

**Chapter 4**

**DISCUSSION**

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As mentioned earlier the objectives of the present investigation are :

1. To see the impact of air pollution on the distribution and diversity of herb and shrub species.
2. To find out the differential response of tree species growing near industrial area.
3. Exposing potted tree saplings to different levels of ambient air pollutants, and
4. Usefulness of some reducing agents as mitigators of pollutant injury.

All the results obtained are discussed under separate subheadings along with pertinent literature for supporting the observations. The salient feature of the present study is that conclusions are drawn taking a holistic approach. While inter-relating the data some of the inferences are repeated as the omission could not convey the message effectively.

#### **4.1 AMBIENT AIR QUALITY**

Pollutant concentrations in the ambient air are influenced by many factors and always vary from one locality to another. It is reported that high pollutant concentrations which cause acute injury, usually occur during production processes, accidents at the pollution source, or during unfavourable weather conditions. As a result of increasing density of industrial and domestic areas and traffic the pollution situation is characterized by persistent effects of low ambient concentrations. Plants have little or no chance for detoxifying absorbed pollutants due to lack of sufficient pollution free-times and/or due to the accumulatory effect in plant organs and soil (Guderian, 1977). Ormrod (1982) observed

that it is unlikely that steady concentrations of mixed pollutants will be found out-doors. In the natural environment peak concentrations may occur at different times for different pollutants. Such patterns may have considerable impact, with the preconditioning of plants by one pollutant affecting their response to a later peak concentration of another. The amount of injury in the field may be underestimated because of the fluctuation of air pollution concentration in the field with upto ten times the daily mean occurring occasionally in industrial areas.

Fluctuating concentrations of pollutants may cause more stress than steady amounts, to which a plant may easily adapt at the cellular and physiological level (Wellburn et al., 1976). Jacobson & Mc Manus (1985) reported that episodic emissions of  $\text{SO}_2$  should be taken for consideration in vegetation effects assessment. As compared to single pollutant response, plants response to mixture of pollutants is altogether different (Amundson et al., 1982; Elkley & Ormrod, 1981<sup>a</sup>; Hofstra et al., 1985; Irving, 1983). Hence in field conditions a mixture of pollutants with wide ranging concentrations is a common phenomenon due to the heterogeneity of industries, augmented with mobile sources of pollution. These fluctuating concentrations play a major role in influencing the surrounding vegetation at physiological level which ultimately affects the plant productivity.

Data on the ambient air quality given in the results clearly indicates the wide range of concentrations of different pollutants. The peak concentrations of different pollutants are the maxima recorded at different localities. These concentrations adversely affect the vegetation growing in the vicinity as already concluded in the above reports. The fluctuating concentrations, broad range of pollutant types and variations in seasonal pattern and conditions create continuous stress on the plants. The intermittent fluctuations

augments the damage. As the monitoring was not continuous, some of the peak concentrations might have been missed. Continuous monitoring could not be carried out because of meagre facilities available and scarcity of funds. Hence, while assessing the impact of air pollution on plant growth these missing concentrations were kept in mind while interpreting the data.

As the industries cover an expansive spectrum, pollutant types are quite varied. Apparently there are so many sources of emissions that periods with elevated pollutant concentrations are outspread. The National Highway No.8 with heavy vehicular traffic also passes through the study area effecting the nearby vegetation (Krishnayya & Bedi, 1986<sup>a</sup>).  $\text{SO}_2$ ,  $\text{NO}_x$  and SPM were chosen for monitoring because of their prevalence in the ambient air, major pollutants influencing vegetation and also for their simple estimations with minimum requirements for equipment and chemicals. These mixtures of pollutants in the presence of sunlight can lead to the formation of secondary pollutants which acts as an auxiliary source for the atmospheric contamination. This heterogeneity of pollutants increased the complexity of air thereby making the plant exposed to stress conditions. Hence, the damage reported in this study should be taken as an effect of all the existing mixtures of pollutants.

In ambient air acute exposures are usually associated with isolated point sources or several point sources that are close together. Chronic exposures, usually associated with multiple or regional sources are more pervasive and are more likely to effect growth and yield without showing obvious symptoms of injury (Heck, 1982). Exposure to  $\text{NO}_x$  in combination with  $\text{SO}_2$  and or  $\text{O}_3$  can alter plant metabolism and productivity at concentrations that do not produce such effects if the pollutants were present alone (Amundson & Maclean NY 1985<sup>3</sup>). Hence, the implications of synergism and antagonism must be

viewed in terms of pollutant concentrations and durations. Dassler & Bortitz (1988) summarised that as per the recent reports from the UN economic and social council the emission threshold values for the protection of sensitive plant species vary depending on the mixture of pollutants and, the greater the number of pollutants taken, threshold concentrations are the lesser for each pollutant. Hence, as the complexity increases the threshold value decreases. All these reports are in conformity with the observations of this study revealing that in ambient air conditions, low concentrations of mixtures of pollutants play a major role.

The order of sensitivity of plant populations to  $\text{SO}_2$  depend primarily on the concentration and duration of exposure (Garsed & Rutter, 1982). Mudd (1982) observed that in most experiments carried out in areas exposed to industrial waste, the degree of pollution is expressed in terms of  $\text{SO}_2$  concentration because it is the main pollutant. Yet in most instances it is accompanied by other pollutants and the field environments are invariably a complexes of mixed pollutant situations at least for some time. So additive effects must be reckoned with. Freer-Smith (1984) concluded that mixture of  $\text{SO}_2$  and  $\text{NO}_2$  clearly represent a potential stress to amenity trees in urban and industrial areas. The response of plants to  $\text{SO}_2$  and  $\text{NO}_2$  are variably depending on species and with continued exposure, can change in kind and extent. In the present study more than additive effects have been taken into account. Correlations have been made only with accumulation of sulphur and ambient  $\text{SO}_2$  concentrations which were found significant. This is because of the major role of  $\text{SO}_2$  individually as well as in synergism at the recorded concentrations.

#### 4.2 GENERAL VEGETATION SURVEY

Results of the general vegetation survey clearly showed the influence of air pollution on plant distribution and species diversity. Both windward and leeward directions showed marked changes in number and abundance of plant species. Close to the source of pollution ( $< 1.0$  Km.) the influence of air pollution in both the directions was almost the same showing a very sparse distribution of species. The minimum area required for a quadrat was very high ( $> 10\text{ m} \times 10\text{ m}$ ). This clearly indicates that the influence of air pollution close to the source of pollution is almost uniform in all directions. Farther than 1.25 km, species distribution was higher on leeward direction showing that plants respond differently to varying concentrations of air pollutants. Smaller concentrations of pollutants in the leeward direction made the plants grow more vigorously compared to the windward. Thus meteorological parameters are seen to be strongly influencing pollutant dispersion and in turn, the vegetation.

