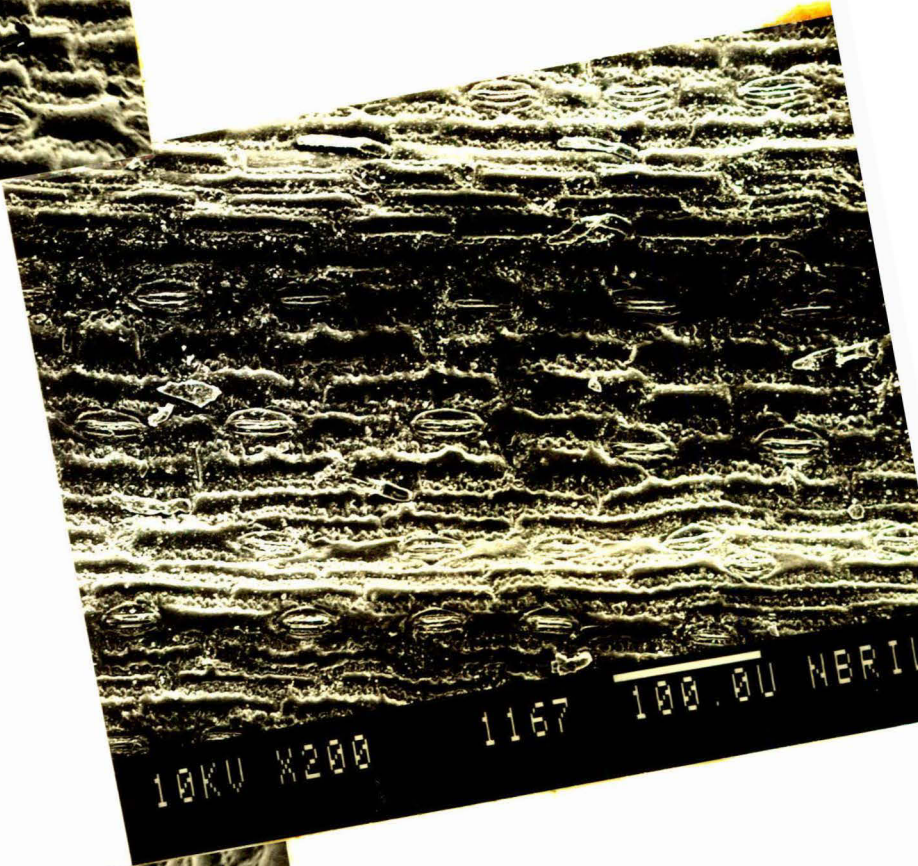


Fig. 3.27 Scanning electron micrograph of stomata on the adaxial leaf surface of Medicago sativa kept as control for 90 days (X1980)

Fig. 3.28 Scanning electron micrograph of stomata on the adaxial leaf surface of Medicago sativa exposed in field near IP Power Plant for 90 days (X1980)

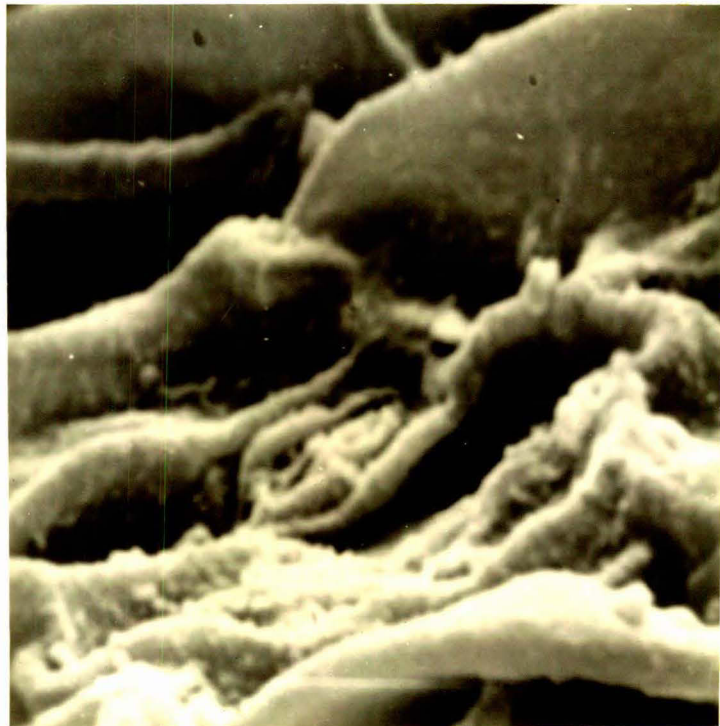
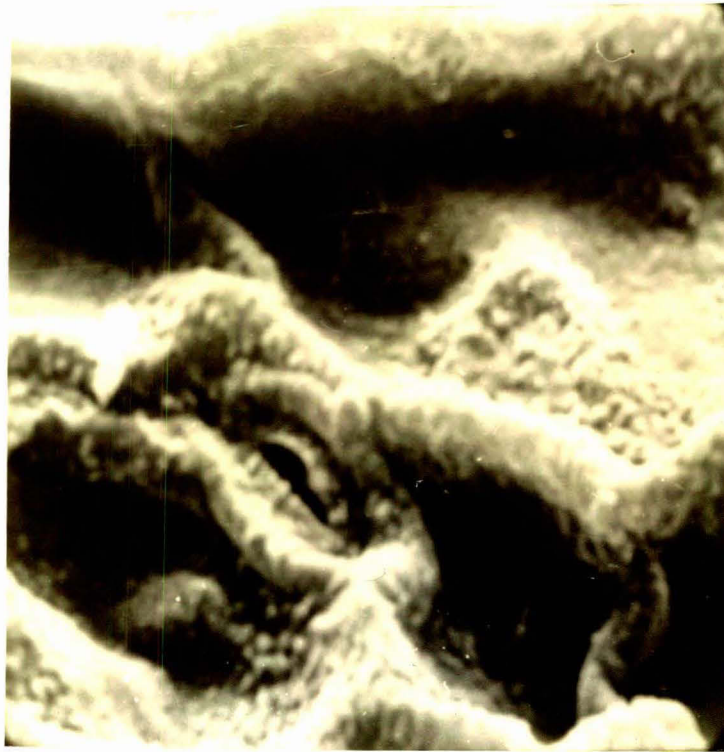


Fig. 3.29 Scanning electron micrograph of stomata on the adaxial leaf surface of Medicago sativa exposed artificially to  $\text{SO}_2$  for 90 days (X1800)

Fig. 3.30 Scanning electron micrograph of stomata on the adaxial leaf surface of Medicago sativa exposed artificially to flyash for 90 days (X1800)

Fig. 3.31 Scanning electron micrograph of stomata on the adaxial leaf surface of Medicago sativa exposed artificially to a combination of  $\text{SO}_2$  and flyash for 90 days (X1980)

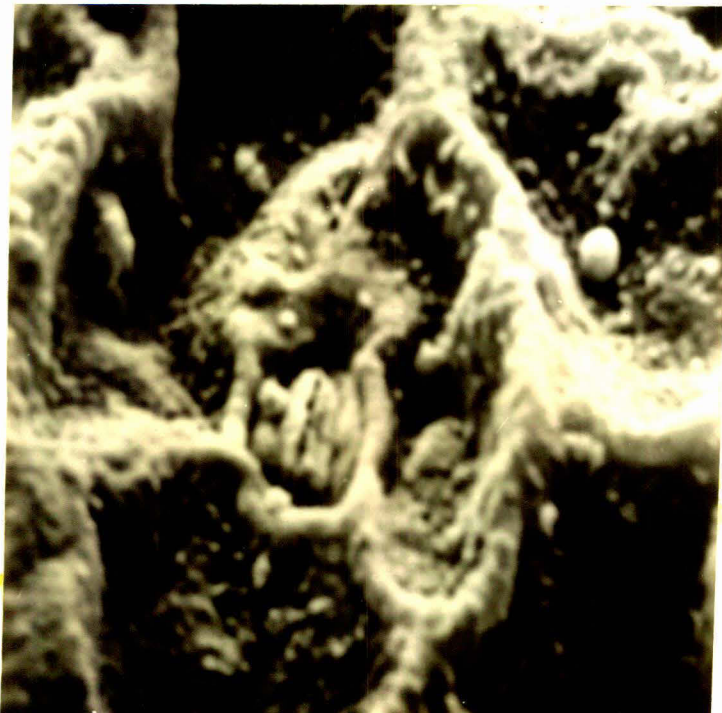


Fig. 3.32 Scanning electron micrograph of stomata on the abaxial leaf surface of Medicago sativa kept as control for 90 days (X1980)

Fig. 3.33 Scanning electron micrograph of stomata on the abaxial leaf surface of Medicago sativa exposed in field near IP Power Plant for 90 days (X1980)

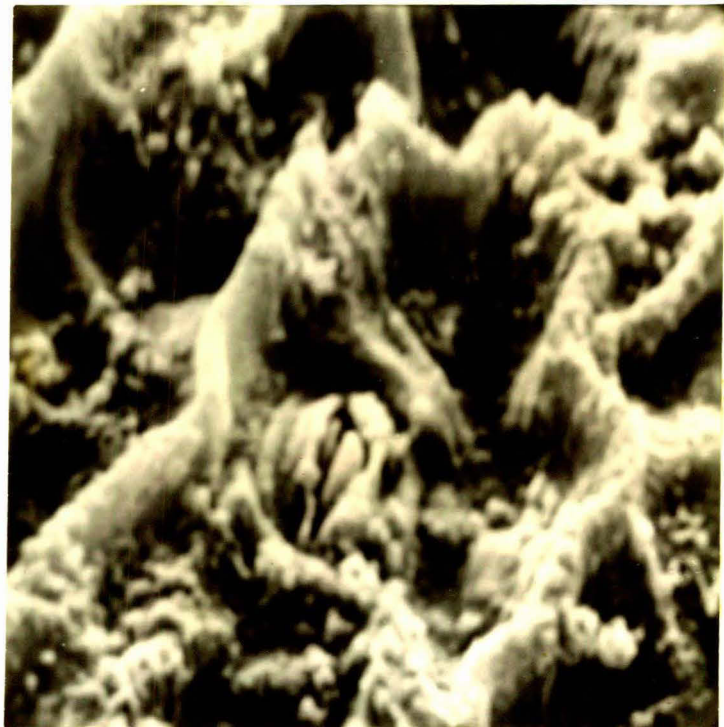


Fig. 3.34 Scanning electron micrograph of stomata on the abaxial leaf surface of Medicago sativa exposed artificially to  $\text{SO}_2$  for 90 days (X1980)

Fig. 3.35 Scanning electron micrograph of stomata on the abaxial leaf surface of Medicago sativa exposed artificially to flyash for 90 days (X1800)

Fig. 3.36 Scanning electron micrograph of stomata on the abaxial leaf surface of Medicago sativa exposed artificially to a combination of  $\text{SO}_2$  and flyash for 90 days (X1980)



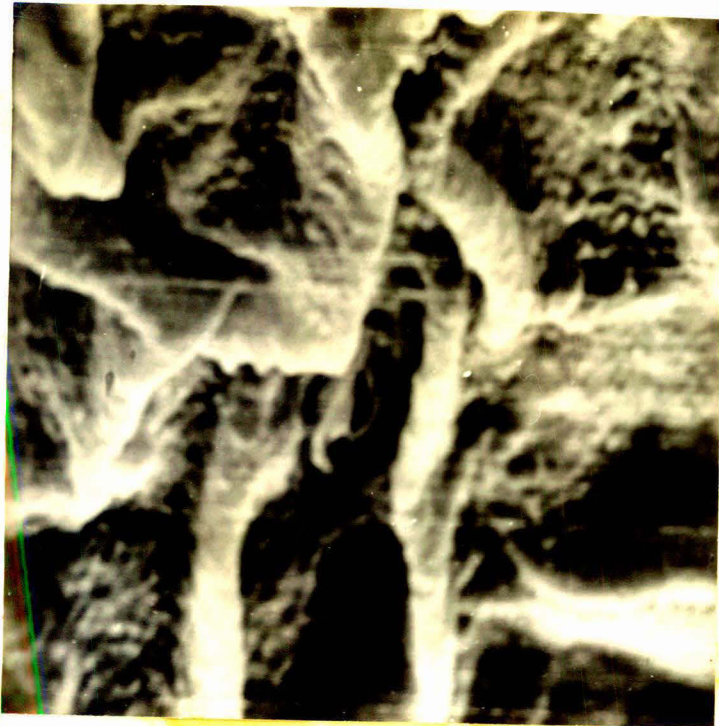
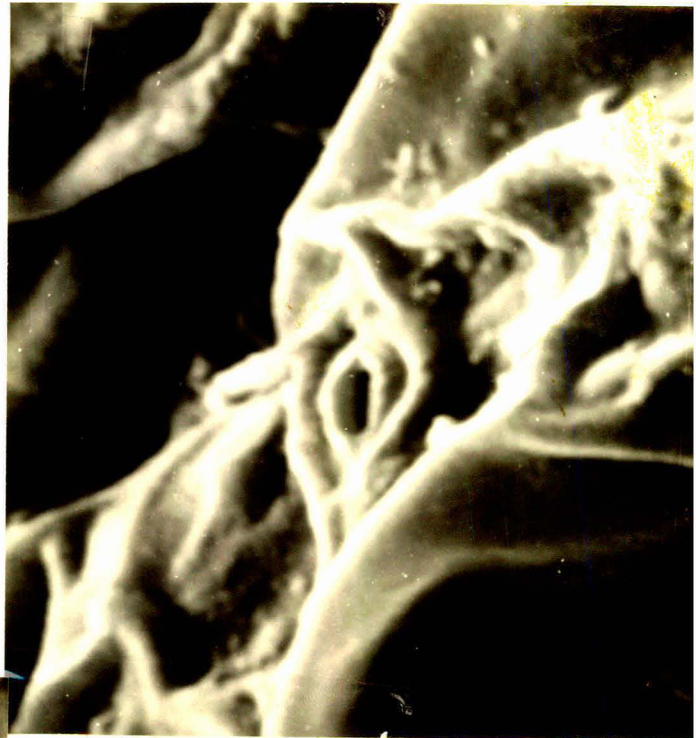
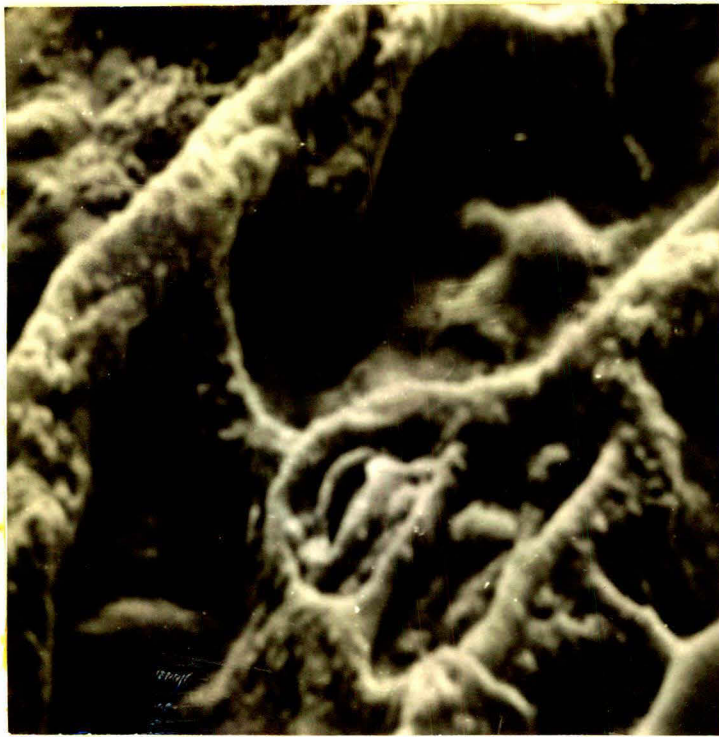


Fig. 3.37 Scanning electron micrograph of stomata on the adaxial leaf surface of Triticum aestivum kept as control for 90 days (X1280)

Fig. 3.38 Scanning electron micrograph of stomata on the adaxial leaf surface of Triticum aestivum exposed in field near IP Power Plant for 90 days (X990)



Fig. 3.39 Scanning electron micrograph of stomata on the adaxial leaf surface of Triticum aestivum exposed artificially to SO<sub>2</sub> for 90 days (X1010)

Fig. 3.40 Scanning electron micrograph of stomata on the adaxial leaf surface of Triticum aestivum exposed artificially to flyash for 90 days (X1090)

