SEED AND SEEDLING TOLERANCE OF CEREAL, OILSEED, FIBRE AND LEGUME CROPS TO INJURY FROM BANDED AMMONIUM FERTILIZERS.

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Faculty of Environmental Sciences

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DECLARATION OF ORIGINALITY

This experimentation and analysis, interpretation and presentation of the results is the original work of the author and has not been submitted for any previous degree or diploma in any University. To the best of my knowledge and belief, this thesis contains no material previously published or written by any other person except where due reference is made in the thesis itself.

Signature:

Christopher W. Dowling
ABSTRACT

Processes causing crop establishment damage from ammonium (NH\textsubscript{4}\textsuperscript{+}) fertilizer placed in close proximity to seed at sowing are generally poorly understood within farming communities of eastern Australia. Currently, the information used to assess establishment hazard includes nitrogen (N) tolerance for a limited range of crop species. Other factors include the N concentration of fertilizer products, with adjustment of the rate expected to be tolerated on the basis of soil moisture and application equipment.

Current recommendations were adapted from North American research spanning a period from the start of the century until the 1990s and some recent Australian research (1960s to 1980s) for a limited range of crops and fertilizer types. The incidence of seedling damage from N fertilizer and fertilizer containing other nutrients appears to have grown in recent years. This may be attributed to use of a wider range of NH\textsubscript{4}\textsuperscript{+} and other fertilizers, a trend for at-sowing application in zero-tillage and expansion of areas of declining soil fertility, particularly N fertility. Other factors include the sowing of new crops with greater fertilizer sensitivity, low tolerance to establishment loss for high value genetically modified seed and modern designs of sowing and application equipment.

The major objective of this research was to investigate ammonia (NH\textsubscript{3}) tolerance of 10 crop species of importance for eastern Australian cropping systems (maize, *Zea mays* L.; cotton, *Gossypium hirsutum* L.; wheat, *Triticum aestivum* L.; barley, *Hordeum vulgare* L.; chickpea, *Cicer arietinum* L.; sorghum, *Sorghum bicolor* (L.) Moench; canary, *Phalaris canariensis* L.; canola, *Brassica napus* L.; panicum, *Setaria italica* L. and sunflower, *Helianthus annuus* L.). Experiments were designed to highlight differences among crops in NH\textsubscript{3} toxicity and osmotic damage potential for commonly used NH\textsubscript{4}\textsuperscript{+} fertilizers. Various strategies were then tested to maintain plant populations within commercially acceptable ranges when affected by NH\textsubscript{3} toxicity and/or high osmotic pressure.
Tolerance of seeds to NH$_3$ toxicity was evaluated in the field and for atmospheric exposure. Response of various crop species to atmospheric-NH$_3$ exposure showed that certain species responded differently in their germination, coleoptile growth and radicle growth in a closed system. Using these 3 parameters as indices of crop response to NH$_3$ toxicity revealed different ranking for some species; the same species showed a different critical NH$_4^+$ concentration for each parameter. Exposing seeds above 200 x 10$^{-4}$ M NH$_4$OH for 72 h was sufficient to significantly reduce or inhibit germination of all 10 species tested. Seed of most species were unaffected by exposure above 20 x 10$^{-4}$ M NH$_4$OH. Species rank, combining tolerance for germination, coleoptile growth and radicle growth was established to relate to likely performance in the field. Decreasing order of tolerance for monocot species was: maize > sorghum > wheat = barley > panicum > canary and for dicot species chickpeas > cotton > sunflowers > canola.

A range of physical and chemical seed characteristics was correlated with NH$_3$ tolerance to investigate tolerance mechanisms. For monocot species, tolerance was related to the seed surface area/volume ratio suggesting that diffusion resistance was an important parameter whereas for dicot species N concentration of seed was negatively correlated with tolerance.

In field experiments where NH$_4^+$-fertilizers were placed with seeds, difference between species in their tolerance of atmospheric-NH$_3$ was insufficient to describe effects of NH$_4^+$-fertilizers on crop emergence. Crop species fell into 3 response categories; high (maize, sorghum, barley and wheat), medium (cotton, canary and sunflower) or low (canola, chickpea and panicum) tolerance to soil NH$_4^+$-N. Soil NH$_4^+$-N concentrations tolerated by the medium and low tolerance group was 50 % and 15 to 25 % respectively, that of the high tolerance group. Generally, NH$_3$ tolerance response for species such as wheat, barley and sorghum was found similar to current recommendations for urea (~0.5 g/m N as urea) but there were significantly different responses to NH$_4^+$-N from different NH$_4^+$-fertilizer products, that are not recognised in current recommendations.
Crop species were ranked for sensitivity to mono-ammonium phosphate (MAP), di-ammonium phosphate (DAP), triple superphosphate (TSP), urea and ammonium nitrate, and categorised according to the fertilizer rate at which significant establishment damage occurred. Ranking of crop species for NH$_3$ toxicity was generally similar across experiments but the NH$_4^+$-N rate tolerated varied with experimental conditions. Urea and DAP caused larger reductions in establishment than equivalent NH$_4^+$-N rates from MAP or ammonium nitrate. The “safe” rate for ammonium nitrate (1 g/m NH$_4^+$-N) was approximately twice that of urea at equivalent NH$_4^+$-N rates. Usually between 20 and 30 % more NH$_4^+$-N was tolerated for MAP than for DAP.

In the absence of NH$_3$ toxicity, osmotic effects of fertilizer products delayed and occasionally inhibited germination. There was significant difference among species in osmotic tolerance; cotton, maize and sorghum (< -0.3 MPa) were more tolerant than sunflower or soybean (> -0.2 MPa).

Strategies to improve crop establishment in the presence of NH$_4^+$ fertilizer such as increasing seeding rate, adding water to the seed furrow, changing fertilizer N source and chemically modifying hydrolysis of urea were identified and tested. For low to moderate rates of seed placed NH$_4^+$-N, increasing barley seeding rate from 25 to 40 kg/ha was found to be successful strategy to maintain establishment when urea rate was increased from 1.1 to 2.3 g/m of seed row. Changing the fertilizer N source and modifying hydrolysis of urea were successful in lowering soil NH$_4^+$ around the seed and thus reducing establishment losses, but osmotic effects also limit the maximum fertilizer application rates. The added cost of these strategies may prevent their widespread adoption.

Complex interactions between crop species, fertilizer product, soil texture and moisture, and application equipment highlighted by the results of these experiments, suggest that simple decision tools are insufficient to provide fertilizer recommendations that meet the demands of modern agriculture. A computer based decision support programme, Fertsafe, was developed
during this study from experiments conducted and papers reviewed, to provide “safe rate” recommendations to apply fertilizer at sowing for a range of crop species, fertilizers, soils and sowing conditions of eastern Australia. Changes to fertilizer application equipment, other crops and fertilizer products will require ongoing research continuously improve and update this decision support tool.
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PUBLICATIONS FROM THIS RESEARCH

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Chapter 1

Introduction

Australian farmers have applied fertilizer, particularly phosphates in cropping and pasture programs relatively early in the development of agricultural production systems (Donald 1965). Many soils, with the exception of the black Vertosols and some grey Vertosols are low in cations, P and organic matter. N fertility was dependent on the original native vegetation and soil type (Cook and Scott 1987; Dalal and Mayer 1990).

In the northeastern Australian cereal belt, most soils with high native N levels have been degraded by exploitative cereal cropping practices from the time they were first cultivated (Dalal and Mayer 1986). In contrast to southern temperate climatic regions, where N levels in poorer soils were increased, and then maintained, by the inclusion of a period of legume pasture ley (Donald 1965), there has been a noticeable absence of pasture leys in northern cropping systems.

As the native fertility of the soils of the region has declined (Graham et al. 1981; Dalal and Mayer 1986) and cropping intensity has increased, the importance of N fertilizer in maintaining productivity has grown (Cook and Scott 1987). Nitrogen fertilizer is commonly the largest variable cash cost in many cropping systems. It has become a key input for profitability in a range of crops and cropping systems due to the run-down in native soil fertility (Dalal and Mayer 1986). Associated with the increased recognition of the need for N fertilizer has been the emergence of ammonium phosphates to replace superphosphate usually applied at sowing.

Climatically, northern Australia presents some unique problems for cropping. Wollin et al. (1987) and Hammer and Munchow (1990) found that a narrow planting window caused by the unreliability of late autumn and early winter rain, low spring rainfall for winter cereal cropping and high reliance on fallow stored
moisture for both winter and summer crops significantly influenced crop management. Unique combinations of climate, soils and crop species have influenced Australian farmers and farm machinery suppliers to modify imported fertilizing application, cultivating and sowing techniques or have invented their own, to maintain relevance to local soil management needs and viable farming enterprises (Wollin et al. 1987; Thomas et al. 1998).

Nitrogen fertilizers have been used for about 30 years on a broad scale in Australia (Pulsford 1978). During that time farming practices changed dramatically, from intensive tillage and bare fallowing to reduced tillage or zero tillage. Change from mechanical to herbicide-based weed control and seed-bed preparation, and subsequent effects on soil tilth, has forced N fertilizer application rates to increase and application methods to change to meet the new soil conditions and higher yield potential (Cook and Scott 1987; Wylie 1997).

In the earliest established N fertilizer use area of eastern Australia, the Darling Downs of Queensland, N fertilizer was originally applied 6 to 8 weeks pre-sowing during mechanical seedbed preparation (Wollin et al. 1987; Incitec Fertilizers 1994). This operation was convenient, enabling farmers to apply N fertilizer during pre-sowing tillage for weed control thereby avoiding crop establishment problems arising from ammonia (NH₃) forming fertilizers such as urea and anhydrous ammonia being placed too close to germinating seed. Reduced germination resulting from these and other fertilizer materials such as potassium chloride (KCl), sodium nitrate, ammonium sulfate and DAP placed in close proximity to seed was recognised in the early 1900s in the USA (Hicks 1900; Sherwin 1923; Truog et al. 1925; Rost 1930; Willis and Piland 1931).

 Adoption of reduced tillage farming has also reduced the opportunity for pre-sowing application of N fertilizer using high-disturbance cultivation equipment. The reduction in mechanical tillage early in a fallow phase created delays in N fertilizer application, moving the application closer to sowing. There has also been an increasing trend for N fertilizer to be applied at sowing (Ward 1987; Robotham 1993). More recently, the availability of genetically modified crop
species and associated increase in seed cost (x 3-5) have intensified the need to reduce seeding rate and keep seedling loss to a minimum.

Two major pathways by which fertilizer may affect germination and early growth are through toxicity concentrations of NH$_3$ in the soil atmosphere and soil solution, and increased osmotic pressure in the soil solution, one or both of which may damage germinating seeds (Cook and Scott 1987; Ward 1987). It was not until after 1950 that significant effort was put into investigating the specific role NH$_3$ in the soil atmosphere played in reducing germination. Since that time, studies by Duisberg and Buehrer (1954), Brage et al. (1960), Allred and Ohlrogge (1964), Blanchar (1967), Bennett and Adams (1970), Colliver and Welch (1970) and Woodstock and Tsao (1986), Bremner and Krogmeier (1989) have all measured the effect of NH$_3$ on germination and establishment.

Most studies of soil atmosphere NH$_3$ effects on germination have been restricted to maize (Zea mays L.), with a smaller number of studies including wheat (Triticum aestivum L.), barley (Hordeum vulgare L.), soybean (Glycine max (L.) Merr.) and peanut (Arachis hypogaea L.). It is difficult to discern the relative NH$_3$ tolerance of these species from the findings of the above authors as most experiments were conducted with only 1 crop species or a single NH$_3$ rate. Woodstock and Tsao (1986) compared soybean, maize and peanut in a soil-less system and found that maize was killed by a lower (x10) concentration of atmospheric-NH$_3$ than soybean. This is contrary to field results of Gerwing et al. (1994) who found that soybean was less tolerant than maize of seed-row placement of a range of ammonium (NH$_4^+$) fertilizers.

The osmotic pressure effect on seed germination has been characterised for several crop and pasture species in the experiments of Dubetz et al. (1959), Redmann (1974) and Young et al. (1983) mostly conducted with osmotic solutions in artificial media. Little tactical fertilizer management information about crop species response to fertilizer derived osmotic pressure from commonly used products such as KCl, and triple superphosphate (TSP), applied in the seed furrow, is available.
The majority of recent field studies of fertilizer effects on crop establishment have been conducted in North America and appear as specific recommendations for localised crops, soil, climate and fertilizer products (Gerwing et al. 1995; Roberts and Harapiak 1997). The findings of Roberts and Harapiak, (1997) have provided a more universal approach for recommending fertilizer in the seed-furrow for a range of winter cereal and oilseed crops. This approach includes modification of the maximum rate for seed-row fertilizer application based on row spacing, within row dispersion of fertilizer by the application tine and soil moisture content.

Experiments by Carter (1967), Mason (1971), Scott et al. (1987), and Scott (1989) have quantified the germination and establishment effects for a limited range of fertilizer products and a narrow range of crop and pasture species and Australian soils. There have been few research reports of effects of fertilizer on crop germination, emergence and establishment for Australian conditions, published since the late 1980s. There are conspicuously few reports on fertilizer effects on summer cereals, summer oilseeds and legumes and the performance of newer fertilizers such as ammonium phosphates. The impact of modern fertilizer application techniques such as application of anhydrous ammonia at sowing and split-boot fertilizer placement on crop emergence are even less well defined.

Because of this paucity of information of this type the potential for fertilizer damage to crop establishment has increased for farmers in eastern Australia. The only widely used benchmark by which the hazard of seed-row placement of fertilizer is assessed is by the N concentration of the fertilizer, disregarding the N form or fertilizer product type (Mills et al. 1996; Mills and McIntyre 1997). In the northeastern Australia cereal belt current recommendations for N fertilizer applied in the seed furrow is a maximum of 25 to 30 kg/ha N placed with the seed in 180 to 250 mm row spacings for most crop species (Ward 1987). In southern Australia 15 to 20 kg/ha N is the maximum rate recommended for cereal crops.
In this thesis, there is frequent use of the terms germination, emergence and establishment. The following definitions describe the use of these terms in this thesis as defined by Collis-George (1987). Germination is the stage of seedling development when active growth first becomes evident. This is usually defined as when the radicle extends 2 to 3 mm outside the seed testa. Emergence is the stage of seedling growth when the seedling emerges through the soil surface. Crop establishment is reached when emerged seedlings could be expected to grow right through to crop maturity with favourable growing conditions (Wood 1987).

Given apparent differences in the tolerance of crops to soil atmospheric NH$_3$ and soil solution osmotic pressure differences within NH$_4^+$-fertilizer products, there is clearly a need to gain a better understanding of the principles of how fertilizers affect crop germination, emergence and establishment. Ward (1987) also highlighted this in a review of the subject. This may be achieved through gaining an understanding of the toxic effects of soil atmospheric NH$_3$ and role of osmotic pressure, from banded NH$_4^+$-fertilizer, on a range of economically important crop species using research techniques that do not severely limit wider interpretation of the results. Avoidance of crop establishment damage depends on developing strategies and decision support information that modifies or helps avoid unfavourable crop establishment conditions created from inappropriate placement of NH$_4^+$-fertilizers.
Chapter 2

Review of Literature

Nitrogen fertilizer is a vital component of many modern cropping systems. Nitrogen use has gained importance in an attempt to maintain or improve crop productivity where soil N fertility is inherently low or where higher N fertility has been depleted by exploitative farming practices.

Application of N fertilizer in the seed-furrow at sowing is a relatively new practice, developed in response to reduced opportunity for application pre (reduced tillage) and post-sowing. Other factor that have also influenced this trend has been a change to fertilizer products used at sowing such as increased use of MAP and DAP in preference to superphosphate, and the further development of combined sowing and fertilizing equipment.

The effect of seed-placed fertilizer on the success of crop establishment depends on a complex interaction of factors that are classified broadly into 5 groups; external environment, application equipment, soil, crop species and fertilizer characteristics.

2.1 FACTORS AFFECTING CROP ESTABLISHMENT WHEN SEED IS SOWN IN CONTACT WITH FERTILIZER

External Environment Factors
Air temperature, relative humidity and wind speed are the most important weather related variables that can affect seed germination and crop establishment in the time from sowing to seedling emergence (Weaich et al. 1996). These climatic variables are closely connected to the germination process through a common factor, soil drying rate. Other factors such as starting soil moisture, soil disturbance level caused by the soil engaging equipment and effectiveness of repacking the seed furrow also modify the crop emergence response to soil drying rate (Ward 1987).
Rate of soil drying is as important as the initial soil moisture during the germination and establishment phase. In the absence of high temperature conditions, emergence failure can result from high soil strength produced by rapid drying resulting from air movement. Soil drying rate is lower at higher relative humidity and the rate of increase in soil strength is reduced (Wood 1987; Weaich et al. 1992; Weaich et al. 1996).

Direct effects of high air temperatures that are sub-optimal for germination are expressed through effects on soil temperature. Emergence failure under sub-optimal high soil temperature conditions can result from low metabolic activity that reduce the rate and extent of coleoptile elongation (Radford et al. 1989; Weaich et al. 1996).

Soil stubble cover also moderates soil temperature maintaining the temperature closer to the optimum promoting more rapid growth. Weaich et al. (1996), in a computer simulation, showed that soil with 2400 kg/ha equivalent stubble cover had a higher minimum temperature, a lower maximum soil temperature and a 69% reduction in accumulated evaporation by the application of mulch. These conditions provided by the mulch were more conducive to low soil strength and higher rates of germination and seedling growth.

**Application Equipment**
Application equipment can have a large effect on crop establishment. Equipment may interact strongly with soil conditions that affect soil drying rate.

Configuration of opening and closing devices that place seed and fertilizers in the soil are frequently matched to soil characteristics to enable maximum seed emergence and vigour (Wollin et al. 1987). The configuration of equipment is frequently less than optimal for the establishment of crops in the presence of fertilizer. Concentrations of toxic ions or osmotic pressure of the dissolved fertilizer in the seed-furrow, or both (Richards 1979) cause fertilizer damage to crop establishment. Fertilizer concentrations in the seed zone are greatly influenced by the amount of soil disturbance and mixing produced by the soil
opening device during the seeding operation (Ward 1987; Roberts and Harapiak 1997).

Soil recompression in the area disturbed by the soil-opening device has been shown to increase crop establishment. Recompression of the soil by implements such as press-wheels reduce soil covering depth (Radford et al. 1989), reduce the soil porosity and increase the seed/soil contact area. In situations where depth of soil cover is a critical factor in successful establishment, the recompression provided by press-wheel or field rollers may be a critical factor for success. Radford et al. (1989) showed that the depth of soil cover was important to the establishment of wheat with the semi-dwarf growth habit. High temperatures (early sowing), the presence of urea and some seed treatment fungicides reduced coleoptile elongation and establishment. Control over sowing depth is therefore an important factor for successful crop establishment where coleoptile length is reduced by fertilizer application in the seed row.

Generally a reduction in porosity, achieved by recompression of the soil helps reduce the soil drying rate in the seed zone by reducing movement of soil water through evaporative processes (Weaich et al. 1996). When fertilizer is applied in the seed row, the reduction in porosity may be beneficial or detrimental depending on the advantages of restricting movement or liberating gaseous NH₃ from the seed row.

Seed-soil contact area together with the soil water potential governs the ability of the seed to reach its critical water content for germination (Wood 1987; Bouaziz and Bruckler 1989). Seed-soil contact depends on both soil structure and soil water content. Thus the effect of soil recompression is dependant on both the soil structure and soil water content (Ward 1987).

**Soil Factors**

The soil water content plays a pivotal part in the seed germination and establishment processes, and has a significant impact on the fertilizer-seed interaction.
The gross osmotic effect of a fertilizer in the soil solution can be described in terms of the equation of Young et al. (1983):

\[ \psi (\text{MPa}) = \frac{\nu RT W_A \phi m}{100V_A} \]

where:
- \( \nu \) = number of moles of ions that can be ionised from one mole of salt
- \( R \) = Universal gas constant
- \( T \) = temperature (K)
- \( W_A \) = molecular weight of the solvent
- \( V_A \) = partial molal volume of the solvent
- \( m \) = molality of the solution
- \( \phi \) = molal osmotic coefficient

For a given rate of fertilizer, the osmotic pressure is therefore inversely related to the volume of water in the soil.

Adsorption and precipitation reactions account for the difference in osmotic pressure between a pure solution of the fertilizer salt and the osmotic pressure in soil solution (Rader et al. 1943).

Soil water content can decrease or increase damage from NH\(_3\) toxicity during seed germination, depending on whether it provides sufficient hydrogen ions to force the NH\(_3\)-NH\(_4^+\) equilibrium to a lower, less toxic soil atmospheric NH\(_3\) concentration. Similarly changes to soil water content creates changes in soil porosity, permeability and solution concentration of NH\(_3\), thereby creating a higher or lower NH\(_3\) diffusion potential. Mahler et al. (1989) found significant interactions between soil matric potential, N rate and N source and the emergence of winter wheat. Lower matric potential generally increases N fertilizer damage, reducing crop emergence.
Soil physical characteristics such as texture and tilth are key factors in the movement of soil water and gases, and adsorption processes associated with movement of fertilizer away from application sites. As the surface area for adsorption increases in response to texture and/or tilth, diffusion of gases and liquid is reduced. Reduced diffusion rate and retention area are a function of increased reactive surface area, reduced soil porosity and increased soil tortuosity (Marschner 1995a).

Both texture and tilth can also have significant effects on the seed germination process though interactions with moisture holding capacity and surface area contact with the seed. Texture and tilth characteristics, that restrict movement of fertilizer salts and gases, tend to favour seed germination conditions. Depending on seed-fertilizer separation, these factors can be complimentary or detrimental. On the other hand they may increase seed-soil contact and help exclude fertilizer salts from the seed row. Alternatively they may restrict the movement of the fertilizer away from the seed row (Papendick and Parr 1966b), depending on fertilizer placement geometry.

The rate at which an ion moves through soil is defined as the effective diffusion coefficient (Dₑ). The effective diffusion coefficient is a function of the diffusion coefficient of an ion in water, water content of soil, an impedance factor related to soil texture and tilth (tortuosity), and the reciprocal of the buffering capacity of the soil for a particular ion (Nye and Tinker 1977). Izzauralde et al. (1990) found that soil titratable acidity, a measure of the NH₃ buffering capacity, was a significant factor in predicting the movement of NH₃ from an application band. The rate of movement of the fertilizer ions is therefore dependent on both physical and chemical characteristics of the soil.

Soil pH influences the relative availability of nutrients and the nutrient ion species balance. The cation species balance is important with NH₄⁺-fertilizers as it controls the NH₃-NH₄⁺ balance, hence the toxicity. Soil pH has the greatest effect at the extremities of an application band and across time because in most application bands in the short term, the pH is dominated by the pH of the product applied (Whitehouse and Leslie 1973).
Soil background salt content may be an influencing factor in germination response to fertilizer salts applied with the seed at sowing. Soils with a high salt load add extra osmotic pressure to the fertilizer band.

Activity of the soil biomass is both responsible for the release of NH$_4^+$ ions from amino-compounds and conversion of NH$_4^+$ to N oxides. Where the NH$_3$ source is urea, urease activity of the soil may influence the response of the seed to the fertilizer. In particular, delays to hydrolysis or reducing NH$_3$ evolution rate till after germination and early seedling growth, may control the damaging effects of NH$_3$ release from urea (Bremner 1995). Transformation of urea to NH$_3$-NH$_4^+$ in the soils of eastern Australia is rapid in moist soil. Campbell et al. (1984) measured urease activities in the range 14 - 28 µg N/g/h for soils of southeast Queensland.

Nitrifier activity controls the rate of reduction of NH$_3$-NH$_4^+$ concentration in a fertilizer application band. The balance between rate of release of NH$_3$ from the fertilizer product and rate of N mineralisation determines the net NH$_3$-NH$_4^+$ concentration at the application site.

Nitrite is an intermediate oxidation product in the pathway from NH$_4^+$ to NO$_3^-$ that is toxic to plants at a relatively low soil concentration (Duisberg and Buehrer 1954). Nitrite accumulation is favoured where pH is above 7.7, due to inhibition of *Nitrobacter*, the bacteria responsible for the conversion of NO$_2^-$ to NO$_3^-$, *Nitrosomonas* bacteria, responsible for the conversion of NH$_4^+$ to NO$_2^-$, have an optimum pH of 8.2, but is still active at 9.0.

**Plant Factors**

Water potential of seed is a key factor in germination of the seed. Imbibition, passive uptake of water at the beginning of germination, is governed by the seed-soil water potential difference (Bouaziz and Bruckler 1989)

Seed germination for some species is either partially or totally restricted at water potentials more negative than wilting point but most species have specific requirement, with respect to moisture availability (Table 2.1).
Initial water potential of a seed can be extremely low, -100 MPa, owing to an enormous matric potential. At the time of visible germination, water potential is less than -1 MPa for all species (Bewsley and Black 1978).

According to Bewsley and Black (1978), germination can be divided into 3 distinct phases: Phase 1 is the imbibition phase that begins with rehydration of enzymes and substrates, initiating some metabolic events to commence soon afterwards. Phase 2 is the lag phase when major metabolic change occurs prior to germination. Seeds may be held in phase 2 by drying or exerting osmotic pressure but will germinate readily if rehydrated.

Table 2.1  Seed water potential for the germination of some crop and pasture species.

<table>
<thead>
<tr>
<th>Species</th>
<th>Seed Water Potential (MPa)</th>
</tr>
</thead>
<tbody>
<tr>
<td>maize</td>
<td>-1.23</td>
</tr>
<tr>
<td>soybean</td>
<td>-0.65</td>
</tr>
<tr>
<td>sugarbeet</td>
<td>-0.35</td>
</tr>
<tr>
<td>subterranean clover</td>
<td>-1.16</td>
</tr>
<tr>
<td>rapeseed</td>
<td>-0.3</td>
</tr>
<tr>
<td>sunflower</td>
<td>-1.3</td>
</tr>
<tr>
<td>sorghum</td>
<td>-0.7</td>
</tr>
<tr>
<td>rice</td>
<td>-0.78</td>
</tr>
</tbody>
</table>

( Hunter and Erikson 1952; Williams and Shaykewich 1971; Young et al. 1983)

Phase 3 is the cell expansion and radicle elongation phase. During this phase water uptake increases due to a decrease in water potential related to an unknown process in germination (Bewsley and Black 1978). This is followed by a decrease in osmotic potential due to post-germination reserve hydrolysis.

It is unlikely that the osmotic pressure exerted by soil within a fertilizer band is high enough to inhibit water uptake during Phase 1. Williams and Shaykewich (1971) found that during imbibition seeds could have matric potentials of -100 MPa. Fertilizer damage is more likely to result from stasis in Phase 2, causing temporary delay in water uptake, which slows, or stops the germination process.
If soil dries rapidly during Phase 2 re-wetting is unlikely to produce satisfactory germination due to the damage done to the enzymes. If the osmotic challenge occurs as a result of rapid drying during Phase 3, then germination is likely to fail (Bewsley and Black 1978).

The effect of the osmotic competition between seed and fertilizer is impacted by soil drying rate that is mediated by an increase in soil solution concentration and its osmotic pressure.

Success of crop establishment after the completion of the initial biochemical activation within the seed is dependent on the coleoptile and radicle elongation (Cummins and Parks 1961). Ward (1987) and Radford et al. (1989) have noted that there are significant reductions in length of coleoptiles among wheat varieties with the Mexican semi-dwarf gene in response to urea and a fungicide applied with seed. High soil temperature and low water potential increased susceptibility to these urea and fungicide effects.

Once roots develop, the osmotic component of the soil moisture potential become more important. Thus increasing salinity will inhibit water uptake by roots reducing shoot extension rate and total establishment (Wood 1987).

Crop species have different minimum seed and soil water requirements for germination. If the soil water remains below the minimum required for germination, the seed will eventually be damaged or destroyed by soil fungi (Helms et al. 1996a).

Various crop species exhibit widely differing degrees of salt tolerance at early growth stages. Varieties within species may also exhibit wide variation in ability to tolerate saline conditions (George and Williams 1964). The species by osmotic pressure interaction can be further modified by temperature. Helms et al. (1996b) showed that for soybean as the temperature increased in the presence of water stress, germination was reduced. Increasing temperature reduced coleoptile length and reduced emergence alone and in combination with seed–furrow applied urea (Radford et al. 1989).
Radicle extension rate determines the ability of germinating seed to access water and nutrient ahead of the soil drying front. Moody et al. (1995a, 1995b) found that soybean root elongation rate was reduced by 10% at a soil solution electrical conductivity of 4.1 dS/m. It was also demonstrated that an NH₄⁺ salt induced calcium deficiency was a primary cause of restricted root growth rather than osmotic effects or NH₃ toxicity for some acidic NH₄⁺-fertilizer products. NH₃(aq.) toxicity reduced radicle elongation rate for soybean where the NH₄⁺-fertilizer product in the soil solution was alkaline.

**Fertilizer Factors**

The osmotic effect of a fertilizer salt on seed is primarily related to the chemistry of the salt and the solution concentration of the salt (Fig. 2.3). Secondary adsorption, precipitation and diffusion reactions modify the effect of the salt. These reactions are in turn related to the interaction between the chemistry of soil and fertilizer salt (Moody 1995a, 1995b, 1995c).

Ammonia potential is a term defined in this study, to describe the potential for fertilizer products containing NH₄⁺ to cause metabolic damage during germination, root or shoot growth. The NH₃ potential of a fertilizer product was developed considering its NH₄⁺ concentration, solution pH, solubility and counter-ion accompanying NH₄⁺. Products such as urea and DAP, that produce a high NH₄⁺ concentration, have high solution pH (Richards 1979; Bremner and Krogmeier 1989; Fan and MacKenzie 1993; Fan and MacKenzie 1995; Moody et al. 1995b) and high solubility, pose the greatest risk to the germinating seed. The risk to crop establishment posed by N fertilizers having high NH₃ potential was also confirmed by Mason (1971) and Mahler et al. (1989). They found that urea was more damaging to crop emergence than ammonium nitrate for a range of crop species. Pairintra et al. (1973) found DAP more damaging than MAP to wheat establishment. In laboratory studies Pairintra et al. (1973) also found that NH₃ volatilization was 300% greater from DAP than from MAP.
Fig. 2.1 Pathways of nutrient toxicity to germinating seeds.

Fertilizers such as MAP and DAP with relatively low counter-ion (phosphate) mobility may also have low NH$_4^+$ mobility (Moody et al. 1995b) resulting in higher concentrations of NH$_4^+$ in the fertilizer application band.

Fertilizer application rate by itself is a poor predictor of potential germination damage. Damage is more closely related to fertilizer concentration in the seed zone and therefore is affected by application rate and the volume of soil through which fertilizer is mixed (Roberts and Harapiak 1997).

The size of a fertilizer reaction zone is initially related to the physical distribution of fertilizer that is determined by application equipment and soil conditions (Anon 1995). Further movement of the nutrient ions occurs by mass flow and diffusive migration of anions and cations away from the fertilizer particles.

Seedbed utilisation (SBU) (Roberts and Harapiak 1997) is a term that describes the physical spread of fertilizer bands and is an integration of application parameters. SBU is calculated as soil area over which fertilizer is spread as a proportion of the total soil area. A high SBU indicates a more dilute fertilizer
band for a given application rate i.e. greater dispersion. SBU is also affected by forward speed of the application implement, width of soil opening devices and delivery tube, force with which product is propelled through the application tube into the soil, and row spacing of application tines.

Manipulation of granule size has been suggested as a possible method of reducing effects of fertilizer on crop establishment (Robotham 1993; Bremner 1995). The 1.5 to 4 mm size range of existing fertilizer products appears to be too narrow to observe these effects.

**Factors Affecting Fertilizer Ion Movement**

Many of the soil factors are linked, directly or indirectly, through their effect on the diffusion process. The basis for mobility of an ion is its diffusion coefficient. In aerated soil, ions diffuse only in pore spaces filled with water and the ions in solution may interact with the soil solid phase through adsorption and precipitation processes. The effective diffusion coefficient of an ion in soil \( D_e \) is distinct from the diffusion coefficient in water \( D_i \) due to of the modifying effects of the soil’s physical and chemical characteristics (Marschner 1995a).

\[
D_e = D_i \cdot \theta \cdot f \cdot \frac{dC_i}{C_s}
\]

- \( D_e \) = effective diffusion
- \( D_i \) = diffusion coefficient in water
- \( \theta \) = volumetric water content of soil
- \( f \) = impedance (tortuosity) factor
- \( \frac{dC_i}{C_s} \) = reciprocal of the buffer capacity for the ion

In free solution all the simple cations and anions of interest in plant nutrition and the simple molecules of molecular weight less than 200, have diffusion coefficients in the range of \( 0.5 - 2 \times 10^{-9} \) m²/s at 25 °C (Nye 1979). Ammonia, with a diffusion coefficient of \( 1.15 \times 10^{-9} \) m²/s, falls about the middle of the range quoted by Nye (1979). The magnitude of the soil effects in modifying diffusion rates of the different ions is illustrated by Marschner (1995a). He
quotes the average diffusion coefficients in soil for NO$_3^-$, potassium and dihydrogen phosphate ions to be 5 x 10$^{-11}$, 5 x 10$^{-12}$ and 1 x 10$^{-13}$ m$^2$/s respectively.

Adsorption and precipitation are principal mechanisms restricting the diffusive migration of ions away from a fertilizer application band. Soil factors central to the adsorption process are cation exchange capacity (CEC), pH and base saturation. Soil water content, pH and solubility products of dominant anion and cation species (Tisdale and Nelson 1975) govern precipitation (Barber 1995).

2.2 DIRECT EFFECTS OF AMMONIA AND OSMOTIC PRESSURE ON PLANT ROOTS AND SEED GERMINATION.

Occurrence of Non Ionised Ammonia in the Soil

Studies by Du Plessis and Kroontje (1964) showed a linear relationship between NH$_3$ loss from the soil and predicted loss based on calculations of the equilibrium of the NH$_3$ + H$_2$O $\leftrightarrow$ NH$_4^+$ + OH$^-$ system. In an unbuffered system, the equilibrium of the reaction lies strongly to the left as the pH rises, being dominated by NH$_3$ in solution (NH$_3$(aq.)). In the soil equilibrium exists between adsorbed NH$_4^+$, NH$_4^+$ in solution, NH$_3$ vapour (NH$_3$(g.)) and NH$_3$(aq.).

Higher pH associated with an NH$_3$ application zone in the soil is likely to favour the NH$_3$(g.) vapour and NH$_3$(aq.). Table 2.2 shows the relative proportions of ionised NH$_4^+$ and non-ionised NH$_3$ in an unbuffered system at 25 °C calculated using the equation:

$$\text{pH} = \text{pK}_a + \log \frac{\text{NH}_4^+}{\text{NH}_3}$$
Table 2.2 Relative proportions of NH$_3$(aq.) and NH$_4^+$ in an unbuffered NH$_4$OH solution with pKa 9.4 (Freney et al. 1983).

<table>
<thead>
<tr>
<th>pH</th>
<th>NH$_3$(aq.)</th>
<th>NH$_4^+$</th>
</tr>
</thead>
<tbody>
<tr>
<td>6</td>
<td>0.04</td>
<td>99.96</td>
</tr>
<tr>
<td>7</td>
<td>0.4</td>
<td>99.6</td>
</tr>
<tr>
<td>7.5</td>
<td>1.2</td>
<td>98.8</td>
</tr>
<tr>
<td>8</td>
<td>3.8</td>
<td>96.2</td>
</tr>
<tr>
<td>8.5</td>
<td>11.1</td>
<td>88.9</td>
</tr>
<tr>
<td>9</td>
<td>28.6</td>
<td>71.4</td>
</tr>
<tr>
<td>9.5</td>
<td>55.6</td>
<td>44.4</td>
</tr>
<tr>
<td>10</td>
<td>80</td>
<td>20</td>
</tr>
<tr>
<td>10.5</td>
<td>92.6</td>
<td>7.4</td>
</tr>
<tr>
<td>11</td>
<td>97.6</td>
<td>2.4</td>
</tr>
</tbody>
</table>

Effect of Ammonia on Seed Germination

Exposure of seeds to NH$_3$ vapour can reduce their viability. Woodstock and Tsao (1986) found that 24 hours exposure to 15 mg/L NH$_3$ vapour killed soybean seed and 1.5 mg/L was required to kill maize. They also found that the damage from lower concentrations increased with the exposure period and that injury was less severe at 5 °C than 25 °C.

The process of germination of a seed begins with water uptake, initially by the embryo, followed by the endosperm (Milthorpe and Mooreby 1974). As the embryo cells reach full turgor, metabolic activity accelerates rapidly, as does the sensitivity of the seed to NH$_3$ damage.

Plant roots appear to be the prime site for assimilation of NH$_4^+$ as they are able to liberate a proton more easily than shoots. The key enzymes involved in this process are glutamine synthetase, glutamate synthetase and glutamate dehydrogenase (Marschner 1995a).

One pathway contributing to the toxic effect of NH$_3$ is its ability to specifically inhibit oxidation of DPNH (diphosphopyridine nucleotide) in the Krebs cycle, thus blocking the transport of electrons from oxidised substrates to oxygen (Vines and Wedding 1960). As this process is concentrated in the most
metabolically active parts of plants, NH₃ has its greatest effects in those plant and seed parts most metabolically active at the time NH₃ is present.

Numerous studies on NH₃ have included measurement of critical levels of NH₃ required for germination damage (Table 2.3). The literature reviewed suggested no common NH₃ concentration at which the germination of most crop species is significantly affected.

Vines and Wedding (1960) found that NH₃(aq.) produced the same effect as NH₃(g.). The effect of NH₃(aq.) concentration in soil water on seed germination and seedling growth was equivalent to that found in the laboratories using NH₃(g.).

The large critical range in Table 2.3 indicates the complexity of the interaction between NH₃(g.) and NH₄⁺ concentration, seed tolerance and soil characteristic such as moisture, texture and tilth. This lack of consistency most likely can be related to different experimental conditions rather than natural variability within the species.

