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#### REVIEW ARTICLE

# Nitrogenous pollutants in the atmosphere: Their assimilation and phytotoxicity

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Most plants possess well-characterized physiological and biochemical systems to absorb nitrogenous gases from the atmosphere and to assimilate the nitrogen into organic nitrogenous compounds. The system is often activated during the exposure of plants to low level of the gases with concurrent increases in organic nitrogen content and growth of the plants. The nitrogenous gases, however, are not the complete replacement of soil nitrogen or other usual modes of nitrogen nutrition and often the exposure to these gases at relatively higher concentrations results in some physiological and morphological aberrations in plants. Apparently, there are some unknown constraints on the optimum utilization of nitrogenous gases as sources of nutrient nitrogen, which are perhaps linked to their phytotoxic reactions.

NITROGENOUS gases from natural as well as anthropogenic sources are important contributors to the atmospheric pollution. Total annual emission of these gases has been estimated to be about  $64 \times 10^8$  tons from natural sources and about  $57 \times 10^8$  tons from man-made sources<sup>1</sup>. Nitric oxide,  $NO_2$ ,  $N_2O$  and  $NH_3$  are the principal components of these pollutants, although trace amounts of other nitrogenous compounds such as  $N_2O_5$ ,  $HNO_3$  vapours, nitrogenous amines and volatile hydrocarbons are also present in the atmosphere. Nitrogen oxides  $(NO_x)$  are formed by the heat-promoted combination of atmospheric nitrogen and

oxygen. Oxidation of nitrogenous compounds in fuel and biological materials and microbial conversion of excess nitrogen fertilizers in the soil also contribute towards NO<sub>x</sub> production.

The phytotoxicity of nitrogenous air pollutants is well established<sup>2-4</sup>. Considering that nitrogen is the most abundant (constituting about 1 to 5% of a plant's dry weight) mineral element in plants and that most of the nitrogenous pollutants are assimilable compounds, these phytotoxic effects are rather inconceivable. But, careful studies with low levels of many pollutants have shown that the pollutants do increase plant growth in some cases<sup>4,5</sup>. The growth of plants in the presence of either NO or NO<sub>2</sub> as a sole source of nutrient nitrogen, however, is not comparable to that with soil nitrate as nutrient nitrogen<sup>4,6,7</sup>. Clearly, there are some constraints on the optimum utilization of NO/NO<sub>2</sub> and perhaps other nitrogenous gases as well, as a source of nutrient nitrogen. The knowledge of the physiological and biochemical basis of phytotoxicity may help in understanding the possible constraints in the role of nitrogenous air pollutants as alternate sources of nitrogen.

## Ammonia

In temperate zone toposphere, the concentration of ammonia has been recorded to be around 10 ppb (ref. 8),

while that above the tropical rain forests, about 15 ppb (ref. 9). The sources of ammonia emission are both natural and anthropogenic. The vegetation acts both as a source as well as sink for the pollutant.

## Absorption and assimilation

Numerous studies have indicated that atmospheric ammonia might be taken in, either through the leaves, or indirectly through the soil. The deposition velocity for the leaves of fescue, maize, oats, orchard grass, sap bean and soybean has been estimated to be in the range of 39 to 539 nmol m<sup>-2</sup> s<sup>-1</sup> (ref. 10), while that for cotton and sunflower, it was 6 to 9 nmol m<sup>-2</sup> s<sup>-1</sup> (ref. 11). The foliar absorption of the gas is primarily stomatal, which accounts for up to 97% of the total uptake<sup>12</sup>. The atmospheric level of NH<sub>3</sub> is the primary factor determining its uptake rate by the plants. Generally the uptake rate increases with increase in ammonia concentration<sup>13,14</sup>. Atmospheric ammonia may combine with SO<sub>2</sub> and humidity present in the air to form (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub> aerosol, which can then be absorbed by the plants. As is the case with gaseous ammonia, the absorption of  $(NH_4)_2SO_4$  aerosol is also stomatal<sup>15</sup>.

Assimilation of absorbed NH<sub>3</sub> has been indicated in several investigations. This has been demonstrated by increased total nitrogen content during NH<sub>3</sub> exposure<sup>16</sup> and also by the use of <sup>13</sup>N labelled<sup>17</sup> or <sup>15</sup>N labelled<sup>13</sup> NH<sub>3</sub>. From the study of pattern of amino acid labelling during <sup>13</sup>NH<sub>3</sub> exposure, it has been suggested that the assimilation takes place through glutamine synthetase-glutamate oxoglutarate aminotransferase (glutamate synthase) (GS-GOGAT) pathway<sup>17</sup>. The foliar NH<sub>3</sub> is readily transported to roots<sup>13</sup>.