Treshow (1984) reported that absence of species is either due to less reproduction and/or inability to compete with other species. In a natural ecosystem the restriction of plant production by air pollutants may be sufficient to reduce the ability of the affected plants to compete successfully. Mudd (1982) said that in forests and woodlands inhibition of seed production diminishes the capacity for natural regeneration and causes in time the disappearance of some species unable to reproduce vegetally. Similarly pollution induced reductions in root growth are likely to have important consequences in perennials such as grasses in which regrowth after cutting and grazing is dependent upon the reserve assimilates stored in the roots (Koziol, 1984). Malhotra & Blauel (1977) concluded that response of vegetation to aerial emissions of  $\text{SO}_2$  are known to range from no measurable response at all through changes in plant metabolism and conditions

of specific depletion. These reports indicate that air pollution affects plants at various physiological, morphological and reproductive levels influencing species distribution and diversity on any landscape.

The gradual reduction of plants in number and species diversity with reduction in distance from the source of pollution seen in this study is attributed to the less reproductive capacity and/or to their sensitivity. The transition point after which distribution of vegetation is almost normal was at a lesser distance on leeward (2.5 Km) as compared to windward direction (3.0 km). Similarly species which are sensitive to pollution fail to establish near the source as the pollution load decreases with increasing distance, the species diversity increases (Guderian & Kueppers, 1980). Vijayan & Bedi (1988<sup>a</sup>) observed increase in diversity and frequency of species while moving away from the source of pollution. Observations and conclusions of this study support all the findings mentioned above.

Higher sensitivity of mesophytes compared to xerophytic species was attributed to their regulated stomatal movement which reduces pollutant entry and minimises its effects. Wilson & Bell (1985) reported that SO<sub>2</sub> tolerant plants may be highly competitive in a sward in the presence of polluted air, but are less fit than SO<sub>2</sub> sensitive plants when grown in a cleaner air. In the present study also higher number of species seen in windward direction at > 2.5 Km. This difference is due to a greater distribution of species on leeward direction and hence elimination of species that cannot stand competition. The larger number of species in windward direction are likely to be due to their adaptation to the existing pollutant concentrations and/or to sparseness of the species resulting in less competition. These in part support the above finding.



### 4.3 TREE SPECIES SURVEY

#### 4.3.1 Growth

Air quality at different localities showed a major impact on the growth of tree species and on their reproductive cycle. The damage was proportional to the concentrations recorded and at localities 3-5 the damage was aggravated due to the wide range of pollutants. Maximum damage in all the three tree species was seen at locality 5 and minimum at locality 1 having maximum and minimum pollutant concentrations respectively. Thus the response of tree populations was mainly dependent on kind/type of pollutant/s and their concentrations. Oleksy & Bialobok (1986) also reported that individual populations from different countries showed statistically significant differences in the extent of needle injury. Deciduous species were more effected compared to evergreen ones and, of the two deciduous species Azadirachta was more effected.

Reduction in the plant productivity is mainly due to the loss of leaf area and premature leaf fall caused by air pollutants. There are many reports on the foliar injury due to gaseous pollutants. Leaf fall was accelerated in conifers exposed to  $\text{SO}_2$  (Garsed & Rutter, 1982). Soybean plants exposed to  $\text{SO}_2$  showed accelerated senescence (Irving & Miller, 1981).  $\text{SO}_2$  increased injury to wheat plants (Bytnerowicz *et al.*, 1987<sup>b</sup>). Reduction in leaf area and increased leaf area damage were reported in paddy plants exposed to air pollutants which ultimately reflected in the reduction of yield (Ayer & Bedi, 1986; Sisodia & Bedi, 1986). Similar findings were reported for potato crop grown in the present study area (Pandya & Bedi, 1986). Parry & Whittingham (1984) reported that air pollutants decreased plant productivity and this may be largely related to a decrease in photosynthetic activity. Similar observation was reported with ozone which showed a linear decrease in

biomass of soybeans (Grunwald & Endress, 1985). Stan et al., (1981) reported increased ethylene evolution by plants due to air pollutants. Bucher (1978, 1981, 1984) also concluded that  $\text{SO}_2$  exposure induced increased ethylene evolution in the foliage of forest trees and produced various pollution symptoms such as epinasty, premature senescence or leaf abscission. This type of accelerated leaf aging due to ozone exposure was atleast partially responsible for declining net photosynthetic capacity (Reich, 1983). In the present study increased leaf fall and leaf area damage observed due to air pollutants had diminished plant productivity which was ultimately reflected in plant growth. Similar conclusion was drawn by Chaphekar et al., (1980).

Ozone induced reductions in photosynthesis were related to decline in growth (Reich & Amundson, 1985). The biomass production was most negatively influenced in Silene cucubalus plants exposed to  $\text{SO}_2 + \text{O}_3$  (Dueck et al., 1986). Reductions in net photosynthesis were found to be proportional to the pollutant concentration (Darrall & Jager, 1984). Baker et al., (1987) showed that  $\text{SO}_2$  (80 - 200 ppb) may have decreased assimilate production in winter barley. In the present investigation leaf fall and leaf area damaged were more in deciduous species and leaf fall was often aggravated due to premature senescence. Lower reduction in Tamarindus revealed its tolerance to air pollution. Greater reduction in the height and girth of the two deciduous species was attributed to less photosynthate production owing to higher leaf damage as quoted above. Addison et al., (1984) also reported that fumigation with  $\text{SO}_2$  (0.34 ppm) significantly reduced visible symptoms of injury. The decrease in NAR of deciduous species was significantly more rapid than of conifers or an evergreen angiosperm. Results and observations of this study are in conformity with this finding. Lesser reduction in the height and girth of Tamarindus was due to less effect on foliage which maintained biomass production thus lessening the pollution damage.

#### 4.3.2 Reproductive cycle

Air pollutants have a major influence on the reproductive cycle of plants. The effect starts from the inception of flowering. Flower mortality and fertilization are very much influenced. Ernst *et al.*, (1985) showed that flowering in Silene cucubalus was completely hampered by  $O_3$  alone or in combination with  $SO_2$ . Similarly flower mortality was 89% for blueberry and 78% for raspberry and this accounted for most of the loss in the reproductive potential (Staniforth & Sidhu, 1984). In the present investigation reproductive cycle of the three tree species was adversely effected by air pollutants. At reference, profuse flowering was seen in Moringa. Of the two deciduous species Azadirachta was more damaged. Flowering was highly influenced and a sudden spurt in the fall was seen whenever higher peak concentrations of pollutants occurred. Loss in flowering was also high in Moringa. This may be due to the excess burden on inflorescence stock owing to greater number of flowers. Reported ethylene evolution from plants under stress conditions was also responsible.

