Long-term annual nitrogen fertilisation of *Eucalyptus regnans* F. Mueller and *Pinus radiata* D.Don: effects on tree growth, soil chemistry and net nitrogen mineralisation.

by

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requirements of the degree of

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Unfertilised **Fertilised (1300 kg N ha^{-1})**

Pinus radiata **plantation planted on Kurosol in the north-east of Tasmania.**

Declaration

I hereby declare that this thesis contains no material which had been accepted for any award of any other degree or diploma in any tertiary institution and that, to the best of my knowledge and belief, contains no material previously published, or written by another person, except when due reference is made in the text of the thesis.

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Dedication

This thesis is dedicated to my mother Maree Vandersluys, whose strength, persistence and compassion in life has always inspired me, and to my father, Frank Charles Ringrose, for his love of the unknown and persistent questioning of everything that is life.

Authority of Access

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List of Publications

Journal articles,

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C. Ringrose and W.A. Neilsen. 2005 Growth responses of *Eucalyptus regnans* and soil changes following periodic fertilization. Soil Sci.Soc. Am. J. 69:1806-1812.

Technical Reports

Ringrose, C., and W.A. Neilsen. 2002. Changes in soil profile characteristics to periodic additions associated growth responses of *Eucalyptus regnans* and *Pinus radiata*. Technical Report, Rayonier 15. Forestry Tasmania, Division of Forest Research and Development, Hobart.

Ringrose, C., and W.A. Neilsen. 2002. Changes in forest floor mineral N availability with timed application of N fertilizer to *P. radiata* and *E. regnans* plantations. Technical Report, 17. Forestry Tasmania, Division of Forest Research and Development, Hobart.

Abstract

In the current competitive market for land in Tasmania, Australia, economic forest production may require large nutrient inputs to optimise productivity per unit area of land. Nitrogen (N) and phosphorus (P) fertilisers are often required at planting and in early stages of tree establishment to achieve rapid early growth and high survival rates. In Tasmania, further application of N and P to plantations ranging in age from 2- to 20-years has also occurred. To effectively manage these plantations, a detailed understanding of nitrogen (N) fertiliser requirements and N retention in forests is required.

Two field fertilisation experiments were used in this study, one in a 20-year-old *Pinus radiata* D.Don plantation growing in the north-east of Tasmania, on a Yellow Kurosol, and one in a 5-year-old *Eucalyptus regnans* F. Mueller plantation growing in the south, on a Brown Ferrosol. Both of these experiments were established in the early 1980s. Treatments at both sites included various combinations of P and N, applied as single-superphosphate and ammonium sulphate through a period of up to thirteen years. This study examines fertiliser-use, efficiency and impact on forest sustainability, including a detailed examination of the soil profile and litter at both sites. Nitrogen cycling was also examined, concentrating on the effect of N fertilisation on N mineralisation in the contrasting surface soil horizons.

After fifteen years of measurements, nitrogen fertilisation significantly increased volume growth at both sites. Two single applications of P alone (totalling 144 kg P ha^{-1}) doubled *P. radiata* stem volume from 78 m^3 ha^{-1} (Nil) to 192 m^3 ha^{-1} (P). Annual N fertilisation for a period of thirteen years (in addition to the P fertiliser) further increased *P. radiata* stem volume from 192 m^3 ha^{-1} (P) to 344 m^3 ha^{-1} ((P)N1Y), at age 34 years. In contrast, applications of P alone (up to a total of 598 P ha^{-1}) had no effect on *E. regnans* growth, while annual N (plus P) fertilisation for thirteen years, doubled *E. regnans* growth from 125 to 281 m^3 ha^{-1}, at age 19 years.

Although fertilisers may be used to increase forest growth there is concern that long-term N application may impact on forest sustainability through changes in the soil chemistry. At both sites in this study, significant changes occurred in the soil profile due to long-term fertilisation. Soil pH decreased due to both N and P fertilisation, at both sites. Significant reductions of 0.7 and 0.3 of a pH unit were associated with the highest rates of fertilisation in topsoil (A1, 0-10 cm) and litter (O2 horizon), respectively. Substantial reductions in exchangeable Mg concentrations were also measured, particularly in the Kurosol.

In association with enhanced growth was a large increase in litter accumulated at both sites. Total litter masses (O1+O2) ranged from 34.4 to 91.6 t ha^{-1} under *P. radiata* and from 21.6 to 102.4 t ha^{-1} under *E. regnans*. At both sites, the O2 horizon masses were significantly greater with annual fertilisation and were a substantial nutrient pool. Under *P. radiata,* O2 horizon mass was 40 t ha^{-1} when unfertilised and over 70 t ha^{-1} with fertilisation, while under *E. regnans* the mass was 14 t ha^{-1} when unfertilised and 77 t ha^{-1} with fertilisation. This indicates that, in the cool temperate climate studied here, litter could be an important pool of nutrients.

Long-term, annual applications of N fertiliser had no significant effect on the annual rate of NNM measured in either the Kurosol or Ferrosol topsoil (0-10 cm). However, average rates in Kurosol topsoil were up to four-fold higher in the annually fertilised treatment. At both sites topsoil *in situ* net N mineralisation (NNM) rates measured at the end of the experiment were low, ranging between 13 and 52 kg N ha^{-1} yr^{-1}. Such low rates of N mineralisation might have been associated with a prolonged period of low rainfall that occurred throughout the 18-month measurement period.

To assess mineralisation independent of microclimatic effects that prevailed during the *in situ* study, rates of NNM were measured during aerobic laboratory incubations. In agreement with *in situ* studies, NNM in the Ferrosol topsoil was not changed by fertilisation. In contrast, the variation in rates of NNM between treatments for the Kurosol topsoil was greater than that measured *in situ,* with fertilised topsoil

mineralising ten times more N than that unfertilised. However, results were highly variable across moisture and temperature treatments.

Despite the high amounts of N and P that had been applied during annual fertilisation, differences in total N and NNM in soil were small and highly variable. This result contrasted with the large differences in total N content and NNM rates in the O2 horizon from both sites. The influence of fertilisation on N cycling in litter produced clear results, i.e. daily rates of mineralisation (measured by aerobic laboratory incubations) were higher in annually fertilised than unfertilised litters, at both sites if incubations went for 7 days or longer. In contrast, low mineralisation rates in both topsoils often produced similar daily rates of NNM regardless of fertiliser treatments. In both topsoils, 60 days was required to produce a significant cumulative effect, resulting from a divergence in NNM rates between the fertiliser treatments during the later stage of the incubation. Increased replication of samples in laboratory experiments did increase the sensitivity of NNM measurements and significant differences between fertiliser treatments were measured.

These results confirm the importance of litter as an N source in cool temperate plantations. The importance of the litter layer in N cycling was particularly evident under *E. regnans,* where N was most concentrated in the litter layer. In addition, the *E. regnans* O2 horizon accumulated significantly more P, S and Ca due to annual fertilisation.

The effects of air-drying, incubation period, moisture content and temperature on NNM in laboratory studies were examined and found to depend on both the site and fertiliser treatment. This study indicated that higher NNM rates in topsoils would occur if soils were not maintained moist prior to and during incubations. This was particularly important for wetter sites, where canopy closure had occurred, resulting in smaller moisture fluctuations, as observed in the Ferrosol topsoil. Laboratory incubation conditions also influenced correlations between nutrient content (total N, P S and Ca) and NNM. For example, NNM rates in the Kurosol topsoil were linearly correlated with N, P and Mg concentrations and pH when incubated at 20°C (p

<0.05), but only with P at 10°C. This study indicated that minimum disruption of soil processes, particularly by drying was essential if accurate measurements of changes in soil N, in the cool temperate environment, were to be obtained. Hence, I could not identify a reliable indicator of NNM for these plantations.

In agreement with the *in situ* study, application of N fertiliser seasonally (June, October, January or April) resulted in a short-term elevation of mineral N (less than six-months), particularly at the wetter Ferrosol site. April fertiliser application provided the longest period of enhancement and October the shortest, indicating that the current operational practice, of applying fertiliser in autumn (March-May), provides an adequate window for fertiliser uptake to occur. These trends also confirm that, independent of the time of fertiliser application, a six-month delay after fertiliser application was adequate to determine long-term fertiliser affects on NNM. This delay allowed mineral-N concentrations to attenuate to a low value that facilitated NNM measurements. In agreement with previous studies, at both sites rainfall appeared to be a strong regulator of mineral N availability after fertilisation. The importance of the litter was also highlighted, because it retained N fertiliser and thereby limited N leaching.

In situ rates of NNM and tree growth in the nil and annual fertilisation regimes in the *P. radiata* plantation were modelled using the process-based model CABALA. The model was developed with forest managers in mind as part of a silvicultural decision support system and links C, water and N flows through the atmosphere, tree and soil. In this study, CABALA was validated for *P. radiata* using parameterisation from published and unpublished data derived from an independent study. Predicted growth increases due to annual fertilisation within 15 % at age 34 years. Limitations to the current simulation were often due to the assumption that P was not limited at this site. Other responses to N movement in the forest system were also adequately predicted. However, this study indicated that functions for N mineralisation and canopy development need to be more sensitive to fertiliser inputs to adequately predict N availability in the mineral soil and litter layers. There currently seems to be an over-dependence on C: N ratio, because it drives predictions of N mineralisation in the

CERES model, but there is doubt that this is an important controlling variable in forest systems. Further study of *in situ* NNM rates of litter horizons would be required to clarify the amount and mechanisms of N recycling that occur in these systems.

Large growth rate increases from N fertilisation in both *P. radiata* and *E. regnans* plantations were often associated with significant changes in soil chemistry and litter accumulation. Substantial reductions in exchangeable Mg concentrations and soil pH indicate that careful site management is required. Significant accumulation of litter under these plantations may act a substantial future source of nutrients, including N availability for further tree growth, particularly when N mineralisation is low and not significantly effected by long-term fertilisation.

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1.1 Introduction

To achieve the required economic return on investment on plantations in Tasmania, Australia, mean annual growth in excess of 20 m^3 ha^{-1}, and probably 25 m^3 ha^{-1} are required (Forestry Tasmania pers. com). Many of the sites available for plantation establishment in Australian have soils of low fertility with low levels of organic matter, which will therefore require fertiliser additions to reach industry targets (Neilsen and Davis, 1984). Sites requiring fertilisation will also become more common as soil organic matter and soil nutrient levels decline due to wood production above sustainable levels (Hamilton, 1965; Raison, 1980; Squire and Flinn, 1981; Waring, 1981). Large losses of organic matter and consequently N during previous harvesting and site preparation practices have been attributed to the decline of growth in successive rotations of plantations (Keeves, 1966; Wise and Pitman, 1981). Economic plantation production cannot be sustained from native soil N alone (Neilsen, 1983).

Many Australian soils require Nitrogen (N) and phosphorus (P) fertilisers at planting, and during the early stages of tree establishment to achieve rapid early growth and high survival rates (Gentle *et al.*, 1965; Judd *et al.*, 1996; Neilsen, 1996). Later-age application of P at P-deficient sites can increase *P. radiata* plantation growth by an average of 10 m^3 ha^{-1} yr^{-1}, for a periods of 10 years or more (Gentle *et al.*, 1965; Neilsen *et al.*, 1984). In Tasmania, substantial later age P fertilising programs have been in place since the early 1970's (with between 60 – 70% of Forestry Tasmania's estate fertilised). Large volume increases in *P. radiata* plantations from multiple applications of N fertiliser also occur (Neilsen *et al.*, 1992; Neilsen and Lynch, 1998), resulting in the development of a later age N fertiliser program in Tasmania since 1990.

Following fertilisation, increases in litter accumulation occur in a range of forest stands (Baker *et al.*, 1986; Nohrstedt, 1990; Theodorou and Bowen, 1990; Neilsen and Lynch, 1998). However in a subtropical climate, litter accession can be low, as measured in mixed Eucalyptus forests at 1.8 to 3.6 t ha^{-1} yr^{-1} (Birk, 1979). In a warm temperate climate in New Zealand, high rates of forest floor organic debris

breakdown in *E. regnans* also resulted in low amounts of accumulation of 4.7 to 11.0 t ha^{-1} (Frederick *et al.*, 1985). The litter layer is important in the cycling, retention and supply of nutrients in forests (Tamm and Popovic, 1995; Neilsen and Lynch, 1998) and after canopy closure the cycling of N through re-translocation in the tree and mineralisation of litter become more important sources for growth of new tissues than nutrients derived directly from the mineral soil (Cromer *et al.*, 1993). In *P. radiata*, Hunter and Hoy (1983) observed an increase in litter mass from 1.82 tonnes litter ha^{-1} with 10.2 kg N ha^{-1} to 4.13 tonnes litter ha^{-1} containing 24.5 kg N ha^{-1}, due to the application of 400 kg N ha^{-1}. However, in a review of many studies by Aber and Melillo (1980) fresh litter was considered a mineral N sink, rather than a source. Fyles and McGill (1987) also indicated that the addition of recent litter material favours N immobilisation, whereas mineralisation dominates in the O2 layer.

In addition to increased N retention in litter layers, increase in the rates of N mineralisation in soil following N fertilisation have been observed in many field and laboratory studies (Johnson *et al.*, 1980; Adams and Attiwill, 1991; Whynot and Weetman, 1991; Aggangan *et al.*, 1998). Increased N mineralisation following P application has also been observed (Waring, 1969; Khanna *et al.*, 1992; Falkiner *et al.*, 1993). However, the literature on the effects of fertilisation on NNM rates is often conflicting.

Although fertilisers may be used to increase forest growth there is concern that long-term N application may affect forest sustainability through changes in soil chemistry. Application of N fertiliser has been identified as a source of increased acidity in soils across a range of sites (Adams and Martin, 1984; Tamm and Popovic, 1995; Homann *et al.*, 2001). Low soil pH decreases the rate at which organic N is mineralised (Adams and Martin, 1984), can increase the availability of Al to toxic levels and can reduce the availability of cations such as Ca and Mg.

To effectively manage and refine fertiliser operations in both Eucalyptus and Pine plantations, a more detailed understanding of fertiliser requirements is essential. This requires increasing our understanding of N retention in these forests and the longevity

of growth responses (Mead and Pritchett, 1975b). Both N retention and subsequent availability relate to rates of mineralisation in the soils under study and are therefore a response of the local environment and N status (Adams *et al.*, 1989b). Measurements of N mineralisation indicate of the amount of N that becomes available to trees for uptake and subsequent growth (Pastor *et al.*, 1984; Adams and Attiwill, 1986). Tasmania has a cool temperate climate, substantially different to the rest of Australia. In mature cool temperate forests, N mineralisation rates are often below 10 kg $ha^{-1}yr^{-1}$ (Adams *et al.*, 1989a), which contrast markedly to N uptake required for plantation growth, of up to 200 kg N ha^{-1} (Cromer *et al.*, 1993; Smethurst *et al.*, 2004a). In Tasmania, NNM rates between 13 and 188 kg N ha^{-1} yr^{-1} were observed in surface soils (0-10 cm) of *Eucalyptus nitens* plantations being established on Kurosols and Ferrosols (Wang *et al.*, 1998; Moroni *et al.*, 2002)

The experimental sites studied in this thesis provided an opportunity to determine N mineralisation as effected by long-term fertilisation in the Tasmanian environment. Studies of long-term applications of N fertiliser in the past have predominantly occurred in the Northern Hemisphere, in the context of high rates of atmospheric N input. In contrast, this study investigates the effects of N fertilisation at rates up to and above operational requirements for plantation growth, in the Southern Hemisphere away from industrial areas. The soils chosen in this study, i.e. a Yellow Kurosol and a Brown Ferrosol, represent the low-to-medium range of productivity of the *P. radiata* and eucalypt estates in Australia. The Kurosol profile is also very similar to the soil researched intensively in the ACT (Khanna *et al.*, 1992) the major difference being that the Tasmanian site is in a cooler environment. The combination of both sites with contrasting soils and plantation species provides an opportunity to compare the effects of long-term fertilisation, in temperate forests, on nutrient cycling and N mineralisation.

To understand how long-term fertilisation affects plantations in the Tasmanian environment, this thesis tests the following hypothesis:

"Long-term application of nitrogen fertiliser increases the growth of *E. regnans* and *P. radiata* plantations, increases the rate of NNM in soil and litter horizons, and results in a redistribution of N in these systems that can be adequately simulated by a process–based model."

The specific objectives were as follows;

1. Review the effects of climate, vegetation, soil and site management (in particular N fertilisation) on NNM rates in forest soils.

2. Determine the effects of long-term fertilisation on:

- growth of a *P. radiata* D.Don plantation and a *E. regnans* F. Mueller plantations on contrasting soils.
- nutrient distribution and pH in the soil, including litter.
- Rates of NNM in litter (O2) and topsoil (A1) in both field and laboratory conditions.

3. To assess mineralisation in the topsoil and litter horizons independent of microclimatic affects that prevailed *in situ*, laboratory experiments were conducted to determine the effects of controlled parameters on NNM, i.e.

- moisture content and air-drying
- incubation period, and
- incubation temperature

4. Test whether NNM or other attributes measured in these plantations could be adequately simulated by a currently available processed-based model.

5. Discuss the implications of long-term fertilisation on site productivity, soil conditions and management.

This thesis is divided into 10 chapters, a flow diagram of the thesis development and outline is given in Figure 1.1.

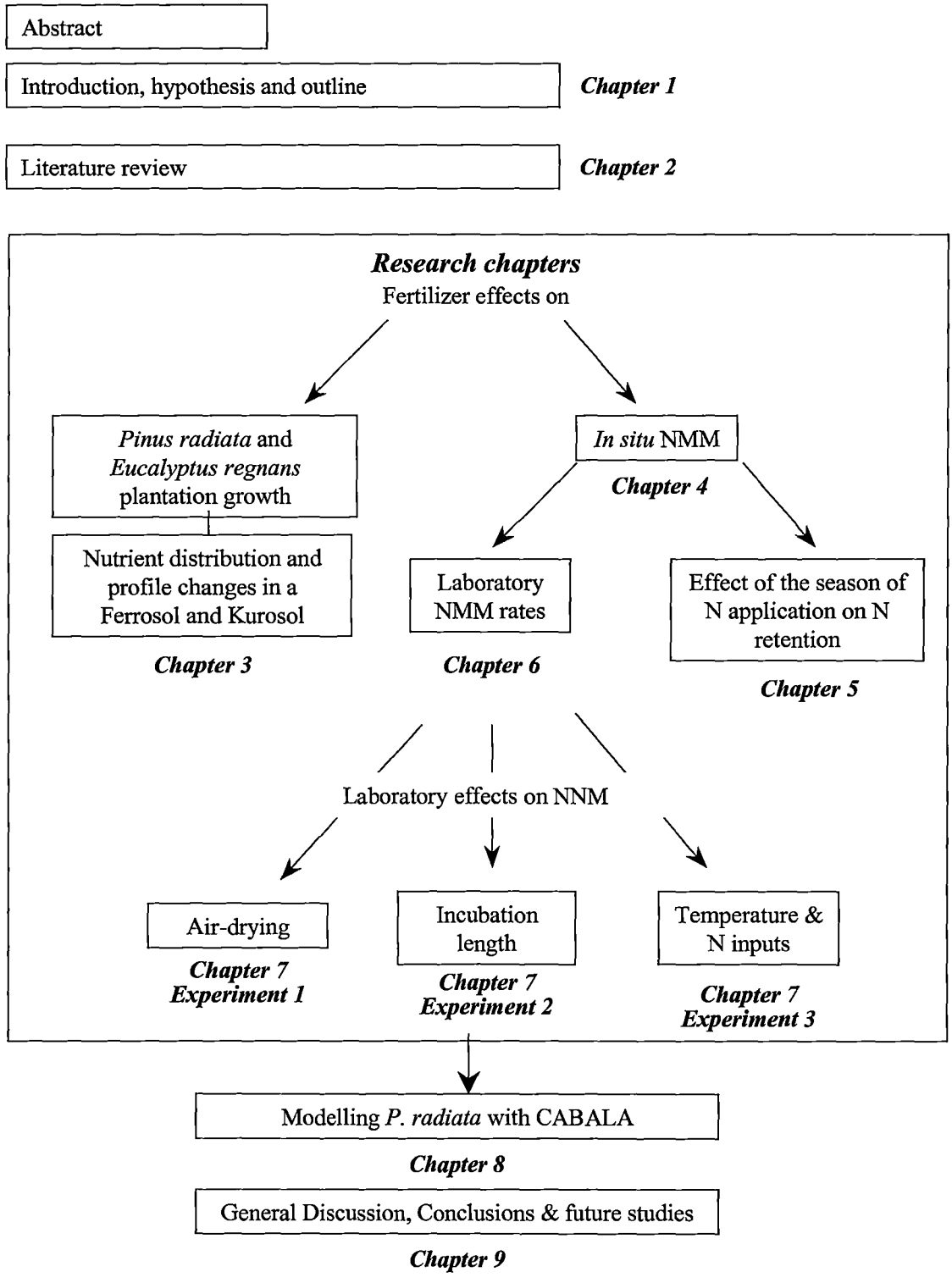


Figure 1.1 Schematic outline of the thesis.

2. Literature Review

2.1 Introduction

Soil is an integral part of the forest and forest growth is a direct product of the quality and quantity of the soil (Grant *et al.*, 1995). The growth of forests depends on the total quantity of nutrients available from the mineral soil and organic matter, on nutrient re-translocation in the tree and on atmospheric inputs. A key to optimising tree growth therefore depends on understanding the complex interaction of processes affecting the long-term availability and cycling of nutrients in the forest environment. These complex processes include factors such as; soil mineralogy, micro- and macro-climates, organic matter and soil mineralisation rates, and the plant-soil relationships of the species occupying the site.

Nitrogen is often the most growth-limiting nutrient in both terrestrial and aquatic systems. The majority of forest plants rely on microbial transformations of organic material to release N for plant uptake and growth. In undisturbed forest soils, inorganic N for plant growth is derived almost entirely from the decomposition of organic forms of N in a process called N mineralisation. Therefore, the efficient conversion of organic N to available mineral forms is vital for plant nutrition. Microbial mineralisation of organic N is influenced by factors such as temperature, moisture, substrate concentration (quantity), and ease of utilisation by, or accessibility to, micro-organisms (quality) (Fyles *et al.*, 1990).

Nitrogen mineralisation has been widely studied as an indicator of site productivity, with productivity in many forest ecosystems increasing with soil N mineralisation (Pastor *et al.*, 1984; Adams and Attiwill, 1986). Nitrogen mineralisation in soils have also been significantly correlated with soil characteristics such as total soil N content, mineral soil N concentration, soil organic C, or soil pH (Nadelhoffer *et al.*, 1983; Pastor *et al.*, 1984; Gower and Son, 1992). In addition, litter amount and quality influence N mineralisation from soil organic matter and that, in turn, influences leaf-litter production and quality. As this interaction is controlled by feedback between vegetation and soil organic matter dynamics, practices that alter the function of either

ecosystem component, may alter the amount of N cycling, and ultimately effect site productivity (Nadelhoffer *et al.*, 1982).

Optimal N fertilisation prescriptions aim to maximise the assimilation of exogenous N into trees, by synchronising N availability with periods of maximum N uptake. To prepare such a prescription it is necessary to understand N availability in litter and soil systems and its subsequent uptake into and retranslocation in the tree. Inherent site properties determine the probability and magnitude of growth response to fertilisation with N and P alone and in combination. The duration of enhanced growth from fertilisation will depend on the duration of increased soil and litter N mineralisation and retranslocation of N within the tree. Forests with inherent low rates of N mineralisation show large growth response to N fertilisation, if P availability is adequate (Raison *et al.*, 1987). However, the duration of growth enhancement due to N fertiliser is often limited to 5 to 10 years post fertilisation (Fagerstrom and Lohm, 1977; Miller, 1981; Fisher and Binkley, 2000). Prolonged responses to tree growth from long-term simulations of N supply from fertilisation have been indicated (Raison *et al.*, 1990; Neilsen *et al.*, 1992; Neilsen and Lynch, 1998). In contrast, single applications of P fertiliser have produced continued response for many years in a number of forest crops (Ballard, 1978; Turner and Lambert, 1986).

Reports on long-term affects of fertilisation on N cycling in forest soil and subsequent N mineralisation vary greatly. Inconsistencies often reflect variations in site characteristic and fertiliser-application management (Aggangan *et al.*, 1998) and limitations when measuring NNM (Adams *et al.*, 1989b). However, increased NNM in forest soil due to N fertilisation have been measured both during laboratory and field incubations (Johnson *et al.*, 1980; Hingston, 1984; Aarnio and Martikainen, 1995), up to six years after application (Smolander *et al.*, 1998). Interactions between microbial utilisation of N and its net N mineralisation depend on a range of site and environmental conditions. Temperature and moisture content together with pH, are among the most important abiotic factors influencing biogeochemical transformations, including N mineralisation (Tietema *et al.*, 1992).

The prediction and realisation of responses as measured by tree growth is only a rough guide to the direct effects of fertilisation on the whole forest system. The role of the soil and internal recycling in the tree and between the tree and soil are important in the development of stand management and fertiliser application regimes. Large losses of organic matter and consequently N during previous harvesting and site preparation practices have been attributed to the decline of growth in successive rotations of plantations (Keeves, 1966; Wise and Pitman, 1981).

Numerous investigations have demonstrated the role of seasonal temperatures on microbial growth and N mineralisation (Foster, 1989; Powers, 1990; Nadelhoffer *et al.*, 1991; Verburg and Van Breemen, 2000). The following review highlights the major factors influencing tree growth and N mineralisation in forests. These are segregated into sections that highlight the principles and ideas tested in the thesis.

2.2 Fertilisation Effects on Plantation Growth and Soil Profile Changes

Many factors can limit the growth and rotation length of a plantation. Factors such as climate, soil physical structure and soil biochemistry interact to form a complex set of indicators of a site's inherent potential for plantation growth. Many Australian soils are old or derived from highly weathered materials and widespread P deficiency has been well documented (Attiwill, 1983). As a consequence, the establishment of plantations such as pines on these P deficient soils commonly depends on the use of P fertilisers at rates between 100 and 300 kg super phosphate per hectare, at the time of planting (Attiwill, 1983). Fertilisation of eucalypt plantations with N and P is also common during the first few years after planting as it leads to substantially increased wood yields (Neilsen *et al.*, 1984; Attiwill, 1996).

Increased plantation growth occurs from later age application of P fertiliser in P-deficient sites (Waring, 1969; Flinn *et al.*, 1979b; Neilsen *et al.*, 1984; Herbert, 1990; Turner *et al.*, 2002). The magnitude and longevity of response to P application depends on soil properties such as the inherent P concentrations, P sorption capacity and soil pH (Ballard, 1978; Pritchett and Comerford, 1982). For example, in *P. radiata* applications of at least 120 kg P ha^{-1} were required to increase growth on

strongly fixing soils in south-eastern Australia (Hopmans and Flinn, 1998). Where adequate P contents are measured in soils and in current *P. radiata* needles, no significant increases in growth due to P application occur (Raison *et al.*, 1990). In contrast, where soil and foliar P levels are low or marginal long-term growth response due to a single application have been measured, extending into a second rotation (Ballard, 1978; Gentle *et al.*, 1986). In a study of to slash and loblolly pine stands on six soil types, Fisher and Garbett (1980) observed soils with high organic matter contents and large reserves of N responded dramatically to P application, but when N was limited, N fertilisation was required in combination with P.

Enhanced growth rates due to N fertilisation are site dependent (Fisher and Garbett, 1980). For, example, Adams and Attiwill (1983) observed no significant increase in growth of 23-year-old *P. radiata,* 30 months after the application of N fertiliser at high rates (500 and 1000 kg N ha^{-1}). Nitrogen excess problems can also occur resulting in no growth response to N application on ex-pasture sites (Birk, 1991). In a study of slash pine growing in the lower coastal plain of the south-eastern United States, Kushla (1980) observed no response to urea applied at rates of 22 and 90 kg N ha^{-1} on young stands of slash pine, while semi-mature stands (9-18 years old) responded to N at rates of 55, 110, 220 kg N ha^{-1}. Limited fertiliser responses in young stands were associated with restricted root growth on wet soils with high surface clay contents.

In Tasmania, large volume increases have been obtained due to annual applications of N fertiliser to *P. radiata* stands (Neilsen *et al.*, 1992; Neilsen and Lynch, 1998). Application of 100 kg N ha^{-1}, on 16-year-old stand of *P. radiata*, for 12 years resulted in fertilised trees having significantly higher N content for all components at all ages than unfertilised trees, and a 150 % increase in growth rate four years after the cessation of fertiliser. Many studies have found that N fertilisation effects conifer growth through the rapid (within six months) increases of foliar N concentration, foliar mass and colour. These increases in foliar N concentration reach a maximum within a year and level off to its original value within a few years (Miller, 1981; Fife and Nambiar, 1997). The rate of wood production, on the other hand, starts to increase

after a time delay of around one year. This positive growth response is then retained for approximately ten years, with a maximum occurring after three or four years (Fagerstrom and Lohm, 1977; Hunter and Hoy, 1983; Neilsen and Lynch, 1998). For example, in a study of six year old *P. radiata* stands, Fife and Nambiar (1997) observed increases in basal area compared to the control of 6, 21 and 29 percent, five years after the application of ammonium sulphate at rate 150, 300 and 600 kg N ha^{-1}, respectively. Nitrogen application increased foliar N concentration in all needles (1-3 years old), and remained elevated well beyond observed N elevations in soil, which declined rapidly to reach pre-treatment within 12 months. Consequently, the authors concluded that the benefit of N application was due to increase in growth rates when environmental conditions were suitable mainly through recycling of N pools in the tree. In agreement, even though Raison *et al.* (1990) observed an increase of growth due to fertilisation with 400 kg N ha^{-1} (ammonium sulphate), no increase in growth occurred during drought conditions.

Although large quantities of N fertiliser have produced growth responses, declines in such responses are associated with decreasing soil N concentrations within the years following application (Williams, 1972; Johnson *et al.*, 1980; Adams and Attiwill, 1983; Hingston and Jones, 1985; Adams and Attiwill, 1991; Khanna *et al.*, 1992; Fife and Nambiar, 1997; Smethurst *et al.*, 2001). Therefore to maintain diverging growth from untreated plantations, repeated applications are required (Neilsen *et al.*, 1992; Neilsen and Lynch, 1998). The period between repeated applications will depend on the period of elevated availability and cycling of applied nutrients in the soil system and the efficiency of internal recycling of nutrients (Switzer and Nelson, 1972). Snowdon (2002) describes short-term growth responses as a Type I response, where the response is characterised by an initial increase in growth that is not sustained in the long-term. In comparison, the responses to P application into the second rotation discussed above are considered a Type II response. That is, P application results in a long-term increase in growth and a change in site quality. The responses observed by Neilsen *et al.* (1992) in *P. radiata* could be considered Type II responses due to sustained increases in growth from multiple fertiliser applications. However, after fertiliser cessation, the response declines indicating a Type I response.

Following fertiliser application nutrients become distributed between the overstorey, ground vegetation, litter and mineral soil horizons. Concurrent with plantation biomass increase, there is an increase in litter mass. The litter layer plays a major role in mineral cycling in forest ecosystems. To improve the understanding of the effects of fertilising on nutrient cycling in plantations it is necessary to quantify the extent to which fertilising increases the turnover of nutrients through the litter layer. At sites with low nutrient levels in the mineral soil, litter quality, as measured in terms of C: N ratio, has been shown to be highly correlated with tree volume growth (Smith *et al.*, 2000).

The upper organic horizons of forest soils can be divided into three layers L, F and H layers. These layers represent different stages in the humification process. The L layer is the litter layer composed of plant products, leaves and twigs, which although weathered retain their original structure. The F layer is the fermentation layer composed of plant remains partially crushed and decomposed, but with tissues that are still recognisable. The H layer is the dark brown or black amorphous humus layer. The transition from the L to the H layer via the F layer is through physical, chemical and biological processes. These processes result in the reduction in the physical structure of the particles, a darkening in the colour, and a lowering of the C: N ratio, relative to the original L layer (Spurr and Barnes, 1980). Often, the strongly decomposed H layer is virtually non- existent. In the Australian system of horizon classification the layers are classified as O1 (L) and O2 (F and H) (McDonald *et al.*, 1990).

Tamm and Popovic (Tamm and Popovic, 1995) in northern hemisphere studies noted the importance of the litter in the retention of base cations within the forest system, and its role in planning management systems for fertiliser additions. The mass and nutrient content of litter is variable between species, stand age, site and fertiliser treatment (Feller, 1978; Baker *et al.*, 1986; Crane and Banks, 1992; Neilsen and Lynch, 1998; Canary *et al.*, 2000) and temporally (Frederick *et al.*, 1985). Turvey *et al.* (1994) observed that litter accumulated on the forest floor at a rate proportional to the productivity of the *P. radiata* stands. They also noted a close link between the

nutrient contents of *P. radiata* and that of the underlying soil. In contrast, Florence and Lamb (1974) observed no correlation between productivity and litter accumulation.

Nitrogen due to fertilisation can accumulate in the tree and soil organic layer in approximately equal amounts (Miller *et al.*, 1979). Neilsen and Lynch (1998) studied the changes of the litter layer over a period of 10 years and demonstrated the importance of this layer in buffering applied nutrients into the rhizosphere. Concurrent with large growth increases from fertilisation, their research also showed increases in litter layer mass (O2) from 15 t ha^{-1} to 50 t ha^{-1} with the N in the litter layer (O1 layer plus O2) increasing from 148 kg N ha^{-1} to 593 kg N ha^{-1}. Hunter and Hoy (1983) observed an increase in litter mass from 1.82 tonnes litter ha^{-1} with 10.2 kg N ha^{-1} to 4.13 tonnes litter ha^{-1} containing 24.5 kg N ha^{-1}, from the application of 400 kg N ha^{-1}. Similar trends were observed by Maier and Kress (2000) in loblolly pines fertilised with urea.

Following fertiliser application a proportion is also lost from a site through leaching and gaseous diffusion (Miller, 1981). With the increasing use of fertilisers, there are increasing concerns about their long-term environmental costs and impacts. Concern over traditional fertilisers, such as urea, result from losses due to N leaching and denitrification. Extensive research in temperate ecosystems across Northern America and Europe have shown that N inputs both from atmospheric and fertiliser additions could lead to soil acidification, depletion of base cations and mobilisation of Al at potentially toxic levels (Adams and Martin, 1984; Matson *et al.*, 2002). Decreases in exchangeable Ca and Mg within the soil profile, associated with acid deposition and harvesting have been linked to declines in forest health and productivity (Watmough and Dillon, 2003).

Acidification has also been reported for many agricultural soils in Australia (Porter *et al.*, 1995). The rate of pH decline is variable and depends on the rate and type of fertiliser application, the soil type and soil parent material (Bromfield *et al.*, 1983; Binkley *et al.*, 1988; Porter *et al.*, 1995). Differences in soil responses are generally

attributed to different clay mineralogy's, which changed the buffering capacities of the soil. Minor or no changes in pH have been observed in some long-term experiments, using either ammonium nitrate or urea (Nohrstedt, 1990; Homann *et al.*, 2001). Other studies, using the same fertilisers, have determined substantial reductions in soil pH, but only at rates of 74 kg ha^{-1} yr^{-1} or more, or at lower rates if combined with P and K fertilisers (Tamm and Popovic, 1995). Similar results are observed in agricultural (cropping) soils. Applications of 45 kg N ha^{-1} as ammonium sulphate have been calculated to lower the pH of an acid sandy loam by about 0.1 pH units. In contrast, a three-fold application would be needed to produce the same effect in a more highly buffered acid clay loam (McGarity and Storrier, 1986). The application of Urea to agricultural soils has been shown have a less acidifying effect than either ammonium sulphate or ammonium nitrate (Porter *et al.*, 1995). In Tasmania, a range of N and P fertilisers have been used in forest production, current applications are predominantly urea or a blend of urea and di-ammonium phosphate (UDPA) (Elliott and Hodgson, 2004).

In several studies significant decreases in the soil solution pH have occurred in subsoils, ranging from 0.5 to 1.9 units (Vestgarden *et al.*, 2001). Fertilising with ammonium sulphate has also resulted in soil solution pH declines in subsoils beneath Eucalypt plantations in Tasmania (Smethurst *et al.*, 2001). In this study the relationship between rate of N application and pH decline was predominantly linear on log-linear, and was effected by the rate of fertiliser application rather than the timing of applications.

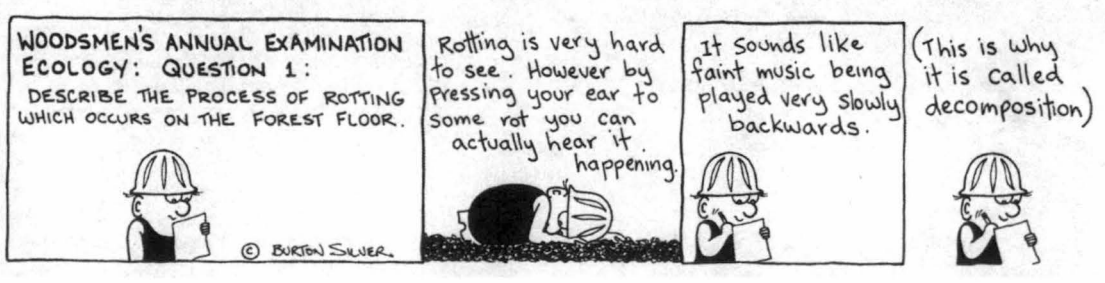
2.3 Soil organic matter, soil fertility and Nitrogen Mineralisation

Soil organic matter is an integral component in the maintenance of soil fertility and N availability in a forest ecosystem. The majority of forest plants rely on microbial transformations of organic material to release N for plant uptake and growth. In undisturbed forest soils, inorganic N for plant growth is derived almost entirely due to the decomposition of organic forms of N. Subsequently, the efficient conversion of organic N to available mineral forms is vital for plant nutrition. Soil organic matter also helps maintain the structural health of the soil, provides cation exchange sites,

and provides energy for heterotrophic soil micro-organisms (Adams and Attiwill, 1988).

For nutrients bound in the SOM to become available for plant uptake, they must first be released through a sequence of transformations. The ultimate conversion of organic matter forms of nutrients to inorganic compounds is called mineralisation. This nutrient release is predominantly a two-step process, initially a fauna-mediated particle size reduction, followed by a predominantly microbial mediated mineralisation of the organic substrate. A review of fauna mediated decomposition has been conducted by many authors including Waring and Schlesinger (1985). The microbial mediated processes have also been reviewed at by many authors including Attiwill and Leeper (1987). The primary outcome of decomposition is a reduction in both litter mass and carbon concentration.

During decomposition, readily available (labile) polysaccharides decompose first by exo- and endo-enzymes (Armson, 1979). This leaves behind an accumulation of more recalcitrant chemical structures such as alkyl, aromatic (aryl and o-aryl), and carbonyl materials, which decompose more slowly (DeMontigny *et al.*, 1993; Mathers *et al.*, 2000). The timing of this decomposition process was described by Berg (2000) as a two-phase model. In the early stage (months to one year) decomposition rate is controlled by climate, major nutrients (N, P and S) and water-soluble nutrient concentrations. Water-soluble components decompose quickly (in a few months) before reaching relatively stable levels. Subsequently, the concentrations of major nutrients and lignin start to increase, and dominate later stage decomposition rates.



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The relative amount of organic matter in a soil at a steady state can vary greatly with climatic conditions, soil acidity, drainage conditions, inorganic nutrients and soil parent material (McLaren and Cameron, 1996). Different organic components are utilised to very different extents by soil microbes and will therefore remain in the soil for different periods of time. Decomposition rate of fresh plant litter may decrease from 0.1 percent per day in fresh litter to 0.00001 percent per day or lower in more completely decomposed material (Berg, 2000). A number of models have been developed to determine long-term changes in organic matter over time, in both soil and litter fractions (Campbell and Doeg, 1989). Due to the complex nature of decomposition and the chemical change in the litter as decomposition continues from readily decomposable to recalcitrant chemical compounds, the indices based on initial litter quality often have limitations in predicting long-term decomposition rates (Corbeels, 2001). Models of organic matter transformations generally divide the various organic matter constituents into plant residues and SOM. These components are then further divided into fractions characterised by their rate of transformation (i.e. fast, slow and resistant). In their review of modelling litter quality effects on decomposition, Paustain *et al.* (1997) concluded that in the wide array of litter decomposition models available the major differences were associated with, whether the model accounted for microbial turnover and the formation of secondary products and whether the pools of plant compounds and decomposition products were grouped into discrete or continuum pools.

In agricultural soils, the RothC model was developed to model organic C turnover from the Rothamsted long-term field experiments. This model was one of the first widely used C transformation models. The model assumes six pools of C are available for decomposition, these are defined by: (i) decomposable plant material (DPM); (ii) resistant plant material (RPM) (iii) slow microbial biomass pool (BIO-S); (iv) fast microbial pool (BIO-F); (v) humidified organic matter (HUM); and organic matter inert to biological attack (IOM). In this model each pool decomposes at different rates and each rate is modified by climate and a plant protection factor. Although these decomposition rates were developed in agricultural soils, the constants were found to be also suitable for soils under plantation systems (Paul and Polglase, 2004b). The CENTURY model (Campbell and Doeg, 1989), also incorporated multiple SOM

compartments (active, slow and physically protected) and simulates decomposition rates that vary as a function of monthly soil temperature and precipitation, including both N and C flows.

In most terrestrial systems, the flow of C and N are closely interrelated. Generally, mineral N does not accumulate in undisturbed forest sites since C inputs are high and N is the element limiting decomposition (Paul and Juma, 1981). Simultaneously to N mineralisation, microbial biomass utilises available N for its development, a process known as immobilisation. Whether the mineralised N is immobilised again mainly depends on the C: N ratio of the microbial biomass. Immobilisation is recognised as a major process in eucalypt forest soils (Adams and Attiwill, 1986). A review of the N cycle in forested ecosystems and a summary of the complex pathway and interactions of N in the forest has been given by Carlyle (1986).

Nitrogen mineralisation has been widely studied as an indicator of site productivity, with productivity in many forest ecosystems increasing with soil N mineralisation (Pastor *et al.*, 1984; Adams and Attiwill, 1986). The quantity of soil N mineralised in a given time depends on temperature, available water, rate of oxygen replenishment, pH, amount and nature of plant residues, and content of other nutrients (Stanford and Smith, 1972). Observed N mineralisation rates over time, in laboratory incubations, generally follow one of four patterns: (I) Immobilisation of N during the initial period of incubation, followed by mineralisation of N in the later period (Haque and Warmsley, 1972). (II) A rate of release that decreases with time. In 15 out of 39 soils studied by Stanford and Smith (1972) there were slight to marked tendencies for the rates on NNM to decline with continued incubation during 30 weeks. Adams *et al.* (1989b) also found a decrease in the rate of NNM with increasing time of incubation. (III) A steady linear release with time during the whole period of incubation. Tabarabai and Al-Khafaja (1980) found when incubating soils at 20 to 35 °C cumulative N mineralisation was linear with time. (IV) A rapid release of nitrate during the first few days, followed by a slower, linear, rate of release (Stanford and Smith, 1972; Feigin *et al.*, 1974; Bonde and Rosswall, 1987). The pattern that applies, depends largely on the manipulation of the substrate before incubation occurs, and the

relative change in the incubation parameters given above, from the native state. For example, Bonde and Rosswall (1987) established that a soil responds to drying and re-wetting with a flush of C and N mineralisation, the magnitude depends on soil characteristics.

Several techniques including *in situ* cores and *in situ* polyethylene bags have been validated and used to study N mineralisation in the field (Richards *et al.*, 1985; Raison *et al.*, 1987; Smethurst and Nambiar, 1989b). In the laboratory, N mineralisation studies follow two basic methodologies, anaerobic incubations and aerobic incubations. Rates of mineralisation using laboratory incubations may or may not be correlated with rates of mineralisation in the field. For example, Connell (1995) found a poor correlation ($R^2 = 0.20$) between laboratory incubation and *in situ* field measurements, with mineralisation rates of up to 10-times greater in the laboratory than the field. These differences are often associated with the large changes in soil structure (sieving and mixing) and moisture content (air-drying and re-wetting) that occur during sample preparation (Birch, 1958; Lund and Goksoyr, 1980; Van Gestel *et al.*, 1993), which are discussed in section 2.1.

In situ core methods have been extensively reviewed by a number of authors (Raison *et al.*, 1987; Adams *et al.*, 1989b; Smethurst and Nambiar, 1989b), and the technique was confirmed in the Biology Forest Growth (BFG) experiment for estimating rates of N mineralised, uptake and leaching (Raison and Myers, 1992). In their review on *in situ* N mineralisation studies Adams *et al.* (1989b) summarised some of the effects of *in situ* core methodology, as a result of altering the soil environment; " (i) cessation from carbon input through decomposing litter and from fine root turnover; (ii) increased C inputs from fine-root turnover; (iii) modification of the moisture and temperature regimes relative to the bulk soil; and (iv) accumulation of inorganic-N."

Incubation of soils *in situ* have been undertaken over a range of periods, between 7 and 90 days, dependent on the expected NNM rate and field moisture fluctuations (Raison *et al.*, 1987; Adams *et al.*, 1989b; Smethurst and Nambiar, 1989b; Goncalves and Carlyle, 1994; Carlyle *et al.*, 1998b). Raison *et al.* (1987) observed NNM rates in

cores increased linearly with time, resulting in an appropriate containment period between 30 and 90 days. In their study, Adams *et al.* (1989b) recommended quite short containment periods of one to two weeks, to reduce the impact of containment on moisture fluctuations. However, they also concluded that "relatively long containment periods (> 4 weeks) may be useful for comparative purposes, and their use may be dictated by the practicality of visiting distant forests."

Rates of N mineralisation depend on many factors, including management practices such as clear-felling, ripping, mounding, slash burning and fertilisation (Raison *et al.*, 1987; Smethurst and Nambiar, 1990a; Connell *et al.*, 1995). In undisturbed soils under mature forests, rates of N mineralisation are usually low, ranging from 1 to 100 kg $ha^{-1}yr^{-1}$ (Binkley and Hart, 1989). The NNM rate depends greatly on the climate zone. In mature cool temperate forests, N mineralisation rates are often below 10 kg $ha^{-1}yr^{-1}$, while rates in mature tropical forests can be greater than 800 kg $ha^{-1}yr^{-1}$ (Adams *et al.*, 1989a). McClaugherty *et al.* (1982) reported rates of N mineralisation for temperate forests to range from 50-300 kg $ha^{-1}yr^{-1}$, while above ground demands for N range from 50-150 kg $ha^{-1}yr^{-1}$. In *P. radiata* sites in southern Australia, Carlyle *et al.* (1998a) observed *in situ* NNM rates between and 74 kg N ha^{-1} yr^{-1}, while Theodorou and Bowen (1983a) estimated rates around 50 kg N ha^{-1} yr^{-1}. In Tasmania, annual rates of NNM in mature eucalypt forests are reported between 16 and 51 kg N ha^{-1} (Adams and Attiwill, 1988), and in young eucalypt plantations between 13 and 188 kg N ha^{-1} (Wang *et al.*, 1998; Moroni *et al.*, 2002).

In terms of the proportion of total N present, annual rates of NNM are generally between 1 to 3 %. In south-eastern Australia under various management practices Connell *et al.* (1995) measured in vitro rates of 2 to 67 kg ha^{-1}, this amounted to between 0.1 and 3.1 % of the N to 30 cm depth. Cole (1995) also reported N mineralisation of 1 and 2 % of the total SOM in temperate forest. Mineralisation rates also varied greatly with altitude ranging from 20 to 80 kg $ha^{-1}yr^{-1}$. A value of 80 was considered adequate for plant growth, while a value of 20 indicated limited N availability.

2.3.1 Species Effects on Nitrogen Mineralisation

Important feedbacks occur between plant characteristics and nutrient availability in terrestrial ecosystems. The release of N from litter is an important process in the cycling of N in an ecosystem. On an ecosystem level, litter amount and quality influence N mineralisation from SOM and that, in turn, influences leaf-litter production and quality. As this interaction is controlled by feedback between vegetation and SOM dynamics, practices that alter the function of either ecosystem component, may alter the amount of N cycling, and ultimately effect site productivity (Nadelhoffer *et al.*, 1982). In addition, the nature of decomposition of litter influences the amount of C retained in the litter and transferred to the soil and effects the ability of a forest site to store C. This has important implications for carbon accounting and potential C trading schemes (Paul and Polglase, 2004a). Rates of decomposition and subsequent C storage in the litter and soil are also important when considering the possible effects of global warming on CO_2 emissions (Comins and McMurtrie, 1992). In a review of the effects of global warming on organic C storage, Kirschbaum (2000) concluded that warming was likely to reduce carbon storage by stimulating decomposition rates more than net primary production (NPP). In a review of soil responses to climate change Anderson (1992) noted that soils in cooler climates generally had greater soil organic matter in a less advanced stage of decomposition than soils in warmer climates and that the same principles applied to wetter and drier climates. However, warm soils with moderate base status are associated with high quality litter. Anderson (1992) also noted that litter quality, in terms of N, lignin and other factors, that changed the rate of decomposition, had a greater influence on soil organic matter in cooler climates.

In a forest soil, litter is the predominant form of organic matter addition (Armson, 1979). Litter is defined as the addition of freshly fallen leaves, twigs, stems, flowers, fruit, bark and roots. Additions vary with the type of forest, eg. forest species, successional stage, soil type, topography and climatic zone. These variables determine the quantity and quality of litter produced and subsequent composition of SOM. After canopy closure the cycling of N through translocation in the tree and mineralisation of litter becomes a more important source for growth of new tissues than nutrients derived directly from the mineral soil (Cromer *et al.*, 1993). The importance of litter

in nutrient availability for tree growth was highlighted by Grier *et al.* (1981) who suggested there is a progressive self isolation of trees roots from the underlying soil, as additional fine root growth occurring as the trees ages, shift upwards into the decomposing litter layer. The forest floor has been noted as a major storage pool for N and P in colder sites, often exceeding the content in vegetation (Johnson, 1995). In South Australian *P. radiata* plantations, Lamb and Florence (1975) observed that the litter layer represented a considerable proportion of the above ground organic matter, containing up to 55% of the total N and 30 % of the total P.

Although annual leaf and needle litter production, and N returned in leaf litter are strongly correlated with estimated N uptake (Nadelhoffer *et al.*, 1982), the rate of decomposition is often not correlated with N mineralisation across sites and vegetation types (Harris and Riha, 1991). No unique relationship has been found between mineralisation and litter quality (Heal *et al.*, 1997). In soils, litter decomposition is related both to the quality of the litter (Melillo *et al.*, 1982) and the soil matrix (Skene *et al.*, 1997). Differences between species in rates of litter decomposition are often not responsible for the differences in N availability. For example, Prescott and Preston (1994) observed that N mineralisation rates were not proportional to C mineralisation rates and respiration under three litter layers (western red cedar (*Thuja plicata* Donn ex D. Don), western hemlock (*Tsuga heterophylla* (Raf.) Sarg.)) and Douglas fir (*Psuedotsuga menziesii* (Mirb.) Franco)), even though nearly identical rates of mass loss of foliar litter in the three species occurred. Such discrepancies between rates of C mineralisation and net N mineralisation may result from differences in an often substantial re-immobilisation of mineral N by micro-organisms (Davidson *et al.*, 1992). The ratios of gross N mineralisation to NNM range between 2 to 100 in forest soil (Binkley and Hart, 1989). In addition, after N fertilisation up to 98 % of the N may be immobilised (Strader and Binkley, 1989).

Aggangan *et al.* (1999) observed that changes in N immobilisation were related to the amount and corresponding degree of incorporation of the litter into the soil. Furthermore, decomposition of litter can be reduced when N inputs are increased. For example, in the BFG experiment decomposition of litter was slowed when fertiliser

(400 kg N ha^{-1}) was applied. Over a three year period the proportion of litter lost was 28 % in unfertilised litter compared to 17 % in the fertilised (Raison *et al.*, 1990). In contrast, (Ribeiro *et al.*, 2002) observed that increased N, P and S concentrations in *E. globulus* leaf litter through fertilisation application did not cause significant increases in decomposition. Kelly and Henderson (1978) observed significant decreases in decomposition of white oak (Quercus alba L.) litter due to P addition but little effect from N addition. Skene (1997) suggested that physical protection by inorganic matrices was the limiting factor in the decomposition of high quality substrates (such as straw), whereas chemical protection was the limiting factor for low quality substrates such as Eucalyptus litter.

Vegetation species occupying a site influence the availability of cycling nutrients and are recognised as having a greater impact on soil N dynamics than microclimate variations (Gower and Son, 1992; Prescott and Preston, 1994). Although this is well recognised, the specific influence of vegetation on N mineralisation, separation of its influence from other factors, such as climate, soil, time, and topography are difficult to determine, due to the predisposition for species to grow on different sites. As a result, although some consistent trends have been identified (ie. low N mineralisation under pines), contradictory findings have been common (Prescott and Preston, 1994). However, recent studies in forests of several species on the same site have demonstrated distinct differences in N availability between species (Adams and Attiwill, 1986; Ellis and Pennington, 1989; Gower and Son, 1992; Prescott *et al.*, 1993; Prescott and Preston, 1994). For example, Jurgensen *et al.* (1986) observed that compared to *E. regnans* the litter layer under *P. radiata* contained more N, P, K and Mg. Differences in the nutrients in the litter layer were seen in plantations as young as four-years-old and regression analysis indicated that the differences between the two species increased with stand age. Gower and Son (1992) also observed significant differences between five species litter NNM rates measured in the laboratory and *in situ*, even though annual litter fall N content were similar between species. In Australia, Adams and Attiwill (1986) observed that higher rates of mineralisation and nitrification occurred in Ash forests *(E. delegatensis, E. regnans*), forests of greater productivity, than forest dominated by *E. obliqua* (Messermate) and *E. sideroxylon* (Red ironbark) and forests of lower productivity. It was noted that in lower

productivity forests immobilisation was almost equivalent to mineralisation, and nitrification was insignificant (Adams and Attiwill, 1986).

Tree species effects on nitrification have been strongly correlated particularly when N fixing species are included (Ellis and Graley, 1987; Ellis and Pennington, 1989). Nitrification can be promoted by the presence of *Acacia dealbata* and apparently inhibited by *Leptosperum lanigerum* (Ellis and Graley, 1987). Nitrogen fixing species such as *Acacia* also have a strong influence on soil N properties resulting in high concentrations of total N, mineralisable N and nitrate, while other soil properties such as pH, loss on ignition and total P remain unrelated to N mineralisation (Ellis and Graley, 1987).