<table>
<thead>
<tr>
<th>Researcher &amp; Dates</th>
<th>Crop</th>
<th>Critical Concentration</th>
<th>Comments</th>
</tr>
</thead>
<tbody>
<tr>
<td>Colliver &amp; Welch</td>
<td>maize</td>
<td>&gt;1000 mg/kg (NH₃ &amp; NH₄⁺)</td>
<td>soil</td>
</tr>
<tr>
<td>(1970)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hunter &amp; Rosenau</td>
<td>maize</td>
<td>&gt;32% NH₄⁺ saturation of soil</td>
<td>closed container</td>
</tr>
<tr>
<td>(1966)</td>
<td></td>
<td>1 mg NH₃/L air</td>
<td>closed container</td>
</tr>
<tr>
<td>Hunter &amp; Rosenau</td>
<td>maize</td>
<td></td>
<td>Soil in pots</td>
</tr>
<tr>
<td>(1966)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Duisberg &amp; Buehner</td>
<td>barley</td>
<td>&gt;450 mg/kg NH₄-N</td>
<td></td>
</tr>
<tr>
<td>(1963)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Allred &amp; Ohlrogge</td>
<td>maize</td>
<td>0.063 – 0.25 mm Hg NH₃</td>
<td>closed containers</td>
</tr>
<tr>
<td>(1963)</td>
<td></td>
<td></td>
<td>solution culture</td>
</tr>
<tr>
<td>Bennett and Adams</td>
<td>cotton and sorghum</td>
<td>0.15 mM - 6.0 mM NH₃ (aq)</td>
<td></td>
</tr>
<tr>
<td>(1970)</td>
<td>roots</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Blanchar (1967)</td>
<td>maize</td>
<td>12 mM NH₃ (aq.)</td>
<td></td>
</tr>
<tr>
<td>Schenk and</td>
<td>cucumber roots</td>
<td>0.06-0.24 mM</td>
<td></td>
</tr>
<tr>
<td>Wehrmann (1979)</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
In general, it could be said that both NH₃(g.) and NH₃(aq.) cause NH₃ toxicity to seed. Damage can commence as soon as the seed-NH₃ contact takes place due to the free passage of NH₃ through biological membranes (Kleiner 1981; Findenegg 1987), and is intensified by the duration of exposure, the NH₃ concentration, crop species tolerance and seed metabolic activity.

There appears to be little evidence that NH₄⁺ has any direct effect on germination. In the many papers reviewed, measurements are quoted as NH₃ plus NH₄⁺ giving no real indication of the injurious level of NH₃(aq.). To convert NH₄⁺-N (mg/g) to concentration of NH₃(aq.), Jacobsen et al. (1986) suggest that the following equation best describes the relationship in an NH₃ band in soil.

\[
\text{NH}_3\text{(aq.) (mM)} = -5.74 \times 10^{-4} + 3.38 \times 10^{-5} \text{(pH)} \times 1.98 \text{(NH}_4^+\text{-N) (mg/g)}
\]

**Osmotic Pressure**

Except under unusual conditions, the osmotic pressure of the soil solution should never become high enough to injure the crop when fertilizer is uniformly broadcast. When fertilizer is localised in a small zone such as that created by band application however, the soluble portion of the fertilizer dissolves only in the soil moisture surrounding the application zone (Rader et al. 1943). This results in local areas of salt solution many times that of the soil solution outside the application zone. High soil solution concentration of fertilizer salt may injure plants by reducing water availability during germination (Dubetz et al. 1959) and/or coleoptile and radicle elongation (Marschner 1995), and in severe cases desiccating young roots. In general, if no plant injury ensues from band placement of fertilizer, higher efficiency of fertilizer result from the rapid access by the crop, reduced weed access to fertilizer and a reduction of soil processes that fix or immobilise nutrients from the soil solution. Osmotic effects of fertilizers result from the rate of diffusion of fertilizer from the application band. Diffusion rate is dependent on factors such as soil compaction, soil moisture, soil permeability, organic matter content, nature of the soil colloids, fertilizer salts involved and the salt
concentration in the soil solution at the time of fertilizer application (Cummins and Parks 1961). Fertilizer salts differ greatly in their effect on the soil solution. The propensity for individual, and combinations of fertilizer salts to create osmotic damage has been explored by Rader et al. (1943) who created tables of fertilizer salt indices (Table 2.4). Ammonia toxicity and increased osmotic pressure from fertilizer placed in close proximity to seed are the 2 most common causes of fertilizer related crop establishment failure (Olsen and Dreier 1956; Carter 1967; Tanaka and Fujinuma 1974).

**Seed Germination**

Availability of moisture is one of the major factors controlling germination. Moisture available for germination is determined by the osmotic and matric potential of water in the seed-bed and the hydraulic conductivity of the water from the substrate to the seed.

**Table 2.4 Salt indices of some fertilizers commonly used in eastern Australia (Rader et al. 1943).**

<table>
<thead>
<tr>
<th>Fertilizer</th>
<th>Salt Index</th>
</tr>
</thead>
<tbody>
<tr>
<td>anhydrous ammonia</td>
<td>47</td>
</tr>
<tr>
<td>ammonium nitrate</td>
<td>105</td>
</tr>
<tr>
<td>ammonium sulfate</td>
<td>70</td>
</tr>
<tr>
<td>urea</td>
<td>75</td>
</tr>
<tr>
<td>monoammonium phosphate</td>
<td>30</td>
</tr>
<tr>
<td>diammonium phosphate</td>
<td>35</td>
</tr>
<tr>
<td>triple superphosphate</td>
<td>17</td>
</tr>
<tr>
<td>single superphosphate</td>
<td>7</td>
</tr>
<tr>
<td>potassium chloride</td>
<td>116</td>
</tr>
<tr>
<td>potassium sulfate</td>
<td>46</td>
</tr>
</tbody>
</table>

During germination of seeds 3 distinct phases can be distinguished; (1) imbibition of water absorbed into the seed; (2) a pause during which time
enzymatic transformation and initiation of meristematic differentiation take place; and (3) the start of growth with radicle elongation and emergence through the seed coat (Mayer and Poljakoff-Mayber 1959). This sequence of stages governs water uptake from the soil or solution.

Soil water osmotic potential is the energy required to move water against the forces of hydration of salts. Electrical attraction between the oxygen side of the water molecule and cations and between the hydrogen of the water molecule and the anions in solution reduces the potential energy of the water molecule.

Imbibition of water is passive. Movement of water into the seed during this first stage of germination is due to the differences in water potential in the seed and that of the surrounding solution (Mayer and Poljakoff-Mayber 1959).

Germination rate decreases with decreasing external water potential and for each species there is a critical value of water potential below which germination will not occur (Hadas and Stibe 1973).

Differences in germination capacity under the effect of low osmotic pressure may be exhibited in 3 measurable ways. As osmotic pressure decreases, total germination % decreases. As osmotic pressure decreases, the rate of germination decreases and a point will be reached, which is variable among species and cultivars, where germination is inhibited.

Seeds of each species and some sub-species have their own rate of germination (Young et al. 1968). Because seeds of different species react differently to reduced osmotic pressure, there is a confounding effect of germination rate in direct comparisons. In many non-irrigated situations, it is assumed that germination rate is an extremely important characteristic governing the chances of successful establishment.
Sharma (1973) found that fast rates of germination were affected by the osmotic potential and the type of plant but not the nature of the osmotic medium in the range of compounds tested. Germination rate was greatest for the dicotyledonous seeds compared with the grasses. This was attributed to the favourable internal seed structure of the dicotyledonous seeds. For example, many monocotyledonous species contain germination inhibitors that delay germination. The presence of a lemma/palea around some grass seed caryopses has been found to reduce injury (Scott 1989).

Hunter and Erikson (1952), George and Williams (1964) and Emmerich and Hardegree (1990) found species sensitivity to low water potential for a range of crop and pasture species including maize, beans (*Phaseolus vulgaris* L.) and sugarbeet (*Beta vulgaris* L. subsp. *vulgaris*), barley, strawberry clover (*Trifolium fragiferum* L.) and Ladino clover (*Trifolium repens* L.) and Buffel grass (*Cenchrus ciliaris* L.). Emergence was generally prevented by osmotic pressures greater than -0.8 MPa for sensitive species and greater than -1.6 MPa for more tolerate species.

**Root and Shoot Elongation**

For circumstances where fertilizer is placed with or in close proximity to germinating seed, it is not only the effect on the germination process that determines the success or failure of the crop to establish. It also depends on rate and magnitude of root and shoot extension.

Generally, there is a marked reduction in both root and shoot elongation in most grass species as the osmotic pressure is increased. For some species this effect can differ between the root and shoot as demonstrated by Young *et al.* (1968).

Adams (1966) and Bennett and Adams (1970a) concluded that calcium (Ca) deficiency induced by high concentration of NH$_3$ in the soil solution was a causal factor in seedling damage from DAP.
Moody et al. (1995a) showed that apart from NH₃ toxicity and osmotic effects, Ca deficiency can cause restriction in root growth in proximity to DAP and MAP granules.

For band applied NH₄⁺-fertilizers ability of roots to explore the soil is dependent on the proximity of and location to the fertilizer band in relation to seed. The chance of root growth restriction due to NH₄⁺-fertilizers increases with decreasing distance between the fertilizer band and germinating seed (Colliver and Welch 1970a). Colliver and Welch (1970b) found that maize roots would not grow into a soil layer containing NH₄⁺ and NH₃ at 1485 mg/kg.

**Species Sensitivity To Fertilizer In Close Proximity To The Seed**

Given that differential response to both osmotic pressure (Sharma 1973) and NH₃(g.) concentration (Woodstock and Tsao 1986) have been recorded it would be expected that there would be a diversity in species response to the combined presence of both effects in some fertilizer compounds.

Carter (1967) concluded from his studies that there were differences in species susceptibility to fertilizer damage during germination. He concluded that generally crucifers such as swede turnips (Brassica rapa L. var. depressa D.C.) and chou moellier (Brassica oleracea L. var. acephala D.C.) were the most sensitive. Legumes such as cowpeas (Vicia sinensis L.) and subterranean clover (Trifolium subterranuem L.) were intermediate and grasses and cereals least sensitive. He also found that there were differences between more closely related species such as wheat and oats. Dubetz et al. (1959) found differences for species sensitivity for maize, beans (Phaseolus vulgaris L.) and sugarbeet (Beta vulgaris L. subsp. vulgaris).

**Plant Establishment**

After a seed has germinated, crop establishment may be affected where roots or shoots encounter high NH₃(g.) concentrations. Research by Allred and Ohlrogge (1964), Hunter and Rosenau (1965), Parr and Engibous
(1967), Bennett and Adams (1970b) and Colliver and Welch (1970b) has shown the detrimental effect of NH$_3$ from both anhydrous ammonia or DAP on root and shoot growth.

On encountering high NH$_3$ concentrations, radicles were stunted, were more branched and thicker near the seed, and had a brown scorched appearance at the tip (Colliver and Welch 1970b). Leaves may also become flaccid where they have emerged from the soil (Bennett and Adams 1970b).

Some parts of an establishing plant are more sensitive to NH$_3$ than others. Allred and Ohlrogge (1964) established that the most sensitive part of maize seedlings was the primary seminal roots, followed by the lateral seminal roots and the plumule. The plumule was believed to be as sensitive as the primary seminal roots due to the rate of metabolic activity, but was less susceptible as it was protected by the coleoptile.

Bennett and Adams (1970b) and Colliver and Welch (1970b) have reported the critical levels for toxic effects on root growth. The levels recorded were 0.13 to 0.17 mM NH$_3$(aq.) for sudan grass roots and 0.17 to 0.22 mM NH$_3$(aq.) for cotton by the former authors and 1172 to 1485 mg/kg (NH$_3$+NH$_4^+$) in maize by the latter authors.

During establishment near the NH$_3$ band, plant growth may also be restricted by the concentration of NO$_2^-$. Nitrite accumulates when pH conditions exist that are unfavourable for nitrifying bacteria that convert NO$_2^-$ to NO$_3^-$. As a result phytotoxic levels of NO$_2^-$ may accumulate.

Levels of NO$_2^-$ from 4 to 80 mg/kg have been found in anhydrous ammonia bands within a week of application (Whitehouse 1972).

Studies by Duisberg and Buehrer (1954) found that up to 26 mg/kg of NO$_2^-$ was not toxic to plants, however Colliver and Welch (1970a) found that
concentrations in excess of 40 to 50 mg/kg of NO$_2^-$ caused maize seedling root injury in nutrient solution studies.

In summary, it appears that there is possibility of NO$_2^-$ toxicity to plants growing in close proximity to an NH$_4^+$ band.

2.3 PROPERTIES AND APPLICATION STRATEGIES FOR SOME COMMON NITROGEN FERTILIZERS.

Properties of ammonia-forming fertilizers

Ammonia is a colourless gas under standard conditions of temperature and pressure (25 °C and 0.103 MPa). It has a molecular weight of 17.03 and represents the -3 valence of N.

Ammonia is the basis for most N containing fertilizers and among the N fertilizers applied directly to soil, anhydrous ammonia, has the highest N concentration (82 %). Natural gas and N$_2$ gas from the air are the raw materials for NH$_3$ manufacture. The natural gas is reacted with steam in the presence of nickel catalysts, at a high temperature (about 800°C) to yield hydrogen, carbon monoxide (CO) and carbon dioxide (CO$_2$). Nitrogen, taken from the air is added to the hydrogen (H$_2$) and the mixture is passed over another catalyst at high temperature and pressure to produce NH$_3$ (Hignett 1978a).

The production of urea involves conversion of CO produced in the breakdown of natural gas during NH$_3$ production, to CO$_2$. One volume of CO$_2$ is reacted with 2 volumes of NH$_3$ to form urea (Fig. 2.2). The reaction, carried out at high temperature and pressure, gives a solution that must be evaporated to produce solid urea. Urea has the highest N concentration (46 %) of any solid N fertilizer (Hignett 1978b).

Sulfate of ammonia was the major N fertilizer used in the early 1960s (Pulsford 1978). The majority of this was produced as a by-product of making coke in steel manufacture. During the coke making process NH$_3$ is produced from the N in the coal. This is then neutralised with sulfuric acid
resulting in the production of sulfate of ammonia. Sulfate of ammonia is also produced by reacting \( \text{NH}_3 \) and sulfuric acid at production facilities for fertilizer \( \text{NH}_3 \).

Ammonium nitrate is a N fertilizer which has a wide range of applications. It is manufactured by vaporising \( \text{NH}_3 \) and steam, and reacting this with nitric acid to produce a 94 % ammonium nitrate solution. This is then concentrated and granulated.

When manufacturing ammonium phosphate compound fertilizers, mono (MAP) and di-ammonium phosphate (DAP), \( \text{NH}_3 \) and phosphoric acid are reacted in the appropriate molar proportions to make the various N-P grades (Hignett 1978c). The resultant slurry is then granulated.

Key properties of a range \( \text{NH}_4^+ \) containing fertilizers are summarised in Table 2.5.

![Fig. 2.2 Summary of manufacture of a range of N fertilizers.](image-url)
Fertilizer toxicity to germinating seeds and growing plants can arise from a number of ions and compounds associated with the fertilizer. Chloride and NH₃ are the most common agents of toxicity and to a lesser degree boron and other trace elements. Biuret, is formed in small quantities (< 1.5 %) during the urea manufacturing process and can affect seedling development but has little effect on germination (Hunter and Rosenau 1966).

Table 2.5  Selected properties of ammonium fertilizers (Glendinning 1990).

<table>
<thead>
<tr>
<th>Fertilizer Product</th>
<th>anhydrous ammonia</th>
<th>urea</th>
<th>ammonium sulfate</th>
<th>ammonium nitrate</th>
<th>MAP</th>
<th>DAP</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chemical formula</td>
<td>NH₃, (NH₂)₂CO</td>
<td>(NH₄)₂SO₄</td>
<td>NH₄NO₃</td>
<td>NH₄H₂PO₄</td>
<td>(NH₄)₂HPO₄</td>
<td></td>
</tr>
<tr>
<td>pH (sat. soln.)</td>
<td>14</td>
<td>10.7</td>
<td>4.5</td>
<td>5.5</td>
<td>4.7</td>
<td>7.4</td>
</tr>
<tr>
<td>Solubility (kg/100 L @ 20°C)</td>
<td>85</td>
<td>107</td>
<td>75</td>
<td>192</td>
<td>38</td>
<td>70</td>
</tr>
</tbody>
</table>

**Urea**

Urea, while as an intact molecule, has been found to be non-toxic to germinating seeds exposed at N rates up to 66 kg/ha (Hunter and Rosenau 1966). However, biuret, a “double” urea molecule formed from urea held at high temperature (>150 to 170 °C) during manufacture, and the NH₃ evolved following the breakdown of urea may be toxic at significantly lower application rates (Hunter and Rosenau, 1966). The osmotic effect of urea even after hydrolysis to NH₄⁺ will not reach the osmotic pressure resulting from equivalent rates of other NO₃⁻ and NH₄⁺-fertilizers.

Conversion of urea to NH₃ is, for the majority of soils, facilitated by the presence of urease enzymes, although a small amount of conversion is facilitated through chemical processes mediated by soil pH. This conversion is rapid (1 to 2 days) under warm moist conditions, a significant factor in the development of high concentrations of NH₃ in the fertilizer application bands (Whitehouse and Leslie 1973).

Urea application in broadacre cereal, oilseed and fibre crops is commonly associated with a pre-sowing cultivation operation. Application timing and soil placement depend on soil moisture content, weed population, and the
probability of follow up rainfall. Applying urea at sowing with the seed as a standard practice has been avoided but when used as a last resort, such as where extreme wet or dry soil conditions preclude pre-sowing application, the effects on crop establishment varied widely. Poor emergence in crops is frequently observed at recommended rates in some areas and farmers frequently comment that they are able to regularly apply urea successfully with seed at greater rates than recommended without affecting crop establishment. The prime factors involved in the variability of response are commonly soil drying rate post-sowing, which is affected by both soil type and level of disturbance caused by application equipment, and the effective rate in the seed-row in contact with seed (Radford, personal communication 1996).

However, with increased popularity of the urea-ammonium phosphate blends and application in wider row spacings (22 to 25 cm), the seed-row concentration of NH$_3$ forming fertilizers is generally increasing.

Although ammonium sulfate was the most commonly available solid N fertilizer in the early days of N use in cropping in Australia, urea use outstripped ammonium sulfate during the 1960s as the price declined and a local source of urea became available (Pulsford 1978). This growth has continued into the 1990s with urea-N making up the major proportion of the total Australian N market as indicated by urea sales growth in northwest New South Wales and southeast Queensland (Fig. 2.3).

Urea application in broadacre cropping is commonly associated with a pre-sowing cultivation where it is drilled into the soil using an air seeder attached to a trash-worker cultivator, a scarifier, or combine drill (Ward 1987). Application timing and soil placement for pre-sowing application of urea are similar to anhydrous ammonia.
Application of urea at sowing can be achieved without sacrificing crop establishment by either minimising the concentration of product in contact with the germinating seed or placing the urea away from the seed at sufficient distance that the urea will not affect crop establishment. Various modifications of airseeders and ground tools are now available to place urea safely at sowing (Robotham 1993; Desbiolles 1998).

Where no N fertilizer has been applied pre-sowing or at sowing, urea applications are surface broadcast and incorporated mechanically, by irrigation or by rainfall.

**Anhydrous Ammonia**

**High pressure application methods**
The first anhydrous ammonia application equipment used in Australia was designed around application equipment developed in the USA. For irrigated row crops, narrow or knife tines were used with varying success. Successful
NH₃ retention with this configuration depended on the soil moisture, texture, tilth, application depth and the efficiency of furrow covering devices. Knife tine application is still widely used in the cotton industry where NH₃ is applied 20 to 40 cm deep as a part of the tillage programme.

Application methods were modified from 1963 to 1965 on the Darling Downs for clay soils that predominate there. Modification was aimed at reducing the high level of soil disturbance and poor NH₃ retention. Shallow application with knife tines increased the amount of direct NH₃ loss. Deep anhydrous ammonia application was abandoned, as in many seasons soil disturbance close to sowing caused unacceptable losses in seedbed moisture and tilth.

The basis of the new equipment was an increase in the number of application outlets, both number of tines and number of outlets per tine. These modifications reduced the application rate per unit length of row, allowing shallower application depths with acceptable NH₃ retention and a reduction in seedbed disturbance.

Further developments in application equipment took place from 1968 to 1970 as anhydrous ammonia application was incorporated during a seed-bed tillage. Equipment developed for this application method had a greater number of tines for weed control than the original anhydrous ammonia applicators. This also provided a more uniform soil tilth for the following application tines (Incitec Fertilizers 1994).

With broader adoption of reduced tillage farming systems during the 1980s, opportunities for anhydrous ammonia application into "ideal" soil conditions were greatly reduced. Soils were no longer cultivated to provide the necessary conditions for anhydrous ammonia application, and other N products were substituted. Fitting of anhydrous ammonia application equipment to primary and secondary tillage implements (chisel ploughs and scarifiers) was the next development that allowed anhydrous ammonia to remain a viable N alternative.
**Low pressure anhydrous ammonia application**

With further moves to reduced tillage, towards zero tillage, effective application of anhydrous ammonia again became a problem. Unacceptable product loss was incurred because of the inability of application equipment to cover the NH₃ vapour with soil before it escaped to the atmosphere.

Development of the Cold Flo® method of anhydrous ammonia application addressed the underlying problem with conventional application, vapour velocity. Cold Flo® uses the powerful refrigerant properties of NH₃ to produce "supercool" liquid NH₃ (-33.3°C) at atmospheric pressure (about 85% of the total volume) and a warm low pressure vapour (about 15% of the total volume).

The success of the Cold Flo® equipment resulted from the elimination of pressure from the system as well as the delayed vaporisation of the liquid component. These factors in combination reduced the need for rapid furrow closure. However failure to close the tine furrow after Cold Flo® application still produced unacceptable losses for hours after application (Incitec Fertilizers 1994).

**Ammonium Phosphates and Urea/Ammonium Phosphate Blends**

The ammonium phosphates, MAP and DAP are the most widely used P fertilizer sources used in cropping. These fertilizer products are normally applied at sowing in contact with the seed providing potential for reductions in germination and seedling establishment where rates are too high. Differences in crop species susceptibility to establishment damage from MAP or DAP is generally not recognised in current “safe” rate recommendations.

There is greater potential for establishment damage from DAP because of the ease with which a second NH₄⁺ ion dissociates from DAP and its alkaline

*Cold Flo is a registered Trademark of USS Agri Steel*
pH prior to nitrification which together create a high NH$_3$ concentration (Hood and Ensminger 1964; Stevenson and Bates 1968; Moody et al. 1995b).

Crop establishment damage from fertilizer blends containing ammonium phosphates and urea may differ depending on the pH of the ammonium phosphate used and the proportion of urea and ammonium phosphate in the blend.

2.4 AMMONIA ADSORPTION PROCESSES

Clearly, retention of NH$_3$ in soils is a complex process and has been the subject of research for the last 40 years. Much is known about the individual components under limited experimental conditions, but the broader interactive view is still largely unexplained.

Each of the various components i.e. CEC, soil moisture, soil tilth, application depth, soil pH and soil texture have been correlated with NH$_3$ retention. The degree of correlation between many of these components and NH$_3$ retention probably depends on the soils and experimental conditions used, when measurements have been made on a single factor basis.

The process of adsorption in this review is defined as the chemical and physical reactions that hold NH$_3$ in the soil. This is distinct from mechanical retention, which includes all factors both internal and external to the soil that allow NH$_3$ to be trapped in the soil for adsorption.

Efficient anhydrous ammonia soil incorporation requires a balance between mechanical retention processes and longer-term adsorption reactions. For other NH$_4^+$ forming fertilizer products the mechanical retention process is relatively less important.

The methods of NH$_4^+$ adsorption in Australian soils and their relative importance have undergone little or no investigation. The majority of the
basic information has been generated in the USA and Europe and more recently in India.

These processes indirectly affect the NH₃ toxicity to seed, as it is the ability or inability of the soil to hold NH₃ that is a major moderating factor affecting NH₃ vapour concentration in the soil atmosphere.

**Physical Adsorption Processes**

Physical mechanisms of NH₃ retention are of 2 types. The first are those that render NH₄⁺ unavailable for nitrification such as entrapment of NH₃ between clay lattices and the replacement of clay lattice interlayer cations with NH₄⁺ (Stanford and Pierre 1946). The second mechanism is characterised by easily reversible retention releasing NH₃ for subsequent transformations to NO₃⁻. This mechanism is only present when there is a positive pressure of NH₃ in the soil e.g. around the injection site. As soon as the pressure of NH₃ in the gaseous phase decreases, physically adsorbed NH₃ reverts back to the gaseous phase and diffuses through the soil until it is chemically adsorbed or lost to the atmosphere (Mortland 1955). Sites for most chemical adsorption are soil minerals and organic matter.

**Chemical Adsorption Processes**

Chemical adsorption of NH₄⁺ is characterised by difficulty in reversing the reaction because of the bond strength and subsequent amount of energy to reverse the reaction. As a result, these sites are important in retaining NH₄⁺ so it is not converted back to the gaseous phase and lost to the atmosphere or present in toxic levels to seeds (Mortland 1958).

**Adsorption by Soil Minerals**

The chemical adsorption reaction that results in the greatest stability of NH₃(g.) in mineral soils is the formation of NH₄⁺ as a result of a significant change in energy levels of the participating atoms and ions (Parr and Papendick 1966b). Ammonium ions are formed when NH₃ molecules are able to react freely with hydrogen ions. The NH₄⁺ ions have a positive
electrical charge and are attracted by the negative charge provided by soil minerals and organic matter.

Cornet (1943) showed that in acidic soils cation exchange sites on clay minerals were the most common source of hydrogen ions.

\[
\text{clay O - H} + \text{NH}_3 \leftrightarrow \text{clay - O - NH}_4^+ 
\]

In acidic soils another possible adsorption process is associated with aluminium ions. Russell (1965) proposed that NH\textsubscript{4}\textsuperscript{+} ions on clay mineral surfaces cause acid reactions. When NH\textsubscript{3} molecules react with aluminium saturated clays, NH\textsubscript{4}\textsuperscript{+} ions are formed from the hydrogen ions provided from the hydrolysis of water. In the alkaline pH range the NH\textsubscript{3} molecule may also react with hydrogen ions arising from hydroxyl groups associated with silicon on the edges of clay minerals, or in amorphous material in the soil (Mortland 1966).

\[
-\text{Si-OH} + \text{NH}_3 \leftrightarrow -\text{Si - NH}_4^+ 
\]

In a low pH environment, these sites will convert NH\textsubscript{3} molecules into NH\textsubscript{4}\textsuperscript{+} ions but as the pH increases, NH\textsubscript{4}\textsuperscript{+} may re-convert to NH\textsubscript{3}.

Clays saturated with calcium and magnesium ions (alkaline condition) may also convert adsorbed NH\textsubscript{3} to NH\textsubscript{4}\textsuperscript{+} (Mortland et al. 1963). It is thought that the polarisation forces of these cations associated with swelling clay minerals cause an increased dissociation of water. Russell (1965) showed that this type of reaction was easily reversible when the NH\textsubscript{3} concentration decreased and that the complex formed by magnesium was more stable than that formed with calcium. In some systems this conversion may be persistent enough to allow uptake of NH\textsubscript{4}\textsuperscript{+} by plants and microorganisms, or to be fixed by more permanent chemical adsorption processes.
Another unstable adsorption process involves the displacement of water molecules from hydrated exchange metal ions (M) on the exchange complex by NH$_3$ molecules (Mortland 1966).

\[
\text{M(H}_2\text{O)}_n\text{(clay)} + m \text{NH}_3 \rightleftharpoons \text{M(NH}_3)_m\text{(clay)} + n\text{H}_2\text{O}
\]

Increase in soil moisture will cause the reaction to reverse and NH$_3$ vapour to diffuse through the soil to be absorbed or lost to the atmosphere.

**Adsorption by the Soil Organic Matter Fraction**

The organic fraction of the soil is relatively more reactive in relation to the amount of NH$_3$ vapour present than the inorganic fractions. Sohn and Peech (1958), Burge and Broadbent (1961), Nommik and Nilsson (1963), Young (1964) and Mortland (1966), found NH$_3$ adsorption closely correlated with the percentage of organic matter/or carbon concentration of the soil. Young (1964) established a linear relationship between total NH$_3$ retention and CEC for soils of wide ranging carbon contents.

\[
i.e. \text{mg/kg NH}_3 \cdot \text{N} = 147 + 596 \times \% \text{ organic carbon} + 43.4 \times \% \text{ clay}
\]

The capacity of soil organic matter to adsorb NH$_4^+$ ions in a non-exchangeable form has been known for over 4 decades. This process has been found to be pH dependent (Broadbent *et al.* 1958; Nommik and Nilsson 1963) increasing as the pH increases above neutral. This fixation has been associated with oxidation reactions but not totally dependent on them. Broadbent *et al.* (1958) showed that over a short duration NH$_4^+$ fixation was independent of oxygen partial pressure, suggesting that reactive organic groupings capable of NH$_4^+$ fixation may be present in soils in large enough quantities to permit rapid initial fixation of NH$_4^+$. The availability of NH$_4^+$ fixed by organic matter for plant uptake is not clear.
Nitrite (NO$_2^-$) ions formed as an intermediate during biological oxidation of the NH$_3$ molecule to NO$_3^-$ may also react chemically with the soil organic matter fraction (Sohn and Peech 1958).

Thus retention of NH$_3$(g.) by both mineral and organic fractions appears to be due to cation exchange sites. Later research by Parr and Papendick (1966a) and others concluded that soil moisture and other chemical adsorption pathways are also important in NH$_3$ adsorption.

More recent research by Izaurralde et al. (1987) has approached the measurement of NH$_3$ retention not by measuring and correlating single soil components, but instead by using titratable acidity, to estimate NH$_3$ retention capacity. Use of titratable acidity as an estimator of NH$_3$ originates from the work of Mortland (1966) who recognised that high alkalinity created by NH$_3$ in soils creates conditions for the release of hydrogen ions from several sources. Thomas and Hargrove (1984) further defined the most likely source of the H$^+$ as being from hydrolysis of cation exchangeable Al$^{3+}$, non-exchangeable polymeric Al and Fe hydrous oxides, as well as organic matter.

Izaurralde et al. (1986) concluded that the following reaction best describes the NH$_3$ retention mechanism in soil and that the titratable acidity method of determining retention was a better parameter than CEC to use in a predictive model.

\[
\text{NH}_3 + H^- \text{- soil} \leftrightarrow \text{NH}_4^- \text{- soil}
\]

2.5 FACTORS AFFECTING ADSORPTION, RETENTION AND MOVEMENT OF AMMONIA.

Soil Moisture

A number of researchers (Jackson and Chang 1947; Blue and Eno 1954; Stanley and Smith 1956; Parr and Papendick 1966a) have shown that soil moisture plays some part in the NH$_3$ retention process. Some have
implicated soil moisture directly as a part of the physical and chemisorption reactions, while others have shown indirect effects through effects of NH$_3$ on air-filled porosity and soil tilth. The most plausible explanation for the effect of soil moisture on NH$_3$ retention is that moisture effects physical and/or chemical reactions with soil, as proposed by Parr and Papendick (1966a).

When anhydrous ammonia is applied at high soil water content, soil moisture supplies the "initial capacity" to adsorb NH$_3$, due to its solubility in water. However, the ability of a soil to retain NH$_3$ is more characterised by the quantity of NH$_3$ retained when soil is in an air-dry condition.

Movement of NH$_3$ away from the application site is restricted at high moisture content, and high concentrations of NH$_3$ (g. and aq.) exist at close proximity to the injection point. However, this effect appears to be only temporary with the movement of NH$_3$ being greater after 36 hours in moist soil than in air dried soil (Papendick and Parr 1966a).

*Soil Texture And Tilth*

Most studies suggest that soil texture plays a major part in the retention of soil applied anhydrous ammonia but has little effect on other NH$_4^+$-fertilizers when placed under the soil surface. Jackson and Chang (1947), Stanley and Smith (1956), McDowell and Smith (1958) and Goring and Martin (1960) observed that under laboratory conditions soil texture was a primary factor in the ability to retain NH$_3$. Retention is greatest for fine textured materials (clays) and least for coarse textured materials (sand).

Under laboratory conditions, Blue and Eno (1952) attributed the greater losses of NH$_3$ in sandy soils to NH$_3$ movement. The amount of movement is related to the lower number of adsorption sites in sand as compared to clay, the sand being more permeable to gases than clay. However, soil texture is not always a reliable measure for determining the soil's ability to retain NH$_3$. Jackson and Chang (1947) proposed that even in coarse textured soils, if the soil contained no moisture and the NH$_3$ was placed at sufficient depth, NH$_3$ could be successfully retained.
They also hypothesised that a clay soil in an unusually cloddy state would be expected to lose NH$_3$. This type of loss has commonly been observed on Mywybilla clays of the central Darling Downs, Queensland. The soil moisture directly affects the tilth of fine textured soil. In the soils to which anhydrous ammonia is applied in eastern Australia, the interaction between soil texture and soil moisture (as it affects tilth) is of vital importance in the retention mechanism.

From laboratory studies, it would seem that clay soils of high moisture content would be best suited for NH$_3$ retention. In field conditions, this is not the case. Moisture conditions that adversely affect the ability of the soil to effect closure and sealing of the anhydrous ammonia injection furrow, reduce the soils’ ability to retain the applied NH$_3$.

Papendick and Parr (1966a) concluded that: -
"Since dry soil retains at least as much NH$_3$ as moist soil, contrary to popular opinion the soil moisture content per se at the time of anhydrous ammonia application is not a critical factor in the retention of NH$_3$ in the field. However, the soil moisture content is a highly important consideration in providing proper soil physical conditions to ensure rapid and complete sealing of the injection channel."

**Soil pH**

For acidic soils, clay minerals are the most attractive NH$_3$ retention sites, while under alkaline conditions organic matter is more effective (Mortland 1955).

Various researchers (Jackson and Chang 1947; Mortland 1958) found that NH$_3$ adsorption in soil proceeded to pH 10.5 as long as sufficient hydrogen ions were available. When hydrogen ions become depleted, physical adsorption followed.
Whitehouse and Leslie (1973) showed that in an alkaline Waco black earth soil of the Darling Downs Queensland, the pH within 25 mm of the centre of anhydrous ammonia and urea application bands rose from a background level of 8.3 to pH 8.6 to 9, lasting for about 7 days. Between 7 and 30 days after application, there was a marked decline in pH (1.3 to 1.6 pH units) due to nitrification. After 30 days, nitrification was complete and the pH rose in response to the buffering capacity of the soil.

The rise in pH, followed by a decline as nitrification proceeds has been described widely by other authors, such as McDowell and Smith (1958), McIntosh and Frederick (1958) and Parr and Engibous (1967).

Soil pH as a single factor appears to be of secondary importance in NH₃ retention mechanisms. Application depth, soil moisture, clay and organic matter content and soil physical conditions being initially more important.

**Cation Exchange Capacity**

Cation exchange capacity plays an important part in the mechanisms of NH₄⁺ retention, as it is a measure of a soil's ability to hold positively charged ions. Jenny *et al.* (1945) and Brown and Bartholomew (1962) reported that NH₄⁺ adsorption was closely correlated with CEC.

Soil CEC has given varying results when used as a measure of NH₃ retention ability because of its dynamic nature.

Cornet (1943) found that the retention of NH₃ vapour by an acidic hydrogen-bentonite clay was closely related to its CEC. Conversely, Parr and Papendick (1966a) found that NH₃ retention was poorly correlated with CEC, and commented that the relationships established by other investigators were most likely a consequence of the choice of soil and experimental conditions. Izaurralde *et al.* (1987) concluded that soils with similar mineralogy and initial pH, but different textures, would probably exhibit a high correlation between CEC and NH₃ retention. However, such correlations would not provide a useful means to quantitatively estimate the NH₃ retained,
or the size of the retention band in a wide variety of soils because many of
the cation exchange sites do not participate in the soil’s capacity to adsorb
NH$_3$. This is not to say that CEC is not an important mechanism in NH$_3$
retention but it is not necessarily highly correlated with CEC.

**Fertilizer Placement Depth**

Placement as a single factor does not have a major influence on the
retention mechanism. However, it becomes important when considered in
combination with soil moisture, soil texture and soil tilth.

For urea, Campbell *et al.* (1984) found that there was a difference in NH$_3$
retention, which depended on soil texture, moisture content and soil
placement. Losses were greater from light textured, poorly buffer soil,
shallow (1.25 cm) application depth and where urea was broadcast and
mixed into the soil instead of banding.

Whereas placement is relatively unimportant in most other NH$_4^+$-fertilizers
placement may affect retention of anhydrous ammonia. Stanley and Smith
(1956) reported that losses were minimal when anhydrous ammonia was
applied at optimum moisture regardless of depth of placement. However, as
the soil moisture moved away from the optimum in both directions, losses
increased. Losses of NH$_3$ from wet soils occurred because of upward
movement of NH$_3$ gas and evaporation of water. In dry soils, the mechanism
of NH$_3$ loss was postulated to be mass flow to the soil surface because of
pressure.

McDowell and Smith (1958) suggested that an application depth of 15 cm
would be sufficient for adsorption on silt loam and clay soils irrespective of
moisture content, but in dry coarser soils application depth should be
increased.

In furrow irrigation, when dry fertilizers are placed in the seed-bed and water
is applied, movement of the dissolved solute is likely. Depending on the
depth of placement, the fertilizer N may move downward or to the surface through capillary action (Tisdale and Nelson 1975).

**Ammonia Movement**

The interaction between factors that influence NH₃ retention determines the direction and extent of movement of NH₃(g.) away from fertilizer application bands.

These factors therefore determine the diameter of the NH₄⁺-fertilizer band, the likelihood of the NH₄⁺ band reaching the seed row, where the NH₄⁺-fertilizer was applied at sowing, and the chance of NH₃ losses to the atmosphere. In the majority of the papers reviewed that measured the spread of the NH₃-NH₄⁺ band, the distance NH₃ moved was less than 7.5 cm (Table 2.6). This spread was similar across a range of soil types and anhydrous ammonia application rates in the laboratory, and application conditions in the field.

In eastern Australia (Whitehouse and Leslie 1973) and in similar soils (Khrengre and Savant 1977) anhydrous ammonia spread 50 mm from the point of application. In both experiments the soils were kept at field capacity similar to that experienced around sowing time. Hence the conditions that minimise the size of the NH₃ band, i.e. soil moisture and soil texture, occur at sowing in the clay soils of eastern Australia.

The data of Whitehouse and Leslie (1973) suggests that urea diffusion is a minor contributor to size of the NH₄⁺ application band. Urea diffusion (25 mm in 7 days) was limited by rapid hydrolysis to NH₄⁺-N. Retention zones of NH₄⁺ were smaller for urea than an equivalent N rate applied as anhydrous ammonia.