Various explanations have been given for the loss of fruit yield. Mudd (1982) reported that repeated sampling of pollen treated with different levels of pollution revealed that in trees from polluted zones (Pinus sylvestris) the average size of pollen grains was significantly reduced in comparison with those from unpolluted area. Similarly, changes in the quality of pollen wall proteins was seen when pollen grains were contaminated with gaseous pollutants (Ruffin *et al.*, 1983). Krishnayya & Bedi (1986<sup>a</sup>) concluded that plants growing nearer to the National Highway showed reduced pollen germination and pollen tube growth. Wolters & Martens (1987) also reported that pollen germination and pollen tube growth are affected by air pollutants, both *in vivo* and *in vitro* in concentrations that do occur in ambient air. Fertilization of Silene cucubalus plants exposed to  $SO_2 + O_3$  was strongly inhibited (Ernst *et al.*,

1985). These reports clearly indicate the influence of air pollution on fertilization process. Loss in fruit production observed in this study was attributed to the failure of pollen germination and/or to the non-receptivity of the stigma. As percentage fruiting was less in deciduous species as compared to evergreen ones, it was concluded that these processes were more hampered in deciduous species.

Reduction in the yield of plants exposed to gaseous pollutants is a common phenomenon. Irving & Miller (1984) showed that synergism of both  $\text{SO}_2$  and  $\text{NO}_2$  reduced yield by 9 - 25% depending on pollutant dosage. Similar type of decrease in fruit production was seen in both blueberry and raspberry by Staniforth and Sidhu (1984).  $\text{SO}_2$  exposure reduced the yield of wheat plants (Bytnerowicz et al., 1987<sup>b</sup>). Reduction in yield were seen in barley when exposed to fluctuating concentrations of  $\text{SO}_2$  (Baker et al., 1986). Reduction in biomass and yield of fruits in tomato plants grown in the industrial area at Baroda were seen (Bell & Bedi, 1985). Mudd (1982) reported that fluorides and  $\text{SO}_2$  show a direct mode of action on the process of fruiting, they result in a loss of seed yield and in the alteration of fruit quality. Reduction in yield due to air pollutants was reported by Heagle et al., (1983<sup>a</sup>) and Kats et al., (1985). Vijayan & Bedi (1989<sup>a</sup>) reported reduction in the fruit yield of mango, rayan and jamun trees growing at Ranoli, near Baroda. All these support the results and conclusions of this study.

Accelerated senescence as suggested by increased leaf fall may be responsible for the decreased yield in the  $\text{SO}_2$ -exposed soybean plants (Irving & Miller, 1981). In this study also higher reduction in the yield of two deciduous species was attributed to their greater leaf fall and foliar damage. Thompson et al., (1976<sup>b</sup>) showed that reduced yield in alfalfa paralleled increases in concentration of oxidants in the atmosphere. Here also as the atmospheric concentrations

of pollutants increased from one locality to another, reduction in the yield was also increased. Thus higher the pollutant concentration, greater was the yield reduction. Premature fruit fall was maximum in Azadirachta. Though fruit formation was less in Moringa due to high flower mortality and/or to less pollen germination, the retention of fruits produced was more. In Tamarindus both fruit production and retention were comparatively more. This is attributed to its better tolerance and to less damage to flowering. Greater fruit retention in Moringa and Tamarindus was due to the hard, woody stalk of the fruit.

#### 4.3.3 Biochemical parameters

##### Chlorophylls

Chlorophyll is one of the main factors which influence the rate of photosynthesis and plant productivity. Sestak et al., (1971) reported that amount of chlorophyll a, its distribution and amounts of accessory pigments affect the amount of radiant energy fixation. Pollutants damage chlorophyll pigments which hampers photosynthesis and plant productivity. In plants exposed to gaseous pollutants reduction in foliar chlorophyll content was seen (Beckerson & Hofstra, 1979<sup>b</sup>; Krishnayya & Bedi, 1986<sup>b</sup>; Pawar & Dubey, 1983, 1985; Pratt et al., 1983), and these reductions lead to a decrease in net photosynthesis (Reich, 1983; Reich & Lassoie, 1985). Chlorophyll a was more sensitive to SO<sub>2</sub> than chlorophyll b (Malhotra, 1977). Krishnamurthy and Rajachidambaram (1986) reported that the reduction in chlorophyll content was due to its reduced biosynthesis. SO<sub>2</sub> fumigated plants had a lower chlorophyll (Grunwald, 1981). SO<sub>2</sub> degrades chlorophyll molecule to phaeophytin Mg<sup>2+</sup> (Treshow, 1984). Decrease in total chlorophyll content (40%) was reported in young bean foliage exposed to acid rain which reduced photosynthetic competency (Chia et al., 1984). Similarly, O<sub>3</sub> bubbled into a suspension

of isolated chloroplasts inhibited electron transport in both photosystems I & II (Christopher & Heath, 1974). In this investigation chlorophyll content was higher in Tamarindus and reduction was less. Fluctuations in chlorophyll content were more in the two deciduous species from one season to another. Chlorophyll a showed higher reduction compared to chlorophyll b. These reductions were attributed to degradation and/or less synthesis of chlorophyll as reported in the above findings. Higher reduction in Azadirachta and Moringa showed their sensitivity to air pollutants as reported in sensitive subterranean clover (Murray, 1984). Reduction in chlorophylls reduced plant's capacity in harvesting the sun light. Higher chlorophyll content in Tamarindus increased photosynthetic rate thereby enhancing carbohydrate levels. Carlson (1983<sup>a</sup>) reported that high levels of carbohydrates generated due to high CO<sub>2</sub> might be available for repair processes or to provide additional carbon skeletons for combination with, and therefore detoxification of SO<sub>2</sub>. Resistance of Tamarindus is attributable to a similar phenomenon. Greater reductions of chlorophyll content in the two deciduous species clearly affected the plant productivity which retarded height and CBH of the trunk.

Various postulations were made for the pollutant damage on chlorophylls and its impact on plant metabolic system. Sakaki et al., (1983) reported that spinach exposed to O<sub>3</sub> in light, destruction of chlorophylls and carotenoids were seen. Superoxide radical participated in the destruction of pigments. Similarly free radical induced chlorophyll destruction was also reported (Kato & Shimizu, 1985). Inhibition of photosynthesis (Ho & Trappe, 1984; Rao et al., 1983; Mukerji & Yang, 1974) and ATP production (Ballantyne, 1973) were reported in plants treated with pollutants. Sakaki & Kondo (1985) reported that intracellular sulphite accumulated in the protoplasts in an unmetabolized state is responsible for the inhibition of protoplast photosynthesis. Sulphite and sulphate inhibited RUBISCO but sulphite was more inhibitory than sulphate

(Khan & Malhotra, 1982<sup>b</sup>). Thus pollutant effects chlorophylls and other accessory pigments, and also various enzymes involved in CO<sub>2</sub> fixation, leading to less photosynthesis and reduction in plant productivity. Reductions in chlorophylls and various growth parameters seen in this study are in conformity with the above findings.