Fig. 3.41 Scanning electron micrograph of stomata on the adaxial leaf surface of Triticum aestivum exposed artificially to a combination of SO<sub>2</sub> and flyash for 90 days (X990)

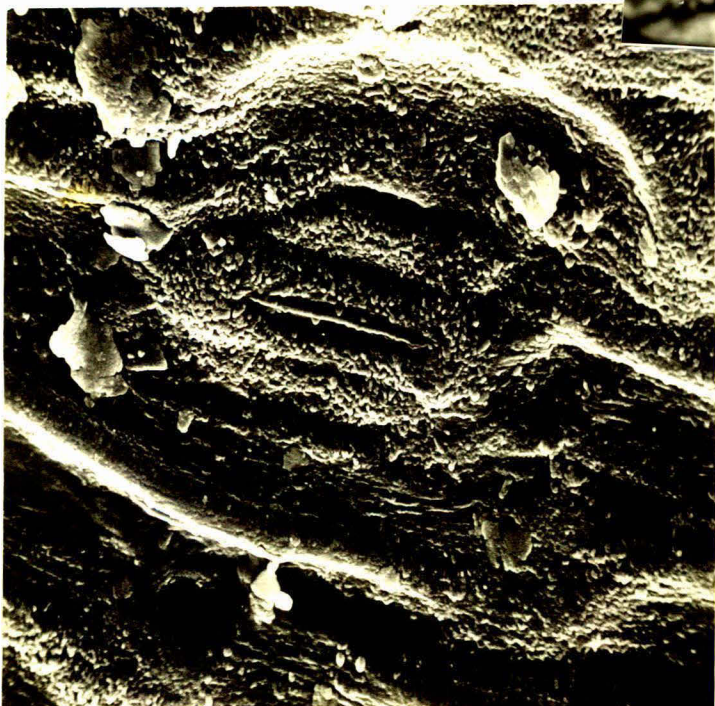


Fig. 3.42 Scanning electron micrograph of stomata on the abaxial leaf surface of Triticum aestivum kept as control for 90 days (X1080)

Fig. 3.43 Scanning electron micrograph of stomata on the abaxial leaf surface of Triticum aestivum exposed in field near IP Power Plant for 90 days (X990)

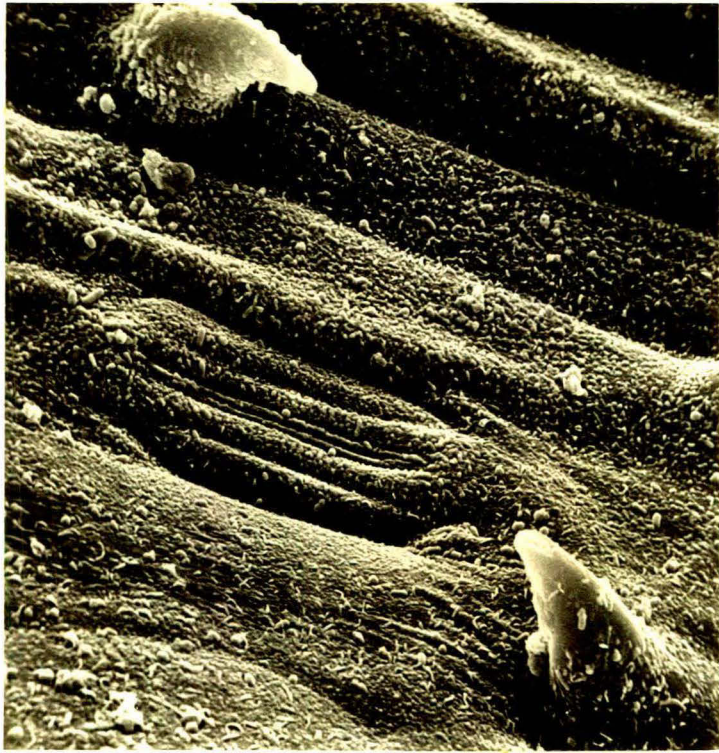


Fig. 3.44 Scanning electron micrograph of stomata on the abaxial leaf surface of Triticum aestivum exposed artificially to SO<sub>2</sub> for 90 days (X1080)

Fig. 3.45 Scanning electron micrograph of stomata on the abaxial leaf surface of Triticum aestivum exposed artificially to a combination of SO<sub>2</sub> and flyash for 90 days (X1080)





Fig. 3.46 Scanning electron micrograph of stomata on the adaxial leaf surface of Zea mays kept as control for 90 days (X1800)

Fig. 3.47 Scanning electron micrograph of stomata on the adaxial leaf surface of Zea mays exposed in field near IP Power Plant for 90 days (X1800)



Fig. 3.48 Scanning electron micrograph of stomata on the adaxial leaf surface of Zea mays exposed artificially to  $\text{SO}_2$  for 90 days (X1890)

Fig. 3.49 Scanning electron micrograph of stomata on the adaxial leaf surface of Zea mays exposed artificially to flyash for 90 days (X1890)

Fig. 3.50 Scanning electron micrograph of stomata on the adaxial leaf surface of Zea mays exposed artificially to a combination of  $\text{SO}_2$  and flyash (X1800)



Fig. 3.51 Scanning electron micrograph of stomata on the abaxial leaf surface of Zea mays kept as control for 90 days (X1800)

Fig. 3.52 Scanning electron micrograph of stomata on the abaxial leaf surface of Zea mays exposed in field near IP Power Plant for 90 days (X2000)

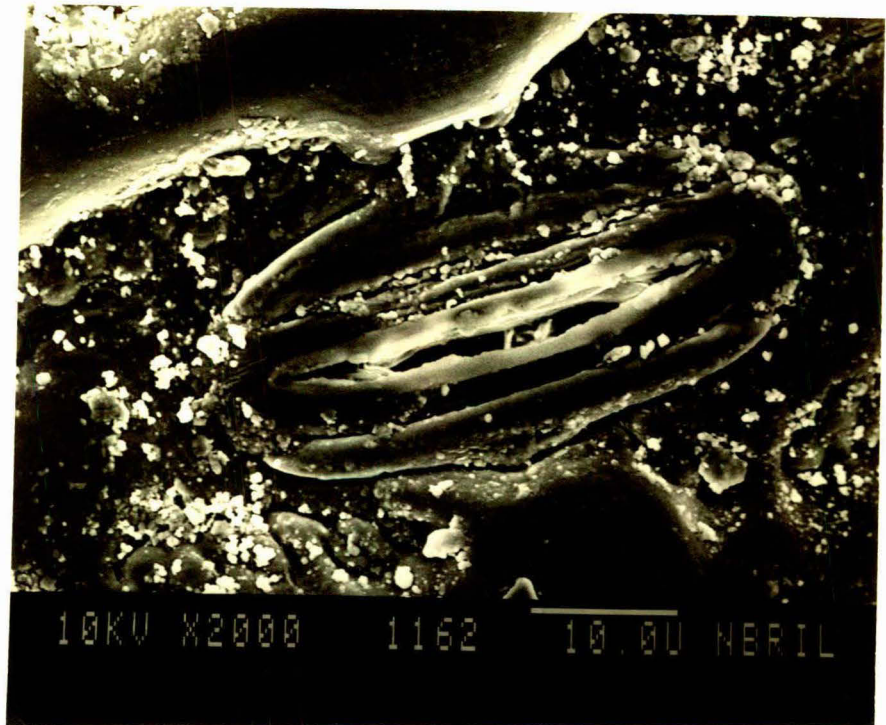
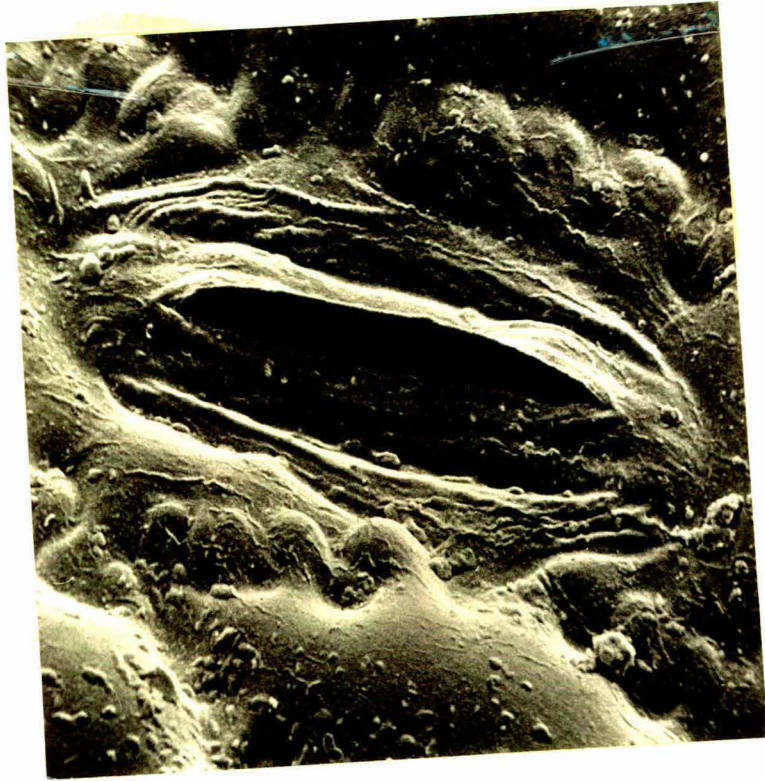


Fig. 3.53 Scanning electron micrograph of stomata on the abaxial leaf surface of Zea mays exposed artificially to  $\text{SO}_2$  for 90 days (X1620)

Fig. 3.54 Scanning electron micrograph of stomata on the abaxial leaf surface of Zea mays exposed artificially to flyash for 90 days (X2000)

Fig. 3.55 Scanning electron micrograph of stomata on the abaxial leaf surface of Zea mays exposed artificially to a combination of  $\text{SO}_2$  and flyash for 90 days (X1890)



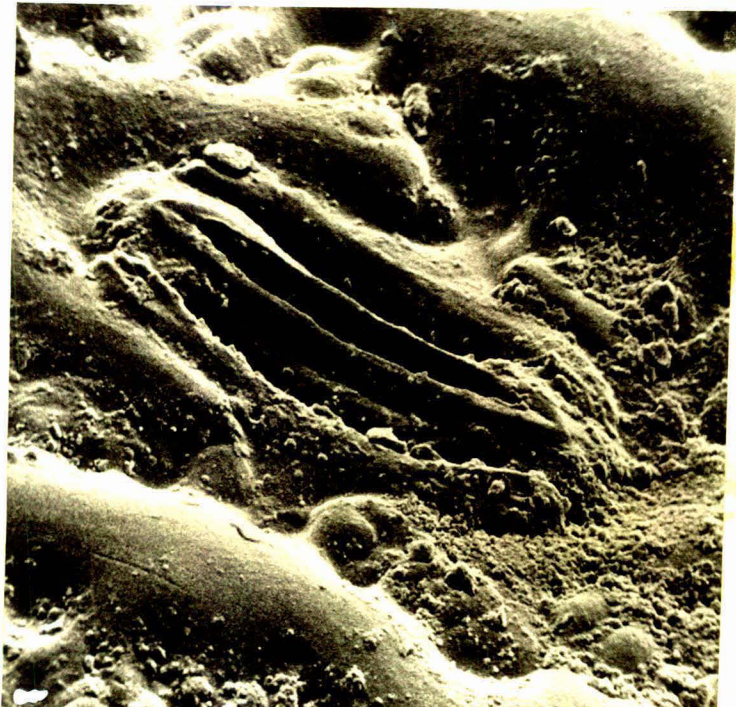
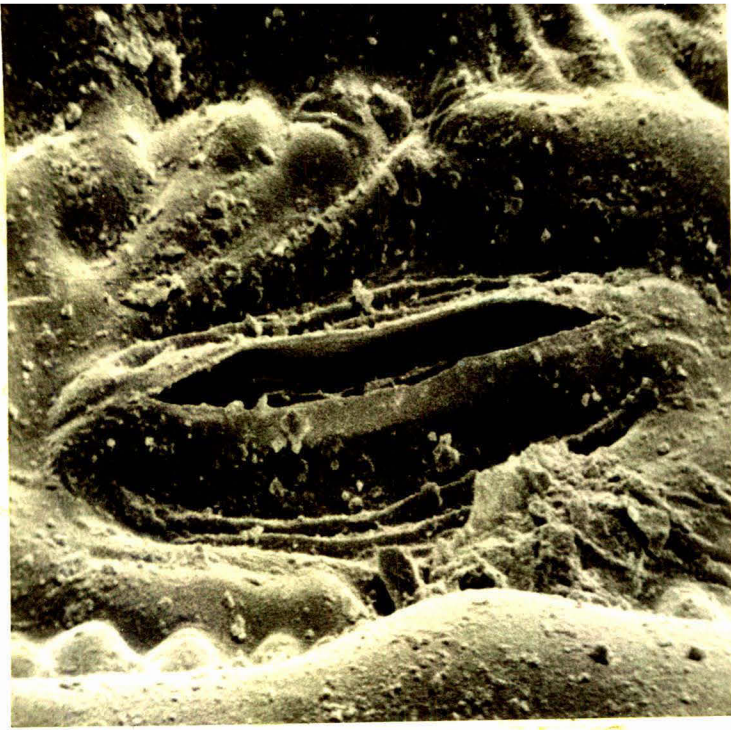


Fig. 3.56 Scanning electron micrograph of the abaxial leaf surface of Triticum aestivum kept as control for 90 days (X202)

Fig. 3.57 Scanning electron micrograph of the abaxial leaf surface of Triticum aestivum exposed in field near IP Power Plant for 90 days (X210)

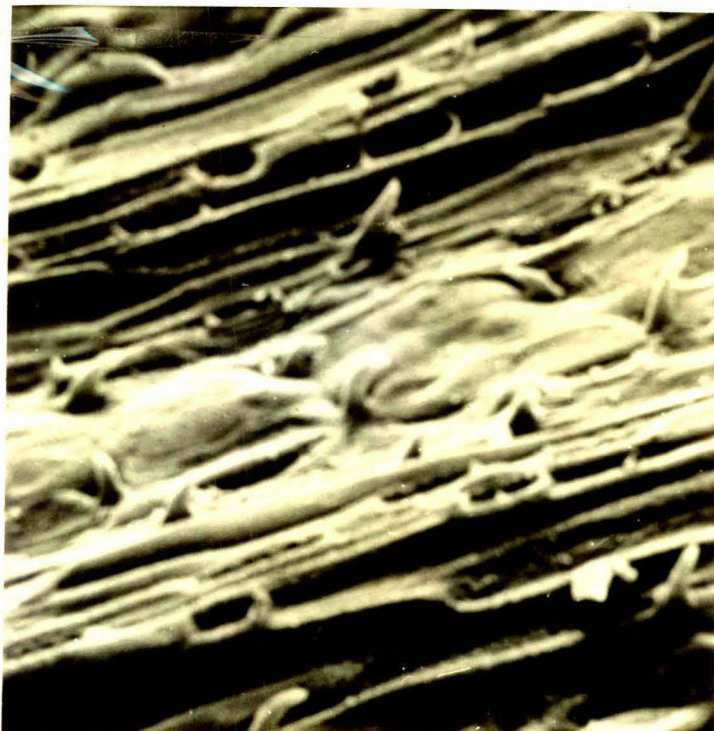


Fig. 3.58 Scanning electron micrograph of the abaxial leaf surface of Triticum aestivum exposed artificially to SO<sub>2</sub> for 90 days (X216)

Fig. 3.59 Scanning electron micrograph of the abaxial leaf surface of Triticum aestivum exposed artificially to flyash for 90 days (X216)

Fig. 3.60 Scanning electron micrograph of the abaxial leaf surface of Triticum aestivum exposed artificially to a combination of SO<sub>2</sub> and flyash for 90 days (X216)

