FINAL REPORT TO THE ENERGY RESEARCH AND DEVELOPMENT CORPORATION (ERDC)

PROJECT TITLE:
DEVELOPMENT OF SECONDARY AMBIENT AIR QUALITY CRITERIA FOR SO₂ AND NOₓ

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Murdoch University Pollution Research Facility (PRF)

The PRF is located on the campus of Murdoch University adjacent to the University Veterinary Farm. It operates twenty-four open-top and six closed-top fumigation chambers. Gas supply, metering and monitoring equipment enable these chambers to be fumigated with the commonly-occurring air pollutant gases; sulphur dioxide, nitrogen oxides, hydrogen fluoride and ozone; in varying combinations and at varying concentrations and frequencies. Plants are propagated in field plots or in glasshouses and then moved into the chambers. Pollution effects on growth and physiology are assessed at the PRF laboratory. Additional laboratory facilities in the School of Biological and Environmental Sciences support these studies. The PRF also operates a weather station and monitors ambient air pollution at remote sites via modem links. This facility, established in 1985 and currently valued at over one million dollars, undertakes contract work for industry customers.
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ABSTRACT

Dose-response relations for the effects of SO\textsubscript{2} and NO\textsubscript{x} (NO and NO\textsubscript{2}) on Australian vegetation were developed using open-top chamber fumigation. This information was used, in conjunction with review of the literature, to formulate secondary ambient air quality criteria for the protection of human welfare in Australia.

A simple linear regression of SO\textsubscript{2} concentration versus response best described the effects of long term, frequent exposures to SO\textsubscript{2} on the growth or yield of Australian vegetation. This regression predicted zero yield or growth loss at 25 nLL\textsuperscript{-1} SO\textsubscript{2} as a 4-h average. Exposure length and frequency were of little importance in determination of the dose-response relationship. Leguminous species and Eucalyptus species showed a no-threshold response due to simultaneous exposure to SO\textsubscript{2} and NO\textsubscript{x}. Both NO and NO\textsubscript{2} were phytotoxic.

To protect human welfare in Australia it is recommended that a one hour arithmetic mean value of 200 nLL\textsuperscript{-1} SO\textsubscript{2} not be exceeded, a four hour arithmetic mean value of 25 nLL\textsuperscript{-1} SO\textsubscript{2} not be exceeded more than once per week and that the sum of the NO and NO\textsubscript{2} concentrations be no higher than 25 nLL\textsuperscript{-1} as a four hour arithmetic mean in the presence of the same levels of SO\textsubscript{2}.
SUMMARY

At present Australia has no national secondary ambient air quality standards for the protection of the environment from damage caused by emissions of sulphur dioxide (SO₂) and nitrogen oxides (NOₓ), the latter comprising nitrogen dioxide (NO₂) and nitric oxide (NO). Primary guidelines designed to protect human health are in use in Australia as health effects have been well characterised by a number of studies throughout the world, with organisations such as the World Health Organisation and the National Health and Medical Research Council recommending criteria for the basis of such guidelines. However, such criteria are inadequate to protect the environment and so secondary criteria are needed based upon the effects of air pollutants on the more sensitive receptors, vegetation and materials.

While data collected on air pollutant effects on materials can be drawn from international research to assess economic loss in specific countries, the database for vegetation effects must be specific to the biogeographic region of concern. The reason for this is that large differences occur in species response due to climate and species type. Australia possesses native flora and climatic conditions quite different to those of the United States of America, Europe and Japan, areas where the most extensive research on plant response to air pollution has been conducted to date. Thus, dose-response relationships established for vegetation in these areas cannot be used to protect natural ecosystems and agricultural and forestry systems occurring in Australia and adapted to Australian environmental conditions.

As Australia's energy requirements are primarily met through the operation of coal-fired power stations, the Energy Research and Development Corporation funded this study to establish secondary ambient air quality criteria for SO₂ and NOₓ - the major emissions from these operations. The specific project objectives were as follows:

1. to review existing dose-response information on the effects of SO₂ and NOₓ on vegetation and materials;

2. to derive dose-response information for the effects of SO₂, NOₓ and their combination on the agricultural crops, forestry crops and native tree species of greatest economic and ecological importance around coal-fired energy generating facilities in Australia; and

3. to use this dose-response information to produce an ambient air quality criteria document for SO₂ and NOₓ which combines an adequate measure of protection for human welfare in Australia, without the imposition of unnecessary emission controls.
The following programme of works was undertaken:

1. compilation and production of literature reviews on the thresholds and dose-response relations of plants and materials exposed to SO$_2$, NO$_x$ and their combination;

2. establishment of an experimental protocol for the development of dose-response information for Australian plant species under conditions relevant to operations of the Australian energy generating industry;

3. planning, construction, testing and operation of an open-top chamber fumigation system for the development of these dose-response relations;

4. selection of the most economically important Australian agricultural and forestry crops, and ecologically significant native tree species occurring in areas that have a high concentration of coal-fired power generating facilities;

5. fumigation of these species and analysis of response data; and

6. review of all available information, to develop ambient air quality criteria for SO$_2$ and NO$_x$ for the protection of human welfare in Australia.

The initial reviews of the literature highlighted the need for information on the response of Australian vegetation to SO$_2$ and NO$_x$ under realistic field conditions. To achieve this, the approach of the United States Environmental Protection Agency and the European Economic Community was adopted whereby an open-top chamber fumigation system was used to establish dose-response relations. This approach effectively and efficiently allows determination of effects under realistic conditions. To ensure that the pollutant exposure conditions used were relevant to those occurring around the coal-fired energy generating facilities in Australia, the research team was guided by regular consultation with an Advisory Committee consisting of representatives from energy generation and pollution control organisations from around Australia.

After three years it has been possible to establish a dose-response database, more comprehensive than that available in all but a few countries of the world, to formulate Australian secondary ambient air quality criteria.

The following points summarise the findings of this project:

1. A review of the international literature established that for both vegetation and materials far more information is available on SO$_2$ than either NO$_x$ or the
interactive effects of \( \text{SO}_2 \) and \( \text{NO}_x \). In addition, \( \text{NO} \) is rarely considered, as the literature focuses primarily on the effects of \( \text{SO}_2 \) and \( \text{NO}_2 \) in combination, with some studies of \( \text{NO}_2 \) alone.

For vegetation, large discrepancies were apparent in data collected using different methodologies and for data from different research facilities. Background ambient air pollution was a major source of this discrepancy.

Based on overseas studies, the concentrations of \( \text{SO}_2 \) reported to have reduced growth of higher plants are a continuous exposure to 16 nLL\(^{-1}\) \( \text{SO}_2 \) over 173 days for perennial ryegrass and 21 nLL\(^{-1}\) \( \text{SO}_2 \) over 28 days for tobacco and cucumber. \( \text{NO}_2 \) alone is reported to have little effect on vegetation until concentrations reach 200 nLL\(^{-1}\), however the threshold for growth effects due to simultaneous exposure to \( \text{SO}_2 \) and \( \text{NO}_2 \) for northern hemisphere species may be as low as 15 nLL\(^{-1}\) \( \text{NO}_2 \) in the presence of similar amounts of \( \text{SO}_2 \). Most studies report synergistic interactions between \( \text{SO}_2 \) and \( \text{NO}_2 \).

Insufficient information is available at present to assess economic loss due to materials damage in Australia from exposure to ambient \( \text{SO}_2 \) and/or \( \text{NO}_x \). Iron, zinc and stone are considered to be the most sensitive materials to damage from \( \text{SO}_2 \) and textiles and nickel are the most sensitive to \( \text{NO}_2 \). Damage functions (dose-response relations) need to be quantified for these materials as a basis for criteria to protect against deterioration of materials in general. Ultimately this will rely on economic assessments and value judgements as to how much damage is acceptable to the community as no true thresholds exist for materials degradation. This is due to the fact that materials lack regenerative capacity and damage is cumulative. For materials of cultural significance, for example, marble statues and limestone buildings, economic loss due to damage is very difficult to quantify, further complicating the establishment of criteria.

2. The results of the three fumigation experiments conducted under this project showed the following:

(i) barley cv. Schooner, oats cv. Echidna, burr medic cv. Santiago, barrel medic cv. Paraggio, white clover cv. Haifa, *Eucalyptus microcorys* and *E. regnans* all showed no-threshold responses to \( \text{SO}_2 \) exposure under a fumigation regime of 4 h/day, daily for four to five months. The remaining species showed thresholds in the range of 100-200 nLL\(^{-1}\) \( \text{SO}_2 \), except for *Pinus radiata* which was unaffected by \( \text{SO}_2 \) concentrations ranging up to 332 nLL\(^{-1}\) \( \text{SO}_2 \).

(ii) sub-clover cv. Trikkala, barrel medic cv. Paraggio, white clover cv. Haifa and all three *Eucalyptus* species studied, *E. microcorys*, *E. pilularis* and *E. regnans*, proved to be very sensitive to exposure to \( \text{NO}_x \), with 40% reductions in biomass in all three eucalypts due to 82 nLL\(^{-1}\) \( \text{NO}_x \) comprising 74 nLL\(^{-1}\) \( \text{NO}_2 \) and a 50% reduction in biomass of white clover due to 87 nLL\(^{-1}\) \( \text{NO}_x \) comprising 29 nLL\(^{-1}\) \( \text{NO}_2 \). These reductions occurred after 4h/day, daily
exposures for periods of four to five months. In contrast, wheat cv. Banks showed an increase in yield due to exposure to 170 nLL\(^{-1}\) NO\(_2\) (189 nLL\(^{-1}\) NO\(_x\)) under this exposure regime.

(iii) for the non-nitrogen fixing species, simultaneous exposure to SO\(_2\) and NO\(_x\) emphasized the role of these gases as nutritional sources of sulphur and nitrogen respectively and of the complex interaction between plant nutritional status and the required ratio of sulphur to nitrogen for growth. For the nitrogen fixing species simultaneous exposure to SO\(_2\) and NO\(_x\) severely affected growth at all concentrations showing a no-threshold response under the exposure conditions considered. Growth depression in the *Eucalyptus* species was less severe due to simultaneous exposure to SO\(_2\) concentrations in the range 125-175 nLL\(^{-1}\) + 82 nLL\(^{-1}\) NO\(_x\) but detrimental effects on growth were still evident at all concentrations.

3. Analysis of the data from the three experiments and a small existing data base for the effects of SO\(_2\) on Australian plant species, allowed establishment of a reliable dose-response curve for predicting growth and yield loss due to long term, frequent exposure to SO\(_2\).

SO\(_2\) concentration proved the best term for prediction of growth and yield response. Daily duration of exposure, frequency of exposure and total length of exposure were found to be far less important in determining the nature of the dose-response relationship. Australian crop, forestry and native plant species all showed similar overall dose-response curves for frequent exposure to SO\(_2\) allowing establishment of a single dose-response curve for Australian plant species. This dose-response curve predicts that the threshold SO\(_2\) concentration for changes in growth and yield is 25 nLL\(^{-1}\) SO\(_2\) as a four hour average for repeated daily exposures over periods of four-five months. The dose-response curve is described by the simple linear regression:

\[
y = 4.81 - 0.19x \quad (r = 0.79, p<0.001)
\]

where

- \(y\) = % change in yield or growth
- \(x\) = SO\(_2\) concentration (nLL\(^{-1}\))

A limited amount of data on acute visible injury from SO\(_2\) exposure indicated that the threshold for such effects was well below 376 nLL\(^{-1}\) SO\(_2\) as a one hour average and possibly as low as 181 nLL\(^{-1}\) SO\(_2\) as a 2.5 hour average, for Australian native species.

4. Multiple regression analysis showed that the predictive ability of the SO\(_2\) dose-response curve for frequent exposure regimes could not be improved by inclusion of NO\(_x\) concentration terms due to the wide variability in species response. However, this response could be categorised according to plant type. All four leguminous pasture species and all three *Eucalyptus* species tested under this project were very sensitive to simultaneous exposure to SO\(_2\) and
NO\textsubscript{x} showing no threshold responses. Multiple regression analysis for these sensitive species gave a statistically significant dose-response curve which also demonstrated that both NO and NO\textsubscript{2} were phytotoxic components of NO\textsubscript{x} emissions to these species.

5. The particular sensitivity of the leguminous pasture species and *Eucalyptus* species tested to simultaneous exposure to SO\textsubscript{2} + NO\textsubscript{x}, has important implications for industrial operations such as coal-fired power stations. Clearly, SO\textsubscript{2} and NO\textsubscript{x} emissions must be kept to a minimum when coal-fired power stations are located near native eucalypt forests or agricultural land using improved leguminous pastures. The sensitivity of these vegetation types should also be taken into consideration when site selection for new energy facilities is being undertaken and in the management of land use around existing and future facilities.

6. Given that insufficient information on materials degradation exists at present, this aspect of human welfare cannot be incorporated into establishment of secondary ambient air quality criteria, therefore the following secondary ambient air quality criteria were established based on the above analyses of vegetation response to protect human welfare in Australia:

* a one hour arithmetic mean value of 200 nLL\textsuperscript{-1} SO\textsubscript{2} should not be exceeded.

* a four hour arithmetic mean value of 25 nLL\textsuperscript{-1} SO\textsubscript{2} should not be exceeded more than once per week.

* in the presence of levels of sulphur dioxide not higher than 25 nLL\textsuperscript{-1}, the atmospheric concentration of nitrogen oxides (the sum of the nitric oxide and nitrogen dioxide concentrations) should be no higher than 25 nLL\textsuperscript{-1} as a four hour arithmetic mean.

These criteria take into account the need for a conservative approach in setting air quality standards from chamber studies to take account of adverse environmental conditions experienced in the field and possible sensitivities of species not considered in the fumigation trials. Simple mathematical reduction of the four hour mean threshold SO\textsubscript{2} concentration of 25 nLL\textsuperscript{-1} for growth and yield effects, to a 24 hour mean was not attempted as data on species exposed to a wide range of exposure regimes indicated that this was overcautious and unnecessary. Such a 24 hour mean would be unduly restrictive of energy generating operations.

The criterion proposed for simultaneous exposure to SO\textsubscript{2} and NO\textsubscript{x} takes into consideration the phytotoxicity of NO, the interaction of both SO\textsubscript{2} and NO\textsubscript{x} with the nutritional requirements of plants and the minimisation of impacts to particularly sensitive species. However, further research is needed to determine whether this criterion is adequate to protect against substantial
losses in the productivity of leguminous pastures and to prevent disruption of the growth of our native eucalypt forests.

Further research is also required to assess the relative phytotoxicity of NO and NO\textsubscript{2} to Australian vegetation at realistic NO to NO\textsubscript{2} ratios typical of industrial NO\textsubscript{x} emissions.

Despite the absence of adequate data on materials degradation at present the criteria proposed above will go a long way towards preventing substantial economic loss due to materials degradation in Australia. However, economic assessment should be conducted in the future to determine whether the losses under the criteria proposed are acceptable to the community.

The scientific criteria proposed above provide a comprehensive base for the establishment of secondary ambient air quality standards for regulation of the energy generating industry without the imposition of unnecessary emission controls. These criteria can also be used to regulate other industrial operations emitting SO\textsubscript{2} or SO\textsubscript{2} and NO\textsubscript{x} and diffuse sources of these air pollutants in urban environments.
RECOMMENDATIONS

The following secondary ambient air quality criteria are recommended for the protection of human welfare in Australia:

* a one hour arithmetic mean value of 200 nLL\(^{-1}\) SO\(_2\) should not be exceeded.

* a four hour arithmetic mean value of 25 nLL\(^{-1}\) SO\(_2\) should not be exceeded more than once per week.

* in the presence of levels of sulphur dioxide not higher than 25 nLL\(^{-1}\), the atmospheric concentration of nitrogen oxides (the sum of the nitric oxide and nitrogen dioxide concentrations) should be no higher than 25 nLL\(^{-1}\) as a four hour arithmetic mean.

These criteria are to be used in conjunction with one another, not in isolation. They are designed to protect against visible injury and losses in growth and yield in Australian vegetation.

It is recommended that further research be conducted to determine the threshold for effects of SO\(_2\) and NO\(_x\) in combination for the particularly sensitive species identified in this project. This is required to determine whether the SO\(_2/NO_x\) criterion is adequate to protect against substantial losses in the productivity of leguminous pastures and to prevent major disruption to growth of our native eucalypt forests.

Definition of the relative phytotoxicity of NO and NO\(_2\) is also required for Australian vegetation. It is recommended that research be conducted on the effects on vegetation of the varying NO to NO\(_2\) ratios typical of industrial emissions.

Economic assessment of materials degradation in Australia due to SO\(_2\) and NO\(_x\) should be conducted in the future to determine whether economic loss due to materials damage, under the criteria proposed above, represents a major cost to the community. Such an analysis can be conducted using the data currently being compiled in overseas countries to establish damage functions (dose-response relations) for materials sensitive to exposure to SO\(_2\) and NO\(_x\).
CHAPTER 1: INTRODUCTION

Following recommendations of a National Energy Research Development and Demonstration Council (NERDDC) workshop on The Scientific Basis of Standards for the Release of Energy-Based Pollutants in Australia, held in 1987, NERDDC (now ERDC, the Energy Research and Development Corporation) funded a study to compile the scientific data base required to develop secondary ambient air quality criteria for sulphur dioxide ($\text{SO}_2$) and nitrogen oxides ($\text{NO}_x$) for the protection of human welfare in Australia. The term nitrogen oxides referring collectively to nitrogen dioxide ($\text{NO}_2$) and nitric oxide (NO). These particular pollutants are of concern to ERDC as they are the major, potentially polluting, atmospheric emissions of coal-fired power stations. In 1989 coal-fired power stations in Australia used 81 million tonnes of coal to supply 90% of Australia’s energy requirements (ESAA, 1990).

$\text{SO}_2$ emissions primarily relate to coal sulphur content with the majority of the sulphur present in coal being emitted as $\text{SO}_2$ if the flue gases are not scrubbed. $\text{NO}_x$ emissions mainly comprise nitrogen oxides created by thermal fixation of atmospheric nitrogen in the combustion flame with a smaller proportion being derived from nitrogen present in the coal (USEPA, 1985). These emissions are primarily in the form of NO at the point of exit from power station stacks. However, NO rapidly oxidizes to $\text{NO}_2$ in the atmosphere until NO concentrations are reduced to about one part per million (Taylor et al., 1987). At this concentration NO is too dilute to react readily with oxygen. However, the conversion of NO to $\text{NO}_2$ is enhanced by sunlight in the presence of photochemical oxidants. Under these conditions NO reacts with ozone ($\text{O}_3$) to produce $\text{NO}_2$ and oxygen. $\text{NO}_2$ then reacts to form $\text{O}_3$ and peroxyacetyl derivatives such as peroxyacetyl nitrate (PAN) further driving the overall conversion of NO to $\text{NO}_2$ (Taylor et al., 1987). Typically $\text{NO}_x$ emissions from power stations are primarily in the form of $\text{NO}_2$ by the time they reach ground level and interact with humans, vegetation and materials.

At present Australia has no national standards enforceable by law for ambient air quality and control is achieved on a state by state basis. In most states to date, air quality control has focused on the use of primary guidelines which are designed to protect human health. These have been established by organisations such as the National Health and Medical Research Council (NHMRC) and the World Health Organisation (WHO). However, these primary guidelines are usually not adequate to protect against losses in agriculture and forestry, disruption of natural ecosystems, damage to materials and deterioration of aesthetics, which can collectively be considered to represent human welfare. For most of the common air pollutants, the environment is more sensitive than human health to air pollution damage.

In recent years most developed nations have set secondary standards to protect human welfare. These standards are based upon scientific criteria which primarily focus on the response of agricultural, forestry and native plant
species to air pollutant exposure. This approach has been taken as many studies have shown that plants are usually very sensitive receptors of air pollutants. Consequently, standards based on plant response criteria are in practice adequate to protect other aspects of human welfare (Jakeman and Gifford, 1987).

The NERDDC workshop concluded that while primary standards for ambient air quality based on those in use in other countries can be applied in Australia, secondary standards must be based on scientific criteria for Australian plant species grown under Australian climatic conditions (Jakeman and Gifford, 1987). This is because response to air pollutant exposure is species specific and dependent on climate. Recent research has shown that Australian flora is often more sensitive to air pollutant exposure than the vegetation of either North America or Europe (Murray & Wilson, 1988a, 1989a; Murray et al., 1990a).

1.1 PROJECT OBJECTIVES AND PROGRAMME OF WORKS

The objectives of this ERDC funded project were:-

1. to review existing dose-response information on the effects of SO$_2$ and NO$_x$ on vegetation and materials;

2. to derive dose-response information for the effects of SO$_2$, NO$_x$ and their combination on the agricultural crops, forestry crops and native tree species of greatest economic and ecological importance around coal-fired energy generating facilities in Australia; and

3. to use this dose-response information to produce an ambient air quality criteria document for SO$_2$ and NO$_x$ which would combine an adequate measure of protection for human welfare in Australia, without the imposition of unnecessary emission controls.

The following programme of works was undertaken:

1. compilation and production of literature reviews on the thresholds and dose-response relations of plants and materials exposed to SO$_2$, NO$_x$ and their combination;

2. establishment of an experimental protocol for the development of dose-response information for Australian plant species under conditions relevant to operations of the Australian energy generating industry;

3. planning, construction, testing and operation of an open-top chamber fumigation system for the development of these dose-response relations;
4. selection of the most economically important agricultural and forestry crops, and ecologically significant native tree species occurring in concentrated areas of coal-fired power generation in Australia;

5. fumigation of these species and analysis of response data; and

6. review of all available information to develop ambient air quality criteria for SO$_2$ and NO$_X$ for the protection of human welfare in Australia.

To derive dose-response relations for Australian flora, the approach used by the United States Environmental Protection Agency (USEPA) and the Programme (EEC) to assess air pollution injury to vegetation was adopted. Both of these bodies independently selected open top chambers for their crop loss assessment networks established to determine dose-response information for major air pollutants. Alternative approaches such as mechanistic and physiological models have virtually been abandoned in both the United States of America (USA) and Europe since the mid 1970s. This was due to both the complexity of the model characteristics, which meant that it would have been decades before the models could have been used predictively, and the impracticality of growing most crops to full maturity in controlled environment chambers. The National Crop Loss Assessment Network (NCLAN), which used open top chambers in the USA, in four years yielded useful data on field grown crops exposed to pollutants (Adams et al., 1985).

Open-top chambers of the same design as those used by the USEPA and the EEC were used in this study in a series of three experiments. A primary concern was to establish the interactive effects of SO$_2$ and NO$_X$ as these pollutants rarely occur in isolation in airsheds surrounding energy generating facilities. Interactive effects between pollutants often greatly reduce the threshold for adverse effects due to a single pollutant alone.

The species of major economic value or ecological significance that occurred in the airsheds of the major energy generating facilities in Australia were selected for study. This included the most economically important cultivars of the agricultural crops wheat, barley, oats, triticale, lucerne, barrel medic, burr medic, sub-clover and white clover. Three *Eucalyptus* species forming major components of the native forests occurring around Australia's major coal-fired energy generating facilities were selected for trial; *Eucalyptus microcorys*, *E. pilularis* and *E. regnans*. These species are also of importance to the forestry industry for hardwood production. In addition, *Pinus radiata* was studied due to its importance as a softwood plantation species nationally.

The experiments were designed to replicate the conditions under which plants are exposed to SO$_2$ and NO$_X$ around major emission sources. To ensure that realistic conditions were used, the research team was guided by an Advisory
Committee consisting of members involved in energy generation and/or pollution control around Australia. This committee comprised representatives from the:

1. Air Quality Committee of NHMRC;
2. Australian and New Zealand Environment Council;
3. Electricity Commission of New South Wales (NSW);
4. Electricity Supply Association of Australia;
5. Environmental Protection Authority of Western Australia;
6. Environment Protection Authority of Victoria;
7. NSW State Pollution Control Commission;
8. Queensland Electricity Commission; and
9. State Energy Commission of Western Australia.

Regular meetings and discussions with the Advisory Committee occurred. Initial discussions led to an emphasis being placed on determining the effects of long term exposures to relatively low concentrations of $\text{SO}_2$ and $\text{NO}_x$. $\text{NO}_2$ emissions were initially considered to be of more importance than NO.

1.2 REPORT STRUCTURE

This final report for the project has been written in sections in line with the project objectives. Chapter 2 presents the review of the international literature on dose-response information for the effects of $\text{SO}_2$ and $\text{NO}_x$ on vegetation and materials. Chapters 3 to 7 cover the experimental work conducted under the project. Chapter 3 discusses general methodology, experimental design and the basis for species selection. Chapters 4-6 each discuss a single experiment, providing specific methodology with a presentation of results and discussion. Chapter 7 draws together the information obtained from the three experiments and all other research conducted on Australian flora and presents dose-response information for Australian crops, forestry and native plant species in a form relevant to establishing ambient air quality criteria. Finally, Chapter 8 develops this dose-response information into secondary ambient air quality criteria for $\text{SO}_2$ and $\text{NO}_x$ relevant to protection of human welfare in Australia.

It should be noted that conversion factors of $1 \text{nLL}^{-1} = 2.62 \mu g \text{ m}^{-3} \text{SO}_2$ (at $25^\circ C$) and $1 \text{nLL}^{-1} = 1.88 \mu g \text{ m}^{-3} \text{NO}_x$ (at $25^\circ C$) have been used throughout this report (SPCC, 1988).
CHAPTER 2: LITERATURE REVIEW

DOSE-RESPONSE INFORMATION ON THE EFFECTS OF SO₂ AND NOₓ ON VEGETATION AND MATERIALS

2.1 INTRODUCTION

SO₂ and NOₓ are the major air pollutants associated with fossil fuel combustion. Emissions are regulated in most countries to prevent severe environmental degradation but there is still concern about the complex interactions between these gases and the environment. Emissions of SO₂ and NOₓ are increasing in Australia as energy generation, mineral processing, manufacturing and transport industries expand. Current emissions in Australia due to all these sources of SO₂ are about 2 X 10⁶ tonnes per annum (Murray, 1989) and NOₓ emissions from our capital cities exceed 270 X 10³ tonnes per annum (Farrington, 1988).

The current approach to preventing environmental damage relies heavily on monitoring of emissions with preventative action if necessary. For planning of new sources, predictions of ground level concentrations are made based on modelling results. These predictions of ground-level concentrations and monitoring results are then compared to standards and guidelines which originate from the USEPA, NHMRC and WHO to determine acceptability. The problem with these standards and guidelines is that they lack defensible criteria in Australia. It is widely acknowledged that one of the greatest weaknesses in the air pollution control system is the lack of information linking ambient concentrations to effects on the relevant receptors of air pollutants.

Many standards and guidelines do not give consideration to the protection of human welfare, as opposed to human health. The guidelines of the WHO and NHMRC were developed to protect human health, but deleterious effects on crops and forests can occur as a result of long term exposure to much lower concentrations than necessary to adversely affect human health.

Knowledge of the interactions between air pollutants is also very important as a single pollutant rarely occurs in isolation. The interactions between SO₂ and particulates on human health have been recognised for decades but the interactive effects of SO₂ and NOₓ on plants and materials have only recently received attention. A major question for the energy generating industry in the USA is whether exposure to SO₂ alone or mixtures of SO₂ and other pollutants at ambient concentrations less than those that cause visible injury in plants, result in permanent economic loss. Billions of dollars rest on the resolution of this issue as to how much more emission control is required if effects do occur (Jones, 1985).
Lefohn and Runeckles (1987) state that in order to develop air quality standards it is necessary to develop scientific criteria defining the relationship between ambient air quality and the potential for adverse effects on the receptors of concern. However, to set standards, these scientific criteria must be considered in the context of prevailing environmental, social, economic and cultural conditions (WHO, 1987). As Padgett and Richmond (1983) explain, standard setting requires judgement to be used in conjunction with science.

The judgement involved in setting standards is to strike a balance between what is practicable to achieve and what is acceptable in terms of effects. There are three main factors to be considered in this judgement (Llewelyn, 1979). These are:

1. the actual effects encountered at various air quality levels;
2. a measure of the penalties associated with these effects in economic or other terms; and
3. the total cost to the community of controlling air quality to various levels.

The establishment of dose-response relationships for various receptors allows the development of predictive models of the effects of air pollutants. These models can be used to establish the scientific criteria for setting air quality standards based on the thresholds for effects (Heck, 1982), these thresholds being the levels above which detrimental effects are likely and below which insignificant or beneficial effects occur.

A dose-response relationship describes the response of a given receptor to a given range of pollutant exposure conditions. The term dose is often defined as the product of the concentration of a pollutant and the duration of exposure to that pollutant (Heck, 1982). However, especially in the case of vegetation, such a definition of dose is often inadequate to provide a useful model to predict response. Throughout this report the term "dose" is used in general terms to refer to any number of possible mathematical terms which can be used to describe pollutant exposure conditions. For vegetation, concentration alone is usually the most useful term for establishment of a dose-response relation (Roberts, 1984a).