### Total Proteins

Proteins are major components of biological membranes and play an important role in various metabolic processes. Decrease in protein content was noticed in plants exposed to SO<sub>2</sub> (Constantinidou & Kozłowski, 1979). Significant decrease in protein content of soybean and peas were noticed due to SO<sub>2</sub> by Skarby (1984). Fumigation of pine seedlings with SO<sub>2</sub> markedly inhibited de novo biosynthesis of proteins in chloroplasts and cytoplasmic fractions and the magnitude of inhibition was dependent on exposure time (Khan & Malhotra, 1983). Treshow (1984) reported that besides inhibiting biosynthesis, pollutants (SO<sub>2</sub> and O<sub>3</sub>) may breakdown existing proteins and increase free amino acid content. Hence, the reductions observed in this study were either due to less synthesis or to the greater breakdown of existing proteins. Higher reductions in deciduous species indicate that these processes were more effected in those two species. Because of these reductions in the availability of proteins various metabolic functions have been effected which lead to the reduction of plant growth. Khan & Malhotra (1983) also concluded that reduction in protein biosynthesis would affect their availability as a structural component of new membranes. Less reductions in Tamarindus revealed that protein turnover rate was comparatively better. Ulrich (1984) reported that higher turnover rate of proteins gives resistance to plants against pollution stress. Resistance of Tamarindus was attributed to a similar phenomenon.

### Ascorbic acid

Ascorbic acid is an antioxidant. It appears to act as a free radical scavenger and protects plants against lipid peroxidation and leaf damage. It may also act in concert with superoxide dismutase to protect the plant tissues against  $O_3$  - induced stress (Lee *et al.*, 1984). In this study reductions in ascorbic acid content were seen in the two deciduous species while Tamarindus showed an increase. Greater content in the evergreen species gives more efficiency in acting as a free radical scavenger which increases its tolerance. Varshney & Varshney (1984) reported that species having high ascorbic acid content are more tolerant to air pollution. An analogous report by Lee *et al.*, (1984) showed that resistant cultivars of soybean and snapbean showed more ascorbic acid in their trifoliate leaves than corresponding susceptible genotypes. These are in consistent with the above findings.

### Glutathione

Glutathione is the most abundant non protein thiol and it reduces dehydro ascorbic acid to ascorbic acid (Jocelyn, 1972) thus recycling ascorbic acid in plant systems. Mapson (1959) reported that one of the main functions of glutathione in the cell is to protect sulphydryl groups in enzymes and structural proteins against oxidation to disulphides, either by acting as a scavenger for the oxidising agents or by repairing oxidised sulphydryl groups via the glutathione disulphide exchange reaction. Here decrease in glutathione content was seen in the two deciduous species and Tamarindus showed an increase. Reductions in deciduous species lessens their capacity in protecting sulphydryl groups of enzymes and structural proteins which damage plant metabolic system, while in evergreen species the protection was enhanced due to the higher content. Rennenberg (1984) reported that an enhanced glutathione content observed upon fumigation with



SO<sub>2</sub> will not necessarily cause disturbance of the thiol status of the cells. It may function in cells of higher plants as a reservoir of reduced sulphur. Alscher *et al.*, (1987) reported that reduced glutathione in an insensitive cultivar of the pea increased during exposure to SO<sub>2</sub>, suggesting that it may protect the photosynthetic apparatus against SO<sub>2</sub> and functions to remove H<sub>2</sub>O<sub>2</sub> in chloroplasts formed due to their response to stress. In this investigation increased GSH content in Tamarindus revealed that it resists air pollution stress to a certain extent in a similar manner (detoxification of H<sub>2</sub>O<sub>2</sub>) and protects the plant from extensive damage. Decrease in GSH content of both the deciduous species made them susceptible.

#### **Total Sulphydryl groups**

Sulphydryl groups have a commendable role in plant metabolism. These are preferred free radical traps. Sulphydryl groups are required in photosynthetic phosphorylation. A SH dependence has been reported for the light dependent exchange of phosphate between ATP and ADP (Jocelyn, 1972). Distribution of the SH groups inside a plant is influenced by stress conditions. Both accumulation and reduction in its content were reported. Maas *et al.*, (1987<sup>a</sup>) showed accumulation of SH compounds in the plant shoot exposed to both H<sub>2</sub>S & SO<sub>2</sub>. An analogous finding by De Kok *et al.*, (1985) showed that after 1 h exposure to H<sub>2</sub>S a substantial increase in the SH content was already detectable and contrary to H<sub>2</sub>S, SO<sub>2</sub> fumigation did not result in a rapid accumulation of glutathione in spinach roots. Treshow (1984) reported a rapid decrease in ATP and sulphydryls following O<sub>3</sub> damage to the membranes. Some of the studies report the effects of fluctuations in thiol groups. Koziol (1984) concluded that oxidation of SH groups by photochemical oxidants seems sufficient to account for the loss of enzymatic activity. Similarly interaction of sulphite with either the membrane site of the generation of dithiol groups or the regulatory enzymes themselves could

result in an inhibition of light modulation process (Alscher, 1984). Thus fluctuations in SH groups adversely effects these processes and aggravates pollution damage to plants.

Total thiol groups showed a decreasing pattern in the polluted localities, except in Tamarindus, where the response was mixed. Reductions were more in the two deciduous species as compared to the evergreen species (Tamarindus) which also showed higher content in summer. Maintenance of thiol groups content in Tamarindus helps in lessening the inhibitory processes on light modulation, enzyme activities etc., increasing its resistance to air pollution. Rao et al., (1983) reported that  $\text{SO}_2$  inhibits the light dependent build-up of free thiols in the epidermis and whole leaves of pea plants. Wellburn et al., (1981) showed that low atmospheric levels of  $\text{SO}_2 + \text{NO}_2$  produces more than an additive inhibitory effect, leading to the deprivation of ATP production which affects the  $\text{CO}_2$  fixation and thiol formation. Reduction in thiol content observed in this study are attributable to similar effects by air pollutants. Takemoto et al., (1986) showed that the ability to form high thiols and emit more  $\text{H}_2\text{S}$  were important for sulphite tolerance in duckweeds. Evolution of  $\text{H}_2\text{S}$  from plants exposed to  $\text{SO}_2$  were reported (Garsed and Read, 1977; Silvius et al., 1976; Filner et al., 1984) and was seen as a protection mechanism (Staszewski, 1985). Higher production of thiols in Tamarindus exhibited that it also adapts a similar mechanism for its tolerance.