Absorption and assimilation of NH<sub>3</sub> has the potential of NH<sub>3</sub> serving as a source of nitrogen for plants, especially in those conditions where soil is deficient in nitrogen. Faller<sup>18</sup> has reported that sunflower plants could be grown with atmospheric NH<sub>3</sub> without any additional nitrogen source. But this has not been confirmed in other species, although an increase in plant growth by exposure to NH<sub>3</sub> has been observed in perennial rye grass also<sup>19</sup>.

#### **Phytotoxicity**

Significance of either NH<sub>3</sub> or other nitrogenous gases as a source of nutrient nitrogen is to be viewed in conjunction with their toxic effects on plants. At relatively higher dosage, NH<sub>3</sub> produces visible symptoms of injury such as necrosis, chlorosis and growth reductions<sup>20</sup>. At lower concentrations, the gas may alter the physiology with or without any visible damage. One of the primary effects of gas is to cause alkalinity in the

cell sap; it produces OH in the aqueous medium:

$$NH_3 + H_2O \rightleftharpoons NH_4^+ + OH^-$$
 (pKa 9.24).

Although, there are no data on the effect of gaseous NH<sub>3</sub> on vacuolar or cytoplasmic pH of the cell sap, the pH of the maize root tips incubated in aqueous NH<sub>4</sub> is known to increase<sup>21</sup>. Besides an increase in pH, following physiological and biochemical disorders might be responsible for the toxic symptoms in NH<sub>3</sub>. exposed plants (Figure 1). These disorders might arise either due to excessive accumulation or assimilation of NH<sub>3</sub>. (1) Accumulation of NH<sub>4</sub> inhibits photosynthetic phosphorylation, which may ultimately lead to reduction in net photosynthesis and other photogenerated ATPdependent processes, and in plant growth<sup>22,23</sup>. (2) Ammonia may saturate lipids in cellular membranes<sup>20</sup>, which may increase cellular permeability leading ultimately to plasmolysis and necrosis. (3) Excessive ammonia assimilation may drain out NAD(P)H and ATP, to the extent that there is an active competition between photosynthetic CO, assimilation into carbohydrates and NH<sub>3</sub> assimilation into amino acids. The net result is the inhibition of photosynthesis and plant growth. Evidences for such a competition have been obtained from experiments, where NH, has been shown to increase the flow of recently fixed carbon in the tricarboxylic acid (TCA) cycle intermediates through increased phosphoenol pyruvate carboxylase and pyruvate kinase activities<sup>24,25</sup>. On the other hand, the pollutant inhibits ribulose bisphosphate carboxylase activity in Sinapis alba<sup>26</sup>. This ultimately leads to increased CO<sub>2</sub> flux into amino acid biosynthesis<sup>24,26</sup> and decreased carbohydrate biosynthesis. If this postulate is to be correct, one would expect total or

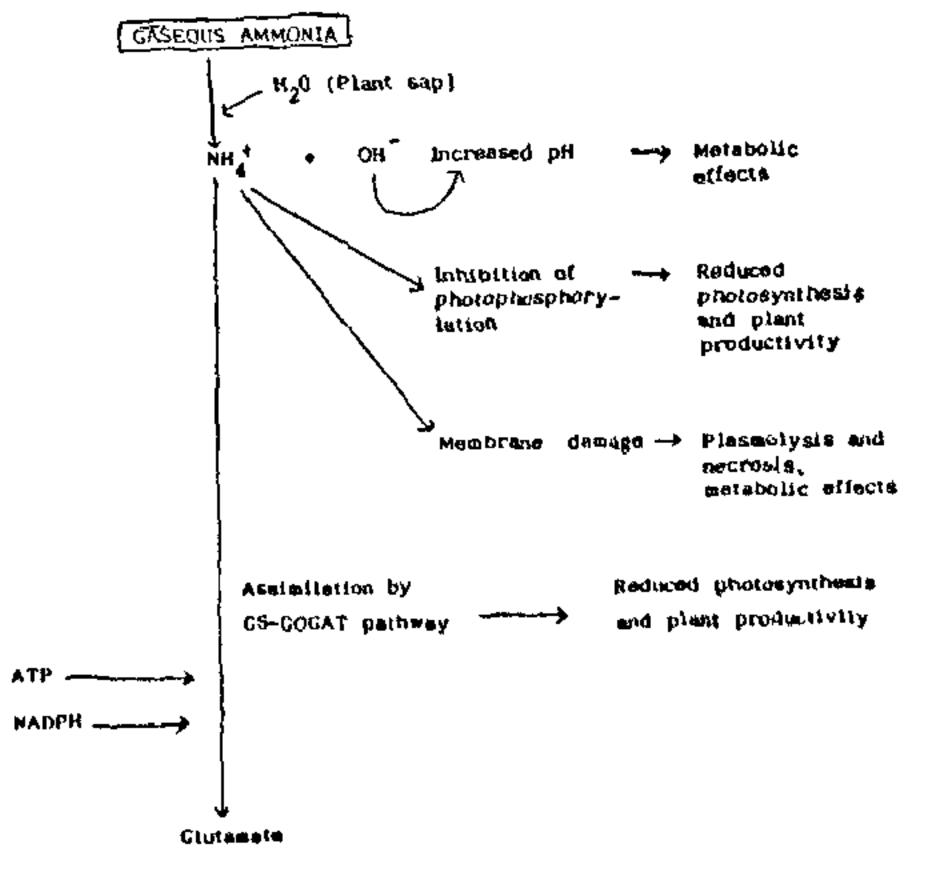


Figure 1. Possible sites of phytotoxic action of NH, and its products.

partial alleviation of NH<sub>3</sub> phytotoxicity under conditions which favour maximum generation of carbon skeletons and photogenerated reductants, such as under high irradiance.