The following review discusses the information currently available on dose-response relationships for vegetation and materials as a basis for the development of Australian secondary ambient air quality criteria. Section 2.2 discusses vegetation and section 2.3 materials.
2.2 VEGETATION

Many attempts have been made to produce empirical models relating pollutant dose to effects on vegetation, especially for parameters such as foliar injury, yield and metabolic change (Roberts, 1984a; Larsen and Heck, 1976; Mansfield et al., 1982). However, the attempt to establish a single dose response curve, which can be applied universally is realistically unattainable. The major reason for this is the large number of biological and environmental factors that affect the extent, severity and the type of injury that occurs in a given plant due to exposure to pollutants such as SO₂ and NOₓ. These factors are shown in Figure 1 and are discussed in detail below. A number of attempts have been made to establish dose-response relationships for particular vegetation types in given countries and some countries have been able to establish secondary ambient air quality standards or guidelines.

The following discussion considers:

1. the factors influencing plant response to air pollutant exposure;
2. the methodology that has been used to collect effects data and establish dose-response relationships for vegetation in other parts of the world;
3. the dose-response data available for the effects of SO₂, NOₓ and their combination and the existing standards and guidelines in use for these pollutants; and
4. the dose-response data currently available for Australian vegetation.

2.2.1 Factors Affecting Response

Factors that affect the rate and amount of pollutant uptake often have the greatest influence on the severity of pollutant effects. The uptake of gaseous air pollutants is usually considered as a series of resistances in the transfer of the gas from the atmosphere to the plant. As most plant surfaces are protected by a waxy cuticle with stomatal pores in this cuticle acting as controlled points for gaseous exchange, stomatal and cuticular resistances are most often discussed with regard to air pollutant uptake. Although other resistance components exist, their effects are small in comparison to the effects of cuticular and stomatal resistance (Fowler, 1985). Of these two, stomatal resistance is the most important factor in determining the uptake of most gaseous air pollutants under dry conditions.

The following discussion of the factors influencing the response of plants to pollutants is dominated by references to the response to SO₂. Much less research has been done on the response of plants to NOₓ and this almost entirely considers only NO₂. However, it is known, for example, that conditions which cause stomatal closure, (low light intensity, low relative humidity) reduce the effects of NO₂ (Law and Mansfield, 1982), as is the case for
Figure 1: Factors that determine the action and fate of $SO_2$ and $NO_x$ in plants. (Adapted from Weinstein, 1977).
SO₂. Krause (1988) claims that the interactions of various factors in the uptake of SO₂ also apply in general for the uptake of NO₂.

The uptake pathway of NO₂ does differ slightly from that of SO₂. Cuticular resistance to SO₂ is considerable. However, NO₂ is more reactive than SO₂ with leaf surface materials as it can react with cuticular wax components leading to damaged surfaces. WHO (1987) state that some, as yet unpublished, studies have shown that significant amounts of NO₂ may enter leaves by penetrating the cuticle. So, although the closure of stomata may reduce the internal dose of NO₂ it may not exclude damage or injury. The extent to which this occurs at ambient concentrations is unknown.

A number of other factors also determine plant response, once uptake has occurred, which largely depend upon the metabolic processes and physiology of the plant concerned and its state of health at the time of exposure. These factors and those affecting uptake are discussed below.

2.2.1.1 Environmental Factors

Stomatal resistance depends upon the degree of opening of the stomata. Each plant also has its own inherent maximum rate of gaseous exchange through its fully open stomata according to the density of stomata and their size. Thus, even under conditions which minimise stomatal resistance, different plants have a different capacity for air pollutant uptake.

The stomata open and close in response to inherent rhythms, light intensity, CO₂ concentration in the sub-stomatal cavity, temperature and water supply to the leaf (Bidwell, 1979). The latter is hormonally controlled and depends on transpiration rate, soil water levels, relative humidity, wind speed and temperature. The stomata of some species have also been shown to open or close in response to SO₂ exposure (Black, 1985). Figure 2 summarises the effects of these parameters on stomatal aperture. Of these parameters light intensity, relative humidity and soil moisture content are the most important in determining the uptake of air pollutants.

As a consequence of the major role that light intensity and quality have on stomatal opening, light is a primary factor controlling SO₂ and NOₓ injury. In the absence of light, most stomata are partially or virtually closed in most species, therefore gaseous exchange is low or minimal.

At mid-day, when stomatal opening is at a maximum in most species, air pollutant injury has been found to be greater than during early morning or late afternoon (Tibbits and Kobriger, 1983). Exceptions to this generalization include plants with specialized metabolism and ecology, especially plants of arid environments, which close stomata during the day to conserve water, and open stomata at night for gaseous exchange.
Figure 2: Factors which affect stomatal aperture.
Srivastava et al. (1975) found that NO₂ uptake in beans was enhanced in the presence of light, when stomata were open. Similarly, Hill (1971) estimated that night-time NO₂ absorption of an oat canopy was only 20-35% of the daytime amount. Yoneyama et al. (1979) found similar results with sunflower plants.

High relative humidity maintains plants in a turgid condition, when adequate soil water is available, and hence, favours the opening of stomata, increasing the sensitivity of plants to air pollutants (Guderian, 1977). Plants maintained at 100% relative humidity are about three times as sensitive to SO₂ as plants at 30% relative humidity (Lacasse and Treshow, 1978). Srivastava et al. (1975) similarly showed that high humidity increased the rate of uptake of NO₂.

The importance of humidity in modifying plant response to SO₂ and NO₂ has been demonstrated by McLaughlin and Taylor (1980) and Thompson et al. (1980) who argue that the current USA secondary standard for SO₂, a 3-h average of 500 nLL⁻¹ SO₂ not to be exceeded more than once per year, will prevent foliar injury in most crops in temperate regions but may be unduly restrictive in semi-arid regions where the low relative humidity reduces the phytotoxicity of gaseous pollutants (Roberts, 1984a).

Soil moisture content plays a major role in stomatal function, as it directly influences the amount of water available to the plant to maintain cells in a turgid state. As stomata close under conditions of plant water deficit, plants are generally less susceptible to air pollutant injury at this time. This has been demonstrated for SO₂ by Zimmerman and Hitchcock (1956). Benedict and Breen (1955, as cited in Amundson and MacLean, 1982) found that after a 4-h exposure to NO₂, plants grown in moist soil developed extensive foliar injury, but little injury occurred under conditions of moisture stress. It is often assumed that desert species are more resistant to air pollutant injury for this reason.

The effects of sulphur dioxide on stomata have now been investigated in a number of species, but the responses are very dynamic and variable and it is not known whether they are reversible or permanent (Black, 1985). Prevailing environmental conditions, the concentration and length of exposure, plant stage of development and leaf age can all modify the response.

The general findings are that at low SO₂ concentrations, 250-1000 nLL⁻¹ SO₂, stomatal opening is enhanced when stomata are already open (Majernick and Mansfield, 1970). However, in some species low SO₂ concentrations produce stomatal closure when relative humidity is less than 40% (Majernick and Mansfield, 1972). Unsworth and Black (1981) believe that initially in these plants, opening does occur at low SO₂ levels but the resultant increase in transpiration induces closure. These plants have developed a mechanism to control stomatal opening in response to evaporative demand. At higher SO₂ concentrations stomatal closure usually occurs (Black, 1985).
It is also argued that changes in carbon dioxide concentration due to the effects of SO$_2$ on respiration and photosynthesis will affect stomatal aperture. Mansfield and Freer-Smith (1984), Unsworth and Black (1981) and Black (1982) believe this is only significant at the higher SO$_2$ concentrations which produce closure.

In contrast, temperature plays only a minor role in affecting stomatal aperture and so pollutant uptake and is generally not considered to markedly affect plant response to SO$_2$ in the range 18-40 °C, (Lacasse and Treshow, 1978). Some recent studies have shown, that low temperatures and SO$_2$ may interact to affect response at a biochemical level. It has been shown that SO$_2$ reduces the freezing resistance of ryegrass (Davison and Bailey, 1982) and spruce (Keller, 1981) and induces cold-stress injury in wheat plants (Baker et al., 1982). A mechanism to explain the reduction in freezing resistance has been proposed which involves a SO$_2$-induced decrease in ascorbic acid concentration and the partial inactivation of the glutathione oxidation-reduction system. This leads to the accumulation of highly reactive oxygen radicals which cause protein aggregation upon freezing and thus tissue death (Murray, 1985a,b).

Wind speed also affects response other than via effects on stomatal aperture. Unstirred air layers around the plant represent important barriers to the entry of gases (WHO, 1987). The thickness of these layers is dependent upon leaf size, shape and orientation, and upon wind speed. As wind speed increases, the boundary layer thickness is reduced, resistance to the movement of gaseous molecules to the leaf surface falls, and hence, uptake rate increases (Unsworth et al., 1985; Ashenden and Mansfield, 1977). The boundary is generally thinner at leaf margins, which could account for injury often occurring at these points, especially in grass species (Taylor and Eaton, 1966). In addition, these are the sites of the greatest transpiration rate, so air pollutant derivatives in the transpiration stream accumulate at leaf tips and margins. Thus, wind speed is important in determining the rate of pollutant uptake and leaf characteristics may determine sites of injury.

Both sulphur and nitrogen are essential elements for plant growth and both NO$_x$ and SO$_2$ at low concentrations can act as supplementary sulphur and nitrogen fertilizers.

Sulphur and nitrogen deficiency can be alleviated by SO$_2$ and NO$_x$ exposure, respectively, and produce enhanced growth. However, pollutant-induced growth stimulations may not always be desirable as susceptibility to insect attack or other environmental stresses may increase because the relationships between various plant nutrients are disrupted (WHO, 1987). In addition, Cowling and Koziol (1982) report that high nitrogen levels can induce sulphur deficiency.

The role of SO$_2$ and NO$_x$ in providing a nutritional source of sulphur or nitrogen, respectively, depends very much on the nutritional status of the
plant at the time of exposure and the relative proportions of sulphur and nitrogen required by the plant. Plants require sulphur and nitrogen in well defined ratios characteristic of their protein composition. Thus, an excess of nitrogen can be deleterious if inadequate sulphur is available for protein synthesis.

The presence of other stresses also influences plant response to pollutant exposure, usually by making a plant more susceptible to injury. This includes diseases, insect attack, competition with other plants and the presence of other toxic pollutants. Under ambient conditions, phytotoxic air pollutants rarely, if ever, occur alone and as such can be considered as another factor affecting uptake and tolerance. SO₂ affects the response of plants to NOₓ and vice versa. The understanding of these effects, however, is minimal. The interaction of pollutants can be antagonistic (less than additive), additive or synergistic (greater than additive).

2.2.1.2 Biological Factors

The biological factors which may affect plant susceptibility to pollutant-induced injury are either ontogenic (relating to developmental stage) or genetic.

Seedlings are relatively resistant to SO₂ due to their lack of fully functional stomata (Halbwachs, 1984). Plants that have just reached physiological maturity are more sensitive to air pollutants than younger or older plants as they have greater rates of gaseous exchange causing greater rates of uptake of airborne gaseous pollutants. Deciduous trees are more sensitive during their early growth periods and show increasing resistance with age (Guderian and Stratmann, 1968, as cited by Halbwachs, 1984).

In some species there are certain growth stages at which they are particularly sensitive to SO₂. Bonte (1982) found that SO₂-induced yield loss was greatest in cereals if they were exposed at the flowering stage. It has been suggested that SO₂ may breakdown the chromosomes of the vegetative and reproductive nuclei of the pollen tube and interrupt the normal flowering process (Ma et al., 1973, as cited in Bonte, 1982).

Plant susceptibility to NOₓ is also determined by age and developmental stage. Amundson and MacLean (1982) summarize the untranslated findings of van Haut and Stratmann. They report that the fumigation of oats during flowering greatly reduced yield, whereas exposure before the initiation of reproductive structures, or during ripening, had no effect. Radish was found to be most susceptible to NOₓ during the period of root expansion. With tobacco and conifers, emerging leaves or needles were more susceptible to NOₓ than mature ones.

Differences in resistance to NOₓ and SO₂ between various species, cultivars or individual plants can be genetically based and attributed to differences in
morphological and anatomical features relating to uptake. Species and
cultivars vary in the number, type, functioning and distribution of stomata
and the physical and chemical nature of the cuticle. Stomatal functioning and
the nature of the cuticle, in particular, alter with leaf age and plant
developmental stage.

Plants also possess differing biochemical and physiological mechanisms for the
storage, re-emission and detoxification of NO\textsubscript{X} and SO\textsubscript{2} which are responsible
for differences in resistance. Genetic factors also determine the activity of
enzymes involved in these detoxification pathways. SO\textsubscript{2} and NO\textsubscript{X}
detoxification mechanisms are primarily light dependent and thus pollutant
uptake which occurs at night or under low light conditions is usually more
injurious than similar doses under high light intensities (NAS, 1977).

2.2.1.3 Pollutant Exposure Conditions

Pollutant exposure conditions, as well as environmental conditions, are of
importance in determining a plant’s response to SO\textsubscript{2} and NO\textsubscript{X}. These
exposure conditions are described by the term "dose" in a dose-response
relationship. Dose is often defined as the product of the concentration of a
pollutant and the duration of exposure. However, this means that the same
dose may result from short-term exposure to high concentrations, as from
long-term exposure to low concentrations, and in many cases plant response to
such different exposures is not equivalent (Roberts, 1984a).

Concentration and exposure duration alone are not the only factors
determining the action of SO\textsubscript{2} and NO\textsubscript{X} on plants. The effect also varies with
frequency and sequence of impact (WHO, 1987). However, there is no
agreement in the literature as to the relative importance of these variables.

Garsed et al. (1982), Jones and Mansfield (1982) and Lane and Bell (1984)
showed from studies involving fluctuating concentrations that long-term
mean concentrations, rather than the peak fluctuations around the mean, are
more important in determining the effects of SO\textsubscript{2} on plants. Likewise, a
fumigation experiment, conducted over two years, demonstrated only small
effects of 282 nLL\textsuperscript{-1} SO\textsubscript{2} peaks within a 37 nLL\textsuperscript{-1} mean, compared with the same
mean at constant concentration (Garsed and Rutter, 1984).

In contrast, Fowler and Cape (1982) claim that the effects of pollutants on
vegetation are strongly dependent on concentration variation with time.
Roberts (1984b) notes that even in diffuse source areas maximum hourly peak
to annual mean ratios may be up to 10:1 for SO\textsubscript{2} and that it is the peak
concentrations that may be most hazardous and of cause for concern. This
view was also confirmed by WHO (1987) in a review of world literature.
However, it must be noted that pollution-free periods can offer a recovery
period for the plant. This is the case if the exposure duration is not too long
and if the pollution-free period is sufficiently long (Guderian, 1977). However,
the duration of the unpolluted periods required to offset detrimental effects cannot conclusively be defined with the current state of knowledge.

As a consequence of the different components of pollutant exposure which determine plant response, a single dose-response relationship cannot be established to cover both short and long term, frequent and infrequent exposure events. In response to this, most regulatory authorities establish variable standards for air pollutants, at least one to protect vegetation from long-term exposure, which may be a seasonal or annual mean, and one to protect vegetation from short-term, high concentration exposures. The latter may be an hourly, or three hourly average value which should not be exceeded or only exceeded on a certain number of occasions over a longer period.

2.2.2 Methodology Used to Collect Effects Data

In many cases dose-response relationships are extracted from the literature retrospectively. This is usually a very difficult exercise as dose and effects have been reported in many different ways and data has been collected using many different experimental techniques under varying environmental conditions (Roberts, 1984b). Fundamentally there are two ways in which effects data can be collected for plants. The first is by the collection of data from plants exposed to polluted ambient air and the second is by actively fumigating plants with gaseous pollutants in a controlled manner.

Studies considering polluted ambient air fall into two categories:

1. Field Studies
These are performed in an uncontrolled environment. Ambient air is monitored for the gas of interest and the status of the plant assessed. Most long-term forest studies are done in this way.

2. Filtration Experiments
Plants are grown in chambers ventilated with ambient, or charcoal-filtered air. Young trees and crops are studied in this way.

Controlled fumigation experiments are ideally performed in "clean" environments or in conjunction with a filtration system. There are primarily two methods:

1. Open-air Fumigations
Controlled concentrations of pollutants are supplied to plants in an open-field situation (such as that described by Miller et al., 1980).

2. Chamber Fumigations
Plants are placed in a chamber which is fumigated with controlled concentrations of gases. These chambers are usually open-top (for
example those described by Heagle et al., 1973 and Roberts et al., 1983a) to reduce the "chamber-effect" and therefore produce data that can realistically be related to the field.

Difficulties arise in comparing the results of experiments performed in such different ways. Studies of polluted ambient air generally indicate greater sensitivity of species to a single air pollutant than studies using controlled fumigation experiments because usually several pollutants are present, but not quantified. WHO (1987) states that future research of polluted ambient air needs a more systematic monitoring of all pollutants. Clearly, discrepancies such as these need to be reconciled when collating dose-response data as results from various studies cannot be simply compared. These discrepancies are further compounded by the multitude of varying environmental conditions under which the experiments were conducted.

In both the United States and Europe, crop loss assessment networks have been established specifically to develop dose-response relationships for economically important crop species. These networks consist of open-top chamber fumigation facilities located throughout the regions of concern. The use of open-top chamber fumigation has proved an effective and efficient method of yielding realistic dose-response information in a relatively short time period. The use of a network of such facilities allows assessment of regional climatic effects.

In the United States it has been concluded that average ambient concentrations of SO$_2$ and NO$_x$ are not high enough or elevated frequently enough to affect crop production on a regional scale although effects may occur on a local scale close to sources (Irving, 1990).

The following sections review the literature available on the effects of SO$_2$, NO$_x$ and their combination on plants, discuss established dose-response relations and existing standards and guidelines.

2.2.3 SO$_2$ Dose-Response Data

2.2.3.1 Agricultural Crops

The lowest concentrations of SO$_2$ that have been found to reduce the growth of higher plants were 16 nLL$^{-1}$ over 173 days for perennial ryegrass (Bell et al., 1979) and 21 nLL$^{-1}$ over 28 days for tobacco and cucumber (Mejstrik, 1980).

Lower concentrations have been reported in the literature that reduce growth or yield but the results have been obtained by using filtration experiments, and hence, the greater sensitivity shown by the plants was probably a response to several pollutants, not solely SO$_2$. For perennial ryegrass in a filtration experiment, the lowest concentration of SO$_2$ at which growth reduction has been recorded is 13 nLL$^{-1}$ over only 28 days (Awang, 1979, as cited by WHO,
Buckenham et al. (1982) using similar techniques reported a yield reduction in barley attributable to ambient air containing \( \text{SO}_2 \) at 19 nL L\(^{-1}\). In conflict with this, Baker et al. (1987) exposed barley to \( \text{SO}_2 \) throughout its growing season and reported that 40-50 nL L\(^{-1}\) stimulated growth and 80-200 nL L\(^{-1}\) depressed growth. WHO (1987) reports that the Buckenham et al. (1982) experimental site was close to a brickworks and the presence of fluorides may have increased plant sensitivity to \( \text{SO}_2 \). Additive effects of hydrogen fluoride (HF) and \( \text{SO}_2 \) have been reported in the literature in other studies (Matsushima and Brewer, 1972; Murray and Wilson, 1988b). Consequently, results need to be considered carefully when assessing \( \text{SO}_2 \) dose-response data.

It is of vital importance to consider the potential presence of several pollutants when establishing air quality criteria. Other pollutants must be monitored, and concentrations documented, for their relative importance to be determined. Roberts (1984b) concluded that the data-base from air filtration studies was not adequate to draw conclusions from, due to the unacknowledged role of other pollutants.

The data base for long-term effects of \( \text{SO}_2 \) on crops is much larger than that for short-term exposures, but not necessarily more conclusive. Even when the same species have been studied under similar conditions, by different research groups, agreement on threshold levels cannot always be reached. For example, after extensive study of ryegrass by Bell and colleagues (Bell et al., 1979; Bell, 1982) they concluded that the threshold for a long-term \( \text{SO}_2 \) response would be in the range 40-80 nL L\(^{-1}\). Cowling and Jones (1978), however, concluded from a similar experiment on ryegrass that 80 nL L\(^{-1}\) was without effect, but 160 nL L\(^{-1}\) could sometimes reduce yield. This type of discrepancy needs to be rationalized when attempting to establish dose-response relationships.

Some recent studies report on the "no-effect" concentrations of \( \text{SO}_2 \) for certain species. Maas et al. (1987a,b) reported that exposure to 250 nL L\(^{-1}\) \( \text{SO}_2 \) for two weeks had no effect on the growth or yield of spinach, red clover or french bean. It should be noted, however, that this dose reduced soybean yield by 20%.

Kohut et al. (1987) reported no effect on wheat of multiple intermittent exposures of up to 363 nL L\(^{-1}\) \( \text{SO}_2 \), for four hours, three times per week. Vicia faba responded by a 12% yield increase to continuous exposure to 21 nL L\(^{-1}\) to 34 nL L\(^{-1}\) \( \text{SO}_2 \) for 56 days (Adaros, et al., 1988).

Irving (1987) after reviewing several studies on corn and soybean concluded that if dose was calculated as concentration multiplied by hours of exposure then doses in the 5000 nL L\(^{-1}\)h range essentially have no effect. Soybeans are generally affected by \( \text{SO}_2 \) doses above 10 000 nL L\(^{-1}\)h, while field corn are shown to be unaffected by doses as high as 24 000 nL L\(^{-1}\)h.

Roberts (1984a) provides one of the most comprehensive analysis of dose-response relations for crops which has been conducted to date. Using results
from a number of chamber studies conducted in the United Kingdom, Roberts analysed a data base consisting of 125 exposures of 21 different species of grasses, horticultural crops and cereals to constant concentrations of \( \text{SO}_2 \).

Roberts examined various combinations of the data points which led to the establishment of two concentration-response curves, one based on the concentration range \( 0-50 \ \text{nL}^{-1} \) representative of ambient \( \text{SO}_2 \) concentrations in the UK, using 35 data points from studies of 9 different species (Figure 3) and one based on data points for the most extensively studied species \( \text{Lolium perenne} \) (ryegrass) using only chamber studies with relatively high air exchange rates (Figure 4).

The regression equation for the 9 species was marginally statistically significant \((r = 0.35, p = 0.05)\) and predicted zero yield loss at \( 21 \ \text{nL}^{-1} \ \text{SO}_2 \) continuous exposure. The regression for a single species, \( \text{Lolium perenne} \), was significant at \( p < 0.001 \) and predicted zero yield loss at \( 35 \ \text{nL}^{-1} \ \text{SO}_2 \) continuous exposure \((r = 0.81)\).

Previously, Bell (1982) had considered the results of 32 long term fumigation exposures of grasses, particularly \( \text{L. perenne} \), in the United Kingdom (UK) and found no correlation between reduction in shoot dry weight and a given dose (concentration \( \times \) days of exposure) of \( \text{SO}_2 \). Normalisation of the log of dry weight for exposure duration (to account for differences produced by harvesting plants with similar growth rates at different times) did not improve the correlation.

Mansfield and Freer-Smith (1981) re-examined these data using Bell's time normalisation method but excluded experiments of less than 40 or greater than 160 days duration and also those using unfiltered polluted ambient air. This gave a significant regression predicting zero yield loss at approximately \( 30 \ \text{nL}^{-1} \ \text{SO}_2 \) continuous exposure.

There were several major reasons for the discrepancy between experiments identified by these studies, these being:

1. the use of polluted ambient air as a control treatment;
2. the use of low rates of air flow in chambers (<1 air change/minute);
3. the comparison of short duration, high concentration exposures with long duration, low concentration exposures;
Figure 3: Linear regression of SO₂ concentration and percentage yield reduction, for 9 crop species exposed to SO₂ continuously in chambers for more than 20 days to less than 50 nLL⁻¹ SO₂. The dotted lines show the 95% confidence limits. (Roberts, 1984).

Figure 4: Linear regression of SO₂ concentration and percentage yield reduction, for Lolium perenne exposed to SO₂ continuously in chambers for more than 20 days to less than 200 nLL⁻¹ SO₂. The dotted lines show the 95% confidence limits. (Roberts, 1984).
4. species variability; and

5. differences in climatic conditions, especially relative humidity.

The concentration-response curves obtained by Roberts (1984a) used a much expanded database and benefitted from the attempts of previous authors to identify reasons for discrepancies between experiments. Thus, Roberts was able to test for significant correlations by applying various restrictions on the data set to find the most applicable set of conditions for comparing various experiments from various studies.

Roberts et al. (1983b) also examined data on intermittent exposures of crops to SO$_2$. Insufficient data were available to obtain concentration-response curves but the following conclusion was tentatively made:

"If fumigation is for approximately 10% of the growth period, the mean must be above 185 nLL$^{-1}$ and the 97.5 percentile value above 260 nLL$^{-1}$ to affect yield".

2.2.3.2 Trees

The database for trees is much less complete than it is for crops. It appears that, generally, trees show greater sensitivity to SO$_2$ than do crops. Several reasons can be offered for this greater sensitivity, and they can be assigned to one of two categories:

1. The greater sensitivity may be an artefact of the experimental design:

   a) the logistical constraints on the controlled fumigation of trees means that, by necessity, it is performed on young trees, which are reported to be more sensitive than mature trees (Berry, 1964);

   b) studies on mature trees have been field studies, and hence, the increased sensitivity could be attributable to the presence of other pollutants;

   c) fumigation studies are often carried out at the pollutant-sensitive stages of fruit set and development, and hence, an apparent greater sensitivity may result (Roberts, 1984b); or

2. The greater sensitivity is real, and attributable to specific characteristics of trees:

   a) trees have lower protein synthesis rates than most crops and consequently a lower demand for sulphur. Accordingly, trees may reasonably be expected to show a greater sensitivity than crops to
SO₂ pollution as they have a reduced capacity to metabolize sulphur (Laisk et al., 1988);

b) a carry over effect of SO₂ from year to year, may exist for trees which is not experienced by annual crops (Roberts, 1984b). The fumigation of trees has been restricted to three to four years only, therefore, the significance of SO₂ to the growth of a tree throughout its lifetime (80-250 years) remains unknown and can only be evaluated on the basis of incremental studies on mature trees at sites where some form of a pollution record is available over a long period (WHO, 1987). Hence, the limited information available on tree effects.

Linzon (1971) surveyed Pinus strobus at various sites near a nickel/copper smelter and noted that in areas with a mean SO₂ concentration of 13-17 nLL⁻¹ a 2.6% annual mortality rate occurred. In a "control" site the mortality rate was 0.7%. In another field study, Farrar et al. (1977) found in Pinus sylvestris that an annual mean below 14 nLL⁻¹ SO₂ had no effect on the frequency of occurrence of this species in a forest stand. A mean of approximately 21 nLL⁻¹ reduced the frequency by 50%. An annual mean above 35 nLL⁻¹ resulted in the absence of this species. The presence of other pollutants could have influenced these results.

Steubing and Fangmeier (1987) studied the effect of 113 nLL⁻¹ SO₂, for four hours once a week over a period of 2 years, on a Beech forest. They reported that this concentration decreased productivity and leaf area indices.

Istas (1980, as cited by Leone, 1985/6) claim that the threshold concentration of SO₂ for injury to apple trees is 100-300 nLL⁻¹.

In a chamber study of four broad-leaved tree species, Garsed et al. (1979) found that fumigation with 56 nLL⁻¹ SO₂ for up to 17 months had no effect on yield in any of the species but that it reduced leaf area in two of the species. In one of the species height was also reduced. One species showed increased leaf senescence and one species was unaffected. The potential for yield loss is evident given a longer fumigation period, possibly of years.

In an indoor chamber experiment Temple (1972) observed visible injury in Ginkgo biloba after 30 days of fumigation with 244 nLL⁻¹ SO₂. That level, for that duration, however, is now considered to be unrealistically high.

Roberts et al. (1983b) also examined data on continuous and intermittent exposure of trees to SO₂. Insufficient data were available to obtain concentration-response curves but the following conclusions were tentatively made:
1. Effects on growth and physiological parameters occur due to continuous exposure to $SO_2$ in the range of 100 nLL$^{-1}$ for several weeks to 50 nLL$^{-1}$ for several years; and

2. If fumigation is for approximately 10% of the growth period, the mean must be above 170 nLL$^{-1}$ and the 97.5 percentile value above 265 nLL$^{-1}$ to produce declines in growth.

Roberts et al. (1983b) stated that these thresholds for trees may be lower under adverse environmental conditions.