Understorey species, although small in biomass, may contribute significantly to nutrient recycling, due to their relatively high concentrations of nutrients to mass and fast turnover (Birk, 1979). For example, Prescott and Preston (2000) observed that variation in NNM among sites was unrelated to overstorey species and decomposition rates. In addition, in Karri wet sclerophyll forests understorey was found to play a key role in nutrient cycling, contributing 30-70 % of the weight of many of the nutrients in the leaf component of the litter (O'Connell and Menage, 1982). However, in Jarra (*Eucalyptus marginata* Donn ex Sm.) dry sclerophyll forests in western Australia O'Connell *et al.* (1978) observed that the main factors determining element concentrations in the litter were soil differences and the overstorey.

The relative importance of the effects of plant species on N mineralisation is critical for assessing the long-term effect of flora and site management. The role of individual plantation species such as *Pinus radiata* on the overall improvement, preservation or degradation of a site has been contemplated by many. Lamb (1976) hypothesised that the poor-quality litter inputs characteristic of *P. radiata* plantation result in site degradation due to the cyclic effect of slow litter degradation and low N mineralisation. In glasshouse experiments, Skinner and Attiwill (1981) were able to show that *P. radiata* decreased the productivity of both native and pasture soil, through changes in N and P availability. Reductions in populations of bacteria, actinomycetes and microarthropods have also been measured in pine litter (Upadhyay

and Singh, 1985). Theodorou and Bowen (1981; 1983a) also observed a decline in bacterial numbers and an increase in fungal numbers under a second rotation *P. radiata* compared with the first rotation (previously pasture). However, both studies involved young age trees in the second rotation and noted that these effects were evident for less than 18 months. Under *P. radiata* plantations of different ages, Birk (1992) observed similar changes, with a decline in total mineral N concentrations under 15 year old plantations compared to younger 2 or 4-year-old stands. In contrast, Ross *et al.* (1995) found microbial biomass C and N and mineralisable N in litter and mineral soil showed no relationship with *P. radiata* stand age up to 33 years. In a comparison under *P. radiata* and *E. regnans* plantations in New Zealand, ranging in ages from 4 to 17 years, Jurgensen *et al.* (1986) observed C: N ratios in the litter were higher under the eucalypts ranging from 38 to56 compared to 23 to 29 under the pines. However, decomposition rates were clearly lower under *P. radiata.*

As a result of the complex integration between the N cycle and microbial turnover, correlations between specific soil and litter parameters and N mineralisation vary considerably. Parameters such as total soil N content, mineral soil N concentration, soil organic C, or soil pH were found not to be significantly correlated with annual soil N mineralisation (Nadelhoffer *et al.*, 1983; Pastor *et al.*, 1984; Gower and Son, 1992; Connell *et al.*, 1995). The N and lignin content of vegetation are the most commonly used variables for characterising litter quality in decomposition models. Models differ in the level of detail used to capture the dynamics of soil organic matter; models with several organic-matter pools may behave differently than single-pool models (Ryan *et al.*, 2000). However, the functions of substrate quality, in terms of litter species and C: N ratios, soil quality, and climatic factors such as temperature and moisture on N mineralisation have been established, and the role of each of these will be discussed in the following sections.

2.3.2 Substrate Effects on NNM

The direction of N transformation, from net mineralisation to net immobilisation, depends on a number of factors. Lignin content and C: N ratios of the soil organic matter are considered two of the most important. C: N ratios are considered good

predictor of leaf litter decomposition especially when the litter substrates have a low lignin content or show a wide range of lignin content (Taylor *et al.*, 1989). There have been various values put forward for the upper and lower limits of the C: N ratio where N mineralisation will occur. In the study of crop residues and litter layers C: N ratios between 24 to 44 have been associated with no NNM or N immobilisation (Edmonds, 1980; Schlesinger and Hasey, 1981; Trinsoutrot *et al.*, 2000). Parfitt *et al.* (1998) observed that C: N ratios of greater than 55 in *P. radiata* litter resulted in no net N mineralisation. However, other research has indicated that NNM occurs at higher C: N ratios, but at a much reduced rate (Van Cleve *et al.*, 1986; Usman *et al.*, 2000). Overall, the direction of N dynamics is influenced by much broader relationships than just C: N ratios. Berg and Ekbohm (1983) observed no fixed C: N quotients for N release, other factors, such as the system in which the litter was incubated were considered important. They compared a mature Scots pine litter and a clear cut Scots pine slash. In the first system, N mineralisation still occurred at a C: N ratio of 109, but in the second, no net N mineralisation occurred over a ratio of 63. In addition, Smethurst and Nambiar (1995) during a study of N mineralisation in a *P. radiata* plantation, revealed that although the C: N ratio decreased, from 38 to 31, during a three year period, N mineralisation also decreased, due to a change in conditions controlling microbial activity. As a result specific relationships between C: N ratio and N mineralisation have been difficult to quantify, and consideration of the system as a whole needs to occur.

In spite of the difficulties in associating N mineralisation directly with C: N ratios, good correlations between litter N and C: N ratios with N mineralisation have been observed by many authors (Pastor *et al.*, 1984; Adams and Attiwill, 1986; Prescott and Preston, 1994; Thomas and Prescott, 2000; Usman *et al.*, 2000). For example, Pastor *et al.* (1984) reported an inverse relationship between litter (ash corrected) C: N ratio and annual NNM for eight forests in Wisconsin, even though NNM was not correlated with N concentration. Conversely, Vitousek *et al.* (1982) found a direct relationship between the amount of N in the annual litter fall and the proportion of litter N mineralised in laboratory incubations. Prescott and Preston (1994) observed a high correlation between NNM rates and initial forest litter concentrations of N %, C: N ratio, lignin % and lignin: N ratios, and Thomas and Prescott *et al.* (2000) observed

that in litter layers P, NO_3^--N and extractable mineral N concentrations were highly correlated with NNM. In contrast, Sollins *et al.* (1984) reported NNM in whole soil and litter layer correlated poorly with the C: N ratio unless N mineralisation was expressed as a proportion of total N (Nm:Nt ratio).

In a study of a wide range of Australian forest soils, Connell *et al.* (1995) also observed soil C, N, P contents, their ratios (C: N, C: P, N: P) and soil texture were poorly correlated with N mineralisation. However, correlations between total soil N and NNM are improved when sites were grouped by primary profile form (Connell *et al.*, 1995) or into strongly and weakly nitrifying soils (Carlyle *et al.*, 1990). In their historical overview of plant litter qualities and decomposition, Heal *et al.* (1997) noted that although the "C/N ratio is accepted as a general index of quality, the relative importance of the different chemical and physical components in different resources and their interaction is a matter of considerable debate." In soils, Adams and Attiwill (1983) noted that in forests were the pools of C and N in soils are large the C: N ratio of the acts as a strong buffer to perturbation of mineralisation patterns.

The presence of other substances, such as lignin and tannins, may also inhibit the activity of soil micro-organisms. In a review by Scott and Binkley (1997), lignin:N ratios in leaf litter explained more of the variation in NNM for the forest ecosystems than any other litter quantity or quality parameters. Lignin is the most resistant component of plant residues entering the soil and is the third most abundant component of plant residues after cellulose and hemicellulose (Cresser *et al.*, 1993). The lignin fraction of the decaying plant material becomes increasingly important as decomposition proceeds (Melillo *et al.*, 1989). Decay-resistant materials such as lignin and possibly lignin-N complexes may be the principle source of N in older litter and organic matter layers, and form important feedback mechanisms in N mineralisation (Pastor *et al.*, 1987). Higher lignin concentrations generally retard decomposition and N release, with a critical lignin:N ratio level given as 50 and above where N availability was substantially reduced (Van Cleve *et al.*, 1986). Inverse relationships have been observed between foliage lignin and NNM and leaf litter fall lignin: N ratio (Pastor *et al.*, 1984; Gower and Son, 1992; Prescott and Preston, 1994).

In contrast, Prescott *et al.* (2000) found no correlation between NNM and percentage lignin or lignin:N ratios in the litter layers. In their study, Prescott and Preston (1994) suggested alkyl C content, rather than lignin per se, may be the component more resistant to decomposition, and a better predictor of decomposition rates. DeMontigny *et al.* (1993) observed similar trends. High tannin levels are also often associated with low N mineralisation in litter (Gallardo and Merino, 1992; DeMontigny *et al.*, 1993). Tannins reduce the biodegradability and humification of organic matter by producing protein-tannin complexes. Overall the complex interactions between species and nutrient availability on decomposition and N supply was aptly summarised by Fisher and Binkley (2000) " General correlations between decomposition and litter N concentration can lead to mistaken inferences about N supply on N decomposition."

2.3.3 Seasonal Temperature Effects on N mineralisation

Nitrogen mineralisation, as a result of microbial utilisation of organic N, intimately depends on the environmental influences on the micro-organisms involved. These include temperature, moisture, substrate quantity and quality (Fyles *et al.*, 1990). Pools of mineralisable N in forest soil often show significant temporal variation (Ellis, 1974; Nadelhoffer *et al.*, 1982; Nadelhoffer and Aber, 1984; Richards *et al.*, 1985). Microbes are influenced by the physical and environmental conditions, and the pattern of mineralisation of organic N subsequently reflects seasonal changes in microbial populations (Theodorou and Bowen, 1981). The intricacy of this pattern is complicated by the wide variety of micro-organisms involved in the process as a whole.

General trends of N mineralisation in temperate regions show increases during the period of late spring and summer as temperature in the forest floor increased, and a subsequent decrease with decreasing temperatures. The maximum rates of N mineralisation reached in summer are often restricted by moisture limitations (Nadelhoffer *et al.*, 1983; Richards *et al.*, 1985; Adams and Attiwill, 1986; Plymale *et al.*, 1987; Foster, 1989). In a summer rainfall climate, the wetter and warmer months favour mineralisation, leading to an increase in the field concentrations of inorganic N throughout the spring and summer. Mineral N concentrations then decline to a

minimum in mid to late winter as the environment becomes progressively drier and colder (Richards *et al.*, 1985). Seasonal trends of NNM are experienced across a range of forest and soil types concurrently (Nadelhoffer and Aber, 1984; Richards *et al.*, 1985; Plymale *et al.*, 1987; Boone, 1992; Gower and Son, 1992) to varying degrees (Adams and Attiwill, 1986; Adams *et al.*, 1989b; Birk, 1992). Nadelhoffer *et al.* (1984) observed pronounced seasonal variations from nine temperate forest ecosystems in Wisconsin. However, seasonal trends where also influenced by secondary peaks in NNM due to leaf fall at deciduous sites. Seasonal *in situ* mineralisation rates were found to be twice that for stands of *E. regnans* aged 80 years than stand ages ranging between 5, 46 and 250 years, which had similar N mineralisation rates (Polglase *et al.*, 1992a). While Birk (1992) observed seasonal fluctuations in mineral N that were more pronounced in the younger stands of *P. radiata* (2 or 4 years old) than older stands (15 years old).

Seasonal ranges for N mineralisation can be dramatic. For example, in a field study of first and second rotation *P. radiata* soils the total mineral N content were three to ten times greater at the end of summer and autumn, than in mid winter (Theodorou and Bowen, 1983a). Furthermore, Vitousek and Matson (1985) showed a nearly ten-fold seasonal range for NNM in mineral soil under clear-cut loblolly pines. Soil temperature and moisture were the primary controls of these NNM seasonal trends. As the warm soil dried later in summer, NNM rates declined. Boone (1992) observed a seasonal change in N mineralisation potential in a mor soil under *Pinus strobus* L. of nearly two-fold for mineral soil (0-15 cm), nearly three-fold for the Oa (4-5 cm forest floor depth) horizon and more than four-fold for the Oe (1-4cm forest floor depth) horizon.

The seasonal trends in N availability and NNM can also vary independently. Although pools of mineral N and P varied with season across forest soils in north-eastern Tasmania reaching a maximum in autumn, no such marked seasonal variation was observed in the rates of NNM (Adams *et al.*, 1989b). Prasolova *et al.* (2000) also observed in soil under hoop pine that although total N and mineral N of the soil did

not differ between a wet and dry seasons, potential mineralisable N displayed different patterns of spatial variability between the two seasons.

Low temperatures often result in no or little net accumulation of inorganic N in soil (Nadelhoffer *et al.*, 1983; Foster, 1989). Seasonal low temperatures impact on the whole N cycle dynamics for a given environment, reduce the rate of organic matter decomposition, thereby reducing N mineralisation (Yin, 1992). Below 4 °C, Foster (1989) found no net accumulation of inorganic nitrogen. Concurrently, low temperatures often occur with reduced plant uptake of N through reducing root growth, soil root transport, and nutrient absorption rate. This reduction in overall plant development can therefore produce high gross N mineralisation in the field and therefore accumulated mineral N (Van Cleve *et al.*, 1981; Yin, 1992). In addition to temporal changes, the ranges of temperatures in which observations are taken also need to be quantified when measuring N availability and NNM. Studies in arctic soils found N mineralisation was more sensitive to changes in temperature above 10 °C than below (Usman *et al.*, 2000). This is consistent with observed enzymatic activity in these temperature ranges. Nadelhoffer *et al.* (1991) also observed that N mineralisation rates and soil respiration were insensitive to temperatures between 3 and 9 °C, but increased by a factor of two or more, between 9 and 15 °C. In a review of research on a range of studies, Kirschbaum (1995) concluded that temperature sensitivity of soil processes (as expressed by the Q_{10} function) was not constant across a range of temperatures, but was far greater at low (<10 °C) than at moderate to high (20-30 °C and above) temperatures.

The period of seasonal change is also considered to influence the rates of N mineralisation. Foster (1989) using *in situ* buried bags in litter horizons of Maple-Birch in central Ontario found NNM appeared to be particularly sensitive to temporal changes in average daily temperatures in the field. A late-summer peak in litter mean daily temperatures triggering a shift from NNM to NN immobilisation as the temperature began to decline. In contrast, Ellis (1974) noted that the direction of seasonal change was not effected by-short-term changes in soil temperature and moisture.

2.3.4 Moisture Effects on N Mineralisation

Significant interactions between temperature, moisture and NNM rate are observed through seasonality in the field and laboratory incubations (Cassman and Munns, 1980; Theodorou and Bowen, 1983a; Zak *et al.*, 1999). In a study predicting litter decomposition under eucalypts and pines using CAMFor (Carbon accounting model, under forests) and GENDEC, Paul and Polglase (2004a) observed that more accurate predictions of litter decomposition occurred when climatic data, temperature and rainfall, were taken into account than when actual values of lignin, cellulose and soluble C content were used. Interactions between moisture availability and temperate are an important factor in the understanding of the overall processes involved in N mineralisation. Clearly, temperature influences physiological activity and, consequently, the demand for substrate. Microbial activity can be limited by diffusion at warmer soil temperatures where high rates of physiological activity create a large demand for substrate. Where as, substrate diffusion is less likely to be limiting at lower temperatures due to the reduced physiological demand. Ellis (1974) observed that in any one stand, the rates of respiration were highly dependent upon the temperature of the soil at the time of sampling, but were considered unrelated to the moisture content, whereas patterns of N mineralisation followed variation in soil moisture content, dominant to temperature. Furthermore, Adams and Attiwill (1986) measured the highest mineralisation rates during summer (February) where both higher temperature and moisture levels were present, while rates of other seasons were generally similar.

Moisture conditions are a major factor controlling survival and activity of micro-organisms in the forest ecosystem. Micro-organisms depend on water for their physiological functions and the activity is effected by supply of dissolved nutrients, dissolved oxygen and exoenzymes (Griffin, 1981). Consequently, a positive correlation is generally exhibited between microbial activity and soil water potential between air-dry and field capacity. A strong relationship is also observed between the soil and litter horizon's water potential and N mineralisation (Powers, 1990; Evans *et al.*, 1998; Prasolova *et al.*, 2000) and nitrification (Tietema *et al.*, 1992). Nitrogen mineralisation, nitrification and respiration have been shown to increase linearly with gravimetric moisture content to a maximum content dependent on local field capacity

value (Tietema *et al.*, 1992). Strong *et al.* (1998b) indicated that water status strongly limits N mineralisation even at the high water potential of -30 to –10 kPa. In a later study Strong *et al.* (1999b) related such changes in NNM due to moisture content to the availability of organic N in soil pores.

In a study of birch (*Betula cordifolia* Regel) and fir (*Abies balsamea* (L.) Mill) forests, Evans *et al.* (1998) observed that patches of litter layers dominated by different species responded differently to annual changes in soil moisture or variables associated with wet and dry years. During an unusually dry year in their study, they found no correlation between NNM and nitrification with soil chemistry (N %, C %, C: N ratio) or abiotic variables (moisture, temperature, pH) in either birch or fir dominated plots. However, in the next wetter year the birch plots exhibited significant positive correlations with N % for NNM and nitrification and a significant correlation between moisture and nitrification. In this wetter year fir plots also showed a positive correlation between moisture and NNM.

Examination of climosequence soils and varying rainfall zones have shown that soil biota response to changes in moisture contents, such as drying and re-wetting are associated with inherent development properties (Birch, 1958; West *et al.*, 1988; Van Gestel *et al.*, 1991; Van Gestel *et al.*, 1993). Changes in NNM rates due to drying, are influenced by the soil biota's pre-adaptation to water fluctuations (West *et al.*, 1988), rainfall zones (Paul *et al.*, 1999), climatic history (Lund and Goksoyr, 1980), previous organic matter depletion by field drying (Degens and Sparling, 1995), previous soil aggregation state (Van Gestel *et al.*, 1991) and soil clay content (Cabrera and Kissel, 1988a; Strong *et al.*, 1999a). In laboratory studies, it is well recognised that environmental conditions most be controlled to obtain reproducible estimates of NNM (MacKay and Carefoot, 1981). Rates of aerobic mineralisation were generally much less for soils incubated *in situ* than for soils incubated in the laboratory (Adams and Attiwill, 1986). A dramatic change in moisture contents prior to laboratory incubation changes the availability of decomposable organic matter through microbial death and physical disruptions. This increases microbial biomass and activity and subsequently mineralisation of C and N (Stevenson, 1956; Birch, 1958; Van Gestel *et al.*, 1993).

The content of clay and organic matter within the soil determines the relative disruption of the physical structure of the soil and subsequently the rate of newly available organic matter release (Cabrera and Kissel, 1988a; Van Gestel *et al.*, 1991; Strong *et al.*, 1999a). In addition, soil C content generally increases with clay content as a result of the physical protection of organic matter within the clay matrix (Anderson, 1992). In a study of particle-size fractions in grassland soils Tiessen *et al.* (1983) observed that the topsoil fine silt fractions had the highest C content of the organo-mineral fractions and that the C: N ratios decreased with decreasing particle size from approximately 15 in sand to around 7 in fine clays fractions. In addition, the highest N contents were generally found in the coarse clay fractions. Their study indicated that particle-size fractionation yielded soil organic matter fractions with distinctly different properties that undergo different transformations during organic matter turnover.

2.4 Fertiliser Effects on N Mineralisation

The literature on the effects of fertilisation on NNM rates is conflicting. Inconsistencies are often a reflection of the effects due to variable nutrient status in the soil studied, the types of fertiliser used and the period during which they are applied (Aggangan *et al.*, 1998). However, increase in the rates of N mineralisation following N fertilisation to forest soils have been observed in many field and laboratory studies (Johnson *et al.*, 1980; Adams and Attiwill, 1991; Whynot and Weetman, 1991; Aggangan *et al.*, 1998).

Due to the large reservoirs of organic N present in the soil and litter pools, fertilisation with N at normal rates (eg. between 100 and 300 kg per hectare) contributes little to current total N pool (Morrison and Foster, 1977). Conversely, the same levels of fertiliser N can contribute substantially to the soil available N pools (Johnson *et al.*, 1980), and result in growth response in plantations (Waring, 1969; Cromer *et al.*, 1975; Miller, 1981; Cromer *et al.*, 1993; Neilsen and Lynch, 1998).

The proportion of nitrification may also increase due to N fertilisation, to the extent that nitrate N may become the newly dominant form of inorganic N (Adams and Attiwill, 1983). Often, the increased mineral N concentrations are relatively short lived, being rapidly transformed into less available forms in the microbial population (Williams, 1972; Johnson *et al.*, 1980; Khanna *et al.*, 1992; Fife and Nambiar, 1997; Smethurst *et al.*, 2001), dependent on the form and frequency of fertilisation (Heilman, 1974; Strader and Binkley, 1989). Enhancement of N concentrations in soil beyond 12 months occurred only at high rates of N fertilisation (600 and 1000 kg of N ha^{-1}) (Miller, 1981; Fife and Nambiar, 1997).

Ammonium is the dominant form of N taken up by eucalypts (Adams and Attiwill, 1986). Under the conditions that prevail in many forest soils, including low pH and intense microbial competition for inorganic N, the conversion of ammonium to nitrate by nitrifying organisms is low and in temperate soils and nitrate concentrations are often low or insignificant (Dyck *et al.*, 1983; Carlyle, 1986). Studies using *in situ* soil incubations in undisturbed soils generally have little nitrate initially and little nitrification during one to two months of incubation (Raison *et al.*, 1987; Carlyle *et al.*, 1990; Connell *et al.*, 1995). In undisturbed forests, the ammonium: nitrate ratio is in the order of 10:1 (Cole and Rapp, 1981). In a study of 27 soils in south-eastern Australia, Connell *et al.* (1995) observed that only soil that had experienced some disturbance (fertilisation, irrigation, ploughing, or burning etc) showed significant nitrification during incubations. Distribution of nitrate in soil profiles usually follows the water regime, rather than specific soil characteristics, such as clay content or particle size (Stevenson, 1982). However, significant anion exchange or other sorption sites will effect the mobility of nitrate in some subsoils (eg in Red Ferrosols, Doyle pers. comm.).

Although no growth response was observed in Adams and Attiwill's (1983) study of nitrification, NNM (as measured by aerobic laboratory incubations) and foliar N content all increased within 12 months of fertilisation. However, inorganic N and patterns of mineralisation approached those of the control plots 30 months after application. Similar increases in soil mineral N concentrations were observed by

Johnson *et al.* (1980) after applying urea (200 kg N ha^{-1}) to a 25-year-old loblolly pine stand (*Pinus taeda*). Twenty days after fertilisation, mineral N (predominantly ammonium) increased in soil to 200 μg g^{-1} and by 161 days they declined to unfertilised concentrations of less than 10 μg g^{-1}. In contrast, McLaughlin *et al.* (2000) observed N applied as ammonium nitrate at 100 kg N ha^{-1} resulted in a significant decrease in NNM and an overall inhibition of organic matter decomposition during the entire growing season. However, long-term changes in NNM have been measured. For example, six years after the application of N fertiliser at 860 kg N ha^{-1} (applied during a period of 7 years) Smolander *et al.* (1998) observed higher rates of NNM (aerobic incubations) compared to unfertilised soil.

In addition to the rate of fertilisation, the type of N fertiliser also influences the amount and patterns of N retention in soil and litter horizons. For example, Williams (1972) compared changes in humus N concentrations in sand dunes after application of ammonium nitrate, ammonium sulphate, urea or sodium nitrate at a rate of 250 kg N ha^{-1} on Scots pine (*Pinus sylvestris* L.). Application of all forms of N significantly increased N availability in the humus layer from 1.4 to 2.0 percent within two months of the final fertiliser application. However, only N applied as urea application showed any significant long-term retention in humus (at 1.52 % after 2 years).

Contrasting affects of fertilisation on microbial biomass, respiration and subsequent organic matter decomposition have also been observed. Decomposition can be inhibited by N decomposition (Berg and Tamm, 1991). Generally, N applied as urea stimulates microbial activity and decomposition, while inorganic forms of N may suppress such activity. For example, Strader and Binkley (1989) observed under Douglas fir stands, N fertilisation as urea increased soil respiration, microbial population numbers, and dehydrogenase activity during the first growing season more than ammonium nitrate. However, the opposite occurred for NNM and neither fertiliser effected N immobilisation. In contrast, Raison *et al.* (1990) observed ammonium nitrate application (400 kg N ha^{-1}) significantly increased soil N immobilisation (128 kg N ha^{-1} 0-40 cm depth) after one year, while, Smethurst *et al.*

(1998) observed no microbial response (estimated by the substrate-induced respiration) to N and P fertilisation in two year old eucalypt plantations.

The role of previous land use is also important when examining the effects of N fertiliser on N mineralisation and organic matter decomposition (Aggangan *et al.*, 1998). Previous land use has a significant influence on NNM (measured in laboratory incubations) decreasing in order of land use with ex-pasture > ex-native forest > native forest (Aggangan *et al.*, 1998). Higher net mineralisation rates were observed in ex-native forest and ex-pasture sites fertilised with N, indicating a faster turnover of organically bound N in response to N application. In contrast, the total amount of potentially mineralisable N declined with application of N, P alone and in combination in the ex-native forest. Site and land use factors also determine the overall ability for trees to compete for nutrients. In *P. radiata* plantations, Smethurst and Nambiar (1995) found trees were unable to take up N mineralised during weed senescence due to previous limited root development.

As discussed previously, application of N fertiliser significantly increases the amount of litter and N concentrations in litter returned to the forest floor. Applying NPK fertiliser (806, 178 and 366 kg ha^{-1}, respectively) during a three-year period to a 12-year-old *P. radiata* plantation increased both litter amount and litter N content (Theodorou, 1990). Fertilisation increased N return in each seasons' litter fall approximately two-fold and over 100 percent annually. However, during a three-year period, N content, immobilisation and release from decomposing litters and decomposition rates, where similar to those unfertilised.

Continued enhancement of growth due to N fertilisation for more than 5 to 10 years depends on repeated applications. This is illustrated by the inability of increased availability of N, alone, to increase rates of decomposition (Theodorou and Bowen, 1990; Prescott *et al.*, 1993), and evidence of often low rates of N mineralisation (Cole, 1995).

Like N fertilisation, P fertilisation impacts on N dynamics are variable and depend on fertiliser type, combinations, application period, site history and site nutrient status, and particularly whether P is limited. An increase in N uptake by enhancing soil N mineralisation has been observed in a range of forest soils (Waring, 1969; Falkiner *et al.*, 1993). For example, application of superphosphate to *P. radiata* plantation increased soil mineral N concentrations by approximately 100 percent, while enhancing NNM for 800 days (Falkiner *et al.*, 1993). In dry sclerophyll eucalypt forest, P application (500 kg P ha^{-1}) increased NNM by 40 percent (Falkiner *et al.*, 1993). Khanna *et al.* (1992) also observed a significant increase in NNM when superphosphate was applied at 200 kg P ha^{-1} to a podzolic soil under ten-year-old *P. radiata*. In addition, P application (200 kg P ha^{-1}) in *P. radiata* resulted in a three-fold increase in N uptake (Falkiner *et al.*, 1993). Short-lived stimulation of mineral N production due to P application was observed by Williams (1972). However, low rates of P fertilisation often have no significant effect of NNM rates. For example, application of 56 kg P ha^{-1} resulted in no significant effect on soil N dynamics using *in situ* buried bags (Javid and Fisher, 1990), and in a soil under young lodgepole pine stands, P fertilisation at the rate of 38 kg P ha^{-1} had no significant effect on NNM, N uptake, or N loss for the total growing season.

Therefore when P is not limiting due to site history such as ex-pastures (Aggangan *et al.*, 1998) or naturally high P content (Johnson *et al.*, 1980; McLaughlin *et al.*, 2000), addition of P fertiliser may have little effect on NNM. Net N mineralisation may actually be reduced after P application as a result of increased microbial activity (Adams and Attiwill, 1991; McLaughlin *et al.*, 2000). However when P is limiting, relatively long-term (beyond three years) increases in NNM can occur (Falkiner *et al.*, 1993).

2.5 Tools for predicting fertiliser responses- process based models

To achieve the required economic return on investment from plantations forest managers need reliable estimates of the long-term effects of silvicultural management practices. It is well recognised that economic plantation growth levels cannot be sustained from native soil N and that large fertiliser programs are required. Often

assessing the site quality of a forest ecosystem in terms of inherent physical, chemical and biological indices does not adequately take into account many soil-plant interactions or the role of nutrient cycling in the forest ecosystem (Schoenholtz *et al.*, 2001), e.g. gradual changes in nutrient pools as the plantation develops. Generally, nutrient requirements by plantation trees are greatest prior to canopy closure and decline significantly in the later stages of stand development when nutrient uptake is primarily driven by wood increment (Cole and Rapp, 1981; Miller, 1981).

Models can summarise the results of many experiments by incorporating hypotheses and conclusions into a quantitative framework. Empirical based models, which use equations to which observed data from many sites have been fitted, do not generally respond to climatic changes and are not suited to simulating management changes such as fertiliser application. One tool increasingly used to inform silvicultural practices, is process-based modelling. Model outcomes can be used to estimate growth responses and therefore economic benefits of silvicultural practices on a rotational length. In addition, they allow the assessment of risk involved both in terms of changing climate affects on economics (i.e. drought) and possible offsite movement of fertilisers.

There are a number of models that have been developed to predict forest productivity including; FOREST BGC (Running and Coughlan, 1988), BIOMASS (McMurtie *et al.*, 1990; McMurtrie and Landsberg, 1992), CENW (Kirschbaum, 1999), PnET (Aber and Federer, 1992), G'DAY (Comins and McMurtrie, 1992), TREGROW (Weinstein *et al.*, 1991) PROMOD (Battaglia and Sands, 1997), 3-PG (Landsberg and Waring, 1997) and CABALA (Battaglia *et al.*, 2004). Each model was developed with different objectives; therefore, different processes are emphasised at different levels of detail.

BIOMASS is a process-based model of *P. radiata* growth incorporating sub models for radiation absorption, canopy photosynthesis, partitioning of assimilate between plant organs, litter fall and stand water balance (McMurtie *et al.*, 1990; McMurtrie and Landsberg, 1992). The model uses a daily climatic data to obtain daily total photosynthesis and transpiration in *P. radiata*. BIOMASS is calibrated against the BFG experiment, which included treatments of irrigation and fertiliser on a *P. radiata*

plantation in Canberra. However BIOMASS was limited by the fact that it does not simulate decomposition and N mineralisation but uses the foliage nutrient concentration as an input to simulate these processes (Ryan *et al.*, 2000). CENW was validated against the BFG experiment and is a comprehensive forest growth model that links the flow of C, energy, nutrients and water in trees and soil organic matter (Kirschbaum, 1999). The model runs on a daily time step, calculating allocation for both fractions of C and N to different plant organs daily. Decomposition is determined by temperature, soil water status and soil organic matter quality based on CENTURY model (Parton *et al.*, 1987). Nitrogen mineralisation, of the active organic matter pool is based on C: N ratios of the organic matter received from the litter. The model was able to successfully simulate dynamics of C, energy, N and water across stands and five treatments over 4 to 5 years. However, the model was unsuccessful in modelling the higher foliar N content (dependent on foliar biomass growth, senescence and N mineralisation and uptake) measured in unfertilised trees in the BFG experiment.

PROMOD predicts growth of forest following canopy closure (Battaglia and Sands, 1997). The model uses simple soil and climate data (monthly or daily time steps) at a site and predicts the closed canopy LAI of a stand, estimates the annual NNP and stand water use of the stand. PROMOD is limited by the fact that it assumes a stand in steady status growth with closed canopy and roots fully occupying the soil volume and does not predict biomass partitioning. Neither does it predict what happens if the stand has a partially closed canopy. This limits its use for silvicultural management prediction such as thinning and pruning. 3-PG is a dynamic process-based model of forest growth that runs on a monthly time step using monthly climatic data (Landsberg and Waring, 1997). 3-PG predicts the time-course of stand development, biomass pools, stand water use and available soil water. The model allows the selection of site factors to be age dependent which simulates silvicultural intervention and aids the study of consequences to changing site conditions such as irrigation and fertilisation or run down of site conditions. However, 3PG had a poor prediction of canopy development and mortality limited has soil and nutrition data. Both PROMOD and 3-PG do not consider the decomposition processes of plant litter.

CABALA is a carbon balance model of plantation growth designed for silvicultural decision support managers (Battaglia *et al.*, 2004). It predicts the time-course of stand

development, water use and available soil water for trees and responds to silvicultural interventions such as thinning and fertilisation. Daily canopy photosynthetic production is calculated using coupled C-water-N models of photosynthetic rate and model of LAI and light interception by tree crowns. Soil N turnover can be calculated using a simple mineralisation model which is a derivation of SNAP, (Paul *et al.*, 2002) or by defining soil C and N pools at the start of the simulation and using process-based N mineralisation model CERES-N, (Goodwin and Jones, 1991). Its primary stand-level outputs are biomass to various pools, available soil water in the tree root zone, tree predawn water potential and a detailed breakdown of site water balance, and the distribution of N in trees and the soil. With N mineralisation tuning, CABALA successfully simulated some aspects of N dynamics in a *E. nitens* plantation in Tasmania (Smethurst *et al.*, 2004a), but predictions beyond 2.8 years were biased mostly by parameters for allocation and plant N concentration that were suited only to much younger plantations.

Models are developed with various objectives and subsequently place different emphasised and different levels of detail on components of any given forest. Models such as CABALA and CENW can be used to compare the long-term effects on N fertilisation on growth and N cycling, as both link the flow of C, N and water on a daily time step. However, both models use N submodels that are based on agricultural soil processes. This is particularly evident in the CENTURY decomposition model used in CENW, which was developed under grazing grasslands.

2.6 Conclusion

Although N is abundant in the atmosphere only a relatively small component occurs in forest soil and less than three percent of this is in the mineral form available for tree growth (Cole, 1995). As most forest trees prefer to take up N in the ammonium form with limited organic N uptake and N fixation, nitrogen mineralisation is an essential component of the N cycle required for plant growth. The process of N mineralisation (NH_4 release) occurs through microbial utilisation of organically bound N forms, as an energy source. As this is a microbially mediated process, it depends on environmental effects such as climate temperature and moisture, and quantity and

quality of organic N. (Fyles *et al.*, 1990). Of N mineralised, 0.5 to 0.01 percent will be released as ammonium and the remainder immobilised by the microbial community (Binkley and Hart, 1989). As N mineralisation is an integral step in the availability of N for plantation growth it has been widely studied as an indicator of site productivity, with productivity in many forest ecosystems increasing with increasing soil N mineralisation (Pastor *et al.*, 1984; Adams and Attiwill, 1986).

An increase in N availability in the short-term also occurs due to application of N fertiliser (Johnson *et al.*, 1980; Adams and Attiwill, 1991; Aggangan *et al.*, 1998). In contrast, limited increases in N mineralisation in the long-term have been measured (Smolander *et al.*, 1998). The difficulty in assessing the long-term effects of N mineralisation may be due to the insensitivity of NNM measurements to low rates of fertilisation. Although there are clear influences of plant species, temperature, and moisture and soil fertility on N mineralisation, the isolation of these effects for interpretation is often less defined. Each factor is often confounded with the site being studied. In addition, the process of *in situ* and laboratory incubations to determine NNM could cause a change in one or more of these factors.

Chapter 3. Growth response of *Eucalyptus regnans* and *Pinus radiata* due to long-term periodic fertilisation and changes in Ferrosol and Kurosol profiles.

3.1 Introduction

In Tasmania the increased demands for wood production on a base of decreasing land availability has resulted in the progression of intensive managed forest into areas with soil of reasonable physical structure but low nutrient status. This trend is combined with increasing demands for faster growth and greater product utilisation from forest sites, intensifying nutrient demand and removal. Prescription N and P fertilisation at planting, and in early stages of tree establishment are required at many sites to achieve rapid early growth and high survival rates (Gentle *et al.*, 1965; Waring, 1972; Judd *et al.*, 1996).

Later age application of P fertiliser on P-deficient sites can increases *P. radiata* plantation growth (Waring, 1969; Flinn *et al.*, 1979b; Neilsen *et al.*, 1984). The magnitude and longevity of response to P application depends on soil properties such as the inherent P concentrations, P sorption capacity and soil pH (Ballard, 1978; Pritchett and Comerford, 1982). Phosphorus application may also increase soil N uptake in plantations (Waring, 1969; Neilsen *et al.*, 1984; Falkiner *et al.*, 1993).

In contrast to P, enhanced growth response due to N application may decline within a few years (Fagerstrom and Lohm, 1977; Miller, 1981; McIntosh, 1982; Fisher *et al.*, 2000). Such declines are related to short-term fluxes in mineral N after fertilisation (Hingston and Jones, 1985; Adams and Attiwill, 1991; Smethurst *et al.*, 2001) and then corresponding decreases in foliar N concentrations (Fagerstrom and Lohm, 1977). Subsequently, to maintain diverging growth from untreated plantations, repeated applications of N fertiliser are required (Neilsen *et al.*, 1992; Neilsen and Lynch, 1998). The frequency of applications will depend on the period of elevated availability, cycling of applied nutrients in the soil system and the efficiency of internal tree recycling of nutrients (Switzer and Nelson, 1972). Falling needle concentrations of N, and declining growth, indicate that applications at between two

and four years are required to maintain growth of *P. radiata* plantations on nutrient poor sites in Tasmania (Neilsen and Lynch, 1998).

Following canopy development, internal redistribution and nutrient return from decomposition become critical processes in supplying nutrients for new growth (Miller, 1981; Weston, 2001). The litter layer is important in the cycling and retention and supply of nutrients in forests (Tamm and Popovic, 1995; Neilsen and Lynch, 1998). Variations in overall mass and nutrient concentration of litter depend on the site, species, stand age, fertiliser treatment, and time of measurement (Feller, 1978; Frederick *et al.*, 1985; Baker *et al.*, 1986; Crane and Banks, 1992; Canary *et al.*, 2000). At sites with low nutrient concentration in the mineral soil, litter quality forms a strong correlation with tree volume growth (Smith *et al.*, 2000) and N mineralisation (Adams and Attiwill, 1986; Gower and Son, 1992; Prescott *et al.*, 1993; Prescott and Preston, 1994).

Although fertilisers may be used to increase forest growth there is concern that long-term N application may impact on forest sustainability through changes in the soil chemistry. Nitrogen fertilisation has been identified as a source of increased acidity in soil chemistry across a range of sites (Adams and Martin, 1984; Tamm and Popovic, 1995; Homann *et al.*, 2001). Low soil pH decreases the rate at which organic N is mineralised (Adams and Martin, 1984), reduces the availability of cations such as Ca and Mg, and could increase the availability of Al to toxic concentrations. Factors including, fertiliser form and rate, the site climate and plant species determine the extent to which the soil pH changes. Soil properties such as, the concentration and nature of organic matter, type and amount of clay, the initial soil pH and the soil buffering capacity also influence the rate of pH change (Adams and Martin, 1984).

The objectives of this experiment were to examine two sites of contrasting soils and tree species and;

1) examine options for improving the growth of both *P. radiata* and *E. regnans* plantations through various periodic applications of late age fertilisation,

2) examine the effect of fertilisation on nutrient distribution in soil profiles,

3) investigate fertiliser effects on forest litter mass and nutrient retention, for long-term nutrient cycling and site management, and

4) evaluate periodic sampling of foliar nutrient concentrations, in both plantations, as an indicator of future growth response.

3.2 Site and Soil Profile Description

Two field experiments, one in *P. radiata* (Latitude 41° 28' S, Longitude 148° 00' E) and one in *E. regnans* (Latitude 43° 17' S, Longitude 146° 54' E), were established to critically evaluate later age fertilising with nitrogen, applied periodically, and in combination with phosphorus.

The Kurosol studied under *P. radiata* in this research is formed on siliceous sediments, which represent a substantial proportion of the *P. radiata* estate in Australia and other *P. radiata* growing areas. Sediments of Devonian-Silurian age comprise the largest proportion of the Tasmanian estate plus extensive areas in Eastern Victoria and Southern New South Wales. The soil profile is also very similar to the soil researched intensively in the ACT (Khanna *et al.*, 1992) the major difference being that the Tasmanian site is in a cooler environment.

The Ferrosol under *E. regnans* is a brown soil formed on basic igneous parent material. Although dolerite is not common outside of Tasmania, the soil is used extensively for eucalypt plantations in Tasmania and is similar to soils developed on basalt used extensively in cool temperate climatic zones where eucalypt plantations are grown.

3.2.1 Kurosol planted with *P. radiata*

The topography of the experimental site was an easterly aspect of about 10 % slope and an altitude of 350 m. The 24 year average annual rainfall was 938 mm spread fairly evenly through the year, with a winter bias. However, annual rainfall was

highly variable. The vegetation on the site before conversion to *P. radiata* plantation was mature *Eucalyptus sieberi* (L.A.S. Johnson) of 20 - 29 m height, with an understorey of moderately dense bracken (*Pteridium esculentum* (Forst.f.) (Photo 3.1).

Photo 3.1. Typical native vegetation of a *Eucalyptus sieberi* and *Pteridium esculentum* before conversion to *P. radiata*.

The profile described was classified as a haplic, mesotrophic, Yellow Kurosol with loamy sand and over silty clay (Isbell, 1996), and a Yellow Podzolic in the Greater Soils Group (Stace *et al.*, 1968). The soil is formed on metamorphosed shales and sandstone of Devonian-Silurian age (Mathinna beds). The likely US soil taxonomy class is Typic Haplohumult. Texture was predominantly silty clay and total soil depth was 0.6 - 0.8 m (Photo 3.2, Table 3.1).

Photo 3.2. Kurosol profile

These soils are widespread in the north-east of Tasmania where the annual rainfall is less than 1000 mm. They form on undulating, rolling steep to low hills, are moderately well drained and normally support dry sclerophyll forest often with *Eucalyptus sieberi* and *E. amygdalina* over an understorey of species such as *Dodonaea viscosa, Davesia latifolia, Pultenaea daphnoides* and *Epacris impressa*. The soils are texture contrast with loamy sand surface horizons over silty clay subsoils of strong to firm strength. Both total N and organic C are naturally low throughout the profile while the surface soil has total P at medium to low levels (Neilsen *et al.*, 2002). The status of total N and P and organic C is described in Grant *et al.* (1995) (Table 3.2).

Table 3.1a Kurosol profile description

HORIZON	DEPTH(cm)	DESCRIPTION
O1	-1.5 to -0.5	Loose litter, comprising pine needles and dead bracken
O2	-0.5 to 0	Decomposing duff layer derived from litter, abrupt boundary
A1	0-10	Dark brown (7.5YR3/2) loamy sand; weak strength, concentration of fine roots and common medium roots, diffuse boundary
A2e	10-20	Greyish brown (2.5Y5/2) fine sand to loamy sand; weak strength, few roots, abrupt boundary
B21t	20-44	Olive yellow (2.5Y6/6) silty clay; strongly developed 20-50mm angular blocky breaking to 5-10mm subangular blocky structure; moderately friable; 10-30% distinct clay skins; moderate strength, 20% rock and 10% gravel present; common medium roots and some concentration of fine roots, gradual transition
B22t	44-58	Olive yellow (2.5Y6/6) silty clay; weakly developed 5-10mm subangular blocky to compact structure; strong strength, 30% rock and 10% gravel present; common medium roots, gradual transition
B3t	58-63	Olive yellow (2.5Y6/6) silty clay; 2-10% <5mm distinct yellowish red (5YR5/8) oxidation/reduction mottles; compact structure; strong strength, 30% rock and 10% gravel present; few fine roots, clear boundary to C horizon
C	63+	Decomposing sandstone

(Neilsen *et al.*, 2002)

Table 3.1b. Kurosol profile description from soil pits for NIL and (P)N1Y treatments under *P. radiata*.

Treatment	Horizon	Sample Depth	Bulk Density	Water-stable Aggregates	Clay	P retention
		(cm)	(g cm^3)	(% > 0.25 mm)	(%)	
NIL	O2	-05 to 0	0.46			
	A1	0-10	1.54	21.3	4	5
	A2e	10-20	1.51	23.6	6	4
	B21t	20-44	1.67	44.3	49	36
	B22t	44-58	1.94	39.6	44	33
(P)N1Y	O2	-3 to 0	0.37			
	A1	0-3	1.71	10.0	4	2
	A2e	3-17	1.71	17.7	7	2
	B21t	17-50	1.9	38.7	33	29
	B22t	50-70	1.04	23.2	29	19

Table 3.1c. Kurosol chemical profile description from soil pits for NIL and (P)N1Y treatments under *P. radiata*.

Treatment	Horizon	Sample Depth	Exchangeable H^+	Exchangeable Al	Exchangeable Acidity	ECEC	BS (%)[1]
		(cm)		(me.100 g^{-1})			
NIL	O2	-05 to 0					
	A1	0-10	1.2	0.9	2.1	16.7	15.9
	A2e	10-20	0.0	1.9	1.9	9.2	6.7
	B21t	20-44	0.5	6.4	6.9	25.2	3.6
	B22t	44-58	0.7	4.2	4.9	21.1	5.0
(P)N1Y	O2	-3 to 0					
	A1	0-3	3.0	1.0	4.0	23.9	5.3
	A2e	3-17	1.1	1.0	2 1	13.4	2.5
	B21t	17-50	1.6	7.8	9.4	20.6	1.4
	B22t	50-70	0.6	4.2	4.7	15.6	1.7

BS = Base Saturation

Table 3.2 Ratings for chemical and physical laboratory analysis, after Grant *et al.* (1995).

Rating	Organic C	Total P	Total N	Water-stable aggregates
	(%)	(ppm)	(%)	(% >0.25 mm)
Very High	>10	-	-	-
High	5-10	>250	>0.6	>70
Medium	2-5	100-250	0.3-0.6	30-70
Low	<2	<100	<0.3	<30

3.2.2 Ferrosol planted with *E. regnans*

The topography of the experimental site was a south-easterly slope of about 10 % and an altitude of 100 m. The average annual rainfall was 1200 mm. Prior to plantation

establishment the site originally carried *Eucalyptus obliqua* of 34 to 41 m height and an understorey of moderately dense bracken with some *Cassinia aculeata* and *Acacia verticillata* (Photo 3.3).

Photo 3.3. Typical native vegetation of *Eucalyptus obliqua* and *Acacia verticillata* before conversion to an *E. regnans* plantation.

Photo 3.4. Ferrosol profile

The profile described was classified as a Haplic, Brown Ferrosol (Isbell, 1996), and a Krasnozem in the Greater Soils Group (Stace *et al.*, 1968). The soil is formed on Jurassic dolerite and Triassic sandstone. The likely US soil taxonomy class is a Hapludalt. The soil was a yellowish-brown to red clay loam soil total soil depth of over 1.0 m (Table 3.1, Photo 3.4).

Table 3.3a Ferrosol profile description

HORIZON	DEPTH(cm)	DESCRIPTION
O1	-4 to -1	Loose litter, comprising leaves and fine branches
O2	-1 to 0	Decomposing duff layer derived from litter, abrupt boundary
A1	0-12	Dark yellowish brown (10YR4/6 to 10YR3/4) clay loam to loam; friable sub-angular structure; weak strength; many medium and fine roots; variable boundary
B21t	12-60	Dark yellowish brown (10YR4/6) distinct strong brown (7.5YR 5/6) oxidation mottles; clay loam; sub-angular blocky structure with peds 2 – 10 cm; weak strength; many medium roots; gradual boundary
B22t	60-90	Dark yellowish brown (10YR4/6 to 10YR5/8) distinct strong brown (7.5YR5/6 to 7.5YR5/8) mottles; clay loam; compact structure ; firm strength; 20-30% sub-rounded 200-600mm dolerite fragments; common fine roots; gradual boundary
B3t	90+	Strong brown (7.5YR5/8) light clay; compact structure; firm strength; 20-50% subrounded 200-800mm dolerite fragments; few fine roots

(Neilsen *et al.*, 2002)

Table 3.3b. Ferrosol profile description from soil pits for NIL and (P)N1Y treatments under *E. regnans*.

Treatment	Horizon	Sample Depth	Bulk Density	Water-stable Aggregates	Clay	P retention
		(cm)	(g cm^3)	(% > 0.25 mm)	(%)	
NIL	O2	-1 to 0	0.67			
	A1	0-12	1.37	84.4	56	47
	B21t	12-60	1.47	88.0	63	57
	B22t	60-90	1.52	85.2	58	54
P1YN1Y	O2	-6 to 0	0.22			
	A1	0-10	1.15	68.9	42	37
	B21t	10-60	1.48	91.5	61	60
	B22t	60-100	1.45	82.1	54	54

Table 3.3c. Ferrosol profile description from soil pits for NIL and (P)N1Y treatments under *E. regnans*.

Treatment	Horizon	Sample Depth	Exchangeable H^+	Exchangeable Al	Exchangeable Acidity	ECEC	BS (%)[1]
		(cm)		(me.100 g^{-1})			
NIL	O2	-1 to 0					
	A1	0-12	1.0	3.6	4.6	32.1	22.7
	B21t	12-60	1.5	6.4	7.8	36.2	7.8
	B22t	60-90	1.4	6 9	8.3	34.3	6.1
P1YN1Y	O2	-6 to 0					
	A1	0-10	2.4	8 3	10.8	36.5	6.2
	B21t	10-60	2.9	16.1	19.0	34.7	3.7
	B22t	60-100	1.9	11.1	13.0	32.2	7.4

BS = Base saturation

These soils are widespread where annual rainfall exceeds 1000 mm. Formed on rolling hills they are well drained and support mainly wet sclerophyll forest dominated by *Eucalyptus obliqua* and ranging to mixed forest, with understorey species such as *Cassinia aculeata, Acacia verticillata, Pomaderris apetala* and *Gahnia grandis*. The soils are distinguished by gradational profiles with clay loams overlying light clays, which vary in colour from yellow brownish to brown. Surface soils generally have high organic C and total P with medium concentrations of total N. In the subsoil these nutrients can be of low to medium status (Table 3.2) (Grant *et al.*, 1995). This site had been logged and burnt in wild fires before plantation establishment and only about 10 cm of topsoil remained (Neilsen *et al.*, 2002).

3.3 Fertiliser experiment establishment

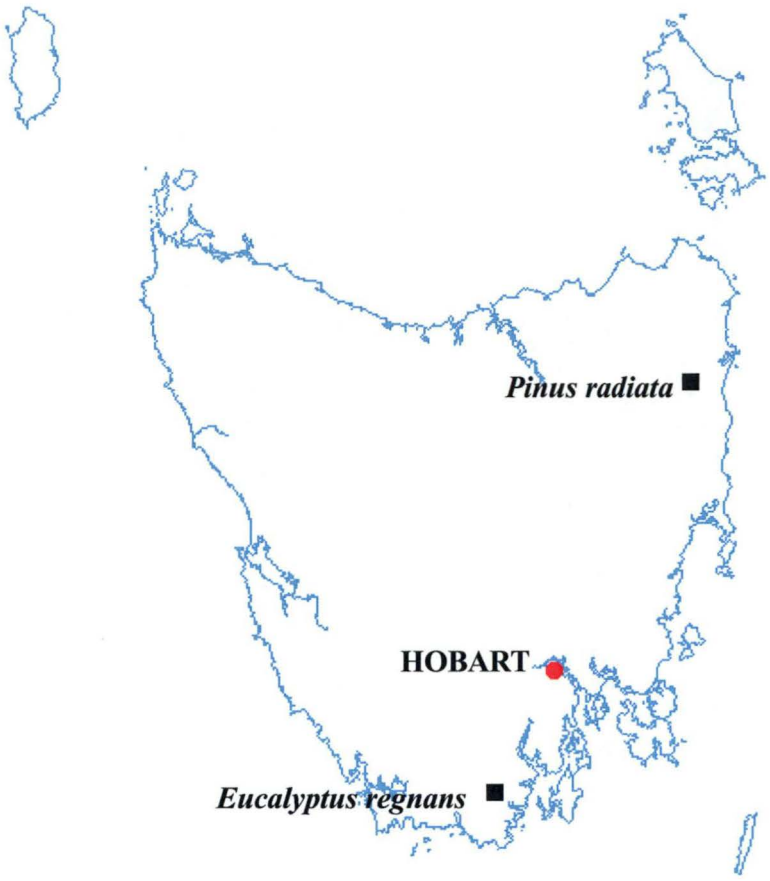
3.3.1 Kurosol planted with *P. radiata*

The fertiliser experiment was established in a 20-year-old *P. radiata* plantation, located in the north-east of Tasmania (Map 3.1). At plantation establishment (1967) seedlings were fertilised with 23 g N (urea) and 12.5 g P (superphosphate) per tree and stocking was 1500 stems ha^{-1} (3 m x 2.2 m spacing). The experiment area was thinned prior to establishment of the fertilisation experiment to approximately 58 % of initial stocking (from 23.6 $m^2 ha^{-1}$ to 18.3 m^2 ha^{-1} mean basal area (BA)). The thinning was mainly aimed at removing small diameter, poorly formed and forked trees. Prior to treatment the experimental site exhibited approximately 56 % dead tops (with no current apical growth). The *Pinus radiata* experiment was established in

an older stand to demonstrate the resilience of *P. radiata* to recover from nutrient limitations on growth. In practice fertilising commences much earlier than this.

The fertiliser experiment consisted of two replicates (blocks) of six treatments (Table 3.4). Twelve rectangular plots each had a total area of 668 m^2. The two blocks were determined on initial tree basal area (BA), with one block being the six highest BA plots (mean BA 20.3 to 20.5 m^2 ha^{-1}) and the other being the six lowest BA plots (mean BA 16.2 to 16.3 m^2 ha^{-1}). In each plot, a sub-plot of 25 trees (288 m^2) was selected for measurement, allowing for a buffer zone of at least two trees between different fertiliser treatments.

Phosphorus was applied at the establishment of the experiment (June 1987), and when need as indicated by foliar analysis, to maintain foliar concentrations close to 0.12%, the concentration considered adequate for good growth. Re-application was carried out six years after initial fertiliser application at age 26, with a total of 144 kg P ha^{-1} applied. The periodic N application experiment was designed with four levels of regular N applications; no N ((P)), N applied every fourth year ((P)N4Y), every second year ((P)N2Y), and annually ((P)N1Y). In addition, two other treatments were included, a treatment with neither N nor P applied (NIL), and a treatment of N plus P applied every two years (P2YN2Y). The rate of N required was determined using previous experiments, as 100 kg N ha^{-1} for each application (Neilsen and Lynch, 1998). Fertilisers used were single superphosphate, ammonium sulphate or mixtures of the two, resulting in large quantities of S and Ca also being applied. Fertilisers were broadcast by hand; rates of elemental N, P, S and Ca applied are given in Table 3.4.