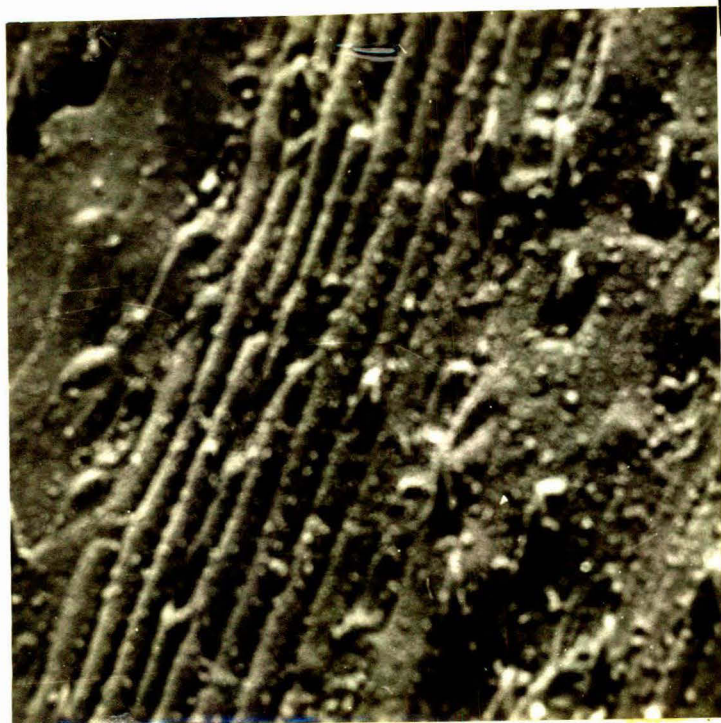
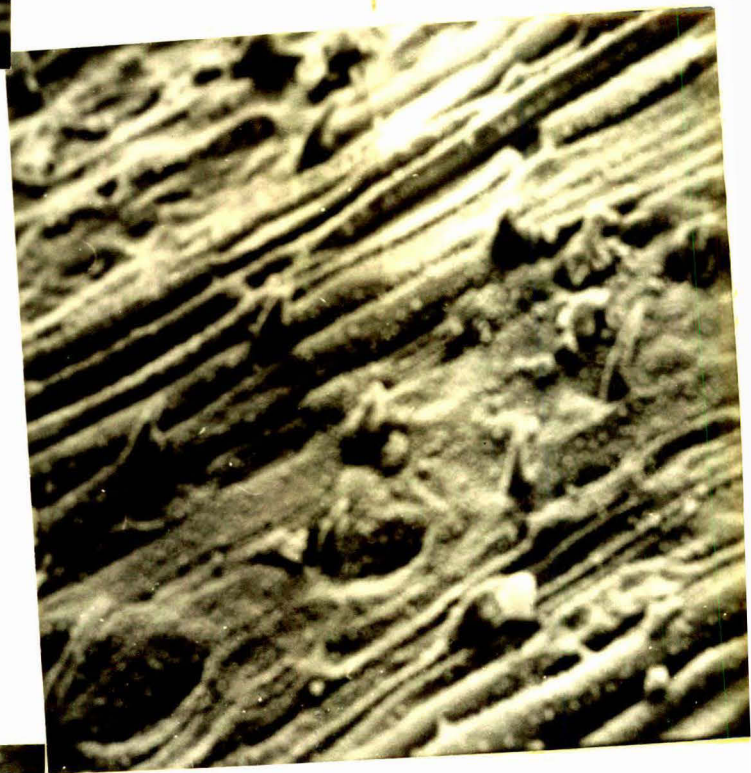
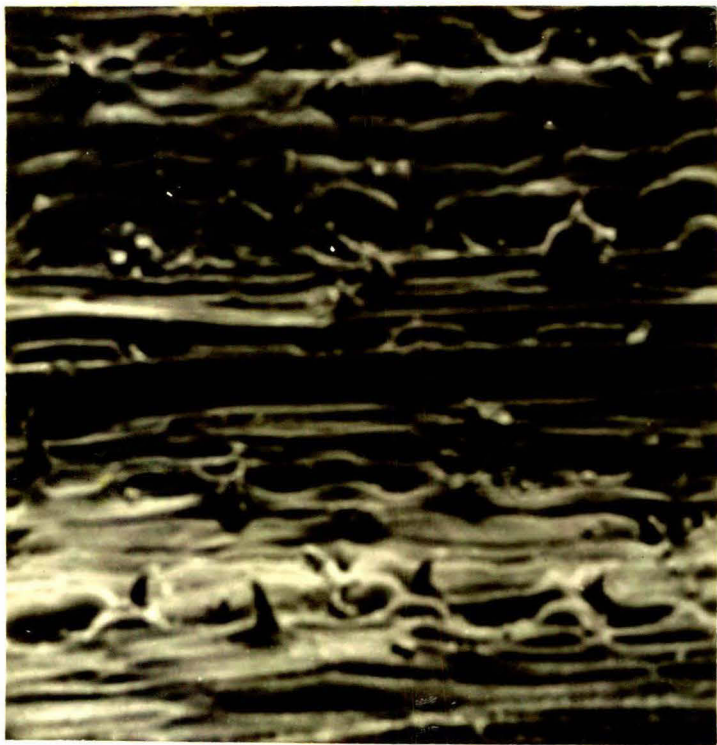


Fig. 3.61 Scanning electron micrograph of trichomes on the adaxial leaf surface of Triticum aestivum kept as control for 90 days (X504)

Fig. 3.62 Scanning electron micrograph of trichomes on the adaxial leaf surface of Triticum aestivum exposed in field near IP Power Plant for 90 days (X504)

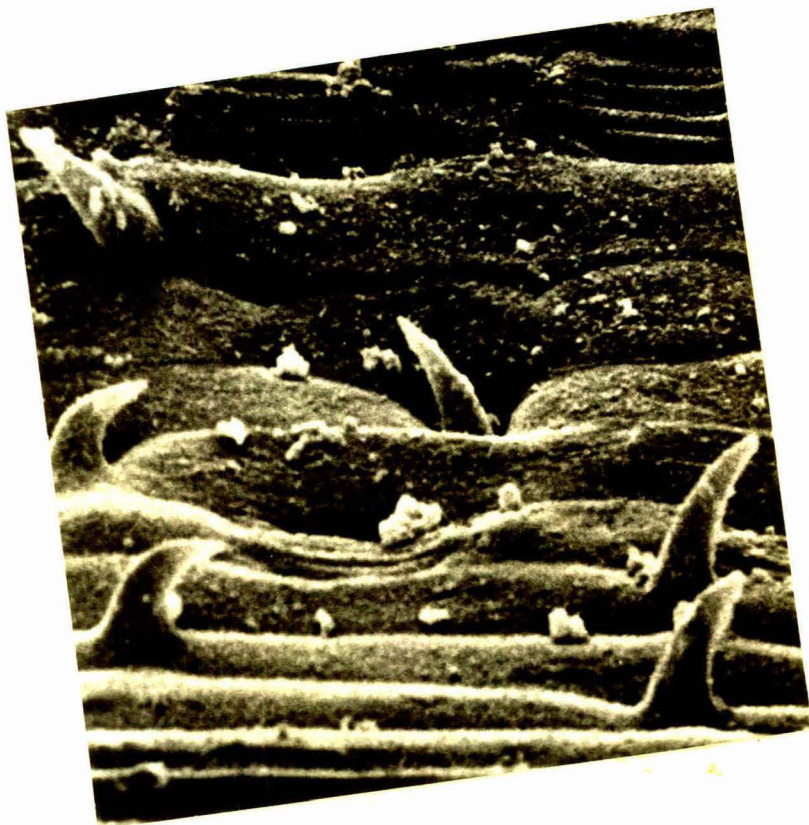
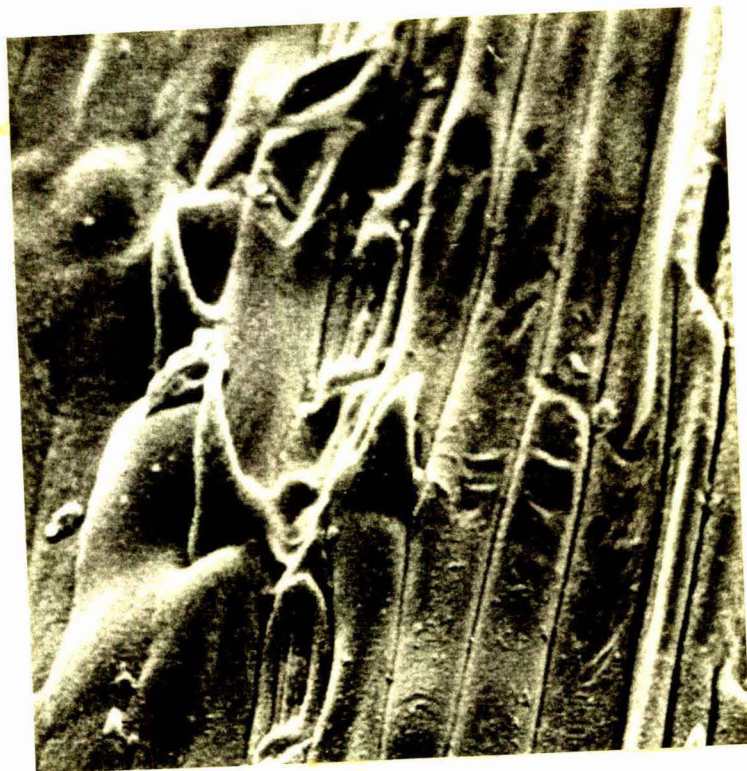


Fig. 3.63 Scanning electron micrograph of trichomes on the adaxial leaf surface of Triticum aestivum exposed artificially to  $\text{SO}_2$  for 90 days (X504)

Fig. 3.64 Scanning electron micrograph of trichomes on the adaxial leaf surface of Triticum aestivum exposed artificially to flyash for 90 days (X504)

Fig. 3.65 Scanning electron micrograph of trichomes on the adaxial leaf surface of Triticum aestivum exposed artificially to a combination of  $\text{SO}_2$  and flyash for 90 days (X504)



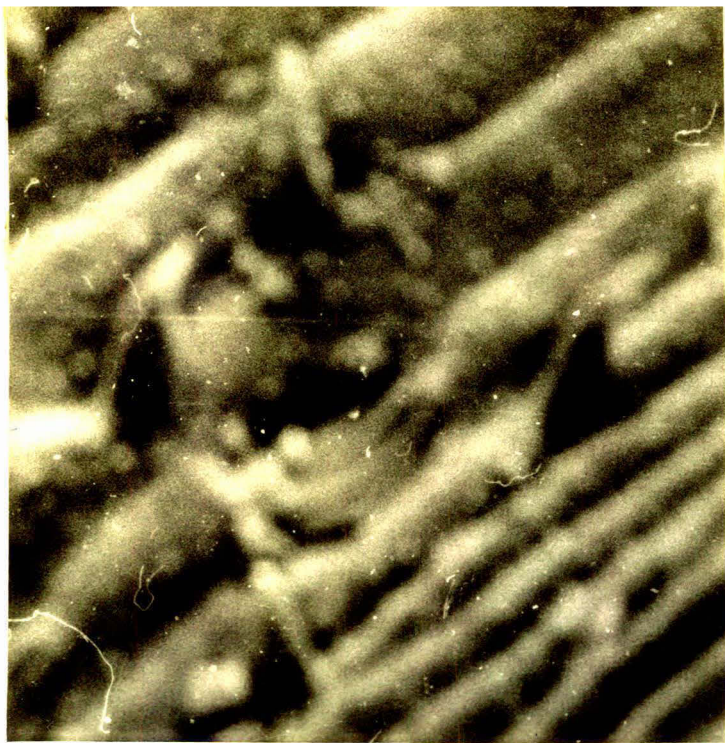


Fig. 3.66 Scanning electron micrograph of trichomes on the abaxial leaf surface of Triticum aestivum kept as control for 90 days (M504)

Fig. 3.67 Scanning electron micrograph of trichomes on the abaxial leaf surface of Triticum aestivum exposed in field near IP Power Plant for 90 days (X504)

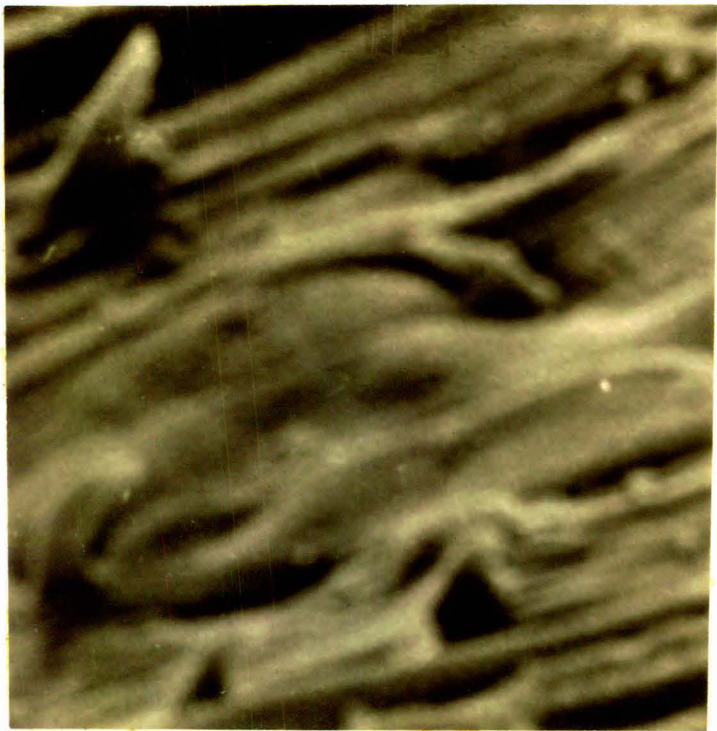
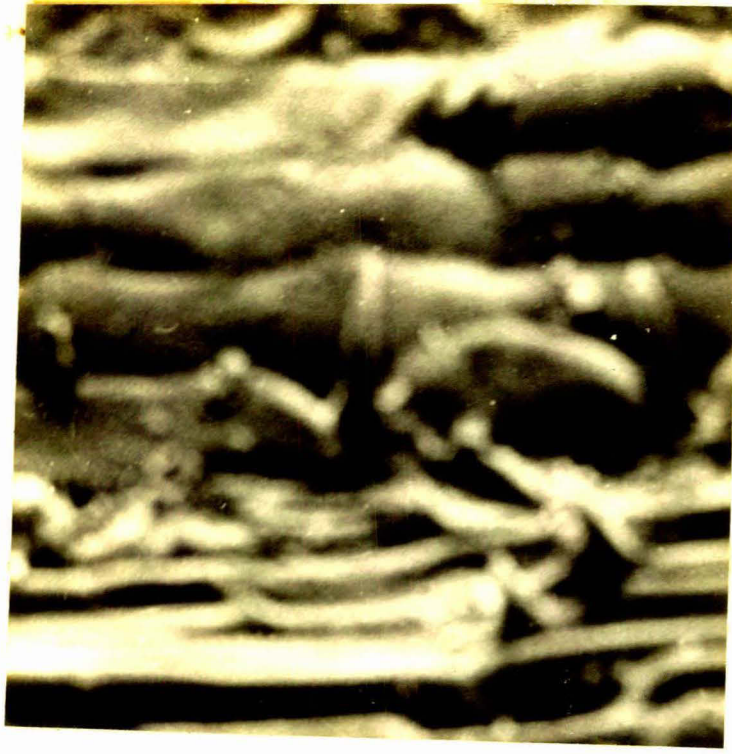


Fig. 3.68 Scanning electron micrograph of trichomes on the abaxial leaf surface of Triticum aestivum exposed to SO<sub>2</sub> for 90 days (X504)

Fig. 3.69 Scanning electron micrograph of trichomes on the abaxial leaf surface of Triticum aestivum exposed artificially to flyash for 90 days (X504)

Fig. 3.70 Scanning electron micrograph of trichomes on the abaxial leaf surface of Triticum aestivum exposed artificially to a combination of SO<sub>2</sub> and flyash for 90 days (X504)