**2.6 TRANSFORMATIONS OF AMMONIA IN SOIL.**

Nitrogen is present in the soil largely (92 to 95 %) in an organic form. Before plants can use N in organic matter, it undergoes chemical changes by microbial processes to mineral forms such as NH₄⁺ or NO₃⁻. The organic N in
plants and animals is normally in its reduced form, the same oxidation state as NH$_3$(g.). But, because NH$_3$ can be oxidised to NO$_3^-$, yielding energy for microbes, NO$_3^-$ is the more common N form found in agricultural soils (Subcommittee on Ammonia 1979b).

**Table 2.6 Soil NH$_3$ band radius reported by authors in a range of soil types and application rates.**

<table>
<thead>
<tr>
<th>AUTHOR</th>
<th>SOIL TYPE</th>
<th>APPLICATION RATE</th>
<th>APPROXIMATE AMMONIA BAND RADIUS (cm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Izauralde et al. (1986)</td>
<td>Silt</td>
<td>20.7 dg N/m</td>
<td>3-5</td>
</tr>
<tr>
<td></td>
<td>Fine silty clay</td>
<td>61.7 dg N/m</td>
<td>5-7</td>
</tr>
<tr>
<td></td>
<td>Fine silty clay</td>
<td>70.8 dg N/m</td>
<td>7.5-10</td>
</tr>
<tr>
<td>Izauralde et al. (1987)</td>
<td>Silt loam</td>
<td>8.4 g N/m</td>
<td>5.5</td>
</tr>
<tr>
<td>McDowell and Smith (1958)</td>
<td>Fine sandy loam</td>
<td>11 g N/m</td>
<td>7</td>
</tr>
<tr>
<td>Whitehouse and Leslie (1973)</td>
<td>Silty loam</td>
<td>11 g N/m</td>
<td>5</td>
</tr>
<tr>
<td></td>
<td>Clay</td>
<td>11 g N/m</td>
<td>3</td>
</tr>
<tr>
<td></td>
<td>Clay</td>
<td>165 kg N/ha</td>
<td>5-6.5</td>
</tr>
<tr>
<td>McIntosh and Fredericks (1958)</td>
<td>Sandy clay loam</td>
<td>132 kg N/ha</td>
<td>5-7</td>
</tr>
<tr>
<td>Eno, Blue and Good (1955)</td>
<td>Fine sand</td>
<td>n/a</td>
<td>2.5-5</td>
</tr>
<tr>
<td>Parr and Engibous (1967)</td>
<td>Fine sandy loam</td>
<td>370 kg N/ha</td>
<td>10-12.5</td>
</tr>
<tr>
<td>Babowitz (1985)</td>
<td>Silt loam</td>
<td>121 kg N/ha</td>
<td>5</td>
</tr>
<tr>
<td>Khengre and Savant (1977)</td>
<td>Clay</td>
<td>800 mg</td>
<td>5</td>
</tr>
</tbody>
</table>

Ammonia is a ubiquitous constituent of the soil, the atmosphere and in water. Its integral association with the N-cycle, is shown in Fig. 2.4.

**Ammonia Fixation**

Since NH$_3$ is primarily applied to the soil to stimulate plant growth, the amount fixed by the clay and/or organic fractions of soil is not available for uptake by plants That which is not fixed is available to plants only after nitrification has taken place.

Mechanisms of NH$_4^+$ fixation in clay are related to the size of the NH$_4^+$ ion, the structure of the clay mineral and the location and magnitude of the electrical charge in the clay mineral. If vermiculite minerals are present in soil, NH$_4^+$ ions may move from a position on the external surface of minerals
to an internal exchange site. Subsequently, $\text{NH}_4^+$ replaces water molecules in the clay lattice and clay lattices collapse rendering the $\text{NH}_4^+$ unavailable for plant uptake or for other transformations.

In an incubation experiment Broadbent (1966) found that the fixation could persist for a long period of time and was unable to detect any decrease in fixation over a 120 day period.

Fixation of $\text{NH}_4^+$ by soil organic matter appears to take place in the lignin derived fraction and is most favoured by conditions of high pH thus conditions within the $\text{NH}_3$ application band are favourable for fixation of $\text{NH}_4^+$ ions by organic matter. Information about the availability of $\text{NH}_4^+$ fixed by organic matter is limited. Burge and Broadbent (1961) found that part of the organically fixed $\text{NH}_4^+$ may be as available as native soil N but not as available as that from mature crop residues. Sohn and Pech (1958) and Opuwaribo and Odu (1975) concluded that at least 50% of the $\text{NH}_4^+$ fixed by the mineral surface soils that they investigated was due to some reaction of $\text{NH}_4^+$ with the soil organic matter.

**Nitrification**

Nitrification rate is an important factor in reducing the toxicity risk of $\text{NH}_3$ from fertilizer bands. Delayed nitrification reduces the risk of N loss from processes such as denitrification and leaching but increases the risk of crop establishment failures. It is important therefore to know the main factors and outcomes of interactions to assess the rate of nitrification and any risk to crop establishment.

Oxidation of $\text{NH}_3$ can take place through chemical, photochemical and biological processes. The importance of non-biological oxidation of $\text{NH}_3$ in soil is not fully understood. However, it appears to be insignificant compared with the biological processes. Non-biological oxidation can be induced by radiation such as sunlight, cosmic rays and natural radioactivity in laboratory conditions (Mortland 1958).
Ammonium that is not fixed by the organic matter or clay fractions of the soil or taken up by plants and other soil microbes can be oxidised to NO$_3^-$ by nitrifying bacteria. This process takes place in two steps, with different bacteria involved in each step. The first oxidation step involves the autotrophic bacteria _Nitrosomonas spp._ and some related genera. They are able to gain the energy necessary for carbon dioxide fixation, growth and reproduction from the oxidation of NH$_4^+$ to NO$_2^-$ (Subcommittee on Ammonia 1979b). The next step involves another group of autotrophic bacteria, _Nitrobacter spp._ and related genera. These bacteria produce NO$_3^-$ from NO$_2^-$, using energy released from these processes for growth.

As a result of the NH$_3$ oxidation pathway yielding energy to microbes during both processes, NO$_3^-$ is the more common mineral N form found in the soil.

Only where soil environmental conditions are moved outside the range that is optimal for normal functioning (e.g. in fertilizer band) of the 2 nitrifying bacterial groups, is NO$_3^-$ not that the dominant form of soil mineral N.

Oxidation of NH$_4^+$ to NO$_3^-$ is important to crop production for 2 reasons, firstly because most crop species take up the majority of their N as NO$_3^-$ and secondly because high concentrations of soil NH$_3$ and NO$_2^-$ are toxic to plant growth.
Fig. 2.4  The nitrogen cycle (source Incitec Fertilizers Analysis Systems Soil Interpretation Manual).
Factors that most affect nitrification rate after an application of NH$_4^+$-fertilizers are the pH in the application band, population of nitrifying bacteria present, concentration of NH$_4^+$ and NO$_3^-$, and soil temperature. Complete transformation of NH$_4^+$ to NO$_3^-$ is most rapid at pH levels in neutral to slightly alkaline soils. Bacteria responsible for conversion of NO$_2^-$ to NO$_3^-$ (*Nitrobacter* spp.) are most efficient in neutral to acid soil conditions, while the neutral to alkaline conditions are favoured by *Nitrosomonas* spp. bacteria that convert NH$_4^+$ to NO$_2^-$.

Soil pH of an anhydrous ammonia or urea application band reduces, spatially away from the point of release and in time as NH$_3$ diffuses away from the injection site. Thus nitrification of applied usually proceeds from the outside of the fertilizer band, inwards (Eno *et al.* 1955; Whitehouse and Leslie 1973).

Various researchers in the USA (Duisberg and Buehrer 1954; McIntosh and Frederick 1958; Izaurralde *et al.* 1987) have recorded increases in pH at the centre of the application band of up to 3.5 pH units. The majority of soils reach a pH of 9 to 9.5 at the point of anhydrous ammonia release in silty loam to clay loam soils. In contrast, Whitehouse and Leslie (1973) found that the pH at the application point in a Waco clay soil from the Darling Downs rose only 0.3 pH units (from 8.3 to 8.6) after application of 168 kg/ha N as anhydrous ammonia. The small rise in pH in the clay soil is most likely attributable to the buffering capacity conferred by the clay content (77 %) and clay species (montmorillonite). Lower pH in the band may account for the higher rates of nitrification in this experiment than in those conducted by Eno *et al.* (1955).

As nitrification proceeds from the more favourable pH areas around the outer surface of the NH$_4^+$ application band, the pH of the soil is reduced below its original level. Then, depending on the soils buffering capacity, the pH returns to its original level (Eno *et al.* 1955).
Where soil pH at the centre of the application band falls below pH 5.5 to 5 it is possible for nitrification to be drastically inhibited as was found by Whitehouse and Leslie (1973) in an ammonium sulphate application and also by McIntosh and Frederick (1958). This effect is most likely linked to the preference of *Nitrosomonas* *spp.* bacteria for neutral to alkaline soil conditions and their inhibition in an acid environment.

**Factors Affecting the Nitrification Process**

**Ammonium concentration**

Ammonium-N must be present for nitrifying bacteria to persist. For clay soils of the Darling Downs, Martin and Cox (1956) showed that NH₄⁺-N had a small periodicity with a minimum level occurring from May to August (winter). Their incubation studies suggest that NH₄⁺-N rarely fell below 1 mg/kg, indicating that this may be the minimum level required for the nitrification process in this soil. The highest concentrations recorded from fallow plots were 2 to 3 mg/kg NH₄⁺-N.

The effect of NH₃ on nitrifying bacteria is related to the concentration per unit volume of the soil rather than the rate applied per hectare. When NH₄⁺-fertilizer is applied to soil, the resulting soil NH₃ concentration will depend on the NH₄⁺ rate per unit area, number of tines per unit area, number of outlets per tine and soil characteristics involved in adsorption and retention of NH₃.

Broadbent *et al.* (1958) found that the rate of nitrification of NH₃ increased as the concentration in soil increased until one or more of the following conditions limited further nitrification:

(a) excessively high pH resulting from the application of NH₃.

(b) toxic effects of free NH₃ causing toxicity to *Nitrosomonas* *spp*.

(c) excessively low pH, resulting from the formation of nitric and nitrous acid, inhibiting *Nitrobacter* *spp.* activity.
(d) osmotic effect of the NO₃⁻ ion on nitrifying bacteria.

The pH effects and the presence of free NH₃ may interact to have direct and/or indirect effects on nitrification as described in the previous section.

Eno and Blue (1955) and Broadbent et al. (1958) found that pH of 8.5 and above may inhibit nitrification. However, Whitehouse and Leslie (1973) found significant nitrification in black clay soils at pH 8.6. It would appear from the differences between the two findings, that pH per se may not be as important to nitrification as the concentration of free NH₃.

Ammonium-N concentrations between 1300 and 3250 mg/kg have been measured at the centre of anhydrous ammonia application bands by various researchers (Broadbent et al. 1958; McIntosh and Frederick 1958; Duisberg and Buehrer 1954; Whitehouse and Leslie 1973). The lower NH₃ concentrations were in coarse textured soils whereas higher concentrations were generally associated with fine textured soils. Although the concentration of NH₃ at the centre of the bands varied greatly, the diffusion zone where nitrification began was restricted to soil with NH₄⁺-N concentrations less than 280 and 400 mg/kg (Duisberg and Buehrer 1954; Eno et al. 1955; McIntosh and Frederick 1958).

Ammonium-N concentrations after application were found to decrease as nitrification proceeded from the outside of the application band toward the centre. Persistence of the NH₄⁺ was related to the soil type and the initial NH₃ concentration in the application band. The time taken for nitrification to commence at the centre of the application band varied from 14 to 30 days in the papers reviewed.

**Soil nitrifying microbial population**

Nitrification in soil is insignificant after soil has been sterilised. The minimum number of nitrifying organisms required to form NO₃⁻, and the minimum number for maximum NO₃⁻ production are not precisely known. Little
increase in rate appears to occur when the population exceeds 100,000/g. The interaction between population and soil temperature is more closely related to nitrification rate than population alone. Sabey et al. (1959) found that increasing the initial population of nitrifiers caused a decrease in the delay period (before maximum nitrification was reached) but did not affect the maximum.

**Soil Temperature**
The effect of soil temperature on nitrification is also influenced by other factors such as microbial population and soil moisture. Nitrification has been found to take place in a temperature range from 0°C to above 35°C, with an optimum range of 15°C to 25°C (Frederick 1957; Sabey et al. 1957). The effect of sub-optimal temperature conditions was found to be associated with reduced rates of nitrification as well as longer delay before nitrification commenced. Justice and Smith (1962) also found a similar effect but also found that under such conditions NO₂⁻ was the major product. Extremes of temperature appear to be more inhibitory to NO₂⁻ oxidisers than NH₄⁺ oxidisers.

In an NH₃ band, the length of the delay period before significant nitrification begins has been found to be due to combined effects of overcoming inhibition caused by the NH₃ concentration and the time taken to build up the nitrifier population (Frederick 1957). Due to the variation in maximum nitrification rate and delay period between soils, Sabey et al. (1959) concluded that factors other than temperature and nitrifier population inherent in soil, also affect nitrification.

**Soil Moisture**
Moisture content of the tilled soil layer fluctuates according to the balance between rainfall and evaporation. Under extremes of soil moisture, nitrification of NH₃ can be disrupted. Justice and Smith (1962) found that at low soil moisture levels (near wilting point) the initiation of nitrification was delayed and there was an increase in NO₂⁻ accumulation.
Nitrification is inhibited at high soil moisture levels. Parker and Larson (1962) found that the transition from favourable to unfavourably high moisture levels for nitrification was abrupt rather than gradual. They also suggested that during periods of high soil moisture, losses of NO$_3^-$ by leaching and denitrification might be more important in depressing the NO$_3^-$-N concentration of the soil than would inhibition of nitrification. Under anaerobic conditions resulting from waterlogging, nitrification of NH$_3$ ceases because of a lack of oxygen to fuel the oxidization reactions (Frederick and Broadbent 1966). In the studies by Justice and Smith (1962) and Parker and Larson (1962), extremes of temperature exacerbated the effects of extremes of soil moisture level to slow nitrification.

**Nitrification within an ammonium fertilizer application band**

Immediately following soil injection of an NH$_4^+$-fertilizer into soil, adsorption of the NH$_3$ produces an NH$_4^+$ concentration and pH gradient around the injection site. Nitrification begins at the outside edge of an NH$_3$ band where the NH$_3$ concentration and pH are favourable for the functioning of the nitrifying microorganisms (Eno et al. 1955). As the concentration of NH$_4^+$ and pH gradients decrease, the rate of nitrification increases towards the centre of the application band until the NH$_4^+$ is completely nitrified or a pH low enough to limit nitrification is reached.

In a black clay soil of the Darling Downs, Whitehouse and Leslie (1973) found that nitrification in the NH$_4^+$ band had begun between 1 and 7 days after application and that large amounts of NO$_3^-$ were present by 30 days. In the same study, pH measurements suggested that nitrification was largely completed by 30 days as the pH, under the influence of the buffering capacity of the soil, increased after 30 days. Up to 1,000 mg/kg of NO$_2^-$ and NO$_3^-$ had accumulated at the centre of the application band after 30 days. By 90 days significant amounts of NO$_3^-$ were found below the original application band due to regular surface watering after application. Downwards movement of NO$_3^-$ due to watering reduced the concentration of NO$_3^-$ and NO$_2^-$ to less than 500 mg/kg at the application centre.
2.7 AMMONIUM FERTILIZER USE, CROPS AND SOILS IN EASTERN AUSTRALIA

The History of Nitrogen Fertilizer Use in eastern Australia.

Improvement of cereal crop yields using N fertilizer was researched as early as the late 1920s and early 1930s in the southern states of Australia. The majority of this research was concentrated in South Australia and Victoria (Russell 1957). In their virgin state soils of the southern Region are inherently low in total N concentration (0.085 %) compared with other cereal growing areas of the world, and lower than the soils of north west New South Wales and southern inland Queensland (0.14 - 0.21 % total N) (Graham et al. 1981; Dalal and Mayer 1986).

In the temperate southern cereal and mixed cropping belt of Australia a rotational farming system involving the used of subterranean clover (*Trifolium subterraneum* L.) fertilized with superphosphate and grazed by sheep was developed during the 1920s. This rotation became an integral part of building the N fertility of soil before a cropping phase (Leeper 1970). During the 1950s, researchers were able to show increases in soil total N concentration from 0.085 % to 0.139 % over a period of years (Donald and Williams 1954). Several successive winter cereal crops were able to be grown without developing N deficiency after a 3 to 5 year period of legume-based pasture. However, the benefit from legume pasture rotations was not so pronounced on higher N fertility soils.

During the 1940s and 50s, extensive areas of grazing land in northwest New South Wales and southern Queensland were brought into cereal crop production. The first areas selected for cereal cropping were the deep fertile black clay soils of the Darling Downs, Queensland and the North Western Slopes of New South Wales. These soils, being inherently high in N in their virgin state, did not show any grain yield increase by the introduction of a legume in the rotation, as had been experienced in the south. By the 1960s researchers such as Holford (1980), Holford (1990) and Littler (1984)
showed grain yield and grain protein increases in wheat after lucerne (*Medicago sativa* L.) pasture, indicating that a decline in N fertility had occurred.

Martin and Cox (1956) showed that in the heavy clay Vertosols (Isbell 1996) of southern Queensland, during the first 25 years of cultivation, a loss rate of 0.8% per annum of the total N content had occurred from the top 0.15 m. A reduction in crop N uptake of 1.7 - 2.4 % per annum was recorded on similar soils of the same region by Dalal and Mayer (1986).

By 1962, Colwell and Esdaile (1966) showed that incipient N deficiency was affecting crops in many areas of NSW and by 1985 N deficiency was widespread (Doyle 1977; Holford and Doyle 1992). Hart (1962) and Leslie (1968) observed that N deficiency commonly occurred in crops grown on soils in southern Queensland continuously cropped for 50 - 80 years, the first sign being a reduction in the N concentration of the grain.

The rate of decline in N fertility escalated in the north-eastern cereal belt of eastern Australia due to the economic slump in the pastoral industries during the 1960s and 1970s when much grazing land was converted to continuous cereal cropping (Hamblin and Kyneur 1993). Littler (1961) showed that wheat profitability could be increased with the use of N fertilizer in land continuously cropped over long periods on the Darling Downs. He was able to produce yield increases of 29 - 69 % and grain protein increases of 1.8 to 2.2 % by applying 50 kg/ha of N as urea. Steady growth in N use on cereals occurred after the mid 1960s (Pulsford 1978).

The initial reaction of cereal growers to soil fertility decline was to fallow for a longer period and/or grow crops other than wheat, for which quality (protein %) standards were not as demanding. As a result barley and sorghum crop areas expanded rapidly during the 1960s and 1970s. By the late 1970s, N deficiency in barley and sorghum crops even on land that had been long fallowed led to increased use of N fertilizer.
In southern Australia, the higher relative profitability of cereal crops as compared with pastoral enterprises changed the relative importance of legume pastures during the 1960s and 1970s. The productivity of legume pastures was allowed to decline, the length of the legume phase was shortened and higher yields of cereal crops were sought. These factors, combined with better control of wheat root pathogens and weeds, led to an increase in the occurrence of N deficiency in winter cereals following a pasture phase. Nitrogen fertilizer consumption increased rapidly in southern Australia in response to the higher profits available with higher cropping frequency. The N fertilizer consumption rate further increased with rapid expansion of canola in the late 1980s (A. Good personal communication 1998). Growing canola provided the benefits of a winter break-crop between successive cereal crops previously only gained from a pasture phase (Colton and Sykes 1992).

Concern for declining protein concentration and saleability of wheat produced in southern Australia, and the reducing volume of Prime Hard quality wheat (premium quality white spring wheat with protein concentration of 13 % or greater) (Nicol 1994) and decrease in soil N fertility in the northern area, has also been a driving force behind the increasing use of N fertilizer in the region (Hamblin and Kyneur 1993; Angus and Pitson 1994).

**Soils**

In eastern Australia, the 2 nutrients that most commonly limit crop growth are P and N (Russell 1957; Dalal and Probert 1997). Urea is the most widely used straight N fertilizer and the ammonium phosphate fertilizers, MAP and DAP, are the most widely used phosphate fertilizers containing N used in cropping. Anhydrous ammonia use is applied to soils with silty-clay to heavy-clay textures. The chemical and physical characteristics of these soils make them suitable for injection of anhydrous ammonia, although a lack of NH$_3$ retention may be a problem in some wet seasons if anhydrous ammonia is applied to wet clay soil (Ward 1987). The characteristics of some major soil types found in the regions where NH$_4^+$-fertilizers are used are summarised in Table 2.7
Table 2.7 Selected characteristics of 3 soils used for broadacre cropping where NH₄⁺-fertilizers are used (Moody personal communication 1996).

<table>
<thead>
<tr>
<th>Soil Classification</th>
<th>Exchangeable (cmolc/kg)</th>
<th>Water Soluble (cmolc/kg)</th>
<th>pH</th>
<th>CEC</th>
<th>Ca (cmolc/kg)</th>
<th>Mg (cmolc/kg)</th>
<th>Clay %</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vertosol</td>
<td></td>
<td></td>
<td>6 - 9</td>
<td>30 - 80</td>
<td>14 - 40</td>
<td>5 - 30</td>
<td>0.8 – 2</td>
</tr>
<tr>
<td>Kandosol</td>
<td></td>
<td></td>
<td>4 - 6</td>
<td>1.5 - 7</td>
<td>1 - 7</td>
<td>0.1 - 5</td>
<td>0.1 -0.3</td>
</tr>
<tr>
<td>Ferrosol</td>
<td></td>
<td></td>
<td>4 - 6.5</td>
<td>8 - 15</td>
<td>2 - 12</td>
<td>2 - 12</td>
<td>0.2 - 0.6</td>
</tr>
</tbody>
</table>


Crops and Nitrogen Fertilizer Use

Nitrogen fertilizer is applied to a large variety of crop species grown in eastern Australia, including winter and summer cereals, oilseed and fibre crops. The areas of crops and average N fertilizer products used are summarised in Tables 2.8 and 2.9 respectively. As a result of the moisture holding capacity of the clay soils and the rainfall pattern in northern New South Wales and Queensland, both summer and winter cereal, oilseed and fibre crops can be grown across the majority of the area. Winter cereals, grain legumes and oilseeds dominating in the southern more temperate climate. Nitrogen fertilizer has become an essential input for winter and summer cereals in dryland northern cropping systems (Holford 1990; Holford and Doyle 1992; Dalal and Probert 1997).

In some years favourable seasons allow opportunity cropping (3 crops in 2 years) in the northern summer cropping area. The increase of frequency of cropping has further increased and rate of supplementary N applied to dryland cereal crops. Increased cropping frequency combined with a general reduction in mechanical cultivation has reduced the opportunities to apply N fertilizer with a tillage event. Similarly, the dry summer period in southern Australia often prevents effective pre-sowing application of N fertilizer because of very dry soil conditions.
Table 2.8  Crop areas of major N consuming crops (1992-93) (ABARE, 1993).

<table>
<thead>
<tr>
<th>Crop</th>
<th>NSW</th>
<th>VIC</th>
<th>QLD</th>
<th>WA</th>
<th>SA</th>
<th>TAS</th>
</tr>
</thead>
<tbody>
<tr>
<td>barley</td>
<td>552</td>
<td>535</td>
<td>183</td>
<td>605</td>
<td>1014</td>
<td>12</td>
</tr>
<tr>
<td>canola</td>
<td>73</td>
<td>18</td>
<td>-</td>
<td>11</td>
<td>6</td>
<td>-</td>
</tr>
<tr>
<td>chickpea</td>
<td>30</td>
<td>70</td>
<td>28</td>
<td>1</td>
<td>16</td>
<td>-</td>
</tr>
<tr>
<td>cotton</td>
<td>175</td>
<td>-</td>
<td>87</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>sorghum</td>
<td>113</td>
<td>1</td>
<td>319</td>
<td>1</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>sunflower</td>
<td>23</td>
<td>1</td>
<td>34</td>
<td>-</td>
<td>1</td>
<td>-</td>
</tr>
<tr>
<td>wheat</td>
<td>1800</td>
<td>950</td>
<td>800</td>
<td>4000</td>
<td>1550</td>
<td>1</td>
</tr>
<tr>
<td>Total</td>
<td>2766</td>
<td>1575</td>
<td>1451</td>
<td>4618</td>
<td>2587</td>
<td>13</td>
</tr>
</tbody>
</table>

Where N fertilizers were traditionally applied pre-sowing, usually during a tillage event with conventional cultivation practices. Nitrogen fertilizer application has now moved closer to sowing and there is a growing trend toward application of N fertilizers at sowing as indicated by the number of sowing equipment manufacturers advertising attachments for N fertilizer placement at sowing.

Table 2.9  Fertilizer N consumption for cereal, oilseed and fibre crops in Australia 1992-93 (Incitec Fertilizers, personal communication 1996).

<table>
<thead>
<tr>
<th>(000 tonnes)</th>
<th>NSW</th>
<th>VIC</th>
<th>QLD</th>
<th>WA</th>
<th>SA</th>
<th>TAS</th>
</tr>
</thead>
<tbody>
<tr>
<td>elemental N</td>
<td>94</td>
<td>20</td>
<td>74</td>
<td>130</td>
<td>13</td>
<td>1</td>
</tr>
<tr>
<td>ammonium phosphate</td>
<td>120</td>
<td>96</td>
<td>25</td>
<td>430</td>
<td>138</td>
<td>8</td>
</tr>
<tr>
<td>Total</td>
<td>214</td>
<td>116</td>
<td>99</td>
<td>560</td>
<td>151</td>
<td>9</td>
</tr>
</tbody>
</table>

1. elemental N is the cumulative amount of N contained in the various N containing fertilizers

For summer crops that are grown on wide row spacing (60 to 100 cm), N fertilizer application at sowing presents few problems. Extra tines can be added between seed sowing tines to separate the placement of the fertilizer and seed. For winter crops with narrower row spacings, 150 to 250 mm, seed and fertilizer placed down separate tines results in small separation distances and likely disturbance of the seed by the fertilizing tine.
Chapter 3

General Materials and Methods

This chapter contains information about 11 experiments, the results of which are reported in Chapters 4 to 7. Four experiments were conducted in laboratory or greenhouse conditions, the remaining 7 were field experiments. Common to the field experiments were crops, sites, soil sampling and analytical methods and application equipment for which details are presented in this chapter. Materials and methods specific to an experiment and modifications of standard practices presented in this chapter are described in detail in the materials and methods of the relevant chapter.

Throughout this thesis, references are made to germination, emergence and establishment. These terms refer to distinctly different phases of establishment of a population of seedlings. In these experiments, germination is defined as the processes up to and including early radicle and coleoptile extension but before emergence above the soil surface (Collis-George 1987). Establishment was where multiple seedling counts were taken and the final population reflected seedling survival (Wood 1987). In experiments where seedlings were counted once only, the term ‘emergence’ has been used.

3.1 SITES AND SOILS

Experiments 4-1, 4-2, 6-3 and 7-1 were conducted at the University of Queensland, Gatton College, Field Research Station at Lawes, Queensland (152° 20'E, 27° 36'S). The soil is a Vertosol, of the Blenheim series (Powell 1982). Soil in the experimental area has a uniform profile of medium clay with an organic carbon content of 1.3 % and a cation exchange capacity of 31 to 37 cmolc/kg. Soil pH for Experiment 4-1 was 7.7 and pH 6.6 for other experiments. High soil pH, electrical conductivity and coarser tilth for Experiment 4-1 were associated with a higher % of cations as sodium (4 % vs 1.9%) and a lower calcium to magnesium ratio (1.06 vs 1.5) compared to those of the nearby site.
for Experiment 4-2. Surface soil (0-10 cm) was gathered from around the site of Experiment 4-2 and used for the pot Experiment 6-2.

Experiment 7-3 was established on a Vertosol, a Mywybilla soil of the Jimbour series (Vandersee 1975) on the property ‘Colonsay’, Formartin, Queensland (151°30’ E, 27°18’S). Surface soil, 0-10 cm, gathered from Formartin was also packed into trays (7.5 cm deep) for Experiment 6-1. More detailed soil characteristics are contained in Table 3.1.

Table 3.1 Selected soil chemical and physical characteristics of Lawes, Formartin and Dalby soils.

<table>
<thead>
<tr>
<th>Soil Parameter</th>
<th>Lawes</th>
<th>Formartin</th>
<th>Dalby</th>
</tr>
</thead>
<tbody>
<tr>
<td>Colour</td>
<td>black</td>
<td>black</td>
<td>grey</td>
</tr>
<tr>
<td>Texture</td>
<td>clay</td>
<td>clay</td>
<td>clay</td>
</tr>
<tr>
<td>pH (1:5 water)</td>
<td>6.6 (7.7)</td>
<td>8.4</td>
<td>8.5</td>
</tr>
<tr>
<td>Nitrate - N (1:5 water) (mgkg)</td>
<td>10 - 78</td>
<td>2 - 15</td>
<td>5 - 7</td>
</tr>
<tr>
<td>Ammonium - N (KCl) (mgkg)</td>
<td>5 -13</td>
<td>1 - 4</td>
<td>2 - 5</td>
</tr>
<tr>
<td>Buffer pH (Melich)</td>
<td>6.4</td>
<td>6.6</td>
<td>6.6</td>
</tr>
<tr>
<td>Phosphorus (Colwell) (mgkg)</td>
<td>150</td>
<td>34</td>
<td>15</td>
</tr>
<tr>
<td>Organic Carbon % (Walkley-Black)</td>
<td>1.3</td>
<td>1.1</td>
<td>0.9</td>
</tr>
<tr>
<td>Cation Exchange Capacity (cmol/kg)</td>
<td>31-37</td>
<td>64</td>
<td>45</td>
</tr>
<tr>
<td>Electrical Conductivity se (dS/m)</td>
<td>0.6-2.1</td>
<td>0.6</td>
<td>1.2</td>
</tr>
<tr>
<td>Soil Classification</td>
<td>Ug 5.12</td>
<td>Ug 5.15</td>
<td>Ug 5.41</td>
</tr>
</tbody>
</table>


Experiment 7-2 was conducted at Dalby, Queensland (150°59’ E, 27°06’S). The soil is a grey Vertosol of the Cecilvale series (Vandersee 1975).

3.2 SOWING AND FERTILIZER APPLICATION EQUIPMENT

Sowing equipment consisted of two 305 mm diameter belted cone seed meters manufactured by Kimseed Machinery, WA. The cone seeders were calibrated to meter a predetermined number of seeds and weight of fertilizer material to each sowing tine. A rotary divider driven by an electric motor was used to divide the seed and fertilizer mixture falling from each cone to sowing tines fitted with 100 mm wide points manufactured by Gyral Implements, Toowoomba,
Queensland. The number of tines and row spacing depended on the requirements of the particular experiment but was generally seven 225 mm rows for winter crop species and 2 or four 450 mm rows for summer species. Detailed tine set-up is described in the materials and methods section of the relevant chapter. Each sowing tine was followed by a 100 mm wide pneumatic press wheel with an axle pressure of approximately 30 kg, to firm the soil over the seed row (Mills and McIntyre 1997).

### 3.3 CROP SPECIES

Eleven crop species including maize, sorghum (*Sorghum bicolor* (L.) Moench), panicum (*Setaria italica* L.), wheat, barley, canary (*Phalaris canariensis* L.), chickpea (*Cicer arietinum* L.), sunflower (*Helianthus annuus* L.), canola (*Brassica napus* L. var. *napus*), cotton (*Gossypium hirsutum* L.) and soybean were used in the experiments. For field experiments, species were selected to suit the seasonal conditions to which they were adapted (summer or winter). Details of the species used in particular experiments are contained in materials and methods for each chapter.

### 3.4 SOIL SAMPLING

Two sampling techniques were used where soil samples were taken from fertilizer application bands. For experiments 4-1 and 4-2, soil samples comprising 20, 25 mm diameter cores (approximately 1 kg field moist) were taken from horizontally along the seed-line in each main plot treatment, 24 h after anhydrous ammonia application.

For Experiments 6-1 and 7-3 soil was collected by pushing a 25 mm diameter tube vertically into the planting row. A 25 mm length section of each core, above and below the seed depth, was retained for analysis.

Field moist soil samples were cooled immediately in an insulated box containing ice and transferred to the laboratory. Samples were either prepared for extraction within 4 hours (Experiments 4-1 and 4-2) or frozen
until after the final sequential sampling for Experiments 6-1 and 7-3 to enable all samples to be processed using a single batch of extractant.

3.5 SAMPLE PREPARATION AND ANALYSIS

Soil was thoroughly mixed before sub-sampling for analysis for mineral N concentration.

For all experiments where mineral-N was measured, a sub-sample of moist soil was retained for determining soil water content and pH. One hundred to 150 g of field moist soil was placed in 300 mL of 2 M KCl solution, tumbled end over end for 1 hour and filtered to extract NH$_4^+$-N, nitrite-N (NO$_2^-$-N) and nitrate-N (NO$_3^-$-N) while minimising denitrification and mineralisation in the wet samples. Soil moisture content was determined by drying at 105 °C to constant weight. Soil moisture was used to adjust the soil concentrations of NH$_4^+$-N, NO$_3^-$-N and NO$_2^-$-N by adjusting the extractant solution volume and the weight of dry soil extracted (Rayment and Higginson 1992).

For Experiments 4-1 and 4-2, duplicate soil samples (100 to 150 g) were dried for 96 h at 42 °C in a standard drying oven (air dried), ground to < 2 mm for N analysis and a 10 g sub-sample extracted with 100 mL 2 M KCl solution (Rayment and Higginson 1992).

The concentrations of NH$_4^+$-N, NO$_2^-$-N and NO$_3^-$-N for both field moist and air dried preparation procedures were expressed on a dry soil basis (mg/kg).

Soil pH in the fertilizer NH$_4^+$ band was also determined for use in estimating the soil NH$_3$ concentration. Soil pH was measured according to the method of Bruce and Rayment (1982), NO$_2^-$-N and NO$_3^-$-N were determined by and automated hydrazine reduction procedure for the Griess-Ilosvay reaction for NO$_2^-$-N (Best, 1976) and NH$_4^+$-N was measured by an automated procedure using a modified Berthelot indophenol reaction with alkaline sodium salicylate/nitroprusside/isocyanurate (Crooke and Simpson 1971). Electrical
conductivity was measured according to the method of Rayment and Higginson (1992).

For Experiment 7-3, soil urea concentration was measured according to the method of Bremner (1982).

3.6 DATA ANALYSIS
The Genstat for Windows program (The Numerical Algorithms Group Ltd 1996) was used for most statistical analyses. Analysis of variance was performed and comparison of treatment values was done by least significant difference (l.s.d.) when F-values were significant ($P<0.05$).

To assess crop species tolerance to NH$_3$ toxicity and osmotic pressure as distinct from fertilizer product response (Experiments 6-1 and 6-3) combinations of products and product rates were converted to NH$_4^+$-N rates and relative osmotic index (ROI). Relative osmotic index, calculated by multiplying the salt index (Rader et al. 1943) of the fertilizer product by its application rate (g/m), is an index developed for this thesis to enable comparison of crop establishment damage caused by fertilizer products and blends of products having different salt indices and applied at different rates.

For all field experiments, establishment data was collected on 6 separate occasions over a maximum of 22 days. Data from each observation time were analysed using ANOVA. Data presented is only for the earliest observations where establishment for all species had reach a maximum. This usually corresponded with the time that displayed the lowest coefficient of variability. Some species, with later observations, displayed reduced establishment after they emerged contributing to an increase in variability.

3.7 AMMONIA-AMMONIUM BALANCE IN SOLUTION
Theory
In most biological fluids, NH$_3$ exists in two forms, ionised NH$_4^+$ and the toxic non-ionised NH$_3$ (Subcommittee on Ammonia 1979a). The pH and
temperature of the solution determine the relative proportion of NH₃ to NH₄⁺.

The equilibrium between NH₄⁺ and NH₃ in solution may be represented by

\[ \text{NH}_4^+ \leftrightarrow \text{NH}_3 + H^+ \]

where

\[ \text{Ka} = \frac{[\text{NH}_3][H^+]}{[\text{NH}_4^+]} \]

Solution pH has a large influence on the NH₃-NH₄⁺ equilibrium. and the equation for this effect can be derived by rearranging and transforming the equation above.

\[ \log_{10}[H^+] - \log_{10}\text{Ka} = \log_{10}\frac{[\text{NH}_4^+]}{[\text{NH}_3]} \]

therefore

\[ \text{pKa} - \text{pH} = \log_{10}\frac{[\text{NH}_4^+]}{[\text{NH}_3]} \]

The value for pKa varies with temperature and can be calculated from the equation of Emmerson et al. (1975)

\[ \text{pKa} = 0.09018 + \frac{2729.92}{T} \]

where T is the absolute temperature (K).

The following equilibrium of the reaction is described by Henry’s Law.

NH₃ (gas in solution) ⇔ NH₃ (gas in air adjacent to the solution surface)

i.e. \[ \frac{[\text{NH}_3\text{(aq.) in solution}]}{[\text{NH}_3\text{ gaseous}]} = \text{constant H at a given temperature} \]

In the equation, NH₃ concentration in the gas (NH₃(g.)) and solution (NH₃(aq.)) phases are molar and H is Henry’s constant. The Henry constant for NH₃ varies markedly with temperature (K) according to the relationship ascribed by the Subcommittee on Ammonia (1979).

\[ \log_{10}H = \frac{1477.7 - 1.6937}{T} \]
The concentration of NH$_3$ in the gaseous (NH$_3$(g.)) phase in equilibrium with
NH$_3$ in the liquid phase (NH$_3$(aq.)) can be calculated as follows

$$[\text{NH}_3(\text{g.})] = \frac{[\text{NH}_3(\text{aq.})]}{H}$$

substituting H from equation 3

$$[\text{NH}_3(\text{g.})] = \frac{[\text{NH}_3(\text{aq.})]}{10^{1477.7/T - 1.6937}}$$

It is difficult to directly measure NH$_3$(g.) concentration in the atmosphere in
the head-space above an NH$_4$OH solution and in soil, however in a steady
state system (closed container) and in soil, NH$_3$ concentration may be
estimated from the solution or soil NH$_4^+$ concentration and the solution or soil
pH (Freney et al. 1983).

Concentrations of NH$_3$-NH$_4^+$ species in containment vessels for Experiments
5-1, 5-2 and 5-3 were calculated using equation 4.
Chapter 4

Tolerance of 9 crop species to ammonium fertilizers applied at sowing.