Map 3.1 Location of experimental sites

Table 3.4 Treatments and frequency of application of fertilisers giving total quantities of N, P, S and Ca.

Code	Treatment	Rate (kg ha^{-1})	Frequency	N (kg ha^{-1})	P (kg ha^{-1})	S (kg ha^{-1})	Ca (kg ha^{-1})
NIL		Nil					
(P)	superphosphate	750	twice		144	164	330
(P)N4Y	superphosphate	750	twice	400	144	636	330
	ammonium sulphate	480	4-yearly				
(P)N2Y	superphosphate	750	twice	700	144	990	330
	ammonium sulphate	480	2-yearly				
(P)N1Y	superphosphate	750	twice	1300	144	1698	330
	ammonium sulphate	480	1-yearly				
P2YN2Y	11:5[†]	952	2-yearly	700	322	1190	330

[†] 11:5 (N: P) is a mixture of superphosphate and ammonium sulphate.

All 25 trees on the sub-plot were measured for diameter at breast height over bark (DBHOB) annually from establishment for 15 years. The two tallest trees on each whole plot were measured for height as MDH (mean dominant height), based on the

tallest 50 SPH (stems per hectare) evenly distributed trees over the area. Stand volume was calculated using volume tables (Wilkinson and Neilsen, 1995). A number of symptoms of tree health were assessed including, foliar colour, crown length, crown width, the presence of fused needle, and dead tops (Neilsen *et al.*, 1992).

Foliar sampling was carried out pre-fertilising at age 17, and at ages 22, 23 and 34 years. One and two year old age classes of foliage were sampled. Three selected trees per plot were sampled by climbing and sampling, or using a shotgun to collect twigs. Needles from the three trees were stripped from each branch; each age class was combined on the basis of equal mass and bulked separately for foliar analysis. Samples were prepared as described by Neilsen *et al.* (1992). Drying was carried out at 70°C before grinding in a Wiley mill prior to analysis.

My involvement in the fertiliser experiment was from tree age 27 onwards, prior to this Bill Neilsen, Wally Pataczek, Lindsay Wilson and Martin Piesse managed the experiment. Information prior to tree age 27 has been provided by Bill Neilsen as pers. com. and written communication.

3.3.2 Ferrosol planted with *E. regnans*

The fertiliser experiment was established in a 5-year-old *E. regnans* plantation, located in southern Tasmania (Map 3.1). At plantation establishment (1981) seedlings were fertilised with 25 g N (ammonium sulphate) and 11 g P (superphosphate) per tree and initial stocking of 1333 SPH (3 m x 2.5 m spacing). The fertiliser experiment consisted of two replicate (blocks) of six treatments (Table 3.5). The two blocks were determined on initial BA, with one block being the six highest tree BA plots and the other being the six lowest tree BA plots. Each of the twelve rectangular plots had a total area of 400 m^2. In each plot, a measured sub-plot of 25 trees was selected, allowing for a buffer zone of at least two trees between different fertiliser treatments. All trees on the measured sub-plot were measured for DBHOB annually from establishment for 15 years. A sample of tree heights was measured and volume was estimated using tree volume tables (Wilkinson and Neilsen, 1995).

Because of the lack of long-term data for eucalypt stands, the fertiliser experiment was designed for analysis as an N and P factorial, with applications at two yearly intervals, as well as a level experiment. The level experiment was designed with regular applications of N and P together with the amounts set by the period between applications. There were four levels of N plus P applied, no fertiliser (NIL), every fourth year (P4YN4Y), every second year (P2YN2Y), and annually (P1YN1Y). For the factorial experiment there were, in addition, applications every second year of N only (N2Y) and P only (P2Y). As in the *P. radiata* experiment, fertilisers used were superphosphate, ammonium sulphate or mixtures of the two. Fertilisers were broadcast by hand; rates of elemental N, P, S and Ca applied are given in Table 3.5.

Table 3.5 Treatments and frequency of application of fertilisers applied to the research area giving total quantities of N, P, S and Ca.

Code	Treatment	Rate (kg ha^{-1})	Frequency	N (kg ha^{-1})	P (kg ha^{-1})	S (kg ha^{-1})	Ca (kg ha^{-1})
NIL	Nil						
P4YN4Y	11:5†	952	4-yearly	400	184	680	420
P2YN2Y	11:5†	952	2-yearly	700	322	1190	735
P1YN1Y	11:5†	952	1-yearly	1300	598	2210	1365
N2Y	ammonium sulphate	480	2-yearly	700		826	
P2Y	superphosphate	476	2-yearly		322	364	735

† 11:5 (N: P) is a mixture of superphosphate and ammonium sulphate.

Foliar samples were collected prior to fertilisation treatments at ages 10 (just prior to the sixth annual fertilisation, 12, 14 and 20 years, three years after the final fertiliser treatment). Three selected trees per plot were sampled using a rifle or shotgun to collect branches or twigs from the upper third of the crowns. Leaves from the three trees were stripped from each branch and combined, on the basis of equal mass basis, for foliar analysis. Samples were prepared as described by Neilsen *et al.* (1992). Drying was carried out at 70°C before grinding in a Wiley mill prior to analysis.

My involvement in the fertiliser experiment was from tree age 16 onwards, prior to this Bill Neilsen, Wally Pataczek, Lindsay Wilson and Martin Piesse managed the experiment. Information prior to tree age 16 has been provided by Bill Neilsen as pers. com. and written communication.

3.4 Sampling and Analysis of Soil profiles and Litter Layers

The impact of fertilisation on forest soils was considered by examining differences in soil profiles between unfertilised and fertilised plots at tree age 34 and 20 in *P. radiata* and *E. regnans*, respectively. Pits were dug, to the depth of 1.0 m, in the centre of each unfertilised and annually fertilised plot and soil horizons were described and sampled for analysis (Photo 3.2 and 3.4). Physical and chemical parameters for each horizon were measured and analysed. Additionally, soils in all plots were sampled by soil-auger (5 cm diameter) to a depth of 50 cm, in 10 cm increments. Soil was collected from each site from the uncultivated zones between tree rows. Each 10 cm increment sample was bulked from each of four auger holes per plot. Resulting in two replicate soil samples for each treatment and depth. Soil used to determine mineral N concentrations were maintained moist and sieved to < 2 mm for chemical analysis. Remaining soils were air dried and sieved to < 2 mm for chemical analysis. Bulk density for soil was calculated from intact cores sampled from soil pits, in 10 cm increments, to a depth of 50 cm.

Litter (forest floor organic debris) was collected using a 25 by 20 cm frame. Litter was collected from each site from the uncultivated zones between tree rows. Litter samples were separated into O1 and O2 litter layers (McDonald *et al.* 1990) (also described in Section 3.2). Five O1 and O2 litter samples were collected from each plot and bulked separately for biomass determination. Only the O2 samples were analysed. O2 litter layers were air-dried and sieved to < 2 mm for chemical analysis. Total nutrient biomass in the litter (O2) and soil was estimated from these measurements.

Foliar and litter samples were digested by acid hydrogen peroxide (Lowther, 1980). The digest was analysed for N, based on the indophenol blue method (Lachat Instruments), P by molybdate blue method (Murphy and Riley, 1962) and Ca, K and Mg by atomic absorption spectrometry (AAS). For soil, total P and N were estimated by semi-micro-Kjeldahl digestion automated colour (Rayment and Higginson, 1992). Total Mg, Ca, K and micronutrients Cu, Fe, Zn and Mn in soil were analysed by AAS following nitric acid digest. Exchangeable Ca, Mg and K were extracted using 1 *M*

ammonium chloride at pH 7.0 (Rayment and Higginson, 1992). Organic C in soil was determined by the Walkley-Black method (Rayment and Higginson, 1992). Soil pH was measured in a 1/5 soil/ distilled water ratio using soil that had not been dried. Mineral N (ammonium plus nitrate) was extracted from samples (< 2 mm fraction), maintained field moist, using cold 2 *M* KCl, (Rayment and Higginson, 1992). Mineral N in KCl extracts was measured using a flow injection analyser (Lachat instruments).

3.5 Data analysis

Total nutrient biomass of *P. radiata* trees was estimated using an equation derived from Neilsen and Lynch (1988) (Table 3.6). Mean tree (based on volume) total content of N, P, Ca and Mg were regressed against mean tree volume and this was applied to the mean tree total volume for the plots in this experiment. Samples based on mean tree volume are unbiased but may have an error of up to 10 % (Crow, 1971).

Analysis of variance and least significant difference (LSD) tests were used to test the significance of treatment and soil profile depths on nutrient content, while group regression analysis was used to evaluate volume growth response (Genstat 5 Committee, 1988). Tests were validated by testing data for normality of distribution, and transforming data where required. Residuals from the model for each variable were examined for normality using diagnostic graphs.

Table 3.6 Regression equations for total nutrient biomass of *P. radiata* trees on Yellow Kurosol formed on Silurian- Devonian sandstone (derived from Neilsen and Lynch 1998†).

Total P	=	76.2	*Mean Tree Volume	+ 8.389	R^2 =0.69
Total N	=	910.3	*Mean Tree Volume	- 37.915	R^2 =0.88
Total Mg	=	218.9	*Mean Tree Volume	+ 0.607	R^2 =0.93
Total Ca	=	814.3	*Mean Tree Volume	+ 19.698	R^2 =0.61

† Wood, bark, branches, cones and roots from each fertiliser treatment had similar concentrations. Foliar concentrations were similar for all elements except N. Foliar N concentrations in the N treated plots were up to 20% higher than in the unfertilised plots. As foliage contained up to 25 % of the biomass N, this could result in an error of up to 5%.

3.6 Results

3.6.1 Kurosol planted with P. radiata

3.6.1.1 Growth and Plantation Health

Total volume growth in the 15 years following the commencement of fertiliser treatments showed increasing *P. radiata* growth due to applied P and with more frequent N applications (Figure 3.1). The greatest response occurred in (P)N1Y, resulting in six times the volume growth of NIL and double the volume growth of the (P) treatment. Phosphorus re-applied at age 26 years resulted in an increasing volume growth of (P) over NIL. During the last six years of measurement there was virtually no volume growth of the NIL (Figure 3.1).

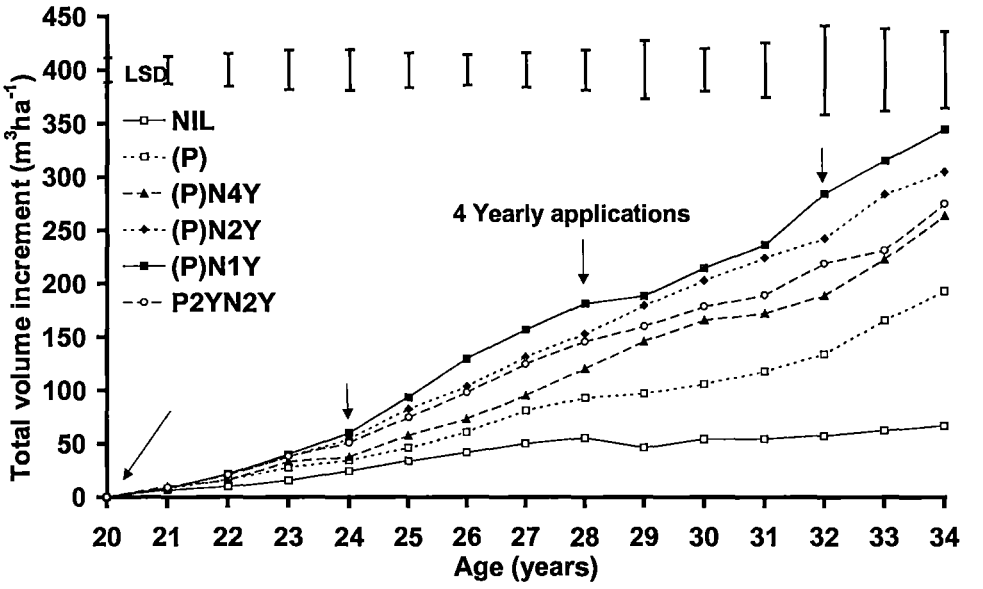


Figure 3.1 Growth of *P. radiata* with various fertiliser treatments following initial application at age 20 years. Bars indicate least significant difference between treatments (LSD).

During the fifteen years of measurement the average volume growth of NIL trees was 4 m^3 ha^{-1} PAI (periodic annual increment), the P treatment was 13 m^3 ha^{-1} PAI, and the (P)N1Y treatment was 27 m^3 ha^{-1} PAI. All N treatments produced 10 m^3 ha^{-1} PAI more than the P only treatment. Enhancement of PAI due to increased frequency of N application was most pronounced in the first seven years, beyond this period (age 27 years) the (P)N1Y treatment advantage over the (P)N2Y declined, and all the N application treatments had showed similar growth rates. The total volume growth

obtained during the 15 years, as a response to the quantity of N applied (fertiliser-use efficiency), showed a decrease in efficiency with annual application compared to applications every second and fourth year.

The health of fertilised trees improved dramatically within three years of initial N applications (Figure 3.2a). Tree health was a visual assessment of foliar colour, crown length, crown width, the presence of fused needle, and dead tops. To be defined as "overall healthy" the tree needed to have a full green crown with apparent no nutrient deficiency symptoms. Overall tree health improved due to P application and was substantially increased when N was also added. Deficiency symptoms, such as colour, crown length, crown width, and the presence of fused needles, decreased in proportion to the amount of fertiliser N applied. Prior to fertiliser treatments, approximately 56 percent of trees had dead tops, with no current apical growth. Dead tops were substantially improved by both P and N application and after the second application of P at age 26 years the dead tops in all fertilised treatments, including the P only, were reduced to zero (Figure 3.2b). However, no trees at age 20 had complete recovery due to soil moisture retention and drought, in particular the last 25 years has been a period of below mean annual rainfalls (Pook and Budd, 2002).

Foliar N and P concentrations varied considerably throughout the sampling times. Foliar N concentrations were marginally deficient prior to fertilisation (Table 3.7). Generally, treatment foliage N concentrations were lower than prior to fertilisation reflecting a dilution effect due to enhanced growth. By the end of the experiment the unfertilised foliar N concentrations had dropped substantially below deficiency levels and below the concentrations in all other treatments. Prior to the initial fertilisation, foliar P concentrations were below 0.08%, well below the level considered deficient. Throughout the experiment foliage from unfertilised trees remained at amounts between 0.08 percent and 0.09 percent, for both N and P, respectively. Even with fertiliser treatment the concentrations of N and P remained marginal throughout the experiment. Only at age 34 years were the N concentrations of the P plus N fertilised treatments significantly higher than those of the NIL and P (Table 3.7). At age 22 and

34 years the P treated trees resulted in a higher foliar P concentrations than the NIL trees.

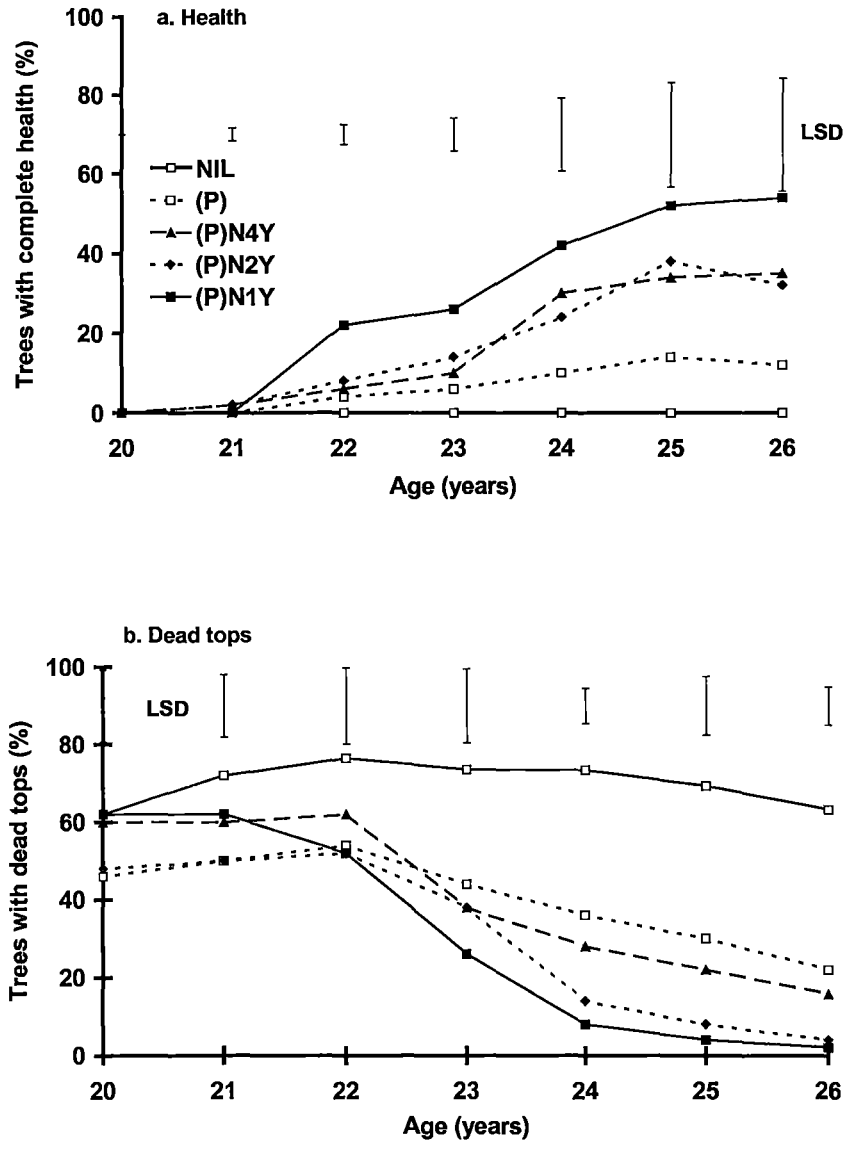


Figure 3.2 *P. radiata* trees with (a) complete health and (b) dead tops (%) for various treatments six years following initial treatment at age 20 years. Bars indicate LSD between treatments.

Further analysis of foliage for calcium, magnesium, potassium, copper, iron and zinc, at the end of the experiment revealed large variations and no significant trends between treatments. Physical parameters such as needle length, weight and colour for both age classes also showed no significant treatment trends. Concentrations of P, N, Ca and Mg in litter were not correlated with either one or two year old foliar

concentrations. Exchaneagble Ca and Mg have been found to be highly variable in the O2 horizon and they were not measured.

Table 3.7 Foliar nutrient concentrations (in one year old needles) for N and P for various treatments following initial treatment at age 20 years in *P. radiata*. Letters indicate significant difference between treatments ($p < 0.05$).

	Age (Years)							
Treatment	17		22		23		34	
***P. radiata* foliar N %**								
NIL	1.11	a	1.04	a	1.25	a	0.85	a
(P)	1.12	a	1.05	a	1.15	a	0.95	ab
(P)N4Y	1.09	a	1.10	a	1.20	a	1.19	b
(P)N2Y	1.18	a	1.16	a	1.30	a	1.11	b
(P)N1Y	1.18	a	1.20	a	1.30	a	1.15	b
P2YN2Y	1.20	a	1.29	a	1.20	a	1.11	b
***P. radiata* foliar P %**								
NIL	0.07	a	0.07	a	0.09	a	0.07	a
(P)	0.08	a	0.15	b	0.12	ab	0.11	b
(P)N4Y	0.08	a	0.10	ab	0.11	ab	0.12	bc
(P)N2Y	0.08	a	0.12	b	0.12	ab	0.10	b
(P)N1Y	0.08	a	0.10	ab	0.09	a	0.10	b
P2YN2Y	0.07	a	0.16	b	0.13	b	0.15	c

3.6.1.2 Fertiliser effects on the soil profile, soil pH and soil chemistry

There were significant differences in nutrient distribution in soil profiles at tree age of 34 years. The heaviest fertiliser treatments resulted in a significant increase in the mass of the O2 litter layer with over 70 t ha^{-1} for the annual fertilised treatment, compared to 26 t ha^{-1} for the (P) (Figure 3.3.). This contributed to a redistribution of nutrients from the mineral soil into the O2 horizon (Table 3.8). In comparison, changes in the mass of the O1 litter layers was not significant between the NIL and annually fertilised treatments, at 17 to 20 t ha^{-1}, respectively.

Under *P. radiata* there was also a significant increase in total N in the 10 – 20 cm soil layer of the (P)N1Y treatment compared to the (P) treatment. However, substantial variations in base nutrient loads generally resulted in no clear differences in total N content at other depths. Concentrations of mineral N in the soil profile were dominated by ammonium (NH_4^+), with nitrate (NO_3^-) commonly below detectable limits. Ammonium increased significantly with the rate of N application in the surface 0-10 cm of the Kurosol (Table 3.9). The remaining profile, to a depth of 50 cm, was generally lower in ammonium than the NIL treatment, suggesting little downward

movement of mineral N. An increased C/N ratio occurred in both the (P)N4Y and P2YN2Y treatments in the surface soil (0-10 cm); with no significant difference throughout the remaining profile (Table 3.10).

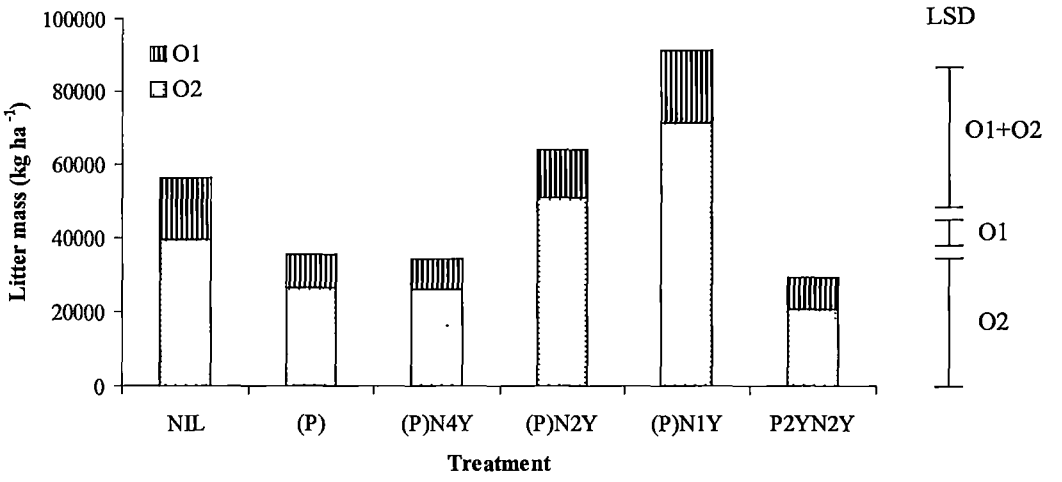


Figure 3.3 Mass of litter O1 and O2 horizons (kg ha^{-1}) for various treatments under *P. radiata*. Letters indicate significant difference between treatments ($p < 0.05$).

Phosphorus application resulted in no significant increase in total P in O2 layer or the surface mineral horizon. However, the total soil P in the top 50 cm of mineral soil was significantly higher for all rates of N application, compared to when P was applied alone. Total organic C in the top 50 cm of the Kurosol profile followed a similar trend to P. The distribution of soil C was concentrated in the surface 0-10 cm. At the highest N application rate soil C content in this horizon doubled from 21 t ha^{-1} in NIL to 41 t ha^{-1} in (P)N1Y (Table 3.8). Increased sulphur contents were observed in the O2 horizon and throughout the upper 50 cm of the mineral soil, but were only significant higher in the highest fertiliser treatments. (Table 3.8).

No significant differences were observed in total Mg or total Ca within the O2 or mineral soil horizons (Table 3.8). However, there was a substantial decrease in the profile total Ca overall, with the highest rate of N application. Base saturation was also substantially reduced due to fertilising, by 50 % to 60 % throughout the profile (Table 3.1c).

Fertiliser treatments significantly reduced pH (1:5 soil: water) by up to 0.7 of a unit in the 0 – 50 cm soil and by 0.3 of a unit in the O2 horizon (Figure 3.4a). Increasing rates of N application resulted in increasing reductions in pH, with (P)N4Y, (P)N2Y and (P)N1Y being significant more acidic compared to (P). At the highest rate, annual, fertilisation resulted in an overall pH reduction from 3.8 to 3.4 units within the surface 0 - 10 cm. The greatest reduction in pH from the annual application of fertiliser occurred in the 20 – 50 cm layer, where the reduction averaged 0.9 units (Table 3.9). Reductions in the order of 0.5 units were recorded throughout the entire soil profiles to the depth of 58 cm.

High rates of N application also resulted in significant reductions in exchangeable Mg (Table 3.9). Significant reductions were measured throughout the 50 cm sampled for all fertiliser treatments (Figure 3.4b). Nitrogen fertilisation reduced exchangeable Ca in the top 50 cm under *P. radiata* by 50 %, compared to the NIL soil and by 75 %, when compared to the (P). All N treatments significantly reduced the exchangeable Ca compared to the (P) to the depth of 30 cm.

Table 3.8 Nutrient content in tree, litter and soil (kg ha^{-1}) for various treatments under *P. radiata*. Letters indicate significant difference between treatments ($p < 0.05$).

Treatment	Tree†	Litter	Soil Depth (cm)					Soil Total
		O2	0-10	10-20	20-30	30-40	40-50	0-50
Total N (kg ha^{-1})								
NIL	135	331 ab	899 ab	726 ab	765 a	835 a	780 a	4006 a
(P)	217	167 a	707 a	405 a	516 a	588 a	554 a	2770 a
(P)N4Y	294	263 ab	949 ab	666 ab	661 a	751 a	798 a	3825 a
(P)N2Y	332	490 b	1272 b	741 ab	680 a	764 a	833 a	4290 a
(P)N1Y	364	717 b	1567 b	837 b	865 a	894 a	791 a	4954 a
P2YN2Y	317	191 a	820 a	532 ab	552 a	655 a	750 a	3310 a
Total P (kg ha^{-1})								
NIL	23	17 ab	75 a	80 a	123 a	142 a	159 ab	579 ab
(P)	30	11 a	82 a	105 a	123 a	130 a	127 a	567 a
(P)N4Y	36	13 ab	137 a	144 a	151 a	182 ab	188 ab	801 b
(P)N2Y	39	26 b	128 a	126 a	141 a	214 b	205 b	814 b
(P)N1Y	42	37 b	119 a	106 a	156 a	195 ab	253 b	830 b
P2YN2Y	38	14 ab	123 a	142 a	149 a	145 a	160 ab	718 b
Total S (kg ha^{-1})								
NIL		34 a	208 a	166 a	185 a	208 a	242 a	1009 ab
(P)		22 a	150 a	137 a	192 a	216 a	236 a	931 ab
(P)N4Y		30 a	106 a	99 a	125 a	250 a	280 a	860 a
(P)N2Y		60 a	223 a	232 a	261 ab	336 ab	360 ab	1412 bc
(P)N1Y		78 a	253 a	175 a	379 b	487 b	433 b	1727 c
P2YN2Y		25 a	168 a	165 a	228 a	275 a	338 ab	1173 ab
Total Ca (kg ha^{-1})								
NIL		127 a	322 a	271 a	157 a	54 a	37 a	841 a
(P)		90 a	404 a	219 a	123 a	63 a	29 a	837 a
(P)N4Y		105 a	331 a	164 a	95 a	59 a	40 a	688 a
(P)N2Y		217 a	367 a	146 a	83 a	82 a	68 a	746 a
(P)N1Y		169 a	260 a	72 a	40 a	37 a	25 a	434 a
P2YN2Y		106 a	326 a	124 a	71 a	61 a	55 a	638 a
Total Mg (kg ha^{-1})								
NIL	42	41 b	201 a	232 a	743 a	1035 a	946 ab	3158 a
(P)	62	21 ab	137 a	170 a	566 a	821 a	760 a	2454 a
(P)N4Y	80	25 ab	171 a	260 a	409 a	792 a	826 ab	2458 a
(P)N2Y	90	48 b	215 a	381 a	523 a	819 a	888 ab	2825 a
(P)N1Y	97	49 b	158 a	150 a	735 a	659 a	666 a	2368 a
P2YN2Y	86	17 a	157 a	205 a	589 a	963 a	1089 b	3004 a
Total C (kg ha^{-1})								
NIL			20984 a	22843 bc	14448 a	12438 a	10565 a	81277 b
(P)			19759 a	13427 a	15596 ab	13184 a	10191 a	72156 a
(P)N4Y			33313 b	18288 ab	21063 b	15887 a	13615 a	102164 d
(P)N2Y			32906 b	27887 c	17047 ab	16154 a	13708 a	107702 e
(P)N1Y			40654 c	20876 b	17481 ab	15328 a	9324 a	103663 d
P2YN2Y			33925 b	21382 b	13480 a	15744 a	12114 a	96646 c

† Calculated from Table 3.5

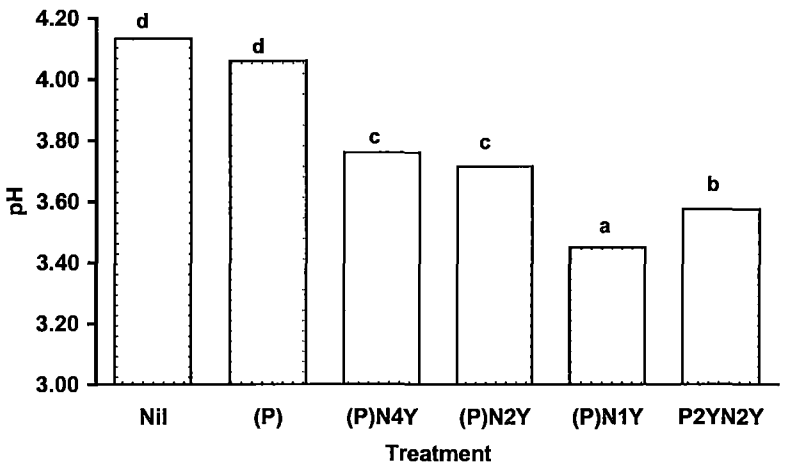
Table 3.9 Litter and soil pH and exchangeable Mg, Ca and K and total NH_4^+ (kg ha^{-1}) for various treatments under *P. radiata*. Letters indicate significant difference between treatments ($p < 0.05$).

Treatment	Tree	Litter		Soil Depth											
		O2		0-10		10-20		20-30		30-40		40-50		0-50 cm	
pH														**Soil Average**	
NIL		4.2	c	3.8	b	3.8	b	4.2	c	4.4	c	4.4	c	4.1	d
(P)		4.1	b	3.9	ab	4.0	ab	4.1	c	4.1	c	4.2	c	4.1	d
(P)N4Y		4.0	ab	3.6	ab	3.7	ab	3.7	ab	3.9	b	3.9	b	3.8	c
(P)N2Y		4.0	ab	3.6	ab	3.6	ab	3.6	b	3.8	b	3.9	bc	3.7	c
(P)N1Y		3.9	a	3.4	a	3.5	a	3.3	a	3.4	a	3.6	a	3.4	a
P2YN2Y		3.9	a	3.6	ab	3.4	ab	3.5	ab	3.6	ab	3.7	ab	3.6	b
Total Ex Mg (kg ha^{-1})														**Soil Total**	
NIL				0.8	b	0.7	c	0.8	c	0.9	c	0.9	b	4.1	c
(P)				1.2	c	0.4	b	0.4	ab	0.6	b	0.7	ab	3.4	b
(P)N4Y				0.6	ab	0.4	b	0.4	ab	0.6	bc	1.0	b	2.9	b
(P)N2Y				0.5	a	0.4	b	0.5	b	0.7	c	0.8	b	2.9	b
(P)N1Y				0.5	a	0.2	a	0.2	a	0.2	a	0.5	a	1.5	a
P2YN2Y				0.4	a	0.2	ab	0.3	a	0.4	ab	0.6	ab	1.8	a
Total Ex Ca (kg ha^{-1})															
NIL				2.3	a	1.6	b	1.1	b	0.4	a	0.5	a	5.8	b
(P)				7.3	c	2.8	c	1.7	c	0.9	a	0.5	a	13.2	d
(P)N4Y				2.4	a	1.2	b	0.9	b	0.7	a	0.5	a	5.8	b
(P)N2Y				2.7	a	1.2	b	1.0	b	1.1	a	0.9	a	7.0	b
(P)N1Y				2.3	a	0.6	a	0.2	a	0.2	a	0.1	a	3.3	a
P2YN2Y				5.7	b	1.6	ab	1.0	b	0.9	a	0.8	a	9.9	c
Total Ex K (kg ha^{-1})															
NIL		76.1	a	0.4	a	0.3	a	0.4	a	0.4	a	0.3	a	1.7	a
(P)		44.8	a	0.5	a	0.2	a	0.2	a	0.3	a	0.3	a	1.5	a
(P)N4Y		46.7	a	0.3	a	0.2	a	0.2	a	0.2	a	0.2	a	1.1	a
(P)N2Y		77.8	a	0.2	a	0.3	a	0.4	a	0.3	a	0.3	a	1.5	a
(P)N1Y		119.8	a	0.4	a	0.2	a	0.3	a	0.3	a	0.2	a	1.4	a
P2YN2Y		36.8	a	0.3	a	0.2	a	0.3	a	0.4	a	0.4	a	1.7	a
Total NH_4^+ (kg ha^{-1})															
NIL		18.3	a	1.0	ab	1.2	b	2.2	c	3.4	c	2.7	b	10.5	d
(P)N4Y		8.1	a	0.5	a	0.4	a	0.5	a	0.7	a	0.7	a	2.7	a
(P)N2Y		7.5	a	1.2	b	1.3	b	1.1	b	0.7	a	0.7	a	4.9	b
(P)N1Y		28.5	a	2.9	c	1.0	ab	1.4	b	1.4	b	1.2	a	8.0	c

Table 3.10 C/N ratios of soil for various treatments under *P. radiata*. Letters indicate significant difference between treatments ($p < 0.05$).

Treatment	Soil Depth (cm)				
	0-10	10-20	20-30	30-40	40-50
NIL	23.1 a	30.1 a	18.9 a	14.9 a	13.6 a
(P)	28.6 ab	33.0 a	30.3 a	22.4 a	18.0 a
(P)N4Y	34.6 b	28.0 a	32.0 a	21.3 a	17.6 a
(P)N2Y	28.2 ab	37.5 a	25.2 a	21.1 a	16.4 a
(P)N1Y	25.9 ab	24.7 a	20.5 a	17.2 a	11.2 a
P2YN2Y	41.4 b	41.0 a	25.1 a	24.3 a	16.3 a

a.



b.

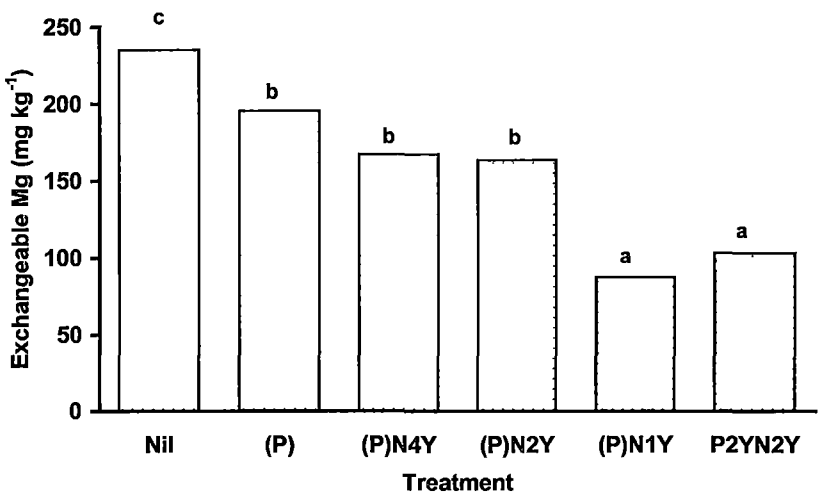


Figure 3.4 Soil (a) average pH for 0-50 cm of mineral soil and (b) sum of exchangeable Mg in the top 50 cm, for various treatments of N and P. Letters indicate significant differences between treatments $p < 0.05$.

3.6.2 Ferrosol planted with E. regnans

3.6.2.1 Growth and Plantation Health

Increasing amounts of N plus P fertilisation resulted in increasing volume growth (Figure 3.5). Volume growth doubled, from age 5 years to age 19 years, from 6 m^3 ha^{-1} PAI in the NIL treatment to 14 m^3 ha^{-1} PAI in the P1YN1Y treatment. Total volume growth obtained during the 15 years, as a response to the quantity of fertiliser applied, showed a decrease in response with annual compared to biannual applications (Figure 3.5).

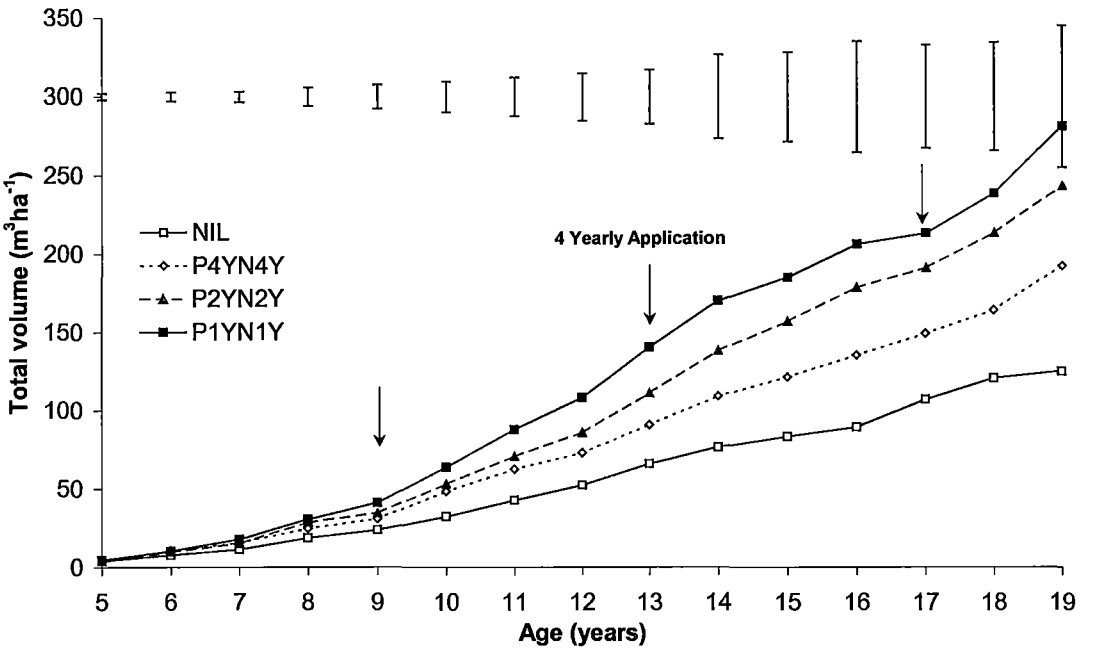


Figure 3.5 Growth of *E. regnans* unfertilised or fertilised with N plus P, annually, two yearly or four yearly, following initial application at age six years. Bars indicate LSD between treatments (p = 0.05).

Significant volume growth increases compared to the NIL treatment due to annual N plus P fertilisation occurred after 300 kg of N ha^{-1} had been applied in both P1YN1Y and P2YN2Y treatments, at ages eight and ten years, respectively. Beyond ten years of age there was no significant increase in volume growth between the P1YN1Y and P2YN2Y treatments. Treatment P4YN4Y did not significantly increase volume growth from the NIL. From ages 14 to 19 years the average PAI's for NIL and the average of the three levels of N fertilisation were 10 and 21 m^3 ha^{-1} yr^{-1}, respectively.

The factorial experiment showed that there was strong growth response to applied N, at the rate of 700 kg N ha^{-1}, but no response to applied P, at the rate of 322 kg P ha^{-1}, during the experimental period and no interaction between N and P (Figure 3.6).

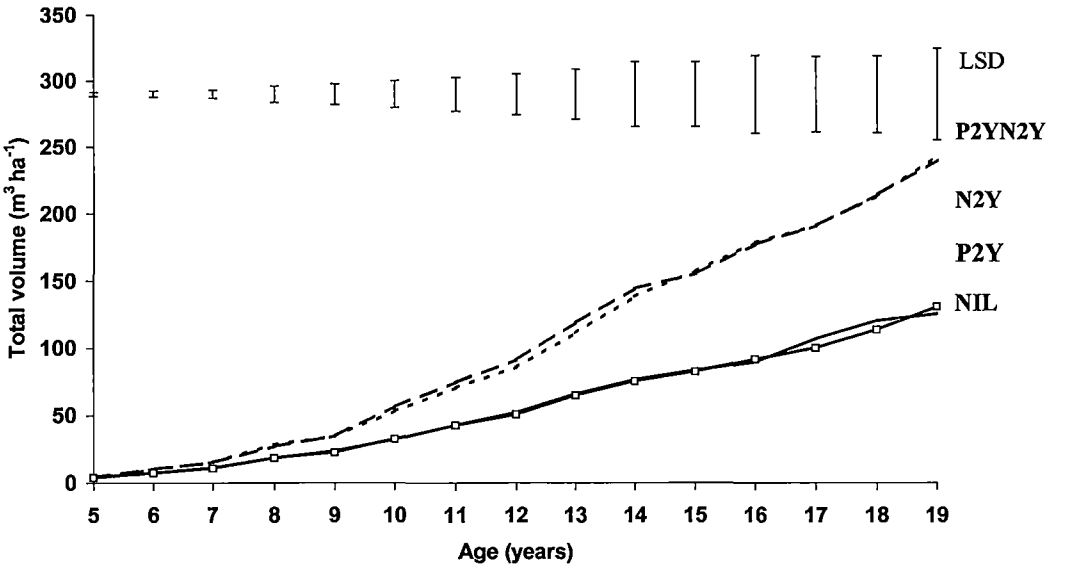


Figure 3.6 Growth of *E. regnans* fertilised with factorial combinations of N and P, applied two yearly following initial application at age six years. Bars indicate LSD between treatments (p = 0.05).

Visual assessment of *E. regnans* health is difficult in older stands and was not undertaken in this experiment. Foliar N and P concentrations varied throughout the sampling times. In general, foliar N concentrations ranged from 1.09 to 1.40 % between treatments, though this was not significant (Table 3.11). In eucalypts, foliar deficiency levels for N is suggested as < 1.1 % and at < 0.1 % in P (Reuter and Robinson, 1997). The foliar concentrations of N and P remained marginal throughout the experiment, except for the P concentration in the P2Y treatment. Despite higher foliar P content in P2YN2Y, (0.16 % P at ages 14 and 20 years) this treatment did not produce more growth than N2Y (foliar P content of 0.10%). This higher P content resulted in a significantly higher N/P ratio for N2Y, varying over the four sampling periods from 16.1 to 18.6, compared to N/P ratios of 7.3 to 12.3 for P2YN2Y. Neither foliar Ca nor foliar S concentrations reflected differences in quantities of these nutrients applied in various treatments. Foliar Ca concentrations were high, ranging from 0.5 % to 1.0 %, while S concentrations were in the range considered adequate, falling between 0.11 % to 0.14 %.

Table 3.11 Foliar nutrient concentrations for N and P for various treatments following initial treatment at age six years in *E. regnans*. Letters indicate significant difference between treatments ($p < 0.05$).

	Age (Years)							
Treatment	**10**		**12**		**14**		**20**	
Foliar N %								
NIL	1.38	a	1.26	a	1.15	ab	1.30	a
P4YN4Y	1.28	a	1.22	a	1.09	a	1.38	a
P2YN2Y	1.31	a	1.23	a	1.17	ab	1.28	a
P1YN1Y	1.34	a	1.27	a	1.24	ab	1.20	a
N2Y	1.40	a	1.30	a	1.29	b	1.24	a
P2Y	1.36	a	1.20	a	1.18	ab	1.21	a
Foliar P %								
NIL	0.10	ab	0.08	ab	0.10	ab	0.10	a
P4YN4Y	0.10	ab	0.09	ab	0.12	ab	0.15	b
P2YN2Y	0.11	b	0.10	b	0.16	b	0.16	b
P1YN1Y	0.16	c	0.14	b	0.14	b	0.17	b
N2Y	0.08	a	0.07	a	0.08	a	0.07	a
P2Y	0.19	c	0.13	b	0.24	c	0.23	c
Ratio N/P								
NIL	13.9	b	16	a	11.7	a	13.2	b
P4YN4Y	12.9	b	14	a	9.2	a	9.5	ab
P2YN2Y	11.4	ab	12	a	7.4	a	7.8	a
P1YN1Y	8.6	ab	9	a	8.6	a	7.2	a
N2Y	17.3	b	20	a	15.9	a	17.4	b
P2Y	7.3	a	9	a	5.0	a	5.2	a

3.6.2.2 Fertiliser effects on the soil profile, soil pH and soil chemistry

The heaviest fertiliser treatment, P1YN1Y, resulted in a significant increase in the mass of both the O1 and O2 horizons compared to NIL (Figure 3.6). The mass of the litter layers increased from 28 t ha^{-1} in NIL to 102 t ha^{-1} in P1YN1Y. The greatest increase occurred in the O2 horizon, which increased from one cm and 14 t ha^{-1} for NIL, to six cm deep and 77 t ha^{-1} for P1YN1Y. This contributed to an accumulation of nutrients in the O2 with significantly more N, P, S and Ca in the annually fertilised treatment (Table 3.12). Nitrogen concentrations in the O2 horizon were 1.22 % in P1YN1Y treatment, compared with 0.88 % in NIL. However, substantial variations often resulted in no clear significant differences in total N, organic C or C/N ratios in the soil (Table 3.13).

The O2 horizon in the NIL treatment contained 122 kg N ha^{-1} compared with 949 kg N ha^{-1} in the P1YN1Y. This seven-fold increase accounted for 60% of the N applied. However, the annually fertilised soil profile to a depth of 50 cm contained significantly less N than the NIL treatment. Soil mineral N was dominated by NH_4^+, with NO_3^- commonly below detectable limits. Mineral N (NH_4^+) content was significantly higher in the O2 horizon for P1YN1Y, than other treatments, while there were no significant differences in mineral N content due to N application in the upper 50 cm of mineral soil (Table 3.14).

Phosphorus application resulted in an increase in total P in the O2 horizon, although this was only significant at the highest rate of fertiliser application. Phosphorus also accumulated in the top 10 cm of the Ferrosol profile in the higher rates of P application. Significantly higher amounts of P were observed in the top 50 cm of all P fertilised soils.

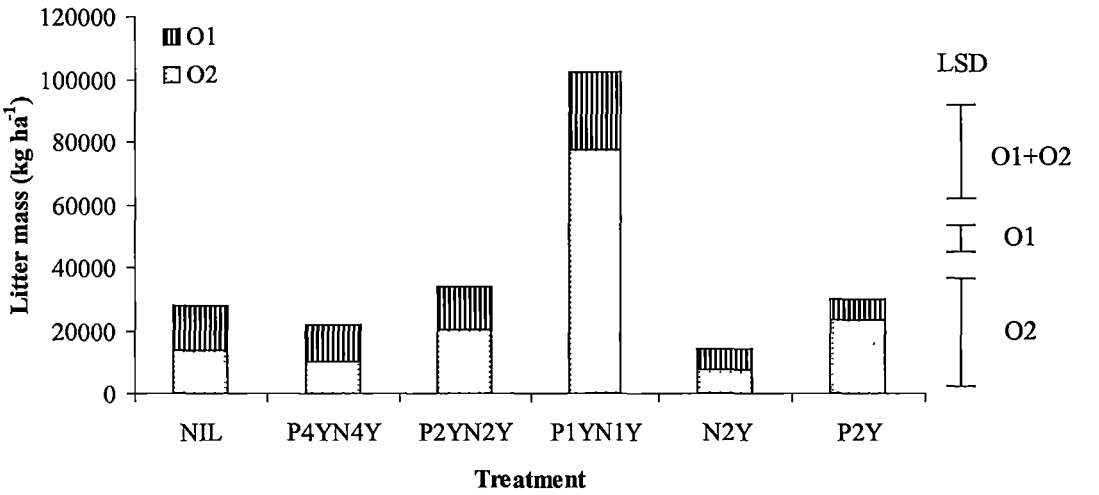


Figure 3.7 Mass of litter O1 and O2 horizons (kg ha^{-1}) for various treatments under *E. regnans*. Letters indicate significant difference between treatments ($p < 0.05$).

Table 3.12 Nutrient content in litter and soil (kg ha^{-1}) for various treatments under *E. regnans*. Letters indicate significant difference between treatments ($p < 0.05$).

Treatment	Litter	Soil Depth										Soil Total	
	O2	0-10		10-20		20-30		30-40		40-50		0-50 cm	
Total N (kg ha^{-1})													
NIL	122	a	2275	b	2008	b	1672	a	841	a	665	a	7460 b
P4YN4Y	97	a	1970	ab	1908	ab	1526	a	1209	a	1046	a	7659 b
P2YN2Y	190	a	2039	ab	1899	ab	1411	a	1087	a	800	a	7236 b
P1YN1Y	949	b	1693	a	1102	a	933	a	733	a	616	a	5077 a
N2Y	78	a	2507	b	1693	ab	1091	a	1005	a	833	a	7129 b
P2Y	237	a	2041	ab	1166	a	902	a	753	a	665	a	5526 ab
Total P (kg ha^{-1})													
NIL	10	a	357	a	379	ab	296	ab	257	a	266	a	1555 b
P4YN4Y	14	a	442	ab	532	b	454	b	354	a	372	a	2154 e
P2YN2Y	30	a	654	cd	507	ab	406	ab	357	a	327	a	2252 f
P1YN1Y	136	b	569	bc	385	ab	360	ab	360	a	380	a	2054 d
N2Y	5	a	359	a	317	a	235	a	209	a	199	a	1320 a
P2Y	32	a	821	d	330	a	244	a	221	a	228	a	1844 c
Total S (kg ha^{-1})													
NIL	13	a	520	ab	485	ab	521	ab	476	a	462	a	2465 ab
P4YN4Y	11	a	485	ab	579	ab	622	b	605	ab	707	ab	2999 bc
P2YN2Y	20	a	560	ab	637	b	685	b	793	b	831	b	3506 c
P1YN1Y	99	b	728	b	705	b	726	b	797	b	778	b	3733 c
N2Y	8	a	447	a	381	a	369	a	376	a	391	a	1964 a
P2Y	26	ab	350	a	384	a	409	ab	453	a	484	ab	2079 a
Total C (kg ha^{-1})													
NIL			48113	a	35023	a	25270	a	14762	a	14013	a	137181 a
P4YN4Y			43785	a	35307	a	25960	a	18511	a	16225	a	139788 a
P2YN2Y			43470	a	43584	a	29353	a	21093	a	14160	a	151659 a
P1YN1Y			56726	a	22288	a	18443	a	15404	a	12077	a	124937 a
N2Y			53802	a	39189	a	29721	a	18290	a	16225	a	157227 a
P2Y			49014	a	27396	a	19691	a	16446	a	13201	a	125748 a
Total Ca (kg ha^{-1})													
NIL	67	a	743	ab	608	a	548	a	467	a	388	a	2754 b
P4YN4Y	61	a	459	ab	509	a	436	a	399	a	280	a	2083 ab
P2YN2Y	121	a	701	ab	589	a	384	a	352	a	266	a	2292 ab
P1YN1Y	674	b	866	b	602	a	579	a	384	a	353	a	2784 b
N2Y	29	a	328	a	276	a	215	a	235	a	255	a	1308 a
P2Y	154	a	1030	b	463	a	400	a	307	a	271	a	2470 b
Total Mg (kg ha^{-1})													
NIL	24	ab	951	ab	1161	ab	1223	b	1155	ab	1118	a	5607 ab
P4YN4Y	14	a	661	ab	728	ab	743	a	924	ab	865	a	3922 a
P2YN2Y	28	ab	625	a	764	ab	776	ab	899	ab	908	a	3972 a
P1YN1Y	51	b	1098	b	1191	b	1298	b	1545	c	1496	a	6628 b
N2Y	10	a	670	ab	676	a	684	a	728	a	844	a	3602 a
P2Y	37	ab	995	ab	1060	ab	1103	ab	1225	b	1193	a	5576 ab

Table 3.13 C/N ratios of soil for various treatments under *E. regnans*. Letters indicate significant difference between treatments ($p < 0.05$).

Treatment	Soil Depth (cm)									
	0-10		10-20		20-30		30-40		40-50	
NIL	21.2	a	17.5	a	16.0	a	17.5	a	21.2	a
P4YN4Y	22.2	a	18.6	a	16.9	a	15.2	a	15.6	a
P2YN2Y	20.3	a	22.4	a	20.9	a	19.4	a	18.0	a
P1YN1Y	32.5	a	20.3	a	19.9	a	21.0	a	19.4	a
N2Y	21.3	a	23.1	a	27.2	a	18.7	a	20.2	a
P2Y	26.0	a	24.9	a	24.0	a	22.8	a	21.0	a

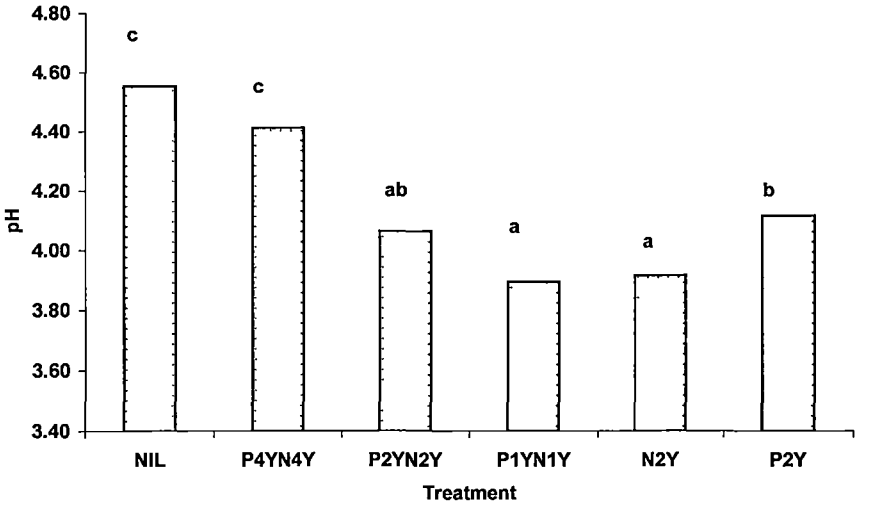
Fertiliser treatments significantly reduced pH (1:5 soil: water) by up to 0.7 of a unit in the 0 – 50 cm soil depth and 0.6 of a unit in the O2 horizon (Table 3.14). Higher rates of fertiliser N resulted in significantly lower soil pH. Treatments P2Y and N2Y also caused significant drops, which were of the same order as that resulting from P2YN2Y. In the O2 horizon, significant declines in pH occurred only at the highest fertiliser rate. At the highest rate, annual fertilisation resulted in an overall pH reduction from 4.6 to 3.9 units, with the greatest reduction occurring in the surface soil (Figure 3.8a). Reductions in the order of 0.5 units were recorded throughout the entire soil profile, to the depth of 90 cm.