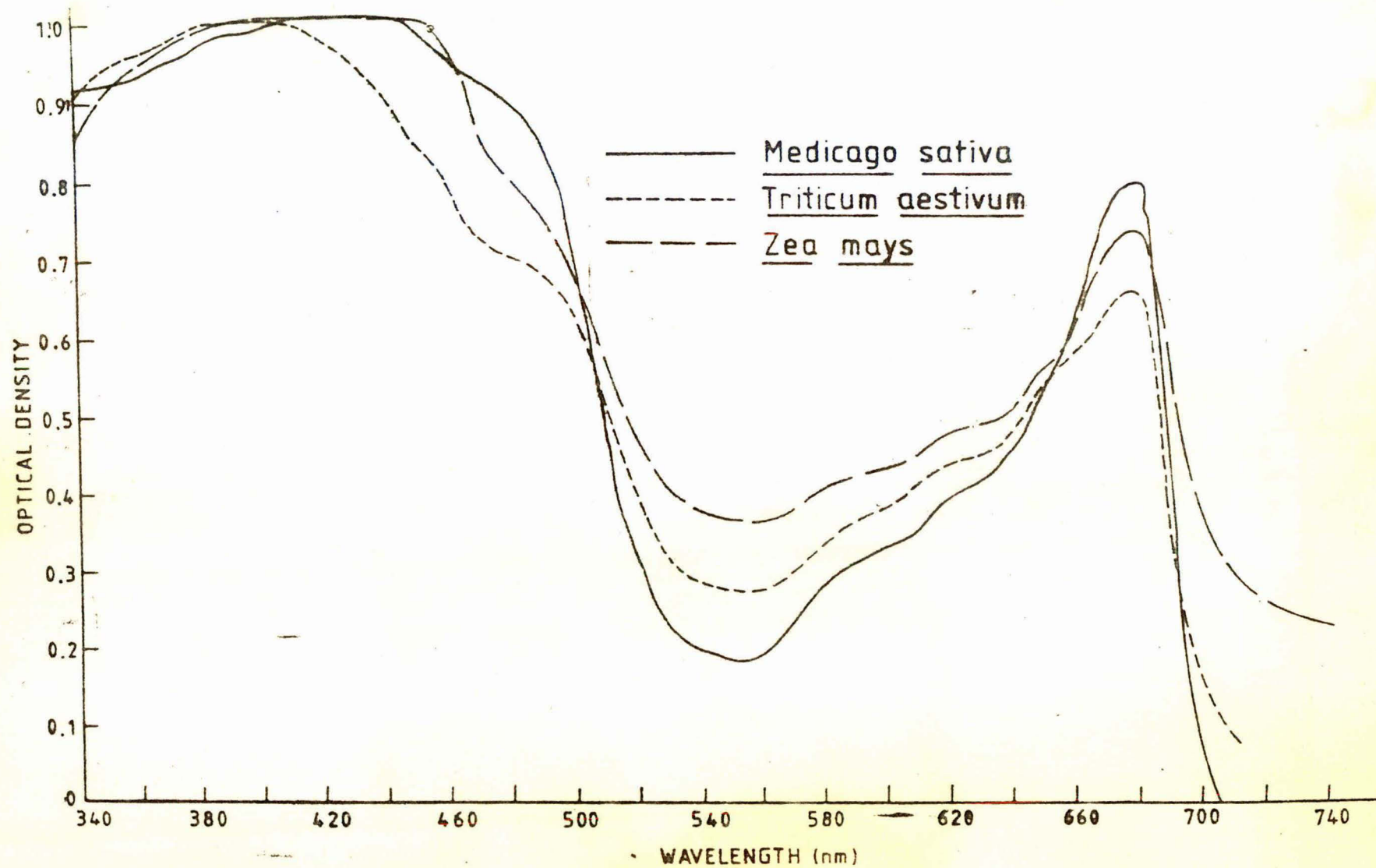


Fig. 3.71 Absorption spectra in visible region of Medicago sativa, Triticum aestivum and Zea mays plants kept as control for 45 days.

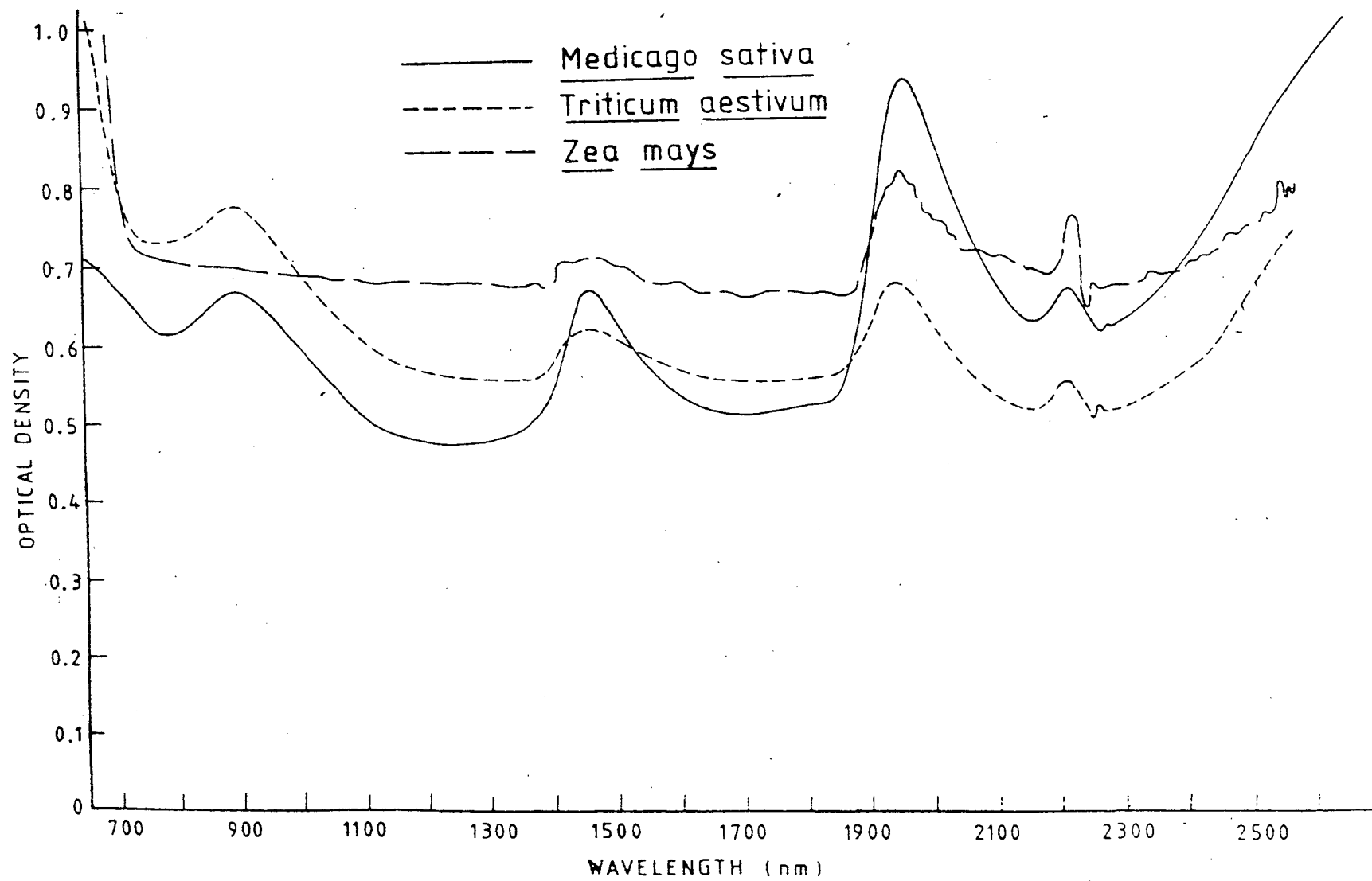


Fig. 3.72 Absorption spectra in infrared region of Medicago sativa, Triticum aestivum and Zea mays plants kept as control for 45 days.

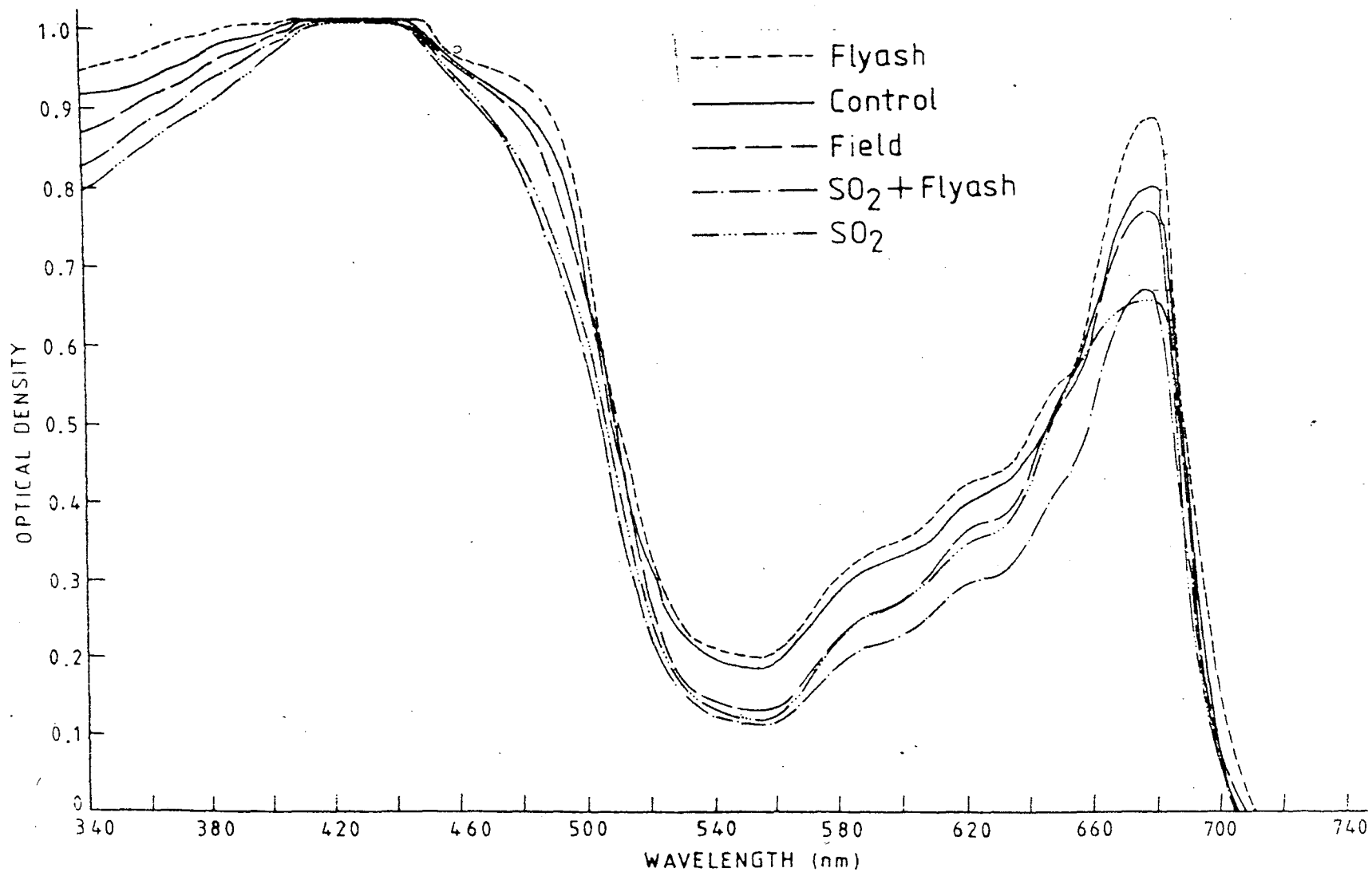


Fig. 3.73 Absorption spectra in visible region of *Medicago sativa* plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash for 45 days (F-A).



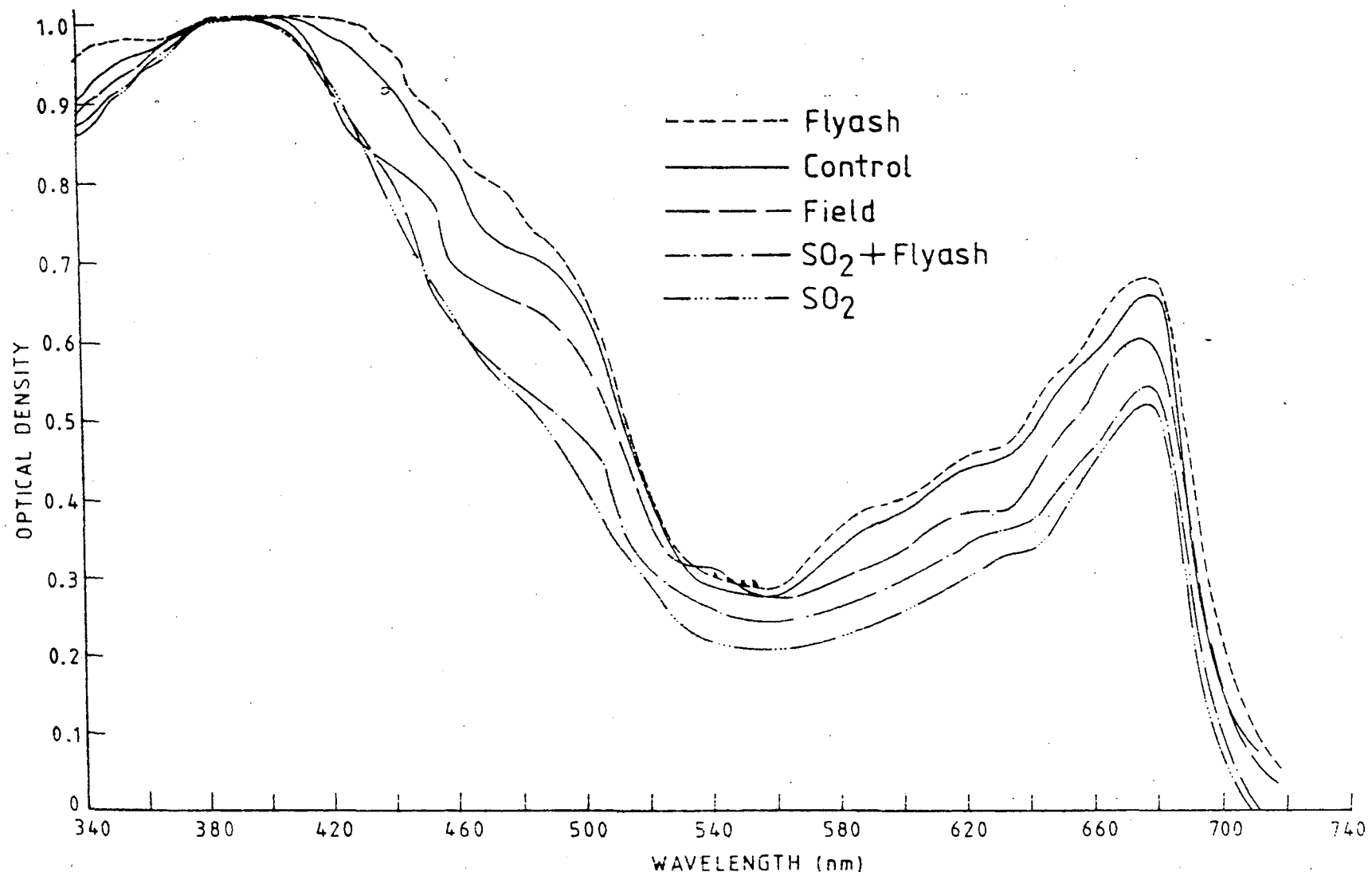


Fig. 3.74 Absorption spectra in visible region of *Triticum aestivum* plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash for 45 days (F-A).