2.2.3.3 Existing Standards and Guidelines for $SO_2$

The International Union of Forest Research Organizations (IUFRO) has proposed two sets of air quality standards for $SO_2$. One is for the protection of forestry trees at most sites and another for trees growing in poor conditions (WHO, 1987). These standards are shown in Table 1 and are based on field observations of tree growth in Canada, Czechoslovakia and Finland (WHO, 1987).

Table 1 IUFRO Air Quality Standards for Sulphur Dioxide (1978), confirmed in 1980 (WHO, 1987).

<table>
<thead>
<tr>
<th>Period</th>
<th>Full Production in most sites</th>
<th>Full Production and Environmental Protection$^a$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Annual average</td>
<td>19</td>
<td>9</td>
</tr>
<tr>
<td>24-h average$^b$</td>
<td>38</td>
<td>19</td>
</tr>
<tr>
<td>97.5 percentile of 30 minute values in growing season</td>
<td>57</td>
<td>29</td>
</tr>
</tbody>
</table>

$^a$ Environmental protection against erosion and avalanches, and to ensure full production in higher regions of mountains, boreal zones, extreme sites etc.

$^b$ The 24-h average may be exceeded 12 times in a period of 6 months.

After an extensive review of European and US literature, WHO (1987) proposed the following guidelines for $SO_2$ for plant species in general:

- annual $SO_2$ average - 11 nLL$^{-1}$
- 24-h $SO_2$ average - 38 nLL$^{-1}$
However, WHO states that these guidelines may not be sufficient under extreme environmental conditions or if other pollutants are present.

The current US secondary ambient air quality standard for SO$_2$ is:

- 3-h SO$_2$ average - 500 nLL$^{-1}$, not to be exceeded more than once/year.

This 3-h standard is designed to protect against acute visible injury in plants and is primarily based on the results of a single study on the species *Fraxinus* (Bennett, 1985).

A 24-h SO$_2$ average of 140 nLL$^{-1}$ and annual SO$_2$ average of 30 nLL$^{-1}$ is used to protect human health in the US. These two standards in conjunction with the 3-h standard for vegetation form the US National Ambient Air Quality Standards (NAAQS) for SO$_2$ (Bennett, 1985).

In Canada a range of ambient air quality objectives have been proposed (Treshow and Anderson, 1989) as shown in Table 2. The acceptable and desirable objectives take into account protection of human welfare.

Table 2: Federal Air Quality Objectives for Canada (Treshow and Anderson, 1989)

<table>
<thead>
<tr>
<th>Averaging Time</th>
<th>Desirable Range $^a$ (nLL$^{-1}$)</th>
<th>Acceptable Range $^b$ (nLL$^{-1}$)</th>
<th>Tolerable range $^c$ (nLL$^{-1}$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1-h average</td>
<td>0 - 172</td>
<td>172 - 344</td>
<td>-</td>
</tr>
<tr>
<td>24-h average</td>
<td>0 - 57</td>
<td>57 - 115</td>
<td>115 - 305</td>
</tr>
<tr>
<td>Annual arithmetic mean</td>
<td>0 - 11</td>
<td>11 - 23</td>
<td>-</td>
</tr>
</tbody>
</table>

$^a$ long term goal for air quality

$^b$ to provide protection against adverse effects on soil, water, vegetation, materials, animals, visibility, personal comfort and well-being

$^c$ level that requires abatement without delay due largely to substantial risks to human health.

The United Nations document on National Strategies and Policies for Air Pollution Abatement (United Nations, 1987) lists a number of ambient air quality standards from around the world a synopsis of which is shown in Table 3. Unfortunately this document gives little indication as to whether these standards are primary (to protect human health) or secondary (to protect
TABLE 3: Ambient Air Quality Standards and Guidelines for SO$_2$ (United Nations, 1987).
All values have been converted from $\mu$g m$^{-3}$ to nL L$^{-1}$ using $1$ nL L$^{-1} = 2.62$ $\mu$g m$^{-3}$ at 25°C.

<table>
<thead>
<tr>
<th>Country</th>
<th>1 hour or short-term average</th>
<th>24 hour average</th>
<th>Annual or long-term average</th>
</tr>
</thead>
<tbody>
<tr>
<td>Austria</td>
<td>19 (Apr-Oct)</td>
<td>38 (Nov-Mar)</td>
<td>27 (Apr-Oct) 57 (Nov-Mar)</td>
</tr>
<tr>
<td></td>
<td>(97.5 percentile of all half hourly averages)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Canada</td>
<td>172-344</td>
<td>57-115</td>
<td>11-23</td>
</tr>
<tr>
<td>Czechoslovakia</td>
<td>191 (half hour)</td>
<td>57</td>
<td>23</td>
</tr>
<tr>
<td>Denmark</td>
<td>95 (annual, 98 percentile)</td>
<td>76</td>
<td>10-153</td>
</tr>
<tr>
<td>Finland</td>
<td>191 (99 percentile)</td>
<td>76</td>
<td>10-153</td>
</tr>
<tr>
<td>Germany (East)</td>
<td>191</td>
<td>57</td>
<td>57</td>
</tr>
<tr>
<td>Germany (West)</td>
<td>191 (annual, half-hour 98 percentile)</td>
<td>19-53 (most areas)</td>
<td></td>
</tr>
<tr>
<td>Greece</td>
<td>95</td>
<td>31</td>
<td>31</td>
</tr>
<tr>
<td>Hungary</td>
<td>191 (half-hour)</td>
<td>57</td>
<td>27</td>
</tr>
<tr>
<td>Iceland</td>
<td>19 (95 percentile)</td>
<td>11</td>
<td></td>
</tr>
<tr>
<td>Italy</td>
<td>95</td>
<td>31</td>
<td></td>
</tr>
<tr>
<td>Luxembourg</td>
<td>95-134 (98 percentile)</td>
<td>31-46</td>
<td></td>
</tr>
<tr>
<td>Netherlands</td>
<td>317</td>
<td>%ile Limit Guide</td>
<td></td>
</tr>
<tr>
<td>Norway (proposed)</td>
<td>57</td>
<td>19-57</td>
<td>10-23 (half year)</td>
</tr>
<tr>
<td>Romania</td>
<td>286</td>
<td>95</td>
<td></td>
</tr>
<tr>
<td>Spain</td>
<td>267 (two hour)</td>
<td>38-57</td>
<td>15-23 (guide value)</td>
</tr>
<tr>
<td>Sweden</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Switzerland</td>
<td>38 (98 percentile)</td>
<td>11</td>
<td></td>
</tr>
<tr>
<td>Turkey</td>
<td>153 (industrial-900)</td>
<td>57</td>
<td>57 (industrial-250)</td>
</tr>
<tr>
<td>USSR</td>
<td>191 (20 minutes)</td>
<td>19</td>
<td></td>
</tr>
<tr>
<td>USA</td>
<td>496$^a$ (3 hour average welfare standard)</td>
<td>139$^a$ (health standard)</td>
<td>31 (health standard)</td>
</tr>
<tr>
<td>Yugoslavia</td>
<td>191</td>
<td>57</td>
<td>15-23</td>
</tr>
<tr>
<td>European Economic Community</td>
<td>Percentile of 24 hour means percentile Limit</td>
<td>50 - annual 31-46</td>
<td>Guide value 15-23 for daily arithmetic mean averaged over 1 yr.</td>
</tr>
<tr>
<td></td>
<td></td>
<td>50 - winter 50-69</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>98 - annual 95-134</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>&lt;3 consecutive days</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Guide value 38-57 for daily peak mean</td>
<td></td>
</tr>
<tr>
<td>World Health Organisation</td>
<td>38</td>
<td>15-23</td>
<td></td>
</tr>
</tbody>
</table>

$^a$ not to be exceeded more than once per year
human welfare). The much quoted EEC standards are for the protection of human health and are derived primarily from those established by WHO and the USEPA (Saunders, 1985).

2.2.4 NO\textsubscript{x} Dose-Response Data

The effects of nitrogen oxides on plants at any level, have almost exclusively only been considered in relation to NO\textsubscript{2}. One reason often given for this is the spontaneous conversion of NO to NO\textsubscript{2} in air under experimental conditions (Mansfield and Freer-Smith, 1981). In addition, at a cellular level it is difficult to distinguish the effects of NO and NO\textsubscript{2} as both form nitrate (NO\textsubscript{3}\textsuperscript{-}) and nitrite (NO\textsubscript{2}\textsuperscript{-}) ions upon dissolution (Wellburn, 1988).

Differences in toxicity of NO and NO\textsubscript{2} are thought to primarily relate to their rates of uptake (Mansfield and Freer-Smith, 1981). Ultimately NO and NO\textsubscript{2} interact with the moist cell walls of plant tissue. Therefore, attention has been given to assessing the relative toxicities of these gases based upon their solubility equilibria in water (Garsed, 1984). NO\textsubscript{2} is highly soluble while NO is only sparingly soluble and for this reason NO has often been considered to be far less toxic to plants than NO\textsubscript{2} (Garsed, 1984).

However, it has been shown for NO\textsubscript{2} and NO that their rate of uptake into plants is not determined purely by their solubility equilibria in water. Fluxes of NO into leaves of sweet pepper (Capsicum annuum) have been found to be only two to three-fold less than those of NO\textsubscript{2} (Law and Mansfield, 1982) despite the large differences in solubility of the two gases. There is strong evidence to suggest that dissolution of these gases in plant solutions is enhanced by the presence of the solutes of plant metabolism (Garsed, 1984). A limited number of studies have shown the effects of NO and NO\textsubscript{2} on photosynthetic rate to be of similar magnitude (Capron and Mansfield, 1976) and additive (Hill and Bennett, 1970). Without further research it must be conservatively assumed that a given concentration of NO has a similar magnitude of effect as the same concentration of NO\textsubscript{2} for the same exposure time, although effects and mechanisms of action at the biochemical and physiological level may be quite different. Wellburn (1990) notes that no beneficial (growth stimulation) effects have ever been shown to result from NO fumigation.

From a review of the literature two other features of the research into the effects of NO\textsubscript{x} on plants became obvious:

1. The majority of studies on the effects of NO\textsubscript{2} alone used unrealistically high concentrations; and

2. Emphasis in the more recent literature has been on the effect of NO\textsubscript{2} in conjunction with SO\textsubscript{2}. Amundson and MacLean (1982) state that except in specific urban locations (for example, Los Angeles, USA) NO\textsubscript{2} concentrations by themselves pose little or no threat to crop productivity.
The data-base for NO₂ effects is rather limited, and collated dose-response relationship data does not exist, as it does for SO₂. The findings of some NO₂ studies are outlined below.

As has been stated previously low concentrations of NO₂ can stimulate plant growth. Mansfield et al., (1982) demonstrated growth stimulation in *Phleum pratense* when exposed to 68 nLL⁻¹ NO₂ for 20 weeks. Whitmore and Freer-Smith (1982) studied another grass, *Foa pratensis*, under similar fumigation conditions, that is 62 nLL⁻¹ NO₂, and found that growth stimulation occurred for the first 11 months, but later in the exposure period growth reduction was evident. This could possibly be explained by a nutrient imbalance having resulted as a consequence of the initial increase in growth.

Godzik et al. (1985) studied six cultivars of radish and concluded that 500 nLL⁻¹ NO₂, for a three hour exposure period, had no effect on plant growth. Gupta and Sabaratnam (1988) similarly exposed soya-bean plants to 500 nLL⁻¹ NO₂ but for seven hours and found a 34% yield reduction. A 60% yield reduction was the response to 1000 nLL⁻¹ NO₂.

The 1-h average National Ambient Air Quality Objective (Canada) for NO₂ is 210 nLL⁻¹. Goodyear and Ormrod (1988) studied the effect of this dose on tomato. They concluded that the objective was adequate to protect vegetation from NO₂ but possibly was not adequate if SO₂ was also present.

Pawloski Sinn and Pell (1984) exposed potatoes to a similar concentration, that is 200 nLL⁻¹ NO₂, but for a longer duration, five hours twice weekly throughout a growing season. They reported an accelerated rate of leaf abscission and reduced tuber number and weight.

Whitmore and Freer-Smith (1982) documented the effects of exposure to 62 nLL⁻¹ NO₂ for 150 days on six broad-leaved trees. They found that only one species displayed a significant increase in growth. The others did show a slight increase when compared with the controls, although this was not significant. Kress et al., (1982) found no effect from 100 nLL⁻¹ NO₂ over 28 days on American sycamore seedlings throughout the exposure period, but the fumigated plants displayed growth stimulation after the experiment ceased.

Continuous exposure of navel orange trees to 64-250 nLL⁻¹ NO₂ for 190 days resulted in a reduction in both the number and weight of fruit at harvest (Thompson et al., 1971). Istas (1980, as cited by Leone 1985/6) suggests that the threshold concentration of NO₂ for injury to apple and pear trees is 800 nLL⁻¹.

From the above discussion the response variation between plants becomes evident. Response varies even between very closely related species, fumigated under the same conditions. Ashenden and Mansfield (1978) found the relative sensitivity of four grasses to be different when exposed to 68 nLL⁻¹ for 20 weeks. Two species showed no effect whilst two showed a growth reduction.
The degree of sensitivity is partly dependent upon plant metabolism. NO₂ entering a leaf dissolves in the extra cellular water and ultimately dissociates into nitrite and nitrate ions (Zeevart, 1976). The effectiveness of the plant metabolism in assimilating or transforming these dissociation products affects its capacity for NO₂ resistance. Evidence exists that the nitrates and nitrites are metabolized by normal mechanisms of nitrogen metabolism (Zeevart, 1976). Hence, the initial stimulatory effect of NO₂ in most plants. The enzymes responsible for the detoxification of NO₂ products are nitrite and nitrate reductase. The localization of nitrate reduction in plants may be linked to their NO₂ susceptibility. In several woody species nitrate reduction is maintained almost exclusively in the roots (Lee and Stewart, 1978 as cited by Amundson and MacLean, 1982). This may make these species more sensitive to NO₂ injury in their leaves.

Irving (1987) states that NO₂ effects on plants are generally not observed until concentrations repeatedly reach 200 nLL⁻¹. These concentrations are rare, and hence, this supports the idea that NO₂ alone poses little threat to vegetation.

2.2.4.1 Existing Standards and Guidelines

The current NHMRC standard for NO₂ is 160 nLL⁻¹ (1-h maximum). The USEPA annual mean standard is 50 nLL⁻¹ NO₂ and the Canadian National Ambient Air Quality Objective is a 1-h average of 210 nLL⁻¹ NO₂. The more recently suggested guideline for NO₂ proposed by WHO(1987) takes the presence of SO₂ into account.

2.2.5 SO₂ and NOₓ Interactions

The interaction of pollutants can induce a response in plants that is either antagonistic (less than additive), additive or synergistic (greater than additive). When plants are exposed to SO₂ and NO₂ simultaneously, a synergistic response is normally detected. This is true for both long-term and short-term exposures (WHO, 1987). Table 4 summarizes the results of interaction studies on four grasses. Tingey et al. (1971) reported synergistic effects of SO₂ and NO₂ at concentrations of approximately 100-200 nLL⁻¹ in soybean, oats, pinto bean, radish, tomato and tobacco. Bennett et al. (1975) reported synergistic effects in radish of a one hour exposure to 500 nLL⁻¹ of both gases.

A study by Godzik et al. (1985) clearly showed the synergistic effect of the combination of SO₂ (150 nLL⁻¹) and NO₂ (210 nLL⁻¹). They exposed six cultivars of radish for a duration of 20 days to NO₂ and SO₂ separately and the combination of SO₂ and NO₂. They reported that NO₂ had no effect on plant growth in any of the cultivars. SO₂ reduced total dry weight in two of the
TABLE 4: Increase or decrease (relative to controls in clean air) in the growth of four grasses exposed for 140 days in winter to atmospheres containing nitrogen dioxide, sulphur dioxide or both. (WHO, 1987).

<table>
<thead>
<tr>
<th>Plant and parameter:</th>
<th>Increase or decrease (%) in growth after exposure to:</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>68 nLL⁻¹ NO₂</td>
</tr>
<tr>
<td>Dactylis glomerata L.</td>
<td></td>
</tr>
<tr>
<td>leaf area</td>
<td>+21</td>
</tr>
<tr>
<td>dry wt green lvs</td>
<td>-7</td>
</tr>
<tr>
<td>dry wt roots</td>
<td>-11</td>
</tr>
<tr>
<td>Poa pratensis L.</td>
<td></td>
</tr>
<tr>
<td>leaf area</td>
<td>-17</td>
</tr>
<tr>
<td>dry wt green lvs</td>
<td>-29</td>
</tr>
<tr>
<td>dry wt roots</td>
<td>-17</td>
</tr>
<tr>
<td>Lolium multiflorum L.</td>
<td></td>
</tr>
<tr>
<td>leaf area</td>
<td>+1</td>
</tr>
<tr>
<td>dry wt green lvs</td>
<td>-10</td>
</tr>
<tr>
<td>dry wt roots</td>
<td>+35</td>
</tr>
<tr>
<td>Phleum pratense L.</td>
<td></td>
</tr>
<tr>
<td>leaf area</td>
<td>+30</td>
</tr>
<tr>
<td>dry wt green lvs</td>
<td>+14</td>
</tr>
<tr>
<td>dry wt roots</td>
<td>+1</td>
</tr>
</tbody>
</table>
cultivars. The combination of \( \text{SO}_2 \) and \( \text{NO}_2 \) reduced the total dry weight in all six cultivars.

Wright (1987) conducted a similar study on birch clones, using lower concentrations over a longer period than in Godzik's experiments. After one year's exposure to 62 nLL\(^{-1}\) \( \text{NO}_2 \) no effect was detected. Stem diameter and dry weight increment were reduced by 62 nLL\(^{-1}\) \( \text{SO}_2 \). The combination of \( \text{SO}_2 \) and \( \text{NO}_2 \) reduced these parameters further.

Godzik et al. (1985) noted that pretreatment with \( \text{NO}_2 \) predisposed plants to \( \text{SO}_2 \) injury. This implies that it is not necessary for concentrations of \( \text{SO}_2 \) and \( \text{NO}_2 \) to peak simultaneously for an interactive effect to be displayed.

Mansfield et al. (1982) were able to produce a dose response curve for the effect of \( \text{SO}_2 \) and \( \text{NO}_2 \) on \( \text{Poa pratensis} \). Plants were exposed for various times to equivalent concentrations of the gases, ranging from 40-100 nLL\(^{-1}\). They noted a growth stimulation at the lowest concentration for the shortest time, that is 40 nLL\(^{-1}\) \( \text{SO}_2 \) + 40 nLL\(^{-1}\) \( \text{NO}_2 \) for 20 days. Inhibition of growth occurred at higher concentrations and/or longer periods. Whitmore (1985) also produced a \( \text{SO}_2 \) + \( \text{NO}_2 \) dose response curve for \( \text{Poa pratensis} \) over a range of concentrations and exposure periods as shown in Figure 5. This displayed an initial stimulation of growth followed by inhibition with a threshold dose of approximately 1500 nLL\(^{-1}\)\(d\) for each gas.

One of the main reasons suggested for the synergistic effect of \( \text{NO}_2 \) + \( \text{SO}_2 \) is the effect of \( \text{SO}_2 \) on nitrogen metabolism. When plants are exposed to both gases the formation of nitrate or nitrite can be inhibited. Nitrite reductase activity commonly rises with exposure to \( \text{NO}_2 \) alone. The presence of \( \text{SO}_2 \), however, can limit or even prevent this substrate-related induction of the reductase enzyme (Wellburn, et al., 1981; Robinson and Wellburn, 1983). The importance of the presence of \( \text{SO}_2 \) becomes very evident, and hence, the reason for its inclusion in recent air quality guidelines for \( \text{NO}_2 \). A Dutch National Health Council study on short-term effects of \( \text{NO}_2 \) led to the conclusion that sensitive plants are adequately protected if the four hour average does not exceed 50 nLL\(^{-1}\) \( \text{NO}_2 \), in the presence of similar \( \text{SO}_2 \) concentrations (Gezondheidsraad, 1979, as cited WHO, 1987). Freer-Smith and Mansfield (1987) concluded from a study of \( \text{Picea sitchensis} \) that 30 nLL\(^{-1}\) \( \text{SO}_2 \) with 30 nLL\(^{-1}\) \( \text{NO}_2 \) over several weeks are doses which fall below the threshold for chronic injury in this species.

In a long-term effects study, Zierock et al. (1986, as cited WHO, 1987) suggested that the threshold of injury in plants may be as low as 15 nLL\(^{-1}\) \( \text{NO}_2 \) when there are similar amounts of \( \text{SO}_2 \) present.
Figure 5: Dose-response relationship of *Poa pratensis* exposed to SO₂ and NO₂ mixtures. Plants were exposed under controlled environment conditions to 7 nLL⁻¹(control), 40 nLL⁻¹(●), 70 nLL⁻¹(▲) and 100 nLL⁻¹(■) of each gas for periods of 4 to 50 days. (Whitmore, 1985).
2.2.5.1 Existing Standards and Guidelines

WHO (1987) reviewed the literature available on the effects of nitrogen oxides on vegetation alone and in combination with other pollutants. The following guideline was proposed:

"In the presence of levels of sulphur dioxide and ozone not higher than 11 nLL\(^{-1}\) and 31 nLL\(^{-1}\) respectively, the atmospheric concentration of nitrogen dioxide should be no higher than 16 nLL\(^{-1}\) as a yearly average of 24-h means and no higher than 50 nLL\(^{-1}\) as a 4-h average" (WHO, 1987).

2.2.6 Dose-response Data for Australian Vegetation

The dose-response data that exists throughout the world for various species cannot validly be used in establishing air quality guidelines for Australian plants under Australian conditions. However, the major difficulty in collating dose-response data for Australian plants is that the data base is exceptionally small.

The only research to have been done in Australia that has resulted in the establishment of a dose-response relationship for some Australian crops is that by Murray and Wilson (1989b). To examine the relationship between SO\(_2\) concentration and crop yield, five crops (wheat, soybean, peanut, navy bean and maize) were grown from seedling to harvest in open top chambers, under ambient climatic conditions. SO\(_2\) was introduced into the chambers for 8 h/day at concentrations of about <0.4 nLL\(^{-1}\), 52 nLL\(^{-1}\) or 107 nLL\(^{-1}\). The responses of the plants varied. Wheat and soybean were very sensitive, with yield reductions of about 5% and 25% at 51 and 103 nLL\(^{-1}\), respectively. Navy beans, maize and peanuts were less sensitive with navy beans and maize showing a yield increase of about 10% at 53 nLL\(^{-1}\) and unchanged yield at 107 nLL\(^{-1}\). Peanuts showed yield reductions of about 5% and 10% at 53 nLL\(^{-1}\) and 105 nLL\(^{-1}\), respectively. These relationships between SO\(_2\) concentration and yield are shown in Figure 6. The dotted line represents a generalised concentration-response curve for these species with a threshold for zero yield at approximately 60 nLL\(^{-1}\) SO\(_2\) due to exposure for 8 h/day, daily over the growing season. This is higher than either of the thresholds for crop species and the single species Lolium perenne established by Roberts (1984a), however, these thresholds were established from continuous exposure regimes rather than intermittent frequent exposures.

At the commencement of this ERDC funded project there was inadequate information to establish a reliable dose-response relationship for SO\(_2\) for Australian native trees or Australian plant species in general and the effects of NO\(_x\) on Australian plants under Australian conditions had not been studied at all, either as a single pollutant or in conjunction with SO\(_2\). However, one study on the response of Araucaria cunninghamii (Hoop pine) to SO\(_2\) and NO\(_x\)
Figure 6: SO$_2$ concentration-response relationships for 5 crop species: wheat (□), navy beans (■), peanuts (★), soybeans (○) and maize (●). A generalized curve for the data is represented by (☆). (Murray and Wilson, 1988).
was conducted during 1989 (Murray et al., 1990a) in conjunction with this project using funding provided by the Queensland Electricity Commission.

Our research group is the only one to date to have studied Australian native species and crops under field climatic conditions. Table 5 lists the data-base available from all other research conducted by our group. This has been primarily concerned with the effects of SO₂.

Several other researchers have studied Australian native species but not under field conditions. O’Connor et al. (1974) studied visible injury in response to SO₂ exposure for a number of Australian species. He exposed 131 tree and shrub species to SO₂ in temperature controlled growth cabinets. The exposure regimes were as follows:

- 300 nL⁻¹ for 27-h
- 1000 nL⁻¹ for 3 or 6-h
- 2000 nL⁻¹ for 4-h
- 3000 nL⁻¹ for 0.5, 1, 2, 4 or 6-h

No evidence of acute or chronic visible injury was found at 300 nL⁻¹ for 27 hours. The most sensitive species all showed visible injury after three hours exposure to 1000 nL⁻¹ and were all species of Eucalyptus. Casuarina was the most resistant genus.

In an overseas study Howe and Woltz (1981) screened 43 ornamental foliage plants for sensitivity to airborne SO₂. They also categorized Eucalyptus as possessing high susceptibility to acute SO₂ injury.

Many species of Eucalyptus have adaptations for arid and nutrient poor environments including sclerophyllous evergreen leaves with thick cuticles, sunken stomata protected by hairs or waxy outgrowths and multi-chambered stomatal cavities. These anatomical and morphological adaptations tend to reduce gaseous exchange. However this does not appear to enhance the resistance of Eucalyptus species to air pollutants. This may be the result of low stress tolerance to air pollutants at a metabolic level or due to the poor stomatal control displayed by some Eucalyptus species (Colquhoun et al., 1984). Another overseas study, which fumigated Casuarina cunninghamiana and an Egyptian variety of E. camaldulensis (Elkley and Ormrod, 1987) found that Eucalyptus plants were very sensitive to SO₂ and NO₂ but, Eucalyptus plants absorbed less gas per unit area of leaf than did Casuarina. This indicates that gaseous exchange rates are not necessarily important in determining the susceptibility of plants to air pollutants.
TABLE 5: Additional data currently available on the effects of SO$_2$ or SO$_2$ and NO$_x$ on Australian native species and crops as established using open-top chambers under field conditions.

<table>
<thead>
<tr>
<th>Pollutant</th>
<th>Species Studied</th>
<th>Pollutant Conc.(mean) &amp; Duration of Exposure</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>SO$_2$</td>
<td>Eucalyptus crebra</td>
<td>0, 43 and 83 nLL$^{-1}$ 24h/day for 40 days</td>
<td>Murray (1984 a)</td>
</tr>
<tr>
<td></td>
<td>Eucalyptus molucaana</td>
<td></td>
<td></td>
</tr>
<tr>
<td>SO$_2$</td>
<td>Trifolium subterraneum (Sub-clover) cv Wooramullup</td>
<td>0, 27, 63 nLL$^{-1}$ 24 h/day for 48 days</td>
<td>Murray (1984 b)</td>
</tr>
<tr>
<td></td>
<td>Loliwm perenne (Ryegrass) cv Tetrarite</td>
<td></td>
<td></td>
</tr>
<tr>
<td>SO$_2$</td>
<td>Medicago sativa (Lucerne) cv CUF101</td>
<td>0, 30, 82 nLL$^{-1}$ 24 h/day for 166 days</td>
<td>Murray (1985a)</td>
</tr>
<tr>
<td>SO$_2$</td>
<td>Trifolium repens (White Clover) cv Regal</td>
<td>0, 30, 82 nLL$^{-1}$ 24 h/day for 63 days</td>
<td>Murray (1985b)</td>
</tr>
<tr>
<td>SO$_2$</td>
<td>Eucalyptus calophylla</td>
<td>0, 47 and 103 nLL$^{-1}$ 24 h/day for 120 days</td>
<td>Murray &amp; Wilson (1988 a, b)</td>
</tr>
<tr>
<td></td>
<td>Eucalyptus marginata</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Eucalyptus gomphocephala</td>
<td></td>
<td></td>
</tr>
<tr>
<td>SO$_2$</td>
<td>Eucalyptus tereticornis</td>
<td>0, 50 and 102 nLL$^{-1}$ 24 h/day for 90 days</td>
<td>Murray &amp; Wilson (1988a, c,d)</td>
</tr>
<tr>
<td></td>
<td>Triticum aestivum (Wheat) cv. Halberd</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Hordeum vulgare (Barley) cv. Clipper</td>
<td></td>
<td></td>
</tr>
<tr>
<td>SO$_2$</td>
<td>Arachis hypogaea (Peanut) cv Virginia Bunch</td>
<td>0, 52 and 108 nLL$^{-1}$ 8h/day for 104 days</td>
<td>Murray &amp; Wilson (1988a, 1989b, 1990a)</td>
</tr>
<tr>
<td></td>
<td>Zea mays (Maize) cv QK958</td>
<td>0, 52 and 108 nLL$^{-1}$ 8h/day for 70 days</td>
<td>&quot; &quot;</td>
</tr>
<tr>
<td>SO$_2$</td>
<td>Glycine max (Soybean) cv Dragon</td>
<td>0, 53 and 106 nLL$^{-1}$ 8h/day for 91 days</td>
<td>&quot; &quot;</td>
</tr>
<tr>
<td>SO$_2$</td>
<td>Phaseolus vulgaris (Navybean) cv Gallaroy</td>
<td>0, 54 and 111 nLL$^{-1}$ 8h/day for 51 days</td>
<td>&quot; &quot;</td>
</tr>
<tr>
<td>SO$_2$</td>
<td>Triticum aestivum (Wheat) cv. Eradu</td>
<td>0, 51 and 102 nLL$^{-1}$ 8h/day for 115 days</td>
<td>Davison (1987) Davison et al (1990)</td>
</tr>
<tr>
<td>SO$_2$</td>
<td>Eucalyptus rudis</td>
<td>0, 50 and 105 nLL$^{-1}$ 8h/day for 123 days</td>
<td>Clarke (1987) Clarke &amp; Murray (1990)</td>
</tr>
<tr>
<td></td>
<td>Eucalyptus calophylla</td>
<td></td>
<td></td>
</tr>
<tr>
<td>SO$_2$</td>
<td>Eucalyptus gomphocephala</td>
<td>0, 38 and 115 nLL$^{-1}$ 24h/day for 126 days</td>
<td>Fulford (1988 ) Fulford &amp; Murray (1990)</td>
</tr>
<tr>
<td>SO$_2$/NO$_x$</td>
<td>Araucaria cunninghamii (Hoop pine)</td>
<td>0.96, 167 nLL$^{-1}$ SO$_2$ 0.76 nLL$^{-1}$ NO$_2$ 2.5 h/day for 112 days</td>
<td>Murray et al (1990a)</td>
</tr>
<tr>
<td>SO$_2$</td>
<td>Acacia saligna</td>
<td>0, 23, 36, 67, 181 nLL$^{-1}$ 2.5 h, 3 days/wk for 20 wks 56 nLL$^{-1}$ 1 h/wk for 20 wks 67 nLL$^{-1}$ 1 h/mth for 20 wks 376 nLL$^{-1}$ 1 h only</td>
<td>Murray et al (1990b)</td>
</tr>
<tr>
<td></td>
<td>Banksia attenuata</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Banksia menziesii</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Eucalyptus wandoo</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Hakea incrassata</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

a  NO to NO$_2$ ratio 1:9
2.3 MATERIALS

There is currently no doubt amongst scientists that $\text{SO}_2$ and $\text{NO}_x$ cause damage to materials used in man-made structures. The extent of this damage, however, is still not accurately quantifiable.