Summary

A trend for application of higher rates of N fertilizer at sowing has increased to present day primarily influenced by changes in tillage practice, fertilizer products, increased cropping frequency and profitability of farming. This trend has increased the need for better knowledge of how crop species respond to NH$_4^+$-fertilizers applied in the seed furrow yet avoid significant crop establishment damage.

Seeds of 9 crop species were exposed to rates of soil NH$_3$ by combinations of anhydrous ammonia application rate and seed-fertilizer band separation distance. Establishment tolerance differences between crop species occurred over a narrow range of soil NH$_4^+$-N concentrations, but species separated into three distinct tolerance groups. Species such as cotton and sunflower (low tolerance group) were able to tolerate only 15 to 25 % of the NH$_4^+$-N concentration tolerated by the most tolerant group, barley, maize and wheat. An intermediate group, sorghum, chickpea and canary tolerated 50 % of the NH$_4^+$-N concentration of the most tolerant group.

4.1 INTRODUCTION

Commercially, fertilizer placement with seed of grain crops at sowing has been a long accepted practice in Australia, even though there is general recognition of possible detrimental effects of the fertilizer on germinating seed (Carter 1967; Mason 1971). Fertilizer placement with seed originated from the need to band phosphate fertilizer with the seed at sowing. This was identified as the most effective application strategy for phosphate application for cereal crops by Stanford and Pierre (1953), Olson and Dreier (1956), Welch et al. (1966), Rudd and Barrow (1973) and Yao and Barber (1986).

In Australia, a trend away from superphosphate to ammonium phosphate fertilizers (MAP and DAP) in cereal cropping began in the 1970s. This trend developed in response to increased availability of ammonium phosphate products, higher concentration of P in ammonium phosphates, and superior
physical attributes for use with sowing equipment. A low requirement for S in most cereal crops and a lower cost per unit of P also encouraged the use of ammonium phosphates. During this time, at average application rates of 40 to 100 kg/ha for ammonium phosphates at standard 180 mm row spacings for broadacre sowing, fertilizer band concentrations of $\text{NH}_4^+$-N were 0.07 to 0.32 g/m. This is below that concentration, 0.36 to 0.45 g/m N (20 to 25 kg/ha N), that was found to cause damage to cereal crops of eastern Australia (Carter 1967).

The trend towards the application of higher N fertilizer rates at sowing has increased to the present day. This trend has been influenced by changes in tillage practices, increased cropping frequency and the desire to delay the expenditure on N fertilizer until the last opportunity in the season. In the northern part of the eastern Australian cereal belt, the last N application opportunity is frequently at sowing, topdressing after sowing usually being unprofitable due to the unreliability of in-crop rainfall (Strong 1982; Doyle and Shapland 1994). These influences have resulted in the N contained in ammonium phosphates being supplemented by blending with urea or by the application of urea or anhydrous ammonia at sowing, as promoted by Robotham (1993).

During the transition from the use of ammonium phosphates to urea blends or straight urea at sowing, the commercially accepted maximum N rate of 25 kg/ha was recommended for the new products, ignoring findings of Carter (1967) and Mason (1971). Based on their research both Carter (1967) and Mason (1971) suggested that urea should not be applied with seed at sowing. Accepted maximum N rates were originally derived for the use of ammonium sulfate, calcium ammonium nitrate and ammonium nitrate.

Changes in fertilizer products since the 1970s have been paralleled by changes in crop species, sowing equipment, row spacing and soil opener type. Changes to fertilizer products, crop species and sowing equipment has increased the need to gain a better understanding of the underlying principles
and basic processes that influence the interaction between seed and NH$_4^+$-N fertilizer during crop establishment.

This experiment was conducted to investigate crop species susceptibility to varying concentrations of NH$_4^+$-N in the seed furrow at sowing. Species tested included most of the commercially important crops in eastern Australia using a selection of cereals, oilseeds and a grain legume.

4.2 MATERIALS AND METHODS

Site
Separate experiments were conducted for winter (Experiment 4-1) and summer (Experiment 4-2) crops at the University of Queensland, Gatton College, Field Research Station at Lawes, Queensland (152° 20'E, 27° 36'S). A pasture composed mainly of native grasses, some broadleaf weeds and Johnson grass (Sorghum halpense L.), preceded the winter crops and a broccoli (Brassica oleracea L. convar. botrytis var. italica Plenck) crop preceded the summer crops (Table 4.1). Different cropping histories, soil chemistry and subsequent soil cultivation practices provided a finer soil tilth for the summer crops than for the winter crops.

More details of the site and soil are contained in the General Materials and Methods, Chapter 3.

Fertilizer Application
Anhydrous ammonia was applied in the 2 studies using the implement described in Chapter 3, the application points were modified to apply NH$_3$ as detailed by Robotham (1993).

The experimental planter was configured to apply anhydrous ammonia down 2 sowing tines spaced 225 mm apart (Plate 4.1). Two adjacent and identically configured tines sowed seeds, but no anhydrous ammonia at the same depth as the adjacent tines. Altering the regulating meter setting (orifice aperture), while keeping a constant ground speed varied anhydrous ammonia application rate. Additional non-sowing tines were attached to the experimental planter to vent anhydrous ammonia outside the 4 rows of each plot. Without venting at
low application rates, the anhydrous ammonia-regulating meter was below its accurate range for uniform application. Increasing the number of tines increased the NH\textsubscript{3} flow rate through the meter to allow uniform application while maintaining a slower tractor speed, more appropriate for good seed placement.

Plate 4.1 Experimental planter and tractor used for sowing Experiments 4-1 and 4-2.

Anhydrous ammonia application rate and distance between the point of NH\textsubscript{3} release and seed placement, either 25 or 50 mm, were varied using application points developed by Robotham (1993), to obtain gradients of soil NH\textsubscript{3}-N concentration around the seed. Seed was placed along a line that tracked immediately behind the point of the tine while anhydrous ammonia was placed in bands either side of the seed row (Plate 4.2).
Plate 4.2  Tine from planter configured to apply ammonia and sow seed in Experiment 4-1 and 4-2.

During anhydrous ammonia application, product flow was continued 5 m beyond the seeded plot area where a quantity of inert blue plastic granules was substituted for seeds. After cutting a face in the soil across the direction of the seed and fertilizer rows to a depth of 250 mm, a pH-sensitive dye solution was sprayed onto the soil cutting, to visibly locate the soil NH$_4^+$ bands. Soil was recovered from the seed-line beyond the seeded portion of the plot by pushing a 25 mm diameter corer centred along the visible line of blue plastic granules midway between the anhydrous ammonia application bands. Accuracy of sample collection was confirmed by the presence of plastic granules along the centre of the core sample. The plot area for seedling emergence counts was therefore not disturbed.
Treatments
Treatments consisted of factorial combinations of anhydrous ammonia applied at 0, 50, 100, 200 or 300 kg/ha of N (0, 1.13, 2.25, 4.5, 6.75 g NH₃-N/m of row), 25 mm and 50 mm separation distances of seed and anhydrous ammonia bands and 9 crop species, maize (cv. Pioneer C84), cotton (cv. DP90), wheat (cv. Cunningham), barley (cv. Tallon), chickpea (cv. Amethyst), sorghum (cv. DK44), canary (cv. Moroccan), canola (cv. Hyola 40) and sunflower (cv. Hysun 44).

Plant Population
Sown seed populations were adjusted for the expected germination % for each seed species in accordance with the desired population range recommended for commercial sowing of the crop species (Table 4.1) (Mills et al. 1996; Mills and McIntyre 1997).

Table 4.1  Seeding rate and target plant population (per m row) for winter crops (Experiment 4-1) and summer crops (Experiment 4-2).

<table>
<thead>
<tr>
<th>Species</th>
<th>Seed sown (seeds per metre row)</th>
<th>Target population (plants per metre)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Experiment 4-1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>barley</td>
<td>20</td>
<td>15</td>
</tr>
<tr>
<td>canary</td>
<td>30</td>
<td>15</td>
</tr>
<tr>
<td>canola</td>
<td>20</td>
<td>12</td>
</tr>
<tr>
<td>chickpea</td>
<td>14</td>
<td>12</td>
</tr>
<tr>
<td>wheat</td>
<td>20</td>
<td>15</td>
</tr>
<tr>
<td>Experiment 4-2</td>
<td></td>
<td></td>
</tr>
<tr>
<td>cotton</td>
<td>16</td>
<td>12</td>
</tr>
<tr>
<td>maize</td>
<td>12</td>
<td>6</td>
</tr>
<tr>
<td>sorghum</td>
<td>12</td>
<td>8</td>
</tr>
<tr>
<td>sunflower</td>
<td>11</td>
<td>5</td>
</tr>
</tbody>
</table>

Seedlings were counted at a 2 to 3 day interval for 19 days after sowing (DAS) at which time establishment had been maximised.

Experimental Design and Layout
Experiments were conducted with a randomised complete block design of sown plots, 5 m long. Main plots were combinations of anhydrous ammonia rate by
separation distance between the seed and the anhydrous ammonia bands, the subplots being the crop species.

**Data Analysis**

Effects of anhydrous ammonia rate and seed to anhydrous ammonia band separation distance on species establishment were analysed using ANOVA. Significant treatment means were separated by a l.s.d. test ($P<0.05$).

To determine the relationship between soil $\text{NH}_4^+$-$\text{N}$ and species establishment response, linear regression models were fitted to transformed data for each species across $\text{NH}_3$-$\text{N}$ rates, assuming common slopes and different intercepts. Regression intercepts were used to group species according to a l.s.d. test ($P=0.05$). Transformation of the data consisted of converting establishment (plants/m) to a relative scale by dividing results for the corresponding species by the establishment for zero control and expressing as a percentage. Transformation was performed to allow comparisons between species with vastly different seeding rates.

The $\text{NH}_4^+$-$\text{N}$ concentration (toxic concentration) required to reduce plant establishment to 50 % of that of the zero control, $\text{TC}_{\text{NH}_4^{50}}$, was derived from the crop establishment and soil $\text{NH}_4^+$-$\text{N}$ concentration regression equation for each crop species.

**4.3 RESULTS**

*Soil Ammonium-N*

**Sample Preparation**

Soils from Experiment 4-1 were processed using both field moist and air dried procedures in order to determine the more appropriate soil handling method to determine $\text{NH}_4^+$-$\text{N}$ concentration of soil recently fertilized with N. Soil extraction using the field moist procedure gave significantly ($P=0.05$) higher soil $\text{NH}_4^+$-$\text{N}$ concentrations than those obtained using the air dried procedure (Table 4.2). As the anhydrous ammonia application rate was increased, the soil $\text{NH}_4^+$-$\text{N}$ concentration increased with both methods of analysis, but the interaction between analysis method and anhydrous application rate was not significant.
These results suggest that drying of soil taken from bands of recently applied anhydrous ammonia with high concentrations (>1000 mg/kg) of NH$_4^+$-N may result in significant NH$_3$ loss prior to analysis but at lower soil NH$_4^+$-N, differences between methods appeared much smaller (Table 4.2).

**Table 4.2** The effect of soil extraction method and anhydrous ammonia application rate on soil NH$_4^+$-N concentration and apparent loss of soil NH$_4^+$-N, Experiment 4-1.

<table>
<thead>
<tr>
<th>Anhydrous ammonia rate (g/m N)</th>
<th>Preparation Procedure</th>
<th>Soil NH$_4^+$-N (mg/kg)</th>
<th>Loss on drying</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>air dried</td>
<td>field moist</td>
<td>mean</td>
</tr>
<tr>
<td>0</td>
<td>14</td>
<td>9</td>
<td>12</td>
</tr>
<tr>
<td>1.13</td>
<td>243</td>
<td>278</td>
<td>261</td>
</tr>
<tr>
<td>2.25</td>
<td>340</td>
<td>418</td>
<td>379</td>
</tr>
<tr>
<td>4.5</td>
<td>678</td>
<td>1018</td>
<td>848</td>
</tr>
<tr>
<td>6.75</td>
<td>765</td>
<td>1107</td>
<td>936</td>
</tr>
<tr>
<td>mean</td>
<td>409</td>
<td>602</td>
<td>193</td>
</tr>
</tbody>
</table>

I.s.d. (P=0.05)

- rate: 301
- procedure: 191
- rate x procedure: ns

At the two highest anhydrous ammonia rates, there was a plateauing of soil NH$_4^+$-N concentration. Air-drying lost proportionally more NH$_4^+$-N during processing than the field moist extraction procedure at these high NH$_3$-N application rates.

Soil NH$_4^+$-N concentrations quoted for both Experiments 4-1 and 4-2 were subsequently determined by field moist analysis and were used in regression analysis and in the calculations of specie’s susceptibility to NH$_3$. 
**Seed Zone Mineral-N**

**Ammonium-N - Experiment 4-1**

As the anhydrous ammonia application rate was increased, soil NH$_4^+$-N concentration in the seed zone increased from 9 to 1156 mg/kg (Fig. 4.1). The maximum soil NH$_4^+$-N concentration, 1156 mg/kg, was measured for 6.75 g/m of anhydrous ammonia-N and a 50 mm seed-NH$_4^+$-N band separation distance. The maximum concentration for the 25 mm separation distance was similar, 1123 mg/kg.

![Graph showing soil NH$_4^+$-N concentration vs. NH$_3$-N rate](image)

**Fig. 4.1** Soil NH$_4^+$-N concentrations (mg/kg) in a 25 mm diameter soil core extracted from the seed-line between application bands. The anhydrous ammonia was applied at 4 rates and 25 (●) and 50 (■) mm seed-anhydrous ammonia separation distances, for Experiment 4-1. The vertical bar is l.s.d ($P=0.05$) for the NH$_3$-N rate - separation distance interaction effect.

Increasing the seed-anhydrous ammonia band separation did not significantly reduce the soil NH$_4^+$-N concentration although the concentration for the 25 mm separation was generally higher than for 50 mm.

**Nitrite-N - Experiment 4-1**

Nitrite-N concentration near the seed was less than 2 mg/kg for all treatments. There were no significant effects of anhydrous ammonia application rate, seed-
anhydrous ammonia band spacing or extraction procedure on NO$_2$-N concentration (data not presented).

**Ammonium-N - Experiment 4-2**

The concentration of soil NH$_4^+$-N increased significantly ($P<0.05$) as anhydrous ammonia application rate was increased (Fig. 4.2).

Soil NH$_4^+$-N concentration for the highest anhydrous ammonia rate with a 50 mm seed-to-fertilizer separation distance was lower than the concentration for the lowest anhydrous ammonia rate with a 25 mm separation distance (Fig. 4.2).

![Fig. 4.2](image)

**Fig. 4.2**  Soil NH$_4^+$-N concentrations (mg/kg) in a 25 mm diameter soil core extracted from the seed line between application bands, 1 DAS. The anhydrous ammonia was applied at 4 rates and 25 (●) and 50 (■) mm seed-anhydrous ammonia band spacings, for Experiment 4-2. The vertical bar is l.s.d. ($P=0.05$) for the NH$_3$-N rate-separation distance interaction effects.

Placing anhydrous ammonia at a greater distance from the seed significantly ($P<0.05$) reduced the soil NH$_4^+$-N concentration at all anhydrous ammonia application rates. Different soil conditions for the 2 experiments appears to be the reason why the greater separation distance (50 mm) between seed and
fertilizer failed to reduce the NH$_4^+$-N concentration in the seed row in Experiment 4-1.

The quantity of NH$_4^+$-N found in the seed zone varied with seed-fertilizer separation distance and anhydrous ammonia application rate. In Experiment 4-1 where surface sealing of soil above the seed-line was not effective there was no difference in seed zone retention for the 25 and 50 mm separation distances (Table 4.3). For Experiment 4-2, where soil tilth was finer, the higher proportions of applied NH$_4^+$-N occurred in the seed zone with the 25 mm separation distance.

Table 4.3 Percentage of applied anhydrous ammonia found as soil NH$_4^+$-N in a 25 mm diameter core around the seed.

<table>
<thead>
<tr>
<th>NH$_3$-N Rate (g/m)</th>
<th>Expt 4-1</th>
<th>Expt 4-2</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>mean</td>
<td>25 mm (%)</td>
</tr>
<tr>
<td>1.13</td>
<td>6.8</td>
<td>8.2</td>
</tr>
<tr>
<td>2.25</td>
<td>5.1</td>
<td>8.1</td>
</tr>
<tr>
<td>4.5</td>
<td>6.4</td>
<td>6.1</td>
</tr>
<tr>
<td>6.75</td>
<td>4.5</td>
<td>4.0</td>
</tr>
</tbody>
</table>

l.s.d (P=0.05) Rate 0.25 0.77 Separation ns 0.48

**Crop Establishment - Experiment 4-1**

Seedling establishment in the zero control plots of most winter crops was low compared with commercially acceptable populations, although the appropriate number of seeds was sown (Table 4.1). Establishment in the control treatments averaged over separation distance were barley 7.8 plants/m (52% of target establishment), canary 7.5 plants/m (50 % of target), canola 5.1 plants/m (42 % of target), chickpeas 9.75 plants/m (81 % of target), and wheat 8.1 plants/m (54 % of target). It was hypothesised that the coarse seedbed tilth related to the soil type and soil preparation prior to sowing was the cause for consistently poor establishment.
As the anhydrous ammonia application rate was increased, seedling establishment declined significantly \((P<0.001)\). At the 2 highest anhydrous ammonia application rates establishment of most species was reduced by more than 50\% (Fig. 4.3).

![Graph showing plant establishment vs NH3-N rate](image)

**Fig. 4.3** Establishment for barley (○), canary (▲), canola (□), chickpea (■) and wheat (●) 19 DAS from the application of 5 rates of anhydrous ammonia at sowing, Experiment 4-1. Means of 25 and 50 mm separation distance. Vertical bar indicates l.s.d. \((P=0.05)\) for anhydrous ammonia rate.

The separation distance of the anhydrous ammonia band and seed at any observation did not significantly affect establishment. Lack of a response to separation distance suggests that seed was exposed to similar anhydrous ammonia concentrations at both 25 and 50 mm separation distances.

Due to vastly different plant populations in the control treatment for different crops, species susceptibility to soil NH\(_3\) was assessed by calculating the reduction in establishment for each anhydrous ammonia rate relative to the zero control treatment. Species were thus separated based on relative reduction in establishment (Table 4.4).
Crop species susceptibility to soil NH$_3$ was significant ($P<0.001$). Canola with 51.1 % relative reduction in establishment was the most susceptible species followed by chickpea (42.4 %), canary (38.4%). Barley and wheat similarly tolerant and significantly ($P<0.05$) more tolerant of soil NH$_3$ than the other species tested with relative reduction in establishment of 30.3 % and 32.7 %.

Table 4.4 Relative reduction in establishment (%) for barley, canary, canola, chickpea and wheat 19 DAS from 5 rates of anhydrous ammonia applied at sowing, 25 mm or 50 mm to both sides of the seed, Experiment 4-1.

<table>
<thead>
<tr>
<th>NH$_4^+$-N rate (g/m)</th>
<th>Crop species</th>
<th>barley</th>
<th>canary</th>
<th>canola</th>
<th>chickpea</th>
<th>wheat</th>
<th>mean</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td></td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>1.13</td>
<td></td>
<td>0</td>
<td>0</td>
<td>16.0</td>
<td>10.7</td>
<td>1.0</td>
<td>5.3</td>
</tr>
<tr>
<td>2.25</td>
<td></td>
<td>3.6</td>
<td>11.7</td>
<td>56.1</td>
<td>11.0</td>
<td>1.0</td>
<td>16.7</td>
</tr>
<tr>
<td>4.5</td>
<td></td>
<td>50.6</td>
<td>66.7</td>
<td>89.0</td>
<td>65.5</td>
<td>55.3</td>
<td>65.4</td>
</tr>
<tr>
<td>6.75</td>
<td></td>
<td>67.1</td>
<td>75.1</td>
<td>94.4</td>
<td>82.5</td>
<td>73.7</td>
<td>78.6</td>
</tr>
<tr>
<td>mean</td>
<td></td>
<td>30.3</td>
<td>38.4</td>
<td>51.1</td>
<td>42.4</td>
<td>32.7</td>
<td></td>
</tr>
</tbody>
</table>

l.s.d. ($P=0.05$)
rate 15.9
species 5.7
rate x species 21.5

**Crop Establishment - Experiment 4-2**

Soil conditions for Experiment 4-2 (summer crops) were apparently more favourable for seedling establishment than for the Experiment 4-1 (winter). Seedling populations in zero control plots averaged over separation distance were cotton 9.6 plants/m (80 % of target establishment), maize 8.6 plants/m (143 %), sorghum 10 plants/m (125 %), sunflowers 11.7 plants/m (234 %). Seedling establishment of greater than 100 % of the target population was thought to have occurred as a result of higher than expected germination percentage. Seeding rates were increased as a result of the poor establishment in winter crops. Summer crops establishment was from 60 % to 100 % of seed sown.
Anhydrous ammonia application decreased establishment of all species as application rate was increased. Generally, the largest decreases occurred between 2.25 and 4.5 g/m of anhydrous ammonia-N (Fig. 4.4). By increasing the anhydrous ammonia band-seed separation distance, establishment increased significantly \( P<0.05 \) (Fig. 4.4). There were significant interactions between separation distance and species \( P<0.001 \) and anhydrous ammonia application rate, separation distance and species \( P<0.001 \).

Species susceptibility to NH\(_3\) was determined by percentage reduction in establish compared to the zero control treatment (Table 4.5).

At 50 mm separation distance of anhydrous ammonia bands and seed, reduction in sunflower establishment (68.5%) was significantly \( P<0.05 \) more than that for maize (8.0 %), sorghum (8.1 %) or cotton (20.2 %) (Table 4.5). Reduction in seedling establishment at 25 mm separation distance was significantly greater for each crop species than for 50 mm spacing \( P<0.05 \). The ranking of species for susceptibility to NH\(_3\) was the same for 50 mm and 25 mm separation distances. Reduction in establishment of sorghum (33.5 % and 8.1 % for 25 mm and 50 mm separation distances) and maize (53.9 % and 8.0 %) were generally less than for cotton (61.6 % and 20.2 %) and sunflower (68.5 % and 39.5 %). Even with low anhydrous ammonia rates (1.13 and 2.25 g/m N), where the fertilizer band was placed 50 mm from seeds, crop establishment response for cotton and sunflower indicate that soil NH\(_3\) concentration was above that tolerated by these species. Only at much higher anhydrous ammonia rates (4.5 and 6.75 g/m N) were significant reductions in establishment recorded for maize and sorghum for the 50 mm separation distance.

**Determining Species Susceptibility to Soil Ammonium Concentration**

An index of crop species susceptibility to soil NH\(_4^+\)-N was the NH\(_4^+\)-N concentration required to reduce establishment to 50 % of the unfertilized control (\( \text{TC}_{\text{NH}_4^{50}} \)).
Table 4.5 Establishment reduction (%) for cotton, maize, sorghum and sunflower 19 DAS from 5 rates of anhydrous ammonia applied at sowing, either 25 mm or 50 mm to both sides of the seed, Experiment 4-2.

<table>
<thead>
<tr>
<th>Anhydrous ammonia rate (g/m of N)</th>
<th>separation distance (mm)</th>
<th>Crop species (%)</th>
<th>cotton</th>
<th>maize</th>
<th>sorghum</th>
<th>sunflower</th>
<th>mean</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0</td>
<td>25</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>50</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>1.13</td>
<td>25</td>
<td>13.1</td>
<td>33.7</td>
<td>0</td>
<td>21.7</td>
<td>17.1</td>
<td></td>
</tr>
<tr>
<td></td>
<td>50</td>
<td>14.9</td>
<td>4.9</td>
<td>9.2</td>
<td>25.5</td>
<td>13.6</td>
<td></td>
</tr>
<tr>
<td>2.25</td>
<td>25</td>
<td>58.6</td>
<td>22.2</td>
<td>0</td>
<td>57.2</td>
<td>34.5</td>
<td></td>
</tr>
<tr>
<td></td>
<td>50</td>
<td>0</td>
<td>2.4</td>
<td>0</td>
<td>27.6</td>
<td>7.5</td>
<td></td>
</tr>
<tr>
<td>4.5</td>
<td>25</td>
<td>27.2</td>
<td>70.0</td>
<td>54.1</td>
<td>96.3</td>
<td>61.9</td>
<td></td>
</tr>
<tr>
<td></td>
<td>50</td>
<td>28.5</td>
<td>10.7</td>
<td>11.1</td>
<td>51.6</td>
<td>19.6</td>
<td></td>
</tr>
<tr>
<td>6.75</td>
<td>25</td>
<td>97.3</td>
<td>89.6</td>
<td>79.7</td>
<td>98.9</td>
<td>91.3</td>
<td></td>
</tr>
<tr>
<td></td>
<td>50</td>
<td>37.3</td>
<td>14.3</td>
<td>12.0</td>
<td>54.8</td>
<td>29.6</td>
<td></td>
</tr>
<tr>
<td>mean</td>
<td>25</td>
<td>61.6</td>
<td>53.9</td>
<td>33.5</td>
<td>68.5</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>50</td>
<td>20.2</td>
<td>8.0</td>
<td>8.1</td>
<td>39.9</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

l.s.d. (P=0.05)
rate x species 24.5
separation x species 15.5
rate x separation 33.1
rate x separation x species 34.6

In a test of homogeneity of regression for data from Experiments 4-1 and 4-2 it was found that there were significant differences between regression equations (P=0.01), significant differences between intercepts (P=0.01) and no difference between slopes. Data for each species was then compared using the linear regression model, \( y = a + 0.0007518 \times x \), with common slope and different intercepts. Regression intercepts, ‘a’ values, were used to group species according to significant l.s.d., \( P=0.05 \) (Table 4.6). Species TC \( \text{NH}_4 \text{ 50} \) were then calculated from species regression equations.
Fig. 4.4  Establishment for (a) cotton, (b) maize, (c) sorghum, (d) sunflowers 19 days after sowing from the application of 5 rates of anhydrous ammonia at sowing, 25 mm (■) and 50 mm (□) to either side of the seed, Experiment 4-2. Vertical bars indicate l.s.d. (P=0.05).
Species were separated into 3 tolerance groups according to the intercept, ‘a’ values, for each species. Barley, canary and wheat were the most tolerant species, sorghum, chickpea and canola were less tolerant and sunflower and cotton were least tolerant (Table 4.6).

Table 4.6 Estimates of NH₄⁺-N concentrations for 50 % reduction in seedling establishment derived from regression analysis of species establishment at each anhydrous ammonia rate and separation distance, and the corresponding soil NH₄⁺-N measurements, for Experiments 4-1(a) and 4-2 (b) (numbers followed by the same letters are not significantly different (P=0.05).

<table>
<thead>
<tr>
<th>Species</th>
<th>Toxic NH₄⁺-N concentration (TCNH₄₅₀) (mg/kg)</th>
<th>pH (at TCNH₄₅₀ calculated)</th>
<th>Regression Intercept (‘a’ value)</th>
</tr>
</thead>
<tbody>
<tr>
<td>barley</td>
<td>1146</td>
<td>8.62</td>
<td>1.3619a</td>
</tr>
<tr>
<td>canary</td>
<td>1121</td>
<td>8.58</td>
<td>1.3428a</td>
</tr>
<tr>
<td>canola</td>
<td>609</td>
<td>7.87</td>
<td>0.9575b</td>
</tr>
<tr>
<td>chickpea</td>
<td>741</td>
<td>8.06</td>
<td>1.0568b</td>
</tr>
<tr>
<td>wheat</td>
<td>1019</td>
<td>8.44</td>
<td>1.266a</td>
</tr>
<tr>
<td>cotton</td>
<td>368</td>
<td>8.15</td>
<td>0.7765c</td>
</tr>
<tr>
<td>sorghum</td>
<td>657</td>
<td>8.57</td>
<td>0.9943b</td>
</tr>
<tr>
<td>sunflower</td>
<td>399</td>
<td>8.19</td>
<td>0.8003c</td>
</tr>
</tbody>
</table>

(pH values at TCNH₄₅₀ were calculated from the regression of NH₄⁺-N and NH₃-N band pH. Data not presented)

Although maize was also tested (data not present), the linear regression fit for maize was not significant and the intercept could not be established with any degree of confidence. Visual assessment of the maize data and the ranking in Experiment 4-2 suggested that maize appears to have higher soil NH₃ tolerance than sorghum.
4.4 DISCUSSION

Two methods to evaluate crop susceptibility to NH₃, species response to anhydrous ammonia application rate and separation by regression with soil NH₄⁺-N gave similar ranking of species susceptibility.

Wheat and barley were consistently the most NH₄⁺-N tolerant winter species (Experiment 4-1), followed by canary and canola. Chickpea was the least tolerant species. The TCₜ₅₀ values suggest that the more tolerant winter species were able to withstand a soil NH₄⁺-N concentration about twice that of less tolerant species. This finding is in general agreement with the current commercial recommendations for winter crops and the general ratings of species groups by Carter (1967). The suggested maximum N fertilizer application rate to wheat is currently 25 kg/ha N and 12 kg/ha N for canary (Crosthwaite and Harvey 1992).

Species soil NH₃-N tolerance ranking for summer crops (Experiment 4-2) differed depending on the ranking method used. Maize and sorghum were consistently more tolerant than sunflower by both ranking methods, but cotton tolerance varied with ranking method. Cotton was similar to maize and sorghum tolerance using application rate means separated by l.s.d., but was similar to sunflowers using the soil NH₄⁺-N regression approach. The greater susceptibility of cotton by the regression approach is in agreement with the anecdotal experience of tolerance to NH₄⁺-fertilizers.

TCₜ₅₀ values for cotton and sunflower suggest that these species are able to tolerate only 60 % of the soil NH₄⁺-N concentration tolerated by sorghum or maize. The relative tolerance ranking of maize, sorghum and sunflower also agrees with commercial observations about these crops (Mills and McIntyre 1997).

The critical soil NH₄⁺-N concentration determined for maize from these experiments, 700 mg/kg, was lower than the critical concentration found by Colliver and Welch (1970b) >1000 mg/kg. Duisberg and Buehrer (1954) found the critical level for barley to be >450 mg/kg whilst for this experiment we found
the critical level to be 1146 mg/kg. Discrepancies may result from methods used to determine critical NH₄⁺-N concentration or from differing moisture content of soils. Colliver and Welch (1970b) used l.s.d. to establish critical concentration and there was no indication of how the critical level was established by Duisberg and Buehrer (1954) whereas in this study critical NH₄⁺-N concentration was designated by that concentration where emergence was reduced by 50%.

In Experiment 4-1 soil conditions unsuitable for seedling establishment appeared to be the cause for poor seedling establishment for all fertilizer rates and both seed-anhydrous ammonia application band separation distances. Soil moisture in the seed placement zone at sowing was 31.3 % w/w which should have been adequate for germination and establishment of the winter crop species.

No soil physical characteristics were measured to explain why separation distance had no effect on soil NH₄⁺-N concentration near seed or on crop establishment of winter crops. There was no visible or olfactory evidence of indirect loss during application. This suggests that the soil conditions did not prevent movement of NH₃ placed 50 mm from the seed back towards the seed line 24 hours after application.

Robotham (1993) suggested that 50 mm seed-NH₃-N band separation distance is adequate to prevent seedling damage of wheat and barley for rates up to 200 kg/ha N (4.5 g/m) as anhydrous ammonia applied either side of the seed. The results for wheat and barley in Experiment 4-1 suggest that under cloddy soil conditions reductions in establishment may be commercially unacceptable at 4.5 g/m even with a 50 mm separation distance.

In Experiment 4-2, where soil tilth was finer, there were clear differences in soil NH₄⁺-N concentrations around the seed resulting from the 25 mm and 50 mm separation distances. Soil NH₄⁺-N concentrations from all anhydrous ammonia application rates and 50 mm separation were lower than those for the lowest anhydrous ammonia application rate for the 25 mm separation. This result
highlights the advantage of a fine soil tilth in crop establishment using this type of fertilizer application equipment. Farmers who are using this type of fertilizer applicator in rough seed-bed conditions should reduce the anhydrous ammonia rate to a maximum of 2.25 g/m N for more tolerant crops such as wheat and barley and to 1.13 g/m N for a less tolerant crop such as canola. They also need to ensure that the equipment is tuned to the range of soil conditions likely to be experienced at sowing and is capable of ensuring that diffusion from the point of anhydrous ammonia injection to the seed-line can be modified with equipment such as press wheels. There is also a need to consider crop species’ NH$_3$ tolerance in the interaction with soil conditions and application equipment configuration.

Similar soil NH$_4^+$-N concentrations for the 4.5 g/m N and 6.75 g/m N anhydrous ammonia rates, in all combinations of anhydrous ammonia rate by separation distance in Experiment 4-1 suggest that a significant loss or displacement of NH$_3$ had occurred during application. Alternatively, the sampling and extraction procedures used were unable to detected the NH$_3$ form or that the NH$_4^+$-N concentration was sufficient to saturate the soil.

Ammonia emissions were detected by smell, from the 4.5 g/m and 6.75 g/m anhydrous ammonia-N application rates during sampling, 24 hours after application. The detection of NH$_3$ in the atmosphere suggested that the anhydrous ammonia application bands had not reached equilibrium and NH$_3$ vapour was still moving away from centre of the application zone to more distant adsorption sites through soil macropores. Robotham (1993), using similar application equipment and a vertical sampling technique, was able to consistently recover 95 % of applied anhydrous ammonia at rates from 50 to 300 kg/ha. Gaseous NH$_3$ loss from the seed zone is unlikely to be the mechanism by which similar soil NH$_4^+$-N concentrations were generated at the two highest anhydrous ammonia-N application rates.

Plateauing of soil NH$_4^+$-N concentration may have resulted from NH$_4^+$ saturation of the soil moisture and soil surface adsorption sites in the soil macropores as gaseous NH$_3$ diffused away from application sites and toward the seed line.
Maximum soil $\text{NH}_4^+$-N concentrations, 965 to 1156 mg/kg, found at the highest anhydrous ammonia rates in this experiment were below the 2850 to 3250 mg/kg $\text{NH}_4^+$-N found at the centre of application bands 24 hours after application by Whitehouse and Leslie (1973). However, maximum concentrations in this experiment were similar to those of Whitehouse and Leslie (1973) per unit of cation exchange capacity (CEC). The CEC for Whitehouse and Leslie’s soil was 73.6 cmol(+) /kg whereas CEC for the Lawes soil was 34 cmol(+) /kg.

By using application equipment that effectively seals the seed-line and choosing soil conditions conducive to sealing the soil after application, high $\text{NH}_4^+$-N concentrations can be prevented in the seed-line, thereby increasing the rate of anhydrous ammonia that can be applied and/or reducing the risk of establishment damage across a wider range of soil conditions.

Nitrite-N was not considered to have had an influence on seedling establishment for these experiments as the concentration measured was less than 2 mg/kg (data not presented). However, it is possible that NO$_2^-$-N accumulation will have occurred after soil sampling (24 hours after anhydrous ammonia application). Whitehouse (1972) found significant NO$_2^-$-N accumulation (4 to 80 mg/kg) in the application band 7 days after application and was at lower levels 15 days after application, where the initial soil $\text{NH}_4^+$-N concentration was less than 2500 mg/kg.

4.5 CONCLUSION
Significant differences in susceptibility of individual crop species to soil NH$_3$ were identified in this experiment. However, the size of the differences between individual species from within a group of species of similar tolerance was not large enough to change currently advised maximum $\text{NH}_4^+$-N rates for application with seed at sowing. Differences in the species tolerance of $\text{NH}_4^+$-N were smaller than changes in soil $\text{NH}_4^+$-N concentration in the seed-line created by different soil conditions.
These data suggest that species can be grouped according to their soil NH$_4^+$-N tolerance and that NH$_4^+$-fertilizer rate recommendations for the species tolerance groupings confirm the accuracy of those in commercial use for ammonium sulfate and ammonium phosphate fertilizers. These experiments also provide NH$_3$-N tolerance ranking for species for which there were no commercial recommendations.

To eliminate the effect of soil type, moisture availability and application equipment on the species’ NH$_3$ response, Chapter 5 investigates the tolerance response of the species used in this experiment, to atmospheric NH$_3$ in containers without soil.
Chapter 5

Tolerance ranking of seed and seedlings for 10 crop species to atmospheric ammonia.

Summary

An experimental technique, involving exposure of seed of 10 crop species to concentrations of atmospheric NH₃ during germination on filter paper, was used to develop NH₃ tolerance ranking by eliminating soil, fertilizer type and application variability, and as much as possible osmotic effects.

Exposure above 200 x 10⁻⁴ M NH₄OH for 72 h was sufficient to significantly reduce or inhibit germination in all species. Tolerance to NH₃ was evaluated for germination %, coleoptile and radicle growth with some species having different critical concentrations for the 3 parameters. Rank, combining tolerance for germination, coleoptile and radicle growth was established to relate to likely performance in the field.

A range of physical and chemical seed characteristics was correlated with NH₃ tolerance to infer tolerance mechanisms. For monocot species tolerance was related to the seed surface area/volume ratio, suggesting that diffusion resistance was an important parameter whereas for dicot species N % of seed was negatively correlated with tolerance.

5.1 INTRODUCTION

The most common method for determining the effect of fertilizer on seed germination and crop establishment has been through field experimentation (Nyborg 1961; Carter 1967; Mason 1971; Diebert et al. 1985; Radford et al. 1989; Gerwing et al. 1994). These experiments usually describe the effects of fertilizer products on germination and crop establishment under defined soil conditions. The interaction of species response with the physical and chemical characteristics of fertilizer and soil, and the design of fertilizer application and sowing equipment may influence results.

Crop establishment damage from NH₄⁺ containing or NH₃-producing fertilizer placed with the seed most commonly is a result of NH₃ toxicity (Warren 1962). Urea, urea blended or compounded with other fertilizers, and to a lesser extent DAP, are products that most commonly cause poor establishment when placed with the seed at sowing. These fertilizer products
have relatively high NH$_3$ potential i.e. they result in a combination of high NH$_4^+$ concentration and moderate to strong alkaline reaction in the soil, leading to and maintaining an emission of free NH$_3$.

In a closed system at equilibrium, gaseous NH$_3$ concentration in the head-space above NH$_4$OH solution remains relatively constant, NH$_3$ concentration being a function of the NH$_4^+$ concentration in solution, temperature and the solution pH. Allred and Ohlrogge (1963), Colliver (1969) and Woodstock and Tsao (1986) in their studies described techniques using NH$_4$OH in closed containers to investigate germination damage from different atmospheric concentrations of NH$_3$.

To gain a better understanding of the tolerance of different crop species to NH$_3$ during seedling establishment, as affected by different fertilizer properties, an experimental technique that eliminated soil and application factors, and as much as possible osmotic effects, was used.