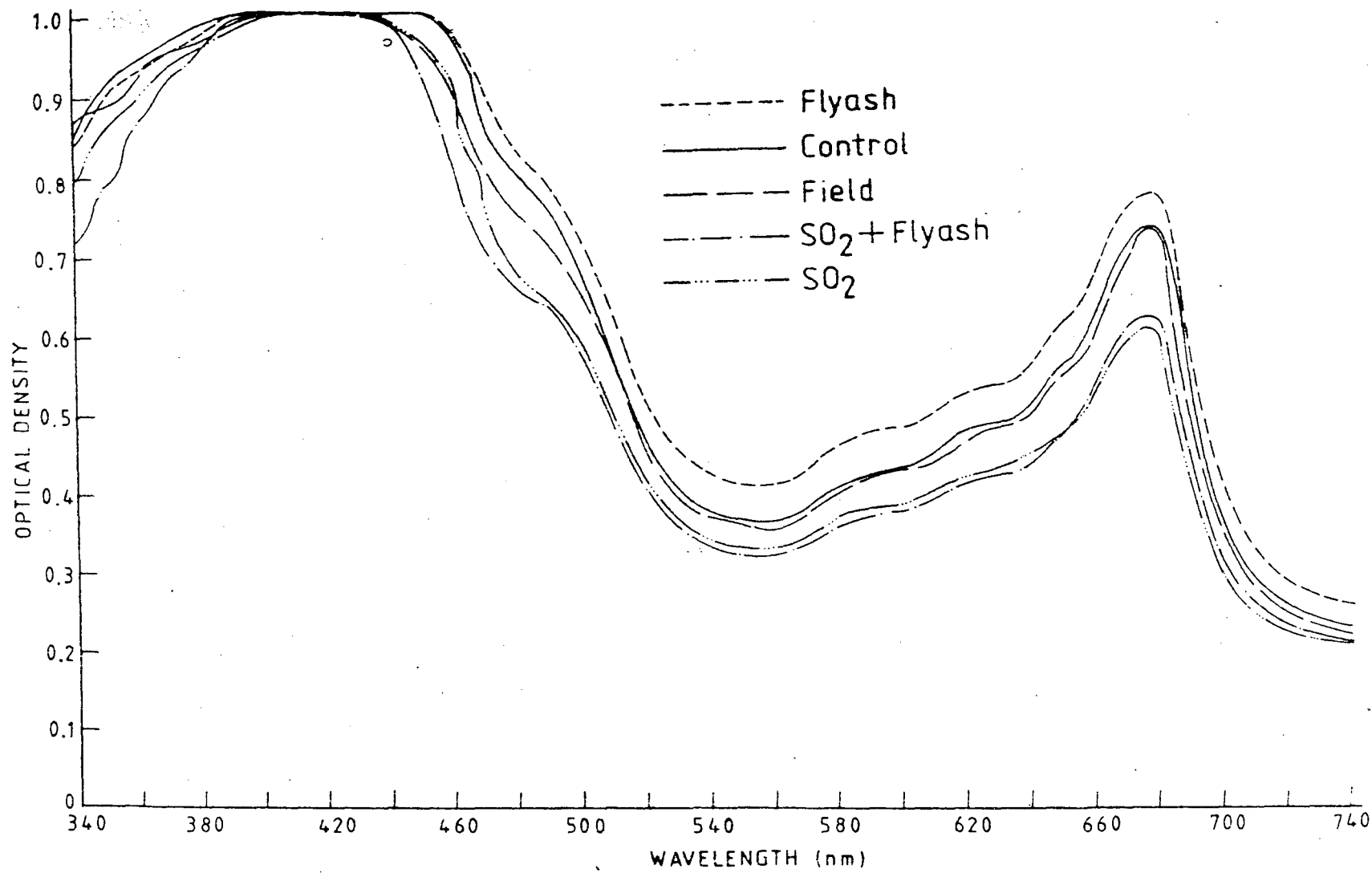


Fig. 3.75 Absorption spectra in visible region of Zea mays plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash for 45 days (F-A).

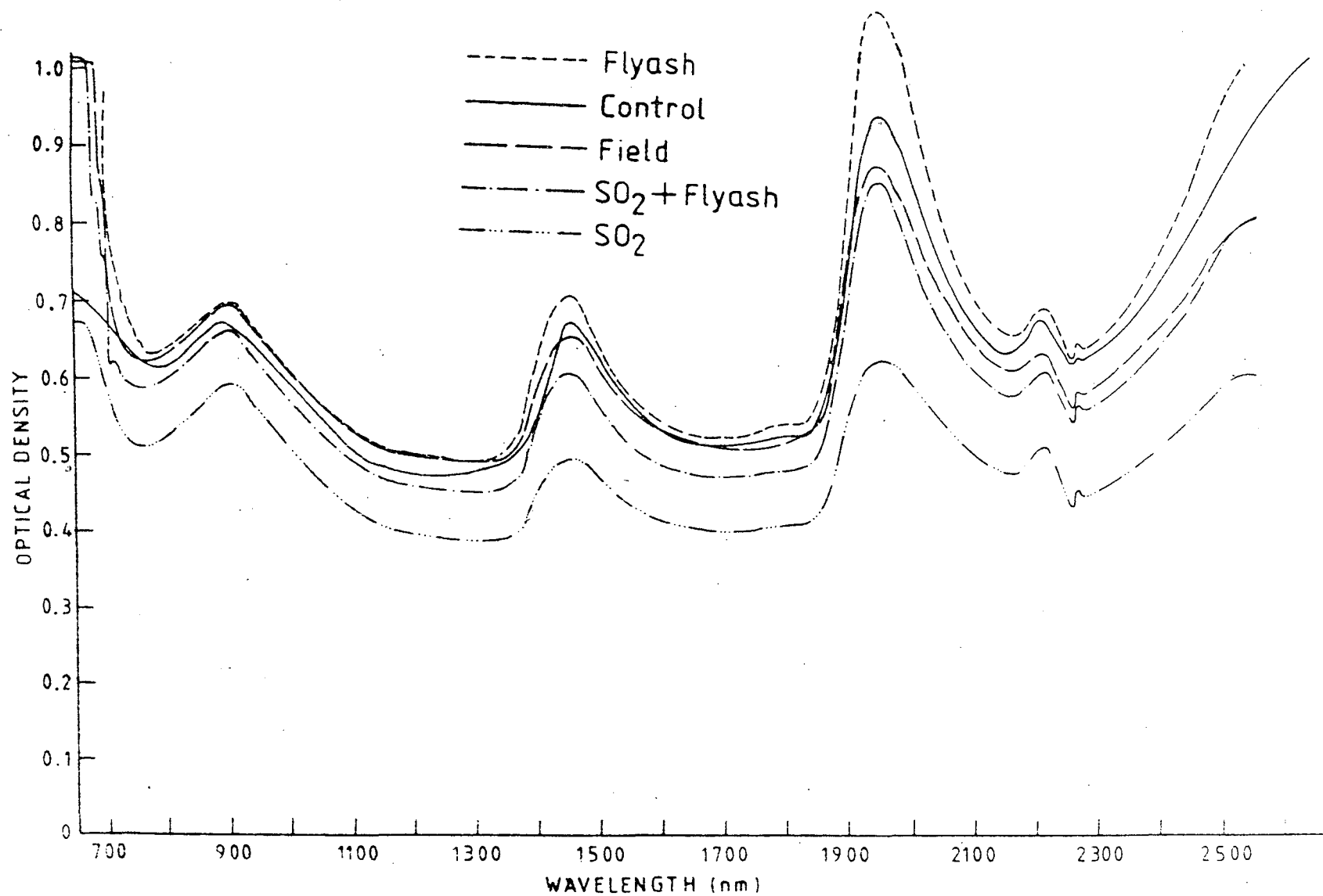


Fig. 3.76 Absorption spectra in infrared region of *Medicago sativa* plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash for 45 days (F-A).

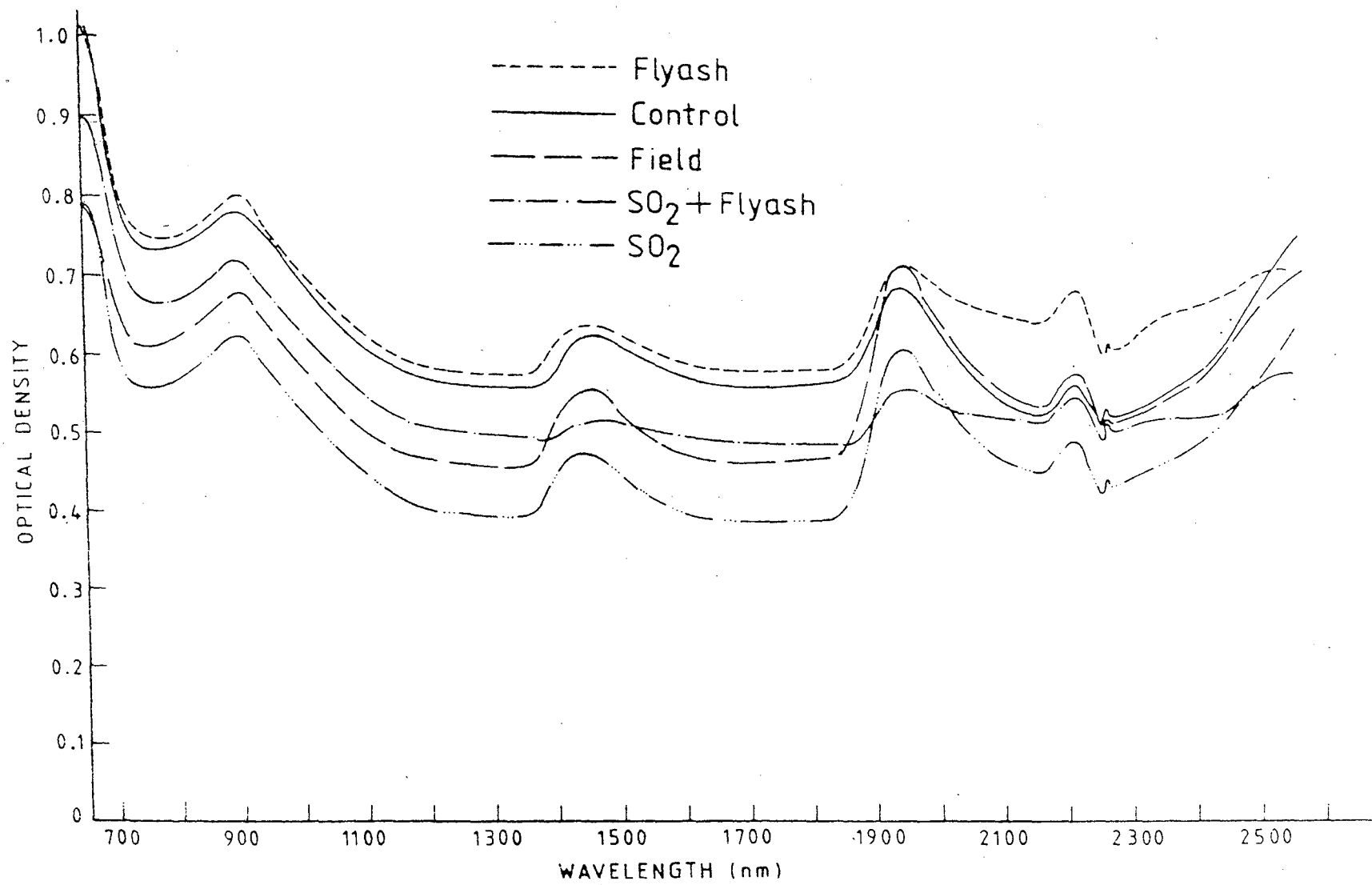


Fig. 3.77 Absorption spectra in infrared region of *Triticum aestivum* plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash for 45 days (F-A).

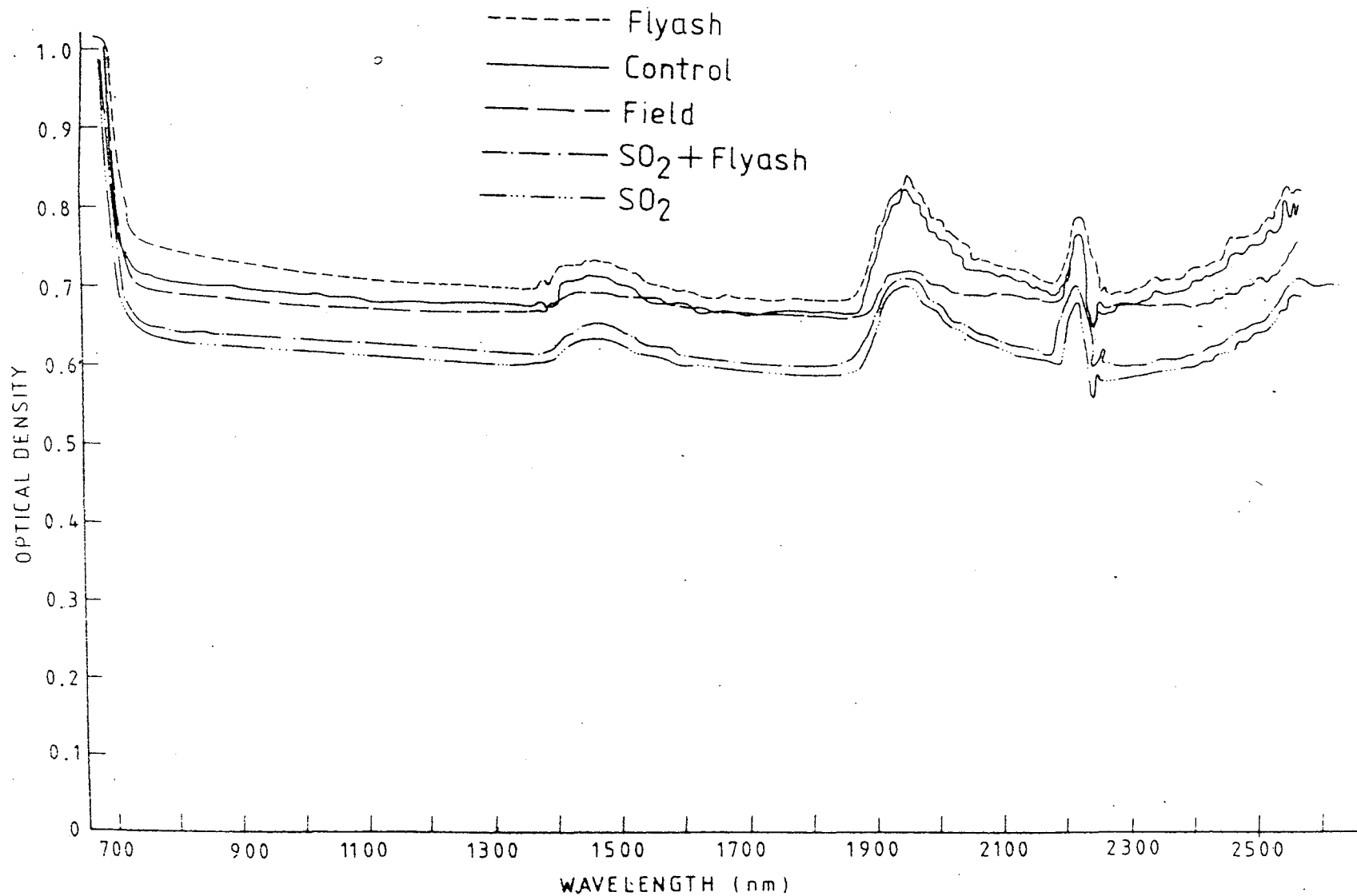


Fig. 3.78 Absorption spectra in infrared region of *Zea mays* plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash for 45 days (F-A).

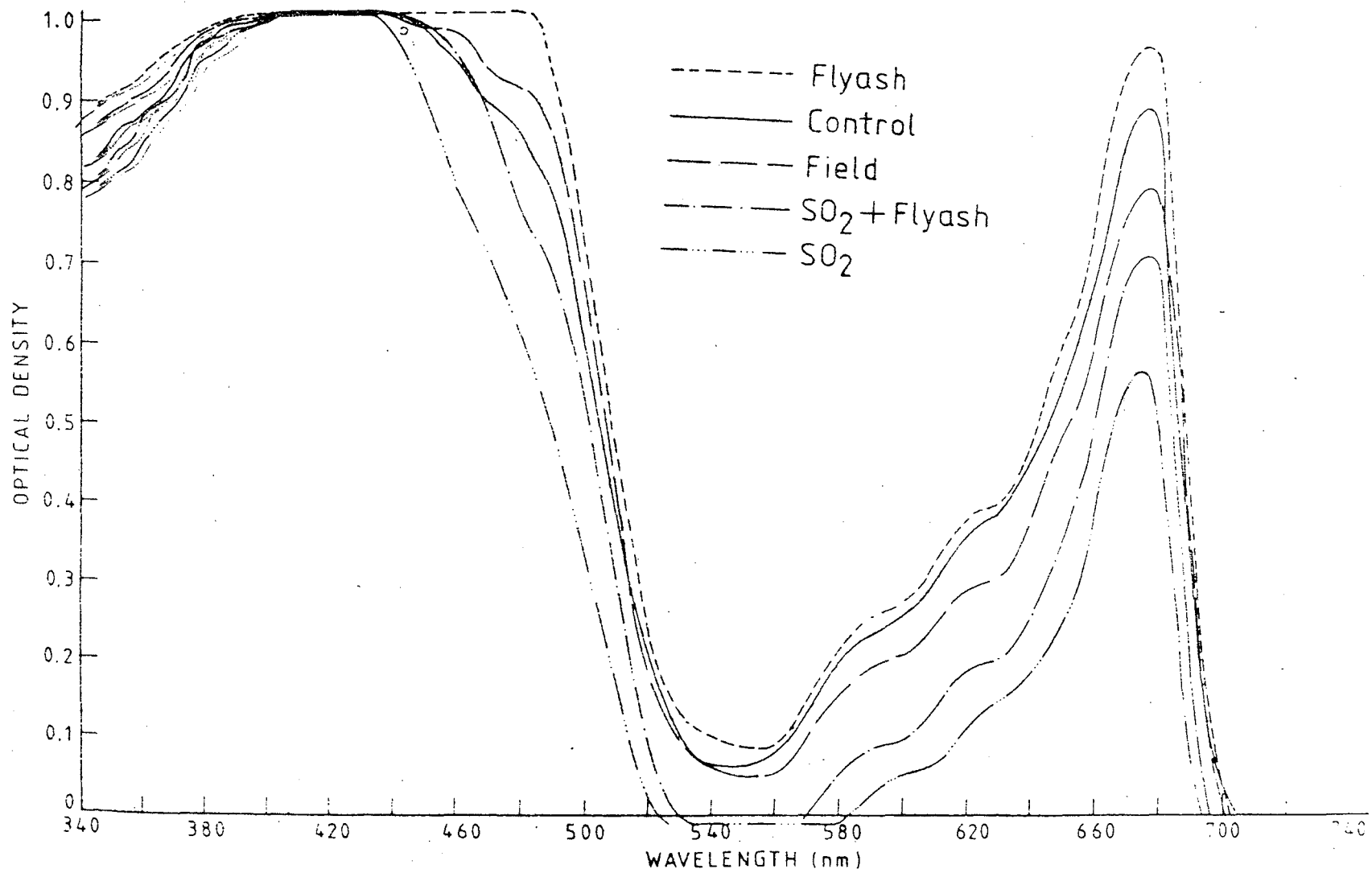


Fig. 3.79 Absorption spectra in visible region of *Medicago sativa* plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash for 90 days (FF-AA).