When considering secondary air quality standards and cost-benefit analyses of air pollution control options, ideally economic assessments of materials damage need to be made. However, there is insufficient information available at present to accurately make such assessments and some believe accurate economic assessments are largely unobtainable for most materials.

Franey and Graedel (1985) claim that despite decades of study the mechanisms responsible for corrosion are still not well characterized. This is in part a consequence of their complexity. Webster and Kukacka (1986) state that while the literature notes that pollutants have adverse effects on building materials, little work is being done to quantify and explain these effects. They claim that the lack of understanding is actually due to a lack of research.

There has been some research on the effects of $\text{SO}_2$ on materials, but the role of $\text{NO}_2$ in materials degradation has so far been investigated only on a limited scale and NO is rarely considered (Johansson et al., 1988; Kucera, 1986).

The following discussion is broadly divided into three sections:

1. the environmental factors influencing the attack rate of $\text{SO}_2$ and $\text{NO}_2$;
2. a summary of the available information on the effects of $\text{SO}_2$ and $\text{NO}_2$ on 19 materials or materials classes; and
3. a discussion of cost assessment.

2.3.1 Factors Influencing Materials Degradation

Atmospheric pollution comes into contact with materials by either wet or dry deposition. Wet deposition entails removal of gases and aerosols from the atmosphere by precipitation, mist and fog, followed by deposition on the material surface. Dry deposition is contact between materials and airborne gases and particles.

Wet and dry deposition often produce different symptoms of decay. For example, wet deposition on marble produces white eroded surfaces whilst dry deposition produces black crusts on the surface (Torraca, 1988). The two types of deposition cause different rates of deterioration. Attack by the dry deposition of pollutants is a slow but continuous process. By contrast wet deposition is often sudden and infrequent with the pollutants being in
relatively dilute acid solutions (Fassina, 1988). In both cases the degradation of materials is largely the result of acid attack.

NO$_2$ reacts with hydroxyl radicals (OH) or H$_2$O to produce nitric acid:

1) $\text{NO}_2 + \cdot \text{OH} \rightarrow \text{HNO}_3$

2) $\text{NO}_2 + \text{H}_2\text{O} \rightarrow \text{HNO}_3 + \text{H}$_2$

Similarly SO$_2$ reacts with several compounds to produce sulphuric acid:

1) $\text{SO}_2 + \text{H}_2\text{O}_2 \rightarrow \text{H}_2\text{SO}_4$

2) $\text{SO}_2 + \text{O}_3 + \text{H}_2\text{O} \rightarrow \text{H}_2\text{SO}_4 + \text{O}_2$

3) $\text{SO}_2$ $\rightarrow$ $\text{SO}_3$

$\text{O}_2$

$\text{SO}_3 + \text{H}_2\text{O} \rightarrow \text{H}_2\text{SO}_4$

For more detail on the chemistry of SO$_2$ and NO$_2$ refer to Fassina (1988).

The production of acid can occur in the atmosphere as well as on the material surface. In polluted areas dry deposition of pollutants is usually more frequent than wet deposition and thus surface reactions are the most important.

Monitoring indicates that approximately 70% of the total deposition of air pollutants is via dry deposition (Sinclair and Weschler, 1986). In the drier areas of Australia this figure may be even higher. Corrosion rates decrease sharply with increasing distance from the emission source indicating that corrosion is largely the result of dry deposition rather than long range transport of pollutants in the form of acid rain (Kucera, 1986). Without moisture in the atmosphere, however, there would be little, if any, atmospheric corrosion even in the most severely polluted environments (Yocum and Upham, 1977; Patterson et al., 1986) due to the importance of moisture in the formation of acid solutions on surfaces. Temperature and sunlight are also important factors that influence the attack rate of SO$_2$ and NO$_2$.

2.3.1.1 Moisture

Material surfaces do not need to be visibly wet for deterioration to take place. Atmospheric humidity is of major importance. Yocum and Upham (1977) report that even at high pollutant levels it has been found that corrosion is minimal at relative humidities below 60%. As relative humidity approaches
80% the protective oxide film on metal surfaces breaks down allowing acid attack and corrosion to begin. At relative humidities above 80% corrosion proceeds rapidly (Sinclair and Weschler, 1986).

Rain exerts its influence in several ways. It acts by:

1. wetting the surface, hence facilitating the conversion of dry deposited SO₂ and NO₂ into their corrosive acid forms;

2. transporting corrosive hydrogen and sulphate ions to the surface;

3. washing away protective layers formed on the surface (Haynie, 1986a); and

4. washing away deposited pollutants from the surface.

The relative potential for materials damage of these effects depends upon the extent of the rainfall, the degree of pollution, the corrosion mechanism and the nature of the corrosion products (Kucera, 1986).

Mists and fogs can cause very rapid deterioration of materials because they provide sufficient water on a material's surface for SO₂ and NO₂ to dissolve and form acid but insufficient water to wash the surface clean. In addition they can collect pollutants over large distances, thus presenting concentrated acid solutions to surfaces.

In porous materials such as stone and marble, capillary rise of moisture is also important in determining the attack rates of SO₂ and NO₂.

2.3.1.2 Temperature

Temperature change effects the rate of degradation by pollutants of materials in two ways (Yocum and Upham, 1977):

1. it influences the rate of the chemical reaction deteriorating the surface; and

2. it influences surface temperature in relation to the dew point. When temperatures fall below the dew point the surface moistens and the rate of deterioration tends to increase.

2.3.1.3 Sunlight

Sunlight is important mainly due to its photochemical activity in the atmosphere. It can influence SO₂ and NO₂ attack rates directly or indirectly by:
1. determining the chemical reactions of SO\textsubscript{2} and NO\textsubscript{2} with other compounds, for example, the photochemical reactions of NO\textsubscript{2} during the day;

2. leading to the production of other damaging agents, such as ozone, which can act synergistically with SO\textsubscript{2} and NO\textsubscript{2}; or

3. causing direct deterioration of certain materials, the effect of which is often not distinguishable from pollutant damage.

2.3.1.4 Other Factors

Air movement determines the rate of supply of pollutants to a surface, affects the boundary layer resistance (Yocum and Upham, 1977) and influences the rate of evaporation. The latter either increases deterioration rates by concentrating acid solutions, or decreases rates by removing the surface moisture film (Rosvall, 1988).

The texture of the material surface determines the type of flow of moisture over the surface. Streams form on smooth surfaces, but flow is broken by heavily textured surfaces. This reduces erosion by water, but pools of acidic moisture may form (Fassina, 1988). The shape of the object also influences the type of flow. Generally, horizontal surfaces are more susceptible to pollution damage than vertical surfaces as deposited pollutants accumulate more readily (Yocum and Upham, 1977).

The order in which substances contact a surface can also be of importance. For example, if copper is initially exposed to unpolluted air a thin protective oxide film develops which resists subsequent pollutant degradation (Yocum and Upham, 1977).

Freezing and thawing can accelerate the pollutant induced deterioration of porous materials, such as concrete and building stone. The resultant cracks in the surface, caused by the physical stress, facilitate moisture and pollutant penetration (Yocum and Upham, 1977; Rosvall, 1988).

The presence of bacteria, fungi, mosses, terrestrial algae and lichens are of particular importance in the deterioration of stone and marble (Rosvall, 1988). They act by:

1. entrapping water molecules, hence prolonging the wetness of the material surface;

2. penetrating surfaces, and facilitating the entry of water and particles; and
3. producing organic acids. The presence of SO\textsubscript{2} increases the activity of thiobacilli (sulphur metabolizing bacteria), leading to increased organic acid production (Rosvall, 1988). Whether gaseous NO\textsubscript{2} is also biologically oxidized is not known. Rosvall (1988) claims that a definite relationship exists between the degradation of stone and the presence of nitrifying bacteria.

The presence of particulate matter also influences the attack rate of SO\textsubscript{2} and NO\textsubscript{2}. Hygroscopic particles such as sea salt prolong wetness of a material surface. Alkaline particles tend to neutralize acidic moisture films, and buffer the surface against the acidity of SO\textsubscript{2} and NO\textsubscript{2} (Lipfert, 1986).

SO\textsubscript{2} and NO\textsubscript{2} can also interact with one another in a variety of ways. Both synergistic and antagonistic effects have been reported under varying environmental conditions and on different materials.

Kucera (1986) reports on study in which the combination of SO\textsubscript{2} and NO\textsubscript{2} at 90% relative humidity caused a rapid corrosion of copper compared to that caused by the gases individually (Figure 7).

Exposure of copper and gold electronic contacts to NO\textsubscript{2} shows no corrosion effect but the synergistic effect of SO\textsubscript{2} and NO\textsubscript{2} is very pronounced (Johansson, 1984, as cited by Kucera, 1986). These synergistic effects are not restricted to the corrosion of metals. Figure 8 shows the same effects on limestone, marble and travertine. A possible explanation for this synergistic effect is that NO\textsubscript{2} oxidizes SO\textsubscript{2} to sulphuric acid:

\[
\text{SO}_2 + \text{NO}_2 + \text{H}_2\text{O} \rightarrow \text{H}_2\text{SO}_4(\text{aq}) + \text{NO}
\]

(Johansson et al., 1988; Kucera, 1986).

By contrast, SO\textsubscript{2} and NO\textsubscript{2} display an antagonistic interaction on the corrosion of galvanized steel. Upon exposure to the atmosphere a protective oxide film is produced on the surface of steel. SO\textsubscript{2} increases the solubility of this film whilst dissolution is reduced by NO\textsubscript{2} (Haynie, 1986a).

From the above discussion the complexity of determining the extent of damage attributable to SO\textsubscript{2} and/or NO\textsubscript{2}, and the conditions under which that damage occurs, becomes clear. In addition to this difficulty, quantitative assessments of deterioration are rarely obtainable, and qualitative assessments are relied upon. This has resulted in a lack of damage functions (dose-response relations) relating pollutant concentrations to materials degradation.
Figure 7: Corrosion of copper in air containing SO$_2$ and/or NO$_2$ at 90% relative humidity. a = 1.3 ppm SO$_2$. b = 3.0 ppm NO$_2$. c = 1.3 ppm SO$_2$ + 3.0 ppm NO$_2$. (Kucera, 1986).
Figure 8: Damage to polished samples of limestone, marble and travertine exposed to air containing SO₂ and/or NO₂ at 22% relative humidity. (Johansson, 1984).
2.3.2 Effects of $\text{SO}_2$ and $\text{NO}_x$ on Materials

Table 6 presents a "degradation potential" ranking of 19 material or material classes for exposure to $\text{SO}_2$ and $\text{NO}_2$ in an urban/industrial area based on that compiled by Graedal and McGill (1986). This ranking is determined by both the sensitivity of each material and typical ambient concentrations of $\text{SO}_2$ or $\text{NO}_2$ in an urban/industrial area relative to those concentrations required to cause deterioration. The ranks are from 1-7 (1 indicating the least potential for degradation and 7 the greatest). Water has been included in this table for comparison because of the major influence it may have, independently of $\text{SO}_2$ and $\text{NO}_2$, on the degradation of most materials. Interactive effects were not quantified but the manifestations of the damage are listed as they are similar for $\text{SO}_2$, $\text{NO}_2$ and water.

From Table 6 it can be concluded that air quality criteria which protect iron, zinc and stone from $\text{SO}_2$, and textiles and nickel from $\text{NO}_2$ would protect materials generally. However, dose-response relations for these materials are still being developed.

The difficulty in determining dose-response relations and ultimately deriving air quality criteria arises from the fact that the rate of deterioration depends not only on the chemical composition of the air but also on all the numerous factors discussed previously, in addition to the physical, mechanical and chemical characteristics of the material itself. Unlike vegetation, the rate of damage to materials can not be correlated directly to $\text{SO}_2$ and $\text{NO}_2$ concentration. Corrosion rates are often not constant with time but change after an initial period, increasing as the surface erodes or decreasing due to the formation of protective layers (Patterson et al., 1986). In addition, as materials lack a regenerative capacity and damage is cumulative there is no true threshold for effects. This implies that secondary air quality standards have to be based on an assessment of unacceptable deterioration and economic loss. Hence, the importance of cost assessment of materials damage.

2.3.3 Cost Assessment

Accurate determination of the cost of air pollution effects on materials is difficult to obtain, unlike crop yield loss, where a monetary value is easier to estimate (Yocum and Upham, 1977). Several issues have to be addressed when translating a qualitative assessment of damage into a quantitative assessment of cost. Four main steps need to be taken (after Yocum and Upham, 1977):

1. evaluation of the relative contribution of a particular pollutant or combination of pollutants to the damage;

2. determination of the extent of the damage;

3. assessment of the significance of the damage; and
TABLE 6: Degradation potential assessment based on material sensitivity and the concentrations of SO₂ and NO₂ typically present in an urban/industrial environment (after Graedel and McGill 1986). The effects of moisture are shown for comparison. 1 = least degradation potential, 7 = greatest degradation potential.

<table>
<thead>
<tr>
<th>MATERIAL</th>
<th>DEGRADATION POTENTIAL</th>
<th>MANIFESTATION</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>H₂O H₂SO₄ H₂NO₂</td>
<td></td>
</tr>
<tr>
<td>Silver a</td>
<td>3 1 2</td>
<td>silver sulphate tarnish forms protective layer</td>
</tr>
<tr>
<td>Copper ab</td>
<td>3 2 2</td>
<td>copper sulphate tarnish forms protective layer</td>
</tr>
<tr>
<td>Bronze c</td>
<td>3 - 2</td>
<td>corrosion, cracking</td>
</tr>
<tr>
<td>Brass b</td>
<td>3 - 2</td>
<td></td>
</tr>
<tr>
<td>Iron ab</td>
<td>7 2 3</td>
<td>corrosion (rust)</td>
</tr>
<tr>
<td>Tin</td>
<td>3 - 1</td>
<td>destruction of zinc carbonate protective layer</td>
</tr>
<tr>
<td>Lead</td>
<td>3 - 1</td>
<td></td>
</tr>
<tr>
<td>Zinc abdef</td>
<td>3 2 6</td>
<td>destruction of zinc carbonate protective layer</td>
</tr>
<tr>
<td>Nickel a</td>
<td>1 5 4</td>
<td>nickel sulphate tarnish</td>
</tr>
<tr>
<td>Aluminium abeg</td>
<td>3 4 5</td>
<td>white powdery deposit on surface</td>
</tr>
<tr>
<td>Stone ahiklmn</td>
<td>3 3 3</td>
<td>exfoliation, cracking, discoloration, erosion, gypsum formation</td>
</tr>
<tr>
<td>Rubber an</td>
<td>1 4 4</td>
<td>cracking, embrittlement, weakening, discoloration</td>
</tr>
<tr>
<td>Paint</td>
<td>1 - 2</td>
<td>discoloration, chalking, softened finish, erosion</td>
</tr>
<tr>
<td>Paper a</td>
<td>7 2 4</td>
<td>embrittlement</td>
</tr>
<tr>
<td>Photographs</td>
<td>1 3 4</td>
<td></td>
</tr>
<tr>
<td>Textiles ano</td>
<td>1 5 4</td>
<td>reduced tensile strength, discoloration</td>
</tr>
<tr>
<td>Leather a</td>
<td>1 - 5</td>
<td>powdered surface, loss of tensile strength, disintegration</td>
</tr>
<tr>
<td>Ceramics/Glass a</td>
<td>1 2 2</td>
<td>change of surface appearance</td>
</tr>
<tr>
<td>Electronic components ab</td>
<td>2 3 3</td>
<td>tarnishes, change in electrical resistance</td>
</tr>
</tbody>
</table>

4. evaluation of the response to the damage e.g. replacement, repair etc.

The costs involved are represented by either:

1. a shortened useful life;

2. the implementation of strategies to prolong useful life, such as:
   
a) over design;

b) the application of protective coatings;

c) the substitution of the material with a more robust and expensive material;

d) more regular cleaning and maintenance; and

e) protective measures indoors such as air conditioning (to reduce relative humidity) or air filtration/purification systems (after Yocum and Upham, 1977).

Also included in the overall costs of air pollution damage should be the cost of research into the effects and the more subtle, delayed economic effect of a more rapid consumption of limited resources (Haynie, personal communication).

The fundamental problem that exists in trying to assess the cost of SO₂ and NOₓ effects on materials is the porosity and uncertainty of the data available on materials damage by air pollution. Haynie (1986b) states that as a result of the variability of available data on paint deterioration, the magnitude of the error in any economic cost estimate will be the same as the estimate. This means the cost could be zero or twice as much as the estimate. Any assumptions made in the economic analysis increase this error further (Haynie, 1986b).

Torraca (1988) reviews the damaging effect of air pollutants on building materials. He claims that with our present knowledge it is impossible to establish accurate models for the deterioration of brittle and porous materials. It may however, be possible to formulate models which are valid for one set of standard microclimatic conditions, for a standard building material of a defined chemical and mineralogical composition, characterized by a standard specific surface (Torraca, 1988).

However, some materials are of cultural significance and are considered priceless and irreplaceable. Stone and marble deterioration is of major concern in Europe and Asia as existing technology lacks proven methods for preserving stone structures that are deteriorating (Lal Gauri and Holdren, 1981). It is very difficult to place a monetary value on damage to such
structures. Feddema and Meierding (1987) note that the most important strategy for reducing deterioration of stone buildings of cultural significance throughout the urban environment is to minimise \( \text{SO}_2 \) emissions as much as possible, as no threshold \( \text{SO}_2 \) concentration can be established.

Estimates obtained from the various methods of economic analysis do give an indication of the magnitude of the cost of air pollution damage for some materials. A conservative estimate of the cost of damage to such materials in Germany for 1980 (prepared by their Federal Agency for the Environment), is as follows:

- Damage to utility buildings: \( \text{US}\$500 \) million per annum
- Corrosion damage: \( \text{US}\$700 \) million per annum
- Additional cleaning costs: \( \text{US}\$300 \) million per annum

These estimates ignore costs of corrosion protection and damage to monuments and other objects of cultural significance (Schubert et al., 1986).

Zinc galvanized steel roofs are considered to be one of the most sensitive metal products to \( \text{SO}_2 \) damage. In the United Kingdom, farmers receive compensation for losses due to early replacement of their galvanized steel roofs. The amount of compensation is calculated according to the concentration of \( \text{SO}_2 \) in air (Pearce, 1985).

In the United States the total cost of \( \text{SO}_2 \) damage to bare galvanized steel roofing, siding and guttering was estimated to be \( \text{US}\$242 \) million dollars for 1980 (Haynie, personal communication). Based on 1974-75 figures, the cost of damage attributable to \( \text{SO}_2 \) due to the need for early replacement of zinc galvanized steel roofs in Melbourne is estimated to be \( \$33000 \) per annum (Martin, 1979).

Future research and economic assessment will slowly answer the question of how much damage is acceptable. The National Acid Precipitation Assessment Programme within the USEPA is currently conducting a major research programme on the effects of air pollutants on construction materials. An objective of this research is to attempt to develop damage functions (dose-response relations) for the major construction materials. The damage functions that emerge will have their applicability checked using corrosion data of standard specimens at experimental sites and by using historical corrosion data produced from both US and European studies. This will improve the database available for assessment of economic losses in Australia. At present insufficient information on economic loss due to materials damage from air pollutants is available in Australia to establish secondary ambient air quality standards that take into account this aspect of human welfare. However, it is likely that the establishment of standards based on scientific criteria for vegetation responses, will go a long way towards protecting against large economic losses from materials degradation.
CHAPTER 3: GENERAL METHODOLOGY

The fumigation concentrations, cultivation procedures, growth conditions and harvest procedures varied slightly for each experiment and therefore will be discussed under the relevant chapter for that experiment. Methodology common to all three experiments and the basis for the species selection are discussed below.

3.1 SPECIES SELECTION

Figure 9 shows the location of all coal-fired energy generating facilities in Australia and thus the areas subject to emissions of $SO_2$ and $NO_x$ from these operations. Based on Figure 9, ecological information and statistics on the economic value of agricultural and forestry crops in Australia, provided by the Agricultural Departments and Forestry Commissions in each state, the following species were selected for study.

3.1.1 Grain Crop Species: Wheat, Barley, Oats and Triticale

3.1.1.1 Wheat

In 1986 wheat had a gross annual value of $2789 million, comprising 37.4% of the gross value of principal crops and pastures in Australia (Australian Bureau of Statistics, 1987). It is grown near the power generating areas of New South Wales, Queensland and South Australia. These states account for 57% of the total wheat production in Australia, with NSW being the greatest producer. The most economically important cultivar in NSW is Banks which is also used extensively in the other states.

3.1.1.2 Barley

In 1986 barley had a gross annual value of $590 million, which comprised 8% of the gross value of principal crops and pastures (ABS, 1987). Barley is grown near the power generating areas of South Australia and Queensland. Production in these states accounts for 51% of Australia's total production of barley, with South Australia alone accounting for 35% (ABS, 1987). The most widely used cultivar in South Australia is Schooner, no current information on cultivar use could be obtained for Queensland.

3.1.1.3 Oats

The gross annual value of oats in 1986 was $185 million, $138 million of this as grain and $47 million as hay (ABS, 1987). It is of particular importance in the power generating areas of New South Wales, South Australia and Western Australia. These states account for 75% of total oat production in Australia (ABS, 1987). Information provided by the Agricultural Departments in these states showed that Echidna was the most widely used cultivar.
Figure 9: The location of energy generating facilities in Australia. (Electricity Supply Association of Australia, 1987)
3.1.1.4 Triticale

In 1986 triticale had a gross annual value of $28 million (ABS, 1987). It is grown mainly in New South Wales with the most important cultivar being Currency which is also widely used in Victoria.

3.1.2 Pasture Species: Lucerne, Sub-Clover, White Clover, Barrel Medic and Burr Medic.

A major land use near power stations in New South Wales, Victoria, Queensland, South Australia and Western Australia is that of improved grazing pasture. Both livestock and crop production increase as a result of the sowing of legumes in pastures to improve soil fertility. The major pasture legumes grown are lucerne, clovers and annual medics. The value of these species to agriculture in terms of soil nitrogen accumulation alone is very difficult to assess. In 1984 the nitrogen accumulation value of subterranean clover alone was estimated to be in the order of $400 million annually (Collins and Gladstones, 1984).

3.1.2.1 Lucerne

In 1986 lucerne had a gross annual value of $99 million, the majority of this as hay (ABS, 1987). Certified seed production figures show the cultivar Siriver to be by far the most important cultivar in use in Australia (Clark and Reed, 1988).

3.1.2.2 Clovers

In 1986 clovers had a gross annual value for seed alone of $10 million (ABS, 1987). The most important clover is subterranean clover (sub-clover) which is sown on an estimated 16 million hectares (Collins and Gladstones, 1984). The clover second to this in importance is white clover. The sub-clover cultivar most widely used is Trikkala and almost all white clover used in Australia is of the cultivar Haifa (Clark and Reed, 1988).

3.1.2.3 Annual Medics

An estimated 50 million hectares are sown to annual medics, 30 million hectares of which are in the wheat and sheep zones of Australia. *Medicago truncatula* (barrel medic) is the most important species followed by *M. polymorpha* (burr medic) (Clark and Reed, 1988). The two annual medics of most economic importance in Australia are barrel medic cv. Paraggio and burr medic cv Santiago (Clark and Reed, 1988).
3.1.3 **Tree Species:** *Eucalyptus pilularis, E. microcorys, E. regnans* and *Pinus radiata*

Emphasis was placed on selecting tree species that were both of ecological significance in native forests close to energy generating facilities and of economic importance to the forestry industry to ensure relevance of the data collected for future resource management and planning. Due to time constraints and the need to grow trees of a reasonable size before fumigation only four species could be selected. Although not native to Australia, *Pinus radiata* was selected as one of these due to its economic importance to the forestry industry as a plantation species occurring in the vicinity of energy generating centres. The remaining three species were selected from eastern states forests due to the concentration of energy generating facilities in the Hunter Valley, NSW, the Latrobe Valley, Victoria and the coastal regions of Queensland close to native forests.

3.1.3.1 *E. pilularis* (Blackbutt)

Blackbutt typically occurs on gentle slopes between the sea and coastal escarpment of the Great Dividing Range and is abundant from as far south as Bega in NSW to as far north as Maryborough and Fraser Island in Queensland (Boland *et al.*, 1984). This species is one of the most important hardwoods in Australia, and is the principal species sawn in coastal New South Wales and south-eastern Queensland (Boland *et al.*, 1984). In New South Wales it comprises 15% of the total volume of log timber (Forestry Commission of NSW, 1986).

3.1.3.2 *E. microcorys* (Tallowwood)

Tallowwood is widely distributed in northern NSW and southeastern Queensland. Its range extends from Newcastle, NSW to Maryborough and Fraser Island, Queensland (Boland *et al.*, 1984). This species comprises approximately 10% of the total volume of log timber in New South Wales (Forestry Commission of NSW, 1986), and is one of the most important logged species in Queensland (Queensland Department of Forestry, personal communication). It is considered to be one of the best native hardwood timbers (Boland *et al.*, 1984).

3.1.3.3 *E. regnans* (Mountain Ash)

Mountain Ash is one of the tallest forest species in the world occasionally exceeding 70 m and often grows in pure stands in open-forest formation (Boland *et al.*, 1984). It occurs in south-eastern Victoria and in Tasmania. It is one of the most important timber species in Victoria with the ash group alone comprising approximately 19% of the sawn timber in this state (Department of Conservation, Forests and Lands, 1987). This species is considered to be the best of the *Eucalyptus* species for use in the Australian pulp and paper industry (Boland *et al.*, 1984).
3.1.3.4 *Pinus radiata*

This species is the most important plantation species nationally, on both privately and publicly owned land. In total, *P. radiata* plantations cover nearly 600,000 hectares, comprising 68% of the total area of plantations. It is the major plantation species in New South Wales, Victoria, Western Australia, South Australia and Tasmania (Department of Primary Industry, 1985).

3.2 FUMIGATION CHAMBERS

Twenty cylindrical open-top chambers (Heagle *et al.*, 1973) were used for each fumigation experiment. Each chamber was 3 m in diameter and 2.4 m high, consisting of a rigid aluminium frame covered by UV-treated PVC plastic. The upper half of the frame was covered by a single layer of plastic and the lower half was covered by a double layer forming an envelope with the inner layer perforated by holes 25 mm in diameter. Air was drawn by a fan through a dust filter and then forced along a duct, into the chamber through the holes in the lower envelope and then out through the open top. The output of the fan was 1 m$^3$s$^{-1}$ enabling about 3.5 air changes per minute.