Exchangeable Mg was also significantly reduced in the soil profile to a depth of 50 cm in the N2Y treatment (Figure 3.8b) and in the surface 10 cm for P1YN1Y (Table 3.14). Significant differences were not observed in total Mg content (Table 3.12). Total Ca in the soil, to the depth of 50 cm, was significantly reduced by N2Y treatment, to 50 % of that in NIL. Base saturation was also substantially reduced due to fertilising, by 50 % to 60 % throughout the profile (Table 3.3c). There was no significant effect of N or P fertilisation on exchangeable K concentrations (Table 3.14).

Table 3.14 Litter and soil pH and concentration of exchangeable Mg, Ca and K and total NH_4^+ (kg ha^{-1}) for various treatments under *E. regnans*. Letters indicate significant difference between treatments ($p < 0.05$).

Treatment	Litter		Soil Depth											
	O2		0-10		10-20		20-30		30-40		40-50		0-50 cm	
pH													**Average**	
NIL	4.8	bc	4.5	c	4.6	b	4.6	b	4.5	b	4.5	b	4.6	c
P4YN4Y	5.0	c	4.4	bc	4.4	b	4.4	b	4.5	b	4.5	b	4.4	c
P2YN2Y	4.9	c	4.0	ab	4.0	ab	4.1	ab	4.1	ab	4.1	ab	4.1	ab
P1YN1Y	4.2	a	3.6	a	3.8	a	3.9	a	4.1	a	4.0	a	3.9	a
N2Y	4.7	b	4.0	ab	4.0	a	3.8	a	3.8	a	4.0	a	3.9	a
P2Y	4.9	c	4.1	b	4.2	ab	4.1	ab	4.1	ab	4.1	a	4.1	b
Total Ex Mg (kg ha^{-1})													**Soil Total**	
NIL			3.3	b	3.4	a	3.2	a	2.3	a	2.2	a	14.3	b
P4YN4Y			2.3	ab	2.5	a	2.7	a	3.7	a	3.4	a	14.7	b
P2YN2Y			1.5	ab	1.9	a	2.0	a	2.6	a	2.7	a	10.5	ab
P1YN1Y			0.9	a	1.8	a	2.4	a	2.4	a	2.4	a	9.9	ab
N2Y			1.2	ab	1.5	a	1.6	a	1.8	a	2.2	a	8.3	a
P2Y			2.8	ab	2.6	a	2.8	a	2.7	a	2.9	a	13.7	b
Total Ex Ca (kg ha^{-1})														
NIL			5.6	ab	5.7	a	5.3	a	3.4	a	2.8	a	22.8	ab
P4YN4Y			5.3	ab	7.2	a	6.1	a	6.0	a	4.4	a	29.1	b
P2YN2Y			8.8	b	7.6	a	5.7	a	5.9	a	4.6	a	32.6	b
P1YN1Y			5.8	ab	5.5	a	5.8	a	5.5	a	5.0	a	27.6	b
N2Y			3.1	a	3.0	a	2.6	a	2.9	a	3.3	a	14.9	a
P2Y			10.2	b	6.2	a	5.6	a	4.5	a	4.2	a	30.7	b
Total Ex K (kg ha^{-1})														
NIL	20	a	0.8	a	0.8	a	0.6	a	0.3	a	0.2	a	2.7	a
P4YN4Y	12	a	1.0	a	1.2	a	0.7	a	0.5	a	0.5	a	3.9	a
P2YN2Y	23	a	0.8	a	1.0	a	0.7	a	0.7	a	0.6	a	3.9	a
P1YN1Y	73	a	0.5	a	0.6	a	0.6	a	0.4	a	0.5	a	2.7	a
N2Y	17	a	0.5	a	0.6	a	0.6	a	0.5	a	0.5	a	2.6	a
P2Y	27	a	0.8	a	0.8	a	0.4	a	0.3	a	0.3	a	2.5	a
Total NH_4^+ (kg ha^{-1})														
NIL	0.62	a	7.85	a	12.91	b	6.79	ab	5.98	a	5.10	a	38.63	b
P4YN4Y	0.16	a	2.23	a	11.13	b	9.45	b	5.27	a	3.57	a	31.65	b
P2YN2Y	0.3	a	4.88	a	5.17	ab	6.57	ab	4.58	a	3.43	a	24.62	ab
P1YN1Y	8.5	b	2.60	a	3.17	a	4.25	ab	4.60	a	3.98	a	18.61	ab
N2Y	0.25	a	4.17	a	4.91	ab	4.80	ab	3.50	a	3.43	a	20.81	ab
P2Y	0.54	a	3.73	a	1.84	a	2.06	a	1.85	a	1.99	a	11.47	a

a.



b.

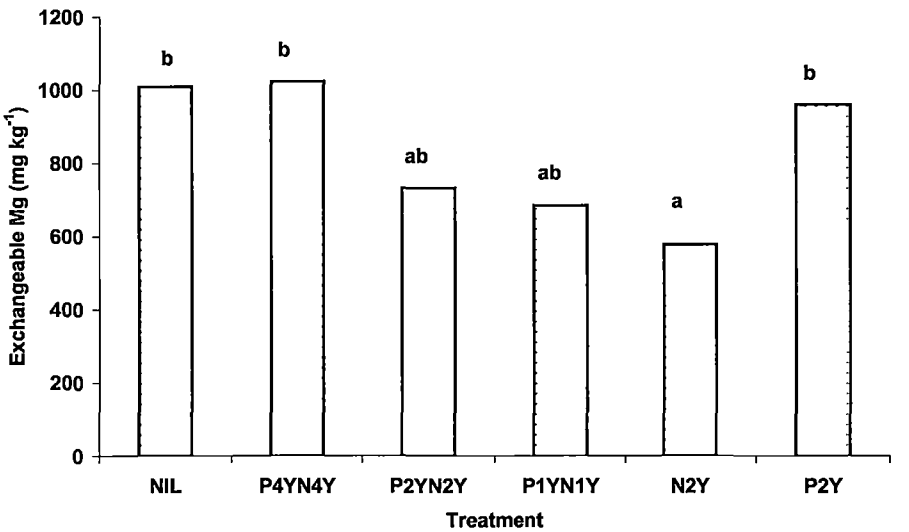


Figure 3.8 (a) Soil average pH levels over the depth of 50 cm and (b) sum of exchangeable Mg in the top 50 cm, for various N and P fertilisation treatments.

3.7 Discussion

3.7.1 *P. radiata* plantation growth

The soil type on which the *P. radiata* experiment was established was defined as P deficient, and large volume growth increases of *P. radiata* have been obtained on this soil type due to applications of P fertiliser (Neilsen *et al.*, 1984). The constraint on *P. radiata* growth due to P deficiency, as measured by low P concentration in the

foliage, was addressed with two single applications of P, resulting in a three-fold increase in stem volume growth. These long-term growth responses are in agreement with the Type II response associated with moderate P applications (Snowdon, 2002). Single applications of P fertiliser have produced continued response for many years in a number of forest crops (Waring, 1969; Ballard, 1978; Gentle *et al.*, 1986; Turner and Lambert, 1986; Comerford *et al.*, 2002; Turner *et al.*, 2002).

Rates of N mineralisation depend greatly on climate, and in cool-temperate forests these are low, sometimes less than 10 kg N ha^{-1} yr^{-1}. Low rates of N mineralisation mean that high growth rates could be achieved only through fertilisation (Binkley and Hart, 1989; Jacobson and Pettersson, 2001). The site used for this research is not uncommon with low native soil nutrients. Such problems will become more common as soil organic matter declines with production above sustainable levels. Economic plantation growth cannot be sustained from native soil N alone. Large growth responses resulting from applied N fertiliser could be sustained by further fertiliser additions, recycling of N within the trees, or supply from mineralisation of accumulated soil N (Neilsen *et al.*, 1992). Growth responses from application of N fertiliser are often short lived and considered a Type I growth response, however multiple applications of N can result in long-term responses simulating a Type II response, as seen in this study. In this study, N application to a *P. radiata* plantation yearly, every second and every fourth year through 15 years resulted in improved growth increases that depended on the amount of N applied. Nitrogen application at the highest rate almost doubled the plantation production from 192 m^3 ha^{-1} with P only to 344 m^3 ha^{-1} on this site during the 15 years of the research. This was in agreement with the large volume increase in *P. radiata* from annual and periodic N application observed in a number of field experiments (Raison *et al.*, 1990; Neilsen and Lynch, 1998). The total growth showed lower fertiliser response efficiency with annual application compared to applications every second or fourth year. Once the *P. radiata* stand was carrying full canopy, applications every fourth years were sufficient to maintain growth. This four-year period has provided the opportunity to develop a viable fertilising program. For economic return on investment in Tasmania, plantation growth in excess of 20 m^3 ha^{-1} mean annual increment MAI, and probably 25 m^3 ha^{-1}

(MAI), are required. Many soils have low organic matter levels and meeting these growth rates requires fertiliser additions.

Reduced volume growth rates in the annually fertilised trees beyond 29 years of age may reflect the limited water availability at this site, as soil water availability was significantly reduced under fertilised trees compared to unfertilised trees (Chapter 4). Raison *et al.* (1990) also observed moisture deficits in a low rainfall area reduced the growth of 10-year-old *P. radiata* plantations growing on podzolic soil. During the four-year growing period studied, they observed an overall 24 percent increase in volume due to N application, but no growth during a drought period. In addition, Crane and Banks (1992) observed the greatest response to N fertilisation in low rainfall plantations occurred when irrigation treatments were included. Annual rainfall was highly variable between years at this site and this is discussed in Chapter 4.

3.7.2 *P. radiata* stand health and foliar nutrient concentrations

Various authors have addressed the effect of repeated fertiliser applications on growth and sustainability issues (Nohrstedt, 1990; Tamm and Popovic, 1995; Nohrstedt *et al.*, 2000). In this study, under *P. radiata* all N application resulted in improved health and volume growth increases, dependent on the amount of N applied.

Foliar P concentration in *P. radiata* reflected the increase in P availability when P was applied. Increases in volume growth in the *P. radiata* were accompanied by rapid increases in stand health. Enhanced overall stand health and needle biomass observations are common after N application to *P. radiata* previously showing severe nutrient deficiencies (Fagerstrom and Lohm, 1977; Hunter and Hoy, 1983; Neilsen *et al.*, 1984; Neilsen *et al.*, 1992). In this study, P application alone resulted in dead tops recovering, but other symptoms were only relieved when N was also added. After fertilisation, N concentrations rapidly increase to a maximum within a year or two and subsequently decline (Fagerstrom and Lohm, 1977; Hunter and Hoy, 1983; Crane and Banks, 1992). Reduced volume growth response to N application after three to four years are also reflected in declining foliage N concentrations.

Foliar nutrient concentrations varied between years, independent of fertilisation. Foliar nutrient concentrations can be influenced by many factors including seasonal differences, soil moisture availability and time since fertilising (Nason *et al.*, 1990). The periodic foliage sampling in this study made it difficult to assess pluses in nutrients due to fertilisation and uptake and retranslocations in the tree. Half to two-thirds of the nutrients required for the new foliage production may result from retranslocation from older foliage (Miller, 1981; Lim and Cousens, 1986). Such retranslocation from foliage was observed in the spring by Fife and Nambiar (1997) in six to ten year old *P. radiata*, but not until late summer and autumn by Crane and Banks (1992) in ten year old *P. radiata*.

Foliar nutrient concentrations are considered good indicators of tree health (Woolons and Will, 1975; Dell *et al.*, 2002). In *P. radiata* deficiency symptoms were associated with low nutrient concentrations. Phosphorus concentrations before treatment, at age 17 years, were well below levels considered deficient (Neilsen *et al.*, 1984), and throughout the experiment NIL trees remained at these low levels. However, limitations of foliage analysis in predicting volume growth response have been observed previously. For example, Benson *et al.* (1992b) observed that there was little similarity between patterns of stem growth in *P. radiata* and N concentration in the foliage. Hunter and Hoy (1983) suggested that foliage responses to N fertiliser were better expressed in terms of needle growth rather than needle N content. However in a study of young *P. radiata* stands (4 to 5 year old), Hunter *et al.* (1987) indicated that foliar N and needle mass could be used to estimate stem volume increments.

3.7.3 Nutrient distribution in soil and O2 horizons

Litter plays an intricate role in the cycling of nutrients in forests. To improve the understanding of the effects of fertilising on nutrient cycling in plantations it is necessary to quantify the extent to which fertilising increases the turnover of nutrients through the litter layer. At sites with low nutrient content in the mineral soil, litter quality, as measured in terms of C: N ratio, can be highly correlated with tree volume growth (Smith *et al.*, 2000). The form and species of litter also has a strong influence

on N mineralisation (Adams and Attiwill, 1986; Gower and Son, 1992; Prescott *et al.*, 1993; Prescott and Preston, 1994).

With fertilisation, the litter layer constitutes a large and significant source of mineralisable nutrients. This was evident in this research. Applications of N at the highest rate resulted in significant increase in O2 biomass, with N mass doubling in the O2 horizon. This was in agreement with work in a range of forest stands that have shown increased forest floor organic matter production and N mass due to N fertilisation (Nohrstedt, 1990; Theodorou and Bowen, 1990; Neilsen and Lynch, 1998). In *P. radiata*, Baker *et al.* (1986) reported a stand fertilised with 960 kg N ha^{-1} during 10 years, which had 15 t litter ha^{-1} (containing 210 kg N ha^{-1} and 18 kg P ha^{-1}) compared to unfertilised plots with 6 t litter ha^{-1} (containing 57 kg N ha^{-1} and 12 kg P ha^{-1}). Under *P. radiata*, Fife and Nambiar (1997) observed, prior to canopy closure, litter amounts increased with increasing N application rates. An increase in litter production of 75 % was also accompanied by an increase in N concentrations in the litter and foliage. This contrasts to this study, where inconsistent foliar concentrations were observed.

3.7.4 Changes in cations and pH under *P. radiata*

Changes in the soil acidity occurred after long-term N additions as ammonium sulphate. There was a general trend of decreasing pH with increasing N application, with the maximum decline of 0.6 units in the (P)N1Y treatment. Extensive research in temperate ecosystems across Northern America and Europe have shown that N inputs both due to atmospheric deposition and fertilisation can lead to soil acidification, depletion of base cations and increased availability of potentially toxic Al (Matson *et al.*, 2002). Acidification has also been reported for many agricultural soils in Australia (Porter *et al.*, 1995).

In the fertilised Kurosol, significant pH changes occurred to at least 50 cm, associated with low organic matter and nutrients. Soil pH declines were greatest in the subsoil (20-50 cm) with decreases up to 1 unit. In several studies and reviews significant pH

decreases, ranging from 0.5 to 1.9 units, in soil solution of subsoil have been determined (Vestgarden *et al.*, 2001; Paul *et al.*, 2003a). Smethurst *et al.* (2001) noted the relationship between the rate of N application and pH decline in the soil solution was predominantly linear or log-linear, and was effected by the amount of fertilisation, rather than the timing of applications. Nitrogen uptake by plants has been attributed to the development of acidic subsurface soil layers (Paul *et al.*, 2003a).

Because of the fertilisers used here, single-superphosphate and ammonium sulphate, large amounts of S, up to 1.7 t ha^{-1}, were applied to the plot. While superphosphate has little impact on active acidity, ammonium sulphate is known to induce soil acidification though the dual impact of nitrification and sulphate and cation leaching. Additions of elemental S and dilute sulphuric acid have been used as a means of acidifying soil for experiments in Europe (Tamm and Popovic 1995). Soils differ in their ability to buffer added S (Binkley *et al.*, 1988).

In a study using similar rates of N fertiliser in Sweden, Tamm and Popovic (1995) observed that application of 1080 kg N ha^{-1} (plus 200 kg P ha^{-1} and 384 kg K ha^{-1}) during 14 years, decreased the pH in the 5-20 cm soil layer by 0.5 pH units, but increased the pH in the litter. In that study, both urea and ammonium nitrate were found to acidify the soil but only at 74 kg N ha^{-1} yr^{-1} or more. At lower amounts (37 kg N ha^{-1} yr^{-1}) pH decline only occurred when N was combined with P and K fertilisers at application. Khanna *et al.* (1992) observed pH changes of 0.8 units in KCl within one year of NPK fertilisation (ammonium sulphate, 400 kg N ha^{-1}, superphosphate, 100 kg N ha^{-1} and potassium sulphate, 10 kg N ha^{-1}). In the current study, application occurred at constant amounts and there was still a significant reduction in pH in the intermediate treatments. In addition, the pH in the litter under *P. radiata*, sampled four years after the final fertilisation had significantly decreased due to N application.

Results from research on the effects of various types and rates of N fertilisers on soil pH have varied. Minor changes in pH of 0.2 to 0.3 units have been observed due to large and or long-term applications of urea and ammonium nitrate (Nohrstedt, 1990;

Homann *et al.*, 2001). However, Nohrstedt (1990) noted that ammonium nitrate application did not result in any clear and persistent signs of soil acidification, nitrate leaching or change in base cations availability. Nohrstedt *et al.* (2000) also observed that 13 years after multiple applications of urea to Norway spruce *Picea abies* (L.), up to a total rate of 2400 kg N ha^{-1}, had no significant effect on pH measured in the humus and mineral soil (to a depth of 10 cm). These results contrast with the significant long-term changes observed in this study, where fertilisation every fourth year resulted in a pH reduction of 0.3 units throughout the profile (0-50 cm).

Soil profile analysis (Table 3.1c) indicated that annual fertilisation substantially increased soil acidity increasing the levels of exchangeable H^+ to a total depth of 50cm. Assuming all NH_4^+ added from the 1300 kg N ha^{-1} (ammonium sulphate) was converted to NO_3^-, a total of 378 kmol H^+ ha^{-1} would be produced. Uptake of mineral N was calculated in fertilised topsoil at a rate of 82 kg N ha^{-1} (Chapter 4), which is equivalent to the consumption of 5.6 kmol H^+ $ha^{-1}yr^{-1}$ or 72 kmol H^+ ha^{-1} in total over thirteen years. Comparing the fertilised and unfertilised Kurosol topsoils indicates that the cation exchange capacity increased from 16.9 to 23.9 meq 100 g^{-1}, while base saturation declined from 16% to 5% (Table 3.1c). The BS% reduction may have resulted from the leaching of cations in association with the added sulphate ions. If the increased cation exchange capacity was utlised by H^+ ions this could account for 114 kmol H^+ ha^{-1} of the available H^+, while the measured reduction in BS% would account for a further 137 H^+ ha^{-1} on the exchange complex. Therefore, the increase in CEC and the decrease in BS% alone could adsorb 67% of the H^+ added by ammonium sulphate fertilisation. These calculated estimates assume full nitrification of the ammonia ions after fertiliser addition, however, low to negligible amounts of nitrate were measured in soil solution and may account for the lower measured change in H^+ ion concentration on the exchange complex of 29 kmol H^+ ha^{-1} (ten-fold less than estimated based on full nitrification of added fertiliser).

In the Kurosol, high rates of N application significantly reduced exchangeable Mg by half throughout the entire profile. Such reductions were also observed in total Mg and Ca content, but due to large spatial variations these were not significant. The depletion of Mg in the profiles was most likely due to uptake by the forest crop and

transfer to the litter, rather than depletion by leaching, associated with N movement. Leaching of Mg and significant reductions of Mg in surface soils due to fertilisation with ammonium sulphate have been observed (Khanna *et al.*, 1992). Exchangeable magnesium is considered to be in low supply when less than 10 μe. g^{-1} soil or when it constitutes less than 6 % of the CEC, and is considered in plentiful supply at values greater than 30 μe. g^{-1} soil (Metson, 1974). These soils therefore have adequate Mg levels for growth. However, low Mg content in some acidic Tasmanian soils has been identified (Neilsen and Dredge, 1999), and given the dramatic reduction of Mg measured in this study any proposals to increase plantation production through fertiliser use need to address possible reduction in soil Mg content in future rotations. The low pH levels measured in these soils indicate that Mg deficiency could well arise. Low levels of foliar Mg and associated indications of deficiencies are now present on 2nd rotation sites on Kurosols in the area studied.

Base saturation was greatly reduced due to fertilisation. Annual N application decreased base saturation by one third throughout the entire profile. Many experiments in Europe and Northern America have shown decreases in exchangeable Ca and Mg in the soil profile. Such losses in base cations associated with N application and timber harvesting, have been linked to declines in forest health and productivity (Watmough and Dillon, 2003).

3.7.5 *E. regnans* plantation growth

Large increases in *E. regnans* stem volume growth due to N fertilisation in this study indicate that the site was limited by N availability. Volume growth at the highest rate of fertilisation was more than double that of the unfertilised. In contrast, there was no measurable response to P alone or when applied in combination with N. Eucalypt plantations response to later age N and P addition are variable and site specific (Cromer *et al.*, 1981; Weston *et al.*, 1991). Ward *et al.* (1985) only found responses in height growth to N and P in combination and no response to P applied alone. In contrast, N fertilising and thinning experiments in *E. regnans,* have shown only a small increase in diameter following urea application at 460 kg N ha^{-1} and no significant increase in volume (Messina, 1992). In the current study, the lack of

growth response to P application in the *E. regnans* plantation indicates that P was not limiting growth at this site. Crane (1978) also noted that response to P in the field were rare. When P is adequate, little or no response to P fertilisation are observed (Fisher and Garbett, 1980; Raison *et al.*, 1990; Raison and Myers, 1992). Ferrosols, due to their high levels of iron oxyhydroxide, have a very strong P fixing capacity (Smethurst *et al.*, 1998). The low pH of these soils between 4.5 and 3.5 and would also have increased the association of P with Fe and Al. The dolerite based soils in the Southern Forests in Tasmania can have P sorption maxima in the order of 3000 ppm. Available P levels, measured by methods such as dilute acid fluoride, are generally around 1 to 3 ppm. A paste extract developed by Mendham, *et al.* (2002) been found to be useful for Ferrosols on basalt, but it has not been fully evaluated for the dolerite based soils. In *P. radiata* plantation in south-eastern Australia, applications of at least 120 kg P ha^{-1} were required to increase growth on strongly fixing soils (Hopmans and Flinn, 1998). Turner and Lambert (1983) indicated that fast growing species of eucalypts remove less P than *P. radiata*. Baker and Attiwill (1985) also observed that *P. radiata* had a greater absolute requirement for P than Eucalyptus. Measures of available P have been shown to be weak indicators of potential growth in Australian Eucalyptus forests due to the activity of mycorrhizal fungi associations while pines do respond (Attiwill and Leeper, 1987). However, P responses have occurred in these soils with alternative Eucalyptus species (Paul Adams pers. com.).

Growth response to N and P fertiliser may depend on several other factors including pest control (Flinn *et al.*, 1979a). The low MAI of the most productive treatment in *E. regnans* in the current study could be partially due to continued insect attack by *Cyrsoptharta bimaculata* (Oliver) (Leon, 1989; Elliott *et al.*, 1993; Neilsen, 1996). These attacks have occurred frequently through the experiment (Leon, 1989). There is some evidence that high P treatments may have been more severely browsed, with significantly more leaf area removed (40 % compared to 25 %) in the treatments with lower P foliar concentration at age 12 years (W. Neilsen pers. com.). In Tasmania, reductions in growth due to insect defoliation of up to 44 % have been observed in 6-year-old *E. regnans* plantations (Elliott *et al.*, 1993). However, *E. regnans* have some of the highest reported productivity in Australia (> 30 m^3 $ha^{-1}yr^{-1}$, over 20-30 year

rotations) (Weston, 2001). The results indicate that N fertiliser response will occur despite this defoliation. However, some distortion of the results is possible.

3.7.6 *E. regnans* stand health and foliar nutrient concentration

This study showed no correlation of growth response with foliar N content. This is in agreement with the findings of Judd *et al.* (1996), but contrary to those of Ballard (1978). Even at applications of N totalling 2180 kg ha^{-1} during a period of six years, Birk and Turner (1992) observed little increase in foliar N concentrations. Numerous authors have associated variations in growth response and foliar nutrient concentrations due to patterns of rainfall soon after treatment (Heilman *et al.*, 1982; Nason *et al.*, 1990; Benson *et al.*, 1992b).

Accumulation of P in *E. regnans* foliage also suggested P was not the limiting factor. When limited by other factors, trees are capable of accumulating P, as inorganic P, beyond the immediate tree requirement (Bennett *et al.*, 1997). Evidence of such response was seen in the accumulation of P up to 300 percent in young *E. globulus* Labill. plantations (Hooda and Weston, 1999). Attiwill (1980) showed that in mature *E. obliqua,* 46 % of the demand for P was met by internal cycling. This efficient use of P by eucalypts could mean that P supplied in fertilisers would also be efficiently recycled and prolong any response. When N was applied, but not P, growth was as good as when both N and P were applied. This was despite low foliar P levels of 0.07 to 0.08% and an N/P ratio of 17, compared with foliar P levels of 0.16% and an N/P ratio of 8 after P fertilisation. This suggests that the requirements for P in *E. regnans* may be lower than indicated in other eucalypt species (Dell *et al.*, 2002). Hopmans and Flinn (1987) observed that growth of *P. radiata* was limited by P deficiency growing on strong P-fixing soils, as indicated by marginal P levels in foliage, even after the application of 240 kg P ha^{-1}.

3.7.7 Nutrient distribution in soil and O2 horizons

In this study, at the highest rate of fertilisation, N, P, S and Ca all accumulated within the O2 horizon. Nitrogen in the fertilised treatments was concentrated in the O2

horizon, the mass of N increased by seven fold due to annual fertilising, equivalent to over 60 % of the N applied. The thicker O2 horizon and higher N concentration in that layer combined to form a significant nutrient pool. This is in agreement with work in a range of forest stands that have shown an increase in litter production and N litter mass due to N fertilising (Hunter and Hoy, 1983; Nohrstedt, 1990; Theodorou and Bowen, 1990; Neilsen and Lynch, 1998; Maier and Kress, 2000). Decomposer organisms are more active and abundant on surfaces, which results in more activity in the finer O2 litter layer, than the O1, and an increase in the availability of inorganic nutrients at the interface between the soil and O2 horizon (Paustain *et al.*, 1997).

In a review of 19 experiments across the United States and Europe, Fenn *et al.* (1998) also notes the importance of soil retaining N, with the majority of labelled N applied in experiments being retained in the soil and litter layer. In contrast, in a warm temperate climate in New Zealand high rates of forest floor organic debris breakdown in *E. regnans* resulted in low amounts of accumulation of 4.7 to 11.0 t ha^{-1} (Frederick *et al.*, 1985), compared to the 102 t ha^{-1} in the annual fertilisation treatment in this study.

Accumulation of P in the litter occurred and was significant at the highest rate of N application, whether P was supplied independent of N or not. Phosphorus also accumulated in the A1 horizon. Turner and Lambert (1986) observed that a single application of P fertiliser (100 kg P ha^{-1}) to *P. radiata* 30 years previously resulted in P accumulation in the litter, which doubled as did Ca, Mg and K, together with a three-fold increase in litter mass. The accumulation of N and P in the litter results in a long-term increase in productivity (Type II response) at this site from multiple fertiliser application.

3.7.8 Changes in cations and pH under *E. regnans*

Changes in the soil pH occurred after long-term N additions as ammonium sulphate. At the highest application rates soil pH declined throughout the profile (0-50 cm) by greater than 0.6 pH units. Generally soil pH declines increased with the rate of N application. In contrast to the Kurosol, declines in soil pH were most pronounced at the 0 to 30 cm depth (0.9 pH units) in the annually fertilised soil. Although there was

a substantial pH decline of 0.6 units in litter fertilised at the highest rate, there was no significant difference between the unfertilised and intermediate fertilisation rates. The reduced pH decline at this site may be associated with an increased capacity for base cation cycling in the tree and litter systems. The addition of plant residues leads to an initial increase in soil pH through the association of organic anions and the biological oxidation of these anions to CO_2 (Paul *et al.*, 2003a). This Ferrosol soil also had a higher clay content and CEC and hence buffering capacity than the Kurosol soil. Bromfield *et al.* (1983) noted that soil parent material, soil texture and hence the buffering capacity changes the rate of pH decline and tends to decrease in the following order: granite > sedimentary >basalt. This was evident in these soils where the rate of pH decline was similar in the annually fertilised soils from both sites, while intermediate fertiliser rates did not affect the more highly buffered Ferrosol.

The rate and prominence of pH change has previously been noted to be extremely variable, dependent on the site and rate and type of fertilisation. The application of urea can effect the pH of agricultural soils as strongly as ammonium sulphate and ammonium nitrate (Porter *et al.*, 1995). In comparison, a range of sites in Tasmania where fertiliser was applied were examined. In an *E. globulus* plantation in northern Tasmania, on a Yellow Kurosol formed on Silurian-Devonian siltstone, the highest rate of N fertiliser, applied as ammonium sulphate, dropped pH throughout soil to a depth of 50 cm by 1 pH unit (Bill Neilsen pers. com.). In another research area in northern Tasmania, on a Yellow Kurosol formed on Precambrian sandstone, application of triple superphosphate at the rate of 70 kg P ha^{-1}, or application of urea for three years at the rate of 100 kg N ha^{-1} yr^{-1}, had no effect on soil pH. However, the application of the two combined significantly reduced soil pH by 0.2 of a unit to 50 cm depth. Doubling the rate of urea for three years (200 kg N ha^{-1} yr^{-1}) significantly reduced soil pH by 0.33 of a unit (Paul Adams pers. com.). On a similar soil to the *P. radiata* site, fertilising with 1346 kg N ha^{-1} during 12 years, mainly as urea, did not significantly reduce pH (Neilsen *et al.*, 1992). It appears that urea, at moderate application rates, could have a lesser impact on soil pH than ammonium sulphate.

Phosphorus treatments also had significantly lower soil pH, compared to NIL. However, there was no cumulative effect of N and P fertiliser in combination. This is contrary to research in agriculture where generally pH decline in cropping soils is not associated with application of P fertilisers (McGarity and Storrier, 1986). For example, in an agricultural cropping soil, application of superphosphate at similar rates to this study (45 to 60 kg ha^{-1} yr^{-1} during 7 years) in general had no effect on pH in the top 7.5 cm of agricultural soil (Manoharan *et al.*, 1995).

Reductions in exchangeable Mg due to fertilisation were significant and in the surface soil there was a depletion of exchangeable Mg by over one third. This was not associated with a significant decline in total Mg content. Base saturation was also greatly reduced due to annual fertilisation by one third in the A1 horizon.

The ample supply of P and Ca through regular superphosphate applications at this site produced an excess in some treatments. Even when applied in excess at this relatively high rainfall site there was no evidence of leaching of these elements. In contrast, the application of N alone significantly reduced the total Ca available in the top 50 cm of soil, compared to the NIL. High rates of N application to the Ferrosol generally doubled exchangeable Al throughout the profile. Below a pH of 4.2 Al is more rapidly released into the soil solution due to aluminium hydroxide buffering more acidic solutions (Matson *et al.*, 2002). Increased Al availability results in inhibited root growth and reduced uptake of Ca (Flinn *et al.*, 1979b). This may be partially responsible for the lack of treatment response in foliar and litter Ca concentrations.

3.8 Conclusion

Plantation productivity can be increased substantially through periodic fertilisation. This study has shown substantial responses to N fertiliser, applied as ammonium sulphate at two diverse sites,

- Volume growth of both stands doubled with annual applications of N fertiliser at 100 kg N ha^{-1} yr^{-1} (totalling 1300 kg N ha^{-1}).

- Although applications every second and fourth year produced less response, these treatments had better fertiliser-use efficiency. In both plantations fertiliser application every second year produced similar growth to annual fertiliser application once 300 kg of N had been applied.
- Two applications of P fertiliser, as superphosphate, totalling 144 kg ha^{-1} produced substantial increases in *P. radiata* volume growth.
- In contrast, applications every second year of N and P together to *E. regnans* resulted in no volume growth difference when compared to N only. This may relate to several edaphic factors or to the Eucalypts capacity to scavenge P from a deep P-fixing soil. Eucalyptus species have a lower absolute requirement for P than *P .radiata* (Baker and Attiwill, 1985).

Nutritional management of plantations requires information on soil and site characteristics, and clear guidelines on where fertilisers are needed, when to apply them, what products to apply and at what rates. While information of this type is available, much of it is restricted and not reliable across sites and regions. The proposition to use fertilisers to lift the performance of plantations on soils of low productivity, as well as the productivity of many satisfactorily performing plantations, needs to be carefully monitored. The effects of fertilisers on soil pH are site specific and varied, but are substantial and progressive. This was seen at both sites with,

- Significant reductions in soil pH at both sites, of about 0.6 of a unit, associated with the highest rates of fertilising. In the Ferrosol the greatest reduction was at the surface, while the greatest reduction was 20 to 50 cm in the Kurosol. In the Ferrosol reductions also occurred with both nitrogenous and phosphatic fertilisers.
- At both sites the reduction in active acidity was matched by reductions reserve acidity as indicated by lower base saturation in high N treatments.
- Decline in pH was linear with quantity of fertiliser applied, although, in some cases, low rates had no effect. The Ferrosol soil also had a higher clay content and CEC and hence buffering capacity than the Kurosol soil.

- Substantial soil pH changes of 0.5 to 1.0 units throughout the soil profile pose serious long-term consequences for productivity directly and through reductions in exchangeable bases. Further evaluation of types of fertiliser and rates of application used in forestry will be needed to select those that will have no impact or at least minimise change. Some forest soils are particularly vulnerable to change with initial low pH and low buffering capacity as seen in the Kurosol profile.

Long-term fertilisations altered the distribution of nutrients in the soil profile with concentration of some nutrients increasing in litter and surface soil horizons. However reductions in base saturation also highlight cation leaching and associated acidification. Reductions in exchangeable Mg were significant at both sites and the decline was associated with lower pH changes in these soils. Under *P. radiata* high rates of N fertilisation reduced exchangeable Mg concentrations by half over the entire profile. The research highlights the balance needed between obtaining growth response and causing detrimental soil effects due to long-term fertiliser application.

Increased N cycling retained in the tree, forest floor and surface soil is important when considering the management of sites for further plantation establishment. Removal of the litter layer and any surface soil due to clearing or burning could result in substantial reductions in the available nutrient pool. Replacement of these nutrients through long-term fertiliser additions could potentially cause significant pH decline over successive rotations. The role of long-term N fertilisation on the availability of mineral N for tree growth is investigated in the following Chapters. The important role of the litter layer in cycling nutrients in a forest system and options for minimising detrimental effects of fertilising on soil are discussed in Chapters 6, 7 and 9.

Chapter 4. *In situ* rates of net nitrogen mineralisation in two Tasmanian forests after 13 years of annual fertilisation

4.1 Introduction

Productivity is often limited in temperate forest ecosystems by low N mineralisation rates, which has led to N fertilisation programs in many countries including; New Zealand, Sweden, Canada, the United States of America, Australia and Portugal (Thomas and Mead, 1992a). Application of N fertiliser at rates between 100 and 300 kg ha^{-1} are common in commercial plantations and can contribute substantially to plant-available N (Johnson *et al.*, 1980). Although the economic costs of fertilisation are often met by the increase in growth, overall recovery of N is commonly less than 30 precent (Mead and Pritchett, 1975a; Heilman *et al.*, 1982). Nitrogen fertilisation increases plant-available N in topsoils for only a short period of time, even at rates as high as 600 kg N ha^{-1}, available N concentrations decline to pre-fertilised within two years (Smethurst *et al.*, 2001). In contrast to the relatively short period of N enhancement, plantation growth rate increases occur due to fertilisation for a period of up to ten years (Hunter and Hoy, 1983; Fife and Nambiar, 1997; Neilsen and Lynch, 1998).

Reports on long-term affects of fertilisation on N cycling in forest soil and subsequent N mineralisation vary greatly. Inconsistencies often reflect variations in site characteristic and fertiliser-application management (Aggangan *et al.*, 1998) and limitations when measuring NNM (Adams *et al.*, 1989b). However, increased NNM in forest soil due to N fertilisation have been measured both during laboratory and field incubations (Johnson *et al.*, 1980; Hingston, 1984; Aarnio and Martikainen, 1995), up to six years after application (Smolander *et al.*, 1998).

Rates of NNM in temperate forest are often low (Adams *et al.*, 1989b). In Tasmania annual rates of NNM in mature eucalypt forests range between 16 and 51 kg N ha^{-1}, and in young eucalypt plantations between 13 and 188 kg N ha^{-1} (Wang *et al.*, 1998; Moroni *et al.*, 2002). In *P. radiata* plantations growing in southern Australia Carlyle

et al. (1998a) observed *in situ* NNM rates between 16 and 74 kg N ha^{-1} yr^{-1}, while Theodorou and Bowen (1983a) estimated rates around 50 kg N ha^{-1} yr^{-1}.

Effects of long-term N fertilisation on N mineralisation rates in the Tasmanian climate have not been studied. In this study, the effect of long-term N or N plus P fertilisation on *in situ* rates of NNM in underlying Ferrosol and Kurosol topsoils were examined. Seasonal variations at these sites were also examined to determine maximum and minimum rates of NNM and establish the optimum sampling times to assess NNM in these environments. Annual rates and seasonal variability in NNM were measured using *in situ* cores.

4.2 Methods

4.2.1 Site selection

Site and soil profile descriptions are provided in detail in Chapter 3 (Section 3.2). Therefore, a limited summary of these details is given here. Nitrogen mineralisation studies were conducted in the two plantations studied in Chapter 3, as they provide contrasting plantation species and soils, and a large body of background information on plantation growth and health during the thirteen-years of fertilisation. One site was located in the north-east of Tasmania and the other in the south (Map 3.1). The climate at both sites was cool temperate, characterised by cool, wet winters, and warm dry summers. Rainfall in the north-east was spread evenly through the year with a winter bias. However, rainfall was highly variable between years. Annual rainfall was obtained using a stand gauge located near the site, at Evercreech, from 1963 to 1986. The local Bureau of Meteorology (BOM) station at Fingal highlights the high variability in rainfall between years ranging from 374 up to 750 mm, annually (Figure 4.1). However, the rainfall average at Fingal weather station was considered lower than that at the plantation. Rainfall at the southern site was more uniform. Soil and profile descriptions were provided in detail in Chapter 3 (Section 3.2). A summary of site details is given in Table 4.1.

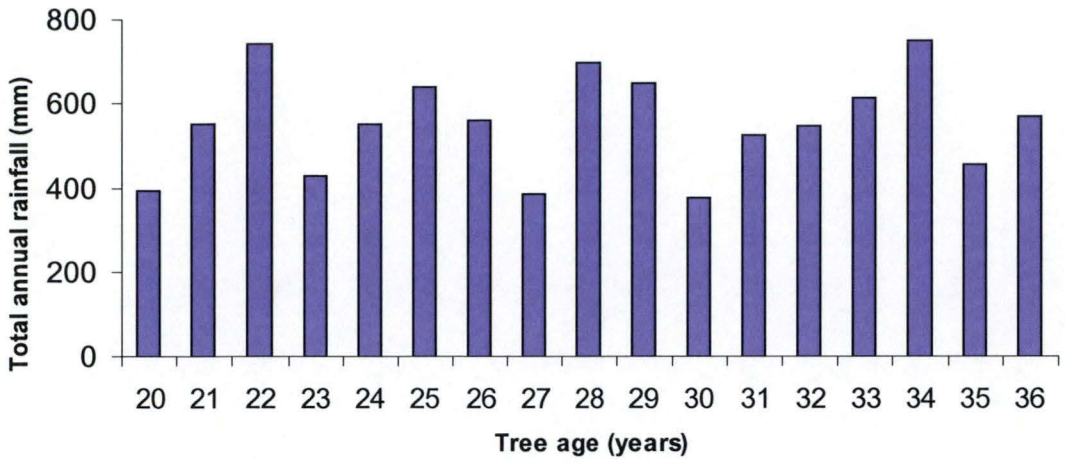


Figure 4.1 Variation in annual rainfall, for the north-east during the period of the fertilisation experiment, measured at the nearest Bureau of Meteorology (BOM) weather station at Fingal, Latitude 41°64' S, Longitude 147° 96' E.

All fertiliser treatment plots studied at both sites were slashed with a brush-cutter to remove understorey weeds one week prior to commencing *in situ* incubations (June 1999). Understorey in the *P. radiata* plantation consisted predominantly of bracken (*Pteridium esculentum* (Forst.f.)) and the highest proportion was in the unfertilised plots. The *E. regnans* understorey was also predominantly bracken, and included some small Blackwood (*Acacia melanoxylon* R. Br.) and cutting grass species. At this site, all the slash was removed to the outside of the plot buffers. In addition, the blackwood stumps were marked with paint so that these areas could be avoided during soil and litter sampling. Understorey species were removed to aid *in situ* technique and reduce the variability across the site, as some understorey species were N fixers. Slash was removed from the plots to ensure a homogeneous soil surface and no confounding of erratic N supply by decomposing slash. Soil sampling predominantly occurred between cultivated tree rows.

Table 4.1 Site description summary

Location	North-east	South
Soil	Yellow Kurosol	Brown Ferrosol
Altitude	350 m	100 m
Aspect and slope	Easterly, 10 %	South-easterly, 10 %
Annual rainfall	938 mm	1200 mm
Species	*P. radiata*	*E. regnans*
Year planted	1967	1981
Experiment established	20-year-old plantation	5-year-old plantation
Final fertiliser application	June 1999	May 1999
Fertiliser treatments	Unfertilised, NIL	Unfertilised, NIL
(studied in this chapter)	Fertilised, (P)N1Y	Fertilised, P1YN1Y
N fertiliser applied	Ammonium sulphate (annually)	Ammonium sulphate (annually)
	100 kg N ha^{-1}	100 kg N ha^{-1}
P fertiliser applied	Superphosphate (twice)	Superphosphate (annually)
	72 kg P ha^{-1}	46 kg P ha^{-1}

At both sites, each fertiliser treatment plot was sub-divided into two sub-plots. Sub-plots were treated separately throughout the *in situ* study. Soil samples from each sub-plot were processed separately in the laboratory, to measure variability between the fertiliser treatment plots. Figure 4.2 and 4.3 is a summary diagram of the site selection, sub-plot selection and *in situ* sampling points, for the *P. radiata* and *E. regnans* experiments, respectively.

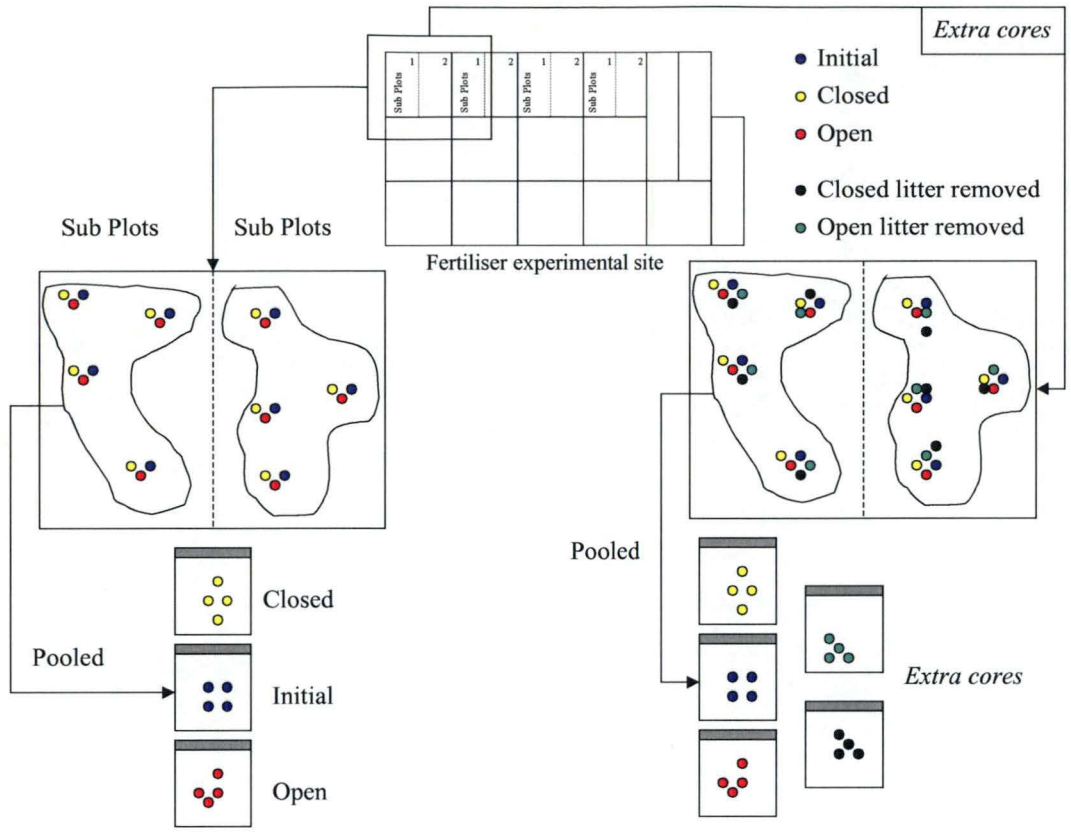


Figure 4.2 *P. radiata* experimental site, sub-plots, sampling points and cores.

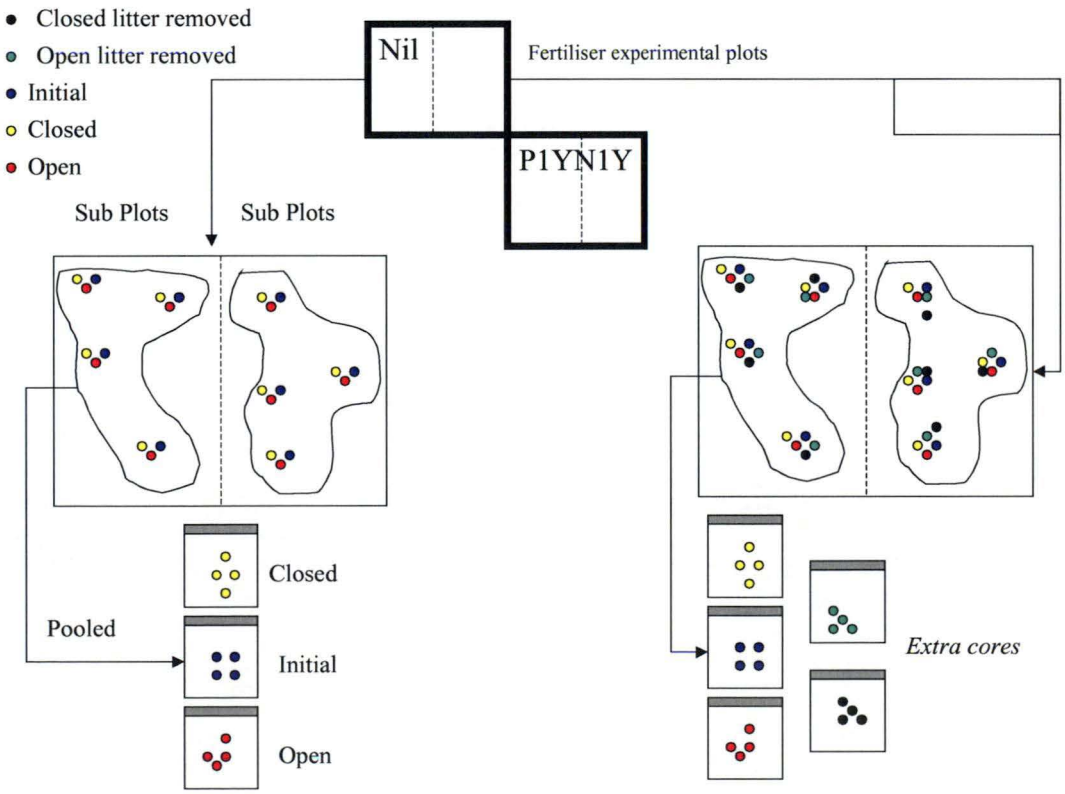


Figure 4.3 *E. regnans* experimental plots, sub-plots, sampling points and cores.

4.2.2 *In situ* core methods

This field study on N mineralisation was established using a modification of the *in situ* core methods from Raison *et al.* (1987). Although Raison *et al.* (1987) used steel cores, I used unperforated PVC cores. PVC cores are cheaper to make and easier to transport. Cores were unperforated to prevent loss of ammonium and nitrate by mass flow and diffusion, as suggested by Hart *et al.* (1994).

Field estimates of NNM using *in situ* cores maintain the natural environmental and temperature fluctuations with as little disturbance to the soil moisture content as possible (except when capped). In summary, soil isolation in PVC pipes was used to estimate NNM from the change in mineral N (NH_4^+ and NO_3^-) during the incubation period. PVC pipes prevent the uptake of soil N by plant roots, thereby limiting N processes to microbial communities in enclosed soil. This experiment also prevented N leaching in some cores by excluding rainfall, via caps on the pipes. The use of both open and closed (capped) cores allowed simultaneous measurements of NNM, N leaching and N uptake.

Cores consisted of 5 cm diameter PVC pipe cut to lengths of 12 or 17 cm (Photo 4.1). The longer pipes allowed for the additional depth of the litter horizon. Individual pipes were marked at 10 or 15 cm to gauge penetration depth on insertion into the soil. Pipes marked at 10 cm were for incubating mineral soil only, and those marked at 15 cm were used when litter horizons were also incubated. In cores marked at 15 cm, only the soil to a depth of 10 cm was isolated. This depth corresponded to the depth of Kurosol and Ferrosol topsoils. At the top of the PVC pipe two holes were drilled to aid removal. The base of each pipe was tapered to aid insertion and minimise soil disturbance.

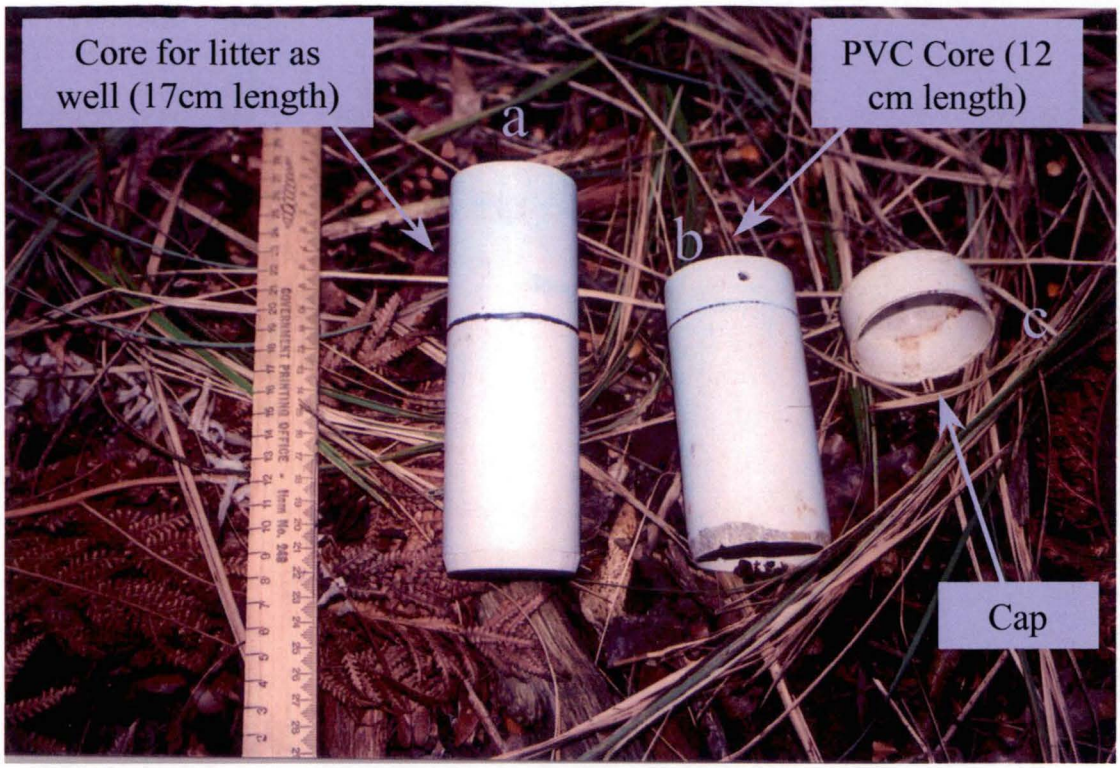


Photo 4.1 Shows 5 cm diameter PVC pipe cut to lengths of 17 cm (a) and 12 cm (b) core, and cap (c) used to cover closed cores during sampling.

Previous *in situ* incubation periods of between 7 and 90 days, have been recommended (Raison *et al.*, 1987; Adams *et al.*, 1989b; Smethurst and Nambiar, 1989b; Goncalves and Carlyle, 1994; Carlyle *et al.*, 1998b). Raison *et al.* (1987) observed NNM rates in cores increased linearly with time, resulting in an appropriate containment period between 30 and 90 days. In this study two-monthly containment periods were chosen as rates of NNM in both soils were expected to be low. In order to examine the seasonal variations in N fluxes at both sites, two-monthly incubations were replicated during an 18-month period (July 1999 to January 2001).

For each sampling point, a minimum of three cores were required, labelled initial, open or closed. At each sampling point, individual cores were set approximately 5 cm apart. The sampling cores were prepared by removing the litter plugs first. The plugs were cut out of the litter horizon using a PVC pipe to the depth of the mineral soil. Once these plugs had been removed, the surrounding litter at the sampling point was removed to the mineral soil interface. This was done to prevent litter entering the mineral soil during pipe insertion.

An initial core of mineral soil was extracted by hammering a PVC pipe to a depth of 10 cm into the soil. The PVC pipe was then removed along with mineral soil. The mineral soil was then pushed out of the pipe and placed in a plastic bag. For the open core, a 17 cm PVC pipe was hammered into the soil to a depth of 10 cm. One of the litter plugs removed earlier was then placed on top of the mineral soil inside. Closed cores were installed in the same manner as open cores, except a PVC cap was placed on top of the protruding pipe to prevent rainfall and leaching in the soil core (Photo 4.2). The remaining litter was placed back around the cores, which remained in the ground for the length of the incubation period.