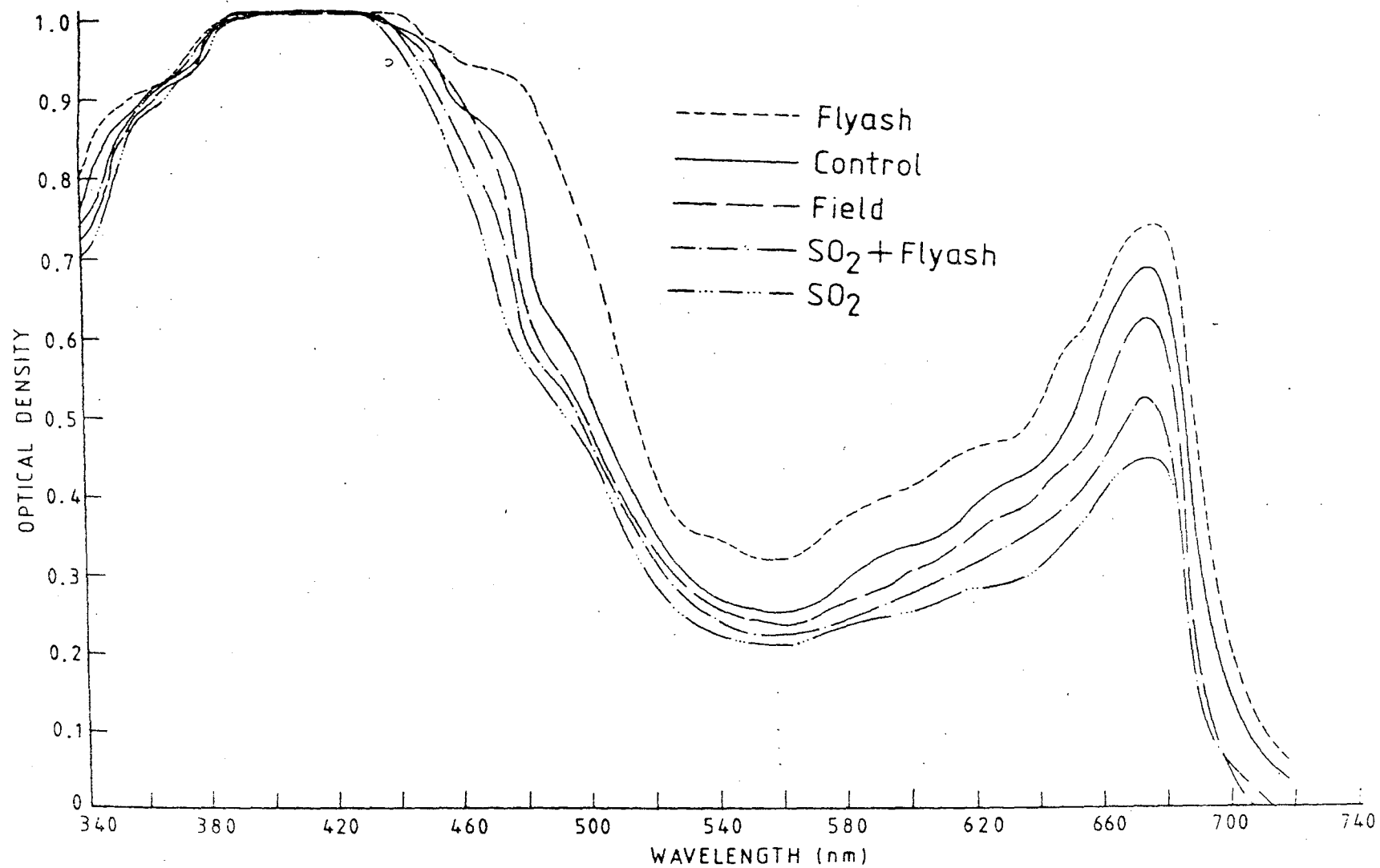


Fig. 3.80 Absorption spectra in visible region of *Triticum aestivum* plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash for 90 days (FF-AA).

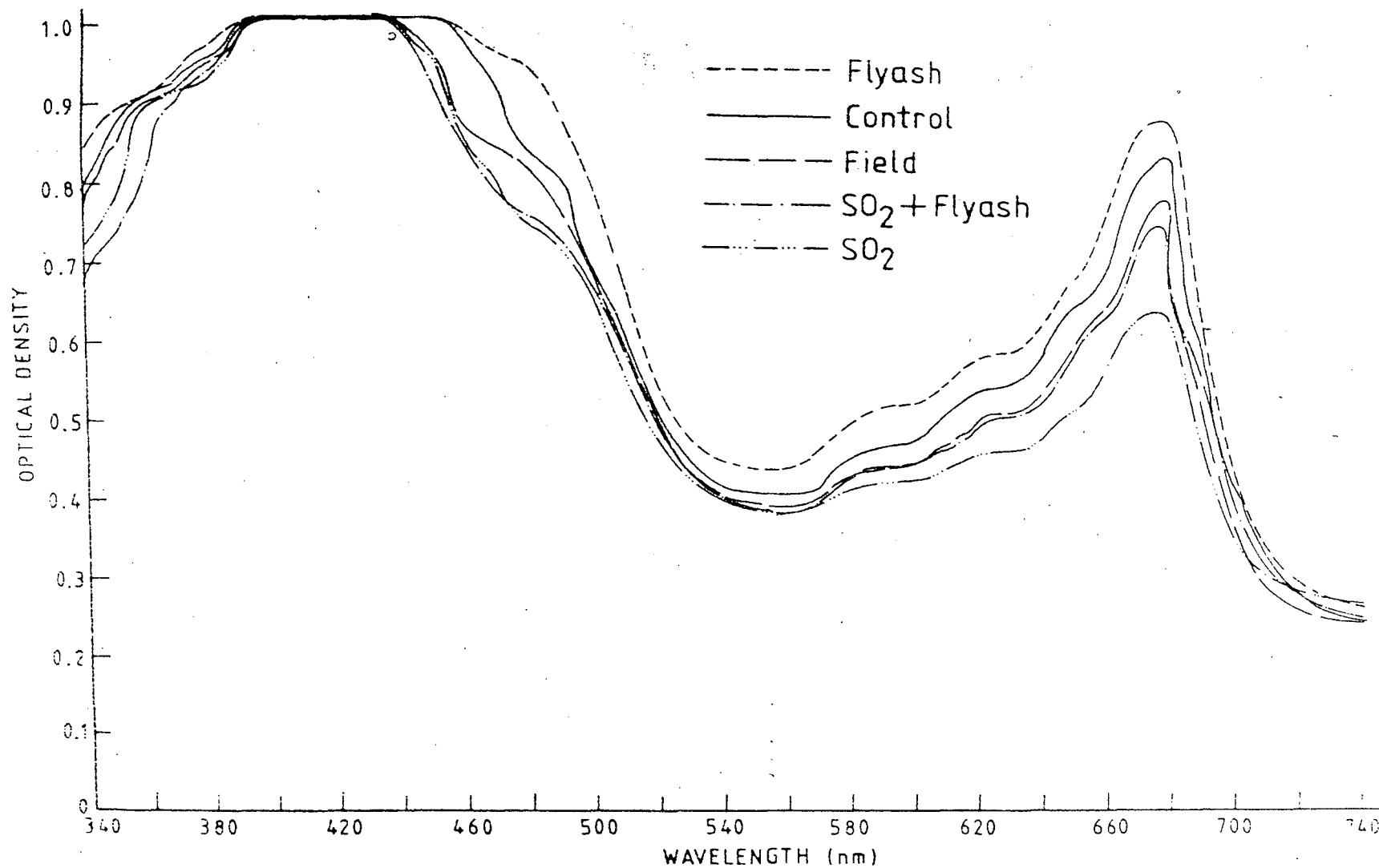


Fig. 3.81 Absorption spectra in visible region of *Zea mays* plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash for 90 days (FF-AA).



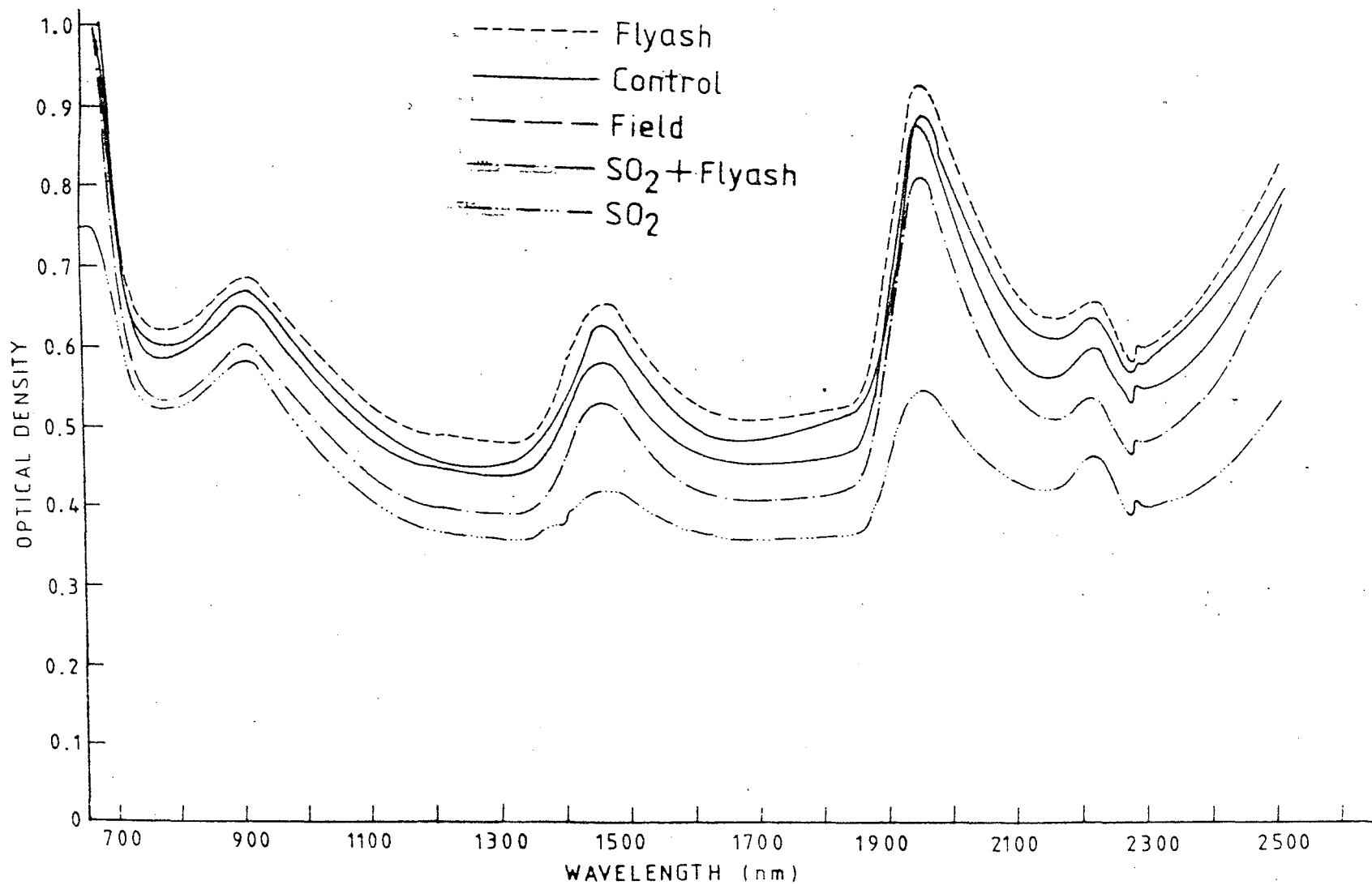


Fig. 3.82 Absorption spectra in infrared region of Medicago sativa plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash for 90 days (FF-AA).

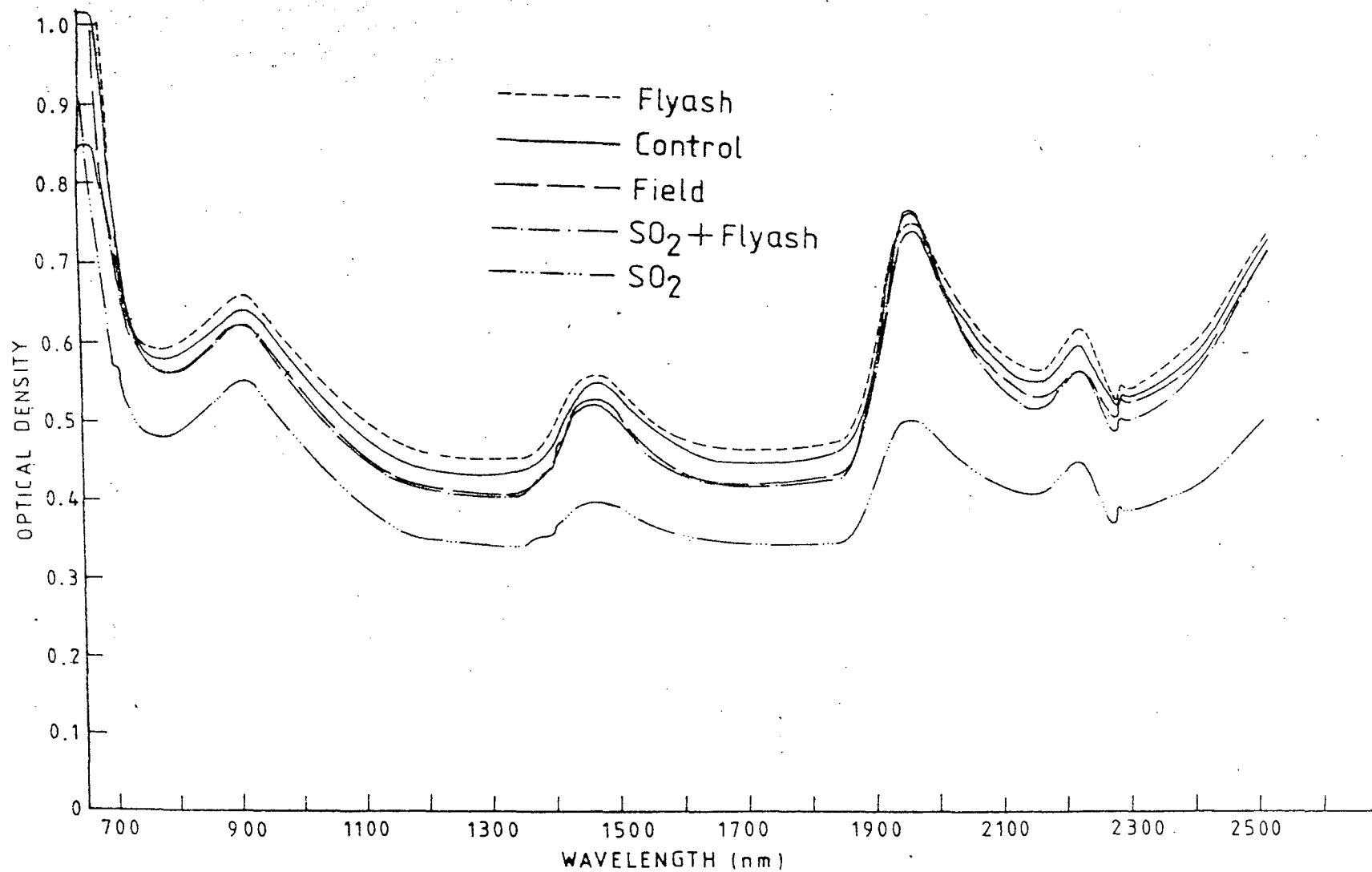


Fig. 3.83 Absorption spectra in infrared region of Triticum aestivum exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash for 90 days (FF-AA).