#### Nitrogen dioxide

Nitrogen dioxide is the most important among NO<sub>x</sub> pollutants. The primary sources of this gas are automobile exhausts, power and heat generation plants and many industrial processes involved in the manufacture of nitrogenous chemicals. In fact, in all types of heat-generating reactions, the atmospheric nitrogen and oxygen combine to form NO, which is readily oxidised to NO<sub>2</sub>:

$$\frac{1}{2} N_2 + \frac{1}{2} O_2 \stackrel{\text{heat}}{\rightleftharpoons} NO (\Delta E, -21.6 \text{ kcal})$$

$$NO + \frac{1}{2} O_2 \rightleftharpoons NO_2 (\Delta E, +4.6 \text{ kcal}).$$

Nitrogen dioxide is involved in the generation of other phytotoxic pollutants such as O<sub>3</sub> and peroxyacyl nitrates (PAN). The average atmospheric concentration of NO<sub>2</sub> is generally low, in the range of 10-50 ppb, but it may reach up to 400 ppb in heavily polluted localities<sup>27</sup>.

#### Absorption and assimilation

Nitrogen dioxide may reach plant system either directly through its foliar deposition or indirectly through rain water or soil deposition. The surface deposition of the gas on the foliage is governed by a variety of plant and environmental factors, including pubescence, cuticular reactivity, foliar hydration states and temperature<sup>28</sup>. The leaf penetration is through open stomata and is governed by various factors including the plant species<sup>29</sup>, plant age<sup>30</sup>, concentration of NO<sub>2</sub> (ref. 30) and a variety of environmental and nutritional factors<sup>31,32</sup>. Direct evidence for the foliar absorption of NO<sub>2</sub> has been obtained by using <sup>15</sup>N isotope of nitrogen. Among the seven species examined by Okano et al.29, the maximum absorption was by three cultivars of Populus hybrids; it was equivalent to 0.3 mg N dm<sup>-2</sup> d<sup>-1</sup>. The uptake rate generally increases with the increase in NO<sub>2</sub> concentration, as has been observed in bean in the concentration range of 100-400 ppb (ref. 33), in potato in a concentration range of 120-430 ppb (ref. 34) and in sunflower and maize in a concentration range of 200-1000 ppb (ref. 35). After its entry to the leaf, NO<sub>2</sub> is rapidly translocated to all other parts of the plant, but for the mature leaves<sup>36</sup>.

Nitrogen dioxide may reach plants through rain

water as well, in the form of HNO<sub>3</sub> and HNO<sub>2</sub>. Because of the high solubility of this gas in the atmospheric humidity, its residence time in the atmosphere is only about one week<sup>37</sup>. The nitrous and nitric acids may be absorbed either by the leaves in contact with the acids or by roots in case the acids are settled down in the soil. Soils, specially the alkaline soils, may directly absorb NO<sub>2</sub> from the atmosphere also<sup>38,39</sup>. There is every possibility that the NO<sub>2</sub> absorbed by the soil is taken up by the plants.

Assimilation of NO<sub>2</sub> has been determined primarily by measuring the increase in organic nitrogen content of the exposed plants. Significant increases in Kjeldahl/total nitrogen due to NO<sub>2</sub> exposure have been observed in the kidney bean, potato, spinach, tomato and many other species<sup>5</sup>. Yoneyama et al.<sup>36</sup> have shown that nitrogen of the gas is first incorporated into ethanol soluble fraction and then gradually into insoluble fraction. By using <sup>15</sup>NO<sub>2</sub>, Rogers et al.<sup>33</sup> have shown that about 65% of the absorbed NO, is incorporated into organic nitrogen during a three-hour exposure period in bean. Murray and Wellburn<sup>40</sup> reported a 267% increase in total amino acid content of tomato (cv. Alisa craig) leaves during an exposure to 1500 ppb NO<sub>2</sub> for 18 h. In sweet pepper (cv. Bell boy) however, the total amino acid content decreased by 22%. Increased amino acid content during NO<sub>2</sub> exposure has been demonstrated in Citrus natsudaidai seedlings<sup>41</sup>, barley roots and shoots<sup>42</sup> and squash seedlings<sup>43</sup> also.