3.3 POLLUTANT DISPENSING AND MONITORING

Dry air was mixed with bottled anhydrous sulphur dioxide (SO$_2$) from a temperature-controlled cylinder and passed through a regulator and series of needle valves to the inlets of the required chambers. The concentration of SO$_2$ in every chamber was monitored using a Thermo Electron, Series 43 pulsed fluorescent ambient SO$_2$ analyser, calibrated with a Thermo Electron, Model 145 calibrator, with NBS traceable certified permeation tubes.

Bottled anhydrous nitric oxide (NO) was mixed with dry air in a cylinder to facilitate the oxidation of NO to NO$_2$ prior to delivery to the chambers for the first two experiments. In the third experiment a greater ratio of NO:NO$_2$ was required. To achieve this NO was delivered directly to the chambers using nitrogen as a carrier gas to minimise oxidation. The ratio of NO:NO$_2$ was controlled by the degree of dilution of NO in the carrier gas. In all three experiments the required amount of NO$_x$ was delivered to each chamber via a regulator and series of needle valves. The concentration of NO$_2$ and NO was monitored in each chamber using a Monitor Labs Chemiluminescent Nitrogen Oxides Analyzer, Model 8840, calibrated with a Thermo Electron calibrator using NBS traceable certified permeation tubes. Pollutant monitoring for both SO$_2$ and NO$_x$ was conducted with a time-sharing system.
3.4 EXPERIMENTAL DESIGN

The basic experimental design consisted of 10 treatments, each duplicated. Each experiment encompassed five different concentrations (levels) of SO₂ and two different concentrations (levels) of NOₓ. Level 1 was ambient air in each case. This design is shown in Table 7.

Table 7: The basic experimental design used in all three fumigation experiments.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Fumigation Level</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Level 1 ambient SO₂ + Level 1 ambient NOₓ</td>
</tr>
<tr>
<td>2</td>
<td>Level 2 SO₂ + Level 1 ambient NOₓ</td>
</tr>
<tr>
<td>3</td>
<td>Level 3 SO₂ + Level 1 ambient NOₓ</td>
</tr>
<tr>
<td>4</td>
<td>Level 4 SO₂ + Level 1 ambient NOₓ</td>
</tr>
<tr>
<td>5</td>
<td>Level 5 SO₂ + Level 1 ambient NOₓ</td>
</tr>
<tr>
<td>6</td>
<td>Level 1 ambient SO₂ + Level 2 NOₓ</td>
</tr>
<tr>
<td>7</td>
<td>Level 2 SO₂ + Level 2 NOₓ</td>
</tr>
<tr>
<td>8</td>
<td>Level 3 SO₂ + Level 2 NOₓ</td>
</tr>
<tr>
<td>9</td>
<td>Level 4 SO₂ + Level 2 NOₓ</td>
</tr>
<tr>
<td>10</td>
<td>Level 5 SO₂ + Level 2 NOₓ</td>
</tr>
</tbody>
</table>

Following review of the international literature and consultation with the Advisory Committee (see Section 1.1) the first two experiments, one on crops, the other on trees, were designed to establish the interactive effects of SO₂, and NOₓ consisting primarily of NO₂. NO₂ has long been considered the more important component of NOₓ emissions with regard to effects on plants. This is due to the oxidation of emissions of NO to NO₂ resulting in relatively high NO₂ concentrations at ground level (see Chapter 1). In addition, NO₂ is usually considered to be more toxic to plants than NO (Krause, 1988). As the project progressed the Advisory Committee reported that relatively high concentrations of NO were being recorded around energy generating facilities in Australia due to low NO to NO₂ conversion rates in stack emissions. This was seen as a cause for concern given the results from the first two experiments which had shown an unexpectedly high sensitivity of the species tested to NOₓ. Subsequently in the final experiment a higher NO:NO₂ ratio was utilised to elucidate the relative importance of NO and NO₂ in NOₓ emissions.
The pollutant concentrations for all three experiments were chosen to simulate worst case scenarios under conditions of inversion break-up and plume grounding. Thus, the decision was made to fumigate for only four hours per day between 1000 and 1400, seven days per week to simulate break-up of the atmospheric inversion layer which typically begins mid-morning. A wide range of SO$_2$ concentrations were used to ensure the threshold of response was determined for each species. Following analysis of the results from the first experiment a lower range of pollutant concentrations was chosen for subsequent experiments as the thresholds were clearly in this lower range.

3.5 ENVIRONMENTAL CONDITIONS

Meteorological recordings were obtained from the Perth Bureau of Meteorology from their recording station located in Wellington Street, East Perth. Mean daily maximum and minimum temperatures were calculated for the fumigation period of each experiment, in addition to mean daily relative humidities for the recording times used by the Bureau of 0900 and 1500. From previous experiments it has been established that mean temperatures are about 1°C higher inside the chambers than outside and relative humidity is about 1.5% lower inside the chambers than outside.

3.6 STATISTICAL ANALYSIS

Fumigation monitoring data were statistically analyzed using Statview 512+ (Abacus Concepts Inc, 1986). Analysis of Variance (ANOVA) was followed by a Scheffe's F-test of the means to indicate duplication of fumigation regimes. As data from duplicate chambers were not significantly different from one another, they were pooled for the descriptive statistics.

Statistical analysis of plant response used data from individual plants. Response variables were subjected to a one-way analysis of variance (ANOVA) followed by a Duncan's Multiple Range test to determine individual treatment effects at the 0.05 level of probability. Homogeneity of variance was tested using Cochran's C-test; where variances showed heterogeneity the data was transformed to achieve homogeneity. Statistical analysis of plant response data was performed using SPSS-X, version 3.0, installed on a Unix/Microvax system.
CHAPTER 4: THE EFFECTS OF SO₂ AND NOₓ ON WHEAT, BARLEY, LUCERNE, SUB-CLOVER AND BARREL MEDIC.

4.1 INTRODUCTION

Wheat and sheep production operate in conjunction in the southern regions of Australia in areas where the crop growing season is five to seven months long. Wheat production is usually alternated with subterranean clover or medic pastures for grazing. This rotation system is important as the clover and medic are nitrogen-fixing legumes which increase the nitrogen content of the soil between wheat crops.

In Australia, wheat is grown primarily for grain with small amounts being used for hay or direct grazing (Reid, 1990). Only wheaten flour has the type of protein required for high-quality, leavened bread-making. Thus, the main use of wheat grain is the production of flour for bread although biscuits, noodles, cakes and pasta products also use wheat flour. The percentage of protein in the grain is of prime importance in determining the suitability of the crop for bread-making. The common bread wheat, *Triticum aestivum*, has grain most suitable for this purpose. The best baking quality grain is that containing 13-14% protein or higher (Reid, 1990). Plant breeding has developed cultivars of *T. aestivum*, for example Banks, that meet this requirement although the quality of a given cultivar will vary from year to year and from location to location according to growth conditions.

Bread wheat cultivars fall into two categories, winter or spring, according to their growing season. Spring wheats are the most commonly grown types in Australia but because of our mild winter and hot summers are usually sown in autumn and early winter. This allows vegetative growth over winter and the development of heads in spring. This ensures that the grain matures before the onset of extreme summer temperatures (Reid, 1990). *T. aestivum* cv. Banks is a high yielding, quick maturing spring wheat.

Barley is the second most economically important crop species in Australia for grain production and can be grown in the same areas as wheat. Barley grain is used for malting and as animal feed and a small amount is also milled for direct human consumption. Barley may also be grown for forage and silage (green stored fodder). The primary use of barley is for malting. This is a process in which the grain is germinated to convert the starches to sugar. The greater the percentage of soluble sugars which can be developed in the malt, the better the malting quality of the barley. Malting requires starchy grain with a relatively low protein content, preferably less than 11% (Reid, 1990). Barley grown for grain is usually planted between April and July and due to its rapid growth and shorter growing season matures before wheat.

Sub-clover, lucerne and barrel medic are legumes that are important for improved pastures used for grazing throughout Australia. The term
improved pasture refers to a pasture which has been sown and artificially fertilized. Legumes increase soil fertility by adding nitrogen to the soil and have a high nutritive value for cattle and sheep as they are very rich in protein. Some fertiliser application is required for optimum growth as legumes used to increase soil-nitrogen are sensitive to phosphorus deficiency (Reid, 1990). Sown pastures and forage crops together are one of Australia's most important agricultural resources as they occupy 12% of non-arid agricultural land and provide most of the feed for the grazing industry. As indicated previously (section 3.1.2) the value of legumes to agriculture through nitrogen fixation and so soil improvement is extremely important to the industry but is very difficult to assess in monetary terms.

Lucerne and barrel medic are both species of *Medicago* with lucerne being the only truly perennial cultivated species of *Medicago*. Lucerne cv. Siriver is a relatively short lived, highly vigorous variety of lucerne which grows actively during winter and is primarily used for hay. Barrel medic is adapted to a wide range of drier wheat-belt regions and is moderately tolerant of high salinity. It is often sown under cereal crops to supply nitrogen during crop growth (Reid, 1990).

Subterranean clover is used extensively for pasture improvement in Australia as it is well adapted to the acid to neutral soils occurring in all temperate areas of Australia. Where soils are more alkaline and growing seasons shorter, annual medics are used. Most cultivars of sub-clover in use are naturally occurring genotypes, however, a few have been improved through breeding. The cultivar Trikkala is one of the breed varieties. It is tolerant of waterlogging and many pests and diseases.

### 4.2 Materials and Methods

**4.2.1 Fumigation Conditions**

Tables 8 and 9 display the characteristics of the fumigation regime used.
Table 8: Fumigation characteristics of the SO\textsubscript{2} exposure regime.
All concentrations are in nLL\textsuperscript{-1}.

<table>
<thead>
<tr>
<th>SO\textsubscript{2} Level</th>
<th>Fumigated 4 h arithmetic mean</th>
<th>Standard Deviation</th>
<th>Percentile</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>10th</td>
</tr>
<tr>
<td>1</td>
<td>&lt;5*</td>
<td>&lt;5</td>
<td>&lt;5</td>
</tr>
<tr>
<td>2</td>
<td>55</td>
<td>24</td>
<td>22</td>
</tr>
<tr>
<td>3</td>
<td>149</td>
<td>59</td>
<td>76</td>
</tr>
<tr>
<td>4</td>
<td>262</td>
<td>96</td>
<td>142</td>
</tr>
<tr>
<td>5</td>
<td>544</td>
<td>154</td>
<td>351</td>
</tr>
</tbody>
</table>

* below the detection limit of the SO\textsubscript{2} analyser.

Table 9: Fumigation characteristics of the NO\textsubscript{x} exposure regime.
All concentrations are in nLL\textsuperscript{-1}.

<table>
<thead>
<tr>
<th>NO\textsubscript{x} Level</th>
<th>NO\textsubscript{2} Concentrations</th>
<th>NO: NO\textsubscript{2}</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Fumigated 4 h arithmetic mean</td>
<td>Standard Deviation</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>&lt;5*</td>
<td>&lt;5</td>
</tr>
<tr>
<td>2</td>
<td>170</td>
<td>60</td>
</tr>
</tbody>
</table>

* below the detection limit of the NO\textsubscript{x} analyser

** n/a = not applicable

4.2.2 Cultivation Procedures and Growth Conditions

Seeds of the following species and cultivars were planted in peat Jiffy pots:

1. Wheat - *Triticum aestivum* cv. Banks;
2. Barley - *Hordeum vulgare* cv. Schooner;
3. Sub-clover - *Trifolium subterraneum* cv. Trikkala;
4. Barrel medic - *Medicago truncatula* cv. Paraggio; and

*Medicago* and *Trifolium* species seedlings were inoculated with a commercial rhizobium inocula (Nodulaid, Sydney) nine days after planting. Three weeks after sowing all species were removed from the glasshouse and 11 plants of each species were randomly assigned to each of the 20 chambers. The soil had been pretreated with the herbicide Roundup (Monsanto, active constituent Glyphosate) and mixed with a potting mix (2 sand: 3 sawdust: 3 wood fibre fines: 4 pinebark) and fertilizer containing both macro and micro nutrients.

Plants were sprayed once during the fumigation with the insecticide Rogor (Chemspray, active constituent Dimethoate) to protect against insect damage. Irrigation was used to supplement natural rainfall when necessary.

Plants were exposed to SO$_2$, NO$_x$ or their combination for four hours per day, seven days per week from 1000-1400 for 108 days between the months of August and December 1988.

The mean ambient daily maximum and minimum temperatures were 23.0°C and 13.4°C, respectively, and the mean ambient relative humidities at 0900 and 1500 were 61.2% and 50.2% respectively.

4.2.3 Harvest Procedure

At harvest, parameters to assess both yield and quality were measured.

Wheat and barley plants were cut at the soil line, separated into shoots and ears and then placed in an oven at 70°C and dried to constant weight. Shoot, ear and grain dry weight and ear and grain number were later measured.

Leaf material was ground to pass a 40 mesh sieve in a Wiley hammer mill prior to total sulphur determination using the oxygen flask combustion method of Hunt (1980). Grain was also ground prior to crude protein analysis using the standard Kjeldahl digestion and distillation determination procedures for total nitrogen. Protein concentration was calculated by multiplying total nitrogen values (%) by the standard factor 6.25 (AOAC, 1984).

The pasture species were cut at the soil line and the length of the longest branch recorded. The plants were then placed in an oven at 70°C and dried to constant weight. Foliar material was ground prior to total sulphur and crude protein determinations, as for the crop species.
4.3 RESULTS

4.3.1 Wheat cv. Banks (Figure 10)

**NO\textsubscript{x}**

Exposure of wheat to NO\textsubscript{x} (170 nL L\textsuperscript{-1} NO\textsubscript{2}) had a significant stimulatory effect on total plant dry weight due to an increase in grain production. Shoot weight and ear number did not increase. Both grain number and grain weight increased, the latter slightly more so resulting in a slight increase in average grain dry weight of 19%, although this was not statistically significant. Despite the increase in total grain weight per plant of 130%, grain protein concentration decreased by 19% showing an overall reduction in grain quality.

Foliar sulphur concentration was unaffected by exposure to NO\textsubscript{x}.

**SO\textsubscript{2}**

Exposure of wheat to SO\textsubscript{2} alone had little effect until concentrations reached 262 nLL\textsuperscript{-1}. At this point plants were barely surviving, grain production had virtually ceased, showing a 79% reduction, and plant biomass had reduced by 90%. At 544 nLL\textsuperscript{-1} the corresponding reductions were 97% and 94%.

Vegetative growth as represented by shoot weight was also decreased by exposure to 149 nLL\textsuperscript{-1} SO\textsubscript{2}. Not enough grain was available to analyse protein concentration at 544 nLL\textsuperscript{-1} SO\textsubscript{2}, however, at 262 nLL\textsuperscript{-1} SO\textsubscript{2} protein concentration in the small amount of grain produced was similar to that of the control plants. Grain protein concentration was reduced slightly by exposure to 149 nLL\textsuperscript{-1} SO\textsubscript{2}.

Foliar sulphur concentration was increased by about the same amount at all SO\textsubscript{2} concentrations considered.

**SO\textsubscript{2} + NO\textsubscript{x}**

At 55 nLL\textsuperscript{-1} and 149 nLL\textsuperscript{-1} SO\textsubscript{2} + NO\textsubscript{x}, total weight, ear weight, grain number and grain weight were significantly increased above the values recorded for SO\textsubscript{2} alone at these concentrations. An additive rather than interactive effect was evident. Ear number, average grain weight and shoot weight showed the same response to SO\textsubscript{2} + NO\textsubscript{x} as to SO\textsubscript{2} alone.

At 262 nLL\textsuperscript{-1} and 544 nLL\textsuperscript{-1} SO\textsubscript{2} + NO\textsubscript{x}, plant growth response was no different from that to SO\textsubscript{2} alone except that at 544 nL L\textsuperscript{-1} SO\textsubscript{2} + NO\textsubscript{x}, grain production was entirely prevented.

Grain protein concentration showed a steady increase in the presence of SO\textsubscript{2} + NO\textsubscript{x} as SO\textsubscript{2} concentration increased. At 149 nLL\textsuperscript{-1} and 262 nLL\textsuperscript{-1} SO\textsubscript{2} +
Figure 10: SO$_2$ and NO$_X$ Concentration-Response Relationships for Wheat cv. Banks Grown in Open-Top Chambers.

Plotted values represent arithmetic means of the response variables. The symbol -$\circ$- represents SO$_2$ only treatments and the symbol -$\rightarrow$- represents SO$_2$ + NO$_X$ treatments. The arithmetic mean NO$_2$ concentration was 170 nL/L and the NO:NO$_2$ ratio was 1:9. For each response curve, values that are labelled with the same letters are not significantly different; the letters w, x, y and z apply to the SO$_2$ treatments whilst the letters a, b, c and d apply to the SO$_2$ + NO$_X$ treatments. If no letters are present on a given curve there are no significant differences. A star (*) next to a value denotes that the SO$_2$ + NO$_X$ treatment is significantly different from the SO$_2$ only treatment of the same SO$_2$ concentration.
Figure 10 (continued)
Figure 10 (continued)
NO\textsubscript{x} protein concentration was not significantly different from the values recorded in the SO\textsubscript{2} only treatments at these concentrations. These results show a strongly interactive effect of SO\textsubscript{2} + NO\textsubscript{x} on grain protein concentration.

At 149 nLL\textsuperscript{-1} SO\textsubscript{2} the presence of NO\textsubscript{x} had no effect on the accumulation of sulphur in plant foliage. However, at 55 nLL\textsuperscript{-1}, 262 nLL\textsuperscript{-1} and 544 nLL\textsuperscript{-1} SO\textsubscript{2}, simultaneous exposure to NO\textsubscript{x} reduced foliar sulphur concentrations to control levels.

4.3.2 Barley cv. Schooner (Figure 11)

NO\textsubscript{x}

Exposure to NO\textsubscript{x} alone had no statistically significant effect on any of the parameters measured in barley, although average grain weight was increased by 12% and grain protein concentration was reduced by 8%.

SO\textsubscript{2}

A negative linear relationship existed between barley growth and SO\textsubscript{2} concentration with growth and yield reductions becoming statistically significant once the SO\textsubscript{2} concentration reached 262 nLL\textsuperscript{-1}. At this point total plant weight, ear weight and grain weight had decreased by 66%, 68% and 68%, respectively.

Grain protein concentration was unaffected at all SO\textsubscript{2} concentrations, whilst foliar sulphur concentration increased at 55 nLL\textsuperscript{-1} SO\textsubscript{2} and above. At 55 nLL\textsuperscript{-1} SO\textsubscript{2} foliar sulphur concentration had increased by 74% and at 544 nLL\textsuperscript{-1} SO\textsubscript{2} a 340% increase had occurred.

SO\textsubscript{2} + NO\textsubscript{x}

Growth stimulation was the main effect of simultaneous exposure of barley to SO\textsubscript{2} + NO\textsubscript{x}. The growth depression afforded by exposure to low SO\textsubscript{2} concentrations alone was ameliorated by the presence of NO\textsubscript{x}. The increase in total plant weight at SO\textsubscript{2} concentrations up to 149 nLL\textsuperscript{-1} was largely attributable to an increased production of grain. Shoot dry weight and ear number did not differ between the SO\textsubscript{2} only and SO\textsubscript{2} + NO\textsubscript{x} treatments but ear weight, grain weight, grain number and average grain weight increased significantly. These effects at 55 nLL\textsuperscript{-1} and 149 nLL\textsuperscript{-1} SO\textsubscript{2} + NO\textsubscript{x} were interactive (greater than additive) whereas at 255 nLL\textsuperscript{-1} and 544 nLL\textsuperscript{-1} SO\textsubscript{2} + NO\textsubscript{x} an additive reduction in growth and yield occurred due to simultaneous exposure to both gases.

Foliar sulphur concentration of barley exposed to SO\textsubscript{2} + NO\textsubscript{x} was significantly lower than that of plants exposed to SO\textsubscript{2} only. At 55 nLL\textsuperscript{-1} SO\textsubscript{2} with NO\textsubscript{x},
Figure 11: SO$_2$ and NO$_x$ Concentration-Response Relationships for Barley cv. Schooner Grown in Open-Top Chambers.

Plotted values represent arithmetic means of the response variables. The symbol $\text{-} -$ represents SO$_2$ only treatments and the symbol $\text{---} -$ represents SO$_2$ + NO$_x$ treatments. The arithmetic mean NO$_2$ concentration was 170 nL/L and the NO: NO$_2$ ratio was 1:9. For each response curve, values that are labelled with the same letters are not significantly different; the letters w, x, y and z apply to the SO$_2$ treatments whilst the letters a, b, c and d apply to the SO$_2$ + NO$_x$ treatments. If no letters are present on a given curve there are no significant differences. A star (*) next to a value denotes that the SO$_2$ + NO$_x$ treatment is significantly different from the SO$_2$ only treatment of the same SO$_2$ concentration.
Figure 11 (continued)
Figure 11 (continued)
sulphur concentration did not increase at all whereas a 74% increase in foliar sulphur concentration occurred due to exposure to 55 nLL\(^{-1}\) SO\(_2\) alone.

Grain protein concentration decreased by 21% at 55 nLL\(^{-1}\) and 149 nLL\(^{-1}\) SO\(_2\) + NO\(_x\) concomitant with the stimulation in grain production previously discussed. This suggests an inability of the plant to maintain grain quality while increasing grain biomass. A strongly interactive effect between SO\(_2\) and NO\(_x\) was evident resulting in greater than additive reductions in grain protein concentration at 55 nLL\(^{-1}\) and 149 nLL\(^{-1}\) SO\(_2\) + NO\(_x\). In the latter case, grain protein concentration was similar to that recorded in control plants but greater than that recorded due to exposure to NO\(_x\) alone.

4.3.3 Sub-clover cv. Trikkala (Figure 12)

NO\(_x\)

The response of subterranean clover to NO\(_x\) was a 56% reduction in shoot dry weight and a 14% reduction in protein concentration. Branch length was unaffected and there was no change in foliar sulphur concentration.

SO\(_2\)

Branch length was initially increased by exposure to SO\(_2\) and then decreased at 262 nLL\(^{-1}\) and 544 nLL\(^{-1}\) SO\(_2\). A maximum increase of 30% occurred at 149 nLL\(^{-1}\) SO\(_2\). Shoot dry weight was decreased at 149 nLL\(^{-1}\) and above. Protein concentration was increased by exposure to 262 nLL\(^{-1}\) and 544 nLL\(^{-1}\) SO\(_2\). Sulphur concentration appeared to increase linearly with SO\(_2\) concentration although not enough sample was available to determine sulphur concentrations for the 262 nLL\(^{-1}\) and 544 nLL\(^{-1}\) SO\(_2\) treatments due to the severe effects of these concentrations on growth.

SO\(_2\) + NO\(_x\)

In contrast to the initial stimulatory effect of SO\(_2\) on branch length, the combination of SO\(_2\) + NO\(_x\) reduced this parameter. Exposure to 55 nLL\(^{-1}\) SO\(_2\) alone increased length by 20% but 55 nLL\(^{-1}\) SO\(_2\) + NO\(_x\) decreased it by 14% showing a synergistic effect.

Biomass was markedly affected by the additional stress of the presence of NO\(_x\). Exposure to 55 nLL\(^{-1}\) SO\(_2\) had no effect on biomass but the addition of NO\(_x\) caused a 67% reduction this being of similar magnitude to the effects of NO\(_x\) alone. The 149 nLL\(^{-1}\) SO\(_2\) treatment caused a 19% reduction in biomass but with the addition of NO\(_x\) a 73% reduction was recorded. The reductions in shoot dry weight due to exposure to SO\(_2\) + NO\(_x\) were additive rather than interactive.
Figure 12: SO$_2$ and NO$_x$ Concentration-Response Relationships for Subterranean Clover cv. Trikkala Grown in Open-Top Chambers.

Plotted values represent arithmetic means of the response variables. The symbol $\rightarrow$ represents SO$_2$ only treatments and the symbol $\leftarrow$ represents SO$_2$ + NO$_x$ treatments. The arithmetic mean NO$_2$ concentration was 170 nL/L and the NO:NO$_2$ ratio was 1:9. For each response curve, values that are labelled with the same letters are not significantly different; the letters w, x, y and z apply to the SO$_2$ treatments whilst the letters a, b, c and d apply to the SO$_2$ + NO$_x$ treatments. If no letters are present on a given curve there are no significant differences. A star (*) next to a value denotes that the SO$_2$ + NO$_x$ treatment is significantly different from the SO$_2$ only treatment of the same SO$_2$ concentration.
Figure 12: (continued)
Sulphur accumulation was initially reduced by the presence of NO\textsubscript{x} at 55 nLL\textsuperscript{-1} SO\textsubscript{2} but at higher SO\textsubscript{2} concentrations NO\textsubscript{x} had no effect on sulphur accumulation. Protein concentration was no more affected by the presence of SO\textsubscript{2} + NO\textsubscript{x} than SO\textsubscript{2} alone.

4.3.4 *Barrel Medic cv. Paraggio* (Figure 13)

**NO\textsubscript{x}**

NO\textsubscript{x} alone reduced shoot dry weight by approximately 50%. The other parameters measured were unaffected.

**SO\textsubscript{2}**

Growth parameters displayed a negative linear relationship with increasing SO\textsubscript{2} concentration, whilst foliar sulphur concentration displayed a positive relationship. Protein concentration was increased by exposure to 262 nLL\textsuperscript{-1} SO\textsubscript{2}.

Foliar sulphur concentration was the only parameter affected by 55 nLL\textsuperscript{-1} SO\textsubscript{2}. A 42% reduction in biomass occurred due to exposure to 149 nLL\textsuperscript{-1} SO\textsubscript{2} and branch length was reduced at 262 nLL\textsuperscript{-1} SO\textsubscript{2} and above.

**SO\textsubscript{2} + NO\textsubscript{x}**

Simultaneous exposure to both SO\textsubscript{2} and NO\textsubscript{x} was substantially more damaging than exposure to either gas alone. In the case of branch length, exposure to both gases showed a greater than additive interactive effect. This reduced branch length substantially at 55 nLL\textsuperscript{-1}, 149 nLL\textsuperscript{-1} and 262 nLL\textsuperscript{-1} SO\textsubscript{2} + NO\textsubscript{x} to well below those values for SO\textsubscript{2} alone at these concentrations. In contrast, the effects of SO\textsubscript{2} + NO\textsubscript{x} on shoot dry weight were additive.

The response curves for SO\textsubscript{2} only and SO\textsubscript{2} + NO\textsubscript{x} were essentially the same for foliar sulphur concentration and protein concentration, although, sulphur accumulation was slightly lower due to exposure to 544 nLL\textsuperscript{-1} SO\textsubscript{2} + NO\textsubscript{x} compared to 544 nLL\textsuperscript{-1} SO\textsubscript{2} alone.

4.3.5 *Lucerne cv. Siriver* (Figure 14)

**NO\textsubscript{x}**

Exposure of lucerne to NO\textsubscript{x} had no statistically significant effect on any of the parameters assessed although shoot dry weight was decreased by 18% and foliar protein concentration increased slightly.
Figure 13: SO₂ and NOₓ Concentration-Response Relationships for Barrel Medic cv. Paraggio Grown in Open-Top Chambers.

Plotted values represent arithmetic means of the response variables. The symbol $\square$ represents SO₂ only treatments and the symbol $\Rightarrow$ represents SO₂ + NOₓ treatments. The arithmetic mean NO₂ concentration was 170 nL/L and the NO:NO₂ ratio was 1:9. For each response curve, values that are labelled with the same letters are not significantly different; the letters w, x, y and z apply to the SO₂ treatments whilst the letters a, b, c and d apply to the SO₂ + NOₓ treatments. If no letters are present on a given curve there are no significant differences. A star (*) next to a value denotes that the SO₂ + NOₓ treatment is significantly different from the SO₂ only treatment of the same SO₂ concentration.
Figure 13: (continued)
Figure 14: SO_2 and NO_x Concentration-Response Relationships for Lucerne cv. Siriver Grown in Open-Top Chambers.

Plotted values represent arithmetic means of the response variables. The symbol - - - represents SO_2 only treatments and the symbol - - - represents SO_2 + NO_x treatments. The arithmetic mean NO_2 concentration was 170 nL/L and the NO: NO_2 ratio was 1:9. For each response curve, values that are labelled with the same letters are not significantly different; the letters w, x, y and z apply to the SO_2 treatments whilst the letters a, b, c and d apply to the SO_2 + NO_x treatments. If no letters are present on a given curve there are no significant differences. A star (*) next to a value denotes that the SO_2 + NO_x treatment is significantly different from the SO_2 only treatment of the same SO_2 concentration.
Figure 14: (continued)
SO₂

Shoot dry weight was decreased by exposure to 262 nLL⁻¹ and 544 nLL⁻¹ SO₂, however, branch length was not reduced until the SO₂ concentration reached 544 nLL⁻¹. Protein concentration was increased by 544 nLL⁻¹ SO₂ but not the other concentrations considered.