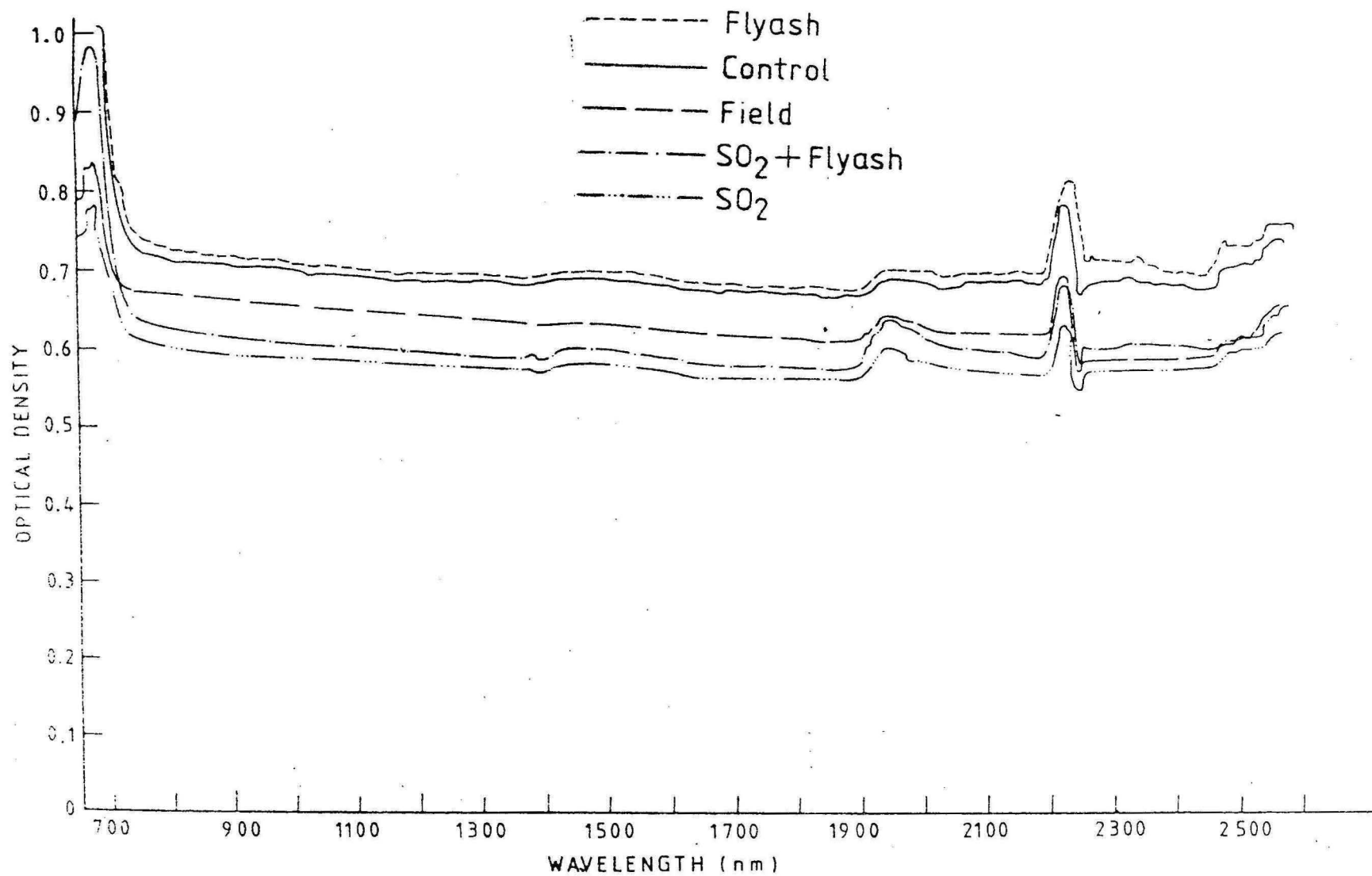


Fig. 3.84 Absorption spectra in infra-red region of *Zea mays* plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash for 90 days (FF-AA).

## DISCUSSION

Observations made on visual leaf injury, growth alteration, total chlorophyll content, epidermal features diffusive resistance, leaf surface temperature and leaf absorbance of Medicago sativa, Triticum aestivum and Zea mays plants exposed in field (IP Thermal Power Plant, New Delhi) and artificially to SO<sub>2</sub> and/or flyash for 45 and 90 days are discussed below in the light of the published literature.

### Visual Injury Symptoms

Chlorotic spots were observed on leaf surfaces of Medicago sativa plants exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash; while Triticum aestivum and Zea mays were unaffected.

In field, near thermal power plant dispersed chlorotic spots, tip and interveinal necrosis were noticed on leaf surfaces of some plants (Scheffér and Hedgcock, 1955; Dubey et al., 1982). However, contrary to our findings, necrotic streaks were observed in grasses (Pandey, 1983). Studies pertaining to impact of a combination of SO<sub>2</sub> and flyash, under laboratory conditions are lacking.

Chlorotic and necrotic injuries were prominent in plants growing in an environment mainly polluted by  $\text{SO}_2$  from copper or nickel smelters (Haywood, 1910; Linzon, 1972) and fertilizer factories (Linzon, 1965; Chaphekar, 1980). In laboratory, more than 140 ppb of  $\text{SO}_2$  under short term duration has been shown to cause visual injuries on leaf surfaces (Hill and Thomas, 1933; Katz, 1949). Medicago sativa is reported to be most sensitive plant, showing bleaching appearance in interveinal region at initial stage, which later on turns ivory or white (Laccaise and Treshow, 1978).

√ In present study, flyash sprayed plants have not shown any visual injuries, however, plants in vicinity of cement factory were found to have necrotic spots (Czaja, 1960), while brown necrotic patches in plants growing in coal unloading areas were noticed (Rao, 1971). Plants sprayed with  $4.7 \text{ g m}^{-2} \text{ d}^{-1}$  cement dust, for two days, had dead interveinal areas (Darley, 1966). In a study, evaluating the comparative impact of three kinds of particulate matter, Pawar et al. (1982) noticed that leaves of Hibiscus amelmoschush sprayed ( $2 \text{ g m}^{-2} \text{ d}^{-1}$  for 30 days) with coal and cement dust had chlorotic spots, while flyash sprayed leaves have not exhibited any visual injuries.

The present study alongwith a comparative view of other studies reveal that it is not necessary that every plant exposed to same dose of a pollutant exhibits visual injuries on the leaf surfaces and a plant species do not respond equally, when exposed to different kinds of pollutants.

Further, it was noticed that although Triticum aestivum and Zea mays have not exhibited visual injuries, yet reduction in their growth parameters was prominent. It also drives support from some studies, indicating the similar responses in plants (Bleasdabe, 1952; Tingley, 1971; Malhotra, 1977). It suggests that a leaf may look healthy in outer appearance but it should not be taken as a criteria for declaring it as resistant species or it is growing in pollution tree zone.

#### Growth alterations

In general, leaf area and biomass and total plant biomass reduced in Medicago sativa, Triticum aestivum and Zea mays plants exposed in field and artificially to SO<sub>2</sub> and/or flyash for 45 and 90 days except for marginal increase in leaf and total plant biomass in plants sprayed with flyash for 45 days. These observations are based on the quantitative data for each parameters, keeping control

under identical conditions of plant age, plant density and soil etc.

Plants in the vicinity of coal based power plants have been reported to be affected adversely (Linzon, 1972; Dubey et al., 1982; Pandey, 1982). These studies are based on qualitative assessment of growth parameters, however, Dubey et al. (1983) estimated phytomas accumulation based on extrapolation of average leaf biomass. In laboratory, responses to plants exposed to a combination of SO<sub>2</sub> and flyash have not been examined. Anyhow, a recent attempt by Dubey et al. (1983) has shown that biomass was less in Cicer arietenum plants exposed to a combination of SO<sub>2</sub> and flyash.

In an environment, where SO<sub>2</sub> is predominantly an air pollutant due to activities copper or nickel smelter, reduced growth of plants has been reported by number of workers (Haywood, 1910; Linzon, 1972; Carlson, 1974). Crittenden and Read (1979) concluded that grasses are more susceptible than other plants when exposed to ambient air containing 50 to 90 ug m<sup>-3</sup> SO<sub>2</sub>. Based on artificial fumigation studies, it was suggested that more than 140 ppb of SO<sub>2</sub> is able to cause adverse effects on growth of plants (Hill and Thomas, 1933; Katz, 1949).

Bell and Clough (1973) reported 46% reduction in ryegrass biomass, on exposure to 1.2 ppm of SO<sub>2</sub> for 9 weeks. Reduced growth of Triticum aestivum was assayed due to exposure to 0.8 ppm SO<sub>2</sub> for 2 hr daily for 60 days. Medicago sativa plants fumigated with air containing SO<sub>2</sub> upto 96 ug m<sup>-3</sup> for 135 days (crop harvested four times) exhibited reduction in morphological parameters.

Studies on plant responses to flyash are very limited, however, reports on plants in vicinity of cement factory have indicated decrease in their growth parameters (Darley, 1966; Singh and Rao, 1980). Growth and yield was reduced in Triticum aestivum plants sprayed with 7 g m<sup>-2</sup> d<sup>-1</sup> cement dust (Singh and Rao, 1978). Cement dust has been reported to be more injurious to Hibiscus amelmoschush plants as compared to coal dust and flyash (Pawar, et al., 1982).

The reduction in area and biomass of leaf, total plant biomass may be attributed to reduction in chlorophyll content as observed in present and in other studies (Rao and LeBlanc, 1966; Malhotra, 1977; Lanrenroth and Dodd, 1981; Dubey et al., 1982). Decrease in net photosynthetic rates (Ziegler, 1972; Sij and Swanson, 1974) and increase in respiration rates (Keller and Muller, 1958),



under pollution stress may contribute significant for the reduced growth. The increase in the leaf and total plant biomass due to flyash spray may be attributed primarily to increase photosynthetic pigment, chlorophyll b, due to shading effect of flyash deposition on leaf surfaces as suggested by some workers (Misra et al., 1978; Pawar et al., 1982; Dubey et al., 1983). Other factors which may cause increase in biomass include increased availability of soluble micronutrients from flyash through leaf surface (Roharman, 1971) and partly due to increased contents of sulphur (Biseewi et al., 1970), calcium and magnesium (Adrino et al., 1978) and zinc (Schnoppinger et al., 1975), in flyash treated soils. An increase in the leaf surface temperature as observed in present study may also be helpful in enhancing the rate of photosynthesis and other biochemical reactions.

Among the plant parts, leaf has been found to be more sensitive, thus may be useful in pollution studies. In this aspect, precautions for identical control should be considered as far as possible, because leaf area and biomass are very much dependent upon environmental factors and soil conditions etc.

### Total chlorophyll content

In plant species selected for present study total chlorophyll content was less, due to exposure in field and artificially to SO<sub>2</sub> and/or flyash for 45 and 90 days with an exception in plants sprayed with flyash for 45 days, it increased as compared to control.

Few studies conducted on the chlorophyll content under pollution stress in field have revealed that chlorophyll content was less in plants kept in the vicinity of thermal power plant (Varshney and Garg, 1980) and those growing near power plant (Dubey *et al.*, 1982; Pandey, 1983). Variation in chlorophyll content in plants due to exposure to a combination of SO<sub>2</sub> and flyash has not been evaluated.

Most of the information available on changes in chlorophyll content due to SO<sub>2</sub>, is for laboratory conditions (Rao and LeBlanc, 1966; Malhotra, 1977). However, Laurenroth and Dodd (1981) measured the chlorophyll content in Agropyron smithii exposed to low doses of SO<sub>2</sub> during ingrowing season for 4 yrs. These investigations have revealed reduction in chlorophyll content.

Quantitative assessment has been made for chlorophyll content in plants growing near cement factory (Auclair, 1976; Singh and Rao, 1980) and Phaseolus aureus sprayed with petro coke dust (Singh and Rao, 1981). In general, reduction in chlorophyll content was observed in these studies, but increase in chlorophyll content was noticed initially in plants sprayed with petro coke dust. Increase in chlorophyll content has also been observed in plants sprayed with flyash (Pawar et al., 1982; Dubey et al., 1983).

The decrease in chlorophyll content under acute exposure of  $\text{SO}_2$  has been attributed to conversion of chlorophyll into phaeophytin as  $\text{Mg}^{++}$  ions at the central position of chlorophyll is replaced by  $2\text{H}^+$  ions (Rao and LeBlanc, 1966). Increase in chlorophyll content due to spray of petro coke dust or flyash may be on account of shading effect caused by deposition of particulate matter which in turn increases the synthesis of chlorophyll b.

## Epidermal Features

The section has been divided into two parts:

(i) Stomata; (ii) Trichomes.

### Stomata

Density of stomata and length and breadth of stomatal pore reduced on both adaxial and abaxial leaf surfaces in Medicago sativa, Triticum aestivum and Zea mays plants exposed in field and artificially to SO<sub>2</sub> and/or flyash for 90 days. Survey of literature reveals, in brief, that most of the workers have reported decreasing pattern at both surfaces in stomatal density and stomatal pore size in plants from areas having major air pollutant such as SO<sub>2</sub> and particulate matter (Sharma and Butler, 1973, 1975; Garg, 1979; Garg and Varshney, 1980), heavy metals (Cu, Zn, Ni) and SO<sub>2</sub> (Caiazza and Quinn, 1980). However, in few plants collected from vicinity of cement factory or urban polluted areas, increase in stomatal density has been reported without specifying the leaf surface (Yunus and Ahmed, 1979, 1982; Srivastava, 1982). There has been no study made so far to study such variations under laboratory conditions, however, the present study is an attempt to assess variation in stomata density and length and breadth of stomata in plants exposed artificially to SO<sub>2</sub> and/or flyash for 90 days.

The factors responsible for such changes are not known but information available on the normal leaf development (Dennett et al., 1970; Charles and Edward, 1979) its alternation due to factors like light intensity (Verbelen and DeGreef, 1979),  $O_3$  (Tang and Mukerji, 1971) and UV radiations (Dickson and Caldwell, 1978) may be useful. Further variations in development of stomata in mutant plants of maize (Maryard et al., 1974) may be helpful to understand the behaviour of stomata under pollution stress.

Decrease in stomatal density and length and breadth of stomatal pore reduces the rate of gaseous exchange in plants in polluted environment, thereby serving as an avoidance factor (Winner and Mooney, 1980).

#### Trichome

Increase in the density and length of trichomes was noticed in Triticum aestivum plants exposed both in field and artificially to  $SO_2$  and/or flyash. It is important to note that all the plants studied so far in relation to variation in trichomes due to air pollution stress under field conditions have shown increase both in density and length (Sharma and Butler, 1973, 1975; Garg, 1979; Calazza and Quinn, 1980; Garg and Varshney, 1980), however,

no attempt has been made under laboratory condition. Although studies pertaining to the cause for such behaviour have not been done, yet information like variation in developmental stage of trichome in mutant plants of maize may provide an insight (Maryard, et al., 1974).

Sharma (1975) suggested that increase in density and length of trichomes enhance the surface area of leaf, thus providing more area for absorption. In other words, absorption of pollutants by the leaf is reduced as a result of more adsorption by trichomes. An indirect evidence to this was provided by Elkley and Ormrod (1979) as he found that peturia cultivars, which are resistant to  $SO_2$  have abundant trichomes as compared to  $SO_2$  sensitive species. Giridhar and Chaphekar (1983) pointed that it is the wet pubescent surface in Solanum melongena, which has high adsorbing capacity than wet smooth surface of Cyamopsis tetragonolaba. However, the dry smooth surface of Cyamopsis tetragonolaba has comparatively more adsorbing capacity than dry pubescent surface of Solanum melongena. A pubescent surface is more likely to have low absorption and high reflection (Johnson, 1975) thereby lowering the leaf and temperature, which in turn will lower the metabolic rates in living cells

and reduces the susceptibility to pollution damage (Sharma and Butler, 1975). Increase in loss of elasticity of leaves due to loss of small folds (Godzik and Sassen, 1978) and extraction of wax under pollution stress (Koziol and Cowling, 1981) may serve as avoidance factors to reduce pollution load.