Several factors influence the incorporation of NO<sub>2</sub> into organic nitrogen. In most studies, the contribution of NO<sub>2</sub> to total organic nitrogen is higher in those plants which are raised at deficient or sub-optimum levels of soil nitrogen<sup>6,42,44</sup>. The increase in organic nitrogen content generally increases with the increase in NO<sub>2</sub> concentration up to a certain level<sup>6,7</sup>. However, at very high concentrations, the organic nitrogen content may decrease as well<sup>45</sup>. In sunflower, 300 ppb NO<sub>2</sub> exerted a nutritional effect on nitrogen-deficient plants, while 2000 ppb NO<sub>2</sub> was phytotoxic at all (0, 5 and 15 mM) nutrient nitrate levels<sup>46</sup>.

Related enzymic determinations have indicated that assimilation of NO<sub>2</sub> involves the established route of inorganic nitrogen assimilation. Nitrate and nitrite produced by the dissolution of NO<sub>2</sub> in the cell sap are reduced by the activities of nitrate and nitrite reductases respectively to generate ultimately NH<sub>4</sub><sup>+</sup> which is then assimilated to glutamate, preferentially through GS-GOGAT pathway. Increase in nitrate reductase (NR) activity following NO<sub>2</sub> exposure has been demonstrated in several systems such as barley shoots<sup>44</sup>, tomato<sup>40</sup>, bean<sup>6,47</sup>, pea<sup>48</sup> and Picea rubers<sup>40</sup> leaves and in the needles of Pinus sylvestris<sup>50</sup>. Increase in nitrite reductase (N<sub>1</sub>R) activity has also been demonstrated in barley shoots<sup>44</sup>, spinach<sup>51</sup> and

tomato<sup>40</sup>. There is a substantial increase in NADH-GOGAT activity in snap bean leaves by exposure to 20-500 ppb NO<sub>2</sub>, although the effect on NADH-GDH (glutamate dehydrogenase) is not very consistent<sup>6</sup>. The activities of transaminating enzymes, glutamate pyruvate transaminase and glutamate-oxaloacetate transaminase are also unaffected by 100 to 1000 ppb NO<sub>2</sub> in pea seedlings<sup>52,53</sup>.

Extrapolating the data from laboratory experiments with NO<sub>2</sub> assimilation, Hanson et al.<sup>54</sup> have calculated annual nitrogen inputs between 0.08 and 1.9 kg ha<sup>-1</sup> yr<sup>-1</sup> for forest canopies in urban environments at prevailing NO<sub>2</sub> levels. Based on annual nitrogen requirements for forest trees, this would contribute about 3% of the tree's annual needs in natural forests and about 10% in urban forests. The nutritive value of NO<sub>2</sub> is reflected in increased growth of plant or plant parts by low levels of NO<sub>2</sub> in some species<sup>6,29,46,55,56</sup>. The stimulation of growth is more apparent at lower soil nitrogen levels than at high levels<sup>6,42,44</sup>, indicating thereby that NO<sub>2</sub> may serve as an alternate source of nutrient nitrogen for plants growing in nitrogen-deficient soils.

## Phytotoxicity

Relatively higher dosage of NO<sub>2</sub> produces visible damage to the plants in the form of bleached, chlorotic or necrotic spots and patches on the leaves. Often the early senescence and abscission of the leaves are also seen. At relatively lower dosage, no visible damage, but the inhibition of vital physiological processes such as that of photosynthesis and chlorophyll metabolism and of enzyme activities is observed<sup>57,58</sup>. This ultimately leads to the reduced plant growth and productivity. Four alternate hypotheses for NO<sub>2</sub> phytotoxicity may be proposed (Figure 2):

Increased acidity. Lee<sup>59</sup> and Zeevart<sup>60</sup> have suggested that damage due to NO<sub>2</sub> is because of increased acidity of the cell. The pH of the leaf juice from bean plants exposed to 5000 ppb NO<sub>2</sub> for 1 h and that from tomato plants exposed to 14000 ppb NO<sub>2</sub> for 1 h decreased by 0.25 and 0.19 units respectively<sup>60</sup>. The magnitude of pH decrease may not appear to be very impressive, but chemical considerations of NO<sub>2</sub> and other gases with acidic or basic reactions indicate the possibility of significant alterations in cellular pH and buffering<sup>61</sup>.

If this hypothesis of increased acidity is to be exclusively responsible for NO<sub>2</sub> phytotoxicity, one would expect alleviation of injury under conditions which favour strong buffering or alkaline environments inside the plant. In one investigation, the addition of 1300-7000 ppb NH<sub>3</sub> (an alkaline agent) vapours in the gas stream reduced the phytotoxicity of NO<sub>2</sub> on celery, bean and pea plants<sup>60</sup>.