Foliar sulphur concentration showed a quadratic response curve due to SO₂ exposure with no further increases in foliar sulphur concentration occurring above 262 nLL⁻¹ SO₂.

SO₂ + NOₓ

Shoot biomass was unaffected by exposure to 55 nLL⁻¹ and 149 nLL⁻¹ of SO₂ alone, however, with the addition of NOₓ, 45% and 46% reductions were observed respectively, showing a strong synergistic effect. This effect was also evident in branch length at 55 nLL⁻¹ and 262 nLL⁻¹ SO₂ + NOₓ. The response curve for protein concentration was unchanged by the presence of NOₓ with protein concentration increasing as SO₂ concentration increased. However, simultaneous exposure to SO₂ + NOₓ reduced the rate of sulphur accumulation in comparison to SO₂ exposure alone.

4.4 Discussion

Exposure to NOₓ, comprising primarily NO₂, at a mean concentration of 170 nLL⁻¹ NO₂ had no detrimental effect on barley or lucerne. However, NOₓ severely reduced the growth of both sub-clover and barrel medic. It also reduced grain protein concentration in wheat but not to below the 13-14% requirement for good bread making flour. In addition, this decrease in protein concentration in wheat was concomitant with a massive increase in grain production resulting in a greater total yield of protein. In sub-clover, both biomass and foliar protein concentration decreased resulting in a reduced and poorer quality feed stock for grazing animals.

Overseas research suggests that plants are not detrimentally affected by NO₂ until concentrations repeatedly reach 200 nLL⁻¹ (Irving 1987). At concentrations below this, growth stimulation has been recorded in several species (Mansfield et al., 1982; Whitmore and Freer-Smith, 1982). Clearly sub-clover and barrel medic are particularly sensitive to NOₓ as reductions in biomass of approximately 50% occurred at 170 nLL⁻¹ NO₂.

The stimulation of grain production observed for wheat may not have occurred in barley due to a lack of sufficient sulphur for protein synthesis. As plants require sulphur and nitrogen in well defined ratios related to the composition of their proteins, barley may not have had sufficient soil supplied sulphur to use the available nitrogen from NOₓ exposure to synthesize...
protein. Barley is faster growing than wheat with a greater incremental biomass production and a greater demand for soil supplied sulphur. As wheat effectively had more available sulphur it is possible that it utilised the extra nitrogen from NO\textsubscript{x} exposure for grain production. When barley had an increased supply of sulphur, in the form of SO\textsubscript{2}, an increase in grain production occurred.

The relative sensitivities of the five species to SO\textsubscript{2} was similar to that for NO\textsubscript{x}. Barrel medic and sub-clover were severely affected by concentrations of 149 nLL\textsuperscript{-1} SO\textsubscript{2} and higher. Wheat showed a reduction in shoot dry weight at 149 nLL\textsuperscript{-1} SO\textsubscript{2} but for both wheat and barley, growth and yield were not severely reduced until exposure to 262 nLL\textsuperscript{-1} and 544 nLL\textsuperscript{-1} SO\textsubscript{2} occurred. Lucerne showed a reduction in shoot dry weight at 262 nLL\textsuperscript{-1} SO\textsubscript{2} but was most severely affected by 544 nLL\textsuperscript{-1} SO\textsubscript{2}. SO\textsubscript{2} exposure had little effect on grain or foliar protein concentrations except at the highest concentrations where increases in foliar protein occurred in sub-clover and lucerne. This probably represents protein production for defence and repair mechanisms, primarily enzymes involved in biochemical detoxification pathways.

The growth response of the five species to the combination of SO\textsubscript{2} and NO\textsubscript{x} was much more complex and varied. All three pasture species were very severely affected whilst grain production in barley and wheat was initially stimulated.

Lucerne, which displayed reasonable tolerance to NO\textsubscript{x} + SO\textsubscript{2} individually, was almost as severely damaged by exposure to the combination of SO\textsubscript{2} and NO\textsubscript{x} as barrel medic and sub-clover. Exposure of these three pasture species to SO\textsubscript{2} alone did not invoke a negative response until concentrations reached 149 nLL\textsuperscript{-1} or 262 nLL\textsuperscript{-1}. However, with simultaneous exposure to NO\textsubscript{x} 55 nLL\textsuperscript{-1} SO\textsubscript{2} reduced growth, on average, by more than 50%.

The degree of sensitivity expressed by the sub-clover and barrel medic to NO\textsubscript{x} and all three pasture species to SO\textsubscript{2} + NO\textsubscript{x} may be linked to the fact that these species are all nitrogen-fixers. Exposure to NO\textsubscript{x} alone, in combination with the nitrogen fixing activity of these species, may have been causing nitrogen toxicity due to excessive nitrogen accumulation.

The presence of SO\textsubscript{2} may have been interfering with the nitrogen fixing mechanism and overall nitrogen metabolism. SO\textsubscript{2} is known to inhibit the activity of nitrite and nitrate reducing enzymes resulting in an accumulation of these ions to toxic levels (Wellburn et al., 1981). Very little is known about the effects of air pollutants on the process of nitrogen-fixation but most studies undertaken to date report inhibitory effects (Sheridan, 1979; Griffith and Campbell, 1987; Tingey et al., 1980).

In contrast to the response of the pasture species, the non nitrogen-fixing species, wheat and barley, benefited from exposure to SO\textsubscript{2} and NO\textsubscript{x} at 55 nLL\textsuperscript{-1}
and 149 nLL$^{-1}$ SO$_2$, displaying significant yield increases. At higher SO$_2$ concentrations detrimental effects were observed.

The stimulation observed in barley and wheat plants exposed to the combination of SO$_2$ and NO$_x$ was the result of a stimulation of grain production, rather than vegetative growth. It is possible that grain production can act as a sink for the products of sulphur and nitrogen metabolism. Laisk et al. (1988) report that the high protein synthesis rates in crops can lead to a degree of tolerance to certain air pollutants. Sulphur and nitrogen are very important in protein synthesis, and hence, wheat and barley would show a high capacity for the use of these elements at the grain production stage. It should be noted, however, that this can only occur when other nutrient requirements are met from the soil. Once these are exhausted the excess sulphur and nitrogen cannot be utilised and toxic derivatives of SO$_2$ and NO$_x$ metabolism may then accumulate.

The lack of grain production in pasture species may be a further reason for their sensitivity as this sink for SO$_2$ and NO$_x$ pollutant derivatives would not be available.

Most studies report synergistic (greater than additive) interactions between SO$_2$ and NO$_x$ resulting in visible injury and reductions in both growth and yield at concentrations below the thresholds for effects of either gas alone (Darrall, 1989; Wellburn et al., 1981). Lucerne and sub-clover both showed greater than additive reductions in shoot dry weight at 55 nLL$^{-1}$ SO$_2$ + NO$_x$, while the reductions in this parameter at other SO$_2$ concentrations and in barrel medic due to SO$_2$ + NO$_x$, were approximately additive. However, effects on branch length in all three species showed a synergistic interaction of SO$_2$ + NO$_x$. Exposure to SO$_2$ + NO$_x$ had no effect on foliar protein concentration.

The increase in grain production at 55 nLL$^{-1}$ and 149 nLL$^{-1}$ SO$_2$ + NO$_x$ in wheat was an additive effect. SO$_2$ alone at these concentrations afforded a slight, although not statistically significant, increase in grain production in addition to the large increase afforded by exposure to NO$_x$ alone which accounted for the increase due to simultaneous exposure to SO$_2$ + NO$_x$. In contrast, SO$_2$ alone at 55 nLL$^{-1}$ and 149 nLL$^{-1}$ slightly reduced grain production in barley and NO$_x$ alone had little effect, but in combination these two gases led to a greater than additive increase in grain production. This no doubt relates to the sulphur to nitrogen ratio required by barley for growth.

Grain protein concentration steadily increased in wheat as SO$_2$ concentration increased in the presence of NO$_x$, overcoming the decrease in protein concentration produced by exposure to NO$_x$ alone and reflecting the increasing availability of sulphur, from SO$_2$, for protein synthesis. However, at the highest SO$_2$ concentrations 262 nLL$^{-1}$ and 544 nLL$^{-1}$ SO$_2$ + NO$_x$ both growth and grain production were dramatically reduced so that total protein mass and so yield, was virtually zero.
In contrast, exposure to 55 nL^{-1} and 149 nL^{-1} SO_2 + NO_x in barley, reduced grain protein concentration while increasing grain production. For the purposes of malting this is not necessarily a detrimental effect provided starch content is maintained. However, a decrease in protein concentration reduces the quality of the grain for animal feed.

The presence of NO_x changed the dose-response curve for foliar sulphur concentration in wheat, barley and lucerne leading to lower total sulphur concentrations than recorded for exposure to SO_2 alone. In wheat and barley this is likely to be a result of the redistribution of sulphur within the plant from the foliage to the grain. In lucerne, changes in the processes of uptake, redistribution, exclusion or reemittance of SO_2 may have been occurring but effects on growth were still severe.
CHAPTER 5: THE EFFECTS OF SO₂ AND NOₓ ON EUCALYPTUS MICROCORYS, EUCALYPTUS PILULARIS, EUCALYPTUS REGNANS AND PINUS RADIATA

5.1 INTRODUCTION

E. microcorys, E. pilularis and E. regnans all occur in Australia's indigenous coastal and highland eucalypt forests where average annual rainfall is between 800 mm and 1300 mm. These forests are the primary commercial hardwood forests utilised in Australia, the most important species being E. regnans (mountain ash) and E. obliqua (messmate) in Victoria and Tasmania, E. microcorys (tallowwood) and E. pilularis (blackbutt) in New South Wales and eastern Queensland and E. marginata (jarrah) and E. diversicolor (karri) in the south-west of Western Australia (Brown, 1988). The term "hardwood" is taken to mean eucalypt timber in most parts of Australia whereas "softwood" refers to the timber of coniferous species (Rodd, 1988).

Eucalypt timber is primarily used for general construction work (houses etc), railway sleepers, mining supports, electricity poles, bridge building and fence posts (Rodd, 1988) although imported softwoods and plantation softwood grown in Australia, primarily Pinus radiata, are increasingly being used for house frame construction.

Eucalypt timber is also used for wood pulp and provides much of Australia's newsprint (Rodd, 1988). Packaging materials such as cardboard and building boards such as "Masonite" are also made from eucalypt pulp. The forests of Victoria and Tasmania provide timber of the best quality for these purposes, primarily species of the ash group, particularly E. regnans, which form pulpwood of low density and tannin content. The export of Australian eucalypt timber as woodchips for pulpwood production mainly uses timber from the far south-east of NSW, Tasmania and south-west Western Australia.

E. microcorys is a particularly valuable timber species due to its dense, durable, greasy wood containing large quantities of natural wax (Brown, 1988). It is highly prized as a flooring timber. E. pilularis yields wood of medium to high density and is fairly durable providing useful timber for general construction (Brown, 1988).

More than 90% of Australia's plantations for timber production consist of softwoods with Pinus radiata being most widely used except in Queensland where conditions suit P. elliottii (slash pine), P. caribaea (Caribbean pine) and the indigenous Araucaria cunninghamii (hoop pine) (Brown, 1988).

These plantations generally produce greater yields than those obtained from indigenous forests. Softwood timber is of medium density, is easily dried and preserved and has longer fibres than hardwood making it more suitable for paper manufacture in some cases (Brown, 1988).
5.2 MATERIALS AND METHODS

5.2.1 Fumigation Conditions

Tables 10 and 11 display the characteristics of the fumigation regime.

Table 10: Fumigation characteristics of the SO₂ exposure regime.
All concentrations are in nLL⁻¹.

<table>
<thead>
<tr>
<th>SO₂ Level</th>
<th>Fumigated 4-h arithmetic mean</th>
<th>Standard Deviation</th>
<th>Percentile</th>
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<td></td>
<td></td>
<td></td>
<td>10th</td>
</tr>
<tr>
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<td>&lt;5*</td>
<td>&lt;5</td>
<td>&lt;5</td>
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<td>50</td>
</tr>
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<td>85</td>
<td>78</td>
</tr>
<tr>
<td>5</td>
<td>332</td>
<td>130</td>
<td>163</td>
</tr>
</tbody>
</table>

* below the detection limit of the SO₂ analyser

Table 11: Fumigation characteristics of the NOₓ exposure regime.
All concentrations are in nLL⁻¹.

<table>
<thead>
<tr>
<th>NOₓ Level</th>
<th>NO₂ Concentrations</th>
<th>NO: NO₂</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Fumigated 4 h arithmetic mean</td>
<td>Standard Deviation</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>&lt;5*</td>
<td>&lt;5</td>
</tr>
<tr>
<td>2</td>
<td>74</td>
<td>36</td>
</tr>
</tbody>
</table>

* below the detection limit of the NOₓ analyser

** n/a = not applicable

5.2.2 Cultivation Procedures and Growth Conditions

Seedlings of the following tree species were grown in pots under field conditions for one year prior to fumigation:
1. *Pinus radiata*;

2. *Eucalyptus microcorys* (Tallowwood);

3. *Eucalyptus regnans* (Mountain Ash); and


A subsample of the 12 month old saplings of each species were selected on the basis of uniform size and appearance, and seven were randomly assigned to each of the 20 chambers and planted in the ground. The youngest fully expanded leaf (YFEL) on the main stem of each sapling was tagged to indicate the stage of growth at the commencement of the experiment, so that height increment during the experiment could be recorded.

Fertiliser application was not necessary and plants were sprayed with the insecticide Rogor (Chemspray, active constituent Dimethoate) only when insect activity was noted. Irrigation took place to supplement natural rainfall when necessary.

Plants were exposed to SO$_2$, NO$_x$ or their combination for four hours per day, seven days per week from 1000-1400 for 107 days during the months of January to April 1989.

The mean ambient daily maximum and minimum temperatures were 30.2°C and 18.9°C respectively, and the mean ambient relative humidities at 0900 and 1500 were 51.6% and 40.6% respectively.

5.2.3 Harvest Procedure

At harvest stem diameter was measured in each tree 10cm above the soil line. Plant height was measured, as was the height of the growth that had occurred during the fumigation period.

The saplings were then cut at the soil-line and separated into stems and leaves. These were placed in an oven at 70°C and dried to constant weight for determination of leaf, stem and total plant dry weight. Leaf material was ground to pass a 40 mesh sieve in a Wiley hammer mill. Leaf material was analysed for total sulphur content by inductively coupled plasma emission spectrophotometry. The sulphur was brought into solution for analysis by nitric-perchloric acid digest.
5.3 RESULTS

5.3.1 *Eucalyptus microcorys* (Tallowwood) (Figure 15)

**NOx**

NOx alone significantly decreased leaf dry weight, total plant dry weight, stem diameter and height. Total plant dry weight was decreased by 41%. Decreases in stem dry weight and height of new growth were also apparent. NOx had no effect on total sulphur concentration in the foliage.

**SO2**

SO2 alone at a concentration of 174 nLL-1 or above, decreased leaf dry weight and total plant dry weight. At 332 nLL-1 total dry weight was reduced to 50% of that of the control plants. Stem diameter was decreased at 122 nLL-1 and above. There were no statistically significant decreases in stem dry weight, height or height of new growth, however, a trend towards growth depression was evident with an initial slight stimulation of height growth at 50 nLL-1 and 122 nLL-1 SO2.

**SO2 + NOx**

The growth depression induced by NOx was at least partially ameliorated by the presence of SO2 at 122 nLL-1 and 175 nLL-1 to give a growth depression of as little as 9% at 122 nLL-1. However, at 332 nLL-1 SO2 + NOx growth depression was of a similar magnitude to that induced by either NOx or 332 nLL-1 SO2 alone. The presence of NOx also slightly alleviated the growth depression induced by either 122 nLL-1 or 175 nLL-1 SO2. These results show a strongly interactive effect between SO2 and NOx with the changes in growth being less than additive for all SO2 concentrations considered. The presence of NOx had no effect on the accumulation of sulphur in the foliage.

5.3.2 *Eucalyptus pilularis* (Blackbutt) (Figure 16).

**NOx**

NOx decreased leaf dry weight, total plant dry weight, stem diameter and height of new growth. Total plant dry weight was reduced by 42% showing a very similar response to *E. microcorys*. Decreases in stem dry weight and height were also apparent. NOx had no effect on total sulphur concentration.

**SO2** (Growth response data for the 50 nLL-1 SO2 treatment have been deleted due to anomalous results)

All growth parameters were reduced by exposure to SO2 at 175 nLL-1 and
Figure 15: \( \text{SO}_2 \) and \( \text{NO}_x \) Concentration-Response Relationships for \( E. \ microcorys \) Grown in Open-Top Chambers.

Plotted values represent arithmetic means of the response variables. The symbol \( \text{SO}_2 \) only treatments and the symbol \( \text{SO}_2 + \text{NO}_x \) treatments. The arithmetic mean \( \text{NO}_2 \) concentration was 74 nL/L and the \( \text{NO}:\text{NO}_2 \) ratio was 1:9. For each response curve, values that are labelled with the same letters are not significantly different; the letters \( \text{w, x, y and z apply to the SO}_2 \) treatments whilst the letters \( \text{a, b, c and d apply to the SO}_2 + \text{NO}_x \) treatments. If no letters are present on a given curve there are no significant differences. A star (*) next to a value denotes that the \( \text{SO}_2 + \text{NO}_x \) treatment is significantly different from the \( \text{SO}_2 \) only treatment of the same \( \text{SO}_2 \) concentration.
Figure 15: (continued)
Figure 15: (continued)
Figure 16: SO₂ and NOₓ Concentration-Response Relationships for *E. pilularis* Grown in Open-Top Chambers.

Plotted values represent arithmetic means of the response variables. The symbol - - represents SO₂ only treatments and the symbol - - represents SO₂ + NOₓ treatments. The arithmetic mean NO₂ concentration was 74 nL/L and the NO: NO₂ ratio was 1:9. For each response curve, values that are labelled with the same letters are not significantly different; the letters w, x, y and z apply to the SO₂ treatments whilst the letters a, b, c and d apply to the SO₂ + NOₓ treatments. If no letters are present on a given curve there are no significant differences. A star (*) next to a value denotes that the SO₂ + NOₓ treatment is significantly different from the SO₂ only treatment of the same SO₂ concentration.
Figure 16: (continued)
Figure 16: (continued)
332 nLL⁻¹. Total plant dry weight was reduced by approximately 40% by both 175 nLL⁻¹ and 332 nLL⁻¹. Total sulphur concentration in the foliage was increased by exposure to 50 nLL⁻¹ SO₂ and above.

SO₂ + NOₓ

Large differences in response were evident, however, few of these proved to be statistically significant due to the large variability in responses of individual plants in each treatment. For the parameters, leaf dry weight and height of new growth, an amelioration of the growth depression induced by NOₓ alone occurred due to the presence of SO₂. This interactive effect was strongest at 122 nLL⁻¹ and 175 nLL⁻¹ SO₂ + NOₓ. A similar response was evident in stem and total dry weight and in total height.

The presence of NOₓ had little effect on sulphur accumulation in the foliage except at 122 nLL⁻¹ SO₂ where the presence of NOₓ slightly decreased total sulphur concentration.

5.3.3 Eucalyptus regnans (Mountain Ash) (Figure 17)

NOₓ

NOₓ decreased leaf dry weight, stem diameter and height of new growth. Decreases in all other growth parameters were also evident. Despite a 43% reduction in total plant dry weight, this decrease was not statistically significant due to variation between the response of individual plants. There was no effect on total sulphur concentration in the foliage.

SO₂

Exposure to 175 nLL⁻¹ or above decreased all growth parameters. Accumulation of sulphur in the foliage occurred due to exposure to 50 nLL⁻¹ and above.

SO₂ + NOₓ

As in the other two eucalypts the presence of SO₂ and NOₓ in combination partially ameliorated the growth depression induced by either gas alone, however, in this species this synergistic effect occurred only at 175 nLL⁻¹ SO₂ + NOₓ. At 50 nLL⁻¹ SO₂ and NOₓ growth depression was more severe than that induced by either gas alone and showed an additive effect for most parameters measured. This growth depression was of similar magnitude to that induced by 332 nLL⁻¹ SO₂ + NOₓ or 332 nLL⁻¹ SO₂ alone. The presence of NOₓ had no effect on sulphur accumulation in the foliage.
Figure 17: SO₂ and NOₓ Concentration-Response Relationships for *E. regnans* Grown in Open-Top Chambers.

Plotted values represent arithmetic means of the response variables. The symbol □ represents SO₂ only treatments and the symbol ● represents SO₂ + NOₓ treatments. The arithmetic mean NO₂ concentration was 74 nL/L and the NO:NO₂ ratio was 1:9. For each response curve, values that are labelled with the same letters are not significantly different; the letters w, x, y and z apply to the SO₂ treatments whilst the letters a, b, c and d apply to the SO₂ + NOₓ treatments. If no letters are present on a given curve there are no significant differences. A star (*) next to a value denotes that the SO₂ + NOₓ treatment is significantly different from the SO₂ only treatment of the same SO₂ concentration.
Figure 17: (continued)
Figure 17: (continued)
5.3.4 *Pinus radiata* (Figure 18)

**NO\textsubscript{x}**

NO\textsubscript{x} alone had no effect on any parameter measured.

**SO\textsubscript{2}**

SO\textsubscript{2} alone had no effect on any growth parameter. As in the *Eucalyptus* species, total sulphur concentration was increased by exposure to all SO\textsubscript{2} concentrations but the response curve was quadratic rather than linear.

Effects on growth may have become apparent with an exposure period longer than four months. The mean value for height of new growth at 122 nLL\textsuperscript{-1} was significantly greater than the mean values at 175 nLL\textsuperscript{-1} and 332 nLL\textsuperscript{-1} SO\textsubscript{2}, indicating a transition from slight growth stimulation to growth depression.

**SO\textsubscript{2} + NO\textsubscript{x}**

Leaf, stem and total plant dry weights and stem diameter were all increased by exposure to 122 nLL\textsuperscript{-1} SO\textsubscript{2} + NO\textsubscript{x}. At 332 nLL\textsuperscript{-1} SO\textsubscript{2} + NO\textsubscript{x} substantial growth depression occurred, with a 35% reduction in total plant dry weight recorded. Exposure to 322 nLL\textsuperscript{-1} SO\textsubscript{2} alone had no effect on growth, neither did exposure to NO\textsubscript{x} alone. Thus, the reduction at 322 nLL\textsuperscript{-1} SO\textsubscript{2} + NO\textsubscript{x} shows a strongly synergistic effect between the two gases.

In the presence of NO\textsubscript{x} the total sulphur concentration in the foliage increased linearly as SO\textsubscript{2} concentration increased.

5.4. **DISCUSSION**

Based on all previous research, NO\textsubscript{x} comprising primarily NO\textsubscript{2} at a concentration of 74 nLL\textsuperscript{-1} would not be expected to affect plant growth, especially not under intermittent fumigation conditions. Yet all three *Eucalyptus* species were very sensitive to the NO\textsubscript{x} exposure regime used, all showing a growth depression of approximately 40% total dry weight after only four months.

These three *Eucalyptus* species were also sensitive to SO\textsubscript{2} exposure showing significant growth depression once the SO\textsubscript{2} concentration reached 175 nLL\textsuperscript{-1}. As a 24-h average this is only 29 nLL\textsuperscript{-1}. This is equivalent to the lowest SO\textsubscript{2} concentration known to affect northern hemisphere tree species (Roberts *et al*, 1983b). Growth depression was also evident at 122 nLL\textsuperscript{-1} in *E. regnans* and at 50 nLL\textsuperscript{-1} and 122 nLL\textsuperscript{-1} in *E. microcorys*. It is likely that the growth depression at these concentrations would have become statistically significant if exposure
Figure 18: SO₂ and NOₓ Concentration-Response Relationships for P. radiata Grown in Open-Top Chambers.

Plotted values represent arithmetic means of the response variables. The symbol — represents SO₂ only treatments and the symbol — represents SO₂ + NOₓ treatments. The arithmetic mean NO₂ concentration was 74 nL/L and the NO:NO₂ ratio was 1:9. For each response curve, values that are labelled with the same letters are not significantly different; the letters w, x, y and z apply to the SO₂ treatments whilst the letters a, b, c and d apply to the SO₂ + NOₓ treatments. If no letters are present on a given curve there are no significant differences. A star (*) next to a value denotes that the SO₂ + NOₓ treatment is significantly different from the SO₂ only treatment of the same SO₂ concentration.
Figure 18: (continued)
Figure 18: (continued)
had continued beyond four months. Of the three *Eucalyptus* species, *E. microcorys* was the most sensitive to exposure to SO₂ and is not likely to be protected by the most stringent guideline for SO₂ currently in existence as recommended by the IUFRO. This guideline states that for full protection of tree species, a 24-h average of 19 nLL⁻¹ SO₂ is not to be exceeded more than 12 times in any given six month period. Yet *E. microcorys* showed a trend towards growth depression at 50 nLL⁻¹ for 4-h/day, equivalent to a 24 hour average of only 8 nLL⁻¹ daily.

The growth reductions in the *Eucalyptus* species due to either SO₂ or NOₓ were less severe when both gases were present in combination especially at 122 nLL⁻¹ and 175 nLL⁻¹ SO₂. For all three *Eucalyptus* species the growth reductions afforded by the interaction of SO₂ + NOₓ were less than additive at most SO₂ concentrations considered. The exception was *E. regnans* which was more sensitive to the combination of SO₂ + NOₓ than the other two eucalypts, showing an additive growth reduction at 50 nLL⁻¹ SO₂ + NOₓ. In this species, 175 nLL⁻¹ SO₂ + NOₓ afforded the least severe effects on growth.

In contrast, *Pinus radiata* was unaffected by exposure to NOₓ or SO₂ alone, although a depression of height growth may have developed at 175 nLL⁻¹ SO₂ or above if fumigation had continued beyond four months. The only treatment regime that led to a significant growth reduction was 332 nLL⁻¹ SO₂ + NOₓ whereas 122 nLL⁻¹ SO₂ + NOₓ tended to increase growth. These effects were both greater than additive, although they resulted in responses of opposite direction. *P. radiata* is less sensitive to SO₂ and SO₂ + NOₓ than some other *Pinus* species which have been studied previously. *Pinus strobus* and *P. sylvestris* are amongst the most sensitive northern hemisphere tree species sensitive to SO₂ (Roberts *et al.*, 1983b) and growth reductions have been reported in other *Pinus* species exposed to between 50 nLL⁻¹ and 80 nLL⁻¹ SO₂ or SO₂ + NOₓ (Freer-Smith, 1985).

The differences in response between the *Eucalyptus* species and *Pinus radiata* most likely relate to the specific ratio of sulphur to nitrogen required by each species for nutrition.

The mechanisms which induce injury or otherwise in plants due to exposure to SO₂, NOₓ or their combination closely relate to the metabolic pathways which allow atmospheric SO₂ and NOₓ to be used as nutritional sources of sulphur and nitrogen respectively. Organic sulphur and nitrogen have a constant ratio of 0.030 in the foliage of *Pinus*, Douglas fir and *Eucalyptus* species, the same ratio as that in foliar protein (Lambert and Turner, 1980). A fine balance exists between the use of SO₂ and NOₓ for growth and the accumulation of toxic metabolic derivatives as determined by the sulphur-nitrogen ratio required by a given species and the availability of nutrients in the soil at the time of fumigation. This balance is clearly evident in the transition of *E. pilularis* from normal growth at 122 nLL⁻¹ SO₂ + NOₓ to growth depression at 175 nLL⁻¹ SO₂ + NOₓ.
The nutritional interplay of SO₂ and NOₓ is also evident in *P. radiata* where 122 nLL⁻¹ SO₂ + NOₓ produced growth stimulation but 322 nLL⁻¹ SO₂ + NOₓ produced severe growth depression.

The large difference in response between *P. radiata* and the *Eucalyptus* species probably relates to the adaptation of the eucalypts to growth on nutrient poor soils such that eucalypts require only minimal nutrition. It is likely that the metabolic pathways they possess cannot cope with high nutrient uptake as afforded by SO₂ and NOₓ exposure or high rates of fertilizer application.