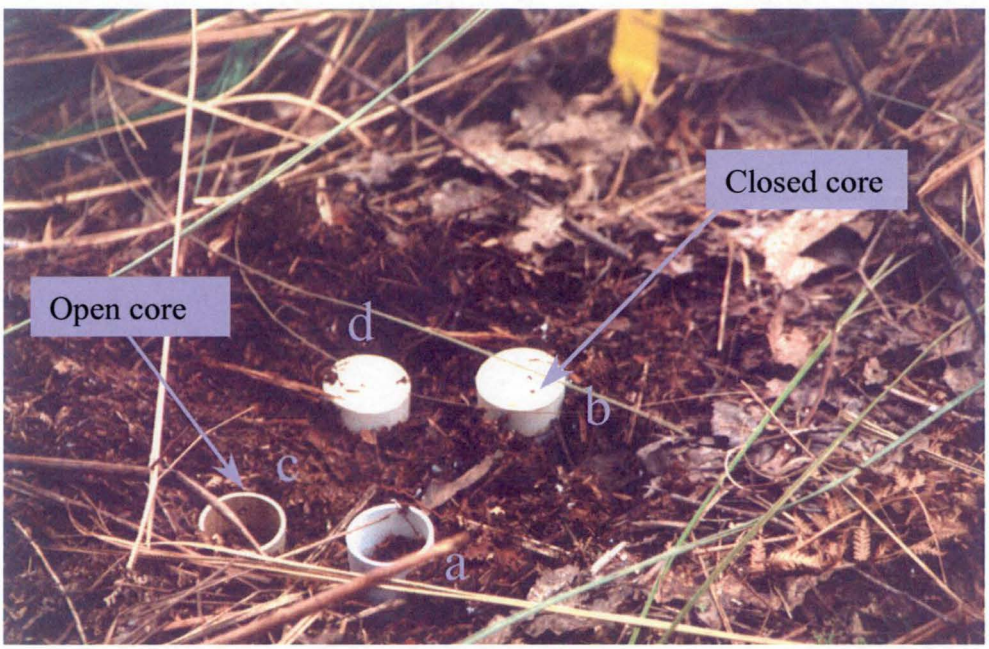


Photo 4.2 Shows a sampling point where all four cores were incubated for two months (a) open with litter, (b) closed with litter, (c) open without litter, (d) closed without litter.

Samples were taken at two-month intervals for 18-months. At the end of each two-month incubation, open and closed cores were removed. On the same day, a new sample point was selected and a new set of cores was added. Four sub-samples were taken from each treatment and sub-plot (Figure 4.2 and 4.3). These four sub-samples (per sub-plot) were pooled into a single plastic bag. Only mineral soil was kept and litter plugs were discarded. The pooled treatment samples were then placed in a "cooler bag" for transport to the laboratory.

Extra cores were added to the sampling points, periodically to determine how the litter horizon influenced N fluxes during incubation. Extra open and closed cores were added during September, July and May. These cores were incubated without the litter plug on top and were called 'open without litter' and 'closed without litter' (Figure 4.2 and 4.3). Hence, during these sampling periods each sampling point had four cores left for the two-month incubation (as shown in Photo 4.2).

4.2.3 Laboratory procedures

All samples were transported to the laboratory within approximately six hours and refrigerated at 2-4 °C until processed. Storage time was kept as short as possible, with a maximum of three days passing before all samples were ready for mineral N extraction.

Each pooled soil sample (an individual bag containing soil from four pooled cores) was processed as follows;

- Samples were thoroughly mixed in a plastic bag, manually dispersing individual soil cores completely, until a friable homogenised soil sample was formed.
- From this homogenised soil, a small sub-sample was taken (~ 50 g), weighed, oven dried at 105 °C for 24 hours, and then reweighed. This oven-dried weight was used to calculate the gravimetric field moisture content at the time of sampling. The oven-dried soil was then heated to 600 °C for four hours and reweighed, to estimate the organic matter content of the soil (Herbert *et al.*, 1995).

From the homogenised soil a large sub-sample was passed through a 2 mm sieve. The < 2mm fraction was then used to determine soil pH, mineral N concentration, and organic C content.

- pH was determined in a 1:5 ratio of soil (< 2mm): distilled water.
- organic carbon was determined by loss on ignition.
- Oven dried weight and loss on ignition was calculated for < 2mm soil as per above.

Mineral N was extracted from fresh < 2mm homogenised soil using 2 *M* KCl (Rayment and Higginson, 1992);

- Duplicate 10 g < 2mm soil samples were shaken in 50 ml of cold 2 *M* KCl for one hour.
- The suspension was then filtered through Whatman No 42 papers to provide clear extracts. All extracts were stored frozen.
- Extracts were defrosted and analysed for nitrate and ammonium using a flow injection analyser (FIA) (Lachat Instruments).

Concentrations of mineral N were corrected using moisture content in the < 2 mm soil faction determined earlier. Topsoil mass per hectare was calculated using the average bulk densities (of replicate treatment plots) determined in Chapter 3. The average bulk density across both treatment plots in the Kurosol topsoil was 1.625 g cc^{-1}, and in the Ferrosol it was 1.26 g cc^{-1}.

4.2.4 Calculations and statistical analysis

Calculations of N mineralisation, leaching and uptake using the *in situ* core methods have been described in detail by Adams *et al.* (1989b). Summaries of the principles are given here.

Net N mineralisation was calculated as the change in ammonium and nitrate values between the soil sampled at the start of each incubation (initial soil core) and that measured in the closed soil core at the end of each incubation period (two months) (Equation 4.1).

$$Nm = Nc(t+1)-Ni(t) \qquad (4.1)$$

Where,
Nm = N mineralisation (ammonium plus nitrate)
t = time
t+1 = time at the end of incubations
Nc = closed soil core mineral N concentration
Ni = initial soil core mineral N concentration

Assuming rates of N mineralisation in the open or closed soil cores are the same, uptake of N by vegetation can be calculated. Nitrogen uptake was calculated as the change in mineral N measured in closed soil cores at the end of the incubation period minus the amount of mineral N measured in initial soil cores in the same time period (Equation 4.2).

$$Nu = Nc(t+1) - \{Ni(t+1) - Ni(t)\} \quad (4.2)$$

Where, Nu = N uptake

Assuming inputs of N due to rainfall are negligible, by comparing the amount of mineral N measured in closed soil cores to that in open cores, the amount of leaching of N during the incubation period could also be measured (Equation 4.3).

$$Nl = No\ (t+1) - Nc\ (t+1)$$

Where, Nl = N leaching

No = open soil core mineral N concentration

Results were analysed using a multiple analysis of variance (MANOVA) procedure of the GenStat software (Genstat 5 Committee, 1988). Means were compared using the treatment interactions (fertiliser and season) least significant differences ($p < 0.05$ and $p < 0.001$), as stated. No data transformation was required. Tests were validated by testing data for normality of distribution, and transforming data where required. Residuals from the model for each variable were examined for normality using diagnostic graphs.

4.3 Results

4.3.1 Kurosol topsoil

Mineral N concentration was enhanced in fertilised topsoil compared to unfertilised topsoil at the beginning of the study (fertilisation had occurred one month earlier) (Figure 4.4). Although fertilised topsoil had a significantly higher mineral N concentration than unfertilised during some measurements, differences between fertiliser treatments during all sampling times were not significant, due to the wide variations in mineral N concentration measured in the fertilised topsoil.

In both fertilised and unfertilised topsoil, rates of monthly NNM were generally low, reaching a maximum of 9 kg N $ha^{-1}month^{-1}$ in fertilised topsoil during the summer of 2000 (Figure 4.5). Immobilisation of N occurred in fertilised topsoil for the first six months of the study. The significantly higher immobilisation of N in fertilised topsoil during the first *in situ* core incubation was probably associated with the recent fertilisation. Beyond six months, NNM rates increased, and although not significantly different, rates were up to six times greater in fertilised topsoil than those unfertilised.

Ammonium was the dominant form of mineral N in both fertilised and unfertilised topsoils, with nitrate generally below detectable limits.

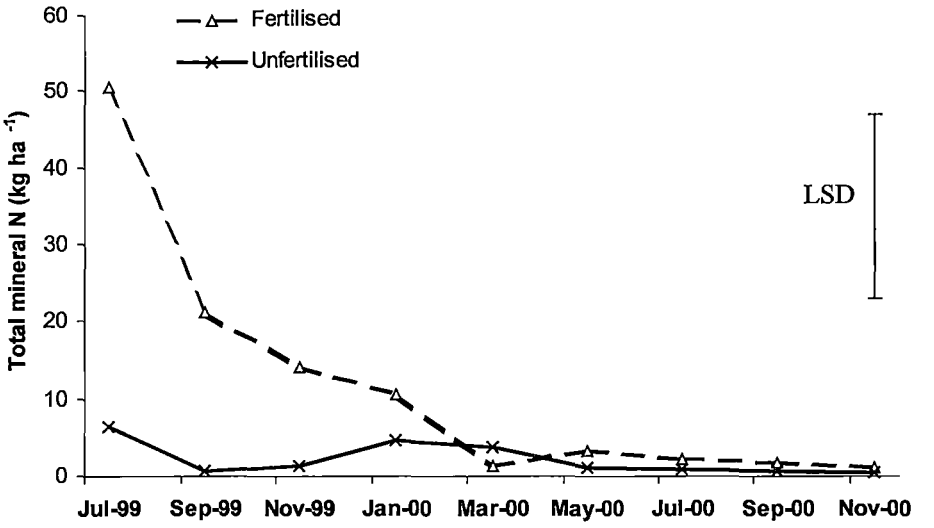


Figure 4.4 Mineral N (kg ha^{-1} in A1 horizon) during the 18-month field study (based on the successive initial core values). Bar indicates LSD of treatment by time, not significant at (p = 0.05).

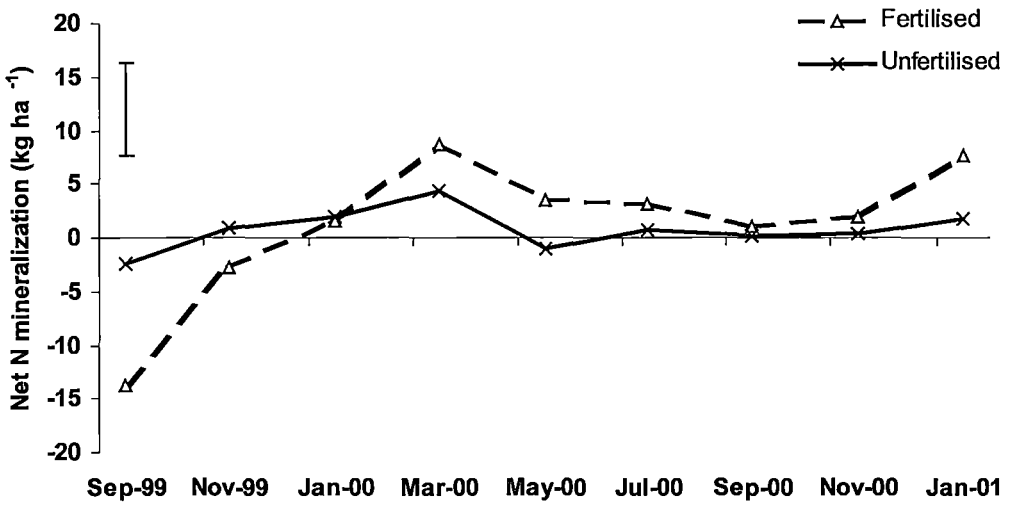


Figure 4.5 Net N mineralisation (kg ha^{-1} in A1 horizon) during the 18-month field study (based on closed minus initial cores). Bar indicates LSD of treatment by time, not significant at (p = 0.05).

Seasonal variability was observed in unfertilised topsoil as a summer peak in NNM of 4 kg N ha^{-1} month^{-1} (Figure 4.6). This peak was associated with a high rainfall event

in January, the time of soil containment. During this period, the moisture content was twice that of the previous spring and summer gravimetric moisture content of 22 % compared to approximately 10 % (Figure 4.7). The large enhancement of mineral N in fertilised topsoil for the first six months of the study obscured any clear seasonal trends. Rates of NNM were similar in fertilised and unfertilised topsoils during the last 12-months of the study.

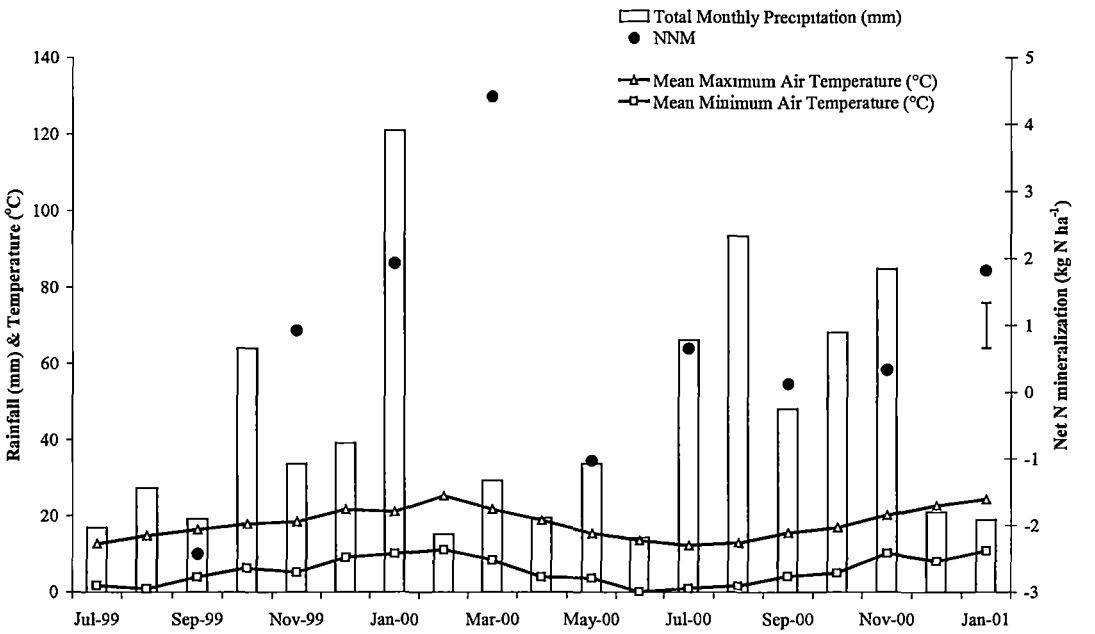


Figure 4.6 Rate of net N mineralisation in unfertilised topsoil compared to climatic data (rainfall and temperatures) from nearest Bureau of Meteorology (BoM) weather station at Fingal. Bar indicates LSD of NNM in unfertilised topsoil during the 18-month study.

During the 18-month study unfertilised topsoil had a significantly ($p < 0.05$) higher moisture content than that fertilised (Figure 4.7a). Unfertilised topsoil had an average moisture content of 24 % almost twice that measured in the fertilised topsoil at 13 %. Although open cores could have up to twice the moisture content of those closed, throughout the entire sampling period this was not significant (Figure 4.7b). Topsoil in open and closed cores also had similar trends in mineral N concentration throughout the experiment. Net N mineralisation was compared to the relative field water content (RFWC),

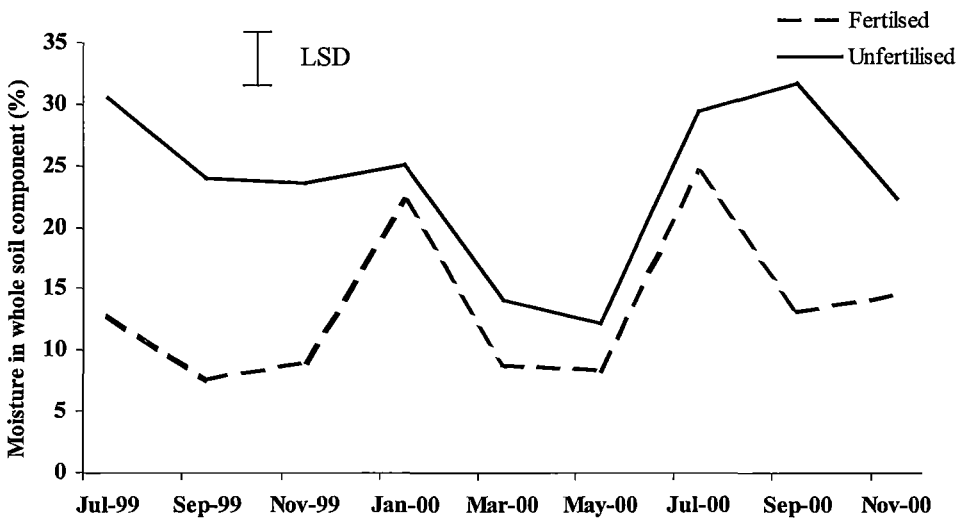
$RFWC = (\theta - \theta LL)/\theta_{UL}-\theta_{LL})$

θ = gravimetric water content θ_{LL}= Lower Limit

θ_{UL} = Upper Limit

The RFWC allows the comparison of a given water content to the lower and upper limits observed in the field. No trend between NNM and RFWC was observed in either fertilised or unfertilised topsoils (Figure 4.8).

a.



b.

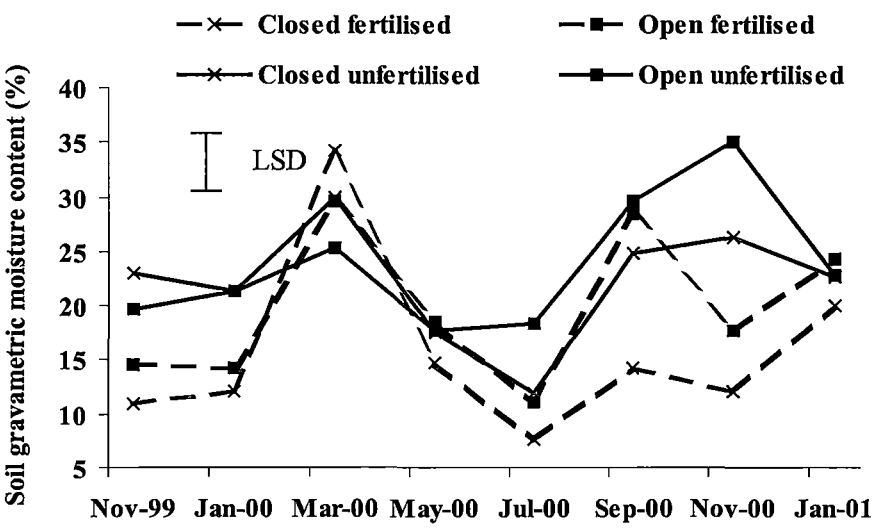


Figure 4.7 Gravimetric soil moisture content measured in topsoil from (a) initial cores (b) closed and open cores (% in A1 horizon). Bar indicates LSD across treatment and time.

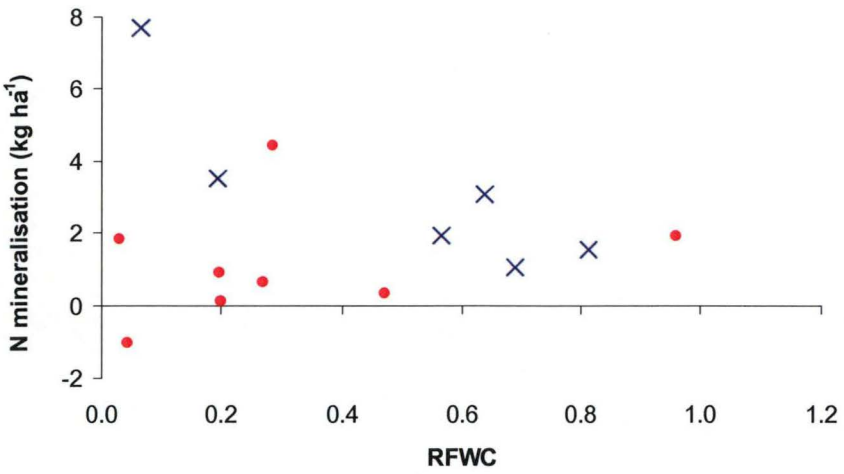


Figure 4.8 Comparison between monthly NNM kg N ha^{-1} and RFWC (relative field water content, in fertilised (circles) and unfertilised (crosses) topsoils.

An *in situ* temperature logger (Stowaway Titbit temperature logger) measured the temperature in the mineral soil at the depth of 5 cm. The logger was located under an even canopy in a plot fertilised every two years, representing the average canopy cover of the experimental site as a whole. The daily maximum soil temperature reached over a two-year period was 19.1 °C and the daily minimum was 5 °C. Average maximum and minimum temperatures over the two-year period are presented in Figure 4.9.

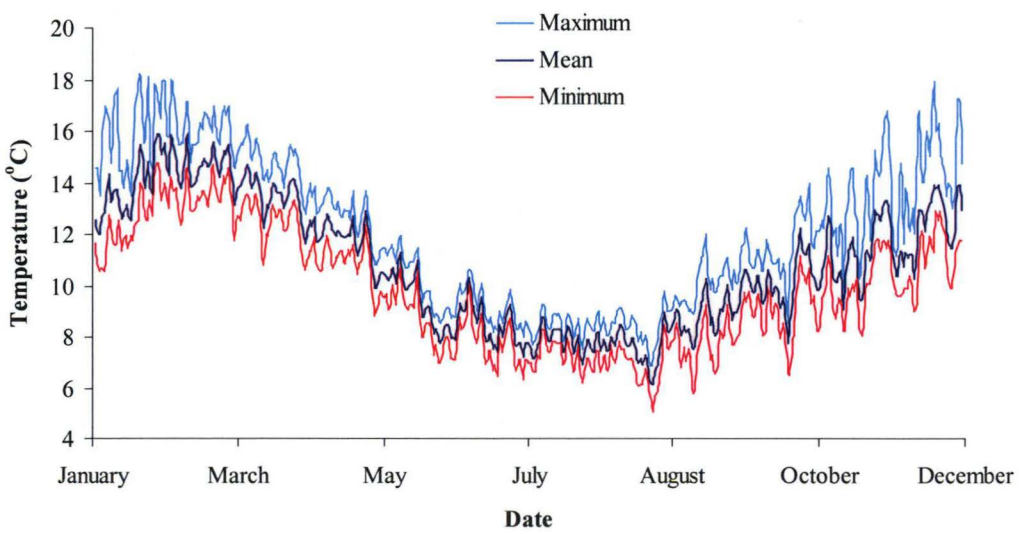


Figure 4.9 Average (over two years) minimum, maximum and mean temperatures (°C) from an *in situ* temperature logger, located in a (P)N2Y plot.

Fertiliser effects on NNM, N leaching and N uptake were compared for the final 12-months of the study (Table 4.2). These measurements examine the long-term affects of fertilisation. Inclusion of the first six months of sampling would have obscured any long-term effects of fertilisation due to the rapid decline in mineral N in recently fertilised topsoil. Fertiliser treatment effects on NNM were significant ($p = 0.045$), while there was no significant effect of fertilisation on N leaching or N uptake. The annual rates of NNM, N leaching and N uptake were calculated by summing *in situ* rates during the 12-month period (Table 4.3). Although there were large increases in the annual N mineralised, N leached and N uptake in fertilised vs unfertilised topsoils during this time, the effect of fertilisation was not significant.

Table 4.2 *In situ* net N mineralisation rate, and calculated N leaching and N uptake values (kg ha^{-1} $month^{-1}$ in A1 horizon).

		In situ period ending					
	Fertiliser treatment	March 2000	May 2000	July 2000	September 2000	November 2000	January 2001
Net N Mineralisation	Fertilised	8.6	3.5	3.1	1.0	1.9	7.7
	Unfertilised	4.4	-1.0	0.7	0.1	0.3	1.8
N leaching	Fertilised	8.7	1.4	0.2	-2.2	-1.6	1.9
	Unfertilised	3.8	-2.7	-0.5	-0.6	-0.8	0.5
N Uptake	Fertilised	18.4	3.3	5.2	2.4	3.1	8.7
	Unfertilised	7.0	2.2	1.4	0.7	0.8	2.2

Fertiliser treatment or time did not significantly effect N leaching. Fertiliser treatment significantly increased average NNM ($p= 0.045$) however, there was no significant fertiliser by time interaction.

Table 4.3 Calculated annual net N mineralisation, N leaching and N uptake (kg ha^{-1} in A1 horizon).

	Fertiliser treatment	Anuual 2000
Net N Mineralisation	Fertilised	51.7
	Unfertilised	12.6
N leaching	Fertilised	16.7
	Unfertilised	-0.4
N Uptake	Fertilised	82.2
	Unfertilised	28.6

Fertiliser treatment did not significantly effect annual N mineralisation, N leaching or N uptake.

4.3.2 Ferrosol topsoil

Fertiliser treatment had no significant effect on mineral N concentration (Figure 4.10), or NNM (Figure 4.11), during the 18-month study. Mineral N concentration and NNM varied widely between sampling times. At this site, *in situ* incubations began two months after the final fertiliser application. In contrast to the fertilised Kurosol, there was no pronounced increased mineral N concentration, or N immobilisation, during the initial sampling periods. Ammonium was the dominant form of mineral N in both fertilised and unfertilised topsoils, with nitrate generally below detectable limits.

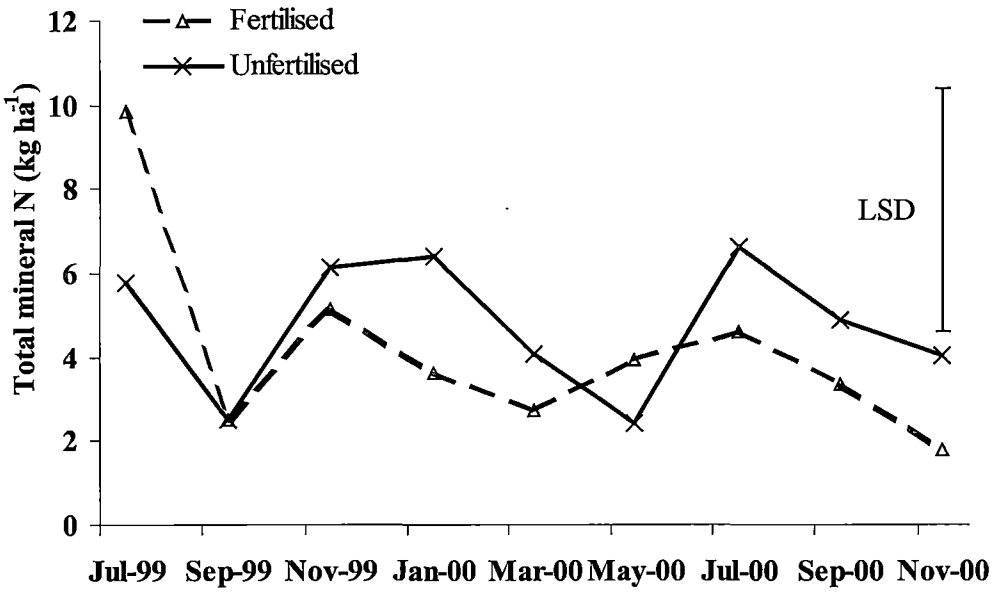


Figure 4.10 Mineral N (kg ha^{-1} in A1 horizon) during the 18-month field study (based on initial cores). Bar indicates LSD of treatment by time. There was no significant difference between fertilised and unfertilised treatments during the period studied.

Although positive at all times, rates of monthly NNM were low, reaching a maximum in unfertilised topsoil during March 2000 of 6 kg N ha^{-1} $month^{-1}$. Large temporal variability combined with high spatial variability (treatment replicates), resulted in no clear seasonal trends in mineral N concentration (Figure 4.10) or NNM (Figure 4.12).

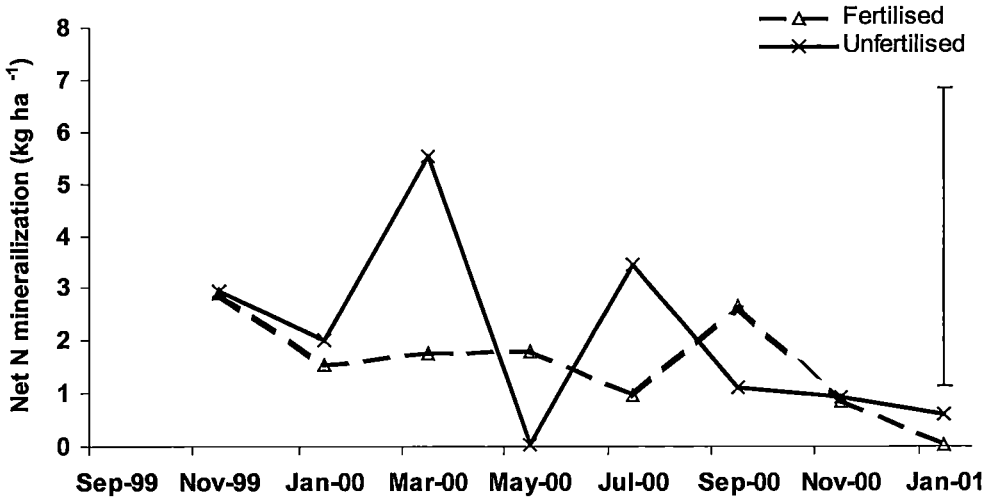


Figure 4.11 Net N mineralisation (kg ha^{-1} in A1 horizon) during the 18-month field study (based on closed minus initial cores). Bar indicates LSD of treatment by time.

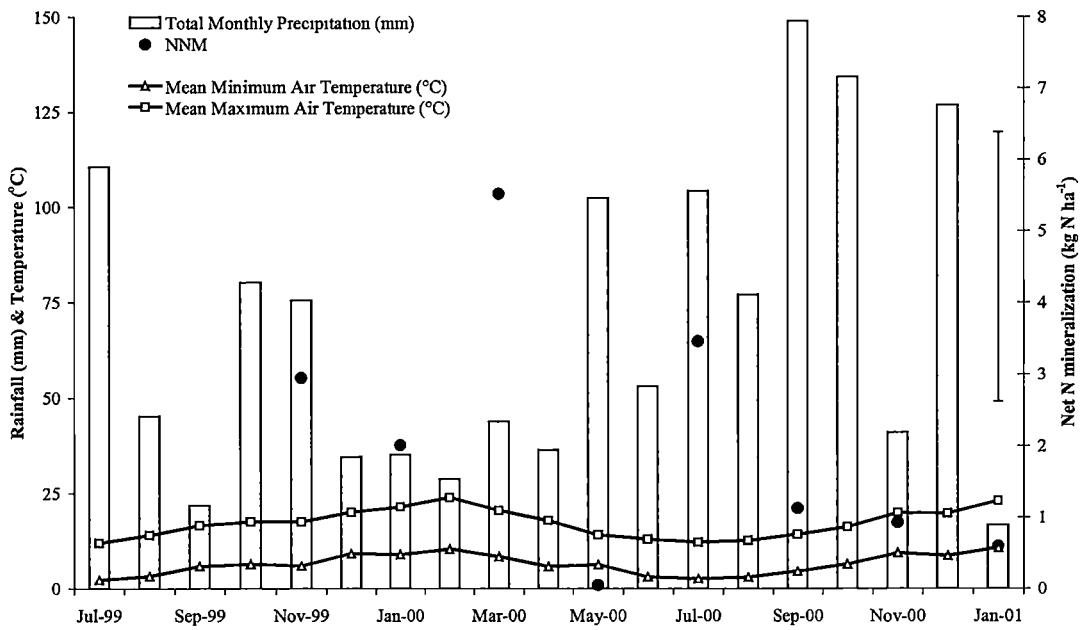


Figure 4.12 Rate of net N mineralisation in unfertilised topsoil compared to climatic data (rainfall and temperatures) from nearest Bureau of Meteorology (BoM) weather station at Latitude 43°31' S, Longitude 147° 02' E. Bar indicates LSD of NNM in unfertilised topsoil during the 18-month study.

Fertiliser effects on NNM, N leaching and N uptake were compared for the final 12-months of the study (Table 4.4). There was no significant effect of fertiliser treatment on NNM, or N leaching during this period. The highest rate of NNM occurred in unfertilised topsoil during March 2000 ($p = 0.058$), all other rates in both fertilised and unfertilised topsoil were similar. Fertiliser treatment did not significantly effect annual NNM, N leaching or N uptake (Table 4.5). However, the average monthly N uptake was significantly ($p = 0.011$) lower in the fertilised (2.5 kg N ha^{-1} $month^{-1}$) than the unfertilised (4.9 kg N ha^{-1} $month^{-1}$) topsoil.

Although an *in situ* temperature logger was placed in the mineral soil at this site the data obtained was corrupted. However, the empirical model SNAP has a submodel to predict daily average temperatures in three soil layers (Paul *et al.*, 2002). By defining the litter layer cover, depth and mass, weed cover, canopy leaf area index along with climatic data the sub model (STUF, soil temperature under forests) calculates the temperature in the mineral soil at 0-10 cm depth. SNAP requires daily weather inputs, in this study the climatic data was obtained from the bureau of meteorology as data derived from the interpolation of local meteorological stations (calculated to within 5 km). For comparison, the P1YN1Y and NIL treatment topsoil temperature was calculated (Figure 4.13). Inputs including soil bulk density, carbon, gravel and clay contents, as well as site parameters such as litter masses and depths, are described in Chapter 3. Leaf area index measurements were assessed at age 36 and were approximately 3 and 2.5, in fertilised and unfertilised trees respectively. These were assessed using both a visual guide (Cherry *et al.*, 2002) and the LICOR LAI2000 (Cherry *et al.*, 1998). Temperatures in the P1YN1Y topsoil ranged from 1.5 °C lower in summer to 0.8 °C higher in winter compared to those in the NIL treatment. The predicted maximum temperature reached 19.6 °C and the minimum was 5.8 °C, these are similar to those observed under *P. radiata*.

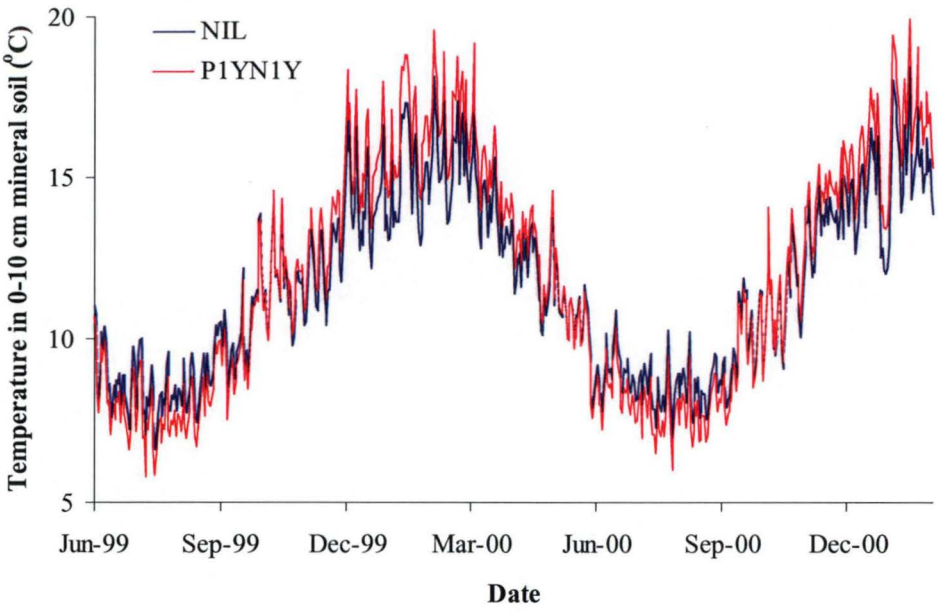


Figure 4.13 Average daily temperature (°C) of P1YN1Y and NIL treatment topsoils (0-10 cm) calculated by the submodel STUF (SNAP).

Table 4.4 *In situ* net N mineralisation rate, N leaching and N uptake (kg ha^{-1}month^{-1} in A1 horizon).

		In situ period ending					
	Fertiliser treatment	March 2000	May 2000	July 2000	September 2000	November 2000	January 2001
Net N Mineralisation	Fertilised	1.8	1.8	1.0	2.7	0.9	0.1
	Unfertilised	5.5	0.0	3.5	1.1	0.9	0.6
N leaching	Fertilised	-5.8	-3.9	-4.9	-2.0	0.4	-0.3
	Unfertilised	1.7	-4.3	-2.3	-1.1	-1.1	-0.4
N Uptake	Fertilised	3.7	1.8	1.6	3.9	2.8	1.4
	Unfertilised	9.9	2.9	2.5	5.3	3.8	4.6

Fertiliser treatment or time did not significantly effect N leaching or NNM. Fertiliser treatment significantly increased average N uptake ($p= 0.035$) however, there was no significant fertiliser by time interaction.

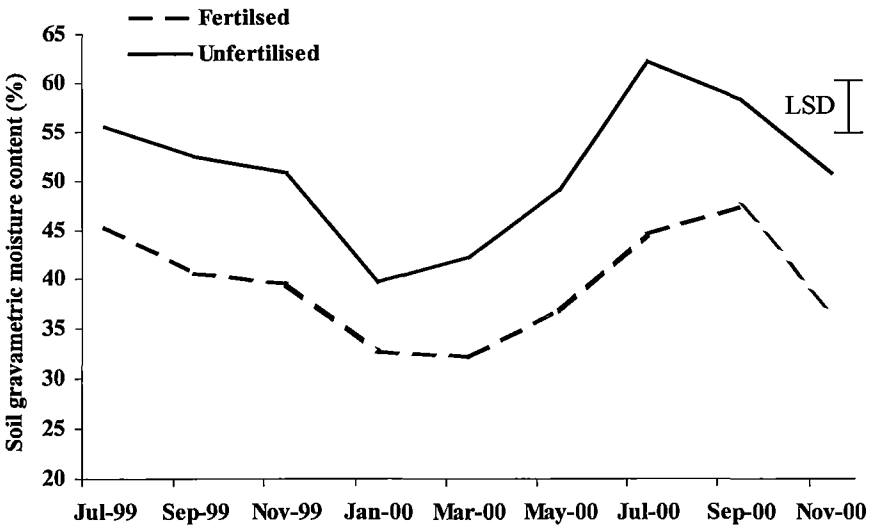
Table 4.5 Calculated annual net N mineralisation, N leaching and N uptake (kg ha^{-1} in A1 horizon).

	Fertiliser treatment	Anuual 2000
Net N Mineralisation	Fertilised	16.1
	Unfertilised	23.3
N leaching	Fertilised	-33.1
	Unfertilised	-15.2
N Uptake	Fertilised	30.6
	Unfertilised	58.2

Fertiliser treatment did not significantly effect annual N mineralisation, N leaching or N uptake. Note the negative leaching values probably relate to drier soil conditions in the closed cores restricting mineralisation with respect to the open cores.

Moisture content in unfertilised topsoil (initial cores) was significantly higher ($p < 0.001$) than that fertilised (Figure 4.14). Nitrogen leaching, calculated as the difference between closed and open soil cores, was generally negative, that is mineral N concentration were higher in open than closed cores. When calculating N leaching, it was assumed that the N inputs of rainfall were negligible, and rates of NNM between open and closed soil cores were the same. Negative N leaching would appear to be a factor of the different moisture conditions in the open vs closed cores. There was a clear trend of higher rates of NNM in open soil cores, which received rainfall. Closed soil cores resulted in a significantly lower ($p < 0.001$) moisture content than open soil cores (Figure 4.14b). Differences in moisture content of up to 15 % occurred during some sampling periods. Differences were often more pronounced in fertilised than unfertilised topsoil, however, there was no core by treatment interaction. No trend between NNM and RFWC was observed in either fertilised or unfertilised topsoils (Figure 4.15).

a.



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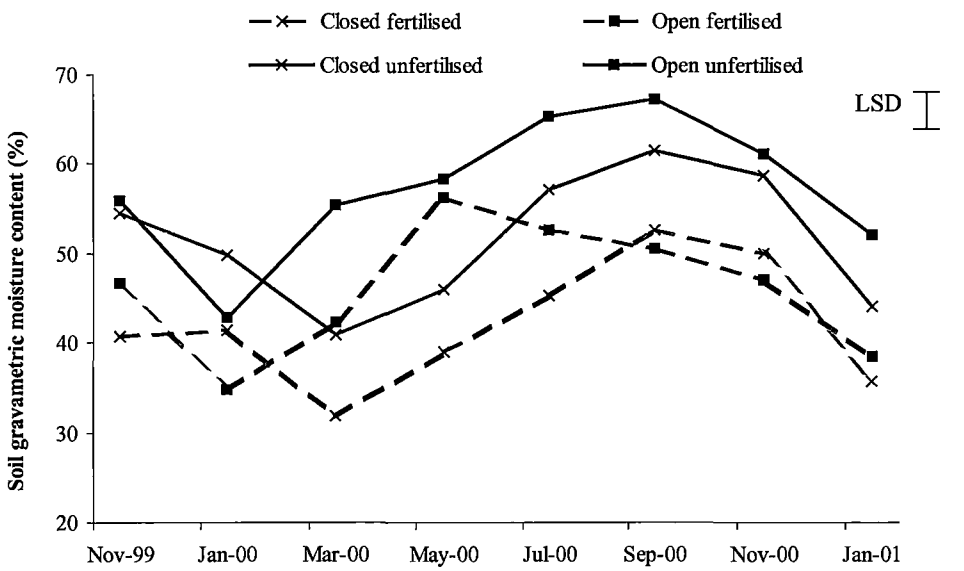


Figure 4.14 Gravimetric soil moisture content measured in topsoil from (a) initial cores (b) closed and open cores (% in A1 horizon). Bar indicates LSD across treatment and time.

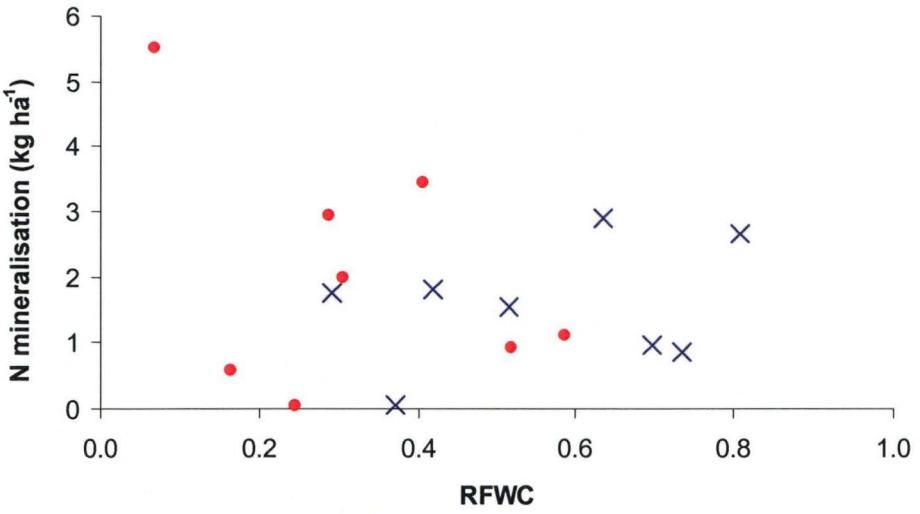


Figure 4.15 Comparison between monthly NNM kg N ha^{-1} and RFWC (relative field water content, in fertilised (circles) and unfertilised (crosses) topsoils.

4.4 Discussion

4.4.1 Kurosol topsoil

Low amounts of N mineralisation occurred in the Kurosol topsoil throughout the 18-month experiment. Net N mineralisation rates were 13 and 52 kg N ha^{-1} yr^{-1} in unfertilised and fertilised treatments, respectively. These measurements are similar to *in situ* NNM rates (between 16 and 74 kg N ha^{-1} yr^{-1}) observed by Carlyle *et al.* (1998a) under nine *P. radiata* sites in southern Australia. Such low rates of NNM, and high site variability, make it difficult to determine the influence of season on NNM in topsoils during the time period studied. However, NNM varied six-fold between the highest and lowest rates, occurring in March and May, respectively. Polglase *et al.* (1992a) also observed an order of magnitude greater mineralisation and nitrification rates measured in spring and summer, to those measured in winter and autumn. Raison *et al.* (1992) observed the highest NNM in spring (10 kg N ha^{-1} $month^{-1}$) and the lowest in winter (0.5 kg N ha^{-1} $month^{-1}$), with a distinct depression occurring in NNM when drought occurred.

Changes in short and long-term N availability due to N fertilisation were observed, but were often not significant. Immediately after fertilisation (one month) mineral N concentrations in topsoil were enhanced ten-fold, compared to unfertilised topsoil.

Elevated mineral N concentrations were evident for the first six months of the study, although this was only significant in the first month. The rapid decline in mineral N observed in the Kurosol have been observed extensively in previous fertilisation experiments, on a range of soils and sites (Williams, 1972; Johnson *et al.*, 1980; Fife and Nambiar, 1997; Smethurst *et al.*, 2001). The magnitude, and rate of decline have been related to the form, and frequency of fertilisation (Heilman, 1974; Strader and Binkley, 1989; Aggangan *et al.*, 1998). Rates of annual NNM were also larger, four times greater, in fertilised Kurosol topsoil than unfertilised, but this was not significant by MANOVA tests. Enhanced NNM in forest soil due to N fertilisation have been widely reported (Johnson *et al.*, 1980; Adams and Attiwill, 1991; Whynot and Weetman, 1991; Connell *et al.*, 1995; Aarnio *et al.*, 1996; Aggangan *et al.*, 1998; Smolander *et al.*, 2000). In the Biology Growth Experiment (BFG) experiment, Raison *et al.* (1992) observed significant increases of two to three-fold four years after fertilisation.

Fertilised Kurosol topsoil had a greater variation in NNM between treatment replicates (82 compared to 22 kg N ha^{-1} yr^{-1}) than between treatments (52 and 13 kg N ha^{-1} yr^{-1}), obscuring any possible treatment effect. N leaching and N uptake were also highly variable between replicates at 82 to 21 kg N ha^{-1} yr^{-1} and 128 to 37 kg N ha^{-1} yr^{-1}, respectively in the fertilised topsoil. The large variability in mineral N cycling between these treatment plots was also observed in later laboratory studies. Examination of the soil analysis, presented in Chapter 3, indicated that total N and P content of fertilised plots were similar (Table 3.8). The only nutrient that was clearly different between plots was Ca, both in total and exchangeable forms, the lower mineralising plot having twice the Ca of the higher mineralising plot. Such a variation would not explain the four-fold difference in NNM. Although higher Ca availability in this plot may have resulted in a potential for stronger immobilisation, thus reducing measured annual NNM rates, this was unlikely as the volume growth of trees on this plot was observed to be slightly higher throughout the entire experiment. Strader and Binkley (1989) and Whynot and Weetman (1991) both emphasised that high spatial and temporal variability during *in situ* studies could obscure treatment effects.

At both sites, each fertiliser treatment plot was sub-divided into two sub-plots at the start of the experiment. Each sub-plot was treated separately throughout the *in situ* study and soils were processed separately in the laboratory. Rates of NNM, N leaching and N uptake were then averaged to give fertiliser treatment effects. This data was not presented as a within treatment replicate as this would constitute pseudoreplication (Hurlbert, 1984). However, it showed that there was limited variability between the sub-plots and that the variability between the fertiliser treatment plots, especially at the Kurosol site, was real.

Nitrogen immobilisation was measured in both fertilised and unfertilised Kurosol topsoils, and was clearly enhanced during the period of measurement immediately after fertilisation. Immobilisation of N was observed in lodgepole pine plantations by Stump and Binkley (1993) and is recognised as a major process in eucalypt forests (Adams and Attiwill, 1986). Raison *et al.* (1992) also observed fertilisation to significantly increase N immobilisation in a Yellow Podzolic topsoil for almost 1 year after its application. The total N immobilised in their study was 147 kg N ha^{-1}. Compared to an average of 33 kg N ha^{-1} in this study. However, in their study they applied a much larger amount of N fertiliser (applying 400 kg N ha^{-1}, as ammonium sulphate) which could explain the larger and longer duration of immobilisation than observed here. The amount and length of immobilisation observed was approximately one quarter of that measured by Raison *et al.* (1992).

Nitrogen uptake was highest immediately after fertilisation at 26 kg N ha^{-1} $month^{-1}$. In examination of the long-term effects (2000 data) the highest N uptake and N leaching (not significant) in both fertilised and unfertilised topsoils occurred in March 2000 in association with increased rainfall after summer. Raison *et al.* (1992) also observed N uptake to be greatest when soil moisture was abundant.

The direction of nitrogen transformation, from net mineralisation to net immobilisation, depends on a number of factors, the C: N ratio of the soil organic matter is considered one of the most important. This is related to the inherent ratio in microbial cells of approximately five to fifteen (Paul and Juma, 1981). In this study,

no significant differences were observed between (P)N1Y and NIL treatment C/N ratios at 25 and 23, respectively (Chapter 3, Table 3.10). This is discussed further in Chapter 7 when other fertiliser treatments are examined.

Ammonium was the dominant form of mineral N in both fertilised and unfertilised Kurosols. Examination of both soil profiles (Chapter 3) showed that the ammonium dominance occurred throughout the soil profile, to the depth of 50 cm. Ammonium is often observed as the dominant mineral N form in fertilised and unfertilised forest soil (Williams, 1972; Johnson *et al.*, 1980; Adams and Attiwill, 1986). Under the conditions that prevail in many forest soils, including low pH and intense microbial competition for inorganic N, the conversion of ammonium to nitrate by nitrifying organisms is low. Thus in many temperate forest soils nitrate concentrations are often low or insignificant (Dyck *et al.*, 1983; Carlyle, 1986). However, in a study of old growth forests, Hart *et al.* (1994) indicated that nitrate immobilisation can be substantial in forests soils and a lack of soil nitrate during incubations is therefore not unequivocal with insignificant nitrification.

As nitrification is more inhibited by low pH than ammonification (Attiwill *et al.*, 1978) this could explain the low and generally undetectable rates of nitrification observed throughout the field study. The Kurosol topsoil was naturally acidic (pH < 4.2), and additions of N and P fertilisers increased this acidity by up to 0.6 pH units (Table 3.9). In a study of 38 podzolic soils, Carlyle *et al.* (1990) observed a clear discrimination between strongly and weakly nitrifying soils using soil pH and observed a distinct switch at a pH of 5.3 below which limited nitrification occurred. As the pH in these soils remained below 5.0 units, nitrification was not expected to occur even after fertiliser additions. Induced nitrification and significant leaching has been reported on sandy soil from fertilisation alone (400 kg N ha^{-1}) and combined fertilisation and irrigation treatments (Raison *et al.*, 1990; Khanna *et al.*, 1992).

4.4.2 Ferrosol topsoil

Nitrogen mineralisation rates were low in both fertilised and unfertilised Ferrosol topsoils at 16 and 23 kg N ha^{-1} yr^{-1}, respectively. Net N mineralisation was also low,

in both cool temperate mature eucalypt forest (<10 kg N ha^{-1} yr^{-1}) (Adams *et al.*, 1989b), and in young eucalypt plantations (13 to 188 kg N ha^{-1} yr^{-1}) (Wang *et al.*, 1998; Moroni *et al.*, 2002).

Large temporal variability combined with high spatial variability resulted in no clear seasonal trends in mineral N concentration or NNM. However, in unfertilised topsoil, NNM rates varied six-fold reaching a maximum in March. Adams and Attiwill (1986) observed N mineralisation peaks during summer, when both higher temperatures and moisture were present, while rates in other seasons were generally similar. This topsoil also showed no short or long-term changes in mineral N concentration, or NNM, due to annual fertilisation of N plus P for a period of thirteen years. In contrast to previous studies, no elevation in mineral N concentration was measured. The final fertiliser application to the Ferrosol occurred two months prior to instillation of the *in situ* studies. This, combined with the higher rainfall present at the southern site, may have resulted in field measurements missing the initial flush of mineral N due to fertilisation. Rainfall at the Ferrosol site following fertilisation was twice that (86 mm) of the Kurosol site (40 mm). Timing of fertiliser application and rainfall affects on mineral N availability are examined in detail in Chapter 5.

When comparing rates of NNM in Ferrosol topsoil, there was a non-significant depression of annual NNM after long-term fertilisation. This is in agreement with McLaughlin *et al.* (2000), who observed a significant decrease in NNM, with an overall inhibition of organic matter decomposition due to ammonium nitrate application (100 kg N ha^{-1}). However, there was also evidence that the N processes in the current study were influenced by the *in situ* core methods used. *In situ* core methods have been extensively reviewed by a number of authors (Raison *et al.*, 1987; Adams *et al.*, 1989b; Smethurst and Nambiar, 1989b), and the technique was confirmed in the Biology Forest Growth (BFG) experiment for estimating rates of N mineralised, uptake and leaching (Raison and Myers, 1992). In Tasmania, rates of NNM were expected to be low, as was observed, so the containment period of two-months was chosen to provide sufficient time for N accumulation to occur without causing detrimental changes. The lack of nitrate suggested that this was well founded.

However, large fluctuations in moisture during this incubation time did impact on the rates of NNM and N leaching measured.

Higher mineral N concentrations were often measured from open soil cores compared to those remaining closed. Differences could be large i.e., up to 200 % greater. Increased rates of NNM were often associated with higher moisture concentration in the open cores during the time of containment. These results suggest that isolating mineralising soil from rainfall could have underestimated field rates of mineralisation in this study. In addition, increased NNM in open cores often resulted in negative estimates of N leaching, with annual leaching rates of –15 and –33 kg N ha^{-1} in unfertilised and fertilised topsoils, respectively. Calculating rates of NNM from soil incubated in open cores may better represent field NNM rates. However, these calculations still resulted in no significant effect of fertiliser treatment on rates of NNM (NIL and P1YN1Y, 39 and 30 kg N ha^{-1} yr^{-1} respectively).