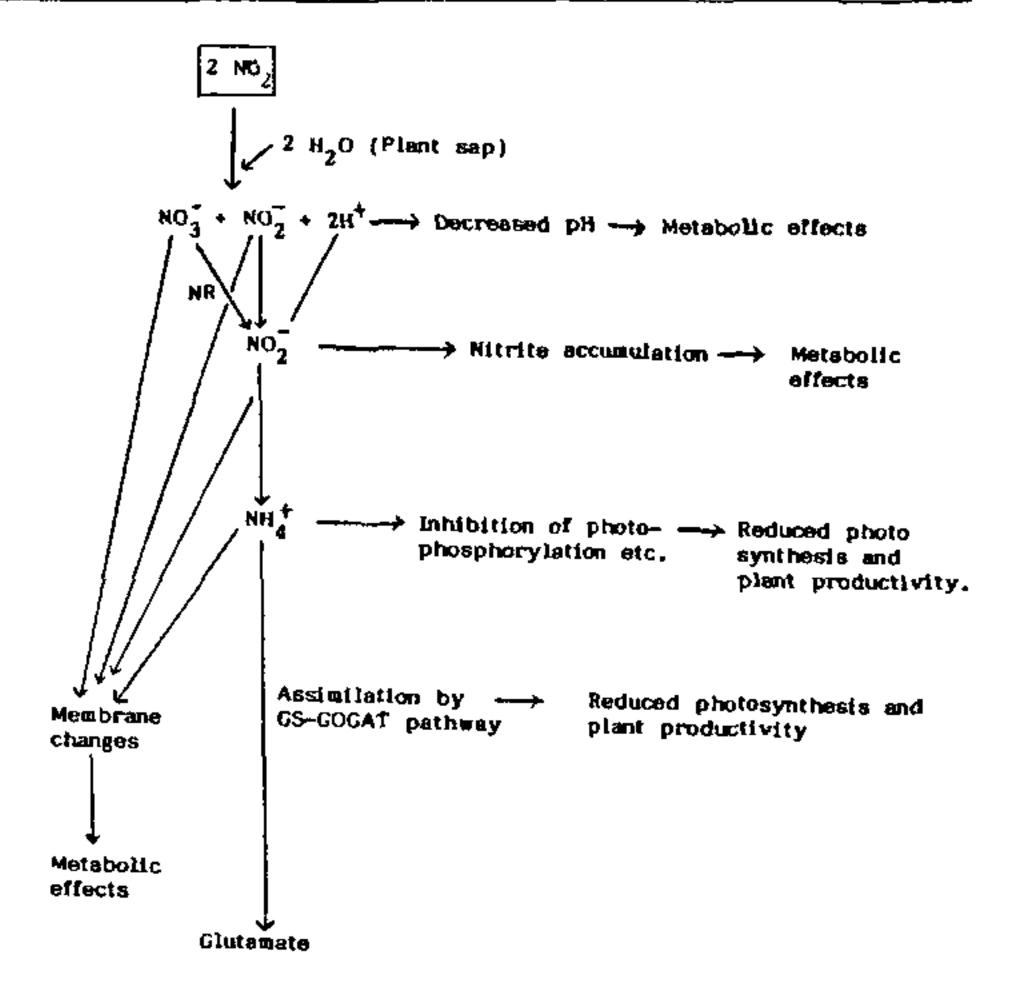


Figure 2. Possible sites of phytotoxic action of NO<sub>2</sub> and its assimilatory products.

Nitrite accumulation. Some observed biochemical effects of nitrite  $(NO_2^-)$  accumulation in plants are: release of Mn from the water-splitting complex in chloroplast thylakoids<sup>62</sup>, effects on thiol-containing enzymes and other proteins<sup>63</sup> and perhaps effects involving free radicals  $(N=0)^4$ . With this background it has been proposed that toxic effects of NO<sub>2</sub> are due to NO<sub>2</sub> accumulation inside the cell<sup>58</sup>. However, the measurement of NO<sub>2</sub> content of NO<sub>2</sub>-exposed plants has yielded variable data; significant accumulation during acute exposure<sup>64</sup> to no accumulation during chronic exposure<sup>6,7</sup>. Further, in squash seedlings, NO<sub>2</sub> content of the leaf and cotyledons increased during an exposure to 4000 ppb NO<sub>2</sub> for 4 h (acute dose) but the final concentration was less than  $10 \,\mu\text{M}$  (ref. 43), ineffective in causing any serious effect. Thus, nitrite toxicity may be relevant under those circumstances only, which favour rapid absorption of NO<sub>2</sub> but decreased reduction of NO<sub>1</sub> generated either from the dissolution of NO<sub>2</sub> or from the reduction of NO<sub>3</sub>.

Ammonium accumulation. It has also been proposed that the phytotoxic effects of NO<sub>2</sub> are due to NH<sub>4</sub><sup>+</sup> accumulation, which arises from the reduction of nitrate and nitrite. Significant accumulation of NH<sub>4</sub><sup>+</sup> following NO<sub>2</sub> exposure has been demonstrated in spinach and kidney bean plants<sup>43,51</sup>. But the interactive effects of externally supplied NH<sub>3</sub>/NH<sub>4</sub><sup>+</sup> and NO<sub>2</sub> seem to be variable. While in vapour form, NH<sub>3</sub> reduced NO<sub>2</sub> injury in some species<sup>60</sup>, in nutrient solution, NH<sub>4</sub><sup>+</sup> augmented the development of visible symptoms of NO<sub>2</sub> injury in bean leaves<sup>65</sup>.