5.2 MATERIALS AND METHODS

Experiment 5-1 - Ammonia exposure period

To determine the most appropriate exposure period and NH$_4$OH concentration range for germination damage, an experiment was designed to describe the germination response for maize, a moderately NH$_3$-tolerant species frequently used for fertilizer toxicity research (Allred and Ohlrogge 1964; Hunter and Rosenau 1966; Colliver and Welch 1970a; Woodstock and Tsao 1986 and Gerwing et al. 1994). The NH$_4$OH concentration range selected was similar to that used by Colliver (1970a).

Fifty seeds of maize (cv. Pioneer C84) were placed in 85 mm petri dishes on Whatman No. 1 filter paper. Filter papers were moistened with either a bathing solution of deionised water or NH$_4$OH solution equal in concentration to that contained in the bottom of the closed container. The different moistening solutions were used to determine the rapidity of germination damage. The difference between NH$_4$OH solutions and deionised water was
thought to be indicative of the speed of diffusion of NH$_3$ into bathing moisture as would be the case in a newly fertilized soil. Petri dishes with lids removed were then placed into the 4.77 L gas-tight container (Plate 5.1) containing 800 mL of either $20 \times 10^{-4} \, M$ or $200 \times 10^{-4} \, M$ NH$_4$OH solution.

Solution concentrations were established by diluting analytical grade NH$_4$OH (16.8-17 $M$) to the approximate required concentration, which was then determined accurately by titrating with an appropriate concentration of dilute sulfuric acid using a Radiometer VIT 90 auto-titration unit.

Seeds were exposed to the equilibrium atmospheric NH$_3$ (NH$_3$(g.)) concentration for either 24 or 72 h in a growth chamber, where a constant temperature $20 \pm 0.5 \, ^\circ C$ was maintained. After the allotted NH$_3$(g.) exposure time, the containers were opened and petri dishes removed. The germinated seeds were counted and transferred to petri dishes containing filter paper moistened with de-ionised water. The petri dishes were placed in the growth cabinet with lids on for a further 72 h. After 72 h each seed was examined for germination; seed with 1 mm of radicle or coleoptile showing outside the testa were considered to have germinated, and radicle and coleoptile lengths were measured.

**Experiment 5-2 - Critical ammonia concentration range**

The objective of this Experiment 5-2 was to ascertain if the NH$_4$OH concentration range for maize was adequate to produce significant germination and growth damage for barley, canary, sorghum, sorghum, sunflower, wheat, cotton and chickpea using the same technique as described for Experiment 5-1, the exposure period was 72 h and the bathing solution was de-ionised water.

Maize and wheat were included as benchmark species to enable germination and seedling growth comparisons with results obtained in similar studies by Blanchard (1967), Allred and Ohlrogge (1964), Colliver (1969) and Woodstock and Tsao (1986).
**Experiment 5-3 - Crop species’ sensitivity to ammonia**

For Experiment 5-3, 50 seeds of the crop species tested in Experiment 5-2 and panicum (cv. Panorama) were exposed to seven NH$_3$(g.) concentrations to more closely define species NH$_3$-tolerance. The methods used were as for Experiment 5-2.

Atmospheric NH$_3$ concentrations were created by placing 800 mL of NH$_4$OH in the bottom of each of 7 containers. Molarities of the NH$_4$OH solutions were 0 (de-ionised water), 7.0 x 10$^{-4}$, 12 x 10$^{-4}$, 29 x 10$^{-4}$, 54 x 10$^{-4}$, 102 x 10$^{-4}$ and 208 x 10$^{-4}$. Concentration of NH$_4$OH and solution pH are presented in Table 5.1, NH$_3$(aq.) and NH$_3$(g.) were calculated from the measured parameters using the equations 1 to 4 in Chapter 3. Each treatment was duplicated in separate closed containers.

**Table 5.1 Concentration of NH$_4$OH (x 10$^{-4}$), pH and calculated concentration of various states of NH$_3$ in the test solutions at a temperature of 20 °C.**

<table>
<thead>
<tr>
<th>[NH$_4$ +NH$_3$] (M)</th>
<th>[NH$_4$] (mg/L)</th>
<th>pH</th>
<th>[NH$_3$ (aq.)] (mg/L)</th>
<th>[NH$_3$(g)] (mg/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>0</td>
<td>5.69</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>7</td>
<td>13</td>
<td>9.24</td>
<td>4.2</td>
<td>0.0019</td>
</tr>
<tr>
<td>12</td>
<td>22</td>
<td>9.64</td>
<td>12.7</td>
<td>0.0057</td>
</tr>
<tr>
<td>29</td>
<td>52</td>
<td>9.69</td>
<td>32.2</td>
<td>0.0146</td>
</tr>
<tr>
<td>54</td>
<td>97</td>
<td>9.74</td>
<td>62.1</td>
<td>0.0281</td>
</tr>
<tr>
<td>102</td>
<td>184</td>
<td>9.99</td>
<td>137.2</td>
<td>0.0620</td>
</tr>
<tr>
<td>208</td>
<td>374</td>
<td>10.35</td>
<td>317.2</td>
<td>0.1430</td>
</tr>
</tbody>
</table>

Measurements were made of some physical and chemical features of the seed suggested by Wedding and Vines (1959), Colliver (1969) and Pulsford (*personal communication* 1992) as being important in seedling injury from NH$_3$ toxicity (Table 5.2). Simple linear regression analysis and correlation coefficients (r) were used to infer seed characteristics that most influenced NH$_3$ toxicity to germination and seedling growth.
For monocot crop species, germination %, radicle and coleoptile lengths were measured to provide an index of NH₃ damage. For dicot crop species, germination % and seedling length were measured.

Data were converted to a relative scale by dividing values by the zero control and expressing as a percentage prior to statistical analysis to enable comparisons between species. This was necessary because of large species differences for germination, coleoptile length and radicle length.

**Data Analysis**

Effects of NH₄OH exposure concentration and separation distance on species germination and seedling growth were initially analysed using ANOVA. Significant treatment means were separated by an l.s.d. test ($P=0.05$).

Piece-wise linear regression analyses were conducted using procedures outlined for STATISTICA 5.1 for Windows (StatSoft Inc., 1996). Critical NH₃ concentrations were derived from the maximum NH₄OH solution concentration at which there was no significant ($P=0.05$) reduction in germination. This point, referred to as the maximum safe rate, was determined using the inflection point of the curve in a piece-wise linear regression analysis.
Table 5.2  Selected seed characteristics and nutrient concentrations for 10 crop species evaluated for tolerance to 
NH₃ toxicity for Experiment 5-3.

<table>
<thead>
<tr>
<th>Species</th>
<th>Seed number</th>
<th>Growth Season</th>
<th>Species group</th>
<th>Critical seed germination moisture</th>
<th>Time to 50% germination</th>
<th>Seed diameter</th>
<th>Seed length</th>
<th>Estimated seed volume</th>
<th>Estimated seed surface area/volume ratio</th>
<th>Seed Nitrogen Conc.</th>
<th>Phosphorus Conc.</th>
<th>Magnesium Conc.</th>
</tr>
</thead>
<tbody>
<tr>
<td>wheat</td>
<td>27500</td>
<td>winter</td>
<td>cereal</td>
<td>55.2</td>
<td>32</td>
<td>2.7</td>
<td>6.1</td>
<td>35</td>
<td>51</td>
<td>1.46</td>
<td>2.08</td>
<td>0.24</td>
</tr>
<tr>
<td>barley</td>
<td>26000</td>
<td>winter</td>
<td>cereal</td>
<td>71.9</td>
<td>32</td>
<td>2.4</td>
<td>7.9</td>
<td>36</td>
<td>60</td>
<td>1.66</td>
<td>2.14</td>
<td>0.44</td>
</tr>
<tr>
<td>sorghum</td>
<td>30000</td>
<td>summer</td>
<td>cereal</td>
<td>51.6</td>
<td>27</td>
<td>4.4</td>
<td>10.7</td>
<td>44</td>
<td>60</td>
<td>1.36</td>
<td>2.97</td>
<td>0.30</td>
</tr>
<tr>
<td>sunflower</td>
<td>14000</td>
<td>summer</td>
<td>oilseed</td>
<td>73.1</td>
<td>30</td>
<td>3.5</td>
<td>10.7</td>
<td>103</td>
<td>118</td>
<td>1.04</td>
<td>3.53</td>
<td>1.02</td>
</tr>
<tr>
<td>cotton</td>
<td>10500</td>
<td>summer</td>
<td>oilseed</td>
<td>77.8</td>
<td>29</td>
<td>3.9</td>
<td>8.8</td>
<td>108</td>
<td>109</td>
<td>1.01</td>
<td>4.56</td>
<td>0.73</td>
</tr>
<tr>
<td>canola</td>
<td>225000</td>
<td>winter</td>
<td>oilseed</td>
<td>85.3</td>
<td>50</td>
<td>2.0</td>
<td>4</td>
<td>12</td>
<td>30</td>
<td>3.00</td>
<td>4.38</td>
<td>0.76</td>
</tr>
<tr>
<td>panicum</td>
<td>150000</td>
<td>summer</td>
<td>cereal</td>
<td>52</td>
<td>56</td>
<td>1.4</td>
<td>2.4</td>
<td>4</td>
<td>6</td>
<td>1.5</td>
<td>2.16</td>
<td>0.27</td>
</tr>
<tr>
<td>maize</td>
<td>3000</td>
<td>summer</td>
<td>cereal</td>
<td>55.2</td>
<td>46</td>
<td>8.5</td>
<td>326</td>
<td>229</td>
<td>0.70</td>
<td>2.09</td>
<td>0.39</td>
<td>0.16</td>
</tr>
<tr>
<td>chickpea</td>
<td>6900</td>
<td>winter</td>
<td>legume</td>
<td>100.9</td>
<td>38</td>
<td>6.2</td>
<td>127</td>
<td>122</td>
<td>0.96</td>
<td>3.17</td>
<td>0.31</td>
<td>0.17</td>
</tr>
<tr>
<td>canary</td>
<td>143000</td>
<td>winter</td>
<td>cereal</td>
<td>64.5</td>
<td>48</td>
<td>1.4</td>
<td>4.9</td>
<td>7</td>
<td>21</td>
<td>3.00</td>
<td>3.23</td>
<td>0.53</td>
</tr>
</tbody>
</table>
Plate 5.1  Gas-tight containers and lids, and stands used to support petri dishes of seeds above NH₄OH solutions used in closed container experiments.

5.3  RESULTS

Experiment 5-1 - Ammonia exposure period

Germination

The combination of moistening the filter paper in contact with seeds with 20 x 10⁻⁴ M NH₄OH solution and lengthening the exposure period of seeds over NH₄OH solution of the same concentration to 72 h significantly (P<0.05) reduced germination. However, germination of maize seeds was still quite high at 92% (Table 5.3).

Germination was reduced to 64 % with 24 h exposure when the NH₄OH concentration was increased to 200 x 10⁻⁴ M, with similar reductions for both
moistening solutions. Germination was reduced to zero after 72 h exposure over the $200 \times 10^{-4}$ $M$ NH$_4$OH.

**Coleoptile length**

Coleoptile length was not significantly affected by exposure to $20 \times 10^{-4}$ $M$ NH$_4$OH concentration. For the 24 h exposure period, increasing the NH$_4$OH concentration from $20 \times 10^{-4}$ $M$ to $200 \times 10^{-4}$ $M$ reduced coleoptile length from 8.2 mm to 4.9 mm when the seed was bathed in water but the difference was not significant when bathed in NH$_4$OH (Table 5.4). Coleoptile elongation was significantly inhibited by increasing exposure period from 24 to 72 h and NH$_4$OH concentration from $20 \times 10^{-4}$ $M$ to $200 \times 10^{-4}$ $M$ in both bathing solutions.

**Table 5.3** Percentage of maize seed germinated after being exposed to 2 atmospheric NH$_3$ concentrations above solutions of 20 and 200 x $10^{-4}$ $M$ NH$_4$OH for 24 or 72 h in a closed system.

<table>
<thead>
<tr>
<th>Solution [NH$_4$OH] ($x 10^{-4} M$)</th>
<th>Exposure time (h) in closed system</th>
<th>Bathing solution</th>
<th>%</th>
<th>mean</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>24</td>
<td>24</td>
<td>72</td>
<td></td>
</tr>
<tr>
<td></td>
<td>water</td>
<td>NH$_4$OH</td>
<td>NH$_4$OH</td>
<td></td>
</tr>
<tr>
<td>20</td>
<td>100</td>
<td>94</td>
<td>92</td>
<td>95</td>
</tr>
<tr>
<td>200</td>
<td>64</td>
<td>60</td>
<td>0</td>
<td>41</td>
</tr>
<tr>
<td>mean</td>
<td>82</td>
<td>77</td>
<td>46</td>
<td></td>
</tr>
</tbody>
</table>

l.s.d. ($P=0.05$)

<table>
<thead>
<tr>
<th>exposure time</th>
<th>concentration</th>
<th>exposure time x concentration</th>
</tr>
</thead>
<tbody>
<tr>
<td>6</td>
<td>5</td>
<td>8</td>
</tr>
</tbody>
</table>

**Radicle length**

Radicle length was significantly ($P<0.05$) reduced by increasing both exposure period and the NH$_4$OH concentration over which seeds germinated (Table 5.5). Greater sensitivity of the radicle growth to NH$_3$ concentration and
exposure time compared with coleoptile growth is in agreement with the findings of Allred and Ohlrogge (1964) and Colliver (1969).

Table 5.4  Maize coleoptile length (mm) after being exposed to 2 atmospheric NH₃ concentrations above solutions of 20 and 200 x 10⁻⁴ M NH₄OH for 24 and 72 h in a closed system.

<table>
<thead>
<tr>
<th>Solution [NH₄OH] (x 10⁻⁴ M)</th>
<th>Exposure time (h) in closed system</th>
<th>Bathing solution</th>
<th>24</th>
<th>24</th>
<th>72</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>water</td>
<td>NH₄OH</td>
<td>NH₄OH</td>
<td>mean</td>
</tr>
<tr>
<td>20</td>
<td></td>
<td>8.2</td>
<td>6.5</td>
<td>7.9</td>
<td>7.5</td>
</tr>
<tr>
<td>200</td>
<td></td>
<td>4.9</td>
<td>5.2</td>
<td>0</td>
<td>3.4</td>
</tr>
<tr>
<td>mean</td>
<td></td>
<td>6.6</td>
<td>5.9</td>
<td>4.0</td>
<td></td>
</tr>
</tbody>
</table>

l.s.d. (P=0.05)

exposure time 1.2
concentration 1.0
exposure time x concentration 1.7

Table 5.5  Maize radicle length (mm) after being exposed to 2 atmospheric NH₃ concentrations above solutions of 20 and 200 x 10⁻⁴ M NH₄OH for 24 and 72 h in a closed system.

<table>
<thead>
<tr>
<th>Solution [NH₄OH] (x 10⁻⁴ M)</th>
<th>Exposure time (h) in closed system</th>
<th>Bathing solution</th>
<th>24</th>
<th>24</th>
<th>72</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>water</td>
<td>NH₄OH</td>
<td>NH₄OH</td>
<td>mean</td>
</tr>
<tr>
<td>20</td>
<td></td>
<td>24.5</td>
<td>27.6</td>
<td>18.6</td>
<td>23.6</td>
</tr>
<tr>
<td>200</td>
<td></td>
<td>6.4</td>
<td>12.5</td>
<td>0</td>
<td>6.3</td>
</tr>
<tr>
<td>mean</td>
<td></td>
<td>15.5</td>
<td>20.0</td>
<td>9.3</td>
<td></td>
</tr>
</tbody>
</table>

l.s.d. (P = 0.05)

exposure time 6.9
concentration 5.5
exposure time x concentration ns
Experiment 5-2 - Species sensitivity to ammonia

Germination

Germination % of all species except barley at the zero NH_4OH treatment was above the minimum expected of certified seed (Mills et al. 1996; Mills and McIntyre 1997). Barley germination at 51% was well below the expected 85 to 95 %, but proportional reduction in germination from increasing NH_4OH concentration was similar to other species (Table 5.6). Germination of all species was unaffected by increasing the NH_4OH concentration to 20 x 10^{-4} M. By raising the concentration to 200 x 10^{-4} M germination was prevented in barley, canary, sorghum, sunflower and significantly reduced in chickpea, maize, cotton and wheat.

Coleoptile elongation

The highest concentration of NH_4OH (200 X 10^{-4} M) caused a significant (P<0.05) reduction in coleoptile length compared with both the zero treatment and 20 x 10^{-4} M in all species except maize. Coleoptile length in maize was similar for the three NH_4OH concentrations (Table 5.7). As the NH_4OH solution concentration was increased from 0 to 20 x 10^{-4} M, the coleoptile length of barley, canary and wheat increased, while for sorghum it was unchanged. The shorter coleoptile length in the zero NH_4OH treatment may have resulted from the osmotic loss of N compounds and carbohydrates from the seed. The harmful action of distilled water on biological material was described by True (1914), Scarth (1924) and Tilford et al. (1924). Conversely, stimulation of coleoptile length at 20 x 10^{-4} M NH_4OH may have been due to increased N supply (Wood 1990).

Radicle length

When the NH_4OH concentration in containers was increased from 0 to 20 x 10^{-4} M, radicle length in all species except sorghum was unaffected (Table 5.8). Sorghum radicle length was increased dramatically from 35.3 mm to 132.8 mm with the increase in NH_4OH concentration.
Table 5.6  Germination % of seeds of 8 crop species after exposure to atmospheric NH₃ above a solution of 0, 20 or 200 x 10⁻⁴ M NH₄OH.

<table>
<thead>
<tr>
<th>Crop species</th>
<th>NH₄OH solution concentration (x 10⁻⁴ M)</th>
<th>0</th>
<th>20</th>
<th>200</th>
<th>mean</th>
</tr>
</thead>
<tbody>
<tr>
<td>barley</td>
<td></td>
<td>51</td>
<td>57</td>
<td>0</td>
<td>36</td>
</tr>
<tr>
<td>canary</td>
<td></td>
<td>60</td>
<td>68</td>
<td>0</td>
<td>43</td>
</tr>
<tr>
<td>sorghum</td>
<td></td>
<td>89</td>
<td>76</td>
<td>0</td>
<td>55</td>
</tr>
<tr>
<td>sunflower</td>
<td></td>
<td>93</td>
<td>93</td>
<td>0</td>
<td>62</td>
</tr>
<tr>
<td>maize</td>
<td></td>
<td>100</td>
<td>95</td>
<td>5</td>
<td>67</td>
</tr>
<tr>
<td>wheat</td>
<td></td>
<td>97</td>
<td>96</td>
<td>17</td>
<td>70</td>
</tr>
<tr>
<td>cotton</td>
<td></td>
<td>81</td>
<td>91</td>
<td>26</td>
<td>66</td>
</tr>
<tr>
<td>chickpea</td>
<td></td>
<td>100</td>
<td>97</td>
<td>29</td>
<td>76</td>
</tr>
<tr>
<td>mean</td>
<td></td>
<td>84</td>
<td>85</td>
<td>10</td>
<td></td>
</tr>
</tbody>
</table>

l.s.d. (P = 0.05)
concentration 5
species 9
concentration x species 15

Table 5.7  Coleoptile length (mm) of the seeds of 5 crop species after exposure to atmospheric NH₃ above a solution of 0, 20 or 200 x 10⁻⁴ M NH₄OH.

<table>
<thead>
<tr>
<th>Crop species</th>
<th>NH₄OH solution concentration (x 10⁻⁴ M)</th>
<th>0</th>
<th>20</th>
<th>200</th>
<th>mean</th>
</tr>
</thead>
<tbody>
<tr>
<td>barley</td>
<td></td>
<td>15.5</td>
<td>21.2</td>
<td>0</td>
<td>12.3</td>
</tr>
<tr>
<td>canary</td>
<td></td>
<td>21.5</td>
<td>29.2</td>
<td>0</td>
<td>16.9</td>
</tr>
<tr>
<td>maize</td>
<td></td>
<td>4.8</td>
<td>8.0</td>
<td>5.7</td>
<td>6.2</td>
</tr>
<tr>
<td>sorghum</td>
<td></td>
<td>21.8</td>
<td>19.9</td>
<td>0</td>
<td>13.9</td>
</tr>
<tr>
<td>wheat</td>
<td></td>
<td>22.5</td>
<td>28.0</td>
<td>1.6</td>
<td>17.4</td>
</tr>
<tr>
<td>mean</td>
<td></td>
<td>17.2</td>
<td>21.3</td>
<td>1.5</td>
<td></td>
</tr>
</tbody>
</table>

l.s.d. (P = 0.05)
concentration 1.8
species 3.3
concentration x species 5.8

¹ Coleoptile length was not measured for dicotyledonous crops.
Radicle length was reduced dramatically in all species when the NH$_4$OH concentration was raised to 200 x $10^{-4} \, M$. Sorghum was the only species to show a significant ($P<0.05$) reduction in radicle length. Its reduction in radicle length was made more evident due to the apparent stimulatory effect of 20 x $10^{-4} \, M$ NH$_4$OH. In the absence of the stimulatory effect, radicle length in other species was not reduced significantly although the change in radicle length due to increased NH$_4$OH concentration was significant when meaned across species (Table 5.8).

**Table 5.8**  Radicle length (mm) of the seeds of 8 crop species after exposure to atmospheric NH$_3$ above a solution of 0, 20 or 200 x $10^{-4} \, M$ NH$_4$OH.

<table>
<thead>
<tr>
<th>Crop species</th>
<th>NH$_4$OH solution concentration (x $10^{-4}$ M)</th>
<th>0</th>
<th>20</th>
<th>200</th>
<th>mean</th>
</tr>
</thead>
<tbody>
<tr>
<td>barley</td>
<td>13.3</td>
<td>17.7</td>
<td>0</td>
<td>10.3</td>
<td></td>
</tr>
<tr>
<td>canary</td>
<td>32.5</td>
<td>29.2</td>
<td>0</td>
<td>20.6</td>
<td></td>
</tr>
<tr>
<td>chickpea</td>
<td>38.8</td>
<td>38.3</td>
<td>3.6</td>
<td>26.9</td>
<td></td>
</tr>
<tr>
<td>cotton</td>
<td>16.7</td>
<td>19.2</td>
<td>8.3</td>
<td>14.7</td>
<td></td>
</tr>
<tr>
<td>maize</td>
<td>29.9</td>
<td>25.2</td>
<td>1.0</td>
<td>18.7</td>
<td></td>
</tr>
<tr>
<td>sorghum</td>
<td>35.3</td>
<td>132.8</td>
<td>0</td>
<td>56.0</td>
<td></td>
</tr>
<tr>
<td>sunflower</td>
<td>11.6</td>
<td>11.1</td>
<td>0</td>
<td>7.6</td>
<td></td>
</tr>
<tr>
<td>wheat</td>
<td>36.1</td>
<td>41.9</td>
<td>1.4</td>
<td>26.4</td>
<td></td>
</tr>
<tr>
<td>mean</td>
<td>26.7</td>
<td>39.4</td>
<td>1.8</td>
<td></td>
<td></td>
</tr>
<tr>
<td>l.s.d. ($P = 0.05$)</td>
<td></td>
<td>17.3</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>concentration</td>
<td>species</td>
<td>31.7</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>concentration</td>
<td>x species</td>
<td>54.8</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

1 Total length of shoot below the cotyledons measured

**Experiment 5-3 - Crop species sensitivity to ammonia**

**Germination**

Germination of all species, except for chickpeas and maize, was significantly reduced at 208 x$10^{-4} \, M$ NH$_4$OH (Table 5.9). The germination of cotton, wheat, canary, panicum and canola declined significantly ($P<0.05$) between
54 and 102 \times 10^{-4} \, M, while barley, sorghum and sunflower did not decline until the concentration was between 102 and 208 \times 10^{-4} \, M.

Table 5.9  Germination (%) relative to zero control for seeds of 10 crop species after exposure to atmospheric NH₃ above 0, 7, 12, 29, 54, 102 and 208 \times 10^{-4} \, M NH₄OH.

<table>
<thead>
<tr>
<th>[NH₄OH] (x10^{-4} , M)</th>
<th>maize</th>
<th>cotton</th>
<th>wheat</th>
<th>barley</th>
<th>chickpea</th>
<th>sorghum</th>
<th>canary panicum</th>
<th>canola</th>
<th>sunflower</th>
<th>mean</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>7</td>
<td>99.5</td>
<td>87</td>
<td>93</td>
<td>106.5</td>
<td>103</td>
<td>107.5</td>
<td>178.5</td>
<td>80</td>
<td>78</td>
<td>102.5</td>
</tr>
<tr>
<td>12</td>
<td>96</td>
<td>93.5</td>
<td>97</td>
<td>105.5</td>
<td>98</td>
<td>112.5</td>
<td>104.5</td>
<td>90.5</td>
<td>103</td>
<td>102.5</td>
</tr>
<tr>
<td>29</td>
<td>94</td>
<td>89.5</td>
<td>98</td>
<td>111.5</td>
<td>102</td>
<td>103.5</td>
<td>115.5</td>
<td>100</td>
<td>91.5</td>
<td>99.5</td>
</tr>
<tr>
<td>54</td>
<td>100</td>
<td>104.5</td>
<td>95</td>
<td>116</td>
<td>102</td>
<td>103.5</td>
<td>179.5</td>
<td>103.5</td>
<td>92.5</td>
<td>103.5</td>
</tr>
<tr>
<td>102</td>
<td>103.5</td>
<td>69.5</td>
<td>86</td>
<td>114.5</td>
<td>103</td>
<td>101.5</td>
<td>69.5</td>
<td>82</td>
<td>40.5</td>
<td>101</td>
</tr>
<tr>
<td>208</td>
<td>72</td>
<td>1.5</td>
<td>5.0</td>
<td>0</td>
<td>102</td>
<td>12.5</td>
<td>4.0</td>
<td>15</td>
<td>0</td>
<td>21</td>
</tr>
<tr>
<td>mean</td>
<td>95</td>
<td>77.9</td>
<td>82</td>
<td>93.4</td>
<td>101.4</td>
<td>91.6</td>
<td>107.5</td>
<td>81.6</td>
<td>72.2</td>
<td>90</td>
</tr>
</tbody>
</table>

l.s.d. (P = 0.05)

- concentration: 9.9
- species: 11.8
- concentration x species: 31.2

Severe darkening of the seed testa as well as discoloration of the bathing solution in petri dishes was observed at the highest NH₄⁺ concentration for all species (Plate 5.2). Discoloration of the bathing solution was most likely related to accelerated leaching of electrolytes from seed exposed to NH₃ during imbibition, as described by Woodstock and Tsao (1986).

**Coleoptile length**

Coleoptile length of species other than barley and panicum was increased for NH₄OH concentrations below 54 \times 10^{-4} \, M and decreased significantly for concentrations above 54 \times 10^{-4} \, M. Coleoptile length for barley and panicum was generally unaffected in the presence of low NH₄OH concentrations but also decreased significantly for concentrations above 54 \times 10^{-4} \, M.
Plate 5.2. Seed coat of most species showed discolouration after exposure to NH₃ above the highest NH₄OH solution concentrations.
Maize coleoptile growth was tolerant of exposure to NH$_3$(g.) above the solution containing 208 x 10$^{-4}$ M NH$_4$OH; barley, sorghum and panicum tolerated up to 102 x 10$^{-4}$ M; canary and wheat were significantly ($P<0.05$) inhibited by NH$_4$OH concentrations greater than 54 x 10$^{-4}$ M (Table 5.10).

Maize was the only species for which the maximum NH$_3$ concentration for unaffected coleoptile growth was higher than for unaffected germination. For all other monocots, coleoptile growth was of equal sensitivity or more sensitive to NH$_3$ concentration than was seed germination.

Although maize was tolerant up to 208 x 10$^{-4}$ M NH$_4$OH, mean coleoptile length response for maize and sorghum were similar (Table 5.10). This appears to be mediated through a significant increase of coleoptile length in response to low to moderate NH$_3$(g.) concentrations. When considering the species mean response to NH$_3$(g.) concentrations, species order of tolerance was maize, sorghum, canary, wheat, panicum and barley.

**Table 5.10** Coleoptile length (%) relative to zero control for seeds of 6 crop monocot species after exposure to atmospheric NH$_3$ above solutions of 0, 7, 12, 29, 54, 102 and 208 x 10$^{-4}$ M NH$_4$OH.

<table>
<thead>
<tr>
<th>[NH$_4$OH] ($x10^{-4}$ M)</th>
<th>maize</th>
<th>wheat</th>
<th>barley</th>
<th>sorghum</th>
<th>canary</th>
<th>panicum</th>
<th>mean</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>7</td>
<td>192</td>
<td>112</td>
<td>86</td>
<td>132</td>
<td>162</td>
<td>89</td>
<td>129</td>
</tr>
<tr>
<td>12</td>
<td>184</td>
<td>105</td>
<td>81</td>
<td>166</td>
<td>126</td>
<td>134</td>
<td>133</td>
</tr>
<tr>
<td>29</td>
<td>176</td>
<td>123</td>
<td>97</td>
<td>163</td>
<td>148</td>
<td>90</td>
<td>133</td>
</tr>
<tr>
<td>54</td>
<td>112</td>
<td>81</td>
<td>108</td>
<td>171</td>
<td>75</td>
<td>58</td>
<td>101</td>
</tr>
<tr>
<td>102</td>
<td>123</td>
<td>51</td>
<td>48</td>
<td>161</td>
<td>42</td>
<td>72</td>
<td>83</td>
</tr>
<tr>
<td>208</td>
<td>138</td>
<td>19</td>
<td>0</td>
<td>76</td>
<td>4</td>
<td>26</td>
<td>44</td>
</tr>
<tr>
<td>mean</td>
<td>128.13</td>
<td>74</td>
<td>65</td>
<td>121</td>
<td>82</td>
<td>71</td>
<td></td>
</tr>
</tbody>
</table>

I.s.d. ($P = 0.05$)
- concentration: 16
- species: 20
- concentration x species: 52
Radicle length

Radicle length of most species was unaffected by NH₄OH concentrations below 54 x 10⁻⁴ M. For cotton however, radicle length increased in response to these low NH₄OH concentrations. Significant reductions in radicle length generally occurred where NH₄OH concentration was either 102 or 208 x 10⁻⁴ M (Table 5.11).

Radicle growth tolerance to NH₄OH exposure varied from 54 to 208 x 10⁻⁴ M across the 10 crop species. Maize and chickpea were tolerant of 208 x 10⁻⁴ M, cotton, barley sorghum and sunflower tolerated up to 102 x 10⁻⁴ M and wheat, canola, canary and panicum tolerated 54 x 10⁻⁴ M.

### Table 5.11 Radicle length (%) relative to zero control for seeds of 10 crop species after exposure to atmospheric NH₃ above solutions of 0, 7, 12, 29, 54, 102 and 208 x 10⁻⁴ M NH₄OH.

<table>
<thead>
<tr>
<th>[NH₄OH] (x10⁻⁴ M)</th>
<th>Species</th>
<th>maize</th>
<th>cotton</th>
<th>wheat</th>
<th>barley</th>
<th>chickpea</th>
<th>sorghum</th>
<th>canary</th>
<th>panicum</th>
<th>canola</th>
<th>sunflower</th>
<th>mean</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>%</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>0</td>
<td>%</td>
<td>104</td>
<td>141</td>
<td>98</td>
<td>103</td>
<td>107</td>
<td>115</td>
<td>72</td>
<td>86</td>
<td>87</td>
<td>98</td>
<td>101</td>
</tr>
<tr>
<td>12</td>
<td>%</td>
<td>87</td>
<td>154</td>
<td>93</td>
<td>87</td>
<td>102</td>
<td>107</td>
<td>84</td>
<td>119</td>
<td>88</td>
<td>100</td>
<td>102</td>
</tr>
<tr>
<td>29</td>
<td>%</td>
<td>88</td>
<td>164</td>
<td>103</td>
<td>108</td>
<td>101</td>
<td>106</td>
<td>113</td>
<td>72</td>
<td>114</td>
<td>89</td>
<td>106</td>
</tr>
<tr>
<td>54</td>
<td>%</td>
<td>96</td>
<td>156</td>
<td>69</td>
<td>106</td>
<td>99</td>
<td>111</td>
<td>82</td>
<td>82</td>
<td>90</td>
<td>103</td>
<td>99</td>
</tr>
<tr>
<td>102</td>
<td>%</td>
<td>155</td>
<td>147</td>
<td>46</td>
<td>73</td>
<td>95</td>
<td>109</td>
<td>33</td>
<td>42</td>
<td>26</td>
<td>101</td>
<td>83</td>
</tr>
<tr>
<td>208</td>
<td>%</td>
<td>97</td>
<td>41</td>
<td>14</td>
<td>0</td>
<td>56</td>
<td>5</td>
<td>6</td>
<td>23</td>
<td>0</td>
<td>20</td>
<td>26</td>
</tr>
<tr>
<td>mean</td>
<td>%</td>
<td>91</td>
<td>113</td>
<td>65</td>
<td>72</td>
<td>83</td>
<td>82</td>
<td>61</td>
<td>66</td>
<td>63</td>
<td>76</td>
<td></td>
</tr>
</tbody>
</table>

l.s.d. (P = 0.05)

- concentration: 18
- species: 21
- concentration x species: 56

The mean species radicle length response to NH₃ exposure indicated that the critical concentration for a species was not necessarily indicative of the way the species responded at lower NH₃(g.) concentrations. Although cotton
was in the group of species tolerant up to $102 \times 10^{-4} \text{ M NH}_4\text{OH}$, it had the highest mean radicle length, 129% (Table 5.11). This appears to be mediated through a significant increase of radicle length in response to low to moderate atmospheric NH$_3$ concentrations. A similar response to NH$_3$ concentration was also found for sorghum radicle in this experiment, this is consistent with the finding for sorghum in Experiment 5-2. The species mean response to atmospheric NH$_3$ concentrations ranked in order of decreasing tolerance was; cotton> maize> chickpea> sorghum> sunflower> barley> wheat - panicum> canola and canary.

**Comparing species germination response to NH$_3$ concentrations using piece-wise linear regression model**

Species were ranked according to germination tolerance to NH$_3$ exposure by determining the maximum safe NH$_3$ concentration by identifying the breakpoint of a piece-wise linear regression model. According to this method, chickpeas showed the greatest NH$_3$ tolerance during germination, being unaffected by exposure to NH$_4$OH concentrations greater than $100 \times 10^{-4} \text{ M}$ (Table 5.12). Maize, barley, sorghum and sunflower tolerated exposure to 90 to $95 \times 10^{-4} \text{ M NH}_4\text{OH}$ and wheat, panicum, canary and cotton tolerated 75 to $80 \times 10^{-4} \text{ M}$. Canola was the least tolerant species with germination reduced by exposure to concentrations above $70 \times 10^{-4} \text{ M}$.

Species tolerance rank for coleoptile growth according to the regression analysis (Table 5.12) was similar to the l.s.d. ($P=0.05$) separation of means. Differences in crop rank between the 2 methods were for wheat, which increased in rank, and barley that decreased in rank according to the regression method. For radicle growth, sorghum and panicum were the only species to change ranking by more than 2 positions. Sorghum appeared less tolerant and panicum was more tolerant to NH$_3$ according to the regression method.
Investigating Seed Characteristics which may affect Ammonia Toxicity

Seed characteristics (diameter, surface area, N, P and magnesium (Mg) content (%), critical seed moisture and time to 50 % germination) were correlated with maximum safe rate of NH₃ for nil effects to germination, radicle and coleoptile growth as determined by the piece-wise linear regression method. There was no clear association between any single seed characteristic and individual species germination tolerance to NH₃. Of the seed characteristics measured, species with large surface areas or small volumes appeared to have lower tolerance to NH₃ than species with a small surface area and large volume. When the crops were regrouped within monocot or dicot species, and analysed, significant correlations were found (Table 5.13).

Table 5.12 The highest concentration of NH₄OH ($M \times 10^{-4}$) at which there were no effects on seed germination, radicle length or coleoptile length of 10 species i.e. the maximum safe NH₄⁺ rate $a$.

<table>
<thead>
<tr>
<th>Species</th>
<th>Germination (%)</th>
<th>Radicle length</th>
<th>Coleoptile length</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>$M \text{NH}<em>4^+ x 10^{-4}</em>{(100r^2)}$</td>
<td>$M \text{NH}<em>4^+ x 10^{-4}</em>{(100r^2)}$</td>
<td>$M \text{NH}<em>4^+ x 10^{-4}</em>{(100r^2)}$</td>
</tr>
<tr>
<td>barley</td>
<td>93 (90)</td>
<td>82 (98)</td>
<td>74 (97)</td>
</tr>
<tr>
<td>canary</td>
<td>78 (94)</td>
<td>70 (98)</td>
<td>80 (97)</td>
</tr>
<tr>
<td>canola</td>
<td>72 (93)</td>
<td>72 (95)</td>
<td>n.a.</td>
</tr>
<tr>
<td>chickpea</td>
<td>101 (83)</td>
<td>94 (99)</td>
<td>n.a.</td>
</tr>
<tr>
<td>cotton</td>
<td>77 (98)</td>
<td>129 (98)</td>
<td>n.a.</td>
</tr>
<tr>
<td>maize</td>
<td>95 (98)</td>
<td>103 (96)</td>
<td>146 (99)</td>
</tr>
<tr>
<td>panicum</td>
<td>81 (96)</td>
<td>82 (99)</td>
<td>81 (76)</td>
</tr>
<tr>
<td>sorghum</td>
<td>91 (98)</td>
<td>93 (99)</td>
<td>138 (93)</td>
</tr>
<tr>
<td>sunflower</td>
<td>90 (99)</td>
<td>87 (98)</td>
<td>n.a.</td>
</tr>
<tr>
<td>wheat</td>
<td>82 (99)</td>
<td>75 (99)</td>
<td>84 (98)</td>
</tr>
</tbody>
</table>

$a$. Breakpoint of a piecewise linear regression ; n.a not applicable

Within the monocots, significant correlations of surface area or seed surface area/volume ratio with germination % ($r=-0.82$) or radicle length ($r=0.82$) suggested large sized, low surface area seeds may be more tolerant to NH₃ during germination. A negative relationship would arise if one of the mechanisms involved in seedling tolerance is resistance within the seed to diffusion of NH₃ into the metabolically active zone.
Within dicot species, N concentration (%) was negatively correlated with germination suggesting that those seeds with high N concentration had lower tolerance to NH₃ than seed with lower N concentration.