Rainfall during the *in situ* study was lower than average at both sites. At the Kurosol site simulated rainfall data from the bureau of meteorology (within 5 km) indicated that the annual rainfall in 1999 was only 690 mm and 713 mm in 2000. At the Ferrosol site the annual rainfall in 1999 was also extremely low for this site at only 760 mm, this increased to 1060 mm in 2000. In the Ferrosol topsoil moisture contents reached field capacity (65 %) only twice during the study, in July and September in open cores from unfertilised topsoil. These low levels of rainfall during this study at both sites may have reduced annual rates of NNM, especially under fertilised trees which often had higher moisture uptake rates (seen as significantly lower soil moisture in the topsoil). This hypothesis was tested in the laboratory studies discussed in Chapter 6 and 7. Using RFWC as a generalised moisture function for N mineralisation allows for microbial adaptation to the local environment, since microbes are adapted to the upper and lower limits of the water contents observed in the field (Paul *et al.*, 2003b). However, in this study NNM in both fertilised and unfertilised soils at both sites were unrelated to RFWC.

Total N measured in the fertilised (P1YN1Y) topsoil was also significantly less than that in the NIL treatment (1.7 t ha^{-1} compared to 2.3 t ha^{-1} respectively, Table 3.13). This could explain the lack of enhanced NNM in the fertilised topsoil and indicates that it may be important to examine N mineralisation in the litter layer at this site, as this was the horizon where substantial increases in total N were measured (Chapter 3, Table 3.13). Investigation of NNM rates in the litter layers (O2) was undertaken under controlled conditions in the laboratory (Chapter 6 and 7).

Ammonium was the dominant form of mineral N in both fertilised and unfertilised, Ferrosol topsoils. Examination of all soil profiles, in Chapter 3, showed that the ammonium dominance occurred throughout the soil profile, to the depth of 50 cm. The naturally low pH (pH < 4.5), and decline in pH in N and P treatments by up to 0.9 pH units (Table 3.15) may have also limited overall mineralisation rates in both the Ferrosol topsoils. Suppression of N mineralisation by soil acidity, and increases in NNM after liming, have been observed in a number of experiments (Attiwill *et al.*, 1978; Page *et al.*, 2003). Denitrification was not expected to contribute to N losses, due to low nitrate concentrations, low soil pH, free draining soils and moisture contents generally below field capacity. However, in a similar study of a Ferrosol under young eucalypt plantations Wang *et al.* (1998) inferred that negative N fluxes were probably due to denitrification. In a study of old growth forests Hart *et al.* (1994) indicated that nitrate immobilisation can be substantial in forests soils and a lack of soil nitrate during incubations is therefore not unequivocal with insignificant nitrification.

In this study only the top 10 cm of the soil profile was examined. Approximately one third of the soil profiles total N, organic C and mineral N was concentrated in the surface 10 cm of mineral soil (Table 3. 8 and 3.13). In contrast, mineral N was dispersed throughout the Ferrosol profile. A more even dispersion of mineral N down the profile often occurred in the higher N fertilisation treatments. Such dispersion of mineral N throughout these profiles might suggest significant amounts of N mineralisation could occur lower in the profile. However, this was considered unlikely as subsoil available C and oxygen would probably become limiting to

microbial growth and hence N mineralisation. The age of soil organic matter also generally increases down the profile and is therefore more resistant to decomposition (Federer, 1983). In addition, microbial distribution is generally concentrated in the surface of a profile (Murphy *et al.*, 1998a). The examination of surface (0-10 cm) soil as an indicator of N availability was qualified in a review by Binkley and Hart (1989). This review noted that the forest floor and mineral soil, to a depth of 15 cm, typically produced half or more of the N mineralised in forest. In a sandy podzol similar to the Kurosol studied in this experiment, Smethurst and Nambiar (1989b) noted that from the top 30 cm of mineral soil, 84 percent of the *in situ* N mineralisation occurred in the in the top 15 cm portion. Below 30 cm, amounts of mineralisable N were very low to undetectable. A strong proliferation of fine roots just below the litter layers also indicated the abundance of nutrients at the surface of the soil profile.

Removal of the understorey weeds one week prior to commencing *in situ* incubations in both *P. radiata* and *E. regnans* plantations, predominantly bracken (*Pteridium esculentum* (Forst.f.)), would have produced an increased number of severed roots in the plots studied. Removal of the understorey would have increased the number of dead roots in both fertilised and unfertilised soils at both sites. However, in the comparison of the fertiliser treatment weed removal was consistent. In a study of weed control effects on N in young *P. radiata* plantation in South Australia, Smethurst and Nambiar (1989a) observed that although mineral N concentrations were 50 to 80 % higher in plots where weeds had been controlled by herbicides there was no significant effect on annual NNM over two years. Enhanced mineral N concentrations in their study occurred for the first six months of measurements. In this study, the first six months of measurements were removed from annual NNM calculations. This was done to remove the last fertiliser application effects from the long-term changes from fertilisation. By removing this first six months, the immediate effects of weed control would have also been limited. Low mineralisation rates at these sites also suggest understorey removal did not stimulate NNM. In their study Raison *et al* (1987) noted that exposure of soil cores to living plants would result in N uptake. Removal of weeds at these sites prevents N uptake by these species. In addition, large woody roots were not generally observed in these cores and therefore would not have resulted in underestimation of NNM. Raison *et al* (1987) also noted

that the severing of roots appeared to have no effect on the patten of accumulation of mineral-N in several undisturbed forest soils for up to 130 days.

The impact of below-ground inputs on soil C and N pools and turnover has received limited attention (Ross *et al.*, 2001) and is generally confined to tench plots with limited or no understorey. In trenched plot studies under a 26 year old *P. radiata* plantation in New Zealand, Ross *et al.* (2001) observed similar amounts of mineral N and NNM between trenched and control treatments after 56 days in a mineral soil (0-10cm). Thirteen years after trenching in an old-growth conifer forest Hart and Sollins (1998) indicated that measurable changes in C and N pools were only observed in laboratory NNM rates. Few detectable changes in C and N pools and processes were observed in the field and no significant changes of annual *in situ* NNM or nitrification were measured. Ross *et al.* (2001) did observe large changes in nitrification from trenching, up to 200-fold increased. As discussed previously little nitrate was observed in either the Kurosol or Ferrosol, which suggests limited effects of understorey removal on severed roots.

It was considered that even though rates of NNM may have been slightly reduced due to increased immobilisation from C additions from served roots, as suggested by Adams *et al.* (1989b), the effects of fertilisation on NNM rates in these soils could still be compared. The effect of roots and increased C additions due to both slashing the site and the placement of cores is limited in later laboratory studies as roots are removal when the soil are sieved (Chapter 6 and 7).

4.5 Conclusions

Long-term annual applications of N fertiliser had no significant effect on the annual rate of NNM in either the Kurosol of Ferrosol topsoils as assessed by MANOVA tests. Rates at the Kurosol site were higher with fertilisation but overall rates on all treatments were low.

The effects of N fertilisation on short and long-term trends in N availability and NNM due to fertilisation varied between soil types;

- In the Kurosol topsoil, with an inherently lower nutrient status, located in a lower annual rainfall area (938 mm), mineral N concentrations increased for a period of around six-months after N fertilisation, before declining to pre-fertilisation levels.
- In contrast, in the Ferrosol topsoil, with an inherently higher nutrient status, located in a high rainfall area (1200 mm), no peak in mineral N concentration was observed two months after N fertilisation.
- Net N mineralisation rates in Kurosol topsoil were low between 13 and 52 kg N ha^{-1} yr^{-1} (not significantly different) in unfertilised and fertilised treatments, respectively.
- Net N mineralisation rates in Ferrosol topsoil were low between 23 and 16 kg N ha^{-1} yr^{-1} (not significantly different) in unfertilised and fertilised treatments, respectively.

Seasonal variations in NNM are difficult to clearly define without sampling for a number of years and the limited time of this study resulted in no significant seasonal effect. However, at both sites there was an overall enhancement in NNM when temperature and moisture levels were higher, during late summer and early autumn, while NNM declined during winter. N uptake was also enhanced during this period. This study indicates that at these low mineralising sites, maximum rates of NNM can be measure during a period of high temperature immediately after rainfall. In contrast, minimum NNM rates depend on either the moisture or the temperature subject to the sites rainfall history. However, minimum rates were generally below half the maximum rate of NNM measured during the rest of the year. Fertiliser treatments followed similar seasonal NNM patterns, indicating that the sampling times selected to measure minimum and maximum rates of NNM could be the same irrespective of the fertiliser treatment.

Results from this study suggest pre-wetting soil cores would allow maximum rates of NNM to be measured at the end of summer and minimum rates at the start of winter.

This hypothesis was tested in the laboratory studies discussed in Chapter 6 and 7. In addition, in chapter 5, I test whether the timing of the fertiliser application alters the maximum and minimum rates of NNM, by assessing the role of the season of fertiliser application on mineral N availability.

At both sites rates, of NNM were low and an incubation period of 60 days was considered necessary to measure changes in NNM rates between fertiliser treatments. However, no significant differences were measured between fertiliser treatments at either site. The role of incubation period on NNM needs to be examined in the laboratory (Chapter 7) to test whether the *in situ* incubation period increased immobilisation and therefore decrease measurable differences between fertiliser treatments.

Chapter 5. Effect of the season of nitrogen fertilisation on the temporal pattern of mineral nitrogen concentrations in litter and topsoil from two contrasting forest soils.

5.1. Introduction

The investigation of *in situ* rates of N mineralisation revealed relatively short-lived elevations in concentrations of mineral N after fertilisation (Chapter 4). Mineral N concentrations decreased substantially within a six-month period in the Kurosol topsoil, while in the Ferrosol topsoil there was no measurable increase in mineral N two months after application. Rapid declines in concentrations of mineral N after fertilisation have been noted in other forest soils (Williams, 1972; Smethurst *et al.*, 2001). The final fertilisation of the Ferrosol site occurred two months prior to instillation of the *in situ* mineralisation cores. This, combined with the higher rainfall at the site, may have resulted in field measurements missing the initial flush of mineral N due to fertilisation. In this study I tested that assumption by examining the rate of mineral N decline in both Ferrosol and Kurosol topsoils for a period of six months following fertilisation.

The previous *in situ* study also indicated low rates of mineralisation throughout the year with a peak occurring in late summer immediately after rainfall, in both fertilised and unfertilised topsoils. Application of fertilisers occurred in May or June prior to the *in situ* study. Although numerous investigations have studied the role of climate, temperature and moisture, and its ultimate impact on soil N mineralisation (Nadelhoffer and Aber, 1984; Boone, 1992; Gower and Son, 1992), limited research is published on the interaction between season and fertilisation on mineral N availability in forest soil. However, research studying foliar N dynamics by both Nason *et al.* (1990) and Thomas and Mead (1992b), observed a strong influence of subsequent rainfall events on N availability. This study aimed to identify whether the timing of fertiliser would effect the period of mineral N enhancement. This has implications for fertiliser efficacy as well as sampling strategies needed to determine long-term NNM rates.

There is an extensive body of research indicating increased litter production due to N fertilisation (Hunter and Hoy, 1983; Theodorou and Bowen, 1990; Neilsen and Lynch, 1998; Maier and Kress, 2000) and subsequent site improvement. In this study, application of P plus annual N almost doubled the mass of the litter (O2 horizon) under *P. radiata* compared to when no fertiliser was applied, while under *E. regnans,* annual application of N, plus P, increased this litter layer five-fold (Chapter 3). Associated with this increased mass, total N content of the litter layers increased by over two-fold and seven-fold under *P. radiata* and *E. regnans*, respectively (Figure 3.3 and 3.7). However, there are few studies on the effect of fertilisation on mineral N availability with the litter layer. In this study I examine how seasonal applications of N fertiliser affect temporal patterns of mineral N concentrations both in the litter and soil of the eucalypt and pine plantations.

5.2 Methods

5.2.1 Experimental design

The experiment was designed to examine mineral N concentrations in the topsoil (A1 horizon) and litter (O1 plus O2 horizons), for a period of six months after N fertilisation (100 kg N ha^{-1}). Each site included five treatments i.e. four different times of N application, and a NIL fertiliser treatment. Nitrogen fertiliser (ammonium sulphate) was applied at the end of the months of June 2000, October 2000, January 2001 or April 2001. All treatments were replicated five times, in a randomised block design.

5.2.2 Site Establishment

This experiment was established at the two sites previously studied (Chapters 3 and 4). The southern *E. regnans* plantation, on a Haplic Dystrophic Brown Ferrosol, and the north-eastern *P. radiata* plantation, on a Bleached Dystrophic Yellow Kurosol. The experiment was located on the previously biannually fertilised plots, ((P)N2Y) and (P2YN2Y) described in Section 3.2 (Table 3.4 and 2.5). At both sites, replicate biannually fertilised plots were available. Due to site debris, both replicate plots where required at the Kurosol site (Figure 5.1), while one plot only was required at

the southern Ferrosol site (Figure 5.2). For further information on the existing long-term fertilisation experimental design, and site details, see Section 3.2.

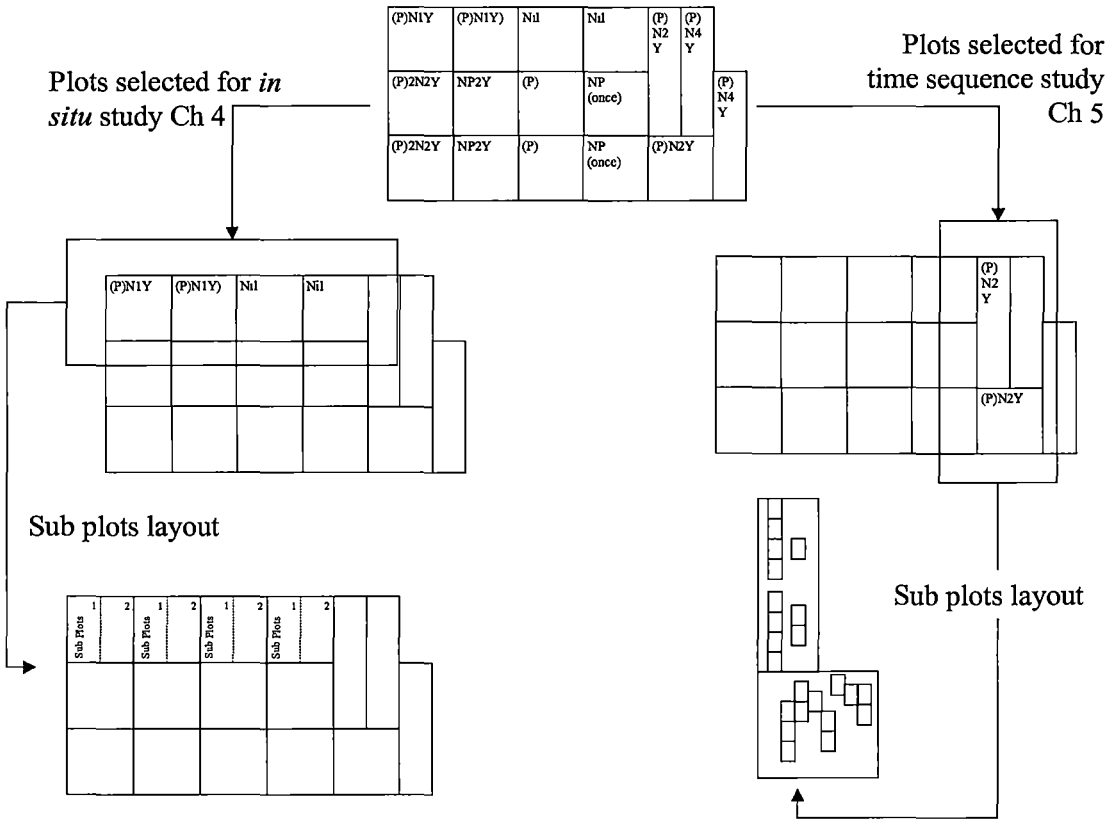


Figure 5.1 indicates the plots selected for sampling and the layout of the sub-plots used in this experiment at the Kurosol site.

Site preparation involved the slashing of understorey species, predominantly bracken, across all treatments. Slash residue was carefully removed to the outside of the plot, to prevent additions to, and disturbance of, the existing litter horizon. Cleared sites were then pegged into 25; 3 x 2 m sub-plots (5 treatments by 5 replicates). Sub-plots were placed between cultivated tree rows, avoiding large debris (rocks and logs).

To ensure even distribution of fertiliser granules, sub-plots were first subdivided into six one-meter square sections. The required amount of N fertiliser (100 kg N ha^{-1}) was also divided into six equal masses and hand broadcast evenly on to each individual square (Photo 5.1).

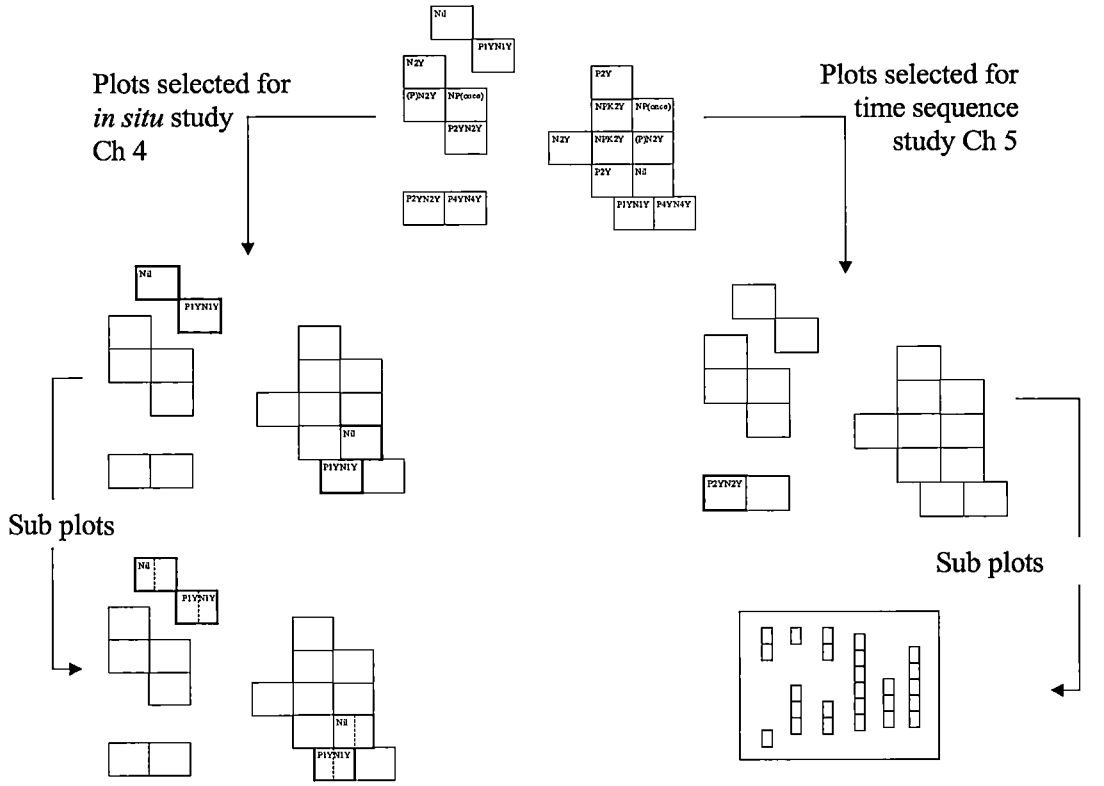


Figure 5.2 indicates the plots selected for sampling and the layout of the sub-plots used in this experiment at the Ferrosol site.

5.2.3 Site Sampling

Litter and topsoil samples were collected monthly for six months after each fertilisation. Unfertilised plots were sampled for the duration of the experiment i.e. a total of sixteen months.

Litter samples were cut using a 5 cm-diameter PVC pipe inserted to the depth of the interface with the mineral soil (up to 6 cm). Once the litter was removed, the underlying mineral soil was cut to the depth of 10 cm using the same device. Four sub-samples of each litter and soil samples were collected from each sub-plot and pooled separately, in plastic bags then placed in a "cooler-bag" for transport.

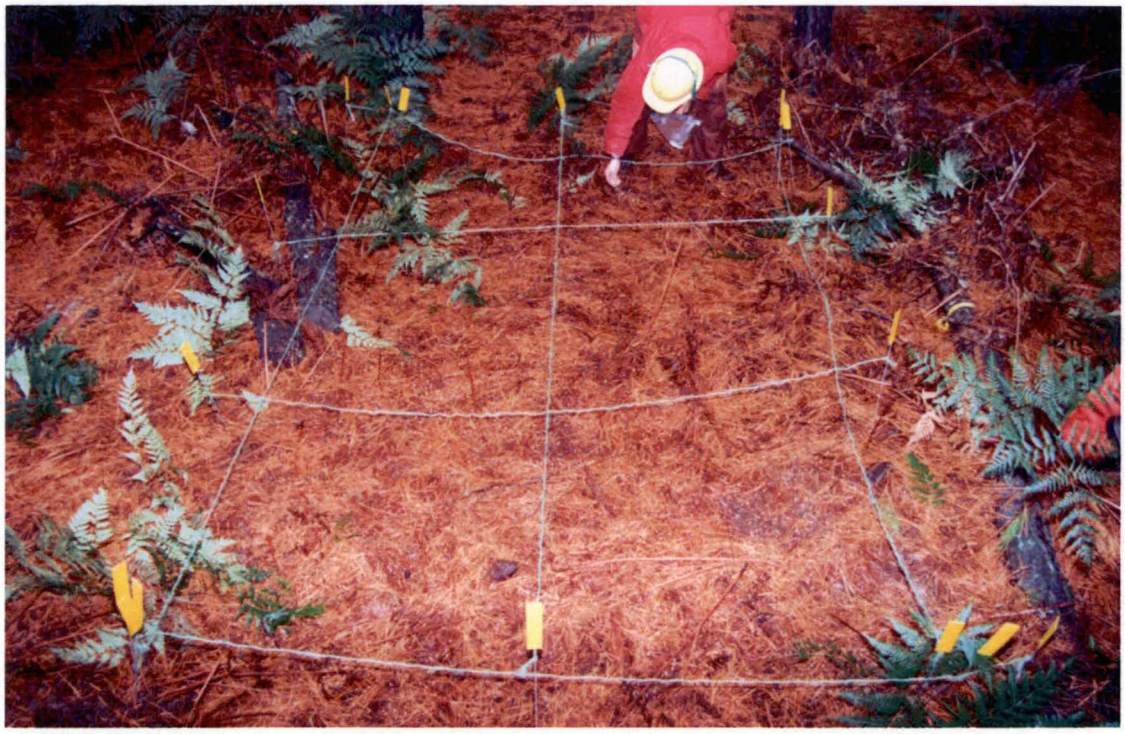


Photo 5.1 Sub-plot layout showing the division of the sub-plot into six sections for even fertiliser application.

All samples were brought to the laboratory within eight hours of sampling, and placed in the cool-store at 2-4°C. The bulked samples were thoroughly mixed to homogenise the material. The samples were prepared in the same manner as used in the field mineralisation study (Section 4.2.3). Gravimetric water contents were determined for each sub-plot by drying at 105°C for 24 hours. The samples were sieved to < 2 mm prior to chemical analysis. Duplicate 10 g sub-samples of < 2 mm samples were shaken in 50 ml of 2 *M* cold KCl for one hour and the extract filtered through Whatman No.42 papers (methods modified from Raymond and Higginson 1992). The resultant extracts were frozen and stored for later analysis of NO_3^- and NH_4^+ using a flow-injection analyser (FIA) (Lachat Instruments). See Section 4.2.3 for further details.

Data analysis

Results were analysed using a multiple analysis of variance (MANOVA) procedure of the GenStat software (Genstat 5 Committee, 1988). Means were compared using the treatment interactions (fertiliser and time) least significant differences ($p < 0.05$ and $p < 0.001$), as stated.

5.3 Results

5.3.1 *P. radiata* litter and Kurosol topsoil

One month after fertilisation, in all treatments, mineral N concentrations were between 10 and 28 times greater in the topsoil, and 10 to 250 times greater in the litter than in unfertilised topsoil and litter. After each fertiliser treatment, enhanced mineral N concentrations, in both fertilised horizons (O1 and A1) decreased within four to five months to concentrations similar to those measured in unfertilised horizons (Figure 5.3 and Figure 5.4).

Seasonal trends in mineral N concentration were not evident in the unfertilised topsoil. Concentrations remained low throughout the experiment reaching a maximum of 4 $\mu g\ g^{-1}$ in September 2000 (Figure 5.5). In contrast, mineral N concentrations in the unfertilised litter increased during summer (December to February). These increases were associated with the period of highest soil temperatures but lowest rainfall, and concentrations declined at the beginning of the autumn rainfalls. The autumn decline in litter mineral N was associated with a small peak in mineral N in the underlying topsoil. Generally, mineral N concentration in the litter was ten times that of the topsoil in the unfertilised treatments (Figure 5.5).

The horizon with the greatest mineral N mass varied depending on the time of fertilisation (Figures 5.3 and 5.4). Fertiliser significantly increased mineral N mass ($kg\ ha^{-1}$) in the topsoil in all treatments ($p<0.05$). However, the amount of mineral N, and time that the significant increase lasted, varied with treatment. Mineral N increases due to fertilisation lasted for three months after the June treatment but only two months after the January and April treatments. Although the October treatment resulted in lower mineral N masses in litter after fertilisation than the other treatments (Figure 5.3b) significant ($p<0.001$) increases in this treatment lasted for four months.

Comparing fertilisation times, one month after fertilisation both January and April had significantly ($p = 0.015$) higher mineral N contents in the *P. radiata* litter than June, which was significantly higher than October (Figure 5.6). During the six months of sampling, the average amount of available mineral N measured in the litter was lower

in the October treatment compared to January and April ($p<0.001$). In contrast in the Kurosol topsoil, although mineral N content was significantly ($p < 0.001$) higher in all treatments for the first two months after fertilising, there was no significant interaction between fertiliser treatment by time in the (Figure 5.7).

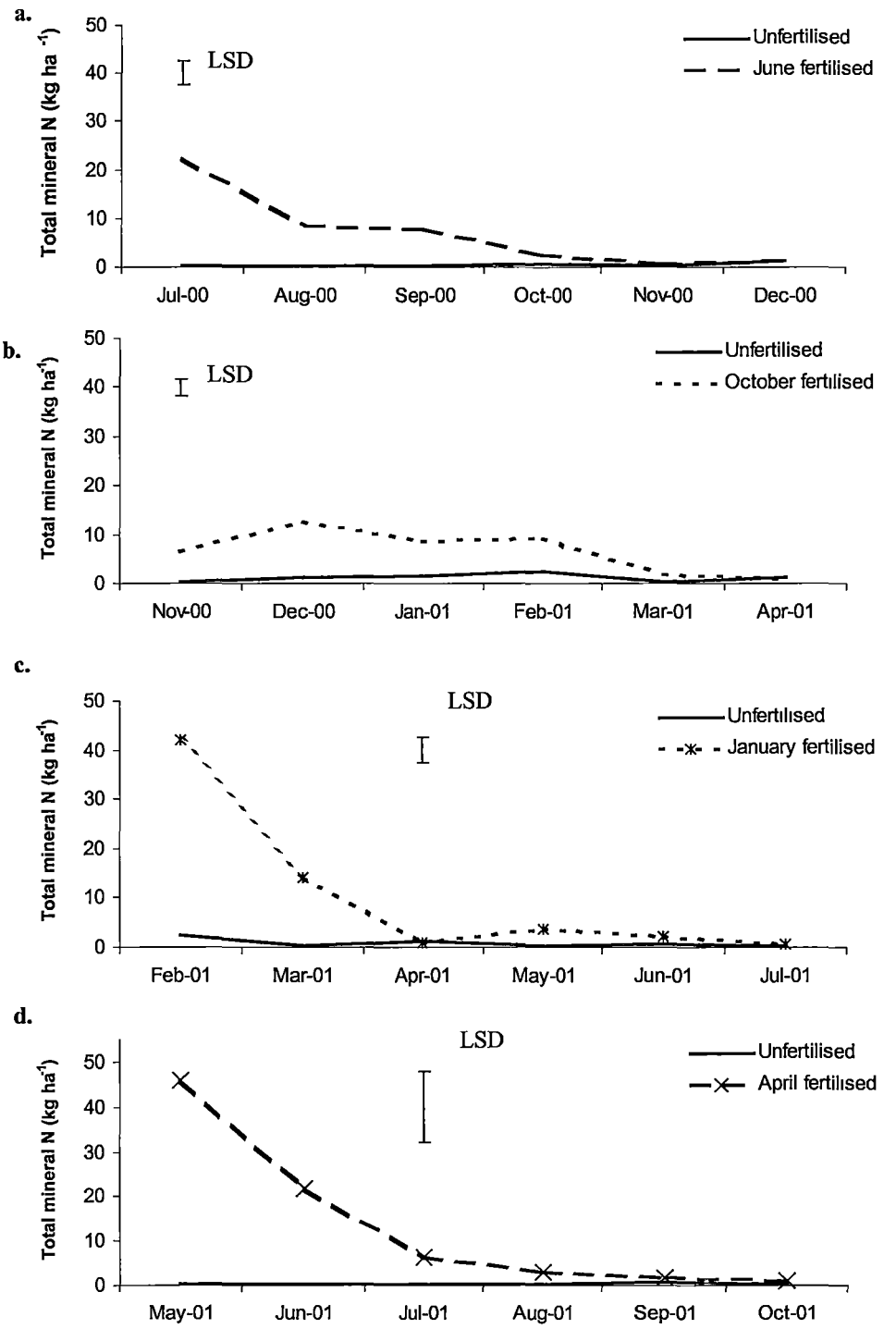


Figure 5.3 Total mineral N mass (kg ha^{-1}) in *P. radiata* litter on the Kurosol topsoil after each fertiliser application (a) June, (b) October, (c) January, and (d) April. Bars indicate LSD for fertiliser treatment by time.

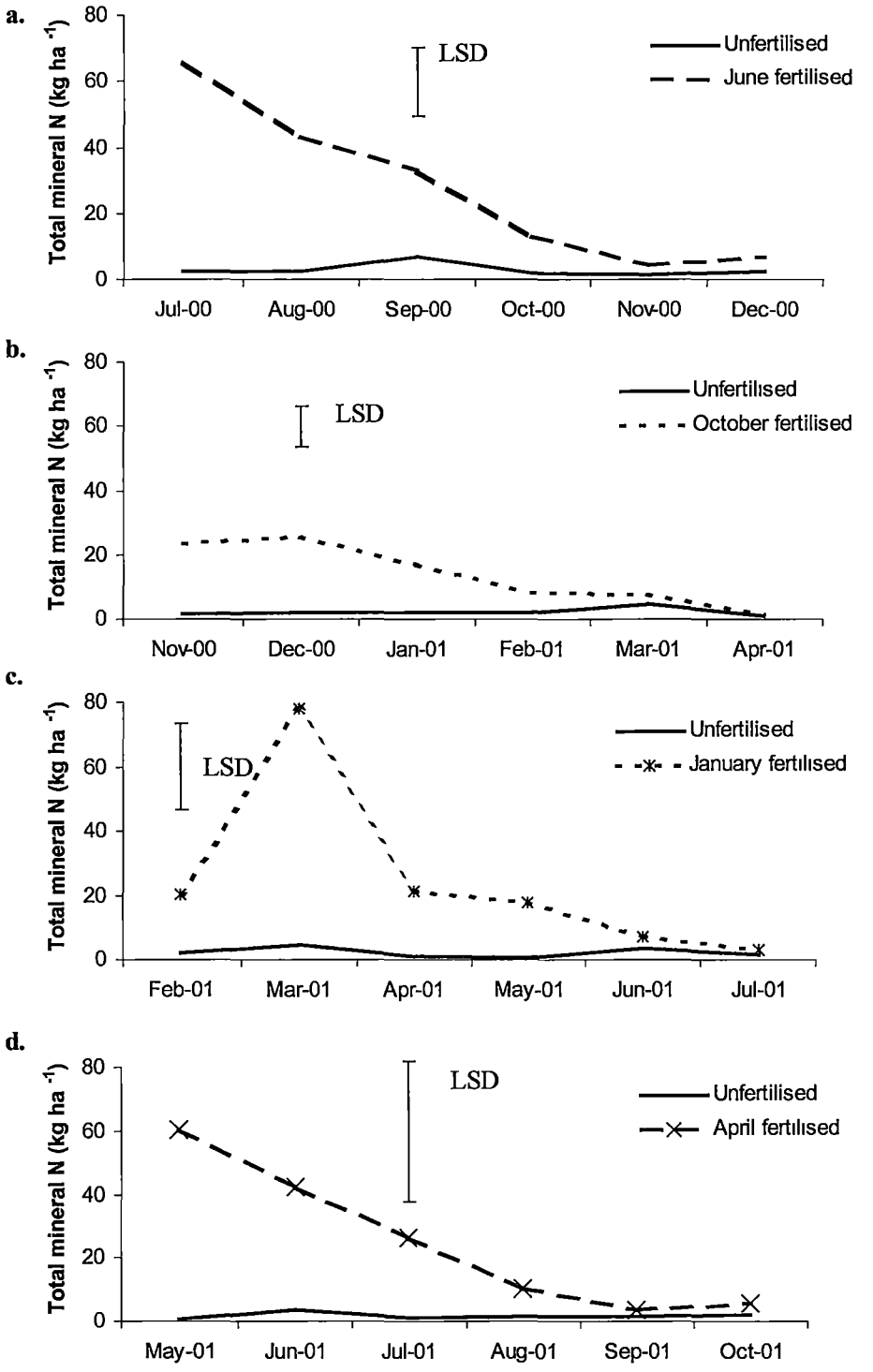


Figure 5.4 Total mineral N mass (kg ha^{-1}) in Kurosol topsoil under *P. radiata* plantation after each fertiliser application (a) June, (b) October, (c) January, and (d) April. Bars indicate LSD for fertiliser treatment by time.

One month after fertilisation, the combined mineral N mass in litter plus topsoil, ranged from 16 to 180 times greater than that unfertilised. This mineral N mass in combined litter plus topsoil samples varied significantly with time of fertilisation. One

month after fertilisation the October treatment had a significantly lower ($p<0.05$) mineral N mass in litter plus topsoil (30 kg N ha^{-1}), compared to other application times. However, by the third month mineral N masses were not significantly different between treatments.

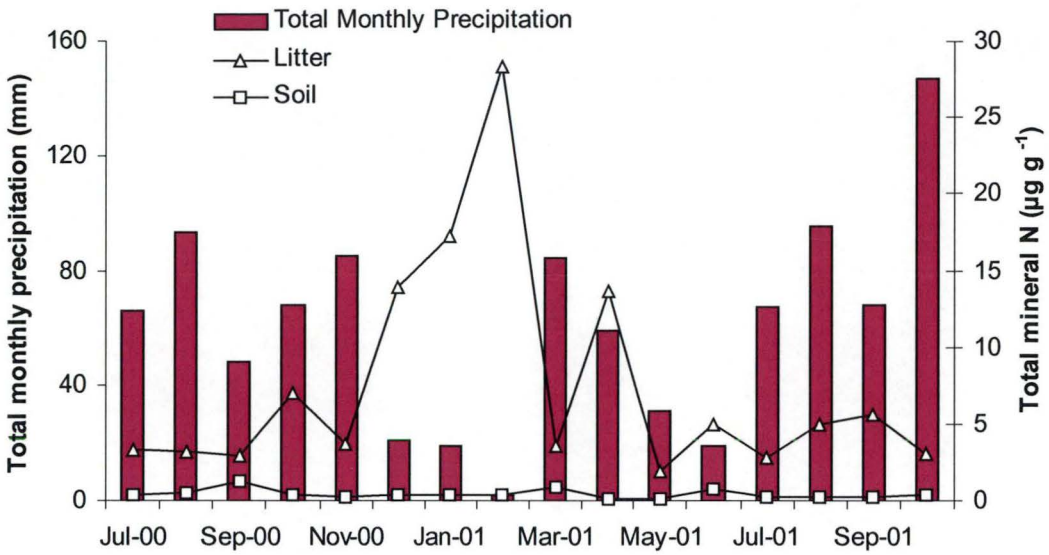


Figure 5.5 Comparison between total monthly precipitation (recorded at Fingal weather station) and mineral N concentrations (µg g^{-1}) measured in the unfertilised *P. radiata* litter and Kurosol topsoil.

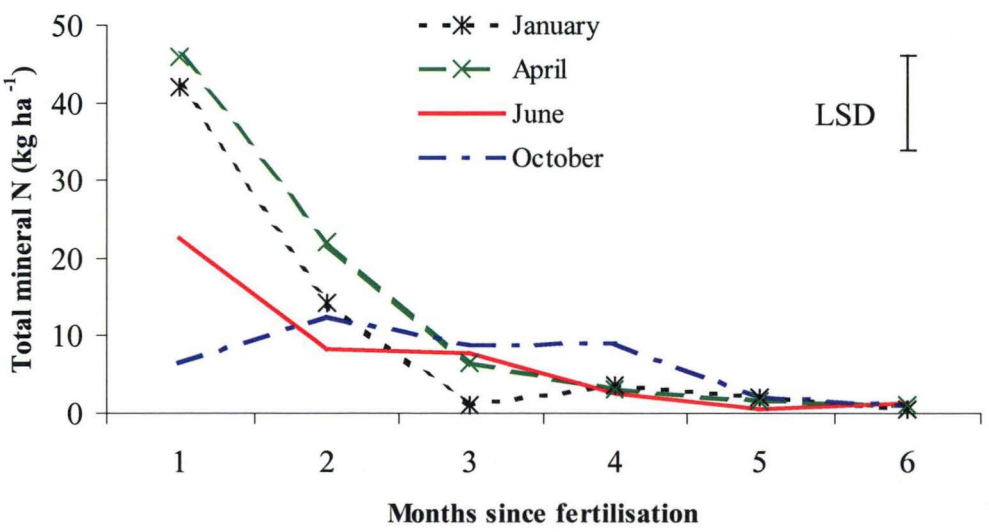


Figure 5.6 Total mineral N mass (kg ha^{-1}) in *P. radiata* litter after each fertiliser application (a) June, (b) October, (c) January, and (d) April. Bars indicate LSD for fertiliser treatment by time.

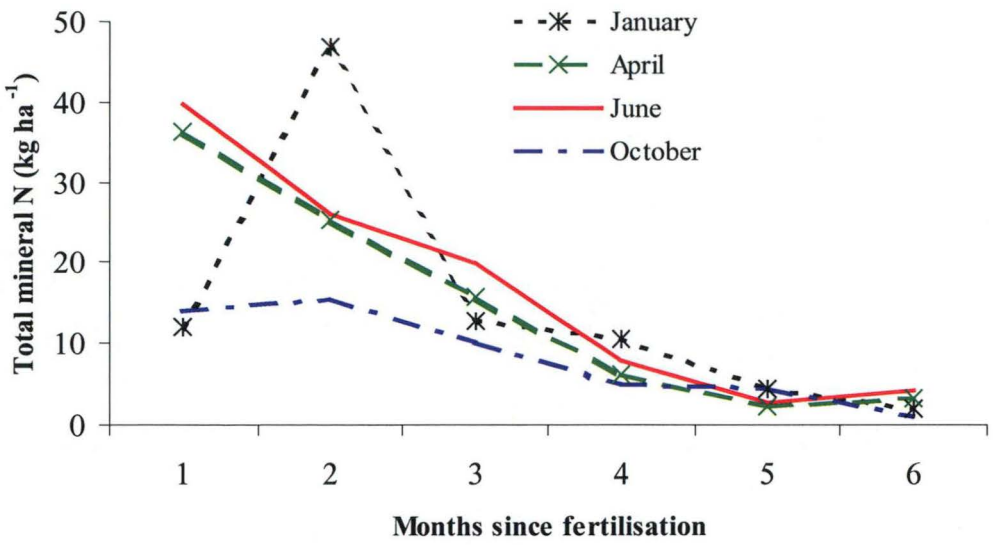


Figure 5.7 Total mineral N mass (kg ha^{-1}) in Kurosol topsoil after each fertiliser application (a) June, (b) October, (c) January, and (d) April. Fertiliser treatment by time interaction was not significant (p = 0.067).

The period of mineral N retention in combined litter plus topsoil was longest after the April treatment, and shortest after the October treatment. Comparing the fertilised and unfertilised sites (taking into account the low mineral N availability in the unfertilised horizon), of the 100 kg N ha^{-1} that was applied in April, 96 % was still present one month after fertilisation. This corresponded to a period of relatively low rainfall in the month immediately after the April application. In contrast, the October application was followed by a period of relatively high rainfall.

Annual temperature range varied between 0.9 °C (minimum) and 26.2 °C (maximum), in July and February, respectively (Figure 5.8). Variations between the monthly maximum and minimum temperatures were highest in January and February and lowest during October and November. No correlations were observed between mineral N and temperature and monthly rainfall trends in unfertilised litter (Table 5.1). In contrast, mineral N concentrations in unfertilised topsoil were significantly correlated with air temperatures (Table 5.1). Maximum and minimum temperatures accounted for 65 percent of the variation in mineral N ($p<0.05$).

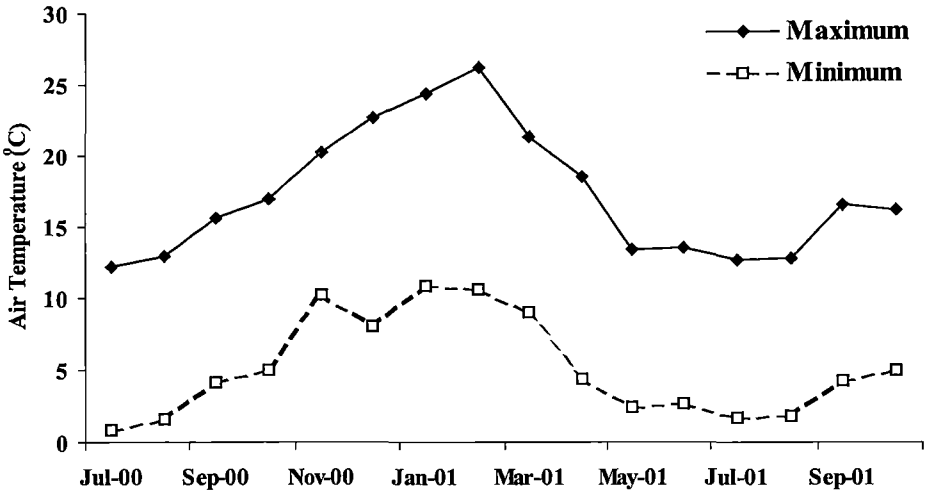


Figure 5.8 Monthly minimum and maximum air temperatures (°C), recorded at Fingal weather station.

Table 5.1 Unfertilised *P. radiata* litter and Kurosol topsoil mineral N concentration correlations with each other and with other climatic variables.

	Max. temperature	Min. temperature	Monthly Rainfall	Soil mineral N	Temperature Range
Litter mineral N	-0.41	-0.39	-0.09	0.42	-0.37
Max. temperature		0.97**	-0.45	-0.65*	0.83**
Min. temperature			-0.32	-0.65*	0.68**
Monthly Rainfall				0.33	-0.66*
Soil mineral N					-0.50*

* Indicates significance value,* $p<0.05$, ** $p<0.001$

Table 5.2 Nitrate –N (g ha^{-1}) measured monthly in Kurosol topsoils for six months after fertiliser treatment.

Application month	Months since application					
	1	2	3	4	5	6
January	0	0	0	0	0	0
April	0	0	0	0	21	0
July	0	2	8	5	0	0
October	0	0	0	0	0	0

Table 5.3 pH measured monthly in Kurosol topsoils for six months after fertiliser treatment.

Application month	Months since application					
	1	2	3	4	5	6
January	3.31	3.48	3.34	3.36	3.45	3.42
April	3.10	3.30	3.31	3.24	3.34	3.31
July	3.35	*	3.41	3.45	3.58	3.53
October	3.54	3.38	3.39	3.39	3.40	3.43

* missing value

Mineral N was dominated by ammonium with nitrate concentrations often below detectable limits (Table 5.2). At its highest, nitrate content was only 2 % of the total mineral N measured during the fifth month after the April treatment. Nitrate contents were not significantly different between fertiliser treatments in the six months measured. Differences also did not occur when seasonal variations in control soils were taken into account (monthly control nitrate content minus monthly fertilised). Over the six month period on average pH was lower ($p < 0.001$) in the April application than in any other treatment (Table 5.3). Time since fertilisation did not significantly effect pH and there was no significant interaction of pH and time. Overall, pH decline in all treatments during the six-month period between 0.1 and 0.3 pH units compared to the control soil. However, there was a significant difference ($p<0.001$) in the pH of control soils during the 16 months of measurement. Taking into account seasonal effect on pH during each six months (monthly control minus monthly treatment pH) there was no effect of fertilisation timing on pH decline in the Kurosol topsoil.

5.3.2 *E. regnans* litter and Ferrosol topsoil

One month after fertilisation, in all treatments, mineral N concentrations were between four and twelve times greater in the topsoil and up to sixty times greater in the litter, than those unfertilised. After each fertiliser treatment, enhanced mineral N concentrations were evident only for the first month's measurement (Figure 5.9 and 5.10). The short period of fertiliser retention in topsoil resulted in significant ($p<0.001$) increases in mineral N measured in June and April treatments only (Figure 5.10). In contrast, all treatments significantly ($p<0.001$) increased mineral N masses in the litter for one to two months, above those unfertilised (Figure 5.9).

Unfertilised topsoil and litter horizons both showed a depression in mineral N concentrations in summer associated with low rainfall events, and a subsequent increase in mineral N corresponding to increasing autumn and winter rainfall (Figure 5.11). This was contrary to the Kurosol site where mineral N peaked in the litter during summer. Maximum mineral N concentrations occurred in winter, at 9 $\mu g\ g^{-1}$ and 17 $\mu g\ g^{-1}$ in topsoil and litter, respectively. Generally, mineral N concentrations in unfertilised litter were ten times those measured in unfertilised topsoil. One month

after fertilisation, the combined mineral N mass (kg ha^{-1}) in litter plus topsoil was seven to twenty five times greater than that unfertilised.

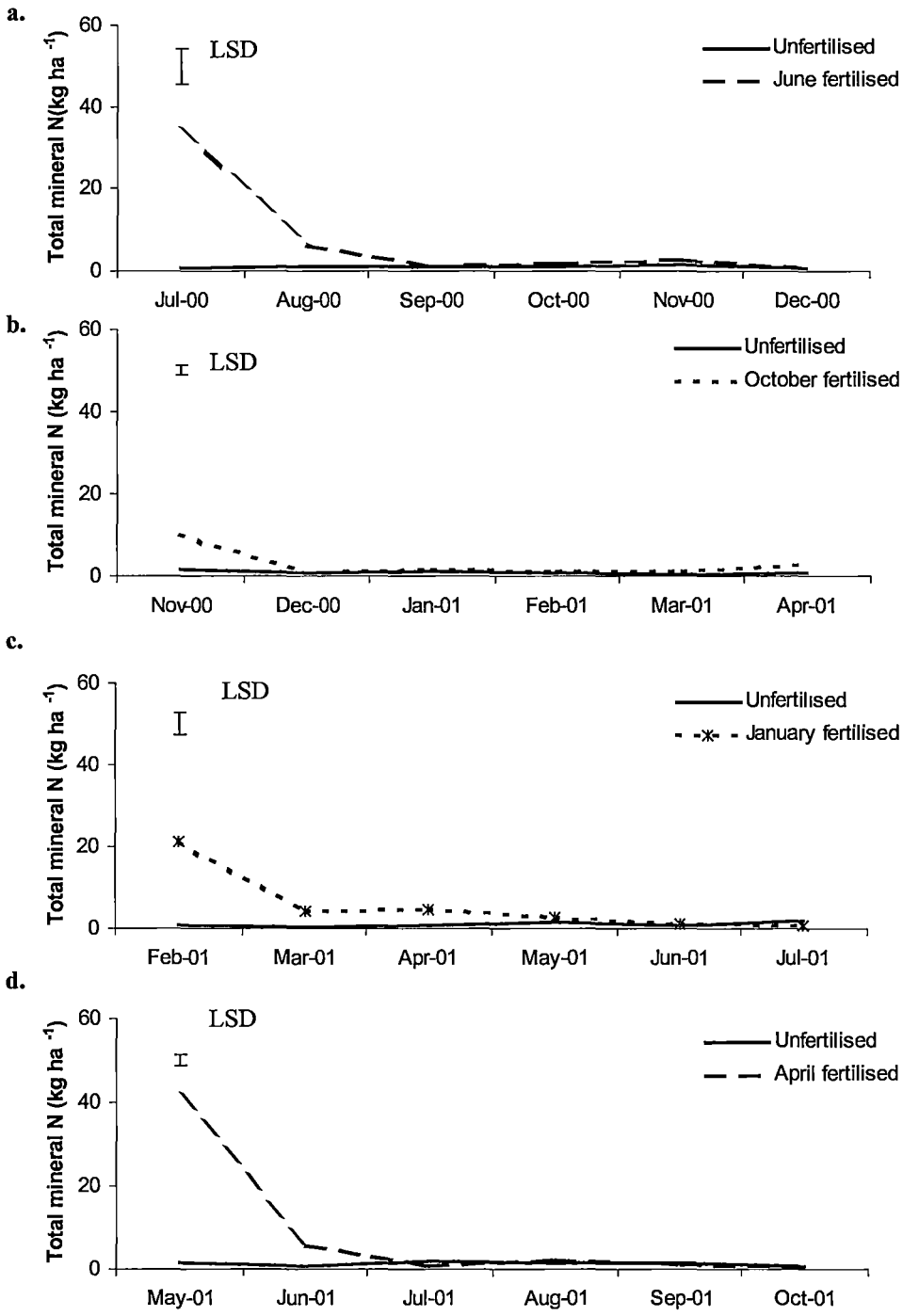


Figure 5.9 Total mineral N mass (kg ha^{-1}) in *E. regnans* litter on Ferrosol topsoil after each fertiliser application (a) June, (b) October, (c) January, and (d) April. Bars indicate LSD for fertiliser treatment by time.

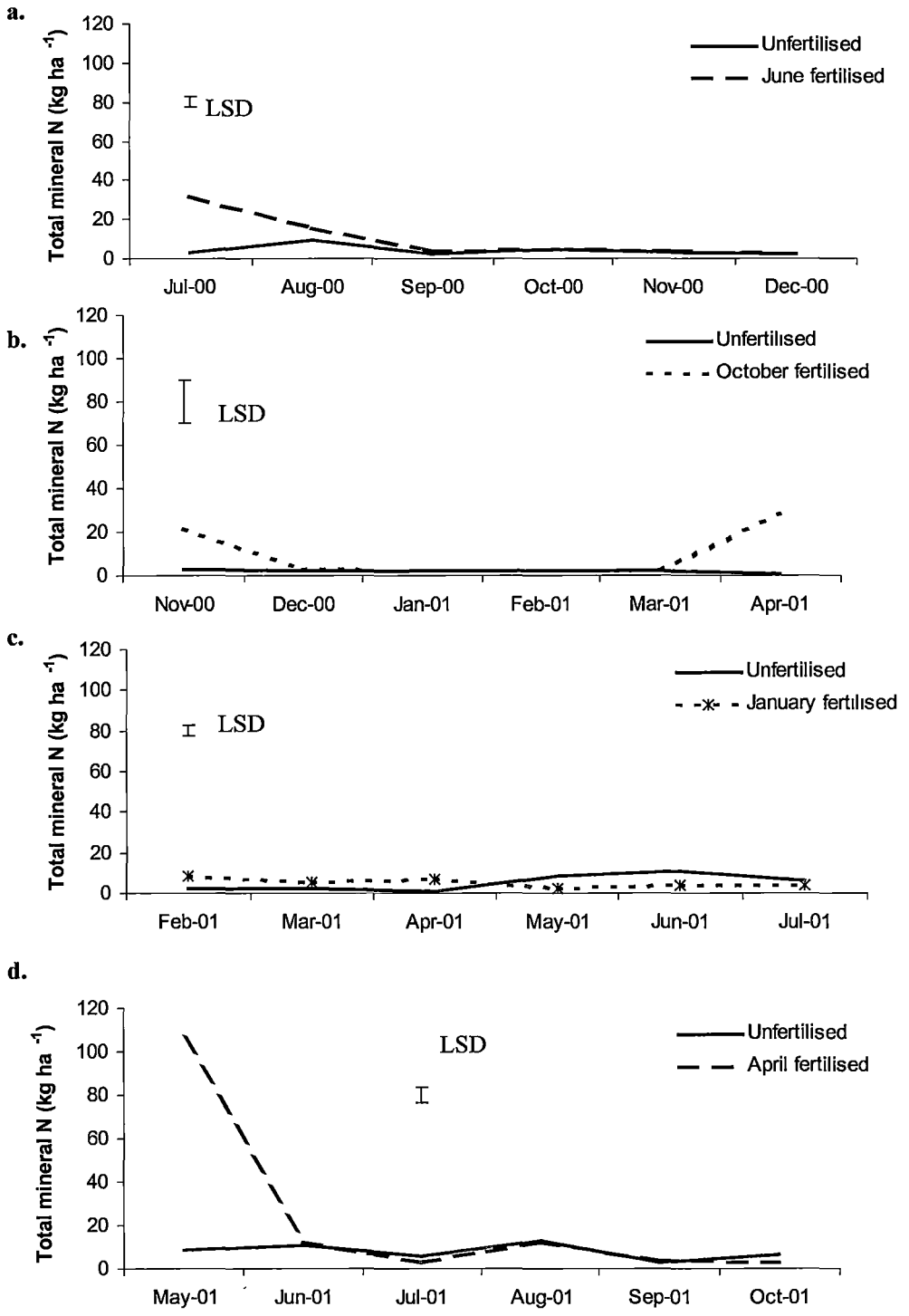


Figure 5.10 Total mineral N mass (kg ha^{-1}) in Ferrosol topsoil under *E. regnans* plantation after each fertiliser application (a) June, (b) October, (c) January, and (d) April. Bars indicate LSD for fertiliser treatment by time.

Comparing fertilisation treatments alone, all treatments had significantly ($p < 0.001$) different mineral N content in *E. regnans* litter just for the first month after fertilisation (Figure 5.12). Mineral N content was highest in April and declined in the

order June, January and then October. Overall, October had a significantly lower average mineral N content during the six months of sampling ($p<0.001$). In agreement with *E. regnans* litter, mineral N content in the Ferrosol topsoil was only significantly higher in the first month after fertilisation ($p < 0.001$) (Figure 5.13). April treatment also had the highest mineral N content in the litter for the first month and on average over the six-month period ($p < 0.001$).