## Diffusive Resistance

Observations for leaf diffusive resistance were taken after both short and long term exposures, and are discussed here separately.

### Short term exposure

Leaf diffusive resistance decreased in plants exposed infield and artificially to  $\text{SO}_2$  and a combination of  $\text{SO}_2$  and flyash. It did not change in flyash exposed plants. Most of the diffusive resistance studies in relation to air pollutants have been carried out taking  $\text{SO}_2$  as an air pollutant. It has been shown that low doses of  $\text{SO}_2$  causes decrease in diffusive resistance (Majersik and Mansfield, 1970, 1971; Unsworth, et al., 1972; Biscoe et al., 1973; Black and Black, 1979). The extent of opening has been shown to vary with environmental factors in some plants like humidity (Majernik and Mansfield, 1970), light intensity (Mansfield and Majernik, 1970), difference in air and leaf temperature, vapour pressure deficit, etc. Olszyk and Tibbitts (1981b) pointed out that younger leaves are less sensitive than a mature leave. The same pattern was noticed in the present study, as 45 days old plant has shown comparatively less decrease in diffusive resistance that the same leaf in 90 days old plants.



The decrease in diffusive resistance or wider opening of stomatal pore under SO<sub>2</sub> pollution stress may be attributed to the change in turgor pressure of epidermal cells, thereby changing the membrane permeability of epidermal cells and consequently the turgidity of the guard cells (Biscoe et al., 1973; Black and Black, 1979). Black and Unsworth (1979a) provided a minor modification in this concept, suggesting that stomatal opening induced by  $\gg$  175 ppb SO<sub>2</sub> is passive and is a result of preferential loss of turgor within the adjacent epidermal cells (subsidiary cells). There is a little evidence of injury to the guard cells at relatively low concentrations of SO<sub>2</sub>. These aspects drive support from the observations such as (i) chemical substances reaching the guard cells usually enter via adjacent epidermal cells (Squire and Mansfield, 1972), (ii) direct absorption by guard cells is likely to be restricted by their cuticle, (iii) guard cells may be actively protected from injury by their ability of chloroplast to convert sulphite into less toxic substances (Libera et al., 1973).

Thus, although the mode of action of SO<sub>2</sub> remains uncertain, it appears probable that injury to epidermal cell membranes is the first sign of action.

The wider stomatal opening under pollution stress may be deleterious to the plants (i) air pollutants entries will be much more rapid causing adverse effects on the morphological, physiological and biochemical processes, (ii) rate of transpiration will be high which will put the plant into water stress conditions in dry climate situations, (iii) it may enhance the frequency of fungal infection in merophyll, which is a site for metabolic processes.

Some studies (Sij and Swanson, 1974; Black and Black, 1979) have been carried out at high dose of  $\text{SO}_2$  which reveal closure of stomatal pore or increase in the diffusive resistance. This behaviour of stomata has been explained as (i) it may be associated with the accumulation of  $\text{CO}_2$  in substomatal cavity following  $\text{SO}_2$  inhibition of photosynthesis (Sij and Swanson, 1974), (ii) due to change in the permeability of guard cell membrane as disorganization of chloroplast membrane was noticed as the first symptom of high concentration of  $\text{SO}_2$  (Wellburn et al., 1976; Malhotra, 1976). Rapid change in cell membrane permeability of lichens due to high concentration of  $\text{SO}_2$  was also observed (Puckett et al., 1976), (iii) Black and Black (1979) observed

death of one or both guard cells at  $\geq 500 \text{ ug m}^{-3} \text{ SO}_2$  but prior to death disorganization of chloroplast of guard cells takes place.

#### Long term exposure

Diffusive resistance increased in plants exposed in field and artificially to  $\text{SO}_2$  and/or flyash for 45 and 90 days. Comprehensive evaluation of variation in diffusive resistance under long term exposure of air pollutants is almost lacking. However, the response of stomata in terms density and length and breadth of stomatal pore has been studied in plants growing in polluted environment (Sharma and Butler, 1973, 1975; Caiazza and Quinn, 1980; Garg and Varshney, 1980) and in the present study. These studies have indicated reduction in stomatal density and length and breadth of stomatal pore, which in a way support the present observations for diffusive resistance under long term exposure.

### Optical Characteristics of Leaf

The leaf surface temperature varies in plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash. Numerically the pattern of change can be written as follows :

Flyash > Control > Field > SO<sub>2</sub> + Flyash > SO<sub>2</sub>

These changes are discussed on the basis which suggests that an imbalance of leaf-environment energy exchange relationship determine the leaf surface temperature.

This relationship can briefly be expressed in the form of the following equation (Idso et al., 1966):

$$Q_a = Q_r + C + LE + P$$

Where,  $Q_a$  = Energy absorbed both in visible and invisible regions

$Q_r$  = Reradiated energy

$C$  = Energy lost or gained by convection

$LE$  = Energy lost as latent heat of vaporisation of water in transpiration

$P$  = Energy used in photosynthesis for CO<sub>2</sub> fixation.

Energy absorbed ( $Q_a$ )

In the present study, observations made for absorption spectrum in visible and infrared regions may be helpful in providing us variation in energy absorbed due to interaction of air pollutants. In general, the leaf absorbance

pattern exhibited by the plants exposed in field and artificially to  $\text{SO}_2$  and flyash and a combination of flyash was found to be in the following sequence both in visible and in infrared regions:

Flyash > Control > Field >  $\text{SO}_2$ +Flyash >  $\text{SO}_2$

It reveals that flyash has caused the increase in the leaf absorbance. It could be due to the deposition of flyash, a greyish black material which may be helpful in the direct absorption of light waves. Eller (1977b) reported that leaves of Rhododendron catawbiense growing along the roadside have shown an increase in leaf surface temperature by 2 to 4°C which on basis of absorption spectra analysis, was suggested to be due to marked increase in leaf absorbance in infrared region (700-1350 nm).

Further, it has been noted that in flyash sprayed plants chlorophyll content was more in the initial stages of plant growth, which in turn be helpful in increasing the absorbance in visible region particularly in the blue and red zones.

On the other hand,  $\text{SO}_2$  has caused maximum decrease in the leaf absorbance both in visible and infrared

regions. It may be attributed to its effectiveness in reducing the chlorophyll content. In addition to this, a factor which probably may affect the leaf absorbance indirectly is the trichome characteristics. It has been reported by number of workers as summarized by Johnson (1975) that a pubescent leaf has low absorbitivity (Gates et al., 1965; Wuenscher, 1970; Eller, 1977a) and usually high reflectivity (Shull, 1929; Billing and Morris, 1951; Gausman and Gardens, 1969, 1973).

#### Reradiated heat ( $Q_r$ )

The relationship between reradiated heat and leaf temperature is given by Stefan-Boltzman equation

$$Q_r = \epsilon \sigma T^4$$

where,  $Q_r$  is reradiation in  $\text{cal cm}^{-2} \text{ min}^{-1}$ ;  $\epsilon$  is leaf emmissivity,  $\sigma$  is the Stefan-Boltzman constant,  $8.130 \times 10^{-11} \text{ cal cm}^{-2} \text{ }^\circ\text{K}^{-4} \text{ min}^{-1}$ , and  $T$  is leaf temperature in  $^\circ\text{K}$ .

According to this relation, as the leaf temperature increases the amount of reradiation from the leaf also increases proportionately. It indicates that reradiation from the flyash sprayed plants will be more followed by plants exposed in field, to a combination of  $\text{SO}_2$  and flyash and to  $\text{SO}_2$  alone.

The direct evidence of an air pollutant on this aspect is lacking, however, in relation to trichomes, Gates et al. (1965) reported a reduction in the long wave emittance in the desert plant cactus, Mammilana lasiacantha, having a dense covering of fine thorns. However, Parker (1968) suggested that surface hairs may act as additional surfaces area from which radiation can occur. On this basis, it may be possible that increase in trichome density and length as observed in the present and several earlier studies, will increase the reradiation of light waves.

#### Convective heat loss (C)

Numerically convective heat loss can be calculated by the method described by Parkhurst et al. (1968). Convective heat loss C, can be represented by the following equation

$$C = h_c (T_1 - T_a)$$

where, C is in  $\text{cal cm}^{-2} \text{min}^{-1}$ ,  $h_c$  is in  $\text{cal cm}^{-2} \text{min}^{-1} \text{ } ^\circ\text{C}^{-1}$  and  $T_1$  and  $T_a$  are leaf and air temperatures in  $^\circ\text{C}$ .

$h_c$  - convective coefficient can be calculated from standard heat transfer theory (Kreith, 1965), taking effective leaf dimension into consideration.

It indicates that comparatively convective heat loss from leaves sprayed with flyash will be more followed by those exposed in field, to a combination of SO<sub>2</sub> and flyash and to SO<sub>2</sub> alone.

The convection coefficient is related to boundary layer thickness, d, by the equation

$$H_c = \frac{3k}{2d}$$

where, k is the thermal conductivity of air,  $6.2 \times 10^{-5}$  cal cm<sup>-1</sup> sec<sup>-1</sup> °C<sup>-1</sup> at 25°C (Kreith, 1965).

The above equations help us to understand that increase in boundary layer thickness will cause decrease in convection coefficient and thus, correspondently decrease the convective heat loss from the leaf. In other words, increase in the length and density of trichomes which causes increase the boundary layer thickness might be helpful in decreasing the convective heat loss from the leaf. In context of present study, it shows that since the increase in density and length of trichomes was less in flyash sprayed plant as compared to others, thus convective heat loss will be comparatively more in flyash exposed plants.



It has been also reported on the basis of experiment that pubescent leaf is warmer than same leaf, when hairs were removed (Heberlandt, 1914; Hendrycy, 1967; Wuencher, 1970). Contrary to this, Wolpert (1962) hypothesized that hairs might increase convection by acting as fins to conduct heat away from the leaf surface. But Uphof (1962) pointed out that it would require the presence of moving water or rapid protoplasmic streaming to rapidly move heat from the leaf surface out into the hairs, since most hairs on the mature leaf area dead empty cells and thus can not act as efficient convectors.

#### Latent heat of vaporization (LE)

Rate of transpiration has got inverse relationship with transpiration resistance, a sum of two resistances (i) stomatal resistance ( $R_s$ ), and (ii) boundary layer or air resistance ( $R_a$ ). It suggests that any change in either of the two resistances will influence the transfer of water vapour and thus, causing the change in the latent heat of vaporization. In the present study increase in the diffusive resistance (mainly stomatal resistance) under long term exposure was comparatively low as compared to other exposed plants, which implies that heat loss due to vaporization was relatively more as compared to other exposed plants.

Boundary layer air resistance ( $R_a$ ) is related to convective coefficient by the following equation (Kreith, 1965)

$$R_a = \left\{ \frac{D_h}{D_{H_2O}} \right\} \frac{\frac{2}{3} (C_p)_{\text{air}}}{h_c}$$

where,  $D_h$  and  $D_{H_2O}$  are the diffusivities of heat and water in air,  $C_p$  is the specific heat of air in  $\text{cal gm}^{-1} \text{ } ^\circ\text{K}^{-1}$ , and  $h_c$  is the density of air in  $\text{g cm}^{-3}$ .

Thus, an inverse relationship between convective coefficient and boundary layer resistance ( $R_a$ ) indicates that decrease in  $h_c$  will increase  $R_a$ . Increase in density and length of trichomes in plants from polluted area or in other words, increase <sup>in</sup> boundary layer thickness causes decrease in convective coefficient ( $h_c = \frac{3K}{2d}$ ) which in turn increases the boundary layer resistance. Thus, the net result will be a decrease in latent heat of vaporization.

#### Energy used in photosynthesis (P)

In the present study, it seems that energy utilization by plants sprayed with flyash was more as compared to plants exposed in field, artificially to a combination of  $\text{SO}_2$  and flyash and  $\text{SO}_2$  alone, as quantitative assessment reveals that biomass was more in flyash sprayed plants than others.

The change in the leaf surface temperature may influence the plant metabolic processes as photosynthesis, respiration etc. The optimum temperature for these processes varies in plant species and therefore to assess the impact of air pollutants, It is desirable to know the optimum temperature range for a process in a specific plant species. A simple classification based on  $C_3$  and  $C_4$  plants for different zones indicating optimum temperature for photosynthesis is given below

$C_3$	Temperate	-	15-25°C
	Tropical and Subtropical )	-	25-35°C
$C_4$	Temperate	-	25-35°C
	Tropical and Subtropical )	-	35-45°C

Although the present study provides an insight into the new aspects in pollution studies in relation to optical characteristics of leaf, diffusive resistance and epidermal features of plants, yet in order to understand the mode of action of such responses, research should be extended in directions given below :

1. A systematic quantitative evaluation of various parameters involved in the leaf - environment relationship ( $Q_a = Q_r + C + LE + P$ ) such as leaf emissivity, convective coefficient, boundary layer thickness, etc.
2. Mechanism of interaction of air pollutants with the developmental stages of stomata and trichomes.
3. Pathways for air pollutant movements within the plant and their effectiveness in affecting enzyme system of plant processes, using radioactive techniques. It will help us to understand the differential responses of plants to the same dose of a pollutant.
4. Studies in physiology and biochemistry of plants under pollution stress for long term will provide us the real picture against the traditional studies carried out at pollutant concentrations, atypical of those reported in a polluted environment.

## SUMMARY

The present study was carried out on Medicago sativa, Triticum aestivum and Zea mays plants exposed in field (in the vicinity of IP Thermal Power Plant, New Delhi) and artificially to SO<sub>2</sub> and flyash and a combination of SO<sub>2</sub> and flyash for 45 and 90 days. Observations were made for visual injuries, plant performance (leaf area, leaf biomass and total plant biomass), chlorophyll content, epidermal features (stomatal density, length and breadth of stomatal pore, density and length of trichomes), diffusive resistance, leaf surface temperature and leaf absorbance. The salient features of these studies are given below.

- Chlorotic spots were observed in Medicago sativa plants exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash for both 45 and 90 days. Triticum aestivum and Zea mays plants did not exhibit any visual injuries.

- In general, reduction in leaf area, leaf biomass and total plant biomass was observed in plants exposed in field and artificially to SO<sub>2</sub>, flyash and a combination of SO<sub>2</sub> and flyash for 45 and 90 days, however, increase in leaf biomass and total plant biomass was

observed in plants sprayed with flyash for 45 days. Changes in Medicago sativa plants were more prominent as compared to Triticum aestivum and Zea mays.

- Total chlorophyll content reduced in plants exposed in field and artificially to  $\text{SO}_2$ , flyash and a combination of  $\text{SO}_2$  and flyash, but with an exception that in plants sprayed with flyash for 45 days, a marginal increase in total chlorophyll content was noticed.

- In epidermal features, stomatal density and length and breadth of stomatal pore reduced in plants exposed in field and artificially to  $\text{SO}_2$ , flyash and a combination of  $\text{SO}_2$  and flyash for 90 days. Trichome density and trichome length were found to increase in Triticum aestivum plants exposed in field and artificially to  $\text{SO}_2$ , flyash and a combination of  $\text{SO}_2$  and flyash for 90 days.

- Under short term exposure, decrease in diffusive resistance was noticed in plants exposed in field (24 h) and artificially to  $\text{SO}_2$  (1 h) and a combination of  $\text{SO}_2$  (1 h) and flyash, at their 45th and 90th day of age (after seedling).

- Diffusive resistance increased in plants exposed under long term for 45 and 90 days in field and artificially to  $\text{SO}_2$ , flyash and a combination of  $\text{SO}_2$  and flyash.

- Leaf surface temperature increased in plants sprayed with flyash for 45 and 90 days, while it decreased in plants exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash for 45 and 90 days. The sequence of change including control can be written as follows:

Flyash > Control > Field > SO<sub>2</sub> + Flyash > SO<sub>2</sub>

- Leaf absorbance was more in plants sprayed with flyash for 45 and 90 days, whereas it was less in plants exposed in field and artificially to SO<sub>2</sub> and a combination of SO<sub>2</sub> and flyash for 45 and 90 days. The sequence of change in leaf absorbance was same as noticed for leaf temperature.

The present research work represents and attempts a comprehensive study of leaf characteristics in relation to pollution stress. In this many new aspects have been examined, such as optical characteristics, diffusive resistance under long term exposure and epidermal characteristics under artificial exposure to SO<sub>2</sub> and flyash and a combination of SO<sub>2</sub> and flyash.

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