Table 5.13 Correlation co-efficients (r values) for germination, coleoptile and radicle growth tolerance to NH₃, with various seed characteristics.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Monocot Germination</th>
<th>Monocot Coleoptile length</th>
<th>Monocot Radicle length</th>
<th>Dicot Germination</th>
<th>Dicot Shoot + Root length</th>
</tr>
</thead>
<tbody>
<tr>
<td>Seed number</td>
<td>0.64</td>
<td>0.65</td>
<td>0.79</td>
<td>0.81</td>
<td>0.53</td>
</tr>
<tr>
<td>Seed volume</td>
<td>0.65</td>
<td>0.68</td>
<td>0.81</td>
<td>0.75</td>
<td>0.61</td>
</tr>
<tr>
<td>Seed surface area</td>
<td>0.72</td>
<td>0.69</td>
<td>0.82*</td>
<td>0.74</td>
<td>0.57</td>
</tr>
<tr>
<td>Surface area/volume</td>
<td>-0.82*</td>
<td>-0.68</td>
<td>-0.72</td>
<td>-0.67</td>
<td>-0.67</td>
</tr>
<tr>
<td>Winter/summer crop</td>
<td>-0.51</td>
<td>0.02</td>
<td>-0.23</td>
<td>0.66</td>
<td>0.65</td>
</tr>
<tr>
<td>Critical moisture</td>
<td>0.10</td>
<td>-0.56</td>
<td>-0.41</td>
<td>0.52</td>
<td>-0.19</td>
</tr>
<tr>
<td>Germination time</td>
<td>-0.44</td>
<td>-0.23</td>
<td>-0.09</td>
<td>-0.33</td>
<td>-0.73</td>
</tr>
<tr>
<td>Seed total N %</td>
<td>-0.33</td>
<td>0.13</td>
<td>-0.28</td>
<td>-0.96*</td>
<td>0.31</td>
</tr>
<tr>
<td>Seed total P %</td>
<td>0.02</td>
<td>-0.18</td>
<td>-0.19</td>
<td>-0.49</td>
<td>-0.12</td>
</tr>
<tr>
<td>Seed total Mg %</td>
<td>0.02</td>
<td>-0.20</td>
<td>-0.18</td>
<td>-0.61</td>
<td>0.26</td>
</tr>
</tbody>
</table>

* indicates correlation co-efficients significant at P=0.05

5.4 DISCUSSION

Experiments 5-1 and 5-2 provided guidelines to the conditions required to reflect the range of responses of crop species to NH₃ toxic condition. The results of Experiment 5-1 indicated that for maize, 0 to 200 × 10⁻⁴ M NH₄OH concentration range was adequate to describe the full range of germination, coleoptile and radicle responses. Exposure period at each concentration was found to be a more important determinant of germination response than the bathing solution in which the seed was initially placed. Significant germination damage could be detected after 24 h exposure when the seeds were placed above 200 × 10⁻⁴ M NH₄OH seed bathing solutions indicating that the onset of NH₃ toxicity was rapid and toxicity was not only mediated through dissolution of NH₃ in the bathing solution, but possibly also by
diffusion of NH₃(g.) directly through the seed coat not in contact with moisture (Woodstock and Tsao, 1986). Total inhibition of germination by exposure above 200 x 10⁻⁴ M for 72 h and the smaller but significant germination reduction from exposure above 20 x 10⁻⁴ M for 72 h suggest that damage may be more significant from short term exposure to high concentrations of NH₃ than from longer term exposure to lower concentrations.

The NH₄OH exposure conditions suggested from Experiment 5-1 were found adequate to produce a full range of germination, coleoptile and radicle response from cotton, wheat, barley, canary, chickpea, sorghum, sunflower in Experiment 5-2. However, 3 concentrations were inadequate to provide significant separation between species’ response.

Within a species, radicle growth was generally less tolerant to NH₃ than seed germination. In only two species, cotton and maize was radicle growth more tolerant of NH₃ than was its germination.

There are significant establishment implications where NH₃ forming fertilizers are placed at sowing near the seed of species for which the maximum safe NH₃ concentration for radicle growth is lower than that for germination. Even where moisture is adequate for germination, radicle elongation may not keep pace with the drying front in the soil. Where this occurs, then seedling establishment may be reduced substantially (Radford et al., 1989).

Radford et al. (1989) and Robotham (1993) observed that NH₃ toxicity derived from urea placed with seed significantly slowed radicle and coleoptile growth in wheat. This was most significant in modern short coleoptile semi-dwarf varieties, particularly when sown deeply or into relatively warm seed beds. They found that poor establishment was not due to impeded germination, but instead, it was due to a failure of the coleoptile to emerge from the soil surface. Also, radicle growth was reduced where seed and fertilizer were placed in close proximity.
Colliver (1969) found that radicle elongation in maize was reduced at a lower NH$_3$ concentration ($7 \times 10^{-4}$ M) than was found in this experiment ($103 \times 10^{-4}$ M). Higher tolerance for maize in this experiment may have resulted from the higher temperature (20.5 °C) at which seed was exposed (13 °C in Colliver 1969) and the shorter exposure time, 3 days versus 11 days, for Colliver (1969). The higher temperature used in this study was selected because it is more representative of soil temperature in cropping areas of Queensland and northwest New South Wales at sowing than the temperature used by Colliver (1969).

For most species, the maximum safe NH$_3$ concentration for satisfactory radicle and coleoptile elongation were in the same concentration range. Using the breakpoint of the piece-wise linear regression to establish the critical concentration for unaffected radicle or coleoptile growth gave a clearer indication of relative crop tolerance to NH$_3$ than the significance level from the l.s.d. test. Relative tolerance to NH$_3$ of the radicle and coleoptile for monocot species are shown in Table 5.14.

Higher tolerance to NH$_3$ of coleoptile than radicle elongation for maize in this experiment is consistent with the findings of Allred and Ohlrogge (1964) and Colliver (1969). These authors hypothesised that this effect resulted from the higher metabolic activity in the radicle during early stages of seedling establishment.

The estimated maximum safe rate for maize of 90-95 $\times 10^{-4}$ M was within the range 63 $\times 10^{-4}$ M to 320 $\times 10^{-4}$ M reported by Blanchar (1967), Allred and Ohlrogge (1964), Colliver (1969) and Woodstock and Tsao (1986), using similar procedures.

The short 24 and 72 h exposure times of this experiment were apparently long enough to significantly interfere with germination of most species, but too brief to decrease radicle or coleoptile growth to the extent reported by
Allred and Ohlrogge (1964), Colliver and Welch (1970b) and Radford et al. (1989).

Table 5.14  Relative tolerance for monocots of coleoptile (C) and radicle (R) elongation to NH₃ toxicity.

<table>
<thead>
<tr>
<th>Crop Species</th>
<th>Relative tolerance of coleoptile and radicle</th>
</tr>
</thead>
<tbody>
<tr>
<td>barley</td>
<td>R &gt; C</td>
</tr>
<tr>
<td>canary</td>
<td>C &gt; R</td>
</tr>
<tr>
<td>maize</td>
<td>C &gt; R</td>
</tr>
<tr>
<td>panicum</td>
<td>C = R</td>
</tr>
<tr>
<td>sorghum</td>
<td>R &gt; C</td>
</tr>
<tr>
<td>wheat</td>
<td>C &gt; R</td>
</tr>
</tbody>
</table>

The presence of low NH₃ concentrations appeared to stimulate growth of radicles and coleoptiles compared with those grown in distilled water. Wood (1990) found that the addition of some nutrients (particularly N, P, K) during germination was beneficial during seedling growth of soybean, sunflower and sorghum. This would explain the stimulation of radicle growth of some species e.g. sorghum at low NH₄OH concentrations.

Ranking of these species in increasing apparent tolerance to NH₃ does not completely agree with commercial experience of their establishment in the field, where N fertilizers are applied in close proximity to seeds at sowing. In northeastern Australia dicot species are classified as more sensitive to N fertilizers applied with seed at sowing than monocots. Thus, lower N fertilizer rates are generally recommended for dicots than for monocots, where fertilizer is to be placed in close proximity to the seed (Dubetz et al. 1959; Carter 1967).

The results of this experiment provide a ranking for tolerance of these species to NH₃. Dicot species were generally found to be no less tolerant to NH₃ during germination than monocots. Thus, the commercial observation about the lower tolerance of dicots to N fertilizer applied with the seed may not be singularly due to NH₃ toxicity. Osmotic or other effects of the fertilizers
or of its management may be causes. For example, dicots are generally grown in wider rows so they receive higher N fertilizer rates per unit length of row.

Generally the grouping of species for tolerance in this experiment was in agreement with that of Experiments 4-1 and 4-2. This indicates that soil conditions were secondary factors in determining establishment, soil NH$_4^+$ concentration and associated NH$_3$ being the primary factor.

5.5 CONCLUSION
The complete inhibition of germination at the highest NH$_4$OH concentration within the 72 h exposure suggested that the exposure period for subsequent experiments need be only 72 h and that NH$_3$ damage to seeds appears to occur early in the germination process.

The highest NH$_4$OH concentration, 200 x 10$^{-4}$ M was sufficient to significantly reduce or completely inhibit germination of all species tested.

Considering the two important phases of seedling establishment, germination and elongation of the radicle and coleoptile, crop species can be ranked by their response to fertilizer in these two phases. Ranks for decreasing tolerance to NH$_3$ concentration during germination for the monocot species was maize > barley > sorghum > wheat > panicum > canary and for dicots, chickpeas > sunflowers > cotton > canola.

To predict field response for a species where seeds are placed in close proximity to NH$_3$ forming fertilizers, it is likely that ranking of the crop in germination, coleoptile and radicle response should be used. After combining the three performance rankings, decreasing order of tolerance for monocot species was found to be maize > sorghum > wheat = barley > panicum > canary and for dicot species chickpeas > cotton > sunflowers > canola.
Although NH$_3$ tolerance ranking of species in field experiments (Chapter 4) and in experiments with gas-tight NH$_3$ chambers (this chapter) were similar, variation among soil types, fertilizer products and application equipment characteristics may have a greater effect on establishment than species response alone.

To better understand crop establishment response to NH$_4^+$-fertilizer, it is important to define the osmotic pressure and NH$_3$ toxicity characteristics of a range of fertilizer products. These fertilizer characteristics are pursued in subsequent chapters.
Chapter 6

Factors affecting seedling injury.

Summary

Three experiments were conducted to separate effects of NH$_3$ toxicity and osmotic pressure for previously studied crop species which represented a wide range of NH$_3$ tolerance. Experiments explored NH$_3$ and osmotic damage to seedling establishment for different fertilizer products with a common anion, in the absence of a fertilizer anion accompanying NH$_4^+$ and for fertilizer products with large differences in osmotic effects.

In the absence of NH$_3$ toxicity, osmotic effects delayed and occasionally inhibited germination. Crops differed significantly for osmotic tolerance, cotton, maize and sorghum being more tolerant than sunflower or soybean and wheat being more tolerant than canola.

Species were ranked according to tolerance for fertilizer product averaged over 5 application rates, and according to the fertilizer rate at which there was significant seedling establishment damage. Species rank for NH$_3$ toxicity and osmotic effects was similar across experiments but the maximum fertilizer rate tolerated changed with experimental conditions. Opportunities to avoid establishment damage are discussed.

6.1 INTRODUCTION

Effects of NH$_3$ toxicity (Allred and Ohlrogge 1964; Colliver 1969; Bennett and Adams 1970b; Woodstock and Tsao 1986) and osmotic pressure (Dubetz et al. 1959; Carter 1964; Young et al. 1968) from fertilizer applied with seed have been long been suspected to reduce germination and/or crop establishment. It is difficult to distinguish between the effects of NH$_3$ toxicity and osmosis because the NH$_3$-NH$_4^+$ complex in the soil solution could also create an osmotic effect on seeds. Compound fertilizers such as MAP and DAP containing NH$_4^+$ and other anions may therefore exert combined effects of NH$_3$ toxicity and osmotic pressure to affect germination and establishment.

Three experiments were conducted to explore the effects of NH$_3$ toxicity and osmotic pressure on previously studied crops that represent a wide range of NH$_3$ tolerance. For Experiment 6-1, fertilizer products MAP, DAP and TSP was selected to have different concentrations of NH$_4^+$, with the same accompanying anion species, phosphate.
TSP, containing no NH$_4^+$, was thought to primarily exert an osmotic effect although in some species there is evidence that the phosphate ion may cause toxicity (Scott 1989). MAP, although containing NH$_4^+$, is thought to have a relatively low potential for NH$_3$ toxicity resulting from its low NH$_4^+$ concentration and low solution pH. Low solution pH within the MAP fertilizer band creates a balance between NH$_3$ and NH$_4^+$ in favour of non-toxic NH$_4^+$ (Vines and Wedding 1960; Moody et al. 1995b). DAP is a product similar to MAP but is recognised as having a higher NH$_3$ toxicity potential due to the higher NH$_4^+$ concentration, and an alkaline pH in solution. Alkalinity within the fertilizer band favours formation and persistence of the toxic NH$_3$ molecule.

The chemistry within the reaction zone may be modified by reactions between banded NH$_4^+$-fertilizers and the surrounding soil. Some reactions in fertilizer bands include a reduced solution and exchangeable Ca and Mg concentration in the presence of DAP, increased soil solution Si and organic C, and reduced calcium activity ratio for both MAP and DAP (Moody et al. 1995b). Moody et al. (1995b) also found that the reduction in the calcium activity ratio due to MAP and DAP had a significant negative effect on root elongation due to Ca deficiency, possibly a factor in post-germination crop establishment failure.

Fertilizers increase soil salt concentration in the application zone hence osmotic pressure of the soil solution. An index of typical increases in soil osmotic pressure from fertilizer materials, salt index, was proposed by Rader et al. (1943). Salt index ranked the osmotic pressure of fertilizers in soil types as compared to sodium nitrate. The salt indices of various fertilizers tested in different soil types differed due to adsorption and precipitation reactions.

Osmotic effects within fertilizer bands were simulated in Experiment 6-2 by applying mannitol, an osmotic compound not translocated into the seed thus excluding confounding effects on germination and seedling establishment of accompanying anions associated with fertilizers. Ammonia toxicity was simulated by the addition of NH$_4$OH to the seed placement zone.
Osmotic effects during germination and seedling establishment were further examined in the field (Experiment 6-3) using 2 fertilizers, MAP and potassium chloride (KCl) with significantly different osmotic effects and soil mobility (Rader et al. 1943).

6.2 MATERIALS AND METHODS

Experiment 6-1 - Species Tolerance to Fertilizer Products MAP, DAP and TSP.

The experiment was conducted under a rain-exclusion structure at Incitec Fertilizers, Toowoomba, Queensland, using trays of a heavy clay soil collected at Formartin, Queensland. More detailed soil chemical characteristics are described in General Materials and Methods, Chapter 3.

Five kg of field moist soil was packed into trays 280 mm wide, 330 mm long and 75 mm deep. The trays were over-watered and allowed to dry until the soil moisture content was about 38 % w/w, suitable for sowing.

Four furrows 30 to 35 mm deep and 25 mm wide were made across each tray. Ten seeds were evenly spaced along each furrow and pressed lightly into the soil. Fertilizer (Table 6.1) was mixed with 10 to 15 grams of soil from the seed furrow and the soil-fertilizer mix was spread evenly along the furrow on top of the seed, and the furrow filled with the remaining soil. Soil in the furrow was lightly pressed to ensure good seed-soil contact.

Sown trays were placed in polyethylene bags and sealed for 3 days until first seedling emergence. All trays were then removed from the bags and subsequently were uncovered for the remainder of the experiment.

Fertilizer products TSP (20.7 % P, 1.4 % S, 15 % Ca, salt index = 17), MAP (10 % N, 21.9 % P, 2 % S, salt index = 30) and DAP (18 % N, 20 % P, 1.7 % S, salt index = 35) were sieved to include only granules with diameters between 2.0 mm and 2.8 mm.
Crop species tested were maize (cv. Dekalb 689), cotton (cv. Siokra V15), sorghum (cv. Pacific Seeds MR Buster), sunflower (cv. Pacific Seeds Hysun 46) and soybean (cv. Pacific Seeds Warrigal).

Emerged seedlings were counted 3, 4, 5, 6, 7, 9 and 14 days after sowing (DAS).

Table 6.1  Five fertilizer application rates for Experiment 6-1 expressed as rates of P and equivalent product rates.

<table>
<thead>
<tr>
<th>P Rate kg/ha</th>
<th>TSP kg/ha</th>
<th>MAP kg/ha</th>
<th>DAP</th>
<th>DAP g/m</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>5.5</td>
<td>27</td>
<td>2.0</td>
<td>25</td>
<td>1.9</td>
</tr>
<tr>
<td>11</td>
<td>53</td>
<td>4.0</td>
<td>50</td>
<td>3.8</td>
</tr>
<tr>
<td>22</td>
<td>106</td>
<td>8.1</td>
<td>100</td>
<td>7.7</td>
</tr>
<tr>
<td>44</td>
<td>213</td>
<td>16.2</td>
<td>201</td>
<td>15.3</td>
</tr>
</tbody>
</table>

All trays were rewatered 11 DAS, by which time moisture had declined to 60% of soil moisture at sowing. Establishment plateaued in the majority of treatments by 14 DAS, at which time trays were watered and seeds that had germinated but had not emerged were recovered from the soil, examined and counted.

Three replicates of each treatment were allocated to trays in a completely randomised design and positioned on a bench in a rain-exclusion shelter. The position of each tray on the bench was randomly reallocated 2, 4, and 6 DAS. Air temperature at the height of the soil surface was from 22°C to 37°C.

To enable species comparisons, establishment data within each experimental replicate was transformed to a relative scale (%) by dividing establishment for each application rate by the establishment for the nil application rate for the appropriate fertilizer product.

**Fertilizer Band Measurements**

Trays with fertilizer treatments applied as described above but not seeded, were established to enable soil analysis for $\text{NH}_4^+$-N, pH, and electrical
conductivity (EC). EC measurements were converted to saturated extract (EC_{SE}) equivalence by adjusting for soil texture as suggested by Shaw (1984).

Soil samples were collected 2 DAS taking a 25 mm diameter vertical core, centred over fertilized rows, to the full depth of the tray. Six cores were taken from each fertilizer row, composited in a plastic bag and frozen.

Soil from each treatment was thawed and divided into 2 sub-samples prior to chemical analysis. One sub-sample was prepared for NH_{4}^{+}-N analysis using the “field moist” procedure described in Chapter 3. Soil moisture, pH and EC were measured on the other sub-sample, after oven drying at 105 °C, using the methods of Rayment and Higginson (1992) for 1:5 soil/water suspension.

**Experiment 6-2 - Simulating ammonia toxicity and osmotic pressure within a fertilizer band.**

Experiment 6-2 was conducted in a controlled climate cabinet to maintain temperature and relative humidity conditions within narrow ranges; the temperature was kept constant at 15 ± 2 °C and the relative humidity was 85 ± 5%. Soil moisture was maintained at about field capacity (35 % w/w) and deionised water was added by weight to ensure constant osmotic pressure in the pots.

The experiment was conducted in 200 mm diameter black plastic pots lined with a water-tight plastic bag to prevent water loss through drainage. Each pot was first filled with 2 kg of air dry soil described as Lawes soil in Chapter 3. One kg of soil was added to each pot after being thoroughly mixed with NH_{4}OH, diluted with deionised water to make the total volume of solution for each treatment 100 mL. Seeds were sown approximately 10 mm deep and the soil drenched with 1100 mL of water containing the predetermined concentration of mannitol to produce the target osmotic pressure.
Treatments consisted of 0, 500 and 1500 mg/kg NH₄⁺-N and osmotic pressures 0, -0.6 and -1.2 MPa. Soil moisture was close to field capacity after the treatments were imposed.

Concentrations of mannitol solution required to produce the desired osmotic pressure (P) were calculated using a derivation of Universal gas equation:

\[
\text{mannitol (g/L) } = \frac{PVm}{RT}
\]

where
V = volume (L)
m = molecular weight of mannitol (182.7)
R= Universal gas constant (0.08205 Litre atmos./degree/mole)
T = temperature (K) (K= 273.15 + 15)
P= osmotic pressure ( MPa= 0.1 x bars)

Background osmotic pressure of the soil was -0.02 MPa, calculated using the equation of Marschner (1995a):

\[-\pi \text{ (MPa) } = EC_{se} \times 0.036\]

The experiment was conducted as a randomised complete block design with a factorial combination of treatments which were duplicated. Five seeds each of canola and wheat were sown in each pot, and establishment was recorded every 2 days until plant number was constant for the majority of treatments.

Establishment rate was the time taken for species in each treatment to reach maximum establishment. If for any treatment all seeds failed to emerge, the time taken for establishment was set at 22 days, the maximum observation time for the experiment.

**Experiment 6-3 - Osmotic effects on cotton establishment in the field with different fertilizers.**
A field experiment was conducted on the Lawes vertosol soil, referred to in Chapter 3, using equipment described in detail in Chapter 3 to place seed and fertilizer together down 2 sowing tines.
Factorial combinations of 0, 4, 8, 16, 32 g/m of MAP (10% N, 21.9% P, salt index = 30) and 0, 2.5, 5, 10 g/m of KCl (50% K, 50% Cl, salt index = 116) were applied in contact with seed at sowing. Eighty cotton seeds were sown along each 5 m plot length. Emerged plants were counted at 2 or 3 day intervals, from 2 to 22 days after first establishment (DAE). Plants were considered established only after the cotyledons were fully expanded.

Soil moisture was 25 % (w/w) 50 to 100 mm below the surface on the day of sowing. Soil temperature was monitored at seeding level for several days prior to sowing to ensure it was above the minimum, 17°C, suggested for satisfactory cotton establishment (Mills et al. 1997).

6.3 RESULTS
Experiment 6-1 - Species Tolerance to Fertilizer Products MAP, DAP and TSP.
Soil moisture at sowing was 46 % (w/w), which is higher than ideal for a commercial sowing operation on this soil due to poor trafficability. Sowing at such a high moisture content was favoured to minimise the effect of soil matric potential on germination.

Emergence of all species; 99 % for maize, 96 % for cotton, 78 % for sorghum, 94 % for sunflower and 82 % for soybean for the nil-fertilizer controls were high when compared within commercially acceptable levels (Mills and McIntyre 1997).

Emerged seedlings were visually examined for effects of toxicity, osmotic effects on seedling growth and population reduction 1 week after trays were rewatered on day 9. No symptoms of growth inhibition or seedling mortality were evident during this period.

The percentage of germinated seeds was generally higher than the percentage that established. Germinated, but unemerged seeds represented the seed population that was affected but not killed by exposure to fertilizer. Seedling
emergence for most species peaked about 9 DAS. Seedlings were only counted when leaves were clearly visible above the soil surface.

Establishment was reduced most severely by DAP and MAP when averaged over species and fertilizer rates (Table 6.2). Ranking of the crop species for their tolerance to co-placed fertilizer and seed was generally similar for different fertilizer products. Tolerance ranking for TSP was maize = sorghum = cotton > sunflower > soybean ($P<0.05$). Ranking for MAP was cotton = maize = sorghum > sunflower = soybean and for DAP was maize = sunflower > cotton > sorghum = soybean.

Another distinguishing feature of species establishment response to TSP, MAP and DAP was the fertilizer application rate where significant establishment differences occurred for each species. Significant reduction in emergence was indicated by a 19 % difference between product-rate-species interaction means (Table 6.2).

No maximum fertilizer rate for safe establishment of maize was found for DAP or MAP. Establishment was not significantly reduced at the highest rate of DAP or MAP applied. For maize the critical rate for TSP was 16.2 g/m. Overall effects of MAP, DAP or TSP on reduced establishment, although statistically significant ($P<0.05$), were only small.

The critical level for DAP in cotton was 4.2 g/m, whereas the highest rates of MAP or TSP applied did not significantly reduce establishment. Cotton establishment was reduced by 23 % by 4.2 g/m of DAP, a further decline of 47 % occurred when the DAP rate increased to 16.2 g/m.

Critical fertilizer rates for sorghum were 8.1 g/m for TSP and 7.7 g/m for MAP, but DAP was tolerated at a lower rate, 4.2 g/m. Reduction in establishment of sorghum at the critical product rates were 52 % for DAP, 54 % for MAP and 43 % for TSP.
Soybean was intolerant of MAP or DAP; establishment was significantly ($P<0.05$) reduced at the lowest rates applied. Response of soybean to TSP suggest that 2 g/m of TSP can be applied safely in contact with seed. Reduced establishment of soybean at the critical fertilizer rate was about 30% for MAP or DAP. At the highest rates of MAP or DAP, establishment was reduced by 90% (Table 6.2).

Table 6.2 Relative establishment (%) of cotton, maize, sorghum, soybean and sunflower resulting from application of MAP, DAP and TSP at 5 rates with the seed and applied to trays of soil in a rain-out shelter.

<table>
<thead>
<tr>
<th>Product</th>
<th>Rate (g/m)</th>
<th>Species</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>cotton</td>
</tr>
<tr>
<td>DAP</td>
<td>0</td>
<td>100</td>
</tr>
<tr>
<td></td>
<td>2.1</td>
<td>90</td>
</tr>
<tr>
<td></td>
<td>4.2</td>
<td>90</td>
</tr>
<tr>
<td></td>
<td>8.4</td>
<td>77</td>
</tr>
<tr>
<td></td>
<td>16.8</td>
<td>30</td>
</tr>
<tr>
<td></td>
<td>mean</td>
<td>77</td>
</tr>
<tr>
<td>MAP</td>
<td>0</td>
<td>100</td>
</tr>
<tr>
<td></td>
<td>1.9</td>
<td>90</td>
</tr>
<tr>
<td></td>
<td>3.8</td>
<td>100</td>
</tr>
<tr>
<td></td>
<td>7.7</td>
<td>95</td>
</tr>
<tr>
<td></td>
<td>15.3</td>
<td>83</td>
</tr>
<tr>
<td></td>
<td>mean</td>
<td>94</td>
</tr>
<tr>
<td>TSP</td>
<td>0</td>
<td>100</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>100</td>
</tr>
<tr>
<td></td>
<td>4</td>
<td>93</td>
</tr>
<tr>
<td></td>
<td>8.1</td>
<td>67</td>
</tr>
<tr>
<td></td>
<td>16.2</td>
<td>89</td>
</tr>
<tr>
<td></td>
<td>mean</td>
<td>89</td>
</tr>
</tbody>
</table>

I.s.d. $(P=0.05)$

- Product 4
- Species 5
- Rate 5
- Product x Species 9
- Rate x Species 11
- Product x Rate x Species 19
The critical product application rate in sunflower was 8.4 g/m for DAP, 1.9 g/m for MAP and 8.1 g/m for TSP. Although the reduction of sunflower establishment from MAP was significant at 1.9 g/m, establishment only declined by a further 5% for MAP rates between 1.9 and 7.7 g/m and was similar to establishment % for equivalent DAP and TSP rates.

**Ammonia toxicity and osmotic effects resulting from fertilizer applications with seeds of a variety of crop species.**

A multiple linear regression analysis was conducted to separate the species according to their response to NH$_3$ toxicity or osmotic pressure. A relative osmotic index (ROI) derived from the salt index and application rate of a fertilizer or mixture of fertilizers (Chapter 3 section 6) and NH$_4^+$-N were used as independent variables and crop establishment as the dependent variable. The regression equation (establishment % = 17.42 - 4.03 NH$_4^+$-N - 0.0028 ROI $R^2=0.51$) was significant ($P<0.001$).

Species response to NH$_3$ toxicity or osmosis was inferred by the significance of the t-statistic for the regression slope ($t=0.2$); species significantly affected were separated according to the slope of establishment decline (Table 6.3).

According to the regression analysis maize was unaffected by the NH$_4^+$-N rate or osmotic pressure exerted by the rates of DAP, MAP and TSP applied.

Cotton and sorghum showed a significant negative establishment response to NH$_4^+$-N rate but their response to ROI was not significant. Sunflower and soybean establishment was significantly correlated to ROI only (Table 6.3). The slope of the soybean response to ROI showed that establishment declined more rapidly than that of sunflower in response to osmotic pressure. The tolerance of soybean to NH$_4^+$-N, relative to maize, is consistent with the findings of Woodstock and Tsao (1986). Non-significance for NH$_4^+$-N and ROI for the regression analysis does not imply that species were tolerant of NH$_3$ or osmotic pressure, but it was thought that this analysis highlighted dominant effects for the crop species and fertilizer products tested.
Table 6.3  Slope values and t-probabilities for maize, cotton, sorghum, sunflower and soybean for regression analysis of NH$_4^+$-N rate, relative osmotic index and seedling emergence.

<table>
<thead>
<tr>
<th>Species</th>
<th>NH$_4^+$-N Slope</th>
<th>t - prob.</th>
<th>Relative osmotic index Slope</th>
<th>t - prob.</th>
</tr>
</thead>
<tbody>
<tr>
<td>maize</td>
<td>0.05</td>
<td>ns</td>
<td>-0.0011</td>
<td>ns</td>
</tr>
<tr>
<td>cotton</td>
<td>-1.31</td>
<td>0.03</td>
<td>-0.0015</td>
<td>ns</td>
</tr>
<tr>
<td>sorghum</td>
<td>-1.34</td>
<td>0.03</td>
<td>-0.0014</td>
<td>ns</td>
</tr>
<tr>
<td>sunflower</td>
<td>-0.23</td>
<td>ns</td>
<td>-0.0016</td>
<td>0.16</td>
</tr>
<tr>
<td>soybean</td>
<td>0.56</td>
<td>ns</td>
<td>-0.017</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

Ammonium-N, pH and Electrical Conductivity in Fertilizer Bands

Electrical conductivity (<4 dS/m) and soil NH$_4^+$-N (<1 mg/kg) for the nil-fertilizer control treatments were not high enough to significantly affect crop establishment (Fig. 6.2 and 6.3).

Soil pH for the DAP application band was consistently higher than MAP and TSP for all rates greater than 2 g/m (Fig. 6.1), and as application rate increased, pH for all products declined. The rate of decline in soil pH was greater for MAP and TSP than for DAP.

Electrical conductivity of the fertilizer application zone was measured to provide information about osmotic effects of the fertilizers. DAP and MAP both increased EC significantly more than TSP as fertilizer application rates were increased (Fig. 6.2). The EC difference between MAP and DAP was not significant. This is consistent with findings of Rader et al. (1943) who assigned similar salt indices of 30 for MAP and 35 for DAP.

Ammonium-N concentration within application bands was significantly different between different fertilizer products (Fig. 6.3). The increase in soil NH$_4^+$-N was in proportion to the quantity of N added as fertilizer product, after adjusting for background soil NH$_4^+$-N concentration. DAP rates applied produced soil NH$_4^+$-N concentrations of 381 to 2197 mg/kg, while concentrations from MAP rates were in the range 166 to 1225 mg/kg.

The electrical conductivity at which establishment was significantly reduced for soybean, sunflower and sorghum was similar for different fertilizer products.
However, sorghum (>6 dS/m) tolerated a higher EC than sunflower or soybean(<6 dS/m) (Table 6.4).

**Fig. 6.1** Soil pH of fertilizer application zones 2 days after application of 4 rates of MAP (%), DAP (©) and TSP (+). Error bar represents the l.s.d. value ($P=0.05$) for the interaction between fertilizer product and application rate.

**Fig. 6.2** Soil $EC_{se}$ of fertilizer application zones 2 days after application of 4 rates of MAP (%), DAP (©) and TSP (+). Error bar represents the l.s.d. value ($P=0.05$) for the interaction between fertilizer product and application rate.
Fig. 6.3  Soil NH$_4^+$-N concentration (mg/kg) of fertilizer application zones 2 days after application of 4 rates of MAP (%), DAP (I) and TSP (+). Error bar represents the l.s.d. value ($P=0.05$) for the interaction between fertilizer product and application rate.

Critical EC$_{se}$ for cotton and maize differed for different fertilizer products, and were higher for MAP and DAP (>6 dS/m) than for TSP (<6 dS/m).

Critical soil NH$_4^+$-N concentration for species was generally in the same range or higher for MAP than DAP. The exception was sunflowers for which DAP was 1896 mg/kg NH$_4^+$-N and MAP was less than 166 mg/kg (Table 6.4).

**Experiment 6-2 - Simulating ammonia toxicity and osmotic pressure of a fertilizer band.**

Final establishment counts of canola and wheat seedlings were taken 22 DAS, 16 days after first plants emerged, and when the number of plants established was constant for the majority of pots. In untreated soil, final establishment of each species was above 90 %. To enable species comparisons, data was transformed to percentage establishment of untreated soil for each species.
Table 6.4 Comparing critical product rate established from l.s.d. \((P=0.05)\), soil \(\text{NH}_4^+\)-N concentration and \(\text{EC}_{se}\) for MAP, DAP and TSP applied to cotton, maize, sorghum, soybean and sunflower.

<table>
<thead>
<tr>
<th>Species</th>
<th>Product rate</th>
<th>(\text{NH}_4^+)-N</th>
<th>(\text{EC}_{se})</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>DAP (g/m)</td>
<td>MAP (mg/kg)</td>
<td>TSP (dS/m)</td>
</tr>
<tr>
<td>cotton</td>
<td>4.2</td>
<td>&gt;15.3</td>
<td>&gt;16.2</td>
</tr>
<tr>
<td>maize</td>
<td>&gt;16.8</td>
<td>&gt;15.3</td>
<td>8.1</td>
</tr>
<tr>
<td>sorghum</td>
<td>4.2</td>
<td>7.7</td>
<td>8.1</td>
</tr>
<tr>
<td>soybean</td>
<td>&lt;2.1</td>
<td>&lt;2.1</td>
<td>2</td>
</tr>
<tr>
<td>sunflower</td>
<td>8.4</td>
<td>1.9</td>
<td>8.1</td>
</tr>
</tbody>
</table>

Increasing the soil \(\text{NH}_4^+\)-N concentration from the background concentration \(<2\) mg/kg to 500 mg/kg significantly \((P<0.05)\) reduced establishment. Canola establishment was reduced from 100 % to 40 % and wheat from 100 % to 80 %. Increasing soil \(\text{NH}_4^+\)-N concentration to 1500 mg/kg completely inhibited establishment of both species (Table 6.5).

Canola was significantly \((P<0.05)\) less tolerant of high osmotic pressure than wheat. At -0.6 MPa osmotic pressure, establishment of canola was 40 % of the untreated soil, whereas at the highest osmotic pressure (-1.2 MPa) canola completely failed to emerge. Wheat establishment was also significantly reduced by high osmotic pressure, but at -1.2 MPa, wheat establishment remained at 50 % of that for untreated soil.

Wheat was more tolerant of combinations of high \(\text{NH}_4^+\)-N and high osmotic pressure than canola.

The time taken to reach maximum canola establishment was not significantly reduced by \(\text{NH}_4^+\)-N at 500 mg/kg and -0.6 MPa osmotic pressure although a small population emerged (Table 6.6).
Table 6.5  Relative seedling establishment (% of nil control) for canola and wheat sown into containers of soil treated with 3 rates of NH$_4^+$-N and exhibiting 3 levels of osmotic pressure.

<table>
<thead>
<tr>
<th>NH$_4^+$-N (mg/kg)</th>
<th>Osmotic Pressure (MPa)</th>
<th>canola</th>
<th>wheat</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0</td>
<td>-0.6</td>
<td>-1.2</td>
</tr>
<tr>
<td>0</td>
<td>100</td>
<td>40</td>
<td>0</td>
</tr>
<tr>
<td>500</td>
<td>40</td>
<td>20</td>
<td>0</td>
</tr>
<tr>
<td>1500</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>mean</td>
<td>47</td>
<td>20</td>
<td>0</td>
</tr>
</tbody>
</table>

l.s.d. ($P=0.05$)

- NH$_4^+$-N: 4.2
- osmotic pressure: 4.2
- NH$_4^+$-N x species: 5.9
- osmotic pressure x species: 5.9
- NH$_4^+$-N x osmotic pressure x species: 10.2

The time taken to reach maximum establishment for wheat was significantly slowed when osmotic pressure was increased from -0.6 MPa to -1.2 MPa at both zero and 500 mg/kg NH$_4^+$-N, but at -0.6 MPa, establishment rate was not significantly different from the untreated control.

**Experiment 6-3 - Comparing cotton establishment in the field with fertilizers of different osmotic effects.**

Low soil moisture content (25% w/w) at sowing resulted in final establishment of 34 plants/5 m in the zero fertilizer treatment, just within the target range of 4 to 7 plants/m of row (Table 6.7), acceptable for rain-grown commercial cotton production, but below 9 to 12 plants/m normally accepted for irrigated cotton (McIntyre and Marshall 1994). Results reported in Table 6.7 are for final crop establishment, 17 DAE, after which there was no further emergence.
Table 6.6  Time taken (days) to reach maximum establishment of canola and wheat sown into containers of soil treated with 3 rates of NH$_4^+$-N and 3 levels of osmotic pressure (n.e.= no emergence).

<table>
<thead>
<tr>
<th>NH$_4^+$-N (mg/kg)</th>
<th>Osmotic Pressure (MPa)</th>
<th>canola</th>
<th>wheat</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0</td>
<td>-0.6</td>
<td>-1.2</td>
</tr>
<tr>
<td>0</td>
<td>7</td>
<td>4</td>
<td>n.e.</td>
</tr>
<tr>
<td>500</td>
<td>6</td>
<td>6</td>
<td>n.e.</td>
</tr>
<tr>
<td>1500</td>
<td>n.e.</td>
<td>n.e.</td>
<td>n.e.</td>
</tr>
<tr>
<td>mean (0 and 500 only)</td>
<td>6.5</td>
<td>5</td>
<td>n.e.</td>
</tr>
</tbody>
</table>

l.s.d. (P=0.05)

<table>
<thead>
<tr>
<th>NH$_4^+$-N</th>
<th>3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Osmotic Pressure</td>
<td>3</td>
</tr>
<tr>
<td>NH$_4^+$-N x species</td>
<td>4</td>
</tr>
<tr>
<td>Osmotic pressure x species</td>
<td>4</td>
</tr>
<tr>
<td>NH$_4^+$-N x osmotic pressure x species</td>
<td>7</td>
</tr>
</tbody>
</table>

MAP at rates above 4 g/m (NH$_4^+$-N rate of 0.4 g/m) without KCl significantly \((P<0.05)\) reduced the number of established plants. In the absence of MAP, applying KCl at rates above 2.5 g/m reduced crop establishment significantly \((P<0.05)\).

Regression analysis for combinations of MAP and KCl rates on cotton establishment 17 DAE showed the combination of individual effects of MAP and KCl was no more damaging to establishment than the effect of KCl or MAP alone. Slope values for the individual regression equations were -0.954 \((t<0.01)\) for KCl and -0.498 \((t<0.001)\) for MAP \((r^2=0.549)\). The greater slope value for KCl suggests that KCl is more damaging to cotton establishment than MAP per unit weight of product.
Table 6.7 Establishment of cotton resulting from seed-furrow placement of 5 rates of MAP and 4 rates of KCl fertilizer, 17 DAE.