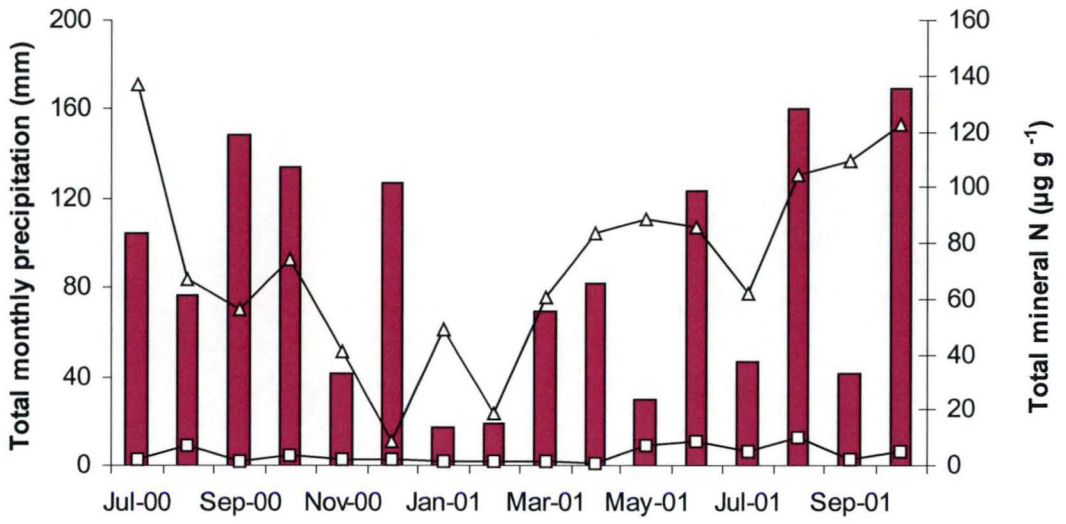


Figure 5.11 Comparison between total monthly precipitation (recorded at Dover weather station) and mineral N concentrations (µg g^{-1}) measured in the unfertilised *E. regnans* litter and Ferrosol topsoil.

Comparing fertilised and unfertilised sites (taking into account low mineral N availability in the unfertilised horizon), of the 100 kg N ha^{-1} that was applied in April, more mineral N was available in the topsoil after the first month than was supplied. In contrast, less than 30 % of the N was measured in the combined topsoil and litter horizons one month after the October application.

Annual air temperatures ranged from 2.6 °C (minimum) and 24 °C (maximum) in July and February, respectively (Figure 5.14). Variations between the monthly maximum and minimum temperatures were highest in January and February and lowest during October and November. No correlation was observed between unfertilised topsoil mineral N content and monthly temperatures and rainfall (Table 5.4). In contrast, even

though temperatures were low during periods of increased mineral N availability, the mineral N content of the litter correlated with temperature, and moisture trends. ($p<0.05$). The difference between mean monthly maximum and minimum air temperature accounted for 81% of the variation in mineral N content in the *E. regnans* litters, while monthly rainfall explained 37% of the variation.

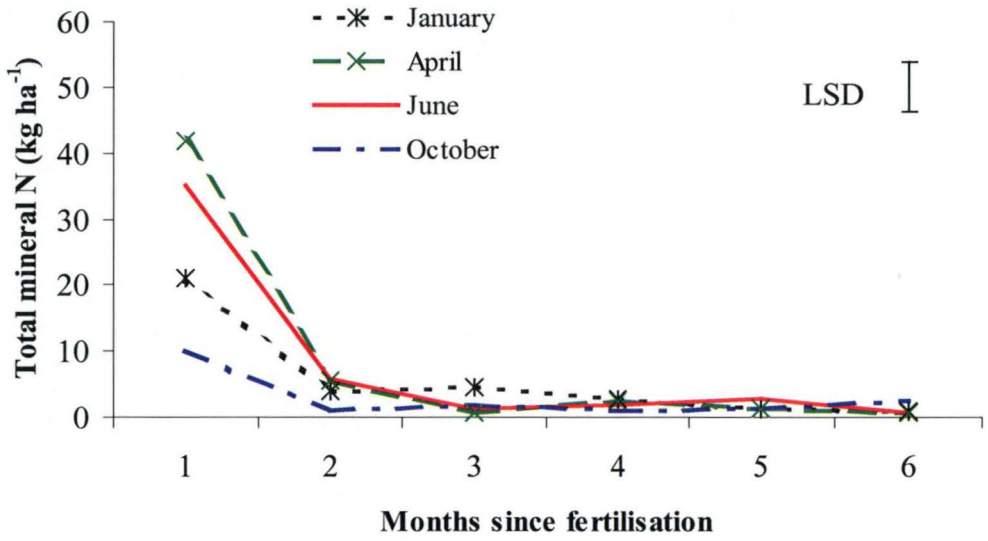


Figure 5.12 Total mineral N mass (kg ha^{-1}) in *E. regnans* litter after each fertiliser application (a) June, (b) October, (c) January, and (d) April. Bars indicate LSD for fertiliser treatment by time.

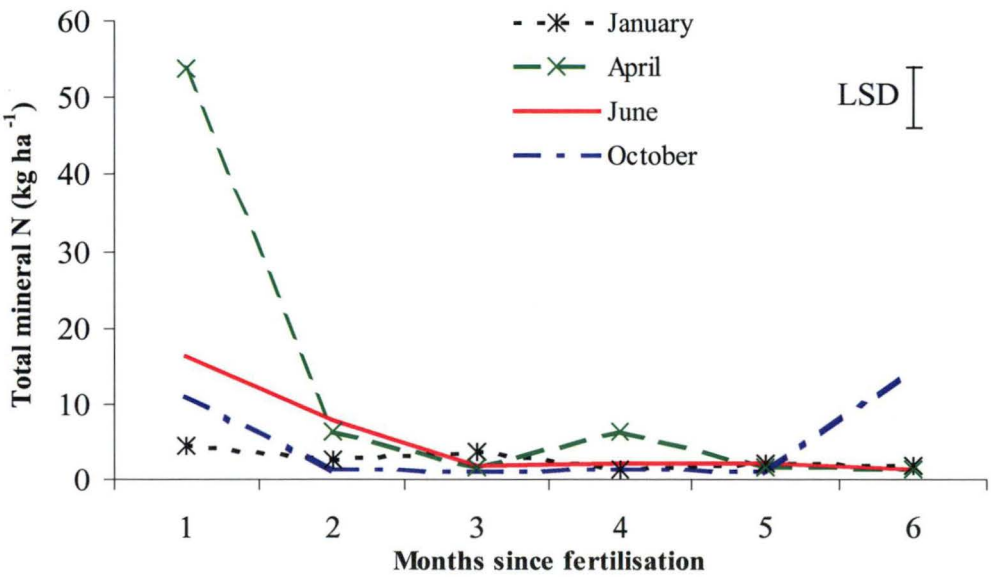


Figure 5.13 Total mineral N mass (kg ha^{-1}) in Ferrosol topsoil after each fertiliser application (a) June, (b) October, (c) January, and (d) April. Bars indicate LSD for fertiliser treatment by time.

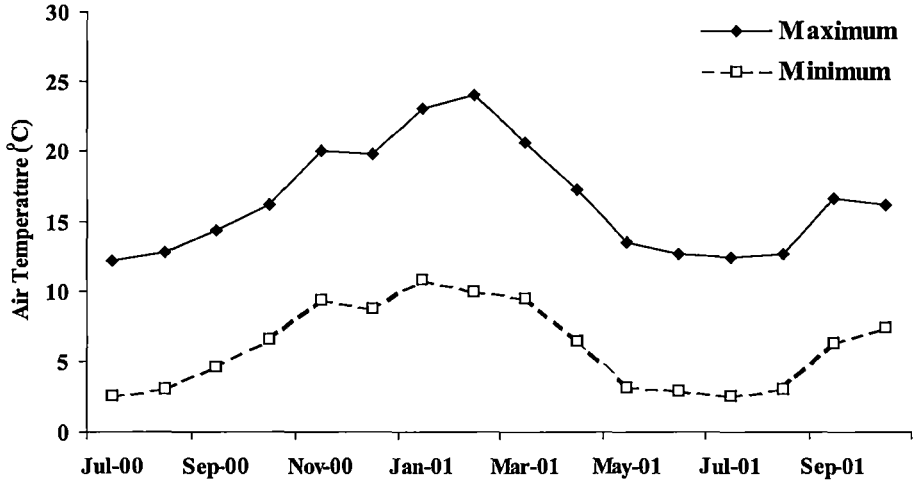


Figure 5.14 Monthly minimum and maximum temperatures (°C), recorded at Dover weather station.

Table 5.4 Unfertilised *E. regnans* litter and Ferrosol topsoil mineral N concentration correlations with each other and climatic variables.

	Max. temperature	Min. temperature	Monthly Rainfall	Temperature Range	Soil mineral N
Litter mineral N	**0.79****	**0.62***	-0.61	**0.90****	-0.16
Max. temperature		**0.96****	-0.41	**0.77****	0.05
Min. temperature			-0.29	**0.54***	0.09
Monthly Rainfall				**-0.54***	-0.08
Soil mineral N					-0.07

Indicates significance value,* $p<0.05$, ** $p<0.001$

Table 5.5 Nitrate –N (g ha^{-1}) measured monthly in Ferrosol topsoils for six months after fertiliser treatment.

Application month	Months since application					
	1	2	3	4	5	6
January	0	0	69	0	91	0
April	0	95	0	0	0	0
July	43	13	0	0	0	0
October	467	0	0	0	0	333

Table 5.6 pH measured monthly in Ferrosol topsoils for six months after fertiliser treatment.

Application month	Months since application											
	1		2		3		4		5		6	
January	4.27	b	4.18	ab	4 18	ab	4.13	ab	4.36	bc	4.05	ab
April	3.98	a	4.04	ab	4 01	ab	3.95	a	4.51	c	3.97	a
July	4.32	bc	*		4.32	bc	4.29	bc	4.40	bc	4.31	bc
October	4.25	b	4.15	ab	4.01	ab	4.29	bc	4.13	ab	4.05	ab

* missing value

Mineral N was dominated by ammonium with nitrate concentrations often below detectable limits (Table 5.5). At its highest, nitrate content was only 7 % of the mineral N measured during the fifth month following January fertilisation. Nitrate contents were not significantly different between fertiliser treatments or between seasonally adjusted fertiliser treatments (monthly control nitrate content minus monthly fertilised) in the six months measured. Both treatment and the month after fertiliser application effected topsoil pH (Figure 5.6). April treatment had the lowest average pH compared to July and January. Overall, pH declined in all treatments during the six-month period between 0.02 and 0.27 pH units compared to the control soil. Seasonal trends in unfertilised Ferrosol topsoil pH were significant during the 16 months of measurement ($p<0.001$) reaching a maximum of pH 3.70 in September and a minimum of pH 3.34 in August. Taking into account this seasonal effect on pH during each six-month treatment (monthly control pH minus monthly treatment pH), fertilisation in October (0.27), April (0.18) and January (0.18) reduced pH significantly ($p<0.001$) more than July (0.02).

5.4. Discussion

5.4.1 *P. radiata* litter and Kurosol topsoil

In all fertiliser treatments there was an elevation of mineral N for up to five months after fertilisation. These agreed with results from Chapter 4, and other field and laboratory research (Johnson *et al.*, 1980; Hingston and Jones, 1985; Khanna *et al.*, 1992). The rate of mineral N decline depended on the horizon and season of fertilisation.

Rainfall immediately after fertilisation influenced N fertiliser retention, with the rainfall following October fertilisation associated with low mineral N concentration and dry conditions following April fertilisation associated with the highest mineral N concentration. These trends agree with Thomas and Mead (1992b), who observed N was retained from urea fertilisation in the soil horizon under 2-year-old *P. radiata* plantations longer during periods of lower rainfall. Nason *et al.* (1990) also related N leaching or immobilisation to rainfall immediately after fertilisation.

Although peaks in mineral N were generally associated with initial fertilisation, there was evidence of delay in N movement between the litter and soil. This was evident in the Kurosol topsoil fertilised in January, with N increases during February and March 2001 associated with a decline in the overlying litter. This flux in mineral N was associated with warm temperatures and a large increase in rainfall, after a three-month dry spell, and as such may also correspond to a flush of N mineralisation due to a drying and re-wetting cycle in the litter and topsoil. Enhanced mineralisation of N due to rainfall (within 4 hours) was observed in agricultural soils by Murphy *et al.* (1998b). This enables a considerable amount of N mineralisation to occur after summer rainfall, even though the soil surface dries rapidly. Data interpretation unfortunately relies on determining just when the rainfall event occurred and at what stage in the re-wetting response cycle the sample was taken. Detailed information on rainfall individual events was not measured during this study. However, a similar peak in mineral N occurred in March of the previous year during *in situ* measurements (Chapter 4), although this was not significant. This was associated with a significant increase in N uptake in both fertilised and unfertilised soils in March 2000, while no differences in N uptake were measured for the remainder of the year. For the remainder of the year N uptake was below 3 kg N ha^{-1} $month^{-1}$ and as such had not significant effect on the mineral N availability in this study.

Canopy cover and litter retention would have also effected moisture regimes. Conifer needles have a hydrophobic surface and small surface area, and as a result they may be slow to take up moisture, but they are effective at retaining moisture (Heal, 1979). Although the moisture content in *P. radiata* litter fluctuated by (1600 percent), the amount was generally higher than that measured in the topsoil. In contrast to the

limited influence of season on topsoil mineral N, the *P. radiata* litter showed a distinct elevation of mineral N during the summer period, which declined dramatically after autumn rainfalls. Although litter horizons are characterised by a high biological activity and frequent and extreme fluctuations in moisture content, these effects are poorly understood (Clein and Schimal, 1993).

In agreement with earlier research the effect of season on mineral N availability was variable (Adams and Attiwill, 1986; Adams *et al.*, 1989b; Birk, 1992). Under *P. radiata* there were no correlations with the climate variables measured and N availability in litter, while the variation of mineral N in topsoil was correlated to monthly air temperatures.

5.4.2 *E. regnans* litter on Ferrosol topsoil

In all fertiliser treatments there was a brief elevation of litter and topsoil mineral N in the first month after fertilisation. The elevation of mineral N for only one-month on this site reflects the generally wetter conditions at this site.

Although peaks in mineral N were associated with initial fertilisation, there was evidence of N mineralisation at the end of measurements in the October treatment (in April 2001). At this time mineral N concentration in the topsoil increased above those observed in the first month after fertilisation, while no corresponding increase occurred in the unfertilised topsoil. This flux in mineral N was also associated with warm temperatures and a period of high rainfall.

In agreement with Chapter 4, little nitrate was measured in the topsoil from both sites. At its highest, nitrate content was only 7 % of the mineral N, which occurred during the fifth month following fertilisation in January. In a study of 38 podzolic soils Carlyle *et al.* (1990) observed a clear discrimination between strongly and weakly nitrifying soils using soil pH and observed a distinct switch at a pH of 5.3 below which limited nitrification occurred. As the pH in these soils remained below 5 units, nitrification was not expected to occur even after fertiliser additions.

Negative correlations between mineral N content in the *E. regnans* litter and monthly rainfall probably result from N leaching during large rainfall events. Moisture content in litters was higher than in topsoils and this difference between the two remained relatively constant, while the moisture content in the litter varying five-fold. This could explain the strong temperature dependent correlation in litter but not in soil. The influence of moisture availability on NNM in the Ferrosol topsoil was previously observed and discussed in the *in situ* study of Chapter 4. During that experiment, NNM in topsoil was reduced by half when additional rainfall was eliminated from *in situ* incubating soil. Nadelhoffer *et al.* (1991) observed that cumulative NNM was significantly related to an organic C by temperature interaction ($r^2 = 0.73$, $p<0.001$), which could explain the correlations with temperature in litter but not topsoil. The previous *in situ* study also indicated although N uptake was highest around the beginning of Autumn (March) there was not significant effect of season on N uptake.

5.5 Conclusions

Enhancement of mineral N was evident in both horizons, at both sites, after each fertiliser treatment. However, the length of time for which this was evident depended on the horizon being studied, the site and the season of fertilisation.

- At the wetter, Ferrosol site planted with *E. regnans*, the flux of mineral N after fertilisation was brief with only a small amount present one month after fertilisation. By the second month, mineral N concentrations were similar to unfertilised horizons. This validates the assumption made in Chapter 4 that the two-month delay from fertiliser application to *in situ* measurements resulted from the flush of mineral N due to fertilisation being missed at this site.
- In comparison, the drier Kurosol site planted with *P. radiata* showed a mineral N flux that lasted between four and five months. These trends are in agreement with the earlier observations made at these sites (Chapter 4) and confirms that a six-month delay after fertiliser application is adequate to determine long-term fertiliser effects on NNM, independent of the time of fertiliser application.
- At both sites the measurement of mineral N after fertilisation was greatest in April treatments, and lowest in October treatments, with similar amounts available in the litter and topsoil. Indicating that current operational practice of applying

fertiliser in autumn provides an adequate window for fertiliser uptake to occur. This is further validated by the increased uptake of N in autumn measured during the *in situ* study (significant only in the Kurosol) (Chapter 4).

- At both sites, one month after fertiliser application in April, mineral N contents were around 80 percent of that applied. In comparison, the October treatment contained around 20 percent. At both sites, the large drop in mineral N following October treatment was associated with high spring rainfalls during this period.
- In agreement with previous studies, rainfall appeared to be a strong regulator of mineral N availability at both sites.
- At both sites, ammonium was the dominant form of mineral N, with nitrate often below detectable limits or less than 10 percent of the total mineral N. The highest proportion measured was 7 percent in the Ferrosol topsoil five months after fertilisation in January.
- At both sites, mineral N concentrations were generally 10 times greater in litter than topsoil in all treatments. Although the litter had higher concentrations of available N the greater density of the topsoil means the total amounts on N in the two horizon types were quite similar.

Chapter 6. Temperature, moisture and fertiliser effects on net nitrogen mineralisation in laboratory incubations of soil and litter from two contrasting sites.

6.1 Introduction

At both sites (Kurosol and Ferrosol), long-term N fertilisation had no statistically significant effect on *in situ* NNM rates (Chapter 4) and after N fertilisation increases in mineral N concentration were observed to be relatively short-lived, decreasing to pre-fertilised levels within a few months (Chapter 5). However, in both studies, changes in soil mineral N, due to N fertilisation were influenced by the rainfall events that occurred after fertilisation. Seasonal effects on N mineralisation are widely reported in the literature (Nadelhoffer and Aber, 1984; Adams and Attiwill, 1986; Plymale *et al.*, 1987; Foster, 1989).

Increased plantation growth through long-term fertilisation can lead to changes in the microclimate of the underlying forest floor. For example, long-term fertilisation can increase the size of the tree canopy (Smethurst *et al.*, 2003), stem volume (Waring, 1972; Hunter and Hoy, 1983; Schönau and Herbert, 1989; Neilsen and Lynch, 1998), and the rate of litter fall, as discussed in Chapter 3. A larger tree canopy and litter layer insulates the underlying soil and dampens fluctuations in both soil temperature and moisture. In addition, the larger canopy can result in a higher rate of transpiration and a subsequent decrease in soil moisture content. Evidence of such trends were seen during the *in situ* mineralisation study, where, for the majority of sample times, there were significantly lower moisture contents under annually fertilised *P. radiata* trees compared to those unfertilised (Chapter 4). As a result, long-term fertilisation creates different microclimates for microbial activity between fertilised and unfertilised soils.

Rainfall during the 18-month study was lower than average, and therefore might have limited NNM. Previous researches have established a strong relationship between water availability and soil and litter N mineralisation rates (Powers, 1990; Evans *et al.*, 1998; Prasolova *et al.*, 2000). These variations in the micro- and macro-climate of certain soils makes it more difficult to extrapolate data generated over the present

relatively short-term field study to the longer-term (20-30 years). To minimise the influence of climatic variations on NNM rates, many researchers have incubated soil under controlled temperature and moisture conditions (Theodorou and Bowen, 1983a; Richards *et al.*, 1985; Carlyle *et al.*, 1998b).

In addition to climatic variations, nutrients applied in fertilisers eventually become distributed between the overstorey, understorey, forest floor, mineral soil horizons and various loss pathways (Miller, 1981), which also changes the patterns nutrient cycling between these compartments. The effect of fertilisation on NNM in litter was not examined in the field study (Chapter 4). However, results from Chapter 3 clearly indicate that changes in total N, due to fertilisation was significant only in litter and not in the top 10 cm of mineral soil. In karri (*Eucalyptus diversicolor* F. Muell.) re-growth forests in Western Australia, fertilisation (200 kg N ha^{-1}) increased annual litter fall by 21 % and N content by 23 % in three years (O'Connell and Grove, 1993) and was associated with an increase in the amounts of nutrients transferred into the litter layer from the soil during decomposition. At these sites it was considered necessary to determine the rates of NNM in the litter, as the litter was a significant nutrient pool in both plantations studied. In addition this present study aimed to determine the optimum temperature and moisture content for N mineralisation in litter (O2) and topsoil (A1) horizons of each of the sites studied, and using these optimum-rates, compare N mineralisation rates from contrasting fertiliser treatments.

6.2 Methods

6.2.1 Field Sampling

At both sites where *in situ* rates of NNM were measured (Chapter 4), four plots were sampled, i.e. two annually fertilised plots and two unfertilised plots (Table 6.1). Samples were collected from uncultivated zones between tree rows using the following procedures.

Table 6.1 Field sampling summary

Site location	Southern Tasmania	North-east Tasmania
Soil	Ferrosol	Kurosol
Species	*E. regnans*	*P. radiata*
Fertiliser treatments	P1YN1Y	(P)N1Y
	NIL	NIL
Horizons	A1 (0-10 cm)	A1 (0-10cm)
	O2 Litter	O2 Litter

Litter was collected using a 25- by 20-cm frame. Samples were separated into two horizons, i.e. the undecomposed organic debris (O1 horizon) and the decomposed organic debris (O2 horizon) (McDonald *et al.*, 1990). There was also a distinct separation between the O1 and O2 boundaries at both sites, with the O2 horizon containing darker particles that were smaller and often matted together by fungi. The O1 horizon was the remaining loose litter on top of the O2. Only the O2 horizon was retained for analysis. Six O2 litter sub-samples were collected and bulked per plot into plastic bags, which were then placed in insulated bags for transportation to the laboratory.

After litter removal, a 25- by 20- by 10 (depth)-cm block of mineral soil was removed, and placed in to a 28-litre plastic bin. At these sites the distinction between layers was clear and relatively easy to separate, with a clear boundary between the O1 and A1 horizon and a marked mass of roots between the two. Site profiles photographs are presented in Chapter 3, which demonstrate this. The mineral soil depth (0-10 cm) corresponds to that used for *in situ* core incubations in Chapter 4. Six mineral soil sub-samples were bulked per plot into a single plastic bin. Topsoil mass per hectare was calculated using the average bulk densities (of replicate treatment plots) determined in Chapter 3. The average bulk density across both treatment plots in the Kurosol topsoil was 1.625 g cc^{-1}, and in the Ferrosol it was 1.26 g cc^{-1}. Litter O2 mass per hectare was calculated using average litter masses per fertiliser treatment (*P. radiata* NIL, 40 t ha^{-1}; (P)N1Y, 71 t ha^{-1}; *E. regnans* NIL, 14 t ha^{-1}; P1YN1Y, 77 t ha^{-1}). For further information on sample collection and calculations of litter mass and

soil bulk densities, see Section 3.4. All samples were brought to the laboratory within eight hours of collection and placed in the cool-store at 2-4 °C.

6.2.2. Laboratory sample preparation

Pooled topsoil (A1) horizons were sieved through a 2 mm sieve to remove rocks, large roots and twigs. The resultant < 2 mm mineral topsoil was then thoroughly mixed and cool-stored (2-4 °C) for two weeks. This period of cool storage was to allow equilibration after the sieving disturbance. Bulk O2 horizon samples were passed through a 4 mm sieve to remove large debris. The resultant < 4 mm fraction was then thoroughly mixed and placed in a square 28 litre plastic bin and cool-stored (as per < 2 mm soil).

Following two weeks cool-storage, all bulked samples were removed from storage and sub-sampled, in duplicate, to determine mineral N concentration (as per methods in Section 3.4). The concentrations of NO_3^- and NH_4^+ measured at this time were set as initial (time zero, T_o) concentrations.

To compare rates of NNM concentrations were converted to kg ha^{-1} using the following equations;

NNM kg Soil/ha = NNM (μg/g) x BD (g/cm^3) x 10^{-9} kg (μg/kg) x 10^8 (cm^2/ha) x 10 cm depth of the topsoil.

NNM kg Litter/ha = NNM (μg/g) x Litter (kg/ha) x 10^{-6} g (μg/g).

6.2.3 Determination of moisture content for laboratory incubations

Three water potentials were chosen for comparison, i.e. just above the permanent wilting point (15 bar) just below field capacity (0.33 bar) and an intermediate value between the two (3 bar). These tensions were applied to both the Ferrosol and Kurosol topsoil using pressure plate apparatus.

From bulk < 2 mm topsoil samples; twelve sub-samples (approximate 30 grams) were removed and packed firmly into individual rubber holding rings. Four packed rings from each plot were then placed on each of the three porous pressure plates and wetted thoroughly. Individual plates were then placed under pressure to produce the designated water potentials of 0.33, 3 or 15 bar, and allowed to equilibrate for three days. Soil samples were then removed and gravimetric water content determined after drying at 105 °C for 24 hours.

Moisture content for litter samples were determined to simulate freely drained litter horizons in the field. From each bulk < 4 mm litter (O2 horizon) sample per plot, four sub-samples were removed and packed firmly into rubber holding rings. Packed rings were placed onto a porous plate and wet thoroughly. Wet O2 horizon samples where then left to free drain (without suction) and evaporate during a period of 36 hours. The remaining mass of water for each sub-sample was then calculated using oven dry weights (105°C, 24 hours).

Remaining bulk < 2 mm soil and < 4 mm litter samples were split for moisture corrections. Depending on the initial moisture content, a sample was dried down (at ambient temperatures) or distilled water was added, by applying a fine spray to the surface of the sample. Once a sample was at the required moisture content, it was thoroughly stirred and returned to cool-store (2-4 °C) for a further two week period of equilibration.

6.2.4 Incubations methods

The design of the experiment included factorial combinations of two sites, two horizons (O2 and A1) and two fertiliser treatments (annual application and unfertilised). There were also four incubation temperatures and three moisture levels for each soil and two incubation temperatures and two moisture levels for each litter. Each temperature-by-moisture treatment was replicated in three individual vials.

Samples of 40 g of < 2 mm topsoil and 30 g of < 4 mm litter were placed into separate 100 ml clear plastic vials. These vials were covered with polyethylene film (brand

name, Gladwrap) to allow O_2 and CO_2 exchange while limiting the loss of water (Connell *et al.*, 1995).

All vials were incubated in the dark two months at one of the treatment temperatures (3, 12, 16 or 22 °C for topsoil and 12 and 16 °C for litter). During incubation, samples were disturbed as little as possible while moisture content was checked fortnightly by weighing. Water content was readjusted, if necessary, by gently spraying with distilled water until the original moistened sample weight was reached. Moisture loss was low at 2 to 4 % by day 28.

Following incubation, samples in vials were thoroughly mixed and a single 10 g sub-sample was removed for extraction with cold KCl (see Section 6.2.2). Determined mineral N concentration was set as the final concentration (T_1).

Table 6.2 Summary of laboratory incubation parameters

Site	Horizon	Temperature (°C)	Moisture Content (% w/w)
North-east	Kurosol topsoil (A1 horizon)	3	10 % (level 1)
		12	15 % (level 2)
		16	20 % (level 3)
		22	
	P. radiata litter (O2 horizon)	12	80 %
		16	120 %
South	Ferrosol topsoil (A1 horizon)	3	30 % (level 1)
		12	45 % (level 2)
		16	60 % (level 3)
		22	
	E. regnans litter (O2 horizon)	12	120 %
		16	180 %

The four temperatures represent the range of temperatures occurring in the forest floor in Tasmania (Chapter 4, Figures 4.9 and 4.13). The three moisture contents in soil samples for each site represent; Level 1, approximately just above permanent wilting point, Level 3, approximately just below field capacity, and Level 2, a water content

mid-way between the other two levels (Table 6.2). All three levels were observed in the field, at each site, during the 18-month field study (Chapter 4).

Data analysis

Results from each plot were compared by the standard error between replicate vials. Overall comparison, of all four plots at each site, were analysed using a multiple analysis of variance (MANOVA) procedure of the GenStat software (Genstat 5 Committee, 1988). Means were compared using the treatment interactions (fertiliser, moisture and temperature treatments) least significant differences ($p < 0.05$ and $p < 0.001$), as stated.

6. 3 Results

6.3.1 Kurosol topsoil

Initial mineral N content of Kurosol topsoil (prior to incubation, T_0) was low, generally less than 10 kg ha^{-1}. On average, fertilised topsoil ((P)N1Y) had mineral N content four times greater than those unfertilised (NIL), 6.7 and 1.9 kg ha^{-1}, respectively (Figure 6.1). However, mineral N contents from the two fertilised topsoil varied, by up to two-fold (Figure 6.1a, b). In contrast, mineral N contents within the two unfertilised soil replicates showed little variation (Figure 6.1c, d).

Following eight weeks incubation, final mineral N content (T_1) in fertilised soil was between three and twenty three times larger than those measured in unfertilised soil. In all four topsoils studied, there was no consistent effect of either temperature or moisture on mineral N content. However, differences between soil collected from fertilised and unfertilised replicates were most pronounced at the higher temperatures of 10, 16 and 22 °C.

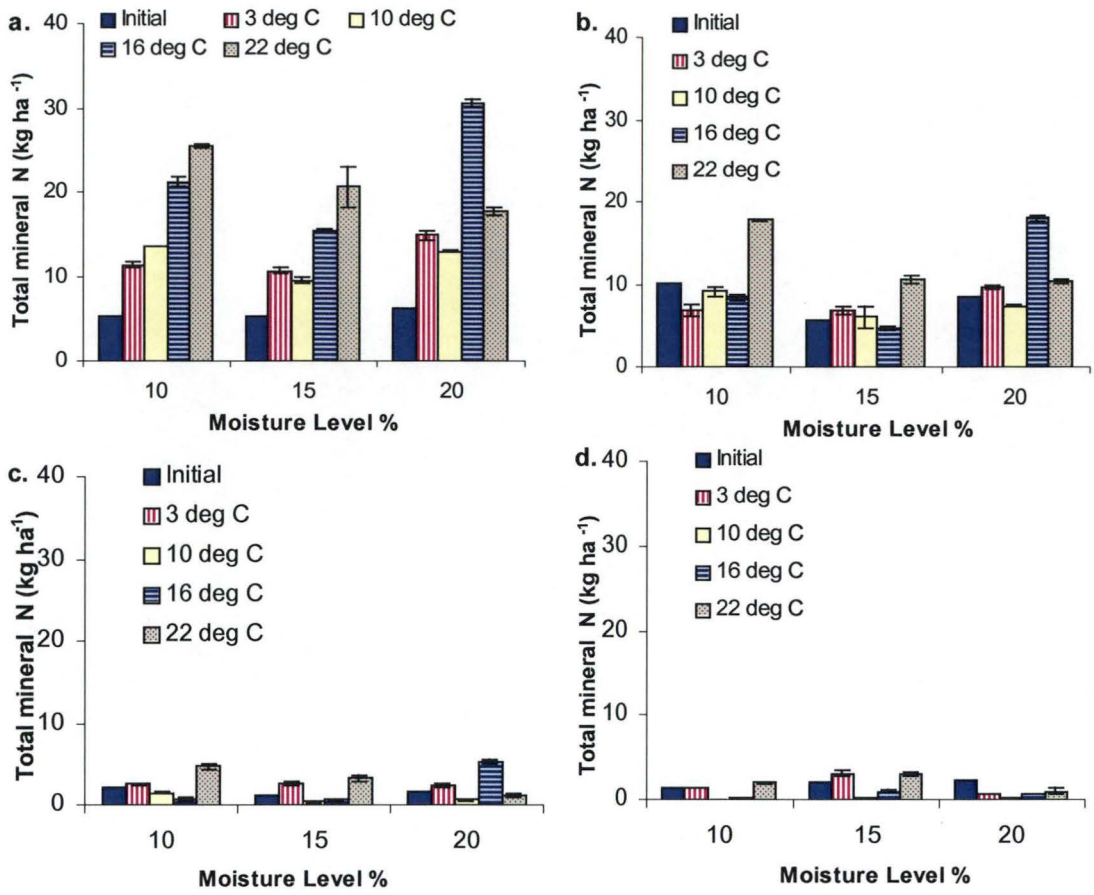


Figure 6.1 Mineral N content of the Kurosol topsoil initially and after two months incubation at 3, 10, 16 or 22 °C, in (a) (P)N1Y plot-1, (b) (P)N1Y plot-2, (c) NIL plot-1, (d) NIL plot-2. Bars indicate standard error (SE) between replicate vials.

Rates of NNM were calculated by subtracting initial mineral N content (T_0) from the final mineral N content (T_1) (see Section 4.2.4 for further details). Due to the relatively high initial mineral N content of plot-2 compared to plot-1, plot-2 topsoil had much lower rates of NNM than plot-1 topsoil (Figure 6.2 a & b). In both fertilised topsoils, the highest NNM rate occurred when incubated at 16 °C and 20 percent moisture, a moisture content that was just below field capacity (24 kg ha^{-1} in plot-1 and 10 kg ha^{-1} in plot-2 per 2 months). Rates of NNM in unfertilised topsoil were very low, and often negative (Figure 6.2 c & d). Positive NNM in unfertilised topsoil was often associated with the treatment (temperature and moisture) that produced high NNM in fertilised topsoil. Maximum NNM in unfertilised topsoil was observed when incubated at 16 °C and 20 percent moisture, the same as the fertilised topsoil.

Overall, when comparing all four plots, fertiliser treatment had a significant effect on mean NNM ($p<0.001$) (Table 6.3). Calculated annual NNM varied, due to incubation moisture and temperature, in fertilised topsoil per plot from –20 to 146 kg N ha^{-1} yr^{-1}, and in unfertilised plots between –14 and 21 kg N ha^{-1} yr^{1}. No other treatment had a significant effect at this level of analysis.

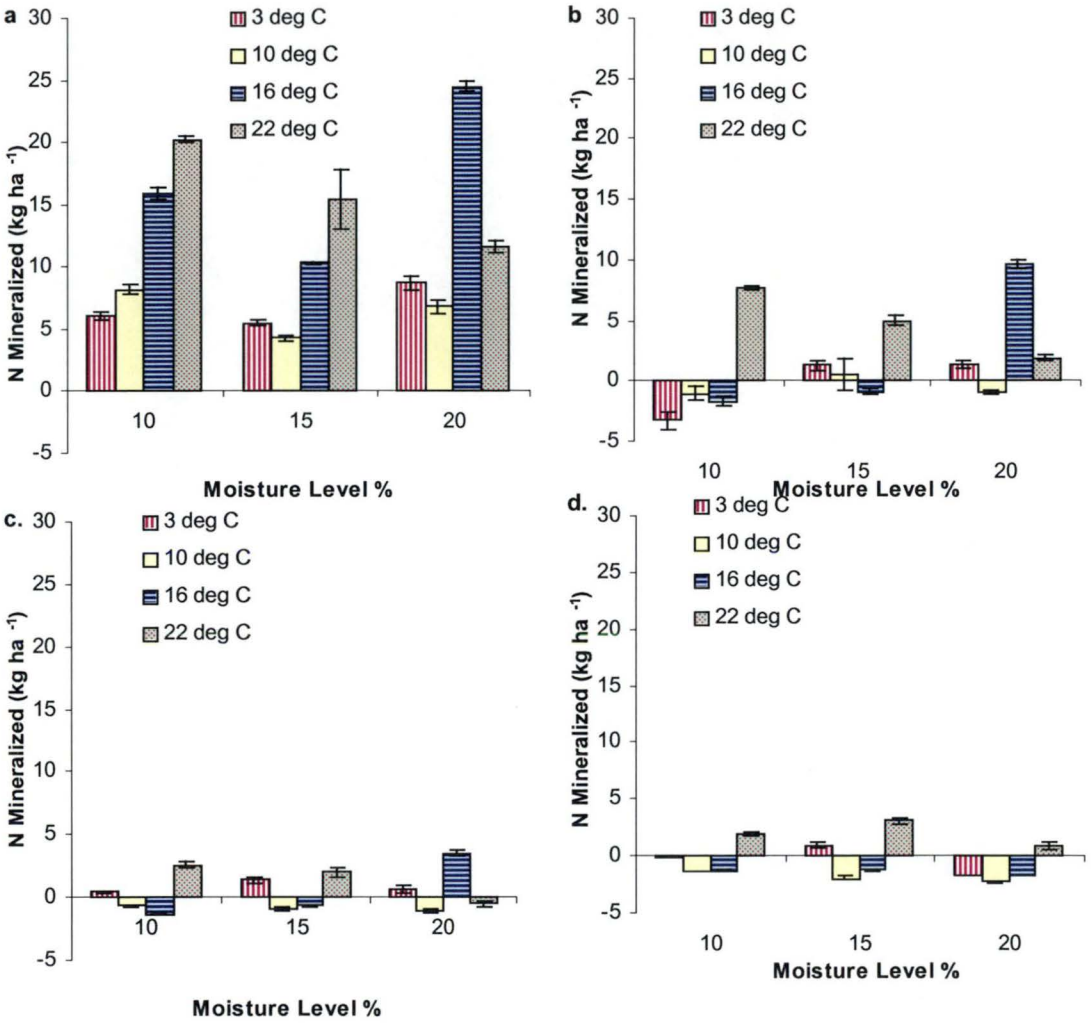


Figure 6.2 Kurosol topsoil net N mineralisation, kg ha^{-1} (a) (P)N1Y plot-1, (b) (P)N1Y plot-2, (c) NIL plot-1, (d) NIL plot-2. Bars indicate SE between replicate vials.

Table 6.3 Analysis of variance comparing mean NNM in Kurosol topsoil from all four plots combined

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Fertiliser	1	299.27	299.27	15.22	<.001
Moisture	2	3.2	1.6	0.08	0.922
Temperture	3	94.64	31.55	1.6	0.196
Fertiliser.Moisture	2	9.12	4.56	0.23	0.793
Fertiliser.Temperture	3	58.72	19.57	1	0.400
Moisture.Temperture	6	84.74	14.12	0.72	0.636
Fertiliser.Moisture.Temperature	6	30.62	5.1	0.26	0.954
Residual	72	1415.54	19.66		
Total	95	1995.85			

Both fertilised and unfertilised topsoil showed an increase in nitrate production at the lower moisture and temperature combinations, but nitrate production was very low (< 0.1 kg N/ha) (Table 6.4). No nitrate was observed prior to incubation.

Table 6.4 Nitrate-N (gram ha^{-1}) in the Kurosol topsoil initially and after two months incubation at various combinations of moisture and temperature. Only positive results are presented.

		Moisture %		
	Temperature (°C)	10	15	20
Fertilized plot-1	Initial			
	3	60		
	10	65	62	
	16			
	22			
Fertilized plot-2	Initial			
	3	33		
	10	53	50	
	16			
	22			
Unfertilized plot-1	Initial			
	3			
	10	46	56	
	16			
	22			
Unfertilized plot-2	Initial			
	3	8		
	10	58	46	
	16			12
	22			

6.3.2 Ferrosol topsoil

Initial mineral N content (prior to incubation, T_0) was similar in the two fertilised (P1YN1Y) and two unfertilised (NIL) topsoils (Figure 6.3). Generally topsoil with the lowest moisture content, just above the permanent wilting point (15 bar), had higher initial mineral N content. This was particularly pronounced in one of the unfertilised plots. Drying unfertilised soil to near wilting point also substantially increased the availability of nitrate prior to and post incubation and at all temperatures (Table 6.5).

Following eight weeks incubation, there were no consistent differences in, final mineral N content (T_1), between fertiliser treatments. Inconsistencies were present between plots and moisture affects. The effect of temperature also varied with fertiliser treatment. Mineral N content in fertilised topsoil generally increased with increasing temperature, while unfertilised topsoil was generally unaffected by temperature.

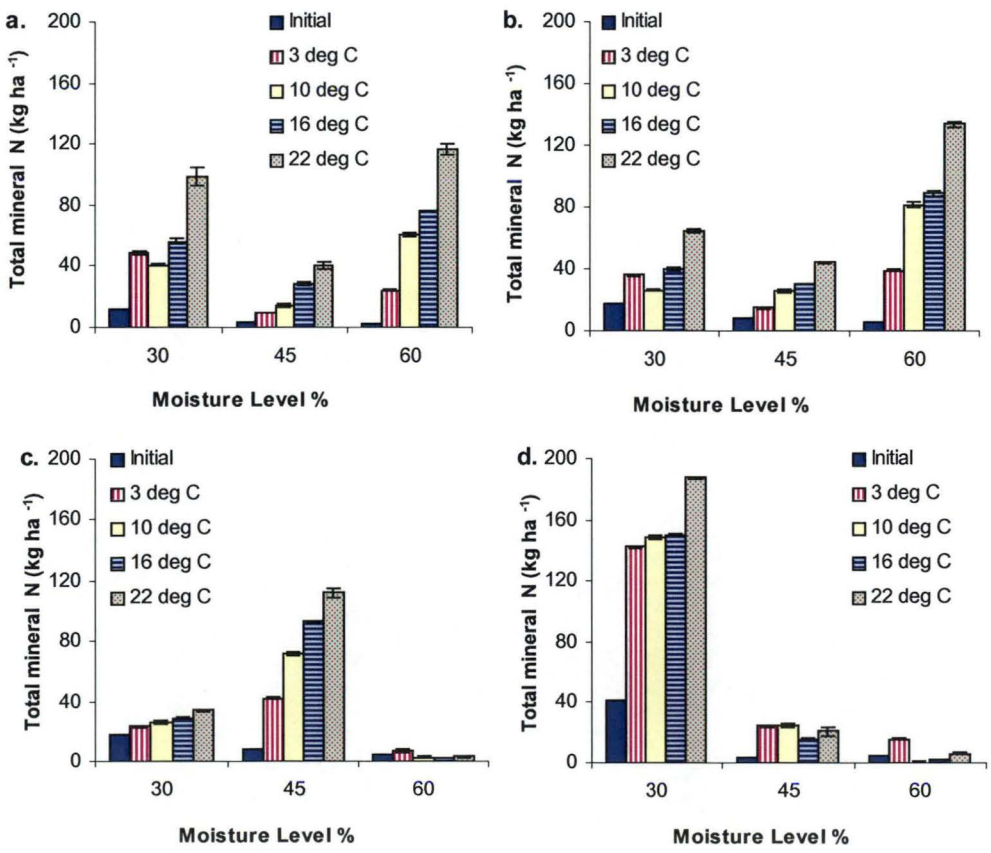


Figure 6.3 Mineral N content of the Ferrosol topsoil initially and after two months incubation at 3, 10, 16 or 22 °C, in (a) P1YN1Y plot-1, (b) P1YN1Y

plot-2, (c) NIL plot-1, (d) NIL plot-2. Bars indicate SE between replicate vials.

Rates of NNM from the two unfertilised topsoils were inconsistent between plots and highly variable, ranging from –4 to 146 kg N ha^{-1} during the two-month incubation (Figure 6.4). In contrast, trends in NNM between the two fertilised plots were similar. In fertilised topsoil, NNM increased with temperature and was slightly depressed at the intermediate moisture content. The optimum temperature for NNM in both fertilised and unfertilised topsoil was 22°C, the highest temperature used. The optimum moisture content was the highest (60%) in fertilised soil, however, this resulted in the highest rate of immobilisation (reduction in N content during the incubation) in unfertilised soil. In contrast to the fertilised topsoil, the inconsistent moisture effects on NNM in the unfertilised topsoil meant that no moisture optimum could be determined.

Table 6.5 Nitrate-N (gram ha^{-1}) in the Ferrosol topsoil initially and after two months incubation at various combinations of moisture and temperature. Only positive results are presented.

		Moisture %		
	Temperature (°C)	30	45	60
Fertilized plot-1	Initial			
	3			
	10	13	5	
	16	1		
	22			
Fertilized plot-2	Initial			
	3			
	10	20		
	16			
	22			
Unfertilized plot-1	Initial	72		
	3	117	10	
	10	124	33	
	16	128	29	
	22	130	13	
Unfertilized plot-2	Initial	30		
	3	501	0	
	10	111	4	
	16	444	0	
	22	443	0	

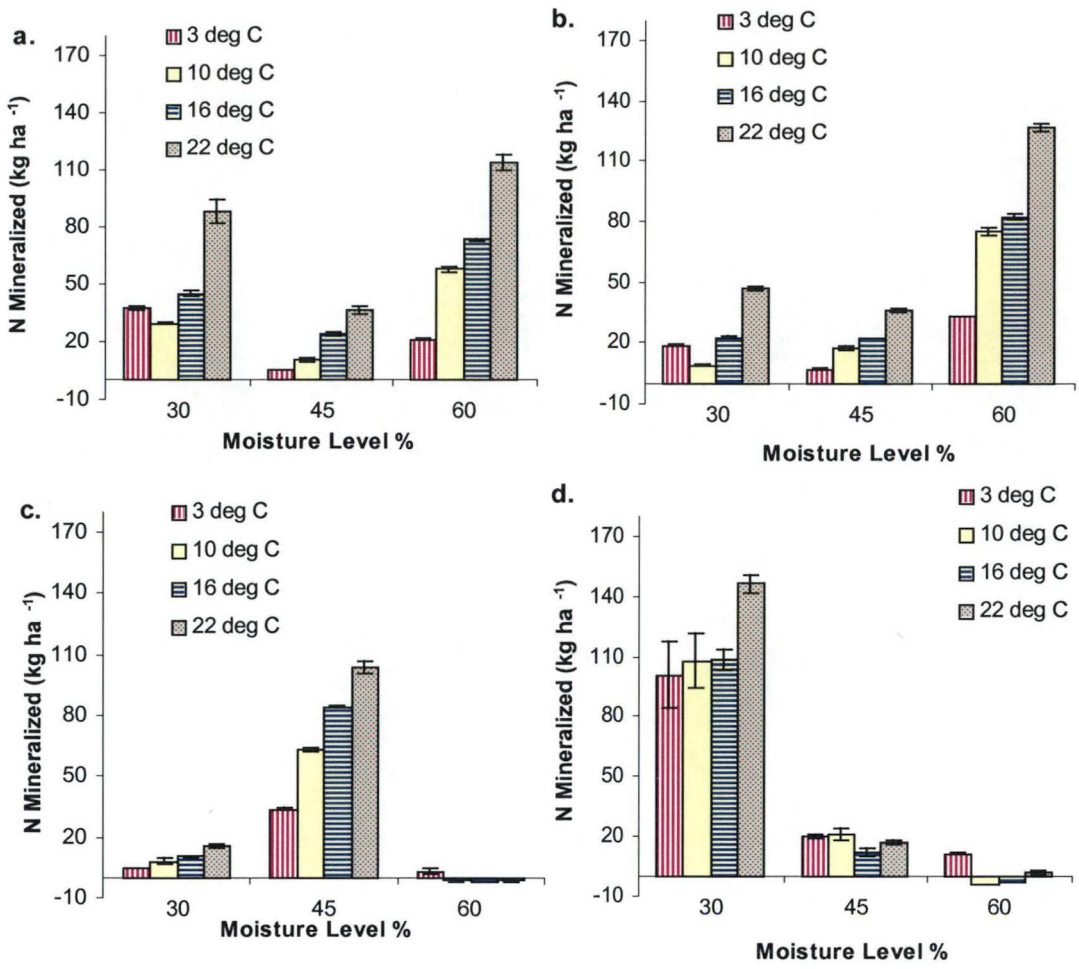


Figure 6.4 Ferrosol topsoil net N mineralisation, kg ha^{-1} (a) P1YN1Y plot-1, (b) P1YN1Y plot-2, (c) NIL plot-1, (d) NIL plot-2. Bars indicate SE between replicate vials.

Overall, when comparing all four plots, no significant difference in mean NNM was observed between fertiliser treatments (Table 6.6). Annual NNM rates ranged between 34 and 764 kg N ha^{-1} yr^{-1} in fertilised topsoil and between –24 and 621 kg N ha^{-1} yr^{1} in unfertilised topsoil. There was a significant interaction between fertiliser and moisture treatment ($p<0.05$) where at the highest moisture treatment (60%) resulted in mean NNM in fertilised soil higher than those unfertilised, while at the lower moisture contents mean NNM was not significantly different.

Table 6.6 Analysis of variance comparing mean NNM in Ferrosol topsoil from all four plots combined

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Fertiliser	1	236	236	0.19	0.660
Moisture	2	1759	879	0.73	0.487
Temperture	3	4396	1465	1.21	0.313
Fertiliser.Moisture	2	13524	6762	5.58	0.006
Fertiliser.Temperture	3	1309	436	0.36	0.782
Moisture.Temperture	6	344	57	0.05	1.000
Fertiliser.Moisture.Temperature	6	1677	280	0.23	0.965
Residual	72	87240	1212		
Total	95	110484			

6.3.3 *Pinus radiata* litter

Initial mineral N content (prior to incubation, T_0) in fertilised ((P)N1Y) litter (O2) was generally ten times greater than unfertilised (NIL) litter (Figure 6.5). However, there was also a large variation, between litters from the two fertilised plots and between litters from the two unfertilised plots.

Following eight weeks of incubation, final mineral N content (T_1) was significantly greater in litter from the two fertilised than litter from the two unfertilised plots. Net N mineralisation rates ranged between 7 and 18 kg ha^{-1} in fertilised litter, while in unfertilised litter NNM rates were generally less than 1 kg ha^{-1}, or negative (Figure 6.5). Net N mineralisation rates increased with increased incubation temperature in both fertilised and both unfertilised litters. As a result, differences in NNM rates between fertilised and unfertilised litter were more pronounced at the higher temperature. In contrast, there were no consistent moisture effects on NNM.

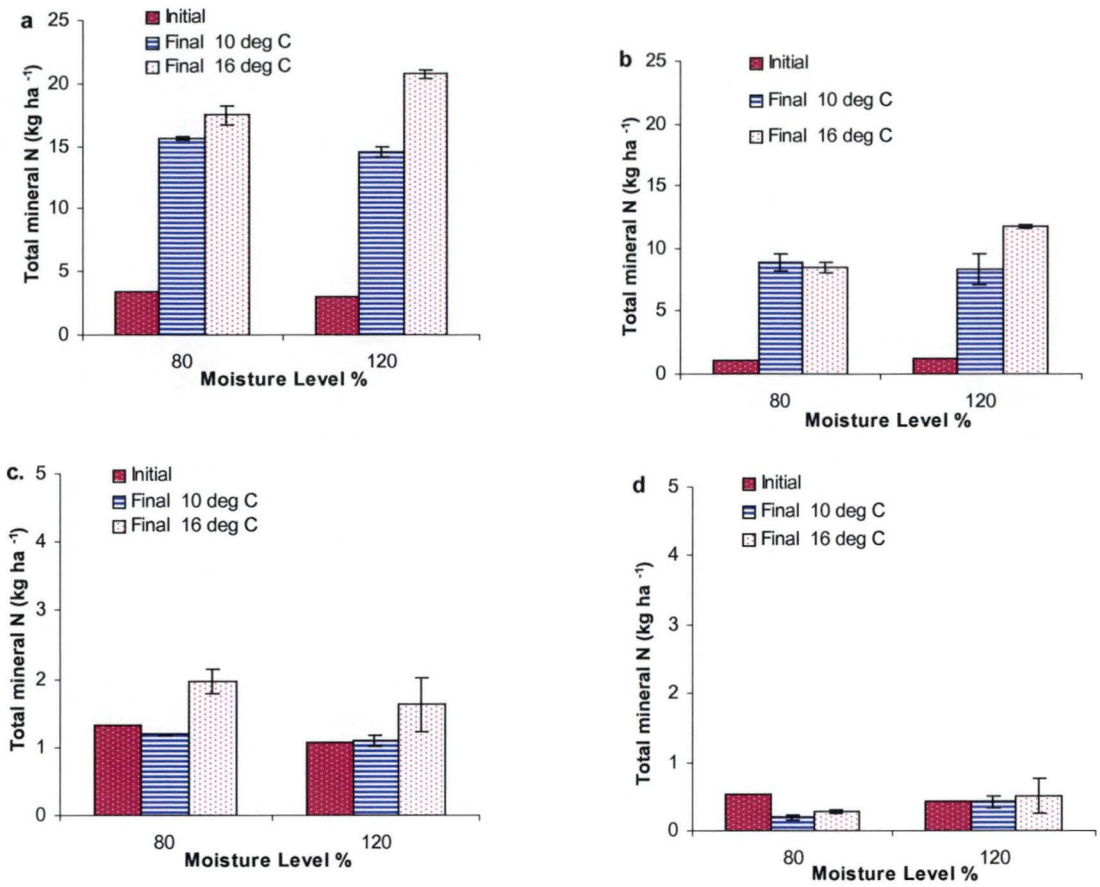


Figure 6.5 Mineral N content (N kg ha^{-1}) of *P. radiata* litter initially and after two months incubation at 3, 10, 16 or 22 °C, in (a) (P)N1Y plot-1, (b) (P)N1Y plot-2, (c) NIL plot-1, (d) NIL plot-2. Bars indicate SE between replicate vials.

Overall, when comparing all four plots, fertiliser treatment had a significant effect on mean NNM ($p<0.001$) (Table 6.7). Annual rates of NNM in the unfertilised litter ranged from –2.1 to 3.9 kg ha^{-1} yr^{-1} compared to 42.8 to 106.2 kg ha^{-1} yr^{-1} in fertilised litter. The maximum NNM rate occurred at the higher temperature and moisture content, 16°C and 120 %, for both fertiliser treatments. Moisture and temperature had no significant effect on mean NNM across all plots.