The pattern of sulphur accumulation in *P. radiata* may further explain the tolerance of this species to SO₂. Exposure to SO₂ alone produced a typical quadratic curve for total sulphur concentration in the foliage with maximum levels reached at about 122 nLL⁻¹ SO₂. It seems that this species can prevent excessive sulphur accumulation in the foliage and perhaps thus avoid toxic effects. In the presence of NOₓ, linear accumulation of sulphur with increasing SO₂ concentration occurred with toxic effects developing at the highest SO₂ concentration. Sulphur accumulation in the *Eucalyptus* species was linear in the presence of SO₂ alone or SO₂ + NOₓ with NOₓ having no effect on the amount of sulphur accumulated. In *P. radiata* the presence of NOₓ enhanced sulphur accumulation in the foliage.
CHAPTER 6: THE EFFECTS OF SO$_2$ AND NO$_x$ ON OATS, TRITICALE, WHITE CLOVER AND BURR MEDIC.

6.1 INTRODUCTION

Oats are grown primarily for grain in Australia with a large proportion being used for livestock feed and only a small amount being milled for human consumption. As for wheat and barley, Australia is a significant world exporter of oat grain, second only to Sweden and Finland for oats, the USA and Canada for wheat and France and Canada for barley (Reid, 1990).

Some oats are grown for grazing, silage or hay production and due to their good growth during winter, can be of importance for winter grazing where winter growth of pastures is poor.

Many of the oat cultivars grown for grain production have been bred for reduced height and resistance to grain lodging and shattering during harvest and transport. The cultivar, Echidna, is a relatively recent advance in this regard due to its breeding from a dwarf mutant oat (Reid, 1990).

Triticale is a relatively new cereal which has been developed from crosses between wheat (*Triticum* sp.) and rye (*Secale* sp.), hence the name triticale. Most triticales are crosses with durum wheats (*Triticum durum*) rather than bread wheats (*Triticum aestivum*) and so the grain is of poorer quality for traditional bread making than bread wheat grain. However, it has good nutritive value, and a higher protein to energy ratio than most traditional feed grains while being of similar digestibility. It also has a high enzymatic activity making it useful as a malting grain for the distilling industry. Some consider that its main use in the future will be for feed grain, particularly for the pig and poultry industries (Reid, 1990).

White clover is a perennial legume, native to Europe and the Mediterranean. It requires fertile soils and especially high phosphorous and potassium levels. It provides high quality feed in pastures with many cultivars being highly productive and resistant to grazing pressure. The cultivar Haifa is a large-leaved erect variety that shows excellent winter growth and has good drought and heat tolerance (Reid, 1990).

Burr medic is another legume species used for pasture improvement and provides good sheep feed. The sown cultivars have been chosen for burrs with short spines to minimise the burring of wool. The cultivar Santiago shows good grazing performance in relatively low rainfall areas (Reid, 1990).
6.2 MATERIALS AND METHODS

6.2.1 Fumigation Conditions

Tables 12 and 13 display the characteristics of the fumigation regime.

Table 12: Fumigation characteristics of the SO₂ exposure regime.
All concentrations are in nLL⁻¹.

<table>
<thead>
<tr>
<th>SO₂ Level</th>
<th>Fumigated 4-h arithmetic mean</th>
<th>Standard Deviation</th>
<th>10th</th>
<th>90th</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>&lt;5*</td>
<td>&lt;5</td>
<td>&lt;5</td>
<td>&lt;5</td>
</tr>
<tr>
<td>2</td>
<td>80</td>
<td>72</td>
<td>7</td>
<td>160</td>
</tr>
<tr>
<td>3</td>
<td>139</td>
<td>90</td>
<td>50</td>
<td>240</td>
</tr>
<tr>
<td>4</td>
<td>255</td>
<td>136</td>
<td>97</td>
<td>437</td>
</tr>
<tr>
<td>5</td>
<td>388</td>
<td>219</td>
<td>146</td>
<td>673</td>
</tr>
</tbody>
</table>

* below the detection limit of the SO₂ analyser.

Table 13: Fumigation characteristics of the NOₓ exposure regime.
All concentrations are in nLL⁻¹.

<table>
<thead>
<tr>
<th>NOₓ Level</th>
<th>NO₂ Concentrations</th>
<th>NO: NO₂</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Fumigated 4 h arithmetic mean</td>
<td>Standard Deviation</td>
</tr>
<tr>
<td>1</td>
<td>&lt;5*</td>
<td>&lt;5</td>
</tr>
<tr>
<td>2</td>
<td>29</td>
<td>23</td>
</tr>
</tbody>
</table>

* below the detection limit of the NOₓ analyser

** n/a = not applicable

6.2.2 Cultivation Procedures and Growth Conditions

Seeds of the following species and cultivars were planted in peat Jiffy pots:

1. Oats - *Avena sativa* cv. Echidna;
2. Triticale - *X. triticecale* cv. Currency;

3. White clover - *Medicago polymorpha* cv. Santiago; and


The white clover and burr medic were inoculated with the relevant commercial rhizobium inocula (Nodulaid, Sydney) at the time of planting. Three weeks after sowing all species were removed from the glasshouse and 10 plants of each species randomly assigned to each of the 20 chambers. Prior to this a 10 cm layer of potting mix (2 river sand: 4 sawdust: 2 wood fibre fines: 2 pinebark) was dug into the first 20cm of soil in each chamber. The soil was then tested for pH and major nutrients by CSBP and Farmers Ltd. Commercial garden lime was applied to bring pH to 6.5 and fertilizer applications made as recommended for each crop.

Plants were sprayed with Dyptrex (Bayer, active constituent Trichlorfon) when required to control caterpillar pests. One application of Kelthane (Hortico, active constituent Difcol) was required to control red spider. Piramor (ICI, active constituent Pirimicarb) and Tilt (Ciber Geigy, active constituent Propiconazole) were also applied once to control aphids and fungal infections (rust and mildew) respectively. Irrigation took place to supplement natural rainfall when necessary.

Plants were exposed to SO$_2$, NO$_x$ or their combination for four hours per day, seven days per week from 1000-1400 between the months of May and November 1990. The white clover and burr medic were fumigated for 149 days before harvest. The oats required longer to mature and were fumigated for a total of 182 days.

The triticale showed very poor growth when first transferred to the chambers in May and many mortalities occurred. For this reason a second batch was sown and then planted out in the chambers in June. Due to this late sowing the triticale could not be grown to full maturity before finalisation of the project. The triticale were fumigated for 131 days in total before harvest.

For the months of May to November inclusive, the mean ambient daily maximum and minimum temperatures were 19.8°C and 10.3°C respectively, and the mean ambient relative humidities at 0900 and 1500 were 71.1% and 52.9% respectively.

6.2.3 Harvest Procedure

As in the previous crop experiment, parameters to assess both yield and quality were measured.
Oat plants were cut at the soil line, separated into shoots and ears and then placed in an oven at 70°C and dried to constant weight. Shoot, ear and grain dry weight and ear and grain number were later measured.

Triticale plants were cut at the soil line and the number of tillers and mature ears recorded. Plants were separated into shoots and ears and then placed in an oven at 70°C and dried to constant weight. One mature ear from each plant was selected prior to drying and dried separately for more detailed analysis. Shoot and total ear dry weight were later recorded. To assess average grain weight the grain number and grain weight were recorded for the single mature ear selected from each plant.

Grain from the oat and triticale plants was then ground to pass a 40 mesh sieve in a Wiley hammer mill prior to total nitrogen determination by standard Kjeldahl digestion and distillation.

The pasture species were cut at the soil line and the length of the longest branch recorded. The plants were then placed in an oven at 70°C and dried to constant weight to determine shoot dry weight. Foliar material was ground in a Wiley hammer mill prior to total nitrogen determination as for the crops. Total sulphur content of the foliage was also determined using ground leaf material and inductively coupled plasma emission spectrophotometry. The sulphur was brought into solution for analysis by a nitric-perchloric acid digest.

Crude protein concentration was estimated for the grain of oats and triticale and the foliage of white clover and burr medic by multiplying total nitrogen values (%) by the standard factor of 6.25 (AOAC, 1984).

6.3 RESULTS

6.3.1 Oats cv Echidna (Figure 19)

NOx

NOx had no statistically significant effect on any of the parameters measured for oats. However, the mean values for grain number and grain dry weight showed an increase of 37% due to NOx. Increases of smaller magnitude also occurred in shoot, ear and total dry weight (13%, 10% and 9% respectively)

SO2*

Shoot, ear and total dry weight all showed significant reductions once the SO2 concentration reached 255 nLL-1 SO2 had no statistically significant effect on ear number, grain number, grain dry weight, average grain dry weight or grain

* Growth response data for the 388 nLL-1 SO2 treatment have been deleted due to anomalous results.
Figure 19: $SO_2$ and NO$_x$ Concentration-Response Relationships for *Avena sativa* cv. Echidna Grown in Open-Top Chambers.

Plotted values represent arithmetic means of the response variables. The symbol $\rightarrow$ represents $SO_2$ only treatments and the symbol $\longrightarrow$ represents $SO_2 + NO_x$ treatments. The arithmetic mean NO$_2$ concentration was 29 nL/L and the NO$_2$:NO$_x$ ratio was 2:1. For each response curve, values that are labelled with the same letters are not significantly different; the letters w, x, y and z apply to the $SO_2$ treatments whilst the letters a, b, c and d apply to the $SO_2 + NO_x$ treatments. If no letters are present on a given curve there are no significant differences. A star (*) next to a value denotes that the $SO_2 + NO_x$ treatment is significantly different from the $SO_2$ only treatment of the same $SO_2$ concentration.
Figure 19: (continued)
Figure 19: (continued)
protein concentration. However, a downward trend was evident for the first three parameters and average grain dry weight showed a trend towards higher values at 80 nLL\(^{-1}\) and 139 nLL\(^{-1}\) but not 255 nLL\(^{-1}\) SO\(_2\).

**SO\(_2\) + NO\(_x\)**

For all parameters except average grain dry weight and protein concentration the presence of NO\(_x\) enhanced the growth and productivity of oats when in the presence of SO\(_2\) at concentrations of 80 nLL\(^{-1}\), 139 nLL\(^{-1}\) and 255 nLL\(^{-1}\) SO\(_2\). This ameliorated the trend towards growth depression induced by SO\(_2\) alone at these concentrations.

Exposure to SO\(_2\) + NO\(_x\) in the SO\(_2\) concentration range used resulted in a growth response similar to that for NO\(_x\) alone, especially at 80 nLL\(^{-1}\) SO\(_2\) + NO\(_x\) where a 37% increase in grain dry weight occurred as for the NO\(_x\) only treatment. Effects at 80 nLL\(^{-1}\) SO\(_2\) + NO\(_x\) were interactive with NO\(_x\) countering a 30% reduction in grain dry weight due to 80 nLL\(^{-1}\) SO\(_2\) alone. At 139 and 255 nLL\(^{-1}\) SO\(_2\) + NO\(_x\) effects were additive.

In the case of grain protein concentration 255 nLL\(^{-1}\) SO\(_2\) + NO\(_x\) significantly increased this parameter to a value greater than that for the NO\(_x\) only treatment. An increase above that for the 255 nLL\(^{-1}\) SO\(_2\) only treatment was also evident. These results indicated that simultaneous exposure to 255 nLL\(^{-1}\) SO\(_2\) + NO\(_x\) produced a greater than additive increase in grain protein concentration.

6.3.2 **Triticale cv. Currency** (Figure 20)

**NO\(_x\)**

NO\(_x\) had no effect on any parameter measured except for a reduction in the number of mature ears at harvest indicating that exposure to NO\(_x\) was delaying maturation.

**SO\(_2\)**

Exposure to SO\(_2\) reduced shoot dry weight and total dry weight at 80 nLL\(^{-1}\) and above.

The particularly low mean value for shoot dry weight and total dry weight recorded for the 80 nLL\(^{-1}\) SO\(_2\) only treatment was due to a particularly wide variation in response between individual plants in this treatment, with the vegetative growth of some plants being more severely affected in this treatment than plants in treatments with higher SO\(_2\) concentrations.
Figure 20: $SO_2$ and $NO_X$ Concentration-Response Relationships for $X. triticosecale$ cv. Currency Grown in Open-Top Chambers.

Plotted values represent arithmetic means of the response variables. The symbol $-$ represents $SO_2$ only treatments and the symbol $-$ represents $SO_2 + NO_X$ treatments. The arithmetic mean $NO_2$ concentration was 29 nL/L and the $NO:NO_2$ ratio was 2:1. For each response curve, values that are labelled with the same letters are not significantly different; the letters w, x, y and z apply to the $SO_2$ treatments whilst the letters a, b, c and d apply to the $SO_2 + NO_X$ treatments. If no letters are present on a given curve there are no significant differences. A star (*) next to a value denotes that the $SO_2 + NO_X$ treatment is significantly different from the $SO_2$ only treatment of the same $SO_2$ concentration.
Figure 20: (continued)
Figure 20: (continued)
Figure 20: (continued)
There was a trend towards decreased ear dry weight with increasing SO\textsubscript{2} concentration but no statistically significant differences were evident between treatments.

Tiller number, representing potential ear number if full maturation had been possible, showed a decrease of approximately 25% at 80 nLL\textsuperscript{-1} SO\textsubscript{2} and above. This decrease became statistically significant at 338 nLL\textsuperscript{-1} SO\textsubscript{2} where it reached a maximum of 39%.

The number of mature ears at harvest was decreased by exposure to 255 nLL\textsuperscript{-1} SO\textsubscript{2} and 388 nLL\textsuperscript{-1} SO\textsubscript{2}. The average grain dry weight of mature ears was decreased by exposure to 388 nLL\textsuperscript{-1} SO\textsubscript{2} with a decrease also apparent at 255 nLL\textsuperscript{-1} SO\textsubscript{2}. In contrast, grain protein concentration was increased by 388 nLL\textsuperscript{-1} SO\textsubscript{2}.

SO\textsubscript{2} + NO\textsubscript{x}

The response curves for SO\textsubscript{2} + NO\textsubscript{x} and SO\textsubscript{2} only were very similar for all parameters except mature ear number.

Exposure to 80 nLL\textsuperscript{-1}, 139 nLL\textsuperscript{-1} and 255 nLL\textsuperscript{-1} SO\textsubscript{2} + NO\textsubscript{x} resulted in a reduction in mature ear number of similar magnitude to that from exposure to NO\textsubscript{x} alone. Only at 388 nLL\textsuperscript{-1} SO\textsubscript{2} + NO\textsubscript{x} did a more substantial reduction occur. The relative magnitudes of the reductions in mature ear number due to SO\textsubscript{2} alone, NO\textsubscript{x} alone and SO\textsubscript{2} + NO\textsubscript{x} indicate an additive rather than interactive effect except at 255 nLL\textsuperscript{-1} SO\textsubscript{2} + NO\textsubscript{x} where an antagonistic (less than additive) effect occurred.

6.3.3 White Clover cv Haifa (Figure 21)

NO\textsubscript{x}

NO\textsubscript{x} decreased branch length and shoot dry weight by 21% and 49% respectively. There was no effect on foliar protein concentration nor on total foliar sulphur concentration.

SO\textsubscript{2}

Branch length and shoot dry weight were decreased by exposure to 80 nLL\textsuperscript{-1}, 255 nLL\textsuperscript{-1} and 388 nLL\textsuperscript{-1} SO\textsubscript{2}. This was accompanied by an increase in foliar sulphur content at the two highest SO\textsubscript{2} concentrations. There was no effect on foliar protein concentration.
Figure 21: \( \text{SO}_2 \) and \( \text{NO}_x \) Concentration-Response Relationships for \textit{Trifolium repens} cv. Haifa Grown in Open-Top Chambers.

Plotted values represent arithmetic means of the response variables. The symbol - ○ - represents \( \text{SO}_2 \) only treatments and the symbol - ● - represents \( \text{SO}_2 + \text{NO}_x \) treatments. The arithmetic mean \( \text{NO}_2 \) concentration was 29 nL/L and the \( \text{NO}:\text{NO}_2 \) ratio was 2:1. For each response curve, values that are labelled with the same letters are not significantly different; the letters w, x, y and z apply to the \( \text{SO}_2 \) treatments whilst the letters a, b, c and d apply to the \( \text{SO}_2 + \text{NO}_x \) treatments. If no letters are present on a given curve there are no significant differences. A star (*) next to a value denotes that the \( \text{SO}_2 + \text{NO}_x \) treatment is significantly different from the \( \text{SO}_2 \) only treatment of the same \( \text{SO}_2 \) concentration.
Figure 21: (continued)
SO$_2$ + NO$_x$

No discernible relationship between the SO$_2$ response curve and the SO$_2$ + NO$_x$ response curve was evident for branch length and shoot dry weight. This was partly a result of the large variability in response between individual plants especially in the SO$_2$ only treatments. In general terms a less than additive interaction was evident at the lower SO$_2$ concentrations in combination with NO$_x$. However, at 388 nLL$^{-1}$ SO$_2$ + NO$_x$ an additive effect occurred whereby branch length and shoot dry weight were reduced significantly below the 388 nLL$^{-1}$ SO$_2$ only treatment. Concomitant with this was an increase in foliar sulphur concentration and protein concentration.

6.3.4 Burr Medic cv. Santiago (Figure 22)

NO$_x$

Exposure to NO$_x$ significantly increased shoot dry weight by 35% but had no effect on branch length. The 10% increase in foliar protein concentration was not statistically significant. There was no effect on foliar sulphur concentration.

SO$_2$

SO$_2$ severely affected the growth of burr medic with reductions in branch length and shoot dry weight occurring at 80 nLL$^{-1}$ SO$_2$ and above. This was reflected by an increase in foliar sulphur concentrations at 80 nLL$^{-1}$ SO$_2$ and above.

Foliar protein concentration was increased at 139 nLL$^{-1}$ and 388 nLL$^{-1}$ SO$_2$ and showed a general upward trend across the entire range of SO$_2$ concentrations considered.

SO$_2$ + NO$_x$

The presence of SO$_2$ lead to a depression of the growth stimulation afforded by NO$_x$ with a synergistic (greater than additive) effect evident in the parameters shoot dry weight and branch length. Foliar protein concentration was increased substantially by the presence of 80 nLL$^{-1}$ SO$_2$ + NO$_x$ but decreased as SO$_2$ concentration increased until at 338 nLL$^{-1}$ SO$_2$ + NO$_x$ protein concentration was similar to that due to 388 nLL$^{-1}$ SO$_2$ alone.

The pattern of sulphur accumulation due to SO$_2$ exposure was similar in the presence of NO$_x$ to that which occurred due to SO$_2$ alone, except at 388 nLL$^{-1}$ SO$_2$ + NO$_x$ where foliar sulphur concentration was much higher than that recorded for 388 nLL$^{-1}$ SO$_2$ alone.
Figure 22: SO$_2$ and NO$_x$ Concentration-Response Relationships for *Medicago polymorpha* cv. Santiago Grown in Open-Top Chambers.

Plotted values represent arithmetic means of the response variables. The symbol $\square$ represents SO$_2$ only treatments and the symbol $\rightarrow$ represents SO$_2$ + NO$_x$ treatments. The arithmetic mean NO$_2$ concentration was 29 nL/L and the NO:NO$_2$ ratio was 2:1. For each response curve, values that are labelled with the same letters are not significantly different; the letters w, x, y and z apply to the SO$_2$ treatments whilst the letters a, b, c and d apply to the SO$_2$ + NO$_x$ treatments. If no letters are present on a given curve there are no significant differences. A star (*) next to a value denotes that the SO$_2$ + NO$_x$ treatment is significantly different from the SO$_2$ only treatment of the same SO$_2$ concentration.
Figure 22: (continued)
6.4 Discussion

Exposure to 87 nL$^{-1}$ NO$_x$ comprising only 29 nL$^{-1}$ NO$_2$ severely affected growth of white clover whilst increasing shoot dry weight in burr medic. As for wheat, NO$_x$ seemed to increase grain production in oats although the 37% increase recorded in grain number and dry weight was not statistically significant. Triticale showed little response to NO$_x$ at these concentrations except for a delay in the maturation rate of ears.

Except for white clover, the responses observed in these crop species were less severe than those observed in the previous crop experiment (Chapter 4) as might be expected due to the lower NO$_x$ concentration used this time. The response of the burr medic, oats and triticale is consistent with previous findings on the effects of NO$_2$ below 200 nL$^{-1}$ which report either growth stimulation or no effects (Irving, 1987). In contrast, the response of white clover shows that this species is highly sensitive to exposure to NO$_x$, although, it is not possible to establish whether NO or NO$_2$ or both were responsible for this reduction. The literature rarely considers NO and tends to assume NO$_2$ is the phytotoxic component of NO$_x$ emissions. However, it is highly unlikely that NO$_2$ alone at a concentration of only 29 nL$^{-1}$ could induce a growth reduction as great as 50%. In the Eucalyptus species (Chapter 5), growth reductions of approximately 40% were induced by a similar concentration of NO$_x$ but comprising 74 nL$^{-1}$ NO$_2$. This preliminary evidence suggests that NO, in addition to NO$_2$, is a phytotoxic component in NO$_x$ emissions.

Triticale, white clover and burr medic were all very sensitive to exposure to SO$_2$ alone showing growth and yield reductions at almost all SO$_2$ concentrations considered, including the lowest concentration of 80 nL$^{-1}$. Oats was more resistant to SO$_2$ exposure, showing significant decreases in growth and yield only at 255 nL$^{-1}$ and 388 nL$^{-1}$ SO$_2$. The former three species were clearly more sensitive to SO$_2$ than the cultivars of wheat, barley and lucerne fumigated in the previous crop experiment (Chapter 4) and possibly also the barrel medic and sub-clover cultivars.

White clover and burr medic were also very sensitive to simultaneous exposure to SO$_2$ + NO$_x$ as occurred in the other pasture species (Chapter 4). In contrast, oats and triticale showed a slight stimulation in yield due to 80 nL$^{-1}$ SO$_2$ + NO$_x$ and showed yield decreases, only at the highest SO$_2$ concentration of 388 nL$^{-1}$ SO$_2$ + NO$_x$. This response was similar to that of the other cereals, wheat and barley.

As discussed previously (Chapter 4) the sensitivity of the pasture species to simultaneous exposure to SO$_2$ + NO$_x$ probably relates to the toxicity of SO$_2$ to the nitrogen fixation process.

The non-nitrogen fixing cereal crops also displayed a degree of tolerance to fumigation as atmospheric SO$_2$ and NO$_x$ can be diverted into grain production
if present in the ratios required to supplement soil supplied sulphur and nitrogen for protein synthesis.

The response of burr medic showed a strongly synergistic interaction between \( \text{SO}_2 \) and \( \text{NO}_x \). The presence of as little as 80 n\text{LL}^{-1} \text{SO}_2 completely reversed the growth stimulation induced by the presence of \( \text{NO}_x \) alone.

In white clover the reduction in growth due to \( \text{SO}_2 + \text{NO}_x \) was generally less than additive but equally as damaging as for burr medic. White clover was very sensitive to exposure to \( \text{NO}_x \) alone as well as \( \text{SO}_2 \) alone.

Triticale showed a similar response to \( \text{SO}_2 + \text{NO}_x \) as to \( \text{SO}_2 \) alone, with the delay in ear maturation rate due to \( \text{NO}_x \) alone and \( \text{SO}_2 \) at 255 n\text{LL}^{-1} \text{ and 388 nLL}^{-1} \) also occurring in the \( \text{SO}_2 + \text{NO}_x \) treatments.

Growth and grain production was increased in oats at 80 n\text{LL}^{-1}, 139 n\text{LL}^{-1} \text{ and 255 nLL}^{-1} \text{SO}_2 + \text{NO}_x \) with greater than additive increases resulting in means of similar magnitude to those induced by exposure to \( \text{NO}_x \) alone, despite the trend towards growth and yield reduction induced by \( \text{SO}_2 \).

Exposure to \( \text{SO}_2 \), \( \text{NO}_x \) or \( \text{SO}_2 + \text{NO}_x \) had little effect on grain protein concentration in oats and thus, the increase in grain production due to 80 n\text{LL}^{-1}, 139 n\text{LL}^{-1} \text{ and 255 nLL}^{-1} \text{SO}_2 + \text{NO}_x \) represented a true increase in yield as grain quality was maintained. At 255 n\text{LL}^{-1} \text{SO}_2 + \text{NO}_x \) grain protein concentration showed a slight increase.

In triticale exposure to both \( \text{SO}_2 \) and \( \text{SO}_2 + \text{NO}_x \) had similar effects on protein concentration resulting in a slight increase at the two highest \( \text{SO}_2 \) concentrations.

Protein concentration was increased in white clover only at 388 n\text{LL}^{-1} \text{SO}_2 + \text{NO}_x \) in conjunction with a very severe reduction in plant biomass. Burr medic showed an increase in protein concentration at the lower \( \text{SO}_2 \) concentrations of 80 n\text{LL}^{-1}, 139 n\text{LL}^{-1} \text{ and 255 nLL}^{-1} \text{SO}_2 + \text{NO}_x \) but this occurred in conjunction with a decrease in biomass. Thus, overall yield was not increased.

In both pasture species at most \( \text{SO}_2 \) concentrations, \( \text{NO}_x \) had little effect on foliar sulphur concentration but at 388 n\text{LL}^{-1} \text{SO}_2 + \text{NO}_x \) a synergistic effect occurred greatly increasing sulphur content of the foliage and consequently the severity of effects on growth.

In the other crop species (Chapter 4) the presence of \( \text{NO}_x \) either reduced or had no effect on the total sulphur content of the foliage. The increase observed in white clover and burr medic may reflect a change in uptake, redistribution or re-emittance processes due to an interactive effect between high \( \text{SO}_2 \) concentrations and \( \text{NO}_x \).
CHAPTER 7: DOSE-RESPONSE RELATIONS FOR AUSTRALIAN CROP, FORESTRY AND NATIVE PLANT SPECIES

7.1 INTRODUCTION

From the previous discussions of each experiment it is clear that the responses of individual species are quite varied and complex especially when simultaneous exposure to SO$_2$ and NO$_x$ occurs.

As the primary aim of this project was to establish criteria to protect against agricultural crop losses, reduction in forestry production and loss of ecosystem integrity, it was considered that use of the parameters total plant dry weight (growth) for the tree and pasture species, and total seed dry weight (yield) for the grain crops, was most relevant in quantifying response for determination of generalised dose-response relationships. To standardise across all experiments the mean total plant dry weight or mean total seed dry weight for each treatment was converted to a percentage change in comparison to the control treatment (ambient air). Thus, a positive percentage change represents growth or yield greater than the control and a negative percentage change represents growth or yield less than the control.

As a small data base also exists for the effects of SO$_2$ on Australian plant species other than those considered under this project (Table 5), determination of a SO$_2$ dose-response relationship incorporating the data from all these species is discussed. However, initially only data from the three experiments discussed in Chapters 4-6 is considered to provide continuity with the later discussion of the effects of SO$_2$ and NO$_x$ in combination.

7.2 SULPHUR DIOXIDE

7.2.1 Species Studied under the ERDC Project

Figure 23 shows the percentage change in comparison to control treatments (ambient air) for each species, due to exposure to SO$_2$ alone.

Clearly the concentration-response relationship is different for each species, especially *Pinus radiata* which was resistant to SO$_2$ exposure over the concentration range considered. Wheat and triticale showed a similar response to SO$_2$ with an initial stimulation in yield below about 200 nLL$^{-1}$ SO$_2$, whereas barley and oats showed a no-threshold response. Burr medic, barrel medic and white clover also showed no-threshold responses while sub-clover and lucerne were little affected until SO$_2$ concentrations reached 100 nLL$^{-1}$ and 200 nLL$^{-1}$ respectively. *Eucalyptus pilularis* showed a threshold around 150 nLL$^{-1}$ but *E. microcorys* and *E. regnans* showed a steady decline in growth across the entire SO$_2$ concentration range considered.
Figure 23: $\text{SO}_2$ concentration-response curves for each species studied under the ERDC grant, grouped according to plant type.
If the percentage change response curves of all these species except *P. radiata* are combined, a statistically significant quadratic concentration-response curve can be fitted to the data \((p = <0.001)\) which accounts for 66% of the data variation \((r = 0.81)\) and shows a slight threshold response (Figure 24). A simple linear regression can also be used to describe this response curve but gives a less reliable prediction of response.

The equation for this concentration-response curve gives a threshold concentration of 7 nLL\(^{-1}\) SO\(_2\) for zero growth or yield loss if exposure is for 4 h/day, daily for four-five months.

All three experiments considered similar long-term, frequent exposure regimes. Thus, use of a "concentration multiplied by hours of exposure" term to describe dose also gave a statistically significant dose-response curve (Figure 25) with a threshold of 4600 nLL\(^{-1}\)h. In a review of several studies on corn and soybean, Irving (1987) also found that SO\(_2\) in the range of 5000 nLL\(^{-1}\)h had no effect on growth or yield. Soybeans were found to be the most sensitive and were generally affected by doses above 10000 nLL\(^{-1}\)h.