<table>
<thead>
<tr>
<th>KCl rate (g/m)</th>
<th>MAP rate (g/m)</th>
<th>0</th>
<th>2.5</th>
<th>5</th>
<th>10</th>
<th>mean</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td></td>
<td>34.0</td>
<td>15.0</td>
<td>12.5</td>
<td>8.0</td>
<td>17.4</td>
</tr>
<tr>
<td>4</td>
<td></td>
<td>26.5</td>
<td>6.5</td>
<td>8.5</td>
<td>8.5</td>
<td>12.5</td>
</tr>
<tr>
<td>8</td>
<td></td>
<td>13.5</td>
<td>9.5</td>
<td>5.0</td>
<td>5.5</td>
<td>8.4</td>
</tr>
<tr>
<td>16</td>
<td></td>
<td>4.0</td>
<td>2.5</td>
<td>2.0</td>
<td>0</td>
<td>2.1</td>
</tr>
<tr>
<td>32</td>
<td></td>
<td>1.0</td>
<td>0</td>
<td>1.0</td>
<td>1.0</td>
<td>0.8</td>
</tr>
<tr>
<td>mean</td>
<td></td>
<td>15.7</td>
<td>6.9</td>
<td>7.2</td>
<td>4.9</td>
<td></td>
</tr>
</tbody>
</table>

I.s.d. (P=0.05)
- MAP rate: 4.2
- KCl rate: 3.7
- MAP X KCl: 8.4

Non-linear regression analysis was conducted fitting an exponential curve of the general form \( y = a + b r^x \) for plant establishment and ROI using the non-linear routine of Genstat 5 (The Numerical Algorithms Group Ltd, 1996). There was a rapid decline in crop establishment as the ROI increased (Fig. 6.4). For the soil moisture conditions at sowing, it is probable that the final cotton population would be lower than commercially acceptable for rain-grown cotton for a ROI higher than 100 which is equivalent to about 3 g/m MAP or 1 g/m KCl.

Data presented in Fig. 6.5 suggest that fertilizer in contact with germinating seed not only affect final plant population, but also affects crop establishment rate. Emergence rate was estimated as the ratio of plants 3 DAE as a proportion of those present 17 DAE. High establishment ratios such as those for the zero fertilizer treatments (70%) indicates a high proportion of seedlings present at 17 DAE were present 3 DAE i.e. there was rapid early establishment. MAP at rates above 8 g/m significantly reduced the establishment ratio, whereas KCl reduced establishment at rates above 5 g/m. At the highest rates of both fertilizers, the establishment ratio declined to about 20 %. The interaction between MAP and KCl was not significant indicating that rate of establishment was related to the greater effect of MAP or KCl individually or that cottons’ response to these products is due to a common process affecting the crops’ emergence.
**Fig. 6.4**  The relationship between ROI derived from a combination of 5 rates of MAP and 4 rates of KCl fertilizer and plant establishment in cotton, Experiment 6-3. (Plants/5m = 3.48 + (30.76x(0.996 ROI)) R²=0.76).

**Fig. 6.5**  Rate of establishment of cotton seedlings (3 DAE/17 DAE) resulting from application of 0, 4, 8, 16, 32 g/m of MAP (I) (means of 4 rates of KCl) or 0, 2.5, 5, 10 g/m of KCl (%) (means of 5 rates of MAP) in the seed furrow. Error bars represents the l.s.d. (P=0.05) value for MAP or KCl mean.
Linear regression analysis of ROI and establishment ratio means, meaned across all levels of MAP or KCl, indicated that there was a close relationship between ROI and establishment ratio; $R^2=0.88$, $P<0.001$.

### Table 6.8 Accumulated ANOVA, mean squares (m.s.), variance ratio (v.r.) and $F$ probability ($F$ (pr.)) for the regression of ROI and cotton establishment rate, grouped for fertilizer product.

<table>
<thead>
<tr>
<th>Change</th>
<th>m.s.</th>
<th>v.r.</th>
<th>$F$ (pr.)</th>
</tr>
</thead>
<tbody>
<tr>
<td>ROI</td>
<td>2372.6</td>
<td>57.1</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Fertilizer</td>
<td>25.83</td>
<td>0.62</td>
<td>0.47</td>
</tr>
<tr>
<td>ROI x Fertilizer</td>
<td>47.1</td>
<td>1.13</td>
<td>0.34</td>
</tr>
</tbody>
</table>

The strong correlation between ROI and establishment rate with similar slopes for MAP and KCl (Table 6.8) suggests that the majority of the delay of cotton establishment was osmotically induced rather than a result of NH$_3$ toxicity, as would have been the case if MAP reduced establishment rate faster than KCl.

### 6.4 DISCUSSION

Lower tolerance of crops to seed-furrow placement of DAP than MAP or TSP for Experiment 6-1 is consistent with the findings of Nyborg (1961), Allred and Ohlrogge (1964) and Hood and Ensminger (1964). Allred and Ohlrogge (1964) concluded that lower tolerance of seeds to DAP than MAP was a result of higher solubility of DAP and the presence of an easily hydrolysable secondary NH$_4^+$ radical liberated from DAP. Ammonium-N concentration and pH of the fertilizer band were higher for DAP than for the other fertilizers, suggesting that the NH$_3$-NH$_4^+$ balance was likely to have favoured toxic NH$_3$ (Du Plessis and Kroontje 1964). Whitehouse and Leslie (1973) found that for an application of 163 kg/ha N as urea or NH$_3$ (alkaline NH$_4^+$-fertilizers), after 7 days soil pH declined by 1.3 to 1.6 as a result of nitrification. The more modest decrease in soil pH for DAP may therefore have resulted from nitrification at the edges only of the fertilizer band included in soil samples as a result of the sampling technique.
All species tested, except for soybean and sunflower, were highly tolerant of TSP during germination. Species with low tolerance to TSP presumably have low tolerance to the osmotic effect of fertilizer or alternatively may be sensitive to P toxicity. Dart et al. (1992) found that soybean and sunflower were more sensitive to low water potential than sorghum and cited that the moisture requirement of sunflower, on a w/w basis, was about double that of sorghum. They also found that high soil temperature further reduced or delayed germination and emergence. The sensitivity of soybeans to osmotic pressure (as indicated by response to TSP) in this study is similar to that described by Moody et al. (1995a, 1995b) who showed that soybean root elongation was reduced by 10 % where the EC$_{se}$ was 4.1 dS/m. In Experiment 6-1 soybean establishment was found to be limited at TSP rates greater 2 g/m which corresponded to EC$_{se}$ values greater 4 dS/m.

An examination of unemerged seeds recovered from TSP and MAP treatments indicated that there had been metabolic activity in the seed, swelling or elongation of the radicle, but germination had not proceeded to coleoptile elongation or cotyledon emergence. Bewsley and Black (1978) indicated that osmotic competition for water from the finish of imbibition to just before radicle elongation, if severe, would inhibit germination, but if moderate would significantly slow the germination process. Observations from TSP and MAP treatments were consistent with germination response to osmotic effects suggested by Bewsley and Black (1978).

Sorghum, cotton and soybean were more tolerant of TSP and MAP than of DAP. For a similar product application rate reduced establishment from DAP was generally greater than for MAP, suggesting that the characteristic of DAP causing germination reduction in these species was the production of NH$_3$. Ungerminated seed recovered from DAP treatments showed no signs of metabolic activity and internal structure was frequently in a state of decay, symptoms consistent with severe disruption of metabolic pathways typical of NH$_3$ toxicity.
Table 6.9 presents the NH$_4^+$-N rates at which significant establishment reductions occurred for each species of crop in Experiment 6-1. Sorghum and maize are the only species tested to which N application in the seed-row is recommended (0.45 g/m N). For sunflower and cotton, it is currently recommended that no N be applied with the seed (Mills and McIntyre 1997). The fertilizer rate at which tolerance to seed placed NH$_4^+$-fertilizer occurred in Experiment 6-1 was generally higher for all species than that for the rates currently recommended for the soil type, and for maize, NH$_4^+$ tolerance was higher in this study than tolerance reported by Colliver and Welch (1970a).

Critical application rates for DAP, MAP and TSP suggested from Experiment 6-1 were significantly higher than currently recommended and higher than the rates suggested by Gerwing et al. (1994) from a field experiment using the same fertilizer products, fertilizer rates and crop species. The high seeding rate used in experiments to satisfy statistical analysis requirements may have influenced the results. Ten seeds per 28 cm of row used for each species is significantly higher than the current recommended seeding rates which are:- for 28 cm of row, 2-4 seeds for maize, 3-5 seeds for cotton, 3-6 seeds for sorghum, 1-3 seeds for sunflowers and 5-8 seeds for soybeans. The difference in seeding rate between Experiment 6-1 and recommended rates in the field is notable in that species that showed the largest contrast in performance were species with the large differences between experimental and field seeding rates. These results suggest that a strategy for maintaining plant population within recommended levels, where the application of fertilizer may reduce emergence, is to increase seeding rate to offset seedling loss due to fertilizer damage.
Table 6.9  Ammonium-N rates from DAP and MAP in Experiment 6-1 and current field recommended rates for northern New South Wales and southern Queensland at which significant reduction in crop establishment occurred for cotton, maize, sorghum, soybean and sunflower (ni. = no information available, nr. = not recommended).

<table>
<thead>
<tr>
<th>Fertilizer Product</th>
<th>Crop Species</th>
<th>Experiment 6-1 $\text{NH}_4^+\text{-N} \ (\text{mg/kg})$</th>
<th>Recommended$^1$ $\text{NH}_4^+\text{-N} \ (\text{mg/kg})$</th>
</tr>
</thead>
<tbody>
<tr>
<td>DAP</td>
<td>cotton</td>
<td>0.75</td>
<td>0.4</td>
</tr>
<tr>
<td></td>
<td>maize</td>
<td>3.02</td>
<td>0.45</td>
</tr>
<tr>
<td></td>
<td>sorghum</td>
<td>0.75</td>
<td>0.45</td>
</tr>
<tr>
<td></td>
<td>soybean</td>
<td>0</td>
<td>ni.</td>
</tr>
<tr>
<td></td>
<td>sunflower</td>
<td>1.51</td>
<td>nr.</td>
</tr>
<tr>
<td>MAP</td>
<td>cotton</td>
<td>1.5</td>
<td>0.4</td>
</tr>
<tr>
<td></td>
<td>maize</td>
<td>1.5</td>
<td>0.45</td>
</tr>
<tr>
<td></td>
<td>sorghum</td>
<td>0.77</td>
<td>0.45</td>
</tr>
<tr>
<td></td>
<td>soybean</td>
<td>0</td>
<td>ni.</td>
</tr>
<tr>
<td></td>
<td>sunflower</td>
<td>0.19</td>
<td>nr.</td>
</tr>
</tbody>
</table>

($^1$ source: Mills et al. 1996 and Mills and McIntyre 1997)

Trends for species’ susceptibility to NH$_3$ toxicity or osmotic pressure could be recognised in their reaction to fertilizer products. Species more affected by DAP were thought to be less tolerant of NH$_3$ and species affected by TSP were thought to be sensitive to osmotic pressure. However, it was not until the species establishment data for each product was compared on a common scale, NH$_4^+$-N rate and ROI that a clear separation of species susceptibility to NH$_3$ toxicity and osmotic pressure was achieved. There were no factors in common or clear species grouping e.g. oilseed vs cereal that provided information about the tolerance mechanism to NH$_3$ or osmotic pressure.

The use of phosphate as an anion in common for comparison of species’ response to NH$_3$ toxicity and osmotic pressure did not exclude possible toxic or inhibitory effects of anions. Hood and Ensminger (1964) found that there were germination tolerance differences between several phosphate compounds, tolerance depended on the effect of the P compound on magnesium activity in enzymatic reactions. This effect was most pronounced for DAP.
It was demonstrated that in the absence of NH$_3$ toxicity, osmotic pressure alone can significantly reduce crop establishment and that there was a significant difference for species’ susceptibility. The significant reduction in canola establishment at -0.6 MPa osmotic pressure is in agreement with Williams and Shaykewich (1971), who found that the critical water potential for rapeseed was -0.3 MPa, while the higher tolerance of osmotic pressure for wheat is similar to the findings of Bouaziz and Bruckler (1989).

Different fertilizer compounds and blended fertilizer products create different risks to crop establishment, when placed with the seed at sowing. The final effect on establishment appears to depend on crop species’ sensitivity to NH$_3$ and osmotic pressure.

Separation of NH$_3$-NH$_4^+$ and osmotic pressure tolerance into individual effects for each species showed that canola is sensitive to both increased NH$_3$-NH$_4^+$ concentration and osmotic pressure. Hence in the field, restricting the fertilizer rate applied with canola seed, based solely on the N rate applied, may not avoid crop establishment reductions where the rate of fertilizer applied, soil salinity and soil moisture conditions could combine to exceed the osmotic pressure tolerance of the species.

Treatments that included high osmotic pressure and high NH$_4^+$ concentration together, resulted in establishment that paralleled the most extreme of either the osmotic or NH$_4^+$ response. This confirms the need for caution with fertilizer products such as urea, having both a high NH$_3$ potential and a high salt index (Rader et al. 1943).

An increase in emergence time is rarely taken into account when assessing the risk of applying fertilizer at sowing, or when assessing the cause of poor crop establishment. Increased time to emerge can have significant effects on crop performance through exposure of the vulnerable shoots and roots to insect and disease attack, and sub-optimal growing conditions. For example, Wanjura (1991) found that when cotton establishment is delayed by up to 100 % of the normal time, lint yield reductions averaged up to 56 %.
Differences in critical rates of fertilizer product between experiments provided evidence that the tolerance level was significantly affected by experimental conditions particularly soil moisture status at sowing. Critical NH$_4^+$-N rates for cotton establishment varied from 0.4 g/m$^2$ for field conditions at low soil moisture, to 1.53 g/m$^2$ for controlled conditions and high soil moisture.

The results of the experiments presented in this chapter suggest that the ranking of species in their tolerance to NH$_4^+$-N and osmotic pressure is relatively stable across a reasonably wide range of sowing conditions. The main outcome that was affected by varying sowing conditions was the actual fertilizer rate tolerated by the species.

Although cotton was shown to be more tolerant of osmotic pressure than most other species, in the absence of NH$_3$ toxicity, there were osmotic limits as indicated by the severe restrictions in emergence to KCl application. High osmotic pressure from KCl was more damaging to cotton establishment than the combined effects of low osmotic pressure and low NH$_3$ per unit weight of product applied.

Establishment results for wheat, canola and cotton suggest that the effects of NH$_3$ toxicity and osmotic pressure act independently, final establishment reflecting each species’ response to the parameter least tolerated such as NH$_3$ toxicity or osmotic pressure.

Currently, recommendations of application of fertilizer in the seed-furrow do not provide critical application rates for fertilizers that contain no N, and there is no recognition of the different chemistry in application bands arising from the application of a range of N fertilizer products. Significant reductions in cotton establishment resulting from KCl and TSP, and strong correlation of ROI, establishment % and establishment rate highlighted deficiencies in the current method of determining damage potential from seed row applied fertilizers.

The ROI concept acknowledges that osmotic effects are a significant factor in germination response. Furthermore, it may provide a means to assess the
germination effects for a range of fertilizer products and rates that are applied to a variety of crop species within the northern cropping region of eastern Australia. It should be of value in decision support systems. More information about species ROI sensitivity to soil texture, moisture % and other soil characteristics is required before the index can be used more universally.

6.5 CONCLUSIONS

Techniques used to separate species’ sensitivity to NH$_3$ toxicity and osmotic pressure provided a clear rank for 5 summer crop species. Generally, the order of rank remained similar for the range of products tested and across the experimental conditions, however maximum fertilizer rate tolerated varied where there were significant differences between experimental techniques.

High seeding rates and high soil moisture modified the effect of high fertilizer rates on crop establishment for cotton, and it is highly likely that the response to seeding rate and soil moisture will be similar for other species. Fertilizer products were generally better tolerated where NH$_4^+$-N content and fertilizer pH was low, but high seeding rate and high soil moisture appeared to moderate the damage to crop establishment from high soil NH$_4^+$-N concentration.

To further investigate the opportunity to modify establishment response to high NH$_4^+$-N concentration and osmotic pressure close to seeds, further experiments were conducted and are recorded in Chapter 7. These were designed to assess the effectiveness of high seeding rate, water addition to the seed furrow and chemically controlling the microbial transformation of N fertilizer in soil, to increase establishment in the presence of high fertilizer rates applied with the seed.
Chapter 7
Practices for modifying the effects of seed furrow placement of fertilizer.

Summary
Fertilizer products are generally better tolerated by germinating seeds where a combination of fertilizer pH and NH$_4^+$-N content or osmotic effects are low. High seeding rates and high soil moisture may alleviate detrimental effects of high fertilizer rates on crop establishment.

Experiments were conducted to investigate the opportunity to moderate detrimental effects on crop establishment due to high NH$_4^+$-N and/or osmotic pressure. Increasing the seeding rate, increasing soil moisture in the seed furrow by water addition at sowing, or modifying the transformation of N fertilizer were studied.

Increasing seeding rate, adding water to the seed furrow at sowing and slowing the rate of N fertilizer hydrolysis successfully increased crop establishment at higher than recommended fertilizer rates. The agronomic and economic limits of these practices are discussed.

7.1 INTRODUCTION
Placing fertilizer salts in direct contact with seed may reduce the establishment of most crop species. The extent of damage depends on the NH$_3$ concentration produced (Allred and Ohlrogge 1964; Colliver and Welch 1970a), osmotic pressure exerted by the fertilizer product (George and Williams 1964; Carter 1967) and interactive effects occurring between soil, fertilizer and crop species. Specific ions released into the soil solution from fertilizer can create toxic effects and seedling damage (Olson and Dreier 1956; Nyborg 1961; Moody et al. 1995b).

Olson and Dreier (1956) found that fertilizer salt was most harmful to germination at low levels of soil moisture availability. They also concluded that soluble N-containing salts were more detrimental than K salts per unit weight, which in turn were more detrimental to germination than phosphate salts. Salt concentration in the soil solution reduces the amount of water that can be imbibed by the seed and retards germination. Germination may not necessarily be prevented by high salt concentrations, but may be retarded (Shrive 1917).
Germination retardation was directly related to reduction in the volume of water absorbed by the seed. Collier (1954) showed that most of the yield reduction caused by fertilizer placed with maize seed, was due to lower plant populations rather than reduced seedling and plant vigour caused by excessive salt concentration.

A sowing technique that is currently used for summer crops in Queensland and northwest NSW and that may help overcome the osmotic effect of fertilizer, is water injection. Water injection consists of applying 100 to 1000 L/ha of water to the seed furrow at sowing and was developed primarily to promote faster establishment of seedlings under drier than optimum sowing conditions and to carry crop protection products, fertilizers and legume inoculant (Ferraris 1992a).

Urea is the most common N fertilizer used across the cereal cropping areas of eastern Australia and is applied at various times and placements to suit the cropping system.

Nitrogen fertilizer is commonly the largest variable cash cost in many cropping systems. It has become a key input for profitability in a range of crops and cropping systems due to the run-down in native soil fertility (Dalal and Mayer 1986). In recent years, there has been a trend toward the application of N at sowing due to the few application windows in a zero tillage management system. It is an effective risk management strategy that avoids costly outlays for N fertilizer when dry conditions prevent sowing. With unreliable planting rain and narrower profit margins, there is an increasing trend to apply N fertilizer at the latest opportunity. In the northern part of the eastern Australian cereal belt, where a lack of reliable in-crop rainfall leads to extremely unreliable responses to topdressing or application post-sowing (Doyle and Shapland 1991), sowing is the latest opportunity to apply N fertilizer.

Urea, before its transformation to NH₄⁺ by hydrolysis, has the unique characteristics of having a purely osmotic effect on seed germination which Rader et al. (1943) rated to be in the mid to high range of the salt indices. After
hydrolysis to form $\text{NH}_4^+$, it is capable of exerting both an osmotic effect on seed germination as well as $\text{NH}_3$ toxicity.

Bremner and Krogmeier (1988) suggested that urease inhibitors could provide a solution to the problem of $\text{NH}_3$ toxicity caused by urea placed in close proximity to germinating seed. In a recent review of the urease inhibitor NBPT (N-(n-butyl) thiophosphoric triamide = Agrotain®\(^1\)) by Grant et al. (1996), it was cited that researchers in the USA and Canada had found that by delaying the conversion of urea to $\text{NH}_3$ by applying NBPT to the urea, establishment was maintained at high rates of seed-applied urea. These comments indicated that the urease inhibitor, NBPT, was capable of preventing the formation of toxic concentrations of $\text{NH}_3$ for a period of time, and the osmotic effects of the fertilizer on seed germination could be observed in the absence of the toxic effects of high $\text{NH}_3$.

As a result of these unique characteristics, it was proposed to compare the effects of urea on establishment, in the presence and absence of hydrolysis. This would enable the osmotic effect only (hydrolysis inhibited) to be compared with the combined effects of $\text{NH}_3$ toxicity and osmosis (with hydrolysis to $\text{NH}_4^+$), on seed germination. If $\text{NH}_3$ toxicity is the major inhibitory effect to germination, then it may be feasible to increase the safe rate of urea applied with the seed when urea hydrolysis is prevented or inhibited.

Experiments in this chapter investigate strategies for reducing detrimental effects of seed placed N fertilizer on crop establishment. One experiment (7-1) examined whether adding water to the seed furrow with fertilizer at sowing would decrease the effect of fertilizer on crop establishment. A second experiment (7-2) examined whether increasing seeding rate would produce a commercially acceptable plant population when seed-placed N fertilizer rate causes seedling damage. Urea and ammonium nitrate were compared with find whether chemical characteristics affect the toxicity of fertilizers containing $\text{NH}_4^+$. A third experiment (7-3) examined whether delaying hydrolysis of urea, by the

\(^1\) Agrotain® registered trade mark of IMC -Global Company, USA
addition of a urease inhibitory compound would increase the rate of urea that could be safely applied with seed.

7.2 MATERIALS AND METHODS

Sowing Equipment

Configuration of the sowing equipment for Experiments 7-1, 7-2, and 7-3 is described in detail in Chapter 3. For Experiment 7-1, MAP was dissolved in water prior to application. Water or fertilizer solution was delivered to the boot of 4 sowing tines spaced at 450 mm apart from 20 L polypropylene drums through a diaphragm pump. Combinations of water and fertilizer rate were achieved by changing fertilizer solution concentration, pump pressure and tractor speed.

For Experiments 7-2 and 7-3 the cone seeders and rotary dividers were configured to meter a predetermined number of seeds and weight of fertilizer material to 7 sowing tines spaced 225 mm apart for winter crops or to 4 tines spaced 450 mm apart for summer crops.

Site and Treatments

Experiment 7-1 - Increasing soil water to improve crop establishment for seed placed fertilizer.

Experiment 7-1 was conducted at the University of Queensland, Gatton College Field Research Station at Lawes, Queensland adjacent to the site of Experiment 4-2. Site and soil details are described in General Materials and Methods, Chapter 3.

A factorial combination of 0, 2, 4 or 8, g/m of MAP (12 % N, 26 % P) and 0, 10, 20 or 80 mL/m of water was applied in the seed furrow at sowing. MAP rates were within the range of rates commonly applied within the seed furrow of cotton and sorghum and were within the response range established for cotton in Chapter 6.
Seventy-five cotton seeds (cv. Deltapine 90) or 100 sorghum seeds (cv. Pioneer Pride) were sown in each 5 m long plot. Treatments were duplicated.

The site was irrigated with 100 mm of water before sowing and the soil moisture was monitored daily. Some plots were sown when moisture reached 35 % (w/w) whilst the others were sown when soil moisture had declined to 25 % (w/w), 10 days later.

Seedlings were counted 7, 9, 12, 15, 18 and 25 days after sowing (DAS). Maximum plant establishment was achieved by 18 days. There was some decline in establishment in some treatments due to fungal attack of cotton seedlings.

**Experiment 7-2 - Improving crop establishment with higher seeding rate or N fertilizers with different chemical characteristics.**

Experiment 7-2 was conducted at Dalby, Queensland. Site and soil characteristics are described in detail in Chapter 3.

Urea (46 % N) applied at rates of 0, 0.6, 1.1, 1.7 or 2.3 g/m (0, 25, 49, 76, 100 kg/ha) and ammonium nitrate (34 % N, 17 % as NH₄⁺-N; 17 % as NO₃⁻-N) applied at rates of 0, 1.3, 2.7, 4.1 or 5.5 g/m (0, 59, 119, 182, 246 kg/ha) contained NH₄⁺-N at rates equivalent to 0, 10, 20, 30 and 45 kg/ha.

Barley (cv. Grimmett) was sown at 25 or 40 kg/ha of seed. Factorial combinations of fertilizer products, fertilizer rates and seeding rates were applied in a completely randomised design. Each combination was duplicated.

Average counts of 5 one-metre randomly selected row lengths were taken for each of 4 sequential observations. Statistical analysis was conducted on the data for the 4th observation when emergence had maximised in all plots.
Experiment 7-3 - Delaying urea hydrolysis to increase the safe rate of urea applied with cotton and sorghum seed.

Experiment 7-3 was conducted at Formartin, Queensland. The field was the collection site for soil used for Experiment 6-1.

A measured quantity of seed, representing 15 seeds/m of row for sorghum (cv. Pacific Seeds Buster MR) or cotton (cv. Siokra 1-4), was placed into the cone seeder hopper with the fertilizer to facilitate close contact between the seed and the urea within the seed row. Seed and fertilizer were divided then directed down 4 tines spaced 450 mm apart. The soil placement was 50 to 70 mm deep using 100 mm wide points manufactured by Gyral Implements, Toowoomba.

Urea used in both experiments was prilled product manufactured by Incitec Fertilizers, Brisbane. The product was selected to ensure uniformity of particle size. Sieve analysis indicated particle size distribution 99.9% >1.18 mm and 98.8 % >1.7mm.

NBPT was applied to urea as the commercial product Agrotain® 0.28 % a. i. 9 days before use.

A measured quantity, 10.4 mL of Agrotain liquid was applied to 1000 g quantities of urea and mixed until the Agrotain spread evenly over the surface of the urea. Even spread was indicated by a uniformly green colour of the treated product.

A nil-urea control treatment enabled the evaluation of possible effects of NBPT on plant establishment. This treatment consisted of diatomaceous earth impregnated with NBPT. Another treatment, consisting of the diatomaceous earth carrier only was included to test whether the carrier affected soil moisture condition within the fertilizer band. A third control treatment without urea, NBPT or carrier was also included.

Urea, with or without NBPT was applied at 0, 0.43, 0.86, 1.29, 2.58 or 5.16 g/m of row for sorghum and 0, 0.43, 0.86, 1.29 or 2.58 g/m of row for cotton which is
equivalent to 0, 10, 20, 30, 60, 120 kg/ha applied in rows spaced 450 mm apart.

Treatments were laid out in a randomised complete block design; each treatment was replicated 4 times with plots 2 m wide and 10 m long.

Soil moisture at sowing was 46 % (w/w) between 2.5 and 7.5 cm, and soil temperature at 7.5 cm was 26 °C. Thirty mm of rainfall was received 9 DAS.

Crop establishment observations were taken at 4, 7, 9 and 17 DAS when emerged plants were counted in 4 one metre lengths of row per plot. Mean establishment 17 DAS, expressed as plants/m of row, was used for statistical analysis.

For each fertilizer rate an extra plot was laid down at the time of sowing adjacent to the experiment. The extra plots were designated for destructive soil sampling and analysis for urea, NH₄-N, NO₃-N and pH within the fertilizer reaction zone. Soil samples from the application band were collected 1, 3, 7, 17 and 38 DAS using the soil sampling method described in Chapter 3.

7.3 RESULTS

Experiment 7-1 - Increasing soil water to improve crop establishment for seed-placed fertilizer.

Establishment for cotton was 85 % of seeds sown and for sorghum 87% of seeds sown, for the zero MAP and zero water treatment. Application of 8 g/m of MAP did not significantly reduce cotton seedling establishment, but reduced sorghum establishment from 82 to 72 plants/5 m (P=0.08)(Fig. 7.1). Sorghum establishment was not significantly affected by 4 g/m.
Established plants of cotton (□) and sorghum (∆) per 5 m of row 18 DAS for MAP applied in the seed row at 0, 2, 4 or 8 g/m. Error bars represent the l.s.d. (P=0.05) value for the species and MAP rate interaction.

For cotton, adding water to the seed furrow at 10 mL/m when the soil moisture content at sowing was 25% w/w significantly increased establishment; there was no further increase in establishment by the addition of water at rates above 10 mL/m (Fig. 7.2). When sowing soil moisture content was 35 % w/w, cotton establishment was similar for 0, 10 and 20 mL/m water addition, 60 to 65 plants /5m, but declined to 55 plants/5m at 80 mL/m (P=0.16).

At 25 % soil moisture content at sowing sorghum establishment increased from 68 to 76 plants/5m with increasing water rates up to 80 mL/m (Fig. 7.2), but at 35% soil moisture content there was no increase in establishment by adding water in the seed furrow.

Just as sorghum establishment increased with water addition to the seed furrow, establishment was also significantly higher when sowing soil moisture content was 35 % than when sowing soil moisture content was 25%. There was no significant effect of sowing soil moisture content on cotton establishment.
Fig. 7.2 Established cotton (●) and sorghum (○) plants per 5 m of row 18 DAS for water applied in the seed row at 0, 10, 20 or 80 mL/m and for sowing moistures of 25% or 35 % (w/w). Error bars represent the l.s.d. (P=0.05) value for the soil moisture and water rate interaction.

There were significant differences within each species in the response to MAP rate or sowing soil moisture content as well as a significant interaction between MAP rate and water rate but the interaction between species, MAP rate and water rate was not significant. This suggests that the species response to MAP and water rate were similar for both cotton and sorghum. Establishment for MAP rates up to 4 g/m was maintained at the nil fertilizer-nil water control level, by adding water to the seed furrows (Fig. 7.3). Water applied to the seed furrow at 10 mL/m was sufficient to produce the maximum water addition response. Adding water when the MAP rate was increased to 8 g/m did not maintain establishment at the nil-fertilizer nil-water control level, but where water was added, crop establishment was significantly better than where no water was added.
**Establishment Rate**

The percentage of seedlings that had emerged 7 DAS as a percentage of emergence 18 DAS was used as a measure of seedling establishment rate, a high percentage indicating rapid establishment.

MAP applied at 8 g/m slowed establishment rate from 81.2 % to 66.1 % (Table 7.1).

![Graph](image.png)

**Fig. 7.3** Establishment of cotton 18 DAS for MAP applied at 0, 2, 4 or 8 g/m and water applied in the seed furrow at 0 (○), 10 (●), 20 (+) or 80 (▲) mL/m. Vertical bar are l.s.d (P=0.05) for the MAP rate and water rate interaction.

Establishment was generally faster at 25 % soil moisture than 35 %. Addition of water to the seed furrow at rates above 10 mL/m increased establishment rate for 25 % soil moisture and maintained it significantly higher than for 35 % soil moisture. The apparent different effects of soil moisture and water added to the seed-furrow on establishment rate might have been due to soil moisture and temperature differences for the 2 sowings, which were 10 days apart.
Table 7.1  Establishment rate of cotton and sorghum in response to rates of MAP fertilizer, seed furrow application of water and at 2 starting soil moistures (25 and 35 % w/w). Establishment rate was indicated by the number of seedlings emerged at the first counting as a percentage of the final establishment.

<table>
<thead>
<tr>
<th>Source</th>
<th>Rate/Level</th>
<th>l.s.d.</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>MAP rate</strong></td>
<td>(g/m)</td>
<td></td>
</tr>
<tr>
<td>mean</td>
<td>0 2 4 8</td>
<td>5.6</td>
</tr>
<tr>
<td><strong>water rate</strong></td>
<td>(mL/m)</td>
<td></td>
</tr>
<tr>
<td>mean</td>
<td>0 10 20 80</td>
<td>5.6</td>
</tr>
<tr>
<td><strong>species</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>mean</td>
<td>cotton</td>
<td>sorghum</td>
</tr>
<tr>
<td><strong>soil moisture</strong></td>
<td>(% w/w)</td>
<td></td>
</tr>
<tr>
<td>mean</td>
<td>25 35</td>
<td>4.0</td>
</tr>
<tr>
<td><strong>soil moisture x species</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>mean</td>
<td>cotton</td>
<td>sorghum</td>
</tr>
<tr>
<td><strong>soil moisture x water rate</strong></td>
<td>(mL/m)</td>
<td></td>
</tr>
<tr>
<td>mean</td>
<td>0 10 20 80</td>
<td>5.6</td>
</tr>
</tbody>
</table>

**Experiment 7-2 - Improving crop establishment with higher seeding rate or N fertilizers with different chemical characteristics.**

Application of urea or ammonium nitrate to the seed furrow at sowing significantly \((P<0.05)\) reduced barley establishment. The reduction in establishment from urea was significantly greater than for ammonium nitrate even though the rate of ammonium nitrate applied was approximately double the urea rate. For the 25 kg/ha seeding rate a barley population less than the target 15 plants/m established when the urea rate was higher than 0.6 g/m (Table 7.2).

For a sowing rate of 40 kg/ha, a barley population less than the target minimum occurred with a urea rate higher than 1.7 g/m. For ammonium nitrate, the
highest rate of application, 5.5 g/m, resulted in acceptable barley populations at both 25 and 40 kg/ha seeding rates.

Averaged over the 2 fertilizers, a similar percentage reduction in establishment from the highest rate of fertilizer was detected for 25 (37 %) or 40 kg/ha (41 %) seeding rates (Table 7.2).

Although the 2 fertilizer products contain NH$_4^+$-N, the concentration of NH$_4^+$-N, and other chemical properties of these products e.g. pH and osmotic pressure, vary greatly. To determine whether different responses in barley establishment for the 2 fertilizers was related to the NH$_4^+$-N rate or to other attributes of these fertilizer products, barley establishment response to NH$_4^+$-N rates from urea or ammonium nitrate was compared (Fig. 7.4).

Establishment response of barley to urea and ammonium nitrate shown in Fig. 7.4 indicates significant differences between the products at common NH$_4^+$-N rates. Establishment response was similar for urea and ammonium nitrate for rates up to 0.4 g/m NH$_4^+$-N. Urea at N rates above 0.4 g/m decreased establishment more than did ammonium nitrate at equivalent NH$_4^+$-N rates (Fig. 7.4). At 1 g/m NH$_4^+$-N establishment was reduced by 51 % for urea and 29 % for ammonium nitrate. The establishment difference between urea and ammonium nitrate increased from 8% at 0.6 g/m to 22% at 1 g/m NH$_4^+$-N. The 9 % reduction in establishment between 0.4 and 1 g/m NH$_4^+$-N for ammonium nitrate was significant ($P<0.05$).
Table 7.2 Establishment of barley resulting from the application of 4 rates of urea or ammonium nitrate and 2 seeding rates.

<table>
<thead>
<tr>
<th>Fertilizer Product</th>
<th>Fertilizer Rate (g/m)</th>
<th>Seeding Rate (kg/ha)</th>
<th>25</th>
<th>40</th>
<th>mean</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>urea</td>
<td>0</td>
<td>16</td>
<td>24.4</td>
<td>20.2</td>
<td></td>
</tr>
<tr>
<td></td>
<td>0.6</td>
<td>14</td>
<td>19.8</td>
<td>16.9</td>
<td></td>
</tr>
<tr>
<td></td>
<td>1.1</td>
<td>12.8</td>
<td>19.0</td>
<td>15.9</td>
<td></td>
</tr>
<tr>
<td></td>
<td>1.7</td>
<td>11.4</td>
<td>16.2</td>
<td>13.8</td>
<td></td>
</tr>
<tr>
<td></td>
<td>2.3</td>
<td>7.4</td>
<td>12.6</td>
<td>10</td>
<td></td>
</tr>
<tr>
<td>mean</td>
<td></td>
<td></td>
<td>12.3</td>
<td>18.4</td>
<td></td>
</tr>
<tr>
<td>ammonium nitrate</td>
<td>0</td>
<td>18.6</td>
<td>25.2</td>
<td>21.9</td>
<td></td>
</tr>
<tr>
<td></td>
<td>1.3</td>
<td>17.0</td>
<td>21.4</td>
<td>19.2</td>
<td></td>
</tr>
<tr>
<td></td>
<td>2.7</td>
<td>15.4</td>
<td>19.8</td>
<td>17.6</td>
<td></td>
</tr>
<tr>
<td></td>
<td>4.1</td>
<td>15.2</td>
<td>18.2</td>
<td>16.7</td>
<td></td>
</tr>
<tr>
<td></td>
<td>5.5</td>
<td>14.6</td>
<td>16.6</td>
<td>15.6</td>
<td></td>
</tr>
<tr>
<td>mean</td>
<td></td>
<td></td>
<td>16.2</td>
<td>20.2</td>
<td></td>
</tr>
<tr>
<td>seeding rate mean</td>
<td></td>
<td></td>
<td>14.2</td>
<td>19.3</td>
<td></td>
</tr>
</tbody>
</table>

I.s.d. (P=0.05)

<p>| | |</p>
<table>
<thead>
<tr>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>product</td>
<td>0.6</td>
</tr>
<tr>
<td>fertilizer rate</td>
<td>1.0</td>
</tr>
<tr>
<td>seed rate</td>
<td>0.6</td>
</tr>
<tr>
<td>product x fertilizer rate</td>
<td>1.4</td>
</tr>
<tr>
<td>product x seed rate</td>
<td>0.9</td>
</tr>
<tr>
<td>fertilizer x seed rate</td>
<td>1.4</td>
</tr>
</tbody>
</table>
**Experiment 7-3 - Delaying urea hydrolysis to increase the safe rate of urea applied with cotton or sorghum seed.**

**Conversion rate of urea to nitrate-N.**

Both urea application rate and NBPT treatment influenced the conversion rate of urea to NH₄⁺.

The urea concentration declined rapidly where no NBPT had been applied and in most treatments had declined to <4 mg/kg 7 DAS (Figure 7.5a). The delaying effect of NBPT on urea hydrolysis had disappeared by 17 DAS. The rain received 9 DAS was likely to have reduced the effect of NBPT by moving urea away from the NBPT.