Thiol groups are preferred free radical traps (Jocelyn, 1972). Fluctuations in the thiol/disulphide status apparently cannot be tolerated by plant cells, because essential functions such as protein synthesis, catalytic activity of major proteins, detoxification phenomena etc., are extremely sensitive to even minor changes in the thiol concentration. Therefore possession of a mechanism for the precise regulation of the cell thiol content will be essential for the survival of a

plant in a continually changing environment (Rennenberg, 1984). Chevrier et al., (1988) reported that regeneration of SH groups by Euglena cells is a part of mechanism involved in the repair of oxidative damage caused by ozone and is an essential step for the initiation of cell division. These clearly indicate the importance of thiol group regulation. Greater reductions in the thiol content of deciduous species indicated that the pollution damage is aggravated which lead to the damage of various processes mentioned above. This reflected in the growth reduction of the tree species.

### **Acid Phosphatase**

It has implication in key metabolic functions such as transportation of stored metabolites (Flinn & Smith, 1967), stomatal opening (Mishra & Panda, 1970) and release of potassium, magnesium and phosphate ions from the phytin complex (Bewley & Black, 1978). Malhotra & Khan (1980) reported that SO<sub>2</sub> inhibits acid phosphatase activity in pine seedlings and this inhibition could create cellular imbalance in the concentration of various phosphorylated intermediates. Higher reductions in Azadirachta and Moringa seen in this study adversely effects all the above mentioned processes leading to injury. Greater increase in Tamarindus compared to Azadirachta in winter and smaller inhibition of acid phosphatase activity showed that it can withstand pollution stress to a certain extent.

### **Peroxidase**

Peroxidase is very sensitive to any stress received by plants and is widely used as a marker for studying stress. Its quantity often increases under stress. Endress et al., (1980) reported that peroxidase levels may be elevated by subjection to air pollution stress and its condition is sensitive to internal physiological condition of the plants.

Khan & Malhotra (1982<sup>a</sup>) suggested that stimulation of peroxidase appears to be due to increase in the production of isoenzymes rather than their activation. The stimulation of peroxidase activity by SO<sub>2</sub> fumigation may be a cellular defence mechanism to eliminate toxic levels of H<sub>2</sub>O<sub>2</sub>. Of the two species they studied (Jack Pine & White Birch), White Birch appeared to be more sensitive as shown by marked increase in peroxidase content. Varshney & Varshney (1985) reported that of the three species Zea mays, Brassica nigra and Phaseolus radiatus taken, control plants of Zea mays exhibited higher peroxidase activity compared to the other two species and postulated that it provides relatively high resistance against SO<sub>2</sub> toxicity. Jager et al., (1985) also concluded that higher peroxidase content in resistant pea cultivars is an indication of a protective mechanism in tolerant plant species. Here increase in peroxidase content as a result of air pollution stress was seen in all the three species studied. Higher content in Tamarindus at all the levels as compared to the deciduous species indicates that the enhanced levels protects against air pollution stress, helping in detoxifying excess levels of H<sub>2</sub>O<sub>2</sub>. Manifold increase in deciduous species revealed their sensitivity to air pollutants. The results and conclusions of this study are in conformity with the above findings.

#### **Foliar sulphur content**

Accumulation of pollutants in plant tissues and their ability in the sorption of pollutants is well documented (Jensen & Jager, 1983; Maas et al., 1987<sup>C</sup>; Milchunas et al., 1983; Pratt et al., 1983; Roberts, 1974). Uptake and accumulation of gaseous pollutants by plants depends on the needs and developmental stage of the plant. These vary from species to species and are influenced by various environmental factors. Hallagren et al., (1982) reported marked diurnal variation in the uptake of SO<sub>2</sub>. Anderson & Mansfield (1979) attributed

the differential response to  $\text{NO}_2$  to the differences in nitrite reduction capabilities of the varieties and their ability to utilise  $\text{NO}_2$  as an atmospheric source of nitrogen. In a similar way Rennenberg (1984) concluded that sulphate reduction in the intact plant takes place predominantly in the leaves and uptake of  $\text{SO}_2$  by leaves can be adjusted to some extent to the plant's needs for sulphur although uptake of injurious amounts cannot be avoided. Olszyk *et al.*, (1987) observed increased total sulphur concentration in leaves with increasing  $\text{SO}_2$  concentration. Plants may be more sensitive in the autumn either because the same environmental conditions that encourage growth also encourage pollutant uptake or because actively growing plants have more air pollutant sensitive tissue. In this investigation increase in foliar sulphur content was seen as the pollutant concentration and age of the leaf increased, in all the three species. Differential response of the species was attributed to their differences in pollutant uptake and its reduction as stated in the above findings.

Roberts and Schnipke (1983) reported that plants from high ambient  $\text{SO}_2$  location contained greater quantities of sulphur than from low ambient  $\text{SO}_2$  site, foliage being the most efficient sorptive tissue. Lauenroth *et al.*, (1979) observed that sulphur accumulation measured within the plant will be determined by concentration of  $\text{SO}_2$ , duration of exposure and is linearly related to treatment concentration. Similarly tolerant plants appreciably showed more sulphur content (Roberts, 1976). Roberts *et al.*, (1986) reported that trees have considerable potential to filter gaseous pollutants from the atmosphere. Similar observations were seen in this study where in linearity in the sulphur accumulation was seen against concentration, normal higher uptake showed that tree species acted as scavengers for pollutants and greater uptake by ever-green species showed that its resistance makes it to absorb higher amounts of  $\text{SO}_2$  as compared to Azadirachta & Moringa.

#### 4.3A OVERALL IMPACT OF AIR POLLUTION ON THE BIOCHEMICAL PARAMETERS STUDIED

Chlorophylls, total proteins, ascorbic acid, glutathione, thiol groups, peroxidase and superoxide dismutase (SOD) are very widely studied in plants against air pollution and other stress conditions. Various mechanisms and postulations were made in depicting their role in plant metabolism inducing tolerance or resistance against gaseous pollutants. A brief review of some of these published reports is given here to draw conclusions for the resistance of plants supporting the observations and inferences of this study.

Schulz (1986) reported that results of SOD, peroxidase, catalase, Chlorophyll *a* and proteins analysed showed that this effective model is sufficient for the interpretation of SO<sub>2</sub> damage. By measuring these it may be possible to estimate the environmental pollution in the atmosphere. Sakaki *et al.*, (1983) observed reduced endogenous levels of O<sub>2</sub><sup>•-</sup> scavengers such as SOD and L-ascorbate in leaves exposed to ozone to one-half. Damage to leaves was as a result of the O<sub>3</sub>-induced destruction of physiological defence against oxygen toxicity. Similarly higher production of ions increases visible injury which appears later. Evidence suggests that membrane lipids are affected by SO<sub>2</sub>. Free radical formation was enhanced during exposure to atmospheric pollution and plants with poorer natural free radical scavenging mechanisms showed sensitivity to pollution earlier, as their membrane components were damaged (Wellburn, 1985).