Membrane damage. Changes in membrane structure and organization may be brought about either by NO<sub>2</sub> itself or by its cellular products, such as H<sup>+</sup>, NO<sub>3</sub>, NO<sub>2</sub> and NH<sub>4</sub>. In ritro experiments with artificial membranes have demonstrated disorganization of membranes in several studies<sup>66-68</sup>. This is apparently brought about by auto-oxidation of unsaturated fatty acids in lipids<sup>69,70</sup>. The lipid oxidation may lead to membrane damage, cellular plasmolysis and ultimately to necrosis. Although, in vivo lipid peroxidation or degradation has not been observed in plants, there is some evidence that NO<sub>2</sub> inhibits membrane biogenesis in plants<sup>71</sup>.

Although, at the moment it is not possible to advance a unified mechanism for NO<sub>2</sub> phytotoxicity, it is possible that more than one mechanisms are at work and their operations depend upon the acute or chronic exposure to NO<sub>2</sub> and also upon other plant and environmental factors, including nutrient nitrogen supply.

#### Nitric oxide

Nitric oxide is also produced in heat-generating reactions, by the combination of atmospheric nitrogen and oxygen under high heat. However, NO is readily oxidised to NO<sub>2</sub> and therefore in most experimental generation of NO, NO<sub>2</sub> is present as a contaminant. For this reason, studies with the absorption and assimilation have been conducted jointly for NO and NO<sub>2</sub>. Appreciable levels of NO may exist in greenhouse atmospheres, where kerosene or propane are burnt for CO<sub>2</sub> enrichment of the greenhouse<sup>72,73</sup>. In these experiments, NO in the range of 350-450 ppb has been detected, which sometimes might reach up to 1000 ppm.

#### Absorption and assimilation

Plants may absorb NO through open stomata, although the rate of absorption is about 12 times lower than that of NO<sub>2</sub> (ref. 74). This is apparently because of lower solubility of NO than NO<sub>2</sub> in water. However, it has also been suggested that NO is more soluble in xylem sap than in distilled water<sup>75</sup>, although the reason for this is not known. Since xylem sap is continuous with the extracellular water in leaf, an enhanced solubility of NO in water may be expected over that predicted by in vitro solubility data.

There are some indirect evidences for NO assimilation by the plants, which is apparently through the GS-GOGAT pathway. The pollutant increases activities of enzymes involved in nitrite assimilation, such as NiR, GDH, GS and GOGAT, in sensitive plants<sup>76</sup>. In tomato plants grown at low soil nitrogen levels, 400 ppb NO increased growth substantially, although it could not compensate for the reduction in growth of

plants due to deficiency of soil nitrogen<sup>4,77</sup>. Further, at higher soil nitrogen levels and at higher NO levels, the pollutant was deleterious to the plant growth<sup>77</sup>.

## Phytotoxicity

Exposure to NO produces necrosis and scorching of the leaves, as observed in Diffenbachia maculata<sup>78</sup>. The mechanism of phytotoxicity is however, unknown, although a few postulates have been advanced. Bennett and Hill<sup>74</sup> suggested that NO forms a complex with Fe of ferredoxin and thus may inhibit photosynthesis and other ferredoxin-dependent processes. In greenhouse environments, NO has been found to be about 4 times more toxic to photosynthesis than NO<sub>2</sub> (ref. 79). However, Pickard and Hewitt<sup>80</sup> observed that NO did not react directly with ferredoxin or nitrite reductase (a ferredoxin-dependent enzyme) under strictly anaerobic conditions. It is likely that NO under cellular conditions is oxidized to nitrosonium ions (NO<sup>+</sup>) and nitrite, which are toxic agents.

### Nitrogen pentoxide

Nitrogen pentoxide is generally present in the atmosphere as a contaminant where  $NO_x$  and  $O_3$  are generated. The following types of chemical reactions may be responsible for  $N_2O_5$  generation in ozonators:

$$NO + O_3 = NO_2 + O_2$$
  
 $NO_2 + O_3 = NO_3 + O_2$   
 $NO_3 + NO_2 = N_2O_5$ 

Absorption and assimilation of  $N_2O_5$  by the plants have been demonstrated through indirect experimentation. Increase in nitrate and other nitrogenous components of plants during  $O_3$  exposure in some angiosperms<sup>81,82</sup> has been attributed to the presence of  $NO_x$ , specially the  $N_2O_5$ , as contaminant in  $O_3$ -generating systems. Nitrogen pentoxide is also considered to be responsible for an increase in NR activity during  $O_3$  exposure<sup>83</sup>.