To further define the nature of plant response to SO\(_2\) under differing exposure regimes all previous research on the effects of SO\(_2\) on Australian vegetation is considered below.

7.2.2 Other Species Studied

Plant response to pollutant exposure has been described in numerous ways in these other experiments.

To standardise across all experiments the mean total plant dry weight or mean total seed dry weight at final harvest was used and converted to a percentage change in comparison to control treatments (ambient air), as for the previous three experiments discussed.

Several major components of pollutant exposure are of prime importance in interpretation of this data set:

1. pollutant concentration;
2. total length of exposure (no. of days);
3. daily duration of exposure (no. of h/day); and
4. frequency of exposure (eg. no. days/wk).

Establishment of a dose term for pollutant exposure that adequately equates to plant response is fraught with difficulties (Kinsman *et al.*, 1988) due to the
Figure 24: The effect of SO₂ concentration on growth and yield of 12 Australian plant species as determined by frequent exposure to SO₂ for 4 hr/day, daily for four-five months, in open-top chambers.
Figure 25: The effect of SO\textsubscript{2} concentration, taking into account the total number of hours of exposure, on growth and yield of 12 Australian plant species, determined by frequent exposure to SO\textsubscript{2} for 4 hr/day, daily for four-five months, in open-top chambers.
complexity of interactions which determine plant response to a given pollutant exposure (see Chapter 2).

Most researchers have found that the arithmetic mean concentration is the best descriptor of plant response if specified for a given range of exposure lengths and frequencies (Roberts, 1984a). Dose calculated as concentration multiplied by hours of exposure is often used but is normally only a useful descriptor of response when considering similar exposure regimes as in Figure 25. The type and magnitude of injury from equivalent doses produced by very different concentrations is rarely equivalent (Garsed et al., 1982; Fowler and Cape, 1982; Roberts, 1984a). If the exposure pattern is highly intermittent with numerous peak pollutant events and low background concentrations, a percentile measure of pollutant concentration can sometimes be useful, for example the 90th percentile concentration (Kinsman et al., 1988).

As the majority of experiments being considered here were designed to study long duration, relatively low concentration, frequent exposures to constant pollutant concentrations for a given number of hours per day, the use of a percentile term for dose is not appropriate. Therefore, it was decided to initially plot plant response against mean pollutant concentration and subsequently consider daily duration of exposure, total length of exposure and frequency of exposure.

The data set available for analysis here comes from studies of 37 different species/cultivars and contains 168 data points. Mean background concentrations of $SO_2$ and $NO_x$ were usually below reliable detection limits of the analysers used ($<5\text{ nLL}^{-1}$) and hence are quoted as zero. The concentrations stated are mean concentrations for the stated daily duration of exposure (eg. 4 h/day).

Only data from one experiment considering $SO_2$ only, Murray et al. (1990 b), were not directly comparable to the others as several treatments used in this experiment were designed to simulate infrequent exposure regimes (1 h/wk, 1 h/mth and 1 hour only). These particular treatments were initially omitted from the $SO_2$ data set for consideration separately.

7.2.2.1 Frequent Exposure Regimes

The initial plot of mean concentration against response, for sulphur dioxide, gave a poor correlation coefficient for a simple linear regression and showed several outlying points not typical of the remainder of the data set. Upon examination these were found to be attributable to four tree and shrub species. The first was $Pinus\ radiata$ as discussed previously. The other three were $Banksia\ attenuata$, $Banksia\ menziesii$ and $Hakea\ incrassata$ which showed large increases in biomass across an exposure range of 23 nLL$^{-1}$ to 181 nLL$^{-1}$ $SO_2$, 2.5-h, 3 days/wk for 140 days. Due to the large differences between the responses of these four species from the response displayed by all other species,
they were omitted from this analysis. Figure 26 shows the plot of concentration against response for the remaining species. A simple linear regression best describes this concentration-response curve (p < 0.001, r = 0.79).

As this data set represented many different species and cultivars, the response of each species/cultivar was then plotted separately to see whether any trends due to species type became evident. Australian native species were plotted separately to crops (Figures 27 and 28 respectively), as it was anticipated that Australian native species might show a basic difference in response compared to crop plants due to basic differences in physiology.

Figure 27 highlights the variability of response of different species of the same genera, *Eucalyptus*, to the same SO$_2$ concentration. For example, exposure to 50 nLL$^{-1}$ SO$_2$ produced a range of responses from -36% for *E. gomphocephela* to +54% for *E. marginata*. This was not due to differences in daily duration of exposure, total length of exposure or environmental conditions as this particular range of response was recorded in the same experiment wherein exposure conditions and environmental conditions were the same for all species. Thus the difference in response is solely attributable to species variation.

Similarly a wide range of response can be seen in the crops, for example, +12% for wheat (*Triticum aestivum* cv. Banks) to -82% for the particularly sensitive species burr medic (*Medicago polymorpha* cv. Santiago) due to exposure to approximately 150 nLL$^{-1}$ SO$_2$ (Figure 28). This range of response occurred between two different experiments but both used an exposure regime of 4 h/day, wheat being exposed for 108 days and burr medic for 149 days. It is also apparent that different cultivars within the same species show different responses.

Figure 29 shows the data for all *Eucalyptus* species plotted according to exposure regime. No trends due to daily duration of exposure are evident. The data points for the 2.5-h, 3 days/wk exposure regime represent the response of a single species, *Eucalyptus wandoo*. It can be seen that exposure to approximately 50 nLL$^{-1}$ SO$_2$ for 2.5-h, 3 days/wk in this species is producing a growth depression of the same order of magnitude as exposure to 50 nLL$^{-1}$ SO$_2$ continuously in another species.

A similar plot for all crop species (Figure 30) shows no apparent trend in response that can be attributed to daily duration of exposure. Exposure for 4 h/day is producing growth depression of the same order of magnitude as continuous exposure and exposure for 8 h/day. When the dose-response data for the crops and natives are compared as in Figure 31 it can be seen that there is little or no difference in the two data sets therefore all species can be pooled by exposure regime and plotted against SO$_2$ concentration (Figure 32). Again no trend attributable to daily duration of exposure is evident and it seems that
Figure 26: The effect of SO$_2$ concentration on growth and yield of 33 Australian plant species as determined by frequent exposure to SO$_2$ in open-top chambers. The conditions of this data set include continuous exposure, exposure for 8hr/day, 4hr/day, 2.5hr/day and 2.5hr, 3 days/wk.
Figure 27: The effect of SO\(_2\) concentration on growth of 13 Australian native plant species as determined by frequent exposure to SO\(_2\) in open-top chambers. The conditions of this data set include continuous exposure, exposure for 8hr/day, 4hr/day, 2.5hr/day and 2.5hr,3 days/wk.
Figure 28: The effect of SO2 concentration on yield of 20 Australian crop species/cultivars as determined by frequent exposure to SO2 in open-top chambers. The conditions of this data set include continuous exposure, exposure for 8hr/day and 4hr/day.
Figure 29: The effect of exposure regime on the growth response of 11 *Eucalyptus* species subjected to frequent exposure to SO$_2$ in open-top chambers.

Figure 30: The effect of exposure regime on the yield response of 20 crop species/cultivars subjected to frequent exposure to SO$_2$ in open-top chambers.
Figure 31: A comparison of concentration-response curves for 20 Australian crop species/cultivars and 13 Australian native plant species as determined by frequent exposure to SO$_2$ in open-top chambers. The conditions of this data set include continuous exposure, exposure for 8hr/day, 4hr/day, 2.5hr/day and 2.5hr,3days/wk.
Figure 32: The effect of exposure regime on the response of 33 Australian plant species subjected to frequent exposure to SO₂ in open-top chambers.
exposure for 2.5-h, 3 days/wk has effects of similar magnitude to daily exposure.

To determine whether total length of exposure (no. of days) is a significant determining factor in the magnitude of the response recorded at the final harvest, total length of exposure was plotted against response for given SO₂ concentrations. Sufficient data were available for only narrow concentration ranges and these are shown in Figures 33 and 34. It is apparent that length of exposure has little effect on the magnitude of response within the range 90-130 days. Experiments harvested at 130 days show a similar range of response to those harvested at 90 days.

As the purpose of this analysis was to establish a SO₂ criteria to protect a range of species under various environmental conditions, a conservative approach considering the full range of dose-response data available to date must be used.

Pooling the data for all species (Figure 26) gave a generalised dose-response curve that could be used to predict the SO₂ concentration at which zero yield or growth loss occurred. This dose response curve is described by the equation:

\[
y = 4.81 - 0.19x \\
r = 0.79, p<0.001
\]

where \( y \) = % change in yield or growth
\( x \) = SO₂ concentration (nL⁻¹)

Zero yield or growth loss is predicted to occur at 25 nL⁻¹ SO₂. This is a mean SO₂ concentration predicted from continuous exposure (8 species, 27 data points), exposure for 8 h/day (7 species, 21 data points), 4 h/day (12 species, 60 data points), 2.5 h/day (1 species, 3 data points) and 2.5 h/day, 3 days/wk (2 species, 10 data points).

The results of this analysis show that the daily duration of exposure was not closely related to yield or growth (Figure 29, 30 and 32) and that there was little difference between the magnitude of response to 4 h/day and continuous exposure. However, the bulk of the data for concentrations above 120 nL⁻¹, which was a major component in determination of the form of the dose-response curve, came from experiments conducted with an exposure regime of 4h/day, daily for four-five months. Thus, the predicted concentration for zero yield/growth loss based on this graph (Figure 26) is best quoted as a 4-h average of 25 nL/L for daily exposures occurring repeatedly for periods of up to five months.
Figure 33: The effect of total length of exposure on response for Australian crop and native species subjected to frequent exposure to SO$_2$ in open-top chambers in the concentration range 50-55 nL/L SO$_2$.

Figure 34: The effect of total length of exposure on response for Australian crop and native species subjected to frequent exposure to SO$_2$ in open-top chambers in the concentration range 95-125 nL/L SO$_2$. 
To test whether the correlation between exposure and response can be improved by using an exposure term other than concentration, three other terms were tested, these being:

1. \( \log \text{SO}_2 \) concentration;
2. \( \text{SO}_2 \) concentration \( \times \) total no. of hours of exposure; and
3. \( \log (\text{SO}_2 \ \text{conc.} \times \text{total no. hours}) \).

None of these terms improved the correlation coefficient. The two terms incorporating the total number of hours of exposure gave very low correlation coefficients, as might be expected, considering the above results.

Thus, concentration is the best descriptor of exposure for the exposure types being considered and the regression equation for Figure 26 gives an initial guideline for the threshold of effects on growth and yield due to long term, low concentration, frequent exposures to \( \text{SO}_2 \). The limited amount of information available on infrequent exposure regimes and also visible injury will now be briefly considered.

### 7.2.2.2 Infrequent Exposure Regimes

Murray et al. (1990b) fumigated five native species at 56 nLL\(^{-1}\) for 1 h/wk, 67 nLL\(^{-1}\) for 1 h/mth and 376 nLL\(^{-1}\) for 1-h only in addition to the range of concentrations for 2.5-h, 3 days/wk as previously discussed.

The results of this single experiment suggest that exposure to 56 nLL\(^{-1}\) for 1h/wk produces changes in growth of the same order of magnitude as similar concentrations for 2.5-h, 3 days/wk, whereas the growth response to 67 nLL\(^{-1}\) for 1 h/mth and 376 nLL\(^{-1}\) for 1-h only seems to be quite different.

However, as there are only a few data points and three of these species showed atypical responses to the frequent exposure regime used (see section 7.2.2.1), it is very difficult to relate these results to the remainder of the data set.

### 7.2.2.3 Visible Injury

Murray et al. (1990b) also considered visible injury due to \( \text{SO}_2 \) exposure. In the experiments previously discussed, injury was assessed primarily in terms of growth or yield changes. Visible injury can be categorised into two types. Acute visible injury is that which develops rapidly, within hours of exposure, due to death of tissue. Chronic visible injury develops over weeks or months due to the disruption of metabolic pathways, primarily those involved in the production or breakdown of photosynthetic pigment.
Four out of five of the native species exposed to 376 nLL-1 for one hour only developed acute visible injury (Murray et al., 1990b) with tissue death evident immediately after exposure. Clearly the threshold SO₂ concentration for visible injury in these species is much lower than 376 nLL-1. Frequent exposure to 181 nLL-1 SO₂ for 2.5-h, 3 days/wk resulted in the development of severe visible injury in these four species (Murray et al., 1990b). Although the exact number of exposures that occurred before onset of visible injury is not known but it was well developed after only eight weeks of fumigation.

O'Connor et al. (1974) found no evidence of acute or chronic visible injury at 300 nLL-1 for 27 hours in 131 Australian tree and shrub species. The most sensitive species all showed visible injury after three hours exposure to 1000 nLL-1. The sensitive species were all of the genus Eucalyptus and included E. regnans. One species common to both acute exposure studies, Acacia saligna, was found by O'Connor et al. (1974) to be quite resistant to acute visible injury from SO₂. This highlights the difficulties of comparing studies conducted using different methodologies. Plants exposed to air pollutants under controlled conditions in growth cabinets (as used by O'Connor) tend to show less severe responses than those exposed under more realistic conditions in open-top chambers.

### 7.3 SULPHUR DIOXIDE AND NITROGEN OXIDES

Determination of a NOₓ dose-response relationship was considered but as data for only a limited number of different NOₓ concentrations are available at present no discernable relationship can be established. Exposure to the two lowest NOₓ concentrations of 82 nLL-1 and 87 nLL-1 led to a range of responses. A 40% growth depression occurred in the three eucalypt species due to 82 nLL-1 NOₓ, comprising 74 nLL-1 NO₂, and a 50% growth depression occurred in white clover due to 87 nLL-1 NOₓ, comprising 29 nLL-1 NO₂. In contrast, burr medic showed a 35% increase in biomass due to exposure to 87 nLL-1 NOₓ, comprising 29 nLL-1 NO₂. In the only other study of an Australian native species to SO₂ + NOₓ exposure, Araucaria cunninghamii (hooppine) showed a significant reduction in height of 11% due to exposure to 85 nLL-1 NOₓ, comprising 76 nLL-1 NO₂, for 2.5 h/day over 116 days (Murray et al., 1990a). Clearly, some Australian species are very sensitive to exposure to NOₓ.

However, establishment of a dose-response relationship for NOₓ alone is not as important as establishment of a dose-response relationship for the interactive effects of SO₂ + NOₓ. This is because NOₓ rarely occurs in isolation around energy generating facilities utilising coal, coal being the primary fuel source for energy generation in Australia at this time. Establishment of a NOₓ dose-response relationship will become important if, for example, there is an increase in energy generation from gas-fired turbines.
Several important points can be noted on the interactive effects of SO$_2$ and NO$_x$ from the studies completed so far. Firstly, nitrogen-fixing leguminous species used for pasture improvement in Australia are particularly sensitive to simultaneous exposure to SO$_2$ and NO$_x$ showing large reductions in productivity if frequently exposed to these pollutants for periods of four to five months. In contrast, cereal crops may show increases in grain production in the presence of NO$_x$ and SO$_2$ concentrations below 200 nLL$^{-1}$, for 4 h/day over their growing season.

Secondly, while Australian native plant species appear very sensitive to NO$_x$ and SO$_2$ exposure individually, the simultaneous presence of low concentrations of SO$_2$ and NO$_x$ can partially ameliorate the growth depression induced by either NO$_x$ or SO$_2$ alone. However, it should be noted that for the *Eucalyptus* species at least, slight growth depressions were still evident due to simultaneous exposure to these low SO$_2$ and NO$_x$ concentrations.

To determine the dose-response relationship for the combined effects of SO$_2$ and NO$_x$ a multiple regression analysis was conducted. Initial inclusion of response data for all species showed that the predictive ability of the SO$_2$ concentration-response curve could not be improved by the inclusion of a NO$_x$ concentration term. This was to be expected given the wide range in response of these species to SO$_2$ and NO$_x$ in combination. All four cereal crops, *P. radiata* and *A. cunninghamii* showed relative resistance to simultaneous exposure to SO$_2$ + NO$_x$. Therefore consideration was given to determining a SO$_2$ + NO$_x$ dose-response curve for the remaining sensitive species, these being the pastures and *Eucalyptus* species. This gave a significant SO$_2$-NO$_x$ concentration-response curve ($p < 0.001$) with both NO and NO$_2$ being significant components in determination of the magnitude of response. This demonstrates that NO is a phytotoxic component of NO$_x$ emissions in addition to NO$_2$. The response curve is described by the following equation:

\[ y = -0.14a - 0.116b - 0.51c - 10.46 \]  

\[ (r = 0.76, p<0.001) \]

where \( y \) = % change in growth or yield  
\( a \) = SO$_2$ concentration (nLL$^{-1}$)  
\( b \) = NO$_2$ concentration (nLL$^{-1}$)  
\( c \) = NO concentration (nLL$^{-1}$)

This shows that there is no threshold for response in these species under the exposure regime being considered. Thus, simultaneous exposure to SO$_2$ and NO$_x$ in combination for 4 h/day, daily for 4-5 months results in a depression of growth or yield at all concentrations for these species.
CHAPTER 8: AUSTRALIAN SECONDARY AMBIENT AIR QUALITY CRITERIA FOR SULPHUR DIOXIDE AND NITROGEN OXIDES

Translation of the data presented in Chapter 7 into ambient air quality criteria for SO\textsubscript{2} and NO\textsubscript{x} requires some consideration of how this has been achieved in other parts of the world. Section 2.2.3 and 2.2.5 has already discussed this in some detail and several important points can be noted:

1. scientific criteria for the setting of standards need to address the effects of long term, low concentration, frequent exposures on growth and yield as well as short term, high concentration, infrequent exposures on the development of acute visible injury;

2. standards usually take the form of maximum one hour, 24 hour and annual averages not to be exceeded; and

3. a conservative approach is required in setting standards from chamber studies to take account of adverse environmental conditions experienced in the field, and the range of responses which occur due to species variability.

It can also be noted that the concentration-response curve obtained for long-term, low concentration, frequent exposures to SO\textsubscript{2} for Australian plant species (Figure 26, Section 7.2.2.1) is consistent in form and of the same order of magnitude as those established by other researchers for non-Australian species. For example, Roberts (1984a) determined a threshold concentration for zero yield loss of 21 nLL\textsuperscript{-1} SO\textsubscript{2} under continuous exposure conditions based on the responses of nine crop species (Figure 3, Section 2.2.3.1). For the single species, *Lolium perenne*, he determined a threshold of 35 nLL\textsuperscript{-1} SO\textsubscript{2} for continuous exposures (Figure 4).

Unlike the work of many other researchers attempting establishment of dose-response data, the data set analysed in Chapter 7 has the advantage that the experiments were all conducted using the same chambers, using adequate air flow rates in a non-polluted environment under similar climatic conditions. Hence, reliable concentration-response curves for long duration, frequent low concentration exposures to SO\textsubscript{2} and NO\textsubscript{x} have been obtained.

As discussed in Chapter 2, insufficient information on materials is available at this stage to establish secondary ambient air quality criteria which take into account both information on economic loss due to materials damage as well as dose-response relations for growth and yield reduction of Australian vegetation. However, establishment of standards based on the scientific criteria for vegetation responses should protect against large economic losses from materials degradation at least in the short term until more information becomes available. At any stage, establishment of criteria with regard to
materials damage will essentially be a question of how much materials degradation is acceptable as no true thresholds exist.

Thus, the following recommendations for the ambient air quality criteria for \( \text{SO}_2 \) and \( \text{NO}_x \) are based solely on vegetation response.

Insufficient data are available at this stage to establish a criterion for short-term, high concentration, infrequent exposures to protect against acute visible injury (see Sections 7.2.2.2 and 7.2.2.3). Acute visible injury has been shown to occur due to a single exposure to \( \text{SO}_2 \) for one hour at an arithmetic mean concentration of 376 nLL\(^{-1}\) \( \text{SO}_2 \), with the indication that the threshold for visible injury effects is substantially lower than this value (Section 7.2.2.3).

Based on this, the international literature and standards established in other countries, it is considered that a one hour criterion for \( \text{SO}_2 \) that reads as follows is sufficient to protect Australian vegetation from acute visible injury:

* a one hour arithmetic mean value of 200 nLL\(^{-1}\) \( \text{SO}_2 \) should not be exceeded.

The establishment of a criterion for longer term exposures to \( \text{SO}_2 \) based on Figure 26 requires some interpretation as a criterion specifying a four hour average concentration not to be exceeded on a daily basis may be difficult to administer and prove too restrictive of industrial operations. The main reason for this is that fumigation patterns around industrial sources fluctuate according to changes in operational and meteorological conditions.

The dose-response data presented in section 7.2.2.1 indicates that for daily exposure to elevated \( \text{SO}_2 \) levels the magnitude of response is not proportional to the number of hours of the exposure. The threshold for effects as determined from Figure 26 is most accurately quoted as a four hour average, however, a simple mathematical reduction of this four hour average to a twenty-four average cannot be justified. The experimental data available suggests that such a twenty-four hour average would be unduly restrictive.

Given that the threshold from Figure 26 is based primarily on daily four hour exposure to \( \text{SO}_2 \) for periods of four to five months, and that a conservative approach should be adopted in the application of chamber studies to allow for interaction with other environmental stresses and species variability, it is considered that a four hour criterion for \( \text{SO}_2 \) should be set as follows:

* a four hour arithmetic mean value of 25 nLL\(^{-1}\) \( \text{SO}_2 \) should not be exceeded more than once per week.

Used in conjunction with the one hour criterion of 200 nLL\(^{-1}\) \( \text{SO}_2 \) not to be exceeded, this should ensure protection of Australian vegetation from \( \text{SO}_2 \) injury under most environmental conditions.
Consideration of the dose-response data for all fourteen species studied so far, in relation to the combined effects of SO\textsubscript{2} and NO\textsubscript{x}, indicates that overall the presence of NO\textsubscript{x} does not change the form of the long-term exposure SO\textsubscript{2} dose-response curve. However, as discussed in section 7.3, there are clearly particular plant types for which this does not hold true. Nitrogen-fixing leguminous species and species of the genus *Eucalyptus* are particularly sensitive to simultaneous exposure to SO\textsubscript{2} and NO\textsubscript{x}, showing a no-threshold response to frequent exposures of 4 h/day over periods of four to five months.

This has important implications for energy generating facilities located near native eucalypt forests or agricultural land using improved leguminous pastures. It should also be taken into consideration when the site selection for new energy generating facilities is being undertaken and in the management of land use around such facilities.

In the case of *Eucalyptus* species, growth reductions are likely to be least severe if emissions can be controlled to produce ground level concentrations of SO\textsubscript{2} and NO\textsubscript{x} of equal proportion, that is, in a ratio of 1:1 SO\textsubscript{2}:NO\textsubscript{x}. Due to the nutritional role of atmospheric SO\textsubscript{2} and NO\textsubscript{x} emissions in the growth of these species and the importance of the sulphur to nitrogen ratio required for growth, a 1:1 SO\textsubscript{2}:NO\textsubscript{x} ratio would afford least disruption to the existing nutritional status of those plants subject to emissions.

For the leguminous pasture species exposure to SO\textsubscript{2} and NO\textsubscript{x} resulted in severe effects on growth at all concentrations considered, although only lucerne and burr medic showed strongly synergistic effects.

At this stage it must be concluded that SO\textsubscript{2} and NO\textsubscript{x} emissions should be kept to a minimum in areas adjacent in improved leguminous pastures and native eucalypt forests.

It is therefore difficult to recommend a criterion for SO\textsubscript{2} and NO\textsubscript{x} in combination until a threshold for effects can be determined for such species. The World Health Organisation guideline for nitrogen oxides states that in "the presence of levels of sulphur dioxide and ozone not higher than 11 nLL\textsuperscript{-1} and 31 nLL\textsuperscript{-1} respectively, the atmospheric concentration of nitrogen dioxide should be no higher than 16 nLL\textsuperscript{-1} as a yearly average of 24 hour means and no higher than 50 nLL\textsuperscript{-1} as a four hour average" (WHO, 1987).

For Australian conditions the following criterion would be more appropriate:

* in the presence of levels of sulphur dioxide not higher than 25 nLL\textsuperscript{-1}, the atmospheric concentration of nitrogen oxides (the sum of the nitric oxide and nitrogen dioxide concentrations) should be no higher than 25 nLL\textsuperscript{-1} as a four hour arithmetic mean.
This takes into account the following points:
1. both nitric oxide and nitrogen dioxide are toxic to the growth and productivity of Australian plant species;

2. based on the responses of all species studied the presence of NO\textsubscript{x} has no determinable effect on the nature of the dose-response relationship established for long term, frequent exposures to SO\textsubscript{2};

3. for those species particularly sensitive to SO\textsubscript{2} + NO\textsubscript{x}, frequent exposure to 4-h average SO\textsubscript{2} and NO\textsubscript{x} concentrations no higher than 25 nLL\textsuperscript{-1} should minimise losses in growth and productivity, although this is primarily an assumption and requires confirmation; and

4. due to the interaction of SO\textsubscript{2} and NO\textsubscript{x} with the nutritional requirements of plants for sulphur and nitrogen and the importance of the sulphur to nitrogen ratio required for plant growth, exposure to SO\textsubscript{2} and NO\textsubscript{x} should be least disruptive of plant metabolism when low concentrations in relatively equal proportions occur.

Given the information currently available, this criterion, when used in conjunction with the two criteria for SO\textsubscript{2} previously quoted, should initially be adequate to protect human welfare in Australia.

Clearly, more research is needed to establish the threshold for effects of SO\textsubscript{2} and NO\textsubscript{x} on particular species and determine whether the SO\textsubscript{2}/NO\textsubscript{x} criterion proposed is adequate to protect against substantial losses in the productivity of leguminous pastures and to prevent disruption of the growth of our native eucalypt forests.

At this stage the relative importance of NO and NO\textsubscript{2} in NO\textsubscript{x} emissions is also still uncertain. It has been established that NO is phytotoxic to Australian plant species but the interactive effects of NO and NO\textsubscript{2} when present in various ratios, as occurs in industrial emissions, are unknown. To a certain extent, NO and NO\textsubscript{2} have differing effects on plant metabolic processes (Wellburn, 1990) and thus some differences in plant growth and yield response could be expected due to NO\textsubscript{x} emissions containing different NO to NO\textsubscript{2} ratios. This requires further research. At present the SO\textsubscript{2}/NO\textsubscript{x} criterion proposed lends equal importance to the effects of NO and NO\textsubscript{2}. Future research may show that one is more significant in determining response than the other.
TECHNOLOGY TRANSFER

Throughout this project the design, results and implications of the work have been discussed with the project Advisory Committee, consisting of representatives from the Air Quality Committee of the National Health and Medical Research Council, Australia and New Zealand Environment Council, Electricity Supply Association of Australia, Environment Protection Authority of Victoria, Environmental Protection Authority of WA, NSW State Pollution Control Commission, Electricity Commission of NSW, Queensland Electricity Commission and State Energy Commission of Western Australia. These discussions proved extremely useful in ensuring that the work remained practical and that the electricity generating industry and government regulators were made aware of the results and implications of the work as well as the facilities available to industry for specific design related work associated with individual projects.

The outcome of this interaction was greater emphasis on the determination of the assimilative capacity of particular environments, to establish the environmental effects of various emission design scenarios and the severity of these effects if any occur, to optimise protection of the environment while minimising costs by avoiding unnecessary emission controls.

A broad range of organisations have sought information about the results of this work from the perspective of predicting impacts of emissions of proposed new projects. It has been found that the best means of providing ready access to the results has been through publication in the open literature. Consequently, to achieve this, the results of this work are in the process of being published in major international scientific journals. In the open literature the information will be most readily available to the electricity generating industry, environmental protection authorities, all heavy industry and private consultants to industry amongst others.

In order to bring the existence of this information to the broader industrial community the project approach and results have been discussed at a number of meetings including the NERDDC-sponsored workshop on Air Quality Standards and the Energy Industry held in Canberra, the CAPER meeting on Air Pollution Effects, the World Clean Air Congress meeting in Sydney and regional meetings with industry and pollution control authorities. In addition the project has been covered by television, radio and print media.

Technologies developed during the course of this project are available to, and used by Australian industry, usually for the prediction of impacts of electricity generating plant. The facilities have recently been used by the Queensland Electricity Commission, the State Energy Commission of WA, CRA and Barrack Mines.
The technology also has a valuable role for the training of undergraduate and postgraduate students in modern environmental monitoring and environmental protection, as this equipment is generally not available in universities and colleges, for reasons of cost. Many students have used the facilities and the preliminary results of this work to understand the principles and practice of environmental monitoring, the impacts of industrial air pollutants, the use of dose-response relationships, and the principles of environmental protection.
PUBLICATIONS FROM THIS WORK TO DATE


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