Treshow (1984) concluded that the free radicals produced can oxidise various cellular metabolites and affect a number of membrane constituents such as SH groups, amino acids, proteins and unsaturated fatty acids. The excess production of free radicals due to pollution stress attack thylakoid membranes or phosphorylation processes. A similar observation

by Dominy & Heath (1985) showed that  $O_3$ -induced plasma membrane permeability changes may be effected by damage to membrane proteins, perhaps by oxidation of amino acid sulphydryl groups to disulphide and sulphenic moieties. High levels of antioxidants prevents this damage which allows stomatal closure thereby preventing  $O_3$  entry into foliar tissues. In plants where adequate levels of antioxidants are not present this response will be sluggish or completely abolished resulting in the exposure of mesophyll cells to ozone. Kato & Shimizu (1987) revealed that defence mechanism against peroxidative chlorophyll degradation (combination of ascorbate, reduced glutathione, glutathione reductase and NADPH) was less active in senescent leaves than in young leaves. Yu Shu-Wen *et al.*, (1982) concluded that when free radical scavengers were used  $SO_2$  injury was less, showing that free radicals do take part in the process of injury.

Foyer and Halliwell (1976) suggested that the cycle constituted by glutathione and ascorbate plays a scavenger role in removing  $H_2O_2$ . Tanaka *et al.*, (1985) concluded that  $O_3$ -tolerant cultivars of spinach plants were found to have high ascorbate and glutathione levels than the other cultivars and ascorbate may participate in the detoxification of  $O_3$ . Mejnartowicz (1984) showed that trees characterized by considerable resistance to the action of  $SO_2$  normally have a high peroxidase activity. Similar findings were reported by Edward *et al.*, (1984). Some plants seemed to react to  $O_3$  by producing increasing amounts of reducing agent ascorbic acid (Skarby, 1984). Similarly  $O_3$  exposure increased ascorbic acid levels in *Sedum album* plants (Castillo & Greppin, 1988). Mudd *et al.*, (1984) concluded that high SOD content in leaves is correlated with resistance to  $SO_2$  damage. Tanaka *et al.*, (1988) said that  $SO_2$  toxicity in plants is caused by active oxygens and that superoxide dismutase participates in counteracting  $SO_2$  toxicity. Bennett *et al.*, (1984) observed

that SOD and the catalase peroxidase enzyme systems serve as interlinked primary protection mechanisms in reducing the potential for cellular injury due to oxyradicals. Common metabolic reductants such as  $\alpha$ -tocopherol and ascorbic acid would further minimise damage due to oxidants present inspite of these protective enzyme systems. Alscher (1984) reported that rate of regeneration of thiol groups, efficient reductive detoxification pathway and efficient photoscavenging system for  $H_2O_2$  are necessary for the removal of sulphite. Longer the residence time of sulphite within the cell in general, and within the chloroplast in particular, the more likely is disturbances in systems such as of light modulation will occur.

Chloroplast metabolism : Bases for  $SO_2$  tolerance :  
Proposed by Alscher (1984) :

1. Rate of regeneration of membrane dithiol groups
2. Rate of photoscavenging of  $H_2O_2$  - SOD, glutathione, ascorbic acid
3. Activity (or inducibility) of sulphite reductase.

Bennett et al., (1984) reported that starch and sugars in carbohydrate - loaded chloroplasts might mitigate injury induced by oxyradicals.

In this study a wide range of parameters were taken to have a broader spectrum in assessing the plant's response to air pollution. As stated in the above citations it is often the free radicals generated due to pollution stress affect the plant to the maximum. Free radical scavengers and antioxidants such as ascorbic acid, glutathione, peroxidase and SOD play a major role in detoxification and generation of these will give a better chance for the survival of plants



in a polluted environment. Chlorophylls play a major role as majority of the above mentioned processes take place in chloroplasts during photosynthesis. Plants having greater amounts of chlorophylls regulate the excess of free radicals generated due to pollution stress and greater amounts of photosynthates produced also minimises the damage. Generation of SH groups including glutathione helps the plant in decreasing the subtle changes. Higher amounts of chlorophylls, ascorbic acid, peroxidase, glutathione and balancing the thiol groups in Tamarindus have given resistance in minimising the pollutant effect on plant growth. Less reductions in growth parameters serve as evidence for this conclusion.

Response of the two deciduous species investigated was negative. Heavy reductions were seen in the quantity of above mentioned metabolites (excepting peroxidase) and though at some points positive response was seen (e.g. acid phosphatase activity in Azadirachta), it could not counteract the damage to a greater extent. This was evident by the higher reductions in growth parameters.

As Azadirachta and Moringa could not generate good amount of free radical scavengers, sensitivity of the species increased, premature leaf fall was aggravated which also hampered the plant productivity. These in effect reflected in the loss of flower and fruit production.

As stated earlier, in field conditions the influence of pollutants is inter-dependent and is often additive. These exert pressure on plant metabolic system and once it is exposed to a pollutant its response to the other pollutant/s varies and in general, plant becomes more susceptible. Beckerson & Hofstra (1979<sup>a</sup>) reported a similar phenomenon. Rennenberg (1984) concluded that plant's response to SO<sub>2</sub> appears to be modified not only by the dosage of SO<sub>2</sub>, but also by physiological

and environmental factors. The threshold may vary considerably between different plant species and even different varieties of the same species and at times within same variety itself. Responses may be modified by the species or varieties of plants, the concentration, duration and frequency of exposure to the pollutant, the degree of pollutant accumulation within sensitive tissues, plant age, nutritional status and prevailing environmental conditions. Similarly the "more-than-additive" effect of low atmospheric concentration of  $\text{SO}_2$  and  $\text{NO}_2$  may be explained partly by failure to induce additional nitrate reductase activity and partly by the combined effects of sulphite and nitrite. These may act by the intermediate formation of free radicals, damaging the photosynthetic membrane and preventing sufficient proton gradients, which would have allowed extra ATP to be formed which is required for protein biosynthesis, thiol formation and for the provision of phosphorylated sugars for export. The results indicate that overall ATP level may be a critical limiting factor. Thus multiple biochemical factors are involved in pollutant injury (Wellburn, 1982). In this way variations in types and concentrations of pollutants influences the plant.