Soil NH₄⁺-N concentration increased significantly ($P<0.05$) 1 DAS. Highest NH₄⁺-N concentrations were found with the highest urea rates (Fig. 7.5b). Hydrolysis of urea was reduced by treatment with NBPT although the difference in NBPT treatments was not significant ($P<0.05$) until 3 DAS. The soil NH₄⁺-N
level for low urea rates declined to background levels, <15 mg/kg, by 17 DAS and high urea rates were at background levels by 38 DAS.

Soil $\text{NH}_4^+$-N concentration peaked 3 DAS in most treatments. The highest $\text{NH}_4^+$-N concentration in the seed line was 570 mg/kg for 5.16 g/m urea without NBPT and 220 mg/kg for the same urea rate, NBPT treated (Figure 7.5 b). Peak $\text{NH}_4^+$-N concentrations generally decreased in proportion to reductions of urea rate. For the 5.16 g/m urea plus NBPT treatment, soil $\text{NH}_4^+$-N concentration remained lower than untreated urea until 17 DAS from which time the plus NBPT treatment was higher until 38 DAS by which time both had decline to less than 5 mg/kg.

Soil $\text{NO}_3^-$-N concentration rose from 8 mg/kg to 54 mg/kg 1 DAS for the 5.16 g/m urea rate without NBPT (Figure 7.5 c). The concentration increased to 243 mg/kg 7 DAS, after which the soil level declined. Increases in soil $\text{NO}_3^-$-N concentration in the seed-line were smaller for lower rates of urea. The rapid decline in soil $\text{NO}_3^-$-N concentration from 7 DAS to 17 DAS in most treatments was probably due to the displacement of $\text{NO}_3^-$-N below the sampling depth. Nitrate movement was most likely promoted by 30 mm of rainfall received 9 DAS. Generally, where NBPT had been applied soil $\text{NO}_3^-$-N concentration was not significant higher than the zero urea treatments, however $\text{NO}_3^-$-N concentration was substantially higher than untreated 5.16 g/m rate 7 DAS where NBPT had been applied. The low levels of $\text{NO}_3^-$-N by 17 DAS may be explained by the combined effects of delay of hydrolysis from NBPT and the rainfall 9 DAS causing movement of urea and/or $\text{NO}_3^-$ below the soil layer sampled.
Fig. 7.5  Soil concentrations of urea (a), NH$_4^+$-N (b) and NO$_3^-$-N (c) in the seed zone (25 to 50mm depth) during the 38 days after application of urea or urea treated with NBPT (0, 1.29, 2.58 or 5.16 g/m) with sorghum or cotton seeds. Error bar represents the l.s.d. ($P=0.05$) for the urea rate, urease inhibitor, sample date interaction.
**Crop Establishment**

Establishment counts for the 3 nil-fertilizer controls for cotton and sorghum indicated that neither inert diatomaceous earth carrier nor NBPT had any effect on final plant establishment. The mean of all 3 nil fertilizer treatments was subsequently used for comparison with urea treatments.

Favourable soil moisture and warm temperatures at sowing were ideal for rapid germination and establishment of sorghum and cotton. Plant counts 4 DAS indicated 50% of seeds sown had emerged in the nil-fertilizer control plots. Soil moisture content of the soil layer to sowing depth declined by 4% (w/w) as a result of soil disturbance due to sowing operations. Rainfall 9 DAS appeared to have little adverse effect on the emerging seedlings. Treatment effects appeared to be well established by the time the rain fell.

In the presence of urea at high rates (2.58 and 5.16 g/m) NBPT addition significantly \( P<0.05 \) increased sorghum establishment compared with where no NBPT was applied (Table 7.3).

The maximum untreated urea rate which caused no significant reduction in sorghum establishment was 1.29 g/m. Urea treated with NBPT could be applied at 5.16 g/m with no significant reduction in sorghum establishment.

The maximum “safe” rate for the untreated urea is similar to the 1 g/m urea maximum rate currently recommended for application in the seed furrow with sorghum.

Response to NBPT in avoiding seedling damage for cotton was similar to that for sorghum for urea rates common to both crops (Table 7.4). The maximum “safe” rate with cotton increased from 1.29 g/m to above 2.58 g/m with the addition of NBPT. Plant population resulting from 2.58 g/m of urea without NBPT was significantly lower than where NBPT had been applied and was visibly unevenly spaced. Generally, where establishment was significantly reduced, plant spacing was observed to be uneven. Uneven spacing at low populations in cotton can create significant management problems which result in reduced yield (Constable, 1982).
Table 7.3  Effect of 5 urea rates and NBPT on the population of sorghum seedlings, 17 DAS.

<table>
<thead>
<tr>
<th>Urea Rate (g/m)</th>
<th>NBPT</th>
<th>0</th>
<th>0.25 %</th>
<th>mean</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>(plants/m)</td>
</tr>
<tr>
<td>0</td>
<td>11.31</td>
<td>11.75</td>
<td>11.53</td>
<td></td>
</tr>
<tr>
<td>0.43</td>
<td>11.13</td>
<td>11.38</td>
<td>11.25</td>
<td></td>
</tr>
<tr>
<td>0.86</td>
<td>11.88</td>
<td>12.5</td>
<td>12.18</td>
<td></td>
</tr>
<tr>
<td>1.29</td>
<td>9.93</td>
<td>10.68</td>
<td>10.31</td>
<td></td>
</tr>
<tr>
<td>2.58</td>
<td>7.43</td>
<td>11.69</td>
<td>9.56</td>
<td></td>
</tr>
<tr>
<td>5.16</td>
<td>3.94</td>
<td>9.63</td>
<td>6.78</td>
<td></td>
</tr>
<tr>
<td>mean</td>
<td>9.27</td>
<td>11.27</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

l.s.d. (P=0.05)

| Urea rate | 0.98 |
| NBPT      | 1.70 |
| Urea rate x NBPT | 2.41 |

The rapidity at which 50 % establishment was reached in sorghum with NBPT suggests that the increasing osmotic effect of increasing urea close to seeds did not significantly influence their establishment rate (Table 7.5). At the same urea rate, sorghum establishment rate decreased more rapidly in the absence of NBPT, suggesting that the delay to establishment resulted from NH₃ toxicity rather than from the osmotic effect of the fertilizer.

For cotton, differences in time taken to reach 50 % establishment and 90 % establishment for 0.43 and 0.86 g /m urea rates, may indicate that some seeds come into more intimate contact with the fertilizer particles than others (Table 7.6). It was hypothesised that seeds distant from fertilizer particles show normal establishment patterns, those closer to the fertilizer particles being slower to emerge or failing to emerge.

Cotton was generally slower to emerge than sorghum, but the effect of urea and NBPT on establishment rate was similar for both species.
### Table 7.4  Effect of 5 urea rates and NBPT on the population of cotton seedlings, 17 DAS.

<table>
<thead>
<tr>
<th>Urea Rate (g/m)</th>
<th>NBPT 0</th>
<th>NBPT 0.25%</th>
<th>mean (plants/m)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>12.25</td>
<td>12.06</td>
<td>12.15</td>
</tr>
<tr>
<td>0.43</td>
<td>12.25</td>
<td>12.38</td>
<td>12.31</td>
</tr>
<tr>
<td>0.86</td>
<td>12.25</td>
<td>12.44</td>
<td>12.35</td>
</tr>
<tr>
<td>1.29</td>
<td>10.12</td>
<td>12.44</td>
<td>11.06</td>
</tr>
<tr>
<td>2.58</td>
<td>7.18</td>
<td>10.38</td>
<td>8.78</td>
</tr>
<tr>
<td>mean</td>
<td>10.81</td>
<td>11.85</td>
<td></td>
</tr>
</tbody>
</table>

l.s.d. (P=0.05)
- Urea rate: 1.76
- NBPT: 1.11
- Urea rate x NBPT: 2.49

### Table 7.5  Effect of 5 urea rates and NBPT on speed of sorghum establishment.

<table>
<thead>
<tr>
<th>Urea rate (g/m)</th>
<th>DAS to 50% establishment</th>
<th>DAS to 90% establishment</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No NBPT</td>
<td>NBPT</td>
</tr>
<tr>
<td>0</td>
<td>4</td>
<td>4</td>
</tr>
<tr>
<td>0.43</td>
<td>4</td>
<td>4</td>
</tr>
<tr>
<td>0.86</td>
<td>4</td>
<td>4</td>
</tr>
<tr>
<td>1.29</td>
<td>4</td>
<td>4</td>
</tr>
<tr>
<td>2.58</td>
<td>7</td>
<td>4</td>
</tr>
<tr>
<td>5.16</td>
<td>dna</td>
<td>4</td>
</tr>
</tbody>
</table>

dna - did not attain establishment %

### Table 7.6  Effect of 5 urea rates and NBPT on speed of cotton establishment.

<table>
<thead>
<tr>
<th>Urea rate (g/m)</th>
<th>DAS to 50% establishment</th>
<th>DAS to 90% establishment</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No NBPT</td>
<td>NBPT</td>
</tr>
<tr>
<td>0</td>
<td>7</td>
<td>7</td>
</tr>
<tr>
<td>0.43</td>
<td>7</td>
<td>7</td>
</tr>
<tr>
<td>0.86</td>
<td>7</td>
<td>7</td>
</tr>
<tr>
<td>1.29</td>
<td>9</td>
<td>9</td>
</tr>
<tr>
<td>2.58</td>
<td>17</td>
<td>9</td>
</tr>
</tbody>
</table>

dna - did not attain establishment %
Relationship between urea rate, soil NH$_4^+$-N concentration and crop establishment.

To establish relationships between urea application rate, soil NH$_4^+$-N concentration in the seed furrow and crop establishment the data for 3 DAS, when the soil NH$_4^+$-N concentration was near its highest, were assessed by regression analysis (Figures 7.6 a and 7.6 b). This sampling also corresponded with a time of high metabolic activity in germinating seeds.

Reduction in sorghum establishment % was highly correlated with the rate of urea applied (R=0.975 without NBPT) and the response was modified by the addition of NBPT (R=0.819 with NBPT).

To better define the difference between urea with and without NBPT, the instantaneous rate of establishment decline was computed from the slope of the establishment response function at 2 g/m, a urea application rate within the responsive range. For untreated urea, establishment was reduced by 13.4 %/g/m at 2 g/m and for urea treated with NBPT the rate of establishment loss was 2.4 %/g/m (Figure 7.6 a).

Cotton was found to be more sensitive to urea than sorghum. The rate of establishment loss in cotton at 2 g/m urea rate was 23.4%/g/m without NBPT, and 11.3%/g/m with NBPT. At low urea rates, addition of NBPT maintained the cotton establishment % at about the control level, but at higher urea rates the establishment was about 70 % of that where no NBPT was applied (Figure 7.6 b).

For sorghum the soil NH$_4^+$-N concentration that produced a significant reduction in establishment \((P<0.05)\), derived from Figure 7.6a, was in the range 190 to 250 mg/kg whereas the range for cotton was 106 to 136 mg/kg (Figure 7.6 b).
Fig. 7.6 Effect of urea or urea treated with NBPT on NH$_4^+$-N concentration and crop establishment for sorghum (a) and cotton (b).
7.4 DISCUSSION

Higher establishment for sorghum and cotton where urea was applied with 
NBPT suggested that the osmotic effect of about 5 g/m of urea applied with 
good soil moisture was not a significant factor in reducing the final population of 
these crops. However, it may have contributed to the slowing of establishment 
rate. Ammonia toxicity appears the most important factor in reducing crop 
establishment when urea is placed in the seed furrow of these crops. The 
effects of biuret and nitrite toxicity were discounted as being factors in the 
establishment response. The findings of Hunter and Rosenau (1966) suggest 
that biuret does not restrict germination and establishment but affects seedling 
growth, and the rate of application required to cause biuret toxic effects in 
maize seedlings was 700 g/ha. The maximum rate of biuret applied in this 
experiment was 550 g/ha. Nitrite concentrations were not considered to be a 
significant factor in the germination and establishment response to NH₃. The 
measured concentrations during the seedling establishment period described in 
Chapter 4, 2 mg/kg, were well below the 26 mg/kg NO₂⁻-N concentration 
suggested by Duisberg and Buehrer (1954) to be toxic to growing plants.

Cotton was more tolerant of MAP placed in the seed furrow than sorghum. The 
critical NH₄⁺-N concentration for cotton was > 0.8 g/m and for sorghum 0.4 g/m. 
The critical concentration for sorghum supports current recommendations 
however, the critical concentration > 0.8 g/m NH₄⁺-N for cotton is significantly 
higher than rates currently recommended. Tolerance for untested dicot species 
has been influenced by the findings of Carter (1967), who concluded that 
legumes, brassica and other dicot species were generally less tolerant of 
ammonia than monocots. Differences between current NH₄⁺-N 
recommendations for cotton and these findings may result from a lack of direct 
fertilizer tolerance experimentation for cotton or difference in cotton response to 
NH₄⁺-N derived from MAP or urea. The critical MAP NH₄⁺-N rate for cotton was 
0.8 g/m and for urea was 0.59 g/m.

Selecting NH₄⁺-fertilizers with differing chemistry can increase NH₄⁺-N rates 
applied in the seed furrow at sowing. For example, barley establishment was 
significantly reduced when 22.5 kg/ha of N as urea (0.5 g/m NH₄⁺-N) or 83
kg/ha of N as ammonium nitrate (0.96 g/m NH$_4^+$-N) were applied. The lower safe rate of urea from this experiment as compared with that found by Mason (1971) may be related to the higher pH of the soils in this study. The soil pH reported for Mason’s experiment was 6.8 (CaCl$_2$ extract) whereas the pH of the Dalby soil was 8.5 (water extract). Higher soil pH would be expected to maintain a higher soil NH$_3$ concentration and the finer texture of the Dalby soil would reduce the NH$_3$ diffusion potential, further increasing the concentration of NH$_3$ in the seed row.

There is clear evidence from soil analysis that NBPT slowed urea hydrolysis for up to 9 days. The delay did not prevent the build up of toxic concentrations of NH$_3$ in the seed row as indicated by the crop establishment response, particularly for urea application rates above 2.58 g/m. Nevertheless, these data suggest that urea rates in excess of 2.58 g/m for cotton or 5.16 g/m for sorghum may be applied safety if treated with NBPT and with favourable soil conditions at sowing. These rates should be regarded as maximum rates to be applied under normal soil temperature and moisture content conditions. Rates in excess of these highest safe rates may cause seedling damage when temperatures are above normal and/or the soil in the seed zone dries rapidly.

Increased sorghum establishment by the application of water to the seed furrow is consistent with the findings of Radford and Neilsen (1985) for the combination of press-wheels and water injection. They found no significant difference in the final plant population for water injection alone, but where a combination of press-wheel and water injection was used; there were significant responses at 4 of 9 experimental sites. They also found that establishment responses were more common at sites with low soil moisture at sowing.

By adding extra water to the seed-fertilizer interface at sowing by water injection, a better crop establishment rate was maintained across higher MAP application rates. Ferraris (1992b) found that seedbed water content and soil temperature dominated the establishment process and that water injection only increased establishment rate and had little effect on final crop establishment. The results for these experiments agree with the findings of Radford et al.
(1985) and Ferraris (1992b) for low fertilizer rates. However, for high rates of fertilizer applied with seed, water addition to the seed furrow significantly improved the final plant stand. When water was added with high fertilizer rates, increased establishment % was thought to be a result of decreased osmotic pressure of the soil solution in the seed zone.

For some crop species, establishment may not exhibit a linear response to water addition as demonstrated by cotton. Under usual sowing moisture contents, water injection to cotton at rates in excess of 20 mL/m are unnecessary but higher rates of water addition, up to 80 mL/m, may be beneficial when sowing into drier than desirable soil.

Mechanisms involved in reduced cotton establishment by the addition of high water rates at 35 % soil moisture were not explored. Crocker and Barton (1957) suggest that in the case of seed soaking, the causal factors involved in reducing germination include oxygen deprivation, exosmosis of soluble constituents and pathogen attack. Any one or a combination of these factors could have been the reason for reduced cotton establishment, with the highest volume of water injected with seeds.

Increasing the seeding rate was successful in offsetting reduced seedling establishment of barley. Across the range of fertilizer application and barley seeding rates, increasing seeding rate was a more successful strategy for maintaining seedling population within the desired range than applying ammonium nitrate instead of urea.

However it would be dangerous to apply this strategy to other crops and products without testing the particular crop species and fertilizer product interactions. Established plant population is not the sole determinant of yield for many crops particularly for crops grown on wide row spacing where evenness of plant spacing within the row (rectangularity) is also important (Willey and Heath 1969). For treatments where significant reductions in plant establishment occurred, seedling damage appeared to be quite irregular along the row and large gaps between plants were common. Thus, species that have the ability to
compensate for reduced plant population through mechanisms such as increased tillering will be best adapted to this strategy.

Establishment appears to occur in several stages where fertilizer is placed in the seed row, as a result of the non-uniform distribution of fertilizer particles along the row. The first plants to emerge are those at a sufficient distance from any fertilizer particle so as to be unaffected by the NH$_3$ and/or osmotic effects. These plants emerged at a rate similar to plants with no urea applied. After establishment of unaffected seeds, the remainder of the seeds emerged at a rate depending on their proximity to fertilizer particles within the seed row. The closer the contact between seed and fertilizer, the slower the establishment. Establishment response for a particular fertilizer product is therefore a function of the total mass of fertilizer as well as the number of fertilizer particles applied.

7.5 CONCLUSION

Three strategies used in these experiments successfully reduced the detrimental effect of fertilizer placement in the seed furrow at sowing. Increasing seeding rates would appear to be the most practical management option, recognising that limitations are most likely to occur for species grown in wide row spacing and for species that have high yield sensitivity to moderate under and over-population.

Ammonia toxicity appeared the most significant and common cause of reduction of seedling establishment. Modifying the NH$_3$-NH$_4^+$ balance in the soil by the application of an N fertilizer product such as ammonium nitrate, that maintains a lower solution pH or prevention of urea hydrolysis were successful strategies to avoid NH$_3$ toxicity. However, these strategies for maintaining plant population are likely to be less economic for most crops than simply increasing seeding rate.
Chapter 8

General Discussion and Conclusion

The effect of fertilizers on crop establishment has been the subject of a large body of research that began in the 1920s peaked in the 1950s and 1960s and waned during the 1970s. Among these studies few Australian experiments have been published in the scientific literature (Carter 1964; Loutit et al. 1968; Mason 1971; Scott et al. 1987; Scott 1989, Radford et al. 1980; Robotham 1993). For the majority of experiments, NH$_3$ toxicity was reported as the main cause of reduced crop establishment (Duisberg and Beuhrer 1954; Allred and Ohlrogge 1964; Bennett and Adams 1970b; Bremner 1995) with osmotic effects (Dubetz 1959; Carter 1964; Young et al. 1968) and hydrofluoric acid from superphosphate (Howe-Morse 1934, Kinra et al. 1962) cited as causes in a lesser number of studies. However, the large body of information has generally provided limited data with which to develop decision support systems due to the usually narrow range of species and products tested within individual experiments, insufficient rates of fertilizer or lack of detailed description of fertilizer application equipment.

Results of experiments undertaken for this thesis show that 10 crop species have different levels of NH$_3$ tolerance based upon a multiple rate response. The magnitude of NH$_3$ tolerance was frequently too small to make clear distinction between species but the range was large enough to allow species to be placed into high, medium and low NH$_3$ tolerance groups. For crop species grown in winter, the higher tolerance group included wheat and barley. Canary was of medium tolerance and the low tolerance group included chickpea and canola. For crop species grown in summer the higher tolerance group was sorghum and maize, panicum was of medium tolerance and the lower tolerance group included cotton and sunflower. These tolerance groupings are in general agreement with N-fertilizer tolerance ranking for wheat, barley and rapeseed (canola) suggested by Mason (1971), wheat and maize from Bremner (1989) and wheat and canola from Henry (1996), but rank of some species were significantly different from findings of other researchers.
Radford et al. (1989) found that barley establishment was more tolerant of urea derived NH$_3$ than wheat. However, they commented that intolerance of wheat was related to a reduction of coleoptile length for some wheat varieties rather than barley being inherently more tolerant. Results of Experiment 5-3 indicated that there was no significant difference between coleoptile length, germination or establishment response of wheat or barley. The wheat variety, Cunningham, used in the Experiment 4-1 (soil-NH$_3$) and Experiment 5-3 (atmospheric-NH$_3$) was suggested by Radford et al. (1989) to be unresponsive to coleoptile shortening from exposure to NH$_3$. In other studies with wheat and oats, Olson and Dreier (1956), Carter (1967), Scott et al. (1987) and Bremner and Kroegmeier (1989) found oats more tolerant of fertilizer-N than wheat while Mason (1971) rated them equally tolerant. Olson and Dreier (1956) suggested that the difference in tolerance in their experiment might have been a result of different seed sources. Although the seed source for each crop species was different for experiments in Chapter 4 and 5, they were the same seed-lot for experiments in both chapters. Ammonia tolerance rank was consistent for most species between Chapters 4 and 5. Hence, seed-lot may be important in obtaining consistency when ranking crop species establishment across a number of fertilizer products and practises where crop species and fertilizer placement interactions are being considered.

Rank for chickpea was not consistent between atmospheric and soil-exposure to NH$_3$. Tolerance rank for chickpea was high when exposed to atmospheric-NH$_3$ but was low when exposed to soil-NH$_3$. Woodstock and Tsao (1986) similarly found soybean more tolerant of atmospheric NH$_3$ than maize which is not consistent with the findings of Gerwing et al. (1994) for soil applied ammonium phosphate fertilizer or for the results of Experiment 6-1. Both Gerwing et al. (1994) and our findings clearly indicate that soybean was least tolerant to all fertilizers and of crop species tested, including maize. Major differences in performance of crop species between studies appear to be related to the experimental method used. Many of the experiments involving assessment of species sensitivity to NH$_3$ toxicity reviewed were typically conducted in artificial conditions and at a single or only a few NH$_3$ or fertilizer rates.
For experiments conducted in the laboratory, both for soil and soil-less culture, temperature can be a significant influence on NH₃ toxicity. Allred and Ohlogge (1964) and Woodstock and Tsao (1986) both found that NH₃ toxicity was exacerbated by increasing temperature as would be expected given the relationship between Henry’s constant and temperature, described by Emmerson et al. (1975). Similarly, crop species germinated outside their normal temperature range may undergo changes in coleoptile and radicle elongation (Ward 1987; Radford et al. 1989) that may influence NH₃ tolerance rank for emergence and establishment.

Tolerance to NH₃ observed in the closed system studies (Chapter 5) was described best by plateau-linear decline regression model. Species were characterised by both the length of the plateau phase (breakpoint) and the slope of establishment (% germination reduction for each increment of NH₃) which is similar to a method by which salinity tolerance thresholds are established for crop species (Marschner 1995b). Less tolerant species such as canary, sunflower, and canola generally had shorter plateau phases than more tolerant species such as sorghum, wheat, and maize. Germination was steady for NH₃ concentrations up to 54 x 10⁻⁴ M for less tolerant species and 102 x 10⁻⁴ M for more tolerant species. Sunflower, cotton, and barley germination response was intermediate, derived from either short plateaus and lesser slopes or longer plateaus and greater slopes.

It was hypothesised that seed tolerance to NH₃ during germination is associated with physical impedance to NH₃ diffusing into metabolically active areas. This was confirmed by the significant correlation of seed surface area-volume with NH₃ tolerance during germination. Scott (1989) has also cited diffusion resistance as a mechanism for tolerance to P fertilizer products applied with seed or as a seed-coat. The length of the tolerance plateau was thought to represent resistance to diffusion while the steepness of decline phase may indicate the ability of a species to cope bio-chemically with increasing NH₃ exposure. Genetic variability relating to physical characteristics such as diffusion resistance of the seed coat (Guttay 1957) and seed vigour may also explain establishment variability within species.
When crop species were exposed to NH$_4^+$-N from a range of fertilizer products such as urea, MAP and DAP (Chapter 6 and 7), tolerance rank was similar to that established from soil and atmospheric-NH$_3$ exposure (Chapter 4 and 5). Wheat (0.4 g/m) and sorghum (0.59 g/m) tolerance to urea-N were similar to currently recommended maximum rate, 0.5 g/m row (Mills et al. 1996; Mills and MacIntyre 1997). Agreement between crop species rank from NH$_3$ exposure and rank from other fertilizer NH$_3$ sources tested in Chapters 6 and 7, suggest that the maximum safe fertilizer rate for species including wheat and sorghum would follow the established rank. Fertilizer-N tolerated by crops of lower tolerance than wheat and sorghum should then be related to the rate established for exposure to NH$_3$(g.) but modified for the ‘NH$_3$ potential’ of the fertilizer product.

The term ‘NH$_3$ potential’ was developed during this study to describe the potential of fertilizers to generate then maintain a soil NH$_3$ concentration relative to urea. Ammonia potential for a fertilizer product was though to be influenced by its NH$_4^+$-N concentration, pH, solubility and accompanying anion.

In past fertilizer recommendations, all NH$_4^+$-fertilizer products have been treated similarly in relation to potential NH$_3$ toxicity (Mills et al. 1996; Mills and MacIntyre 1997). However, there is evidence that NH$_4^+$-fertilizer products differ in potential for establishment damage applied at similar NH$_4^+$-N rates (Cook and Scott 1987). This is clearly demonstrated for cotton, where critical NH$_4^+$-N rates for significant establishment damage from urea was 0.59 g/m and under similar conditions was >0.8g/m from MAP. Establishment damage caused by DAP was generally greater than for MAP at equivalent NH$_4^+$-N rates (Allred and Ohlrogge 1964; Gerwing et al. 1997). Allred and Ohlrogge (1964) suggested that NH$_3$ damage might be greater from MAP applied to soils with soluble calcium carbonate (CaCO$_3$) and pH below 8.4. These are common features of Vertosol soils of the cereal-growing region of northeastern Australia. They suggested that the potential for MAP damage increased as a result of CaCO$_3$ acting as a buffer against the accumulation of hydrogen ions, thereby increasing the NH$_3$ concentration. However, establishment reductions from MAP in the presence of soluble CaCO$_3$ did not reach that of DAP for similar
product concentrations. Hence, caution may be required when extending recommendations for seed-furrow placement of some fertilizer products into soils of significantly different chemical characteristics.

Different establishment responses to $\text{NH}_4^+$-fertilizer products were not a characteristic unique to cotton. Differences were also found for barley response to urea or ammonium nitrate. Cummins and Parks (1961), Mason (1971) and the results of Experiment 7-2 conferred that a range of crops including wheat, barley and maize were more tolerant of ammonium nitrate than urea at equivalent N rates. These results also suggest that the rate of N that can be applied safely as ammonium nitrate is at least twice that of urea ($\text{NH}_3$ potential of 50 %). This indicates a requirement for inclusion of the potential for establishment damage from different fertilizer products to be included in decision support information.

Osmotic effects of fertilizer on establishment differed among crop species but were usually found secondary to $\text{NH}_3$ toxicity (Hood and Ensminger 1964). The majority of research conducted into effects of osmotic pressure on germination is related to reclamation of saline soil with pasture species (George and Williams 1964; Young et al. 1968; Young et al. 1983). A smaller number of studies were concerned with effects of fertilizer salts placed in close proximity to seeds of crop species (Dubetz et al. 1959). Dubetz et al. (1959) tested osmotic tolerance of 3 crop species; maize, fieldbean ($\text{Phaseolus vulgaris}$ L.) and sugarbeet, creating the osmotic pressure with TSP and ammonium nitrate alone and in combination, and mannitol. A major finding of this experiment was that field-bean was significantly less tolerant of high osmotic pressure during germination and maize the most tolerant. Results of TSP treatments for Experiment 6-1 indicated that soybean, another large seeded leguminous crop species, was least tolerant of the 5 species tested. Maize and cotton were among the most tolerant. However, not all grain legumes should be classified as sensitive. Mungbean ($\text{Vigna radiata}$ (L.) R. Wilczek var. radiata) has been found to be reasonably tolerant of seed-placed TSP, MAP and DAP (Dowling unpublished data). Sunflower tolerance of TSP was similar to maize and sorghum which was consistent with the findings of Gerwing et al. (1994) for high
soil moisture conditions, but is lower than the tolerance inferred from critical water potential figures quoted in Table 2.1.

No feature of seeds other than inherent critical water potentials quoted by Hunter and Erikson (1952), Williams and Shaykewich (1971) and Young et al. (1983) for a limited range of crop species, suggest mechanisms of tolerance to osmotic pressure from fertilizer sources.

Where KCl (high salt index) was compared to MAP (moderate salt index, low NH$_3$ potential) using cotton as a test species, KCl was more damaging to seedling establishment and created greater delays to emergence than MAP. For these products osmotic pressure was therefore more damaging than NH$_3$ toxicity. Additional evidence for osmotic pressure reducing establishment was the ability to alleviate the effect of fertilizer by adding water to the seed-furrow at sowing.

Osmotic effects on germination were found to be related to the product of the salt index of Rader et al. (1943) and fertilizer application rate; relative osmotic index (ROI). ROI was used to integrate the osmotic effects of KCl and MAP, fertilizer products with large differences in salt index (116 and 30). In Experiment 6-3, ROI was used to assess whether the osmotic or NH$_3$ effect (from MAP) of the products singularly or in combination was dominant. A lack of clear separation of the establishment effects into product related groups on the establishment response curve suggest that the majority of the fertilizer effect was osmotic. The continuity of the response curve also suggests that where the two products were combined the osmotic effect was additive.

Crop establishment responses to granular NH$_4^+$-fertilizers in Experiments 6-2, 6-3 and 7-1 suggest that there may be 2 mechanisms that allow seedlings to establish in the presence of NH$_4^+$-fertilizers applied in the seed furrow. During application, seed and fertilizer are scattered randomly along the seed furrow, some seeds falling close to fertilizer particles and some more distant. Seeds at greatest distance from fertilizer particles, adjacent to the diffusion zone of the fertilizer, emerge rapidly at about the same time as where no fertilizer has been
applied. Seeds closer to fertilizer particles may germinate, but are affected to varying levels of growth inhibition. This range of effects was observed among the unemerged seeds recovered from the soil in experiment 6-1. Seedlings which emergence more distant from fertilizer particles may be slightly delayed and those closest may be severely inhibited or killed, depending on fertilizer product characteristics such as its particle size and NH₃ potential.

Howe-Morse (1934) found that the orientation of the seed embryo in relation to the diffusion path of fertilizer had a significant effect on germination damage. Multi-stage germination responses to granular products occurred in Experiments 6-2, 6-3 and 7-1 where establishment rate was measured. Establishment rate was high for nil fertilizer or low NH₄⁺-N rates, slowed at intermediate NH₄⁺-N rates, although there was a varying proportion of the population that established quickly, and appeared to increase at high NH₄⁺-N rates for the small population that emerged. The apparent increase of establishment rate at high NH₄⁺-N rates was due to rapid establishment of a small percentage of seed that were assumed to be outside the fertilizer diffusion zone.

Because of the random chance of seed and fertilizer particles being applied in close proximity, it could be expected that plant population in fertilizer affected seed rows would be unevenly spaced and the emergence rate slowed. This was in fact a common observation with high rates of NH₄⁺-fertilizer over a range of species but where seeds were exposed to atmospheric-NH₃ in closed systems in Experiment 5-3, the establishment response was more uniform. Germination and growth response in the closed system was probably more typical of seed in close proximity to fertilizer particles.

**Strategies for Maintaining Establishment**

The most cost effective strategies for maintaining establishment where fertilizer is placed with seed include increased seeding rate and modifying the soil characteristics in the seed-furrow by increasing soil moisture or reducing soil permeability. Increasing seeding rate was a successful strategy to maintain a plant population for barley. However it was thought that increasing seeding rate
was not a practice that would have broad applicability across crop species, being more suited to crops that have mechanisms for compensation for low population such as tillering (Olson and Dreier 1956). For crops such as cotton that have less ability to compensate for low population, a narrower optimum plant population and lower tolerance of large in-row gaps, other more reliable methods of establishment damage avoidance would be more appropriate. Radford and Neilson (1985) suggested that addition of water to the seed-furrow at sowing be not justified as this practice increased establishment rate but not final establishment. The results of Experiment 7-1 suggest that addition of water to the seed-furrow increased total establishment where fertilizer rate limited soil moisture availability for establishment. A water rate of 100 L/ha increased establishment by up to 10 % for soils with reasonable moisture in the seeding layer where fertilizer was applied. Up to 800 L/ha was required for drier soil and higher fertilizer rate. For cropping systems where nutrient deficiencies limit yield and fertilizer rates that can be applied at sowing are restricted to below that necessary for optimum response, there are options to marginally increase fertilizer rate. Use of decision support software that more precisely integrates crop, soil and fertilizer factors that create crop establishment limitations may allow fertilizer rates to be increased safely while maintaining plant population in a desired range. Some other strategies for lowering NH₃ concentration in the seed furrow during seedling emergence are selecting fertilizer products with a lower NH₄⁺ concentration and pH or by chemical inhibition of urea hydrolysis.

**Computer Based Decision Support Software**

A computer-based decision support system was developed to integrate the complex species establishment response to soil, fertilizer product and climate parameters. These interactions were difficult to model at a low level to provide a useful guide for practical fertilizer recommendations. By aggregating parameters to higher order variables and selecting the most critical variables; species NH₃ tolerance and osmotic tolerance, fertilizer NH₃ and osmotic characteristics, soil moisture and chemical characteristics, and fertilizer application equipment configuration to describe the interactions (Appendix 1), a decision support system was developed.
Osmotic pressure changes in the soil are calculated using the data of Rader et al. (1943). These data describe soil osmotic pressure increase for a given weight of fertilizer and soil. Mean osmotic pressure change within the fertilizer band was calculated by combining fertilizer rate, fertilizer band diameter, application row width and mean soil osmotic pressure increase per 100 kg of a fertilizer product. Diameter of the fertilizer band for each nutrient group (ammonium, nitrate, phosphate, potassium) was estimated using nutrient ion movement data from Moody et al. (1995a, 1995b, 1995c) and in-furrow fertilizer dispersion data of Roberts and Harapiak (1997). Mean fertilizer band osmotic pressure was then compared with maximum water potential for a species to determine whether germination was likely to be inhibited by fertilizer.

Osmotic effects on germination were found to be related to the product of Raders’ salt index and fertilizer application rate; (ROI). However there is insufficient data about ROI interactions with important soil and crop parameters to use ROI in the current version of decision support software.

Maximum tolerated \(NH_4^+\)-N rate was determined for a species from relative \(NH_3\) tolerance rank from this thesis considering current commercial recommendations. Maximum fertilizer rate was then adjusted for fertilizer \(NH_4^+\)-N concentration, \(NH_3\) potential of fertilizer products, and soil and application conditions. For fertilizers containing \(NH_4^+\)-N, maximum suggested product rates are calculated for \(NH_3\) and osmotic damage, the recommended maximum application rate being the lesser of the two rates suggested (Appendix 2). For fertilizers containing no \(NH_4^+\)-N, the maximum suggested application rate is based on the limit for osmotic damage.

Sufficient variability was measured within crop species seed-soil-fertilizer interactions during this research to exploit and reduce the risks of establishment damage for fertilizer applied in the seed-furrow at sowing. The incidence of crop establishment damage will be reduced where a better understanding of the fundamentals of the seed-soil-fertilizer interactions are provided to advisers and farmers, together with the logic to predict the outcome on seedling injury. Such information will provide a good basis for predicting the outcome of changes to
equipment and other variables on severity of seedling injury, or for directing
development of new fertilizer products and practices.
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10.


Appendix 1  Crop ammonia and osmotic sensitivity, and key fertilizer product indices used to predict safe rates of seed placed fertilizer in Fertsafe decision support software.

<table>
<thead>
<tr>
<th>Crop Tolerance</th>
</tr>
</thead>
<tbody>
<tr>
<td>Crop</td>
</tr>
<tr>
<td>------</td>
</tr>
<tr>
<td>1</td>
</tr>
<tr>
<td>2</td>
</tr>
<tr>
<td>3</td>
</tr>
<tr>
<td>4</td>
</tr>
<tr>
<td>5</td>
</tr>
<tr>
<td>6</td>
</tr>
<tr>
<td>7</td>
</tr>
<tr>
<td>8</td>
</tr>
<tr>
<td>9</td>
</tr>
<tr>
<td>10</td>
</tr>
<tr>
<td>11</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Tolerance Level</th>
<th>Osmotic Pressure</th>
<th>Max NH$_4^-$ N Rate</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 High</td>
<td>0.8</td>
<td>0.45</td>
</tr>
<tr>
<td>2 Medium</td>
<td>0.6</td>
<td>0.27</td>
</tr>
<tr>
<td>3 Sensitive</td>
<td>0.4</td>
<td>0.135</td>
</tr>
<tr>
<td>4 Tolerant</td>
<td>1.2</td>
<td>0.54</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Product</th>
<th>Product type</th>
<th>Osmotic Pressure</th>
<th>N %</th>
<th>Ammonia Potential</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Ammonia</td>
<td>N 0.0315</td>
<td>0.82</td>
<td>1</td>
</tr>
<tr>
<td>2</td>
<td>DAP</td>
<td>P 0.02286</td>
<td>0.18</td>
<td>0.67</td>
</tr>
<tr>
<td>3</td>
<td>Ammonium sulfate</td>
<td>N 0.04605</td>
<td>0.202</td>
<td>0.25</td>
</tr>
<tr>
<td>4</td>
<td>MAP</td>
<td>P 0.01998</td>
<td>0.1</td>
<td>0.37</td>
</tr>
<tr>
<td>5</td>
<td>Muriate of potash</td>
<td>K 0.07762</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>6</td>
<td>Ammonium nitrate</td>
<td>N 0.06992</td>
<td>0.17</td>
<td>0.5</td>
</tr>
<tr>
<td>7</td>
<td>Potassium nitrate</td>
<td>K 0.04916</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>8</td>
<td>Potassium sulfate</td>
<td>K 0.030778</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>9</td>
<td>Superphosphate</td>
<td>P 0.00521</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>10</td>
<td>Triple super</td>
<td>P 0.006733</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>11</td>
<td>Urea</td>
<td>N 0.05035</td>
<td>0.46</td>
<td>0.9</td>
</tr>
</tbody>
</table>
Appendix 2  Input and output screens of Fertsafe decision support software.

Field Information Input

Crop  
Soil Texture  
Soil Salinity  
Within Furrow Spread  
Application Tine Spacing (m)  
Soil Bulk Density

Fertilizer Details

Fertiliser Product  
Soil Sowing Moisture (max)  
Suggested Maximum Rate (kg per hectare of product)

Osmotic Pressure  
Ammonia Toxicity  
Product Rate Conversion Factor