Nitrogen pentoxide may dissolve in either cellular or acellular water to form nitric acid;

$$N_2O_5 + H_2O = 2 \text{ HNO}_3$$
.

This acidity may be the cause of phytotoxicity (cf. Figure 2).

#### Nitric acid vapours

Dry deposition of nitric acid vapours on leaves has been demonstrated in several studies<sup>84,85</sup>. Cadle et al.<sup>86</sup> have demonstrated three routes of dry deposition in

spruce and pine seedlings; surface deposition, transcuticular deposition and stomatal deposition. About 80% of the HNO<sub>3</sub> deposited via the trans-cuticular and stomatal routes was assimilated by the plant but none of the HNO<sub>3</sub> deposited on the surface.

As discussed earlier, the acidity of the HNO<sub>3</sub> may be the cause of its phytotoxicity (cf. Figure 2).

#### Nitrogenous pollutants and forest decline

The nutritive or toxic effects of nitrogenous air pollutants as observed on laboratory-grown plants are significantly amplified when considered in terms of large ecosystems in nature. Forest is one such ecosystem, where existence of various components is affected by the pollutants. Although there have been periodical reports on the decline of natural forests in the past as well, since around 1975 a rapid decline in forests has been observed in many countries of North America, Europe and Asia. Among various hypotheses proposed by Cowling et al.<sup>87</sup> for the natural forest decline, the important ones are: 1. Soil acidification and aluminium toxicity, 2. General stress, 3. Organic air pollutants, and 4. Excess nitrogen deposition or nitrogen saturation.

The nitrogenous gases may be a factor in forest decline according to nitrogen saturation and soil acidification hypotheses. There has been a growing realization that forest trees are able to absorb large amounts of  $NO_x$  and  $NH_3$  from the atmosphere<sup>32,88,89</sup>. The extra nitrogen acquired from these gases disturbs the physiology of forest trees<sup>87,90</sup>, which have evolved through the ages and learned to live in a self-supporting ecosystem. Increased nitrogen nutrition may cause the following imbalances in the ecosystem: 1. With increased availability of nitrogen, there will be an increased demand for other essential nutrients, such as Ca, Mg and P, that are not similarly supplied from the atmosphere. When supply of these elements in the soil is limited, mobilization of nutrients from older leaves causes senescence and abscission of those foliage and subsequently a reduction in total assimilating capacity of the trees<sup>91</sup>. In this manner, there will be a net reduction in overall plant growth. 2. Excess nitrogen may inhibit epiphytic and soil microbial growth, which otherwise is beneficial to the trees. 3. High nitrogen may increase the susceptibility of plants to other biotic and abiotic stresses<sup>92,93</sup>. This may be due to disruption in normal nutrient cycling and physiological processes. These disturbances include: (a) the inhibition of formation of certain polypeptides, which are perhaps essential for cold tolerance in Pinus sylvestris needles94, (b) imbalances in amino acid concentration and metabolism and in general protein biosynthesis 95,96 and (c) change in carbon allocation pattern<sup>97</sup>.

However, the hypotheses other than the nitrogen saturation hypothesis of forest decline may be equally important (cf. Figure 3), as the contribution of atmospheric nitrogen to total nitrogen content of trees seems to be of minor significance, in some cases. For example, in *Picea rubens* seedlings sprayed with NH<sub>4</sub>NO<sub>3</sub> containing cloud water, only 1.5% of the seedling nitrogen is derived from the cloud water<sup>98</sup>. Similarly, Hanson et al.<sup>54</sup> by extrapolating the data from laboratory experiments estimated that only about 3% of the nitrogen requirements of natural forests could be fulfilled from prevailing NO<sub>2</sub> levels in the atmosphere.

#### **Conclusions**

The following general conclusions about the role of nitrogen air pollutants may be drawn: 1. All nitrogenous air pollutants are absorbed and assimilated by the plants. 2. In some cases, low levels of pollutants increase plant growth, but the growth with the pollutant as a sole source of nitrogen is not comparable to that with nutrient nitrogen. 3. Higher dosage of pollutants generally induces visible and/or physiological disorders, which are perhaps the most important limitations in optimum utilization of air pollutants as