In the present investigation response of each species was different and varying pollutant concentrations have showed a wide range of impact on plants. In the study area pollutants were of a broad spectrum and had shown cumulative effect on the three investigated species which is evident from the high reductions seen in various parameters. Besides, these occasional peaks aggravated the damage and deciduous species were more influenced. Similar type of heavy damage was seen in potted plant - exposure study where negative growth was seen in some seasons. These observations clearly indicate that the response of plant is very much influenced by the fluctuations in pollutant types/concentrations and meteorological conditions. Analysis of variance results clearly indicate

that behaviour of plants at different seasons was different for all the investigated biochemical parameters. Norby & Kozlowski (1981<sup>b</sup>, 1982) reported that changes in temperatures and humidity affect the plant adversely under SO<sub>2</sub> exposure. In this study also temperature and humidity fluctuations in the three seasons influenced plant's response to gaseous contaminants. Response of each parameter was different. Peroxidase was much influenced in monsoon, Chlorophylls were more affected in summer etc. Age of the leaf also influenced the response and it was more in deciduous species. In general upto middle age the response was less and afterwards the damage was aggravated. Processes which lead to senescence were speeded up. Because of the continuous exposure and due to changes in levels and types of pollutants the subtle changes were often irreversible. Higher amount of pollutant intermediates accumulated and excess of free radicals generated aggravated the damage which lead to the death of leaf. Variations in the species and wide range of parameters studied have given a better insight in understanding the impact of low concentrations of pollutants under field conditions.

#### 4.4 POTTED PLANT EXPOSURE STUDY

The objective of this study was to see the response of same aged species grown under different concentrations of ambient air pollutants. To minimise the impact of other environmental factors, care was taken in maintaining same edaphic, nutritional, cultural conditions and sapling variety. Hence the major influencing factor is the variable concentrations of pollutants at different localities. Seasonal variations were also noted. Among the three species studied, Moringa showed maximum growth followed by Azadirachta and Tamarindus. The recovery rate when the pollutant concentrations were minimal was also maximum in Moringa.

#### 4.4.1 Growth Parameters

In the parameters studied, number of leaves and leaf area were maximum affected which ultimately reflected as a whole in the reduction of plant growth. Results of these parameters showed some peculiarity. Moringa and Tamarindus showed similar pattern of results while of Azadirachta's were different. In Azadirachta correlations against pollutant concentration were significant and definite growth at decreasing rate with increasing concentration can be seen. In the other two species fluctuations were wide ranging without any correlation. This is because of the pollutant induced sudden rise and fall in leaf production and also to sudden leaf fall. Thus negative growth was seen for these parameters in Moringa and Tamarindus. There are many reports on the effect of air pollutants on plant foliage, subsequently affecting plant growth and biomass accumulation. Reduction in number of leaves and other plant parts was attributable to a decrease in their formation which decreases plant growth. Ormrod (1982) showed that decrease in leaf production was due to less supply of assimilates. Reich & Lassoie (1985) observed that ozone increased leaf senescence and was linear to the concentration of  $O_3$ . Increased defoliation triggered leaf production. But this did not makeup the difference with control. Similarly greater defoliation had large impact on plant growth (Lauenroth et al., 1985). These reports clearly indicate that the damage caused to foliage by air pollutants does effect plant productivity and it is proportional to pollutant concentration. Defoliation of plants produced higher number of leaves in the follow-up period; but this spurt in production was often less than that of the reference. These fluctuations drastically affected plant growth. The results and conclusions of this study support the above citations.

Many investigators reported reduction in growth of

the plants exposed to pollutants (Koziol *et al.*, 1986; Marie & Ormrod, 1984; Thompson *et al.*, 1976<sup>a</sup>). Plants exposed to ozone had shown linear decline in growth (Amundson *et al.*, 1987). Ayazloo *et al.*, (1980) reported that SO<sub>2</sub> depressed growth in all treatments without any visible symptoms on the foliage. Exposure of spinach to H<sub>2</sub>S reduced the relative growth rate (Maas *et al.*, 1987<sup>a</sup>) and populations of Silene cucubalus were heavily affected in growth by ozone (Ernst *et al.*, 1985). Thus the impact of gaseous contaminants ultimately reflects in the reduction of plant growth. In this investigation all the three species showed a definite growth at decreasing rate with increasing concentration of pollutants. Reductions in growth were of a wider range in Moringa and Tamarindus while Azadirachta always showed higher damage with lesser range. Results obtained support the above findings.

#### 4.4.2 Foliar sulphur content

Sulphur accumulation in the foliar tissues showed that Tamarindus takes more amount of SO<sub>2</sub> from the ambient air/unit leaf area, than Azadirachta and Moringa in the decreasing order. Roberts (1976) reported that tolerant clones showed appreciably more sulphur and foliage absorbs greater SO<sub>2</sub> from the atmosphere. Ernst *et al.*, (1985) also concluded that plant populations in polluted regions may have been selected for tolerance to air pollution to such a degree that the plant has a higher demand for the pollutant. The higher uptake of SO<sub>2</sub> and comparatively less growth reduction in Tamarindus shows that it is more tolerant to pollution stress. This is in conformity with the above findings.

### 4.5 ANATOMICAL OBSERVATIONS

#### 4.5.1 Epidermal Study

There are many reports on the impact of air pollution

on epidermal structure. Epidermal changes effects pollutant absorption by plants. Swieki et al., (1982) observed that mean surface wax on a leaf influences the cuticular resistance of Phaseolus vulgaris. Lendzian (1984) also concluded that waxes embedded within the cuticle form a continuous resistance. Soluble waxes of cuticle play a role in permeation process. Eveling (1986) reported collapse of epidermal cells and altered cuticular surface due to gaseous pollutants. Erosion of surface wax was enhanced, in trees growing in polluted areas and this has important consequences. Cuticle protects against desiccation and also impedes gaseous diffusion into and out of the leaf and thus represents the main barrier to the entry of pollutants. A similar finding by Huttunen & Soikkeli (1984) showed that most obvious effects of air pollutants is the erosion of epicuticular wax structures around the stomata of conifer needles. Pollutant enhanced erosion of the protective cuticle which can have important consequences. These reports clearly indicate the importance of cuticular wax on gas permeation processes and its response to pollutants. Of the three species taken Azadirachta did not show any wax distribution which revealed that it did not have any primary barrier for pollutant uptake. This made it more susceptible to air pollution. Higher amount of cuticular wax seen in Tamarindus revealed that the species had a better mode of protection for the pollutant entry, enhancing its resistivity. Moringa also showed cuticular wax but was comparatively less. Greater deterioration of surface wax in samples from locality 5 clearly showed that pollutants damage leaf surface architecture. Observations of this study are in conformity with above findings.

Stomata are the windows for gas exchange. Along with  $\text{CO}_2$ ,  $\text{O}_2$ , many other gases (inclusive of pollutants) enter into leaf. Metabolically active pollutants ( $\text{SO}_2$ ,  $\text{NO}_2$ ) interfere with plant metabolism and alters the normal processes. Thus entry of pollutants depends on stomatal opening and closing

to a certain extent. Many investigators reported the response of stomata to gaseous pollutants, stomatal opening/closing and size of the stomatal pore are effected. These have a major impact on plant growth as gas exchange is hampered. Elkies & Ormrod (1981<sup>a</sup>) reported that absorption rates of  $O_3$ ,  $SO_2$  and  $NO_2$  into stomates among cultivars differed and generally decreased with longer exposure. Significant reduction in stomatal aperture due to the action of air pollutants was reported by Sharma (1975); Sharma & Butler (1975); Vijayan & Bedi (1988<sup>b</sup>). An analogous finding by Gupta & Ghouse (1986) showed that the size of stomatal pore gets reduced due to coal smoke pollutants. Krishnamurthy & Rajachidambaram (1986) reported that reduction in the rate of photosynthetic  $O_2$  was due to cumulative effect of clogging of stomata reducing gas exchange, shrinking of guard cells and subsidiary cells. Fumigation at the end of flooding period induced partial stomatal closure, injury to leaves and reductions in mean relative growth rates (Norby & Kozlowski 1983). Mansfield & Freer - Smith (1984) observed that stomatal closure which occurs as a result of the inhibitory action of a pollutant on photosynthesis in the mesophyll cannot be looked upon as a desirable way of avoiding stress due to pollution. A true avoidance mechanism would involve closure in advance of stress in the mesophyll, rather than as an event secondary to that stress. In this study stomatal closure and reduction in pore size were maximum in Azadirachta. Changes were less in Moringa and Tamarindus. Clogging of stomata was seen in both the species. In Moringa guard cells showed malformation at inner periphery. Greater adverse effects seen in the deciduous species revealed their sensitivity to air pollution. This hampered gas exchange processes and reduced plant productivity. Response of Tamarindus revealed its resistivity. Results and conclusions support the above findings.

#### 4.5.2 Ultrastructure

There are many reports on the ultrastructural changes in leaf cells when exposed to gaseous pollutants (Thomson *et al.*, 1966; Mejnartowicz, 1984; Miyake *et al.*, 1984). Some of the changes are swelling and curling of thylakoids and reduction in grana. Dixit (1988) showed that bauxite and cement dusts caused disruption of cell structure and plasmalemma was highly vesiculated and separated from the cell wall in Amaranthus and Phaseolus. Treshow (1984) observed granulation of chloroplast stroma in electron micrographs of  $O_3$ -treated tissues which was due to oxidation of SH groups in ribulose biphosphate carboxylase. Huttunen & Soikkeli (1984) showed damage to thylakoids and appearance of light coloured plastoglobuli resembling lipid material were found to be especially frequent in trees within urban areas. They concluded that these ultrastructural changes can be related to some extent to changes in photosynthetic capacity of the mesophyll cells. Similar changes were seen in the present investigation revealing that pollutants affect the ultrastructure of chloroplast and ultimately leads to death of the cell. This hampers plant productivity and reduces plant growth.

Chloroplasts in the leaf cells are the main centres for the photosynthesis and any damage to this organelle hampers photosynthate production. Krishnayya & Bedi (1988<sup>b</sup>) reported that exogenously supplied ascorbic acid mitigates the adverse effects of  $SO_2$  to a certain extent. Lee *et al.*, (1984) concluded that the reversal of  $SO_2$  influence on plastic ultrastructure may be ascribed to the ascorbic acid protection of membrane, against lipid peroxidation. This study indicates that exogenously applied ascorbic acid reverses the effects of  $SO_2$  on the ultrastructure of chloroplast and helps in maintaining their integrity. In conclusion ascorbic acid seems very effective as an antidote to  $SO_2$  damage in Azadirachta.



#### 4.6 ARTIFICIAL FUMIGATION STUDY

Results of the artificial fumigation study showed that ascorbic acid acts as mitigating agent against sulphur dioxide. Untreated saplings exposed to  $\text{SO}_2$  showed almost similar reductions in growth and biochemical parameters as were seen at the field level studies. Deciduous species showed higher damage as compared to evergreen species. Some of the common observations were reduction in growth rate, leaf production, chlorophyll pigments etc. Higher leaf area damage and peroxidase activity were also seen. Conclusions and explanations are the same as mentioned in the previous section. Hence, this part is not elaborated. Importance has been given to the ascorbic acid treatment and its influence on adverse effects of  $\text{SO}_2$ .

##### 4.6.1 Mitigating effects of Ascorbic Acid

There are a few reports on amelioration studies. Some of the reports reveal mitigating effects of various chemicals used against pollutant-induced damage. Sometimes even pollutants when present at low concentrations in nutrient deficit soils can act as nutrients for the growth of the plant. It is important to understand whether adjustment of crop nutrition may be used as a strategy to reduce/mitigate injury by pollutants. Cowling & Koziol (1982) reported that plants growing in soil containing an excess of sulphur may actually have been retarded on treatment with  $\text{SO}_2$  as would be the case with an over supply of fertilizer. Irving (1983) concluded that the net response of a crop to acidic precipitation is the result of the interaction between the positive effects of S + N fertilization, the negative effects of acidity and the interaction of these factors and other environmental conditions such as soil type and presence of other pollutants. Heagle et al., (1983<sup>b</sup>) suggested that physical or nutritional

characteristics of growth substrates will not cause major changes on the magnitude of plant response to chronic doses of ozone when requirements for normal plant growth were met. Ayazloo et al., (1980) reported that sulphur dioxide ( $380 \mu\text{g} \cdot \text{m}^{-3}$ ) depressed growth in all nitrogen treatments after 62 days, without any visible symptoms on the foliage and with some indications that the high level of nitrogen decrease the severity of injury. The level of soil sulphur had few effects on growth and there was little evidence of  $\text{SO}_2$  acting as a nutrient in any of the treatments. Thus response of plants to different pollutant levels depends on various environmental factors and soil nutritional conditions. As majority of these studies were carried out in vitro, response of plants in field conditions varies to a similar pollutant dose because of the wide range of factors influencing its growth.

Various chemical reagents have been used as mitigating agents against pollution injury. Nandi et al., (1984) showed that calcium hydroxide treatment to  $\text{SO}_2$  exposed Vigna sinensis plants lessened the damage. Calcium hydroxide helped in neutralising the acidity of  $\text{SO}_2$ . Similarly amelioration of  $\text{SO}_2$  injury with urea treatment were also reported (Pandey, 1982; Singh & Rao, 1985). Nandi et al., (1981) used potassium ascorbate as an antidote to  $\text{SO}_2$  phytotoxicity. In vitro studies using ascorbic acid as mitigating agent against  $\text{SO}_2$  were conducted by Krishnayya & Bedi (1988<sup>b</sup>), Vijayan & Bedi (1988<sup>c</sup>). Recent research using selective chemical treatments that alter plant metabolic processes prove that certain plant varieties which are naturally quite sensitive to oxidant injury can be transformed into high tolerant plants by such treatments. Ethylene diurea (EDU) gives a 30 - fold enhancement in the foliar tolerance to ozone (Bennett et al., 1984). Roberts et al., (1985) also showed that EDU reduced the appearance of  $\text{O}_3$ -induced symptoms. Lee & Bennett (1982) observed that EDU enhanced tolerance to ozone injury always correlated with