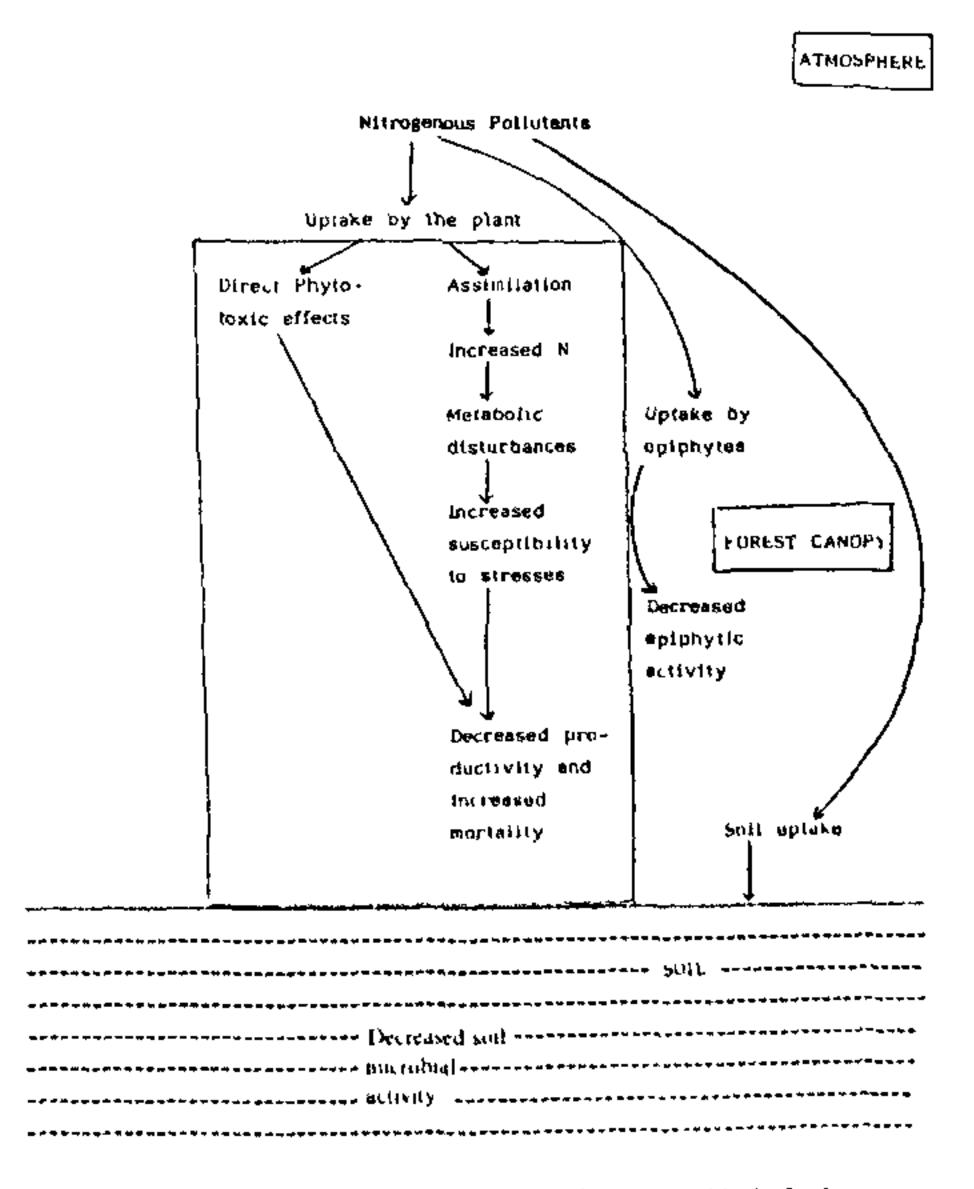


Figure 3. Diagrammatic representation of the possible links between nitrogenous pollutants and forest decline.

nitrogen source. 4. Assimilation of nitrogenous air pollutants by the trees may be a factor in natural forest decline.

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#### RESEARCH ARTICLES

## Close packing and pressure-induced amorphization

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Recently a large number of crystalline compounds have been discovered to amorphize at high pressures. However, the molecular origin of this transformation is not understood. In this article, we show that this is a consequence of a competition between the close-packing and long range order—a principle well known in organic crystallography. We show that the tendency of these systems to acquire higher coordination under compression is sterically hindered at some pressure due to the strong nature of inter-molecular bonding of building blocks. This hindrance drives the system to sacrifice the translational order to achieve better packing. The application of principle is shown through the examples of Ca(OH)<sub>2</sub>, SiO<sub>2</sub>, ice, LiKSO<sub>4</sub>, etc.

From the analysis of the packings of organic molecules, Kitaigorodskii<sup>1</sup> found that 'if the shape of the molecules is such that the formation of a long-range order leads to a larger percentage of empty space than in the absence of such an order in a close-packing arrangement, then no

crystal is obtained'. We show in this article that this principle is also very apt for understanding the mechanism of pressure-induced amorphization of materials. Starting with the first report of amorphization of ice<sup>2</sup> in 1984, presently about 20 substances are known to undergo this crystal-amorphous (c→a) transition under pressure (see Table 1 of Sikka<sup>3</sup>). However, its driving mechanism is still not clearly understood. In our view, the following scenario represents the physical reality. Application of pressure promotes close packing but the shape of the basic building block in the crystal may not favour this. Further compression may result in steric hindrances and ultimately destabilize the parent crystal structure. Formation of a new crystal structure would require a change in the shape of the basic packing unit, e.g. coordination change or dissociation. This step will involve bond breaking/bond making and diffusion of some atoms that may be inhibited at the low temperatures of the high pressure experiments. The