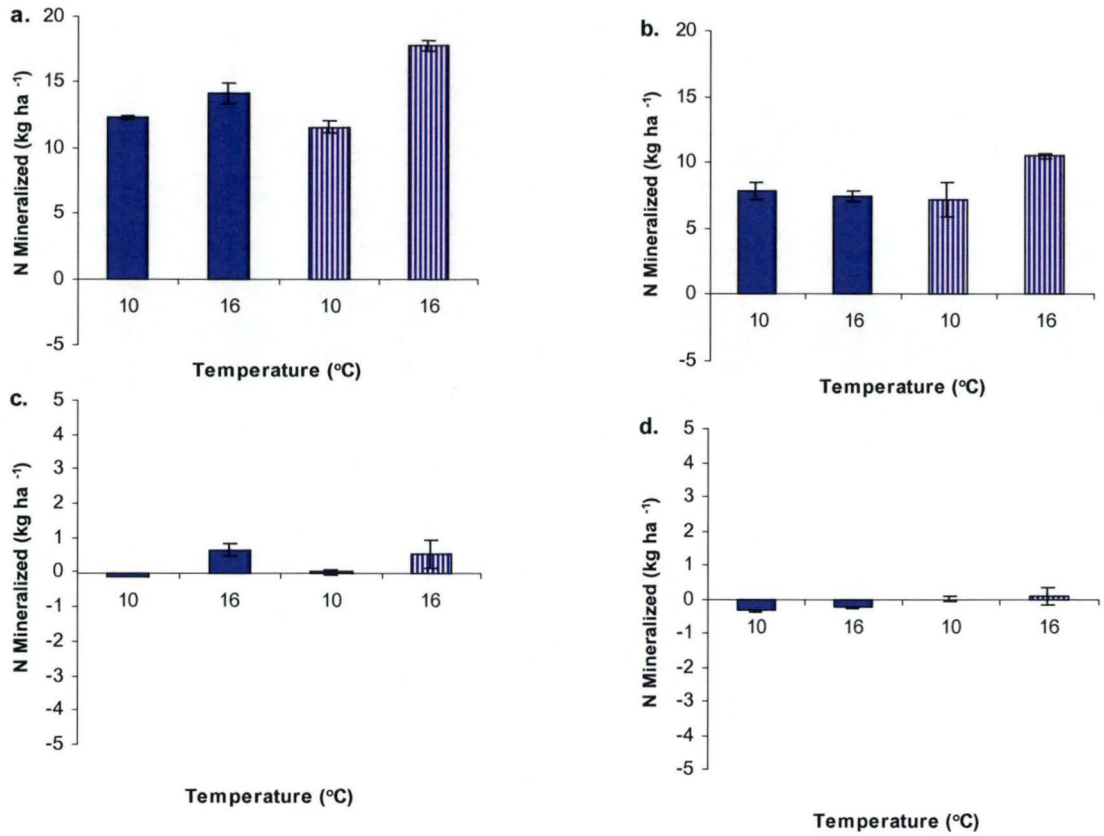


Figure 6.6 *Pinus radiata* litter net N mineralisation, kg ha^{-1} in (a) (P)N1Y plot-1, (b) (P)N1Y plot-2, (c) NIL plot-1, and (d) NIL plot-2. Solid columns are moisture content of 80%, hashed are 120%. Bars indicate SE between replicate vials.

Table 6.7 Analysis of variance comparing mean NNM in *P. radiata* litter from all four plots combined

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Fertiliser	1	485.665	485.665	56.68	<.001
Moisture	1	2.207	2.207	0.26	0.625
Temperture	1	9.753	9.753	1.14	0.317
Fertiliser.Moisture	1	1.251	1.251	0.15	0.712
Fertiliser.Temperture	1	5.622	5.622	0.66	0.441
Moisture.Temperture	1	3.892	3.892	0.45	0.519
Fertiliser.Moisture.Temperature					
	1	4.414	4.414	0.52	0.493
Residual	8	68.546	8.568		
Total	15	581.35			

Table 6.8 *Pinus radiata* nitrate-N (gram ha^{-1}) measured initially and after 2-month incubations at given moisture (%) and temperature (^{o}C). Only positive results presented.

		Moisture	
	Temperature	80	120
Fertilized plot-1	Initial	8	
	10	30	10
	16		36
Fertilized plot-2	Initial	4	6
	10	24	18
	16		31
Unfertilized plot-1	Initial	1	
	10	1	1
	16		19
Unfertilized plot-2	Initial		4
	10		5
	16		13

6.3.4 *Eucalyptus regnans* litter

Initial mineral N content (prior to incubation, T_0) in fertilised (P1YN1Y) litter (O2) (Figure 6.7a, b) were between 5 and 50 times larger, compared to those measured in the unfertilised (NIL) litter (Figure 6.7c, d). Increased moisture content prior to incubation decreased mineral N content in all litters, and generally enhanced the variation in mineral N content between fertiliser and unfertilised plots. Differences between fertiliser treatments were increased at the end of the eight-week incubation (T_1).

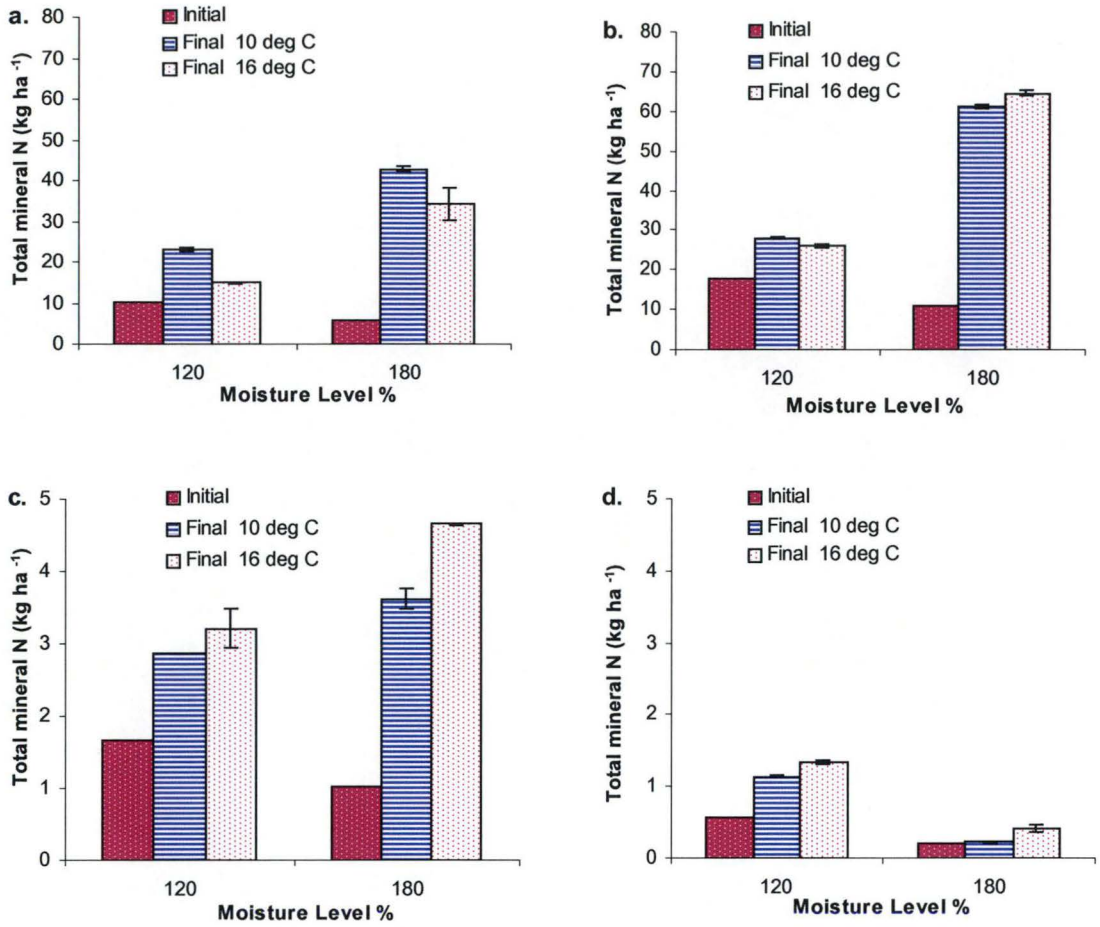


Figure 6.7 Mineral N content (N kg ha^{-1}) of *E. regnans* litter initially and after two months incubation at 3, 10, 16 or 22 °C, in (a) P1YN1Y plot-1, (b) P1YN1Y plot-2, (c) NIL plot-1, (d) NIL plot-2. Bars indicate SE between replicate vials.

Rates of NNM (T_1-T_0) in litter from the two fertilised plots (Figure 6.8a, b) were between 5 and 33 times greater than unfertilised plots (Figure 6.8c, d). There were no consistent effects of temperature or moisture across all four litters. However, higher moisture content increased NNM and the highest NNM rate in all four plots occurred when the litter was incubated at 16°C and 180% moisture.

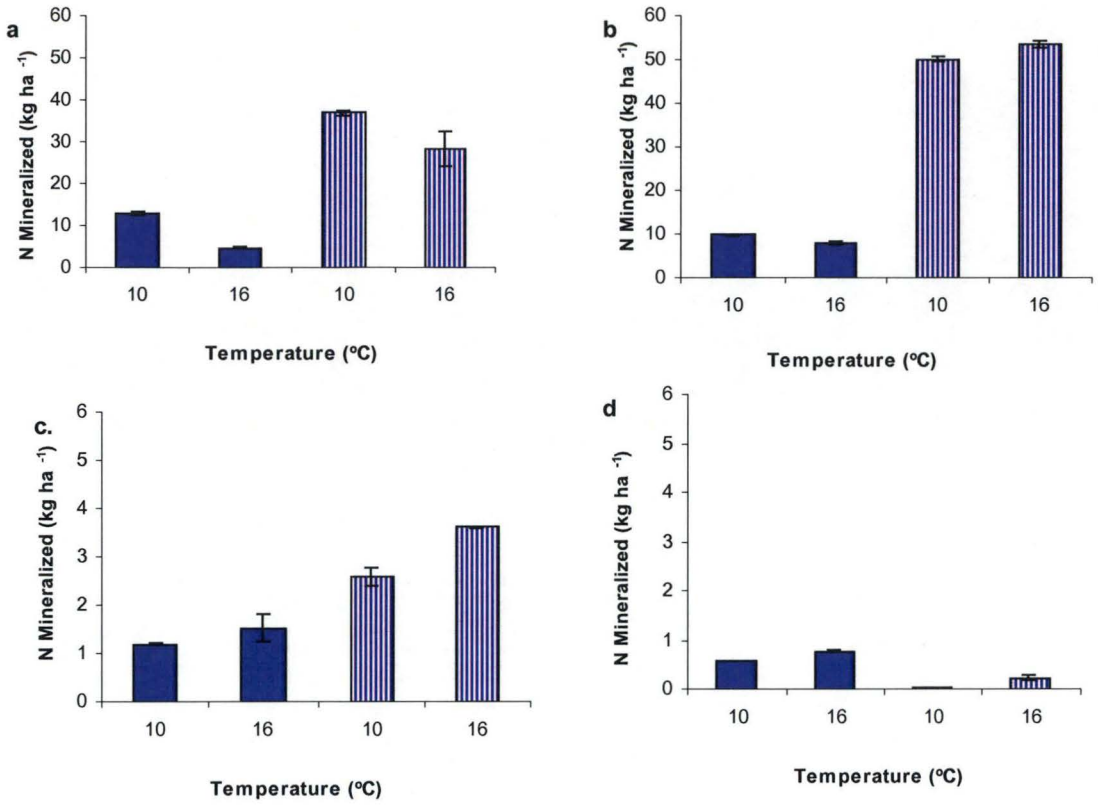


Figure 6.8 *Eucalyptus regnans* litter net N mineralisation, kg ha^{-1} (a) P1YN1Y plot-1, (b) P1YN1Y plot-2, (c) NIL plot-1, (d) NIL plot-2. Solid columns are moisture content of 120%, hashed are 180%. Bars indicate SE between replicate vials.

When comparing all four plots, fertiliser treatment had a significant effect on mean NNM ($p<0.001$) (Table 6.9). Annual rates of NNM in the fertilised litter ranged from 28.2 to 320 kg ha^{-1} yr^{-1} compared to 0.2 to 21.7 kg ha^{-1} yr^{-1} in unfertilised litter. There was no significant effect of fertiliser treatments on nitrate content, which increased slightly by the end of the two-month incubation (Table 6.10). Moisture also significantly increased mean NNM rates across all four plots ($p<0.05$). There was no interaction between moisture and temperature, but there was an interaction between moisture and fertiliser treatment, with fertilised litter significantly increasing with increasing moisture, while unfertilised remained constant. No interaction between treatment, temperature and moisture occurred.

Table 6.9 Analysis of variance comparing mean NNM in *E. regnans* litter from all four plots combined

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Fertiliser	1	2353.61	2353.61	44.38	<.001
Moisture	1	1147.54	1147.54	21.64	0.002
Temperture	1	11.67	11.67	0.22	0.651
Fertiliser.Moisture	1	1068.78	1068.78	20.15	0.002
Fertiliser.Temperture	1	18.48	18.48	0.35	0.571
Moisture.Temperture	1	1.99	1.99	0.04	0.851
Fertiliser.Moisture.Temperature					
	1	1.13	1.13	0.02	0.888
Residual	8	424.29	53.04		
Total	15	5027.47			

Table 6.10 *Eucalyptus regnans* litter nitrate-N (grams ha^{-1}) initially measured and after two month incubation at a given moisture (%) and temperature (^{o}C). Only positive results presented.

		Moisture	
	Temperature	120	180
Fertilized plot-1	Initial		10
	10		37
	16		1
Fertilized plot-2	Initial		
	10	2	9
	16	3	1
Unfertilized plot-1	Initial		
	10	7	49
	16	44	2
Unfertilized plot-2	Initial		3
	10	1	6
	16	6	

6.4 Discussion

6.4.1 *Pinus radiata* litter and Kurosol topsoil

Net N mineralisation in the litter reflected increased N inputs due to fertilisation, which was not always present in topsoil. Previous research indicated a tendency for enhanced rates of NNM in the soil after fertilisation to be relatively short-lived (Williams, 1972; Johnson *et al.*, 1980; Adams and Attiwill, 1983), and depends on the form and frequency of fertilisation (Heilman, 1974; Strader and Binkley, 1989). During current field studies this was also the case, with *in situ* NNM not significantly higher after long-term fertilisation at either site (Chapter 4) and mineral N concentration declined to pre-fertilised levels within five months (Chapter 5). However, results presented in this chapter demonstrated that under controlled

laboratory incubations, significant differences in NNM due to fertilisation could be observed.

Under *P. radiata*, both the fertilised litter and soil had significantly higher rates of NNM than those unfertilised. Increased N cycling in litter due to fertilisation corresponded to a large increase in the total N content and NNM per gram of the O2 horizon (Chapter 3). Previous authors have also observed similar trends from laboratory incubations of soil. Connell *et al.* (1995) observed that heavy fertilisation almost doubled the rate of soil N mineralisation during laboratory incubations. Higher rates of NNM in soil were observed six years after the final application of N fertiliser (860 kg N ha^{-1} applied during a period of 7 years) by Smolander *et al.* (1998), and 22 years after the application of ammonium nitrate (470 kg N ha^{-1}) to a Douglas fir plantation by Strader and Binkley (1989). In agreement with observations in Strader and Binkley (1989) study, increases in NNM from fertilisation in the current study were low, at less than 50 μg g^{-1}. In contrast, increases in NNM in the O2 horizon were up to almost 200 μg g^{-1} (from a maximum concentration of less than 10 μg g^{-1} observed in the unfertilised litter to 195 μg g^{-1} in the fertilised). In the *P. radiata* litter, NNM increased significantly due to fertilisation both due to the increase in total mass of the litter and the rate of NNM per gram of litter.

Significant increases in Kurosol topsoil NNM due to fertilisation determined in the laboratory contrasted to results observed in the field. Although *in situ* rates of NNM were on average four times higher in fertilised topsoil in the field, this was not significant due to large plot variations (Chapter 4). Both the field and laboratory studies highlighted variations between fertiliser plots at this site. Annual rates of NNM in fertilised soil varied between 22 and 82 kg N ha^{-1} yr^{-1} in the field, and between –20 and 146 kg N ha^{-1} yr^{-1} in the laboratory, dependent on the plot studied, incubation temperature and moisture content. In both field and laboratory experiments, NNM rates were up to 20 times higher in plot-1 than plot-2. Such variations indicate that on sites selected as being reasonably uniform (Chapter 3) variation in NNM was very high.

In the laboratory, the effect of the incubation temperature and moisture content varied between the litter and the topsoil. Net N mineralisation in the topsoil showed little variation between 3 and 10 °C, while the highest NNM rates occurred at one of the two higher temperatures (16 and 22 °C). These results concur with Nadelhoffer *et al.* (1991) in a study of six Alaska soils, where C and N mineralisation during aerobic laboratory incubations were insensitive to temperatures between 3 and 9 °C, but increased by a factor of two or more between 9 and 15 °C. Their study also noted that differences in mineralisation rates among soils were greater than differences due to incubation temperature in a single soil.

In the unfertilised topsoil NNM decreased with increasing temperatures from 3, 10 and 16 °C, while the fertilised soil generally increased with increasing temperature. Such trends suggest a change in the microbial population due to long-term fertilisation. In a comparison of soil NNM rates from four forested sites in Michigan, MacDonald *et al.* (1995) observed a significant site, and site by temperature, interaction during laboratory incubations. They observed that differences in NNM a rate between sites were often increased with increasing laboratory temperature, between 5 and 25 °C. Changing temperate optima for microbial populations was also highlighted by Richards *et al.* (1985), their laboratory study revealed distinct microbial ammonifying populations operating at different optimal temperatures, either 15 or 46 °C. Such distinct populations highlighted that a temperature response curve for N mineralisation was actually a composite curve of several different microbial communities. In a review of research on a range of studies, Kirschbaum (1995) concluded that temperature sensitivity of soil processes (as expressed by the Q_{10} function) was not constant across a range of temperatures, but was far greater at low (<10 °C) than at moderate to high (20-30 °C and above) temperatures.

The effect of moisture on NNM rates in the various fertiliser-temperature combinations was not significant in either the Kurosol topsoil or *P. radiata* litter. At this drier *P. radiata* site the Kurosol topsoil underwent constant large moisture fluctuations in the field, and the lowest incubation moisture content, just above permanent wilting point, was not uncommon in the field (Chapter 4).

In this study only the O2 layer was examined in the laboratory, as this layer was a significant proportion of the total litter present (Chapter 3). The aim of this study was to examine the long-term effects of fertilisation on N cycling within each plantation, rather than the immediate effects of the last fertiliser application. Incorporation of the Ol layer into samples would have increased the immediate litter fall influence on NNM (Bauhus, 1996). In addition, field studies had indicated low rates of mineralisation at these sites (Chapter 4), since I measured NNM only, I could not determine whether low gross rate of N mineralisation or high rate of N immobilisation occurred. In their study Fyles and McGill (1987) indicated that addition of recent litter material favours N immobilisation whereas mineralisation dominates in the O2 layer. Aber and Melillo (1980) also reviewed many studies demonstrating fresh litter is a mineral N sink, rather than a source. To prevent increased rates of immobilisation depressing the actual amounts of mineral N available in the field, the O1 layer was not incorporated into the litter samples during laboratory incubations.

6.4.2 *Eucalyptus regnans* litter and Ferrosol topsoil

In agreement with the *in situ* rates of N mineralisation determined in Chapter 4, fertiliser treatment had no significant effect on laboratory NNM rates in Ferrosol topsoil. In contrast, laboratory incubation of the decomposing litter horizon resulted in significantly higher rates of NNM in fertilised litter, generally ten times greater than the unfertilised litter.

Increased N cycling in litter due to fertilisation corresponded to large increases in total N content in O2 horizons and increases in NNM (Chapter 3). Increases in NNM due to fertilisation were associated both with increases in the amount of litter and the rate of NNM within the litter. The average rate of NNM across all moisture and temperatures treatments increased three-fold, from 90 to 327 $\mu g\ g^{-1}$ due to fertilisation. As seen in both studies (reported here and in Chapter 3) increases in N and NNM detected in litter were not always observed in underlying soil, indicating that the litter layer may represent a clearer picture of long-term changes in N cycling at a site, than the soil. Previous investigations have also noted large increases in litter fall N, following N fertilisation (Hunter and Hoy, 1983; Nohrstedt, 1990; Fife and

Nambiar, 1997; Neilsen and Lynch, 1998), but few investigations noted significant changes in soil.

In the laboratory, increased temperatures generally increased NNM in Ferrosol topsoil. This is in agreement with previous research, where inorganic N release increases with increasing temperature between 5 and 40 °C (Theodorou and Bowen, 1983a; Foster, 1989; Beier and Eckerersten, 1998). Temperature affects were expected as temperature exerts strong control over both the physiological activity of soil micro-organisms and their ability to access substrate pools (Ellert and Bettany, 1992; MacDonald *et al.*, 1995). However, the effect of temperature on N immobilisation may result in negative NNM at higher temperatures (Kurosol topsoil, Section 6.4.1).

Large differences in soil moisture content between fertiliser treatments were previously reported (Chapter 4). Differences were associated with increased water uptake by larger trees on the fertilised sites. The importance of soil water availability in controlling the rate of litter decomposition was reviewed by Sommers *et al.* (1981). Rates of microbial processes were generally more rapid near field capacity and linearly decline as water matric potential became more negative (MacKay and Carefoot, 1981; Linn and Doran, 1984). In this study, the effects of moisture on rates of NNM were significant, across fertiliser treatments in *E. regnans* litter and Ferrosol topsoil. The strong effect of moisture on NNM at this site was previously noted in the *in situ* study from Chapter 4, where N mineralisation was limited when moisture additions through rainfall were restricted in closed cores. Strong *et al.* (1998a) also indicated that water status strongly limited N mineralisation even at the high water potential of 0.3 and 0.1 bar (-30 to –10 kPa). By reducing moisture content from –30 to –10 kPa they observed a mean NNM decrease of 16 per cent. However, in the current study the effect of moisture on NNM in the Ferrosol topsoil varied, dependent on the scale of change and previous fertiliser treatment. Paul *et al.* (2003b) also highlighted this function when defining the relationship between soil water content and NNM. In their review they noted that the effect of water content on mineralisation

was dependent on substrate quality. The more easily decomposable the substrate was the more sensitive mineralisation was likely to be to water content.

There was an obvious effect of initial sample disturbances, particularly drying. This effect was most pronounced in unfertilised Ferrosol soil dried to near permanent wilting point. Under such treatment, mineral N content, prior to incubation, was enhanced by up to ten-fold. As no re-wetting occurred, enhanced mineral N due to drying, may reflect bacterial cell lysis, releasing N, or microbial population changes as noted in previous research (Stevenson, 1956; Birch, 1958; Lund and Goksoyr, 1980; Van Gestel *et al.*, 1993). In contrast, rates of NNM at near field capacity moisture content varied greatly between treatments, resulting in immobilisation in unfertilised soil and NNM rates up to 438 kg N ha^{-1} yr^{-1} in fertilised topsoil. Discrepancies amongst topsoils and fertiliser treatments due to differential drying effects on mineral N availability indicate that these effects should be considered in prediction systems for NNM rates. Previous examination of N mineralisation has shown enhancement of C and N due to soil disturbance may result in NNM overestimation (Van Gestel *et al.*, 1993; Murphy *et al.*, 1998b; Pulleman and Tietema, 1999) . However, in soils with a wide range of physical and chemical characteristics under karri forests in southern Western Australia, O'Connell and Rance (1999) were able to use disturbed soils samples to estimate *in situ* NNM rates by plus or minus 20 percent. Incubation of intact cores has been suggested to better reflect field conditions (Adams and Attiwill, 1986; Fyles and McGill, 1987; Raison *et al.*, 1987). Intact cores also maintained the spatial separation between surface litter and humified material lower in the profile, thereby eliminating the possibility than N mineralised from decomposed litter O2 layer material would, for example, be immobilised by undecomposed litter brought into close contact by mixing. Under these circumstances, rates of N mineralisation could be seriously underestimated (Fyles and McGill, 1987). However, results in this study indicated that an extremely high number of intact soil-core replicates would be required to show this in a statistically consistent manner.

6.5 Conclusion

The effect of long-term fertilisation on soil NNM rates in aerobic laboratory incubations was not consistent across sites or various water-temperature combinations, but, at both sites:

- Long-term fertilisation significantly increased mineral N availability and NNM in litter. Depending on the moisture content and temperature at which the samples were incubated, fertilised litter had NNM rates up to 50 times greater (per horizon) than those unfertilised. Increases in NNM due to fertilisation were associated both with increases in the amount of litter and the rate of NNM within the litter. This highlights the importance of site management practices that conserve this high-nutrient component of the forest ecosystem.
- In agreement with *in situ* studies, long-term fertilisation did not significantly increase NNM in Ferrosol topsoil. However, long-term fertilisation did significantly increase NNM in Kurosol topsoils when incubated under control temperatures of 16 °C.

The influence of moisture and temperature on mineral N availability and NNM was strongly site dependent.

- In general, the Ferrosol site, with its higher inherent fertility reacted more strongly to incubation parameters than the Kurosol site.
- Moisture had a significant effect on both initial N availability and NNM in Ferrosol topsoil.
- Changes prior to laboratory incubations, such as air-drying, caused large increases in mineral N in the Ferrosol topsoil, while the effects of temperature on NNM increased at high moisture content.

In laboratory studies, it is well recognised that environmental conditions must be controlled to obtain reproducible estimates of NNM (MacKay and Carefoot, 1981). To obtain an estimate of the overall productivity of a site and the fluxes of N it is necessary to determine optimum rates of NNM. Interactions between moisture availability and temperate are an important factor in the understanding of the overall

processes involved in N mineralisation. The trends observed in this study emphasise the role of the site when selecting incubation parameters. When relative high fertility sites are being measured, the incubation parameters of temperature and moisture could result in variations from N immobilisation to a 500-fold increase in NNM. Even at the relatively poor fertility Kurosol site, this trend was observed when comparing, N-enriched fertilised soil to those unfertilised. Consequently, clear optimum incubation parameters could not be determined. However, general trends indicate that maximum rates of NNM in the laboratory were measured at temperatures between 16 and 22 °C at moisture between 0.33 and 3 bar. The effects of temperature, moisture and timing on NNM rates are investigated further in the following laboratory studies (Chapters 7).

Chapter 7. Effects of incubation period, air-drying and long-term periodic fertilisation on laboratory net nitrogen mineralisation in topsoil and litter samples from two contrasting sites.

7.1 Introduction

Although laboratory rates of NNM have been suggested as an indicator of N availability for tree growth (Adams and Attiwill, 1986), the previous laboratory study (Chapter 6) indicated that *in vitro* rates of NNM depended on the incubation parameters used relative to the site being studied.

Both the field study (Chapter 4) and laboratory study (Chapter 6) of NNM rates showed that changes in moisture availability in Ferrosol topsoil could have a strong influence on rates of *in situ* and *in vitro*. This contrasted with the lack of a significant effect of moisture on NNM in the Kurosol topsoil. Moisture effects in the Ferrosol soil appeared to be greatest when the soils were pre dried to levels just above permanent wilting point (~15 bar). It is important to quantify just how this pre-drying might affect rates of NNM in this and other studies, as many laboratory studies, have pre dried, sieved, and then re-moistened samples prior to standard laboratory incubations (Richards *et al.*, 1985; Bonde and Rosswall, 1987; Robertson *et al.*, 1988; Ross *et al.*, 1995). Thus an experiment was designed to test this. During this research (Chapter 4, 4 and 5) all topsoil and litter samples, including samples collected from the field were maintained moist prior to incubations and N extractions. Differences in sample preparation in the laboratory such as pre-treatments including air-drying may partly explain differences in rates of N mineralisation behaviour in this study as compared to published studies by other workers.

The effects of pre-treatments such as air-drying and sieving on N availability in a soil result from a modification of key physical, chemical and biological processes. These all influence organic matter decomposition and hence N release (Birch, 1958; Bartlett and James, 1980; Seneviratne, 1985; Strong *et al.*, 1999b). Air-drying pre-treatments may therefore alter C and N mineralisation-immobilisation processes, microbial biomass, and microbial activity. (Stevenson, 1956; Soulides and Allison, 1961; Lund

and Goksoyr, 1980; Van Gestel *et al.*, 1993). Air drying and re-wetting also disrupts the soils physical structure resulting in the release organic compounds available for subsequent mineralisation (Van Gestel *et al.*, 1991; Degens and Sparling, 1995).

The laboratory study presented in Chapter 6 indicated that in contrast to field studies (Chapter 4), under controlled laboratory conditions significant differences between NIL and annual fertilisation (1300 kg N ha^{-1}) could be measured. Previous research has shown changes in NNM from fertilisation depends on the soil nutrient status, type of fertiliser applied, and the individual and cumulative rates applied (Heilman, 1974; Strader and Binkley, 1989; Aggangan *et al.*, 1998). Current management of some ex-forest sites in Tasmania may result in the cumulative addition of 500-700 kg N ha^{-1} during each rotation (Smethurst *et al.*, 2004a). At both study sites, intermediate rates of fertiliser application (every two and four years) resulted in cumulative rates between 400-700 kg N ha^{-1} (Table 3.4 and 3.5). Using these fertiliser treatments I wanted to test whether significant changes in NNM could be measured at lower fertilisation rates using laboratory incubation methods.

Under *E. regnans*, the effect of P application was also investigated. At this site P was applied with N at rates ranging between 184 kg P ha^{-1} and 598 kg P ha^{-1} during thirteen years, and was also applied alone biannually, to a total of 322 kg P ha^{-1} (Table 3.5). Although increases in NNM due to P additions have been noted in some cases e.g. Falkiner (1993), other studies have indicated the opposite or no effect of P fertiliser (Javid and Fisher, 1990; Whynot and Weetman, 1991). Growth analysis of the *E. regnans* plantation (Chapter 3) indicated that P was not limiting. Using these treatments, I therefore wanted to examine whether P additions would significantly effect NNM rates in laboratory incubations.

Rates of NNM in the field (Chapter 4) and during the first laboratory study (Chapter 6) were both assessed using 60-day incubation periods. Previous *in situ* incubation periods between 7 and 90 days have been recommended (Raison *et al.*, 1987; Adams *et al.*, 1989b; Smethurst and Nambiar, 1989b; Goncalves and Carlyle, 1994; Carlyle *et al.*, 1998b). At both sites rates of NNM were expected to be low and a linear increase

NNM over time, as indicated by Raison *et al.* (1987) suggested a incubation period of 60 days was required to measure changes in NNM rates between fertiliser treatments at these sites. However, Raison *et al.* (1987) also stated that exposure should not be so prolonged that accumulation of ammonium may induce nitrification in a system which does not otherwise nitrify significantly. In these systems limited amounts of nitrate were observed in the field (Chapters 3, 4 and 5) and during 60 day laboratory incubations (Chapters 5). The lack of increased nitrate production during incubations indicated that the incubation period was not too prolonged. In field and laboratory incubations of soil, the period of incubation was critical when assessing N transformations (Lund and Goksoyr, 1980; Cabrera and Kissel, 1988c; Sierra, 1992; Pulleman and Tietema, 1999; Paul, 2001). To test that an incubation period of 60 days did not increase immobilisation and therefore was an optimum incubation period for NNM it was therefore necessary to examine the role of incubation period on NNM.

In litter layers, studies by Pulleman and Tietema (1999) and Clein and Schimal (1993) have noted that, during laboratory incubation, N release and microbial activity depend on incubation period. However, investigations into NNM rates in litter are limited. Results reported in previous Chapters (2, 4 and 5) showed that there was a large pool of N available for mineralisation in litter of both experimental sites. To obtain an estimate of the overall productivity of a site and the fluxes of nitrogen it was necessary to understand the role of time, on N release from the litter.

The objectives of this study was to determine the effects of various factors on NNM rate, these included conducting laboratory incubations as follows; (1) impact of pre-treatments such as air-drying, (2) impact of incubation period on NNM in both litter and topsoils and (3) the sensitivity of *in vitro* NNM to various long-term N and P fertilisation. In addition, I wanted to examine whether litter and soil horizon chemical characteristics affected the rates of NNM measured in the laboratory at various temperatures. By studying topsoil and litter from the sites studied previously, I also wanted to test the hypothesis that adaptation to soil moisture conditions in the field might reduce the impacts of pre-treatments like air-drying.

7.2 Methods

This study was split into three experiments; Experiment 1 examined the effect of an air-drying pre-treatment on NNM, Experiment 2 examined the role of incubation period on NNM and Experiment 3 examined the effects of intermediate rates of fertilisation on NNM with treatment rates of 400 kg N ha^{-1} to 700 kg N ha^{-1}.

Both sites were examined in this series of laboratory based incubation experiments, i.e. the *E. regnans* plantation on a Haplic Dystrophic Brown Ferrosol, and the *P. radiata* plantation on a bleached Dystrophic Yellow Kurosol.

7.2.1 Field sampling and laboratory methods

Litter (O2 horizon) and topsoil (A1 horizon) samples were collected in July 2002 from both sites by the methods described in Section 6.2.1. At the *E. regnans* site both plots from each of five fertiliser treatments were sampled, NIL, P4YN4Y, P2YN2Y, P1YN1Y and P2Y (collected from the field 28 days prior to the start of the laboratory incubation, day -28) (Table 7.1). At the *P. radiata* site, both plots from each of four fertiliser treatments were sampled, NIL, (P)N4Y, (P)N2Y, and (P)N1Y (collected from the field 29 days prior to the start of the laboratory incubation, day -29) (Table 7.1). For further information on fertiliser treatments see Chapter 3, Tables 3.4 and 3.5.

All samples were brought back to the laboratory within eight hours and placed in the cool-store at 2-4 °C. Samples were prepared in the laboratory for incubation as per Sections 6.2.2. In contrast to Chapter 6, after the determination of mineral N concentrations from pooled plot litter and topsoil samples, samples from plots of the same fertiliser treatment were thoroughly mixed. Mixing of replicate plots resulted in a composite sample for each litter and topsoil, of each fertiliser treatment, for the two sites. Samples were mixed to obtain a representative sample of each treatment. Homogenised litter and topsoil samples were placed back into cool storage for a further one to two weeks to minimise the influences of disturbance on NNM and

allow for air-drying in experiment 1 (day -14 to –7 and 0). A summary of the initial timing and management of samples is given in Table 7.1.

Individual pooled litter and topsoil samples, for each fertiliser treatment, where thoroughly mixed before sub-sampling into vials for incubation. In each experiment between 30 and 40 grams of samples were placed in 100 ml plastic vials and covered with polyethylene film, as per Chapter 6. All samples were maintained moist (~3 bar) throughout the incubation. All vials in Experiment 1 and 2 were placed in a constant 20 °C incubator for the duration of the experiment. The temperature chosen, 20 °C, corresponded to the highest temperature recorded at 5 cm depth in the topsoil under *P. radiata* (Figure 49). In Experiment 3, vials were incubated at either 10 °C or 20°C for 60 days. Vials were checked for moisture losses fortnightly, by weight. Moisture loss during the incubation was found to be very low (~2 to 4 %). Moisture was adjusted once during the experiment, at day 24.

Table 7.1 Timetable for sample collection, an initial laboratory analysis

Day	Process
-28, -29	Field collection of samples
-28	2 week cool-store (2-4 °C) for sieving equilibration
-14	Sub sample individual pooled, plot, samples for mineral N concentrations. Then combine individual plots to form homogeneous treatment samples. One to two week cool-store, for mixing equilibration.
-7	**Experiment 1**. Start of drying treatment, mineral N extraction, T_{-7} and T_0
0	**Experiment 1**. Re-wetting of air-dried samples, start of incubation. Start of all incubations
3, 7, 15, 30	**Experiment 2**. Mineral N extractions at, T_3, T_7, T_{15} and T_{30} **Experiment 1**. Mineral N extraction T_{30}
60	End of all incubations, final mineral N extractions, T_{60}

7.2.2 Experiment 1

The design of the experiment 1 included factorial combinations of two sites, two horizons (O2 and A1), two fertiliser treatments (annual application and unfertilised) and two pre-incubation treatments (air-dried and maintained moist). There were also two incubation periods (30 and 60 days), three replicates, and 96 vials in total (Table 7.2).

Table 7.2 Summary of treatments in Experiment 1

Location	Southern Tasmania	North-east Tasmania
Topsoil	Ferrosol	Kurosol
Species	*E. regnans*	*P. radiata*
Profile	Litter (O2) Topsoil (A1)	Litter (O2) Topsoil (A1)
N inputs	Fertilised (P1YN1Y) Unfertilised (NIL)	(P)N1Y Unfertilised (NIL)
Drying Treatment (prior to incubation)	Air-dried 7 days **Maintained moist**	Air-dried 7 days Maintained moist
Incubation Time	**30 and 60 days**	30 and 60 days
Temperature	20 °C	20 °C
Moisture content **Litter** **Topsoil**	 120 % (w/w) 45 %	 80 % 15 %

*of samples maintained moist prior to incubation and the moisture content of all samples during incubation.

Moisture contents applied to composite litter and topsoil samples in this study (Table 7.2) were the intermediate levels used in the previous laboratory study (Chapter 6). Moisture was adjusted up by adding a fine mist of distilled water until the required weight was reached, and down, by air-drying in the cool-store. All composite samples were adjusted to the appropriate moisture content within 2 days of the laboratory study commencing (at day –7).

Samples were taken for mineral N concentration determination at days –7 and day zero (T_{-7} and T_0). Nitrogen concentration was determined using the methods detailed in Section 6.2.2. At day –7, samples to be air-dried were placed in large flat open dishes for drying at a constant temperature of 20 °C. Moist samples remained covered in the cool-store. Both moist and drying samples were stirred frequently (day –7 to 0).

At day zero, air-dried samples were then re-wetted, with a fine distilled water mist, until the same moisture content as the 'moist samples' was reached. All samples were incubated moist. Thorough mixing after re-wetting was required to completely disperse water throughout the samples, especially with the Kurosol topsoil, which had a tendency to become water repellent. All samples were mixed before sub-samples were transferred to individual vials for incubation.

7.2.3. Experiment 2

The design of the experiment 2 included factorial combinations of two horizons (litter and soil), two fertiliser treatments (annual application and unfertilised), and five incubation periods (3, 7, 15, 30 and 60 days); each treatment was replicated three times (Table 7.3).

Individual pooled litter and topsoil samples, for each fertiliser treatment, were thoroughly mixed before sub-sampling into vials. All vials were placed in a constant 20 °C incubator for the duration of the experiment (day 0 to 3, 7, 15, 30 or 60).

Table 7.3 Summary of treatments in Experiment 2

Location	Southern Tasmania	North-east Tasmania
Topsoil	Ferrosol	Kurosol
Species	*E. regnans*	*P. radiata*
Profiles	Litter (O2) horizon Topsoil (A1) horizon	Litter (O2) horizon Topsoil (A1) horizon
Nutrient Inputs	Fertilised (P1YN1Y) Unfertilised (NIL)	Fertilised ((P)N1Y) Unfertilised (NIL)
Incubation Time (days)	3 7 15 30 60	3 7 15 30 60
Temperature °C	20	20
Moisture content **Litter** **Topsoil**	 120 % (w/w) 45 %	 80 % 15 %

7.2.4 Experiment 3

The design of the experiment 3 consisted of factorial combinations of two horizons, i.e. litter (O2) and topsoil (A1), five (at *E. regnans* site) or four (at *P. radiata* site) fertiliser treatments, two incubation temperatures (10 °C and 20 °C), and three replicates (Table 7.4). Homogenised samples, between 30 and 40 g were placed into 100 ml plastic vials, covered with polyethylene film, as per Chapter 6. Vials were incubated moist at 10 °C or 20°C for 60 days.

Table 7.4 Summary of treatments in Experiment 3

Location	Southern Tasmania	North-east Tasmania
Topsoil	Ferrosol	Kurosol
Species	*E. regnans*	*P. radiata*
Profiles	Litter (O2) horizon Topsoil (A1) horizon	Litter (O2) horizon Topsoil (A1) horizon
Nutrient Inputs **kg N ha^{-1}**	1300 (P1YN1Y) 700 (P2YN2Y) 400 (P4YN4Y) 0 (P2Y) 0 (NIL)	1300 ((P)N1Y) 700 ((P)N2Y) 400 ((P)N4Y) 0 (NIL)
Moisture content **Litter** **Topsoil**	maintained moist 120 % (w/w) 45 %	maintained moist 80 % 15 %
Incubation Time (days)	60	60
Incubation Temperature	20 °C 10 °C	20 °C 10 °C

7.2.5 Data presentation and analysis

Results of all three experiments were expressed as concentration μg g^{-1}. This allowed the examination of the role of both fertiliser treatments and laboratory parameters on NNM across the sites and avoided variations in litter mass and bulk density from excessively influencing calculations. At the end of the experimental results Table 7.15 presents a summary of all experiments in NNM kg N yr^{-1}.

Results were analysed using a multiple analysis of variance (MANOVA) procedure of the GenStat software (Genstat 5 Committee, 1988). Means were compared using the treatment interactions (Experiment 1. fertiliser and pre-drying, Experiment 2. fertiliser and incubation time and Experiment 3. fertiliser and temperature) least significant differences ($p < 0.05$ and $p < 0.001$), as stated. The relationship between mineral N concentrations and NNM were examined by scatter plot and regression analysis. Linear and non-linear (quadratic) were calculated for untransformed data. Relationships were considered significant when ($p < 0.05$). The R squared statistic is given for significant correlations. Tests were validated by testing data for normality of distribution, and transforming data where required. Residuals from the model for each variable were examined for normality using diagnostic graphs.

7.3 Results

7.3.1 Experiment 1. Effects of laboratory air-drying pre-treatment on NNM.

7.3.1.1 Kurosol topsoil and litter under *P. radiata*

Prior to laboratory incubation (days –7 and 0), there was significantly ($p<0.05$) more mineral N in fertilised than unfertilised Kurosol topsoil (Figure 7.1). At day zero there was no initial effect of drying on mineral N concentrations, all concentrations were low, varying between 0.2 and 2.3 $\mu g\ g^{-1}$. After re-wetting and incubating for 30 days, air-drying significantly ($p<0.05$) increased concentration of mineral N above those maintained moist, in unfertilised, but not the fertilised topsoil (Table 7.5). At day 60, both the fertilised and unfertilised air-dried topsoils had significantly higher ($p<0.001$) mineral N concentrations than those maintained moist.

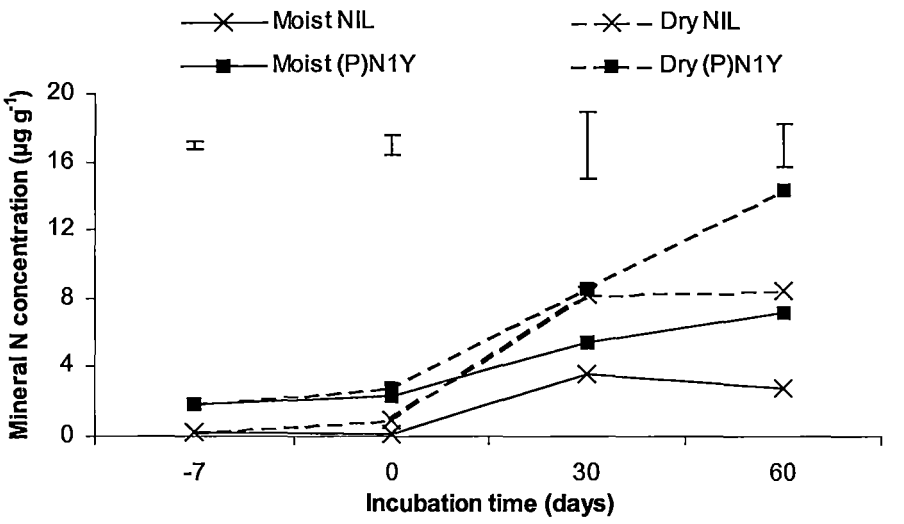


Figure 7.1 Mineral N concentrations (µg g^{-1}) measured in Kurosol topsoil at days –7 to 60. Bars indicate least significant differences (LSD) (p = 0.05) for comparison of fertiliser-by-drying interaction means.

Rates of NNM were similar between fertiliser treatments at day 30, and increased significantly ($p<0.05$) due to air-drying in unfertilised topsoil (Figure 7.2). By day 60 there was a significant ($p<0.001$) increase in both the fertilised and unfertilised topsoils due to the air-drying pre-treatment. There was no interaction between fertiliser and drying treatments at either 30 or 60 days.

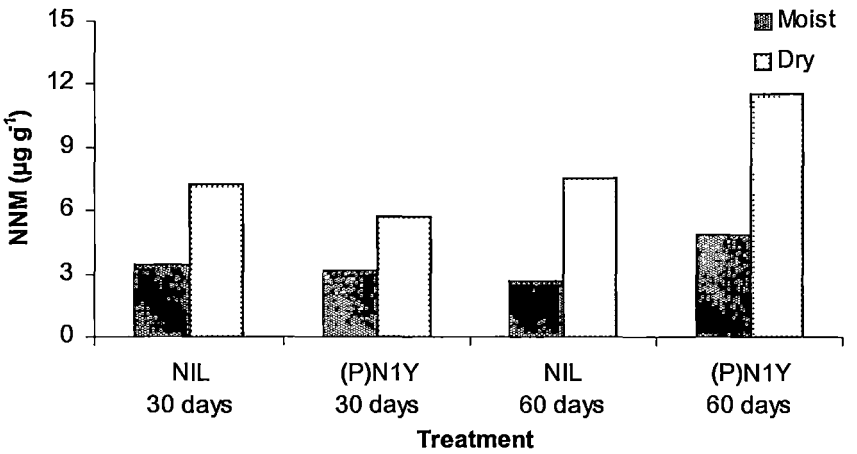


Figure 7.2 NNM (µg g^{-1}) in Kurosol topsoil after 30 and 60 day incubations. There was no significant interaction between fertiliser and moisture treatments.

Table 7.5 MANOVA summery table of treatments (fertiliser and air-drying) effects on Kurosol topsoil NNM, significant differences between means indicated by p values in bold.

	Day	Fertiliser treatment	Drying treatment	Interaction
Daily rate NNM	30	0.48	**0.028**	0.609
	60	**0.004**	**<0.001**	0.306
Cumulative rate of NNM	0	**<0.001**	0.088	0.075
	30	0.379	**0.013**	0.549
	60	**<0.001**	**<0.001**	0.372

Fertilised *P. radiata* litter had mineral N concentrations up to six times higher than that unfertilised ($p<0.001$) (Figure 7.3). In both fertilised and unfertilised litter air-drying significantly increased mineral N concentrations at day zero by around double ($p<0.001$) (Table 7.6). Rates of NNM were similar between air-dried and moist samples at day 30 and day 60 in unfertilised litter (Figure 7.4). However, air-drying had significantly suppressed NNM in the fertilised litter by 20 percent at day 60.

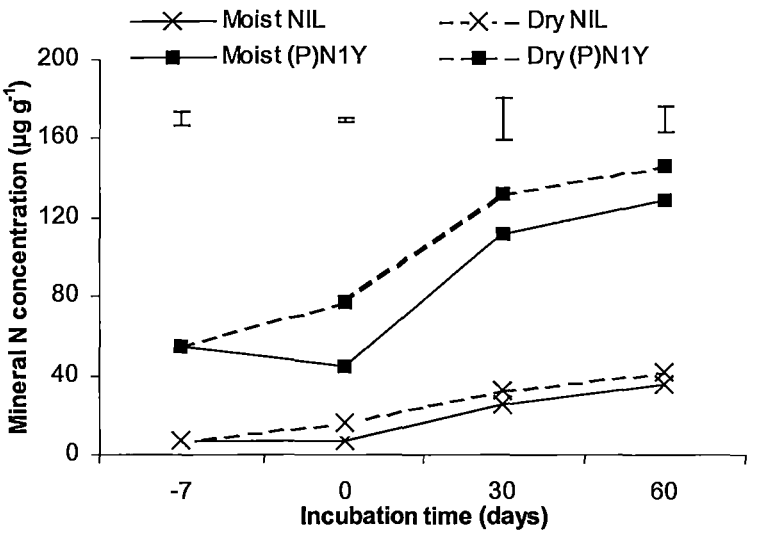


Figure 7.3 Mineral N concentrations (µg g^{-1}) measured in *P. radiata* litter at days –7 to 60. Bars indicate LSD (p = 0.05) for comparison of fertiliser-by-drying interaction means

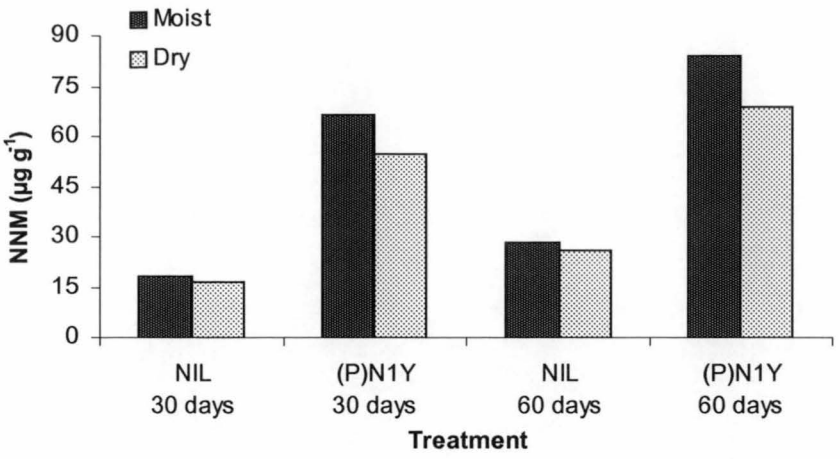


Figure 7.4 NNM (µg g^{-1}) in *P. radiata* litter after 30 and 60 day incubations. There was no significant interaction between fertiliser and moisture treatments.

Table 7.6 MANOVA summery table of treatment (fertiliser and air-drying) effects on *P. radiata* litter (O2) NNM, significant differences between means indicated by p values in bold.

	Day	Fertiliser treatment	Drying treatment	Interaction
Daily rate NNM	30	**<0.001**	0.322	0.445
	60	**<0.001**	0.053	0.145
Cumulative rate of NNM	0	**<0.001**	**<0.001**	**<0.001**
	30	**<0.001**	0.061	0.318
	60	**<0.001**	**0.017**	0.193

7.3.1.2 Ferrosol topsoil and litter under *E. regnans*

Air-drying the samples significantly increased the mineral N concentrations above topsoil maintained moist for the length of the incubation ($p<0.001$) (Figure 7.5). In the fertilised topsoil, mineral N concentrations increased due to air-drying by 5, 15 and 7 times those maintained moist at days 0, 30 and 60, respectively. The effect of pre-drying was less pronounced in the unfertilised topsoil, increasing 2, 4 and 3 times, respectively, with an interaction between fertiliser and drying significant ($p<0.05$) at day 0 and day 30 (Table 7.7).

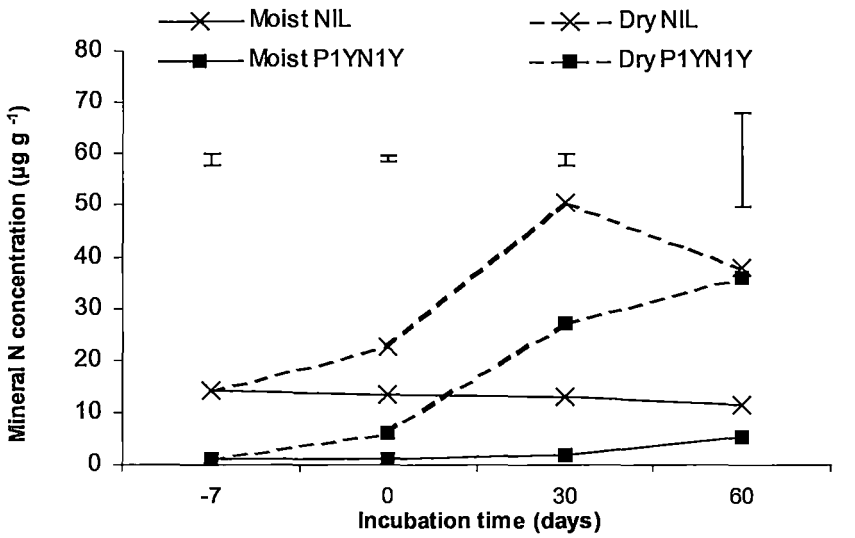


Figure 7.5 Mineral N concentrations (µg g^{-1}) measured in Ferrosol topsoil at days –7 to 60. Bars indicate LSD (p = 0.05) for comparison of fertiliser-by-drying interaction means

Rates of NNM at day 30 and 60 were significantly ($p<0.05$) different between moist and air-dried topsoils and there was a significant interaction between treatments (Figure 7.6). Rates of mineralisation in moist samples were low or negative and around 30 times less than those measured in air-dried topsoil. Differences between rates of NNM due to fertiliser treatment were evident only in air-dried topsoil.

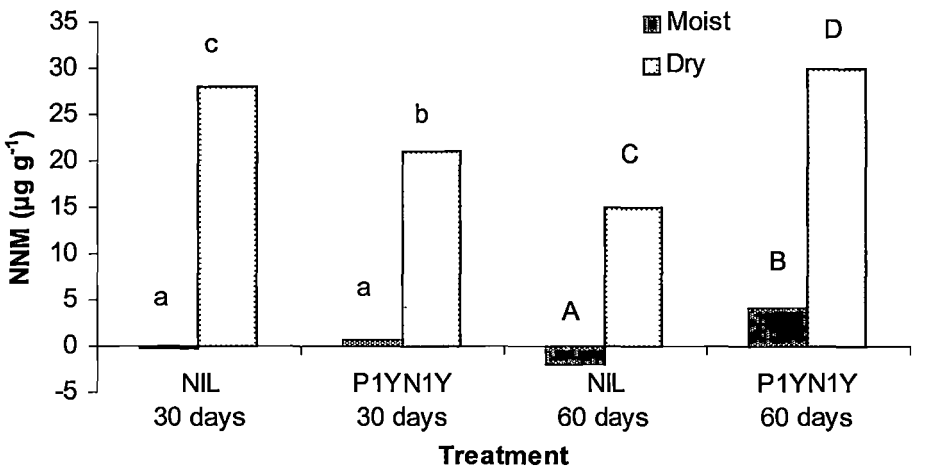


Figure 7.6 NNM (µg g^{-1}) in Ferrosol topsoil after 30 and 60 day incubations. Letters indicate significant differences between fertiliser and drying treatment. Lower case at 30 days, and upper case at 60 days.

Table 7.7 MANOVA summery table of treatments (fertiliser and air-drying) effects on Ferrosol topsoil NNM, significant differences between means indicated by p values in bold.

	Day	Fertiliser treatment	Drying treatment	Interaction
Daily rate NNM	30	**0.004**	**<0.001**	**<0.001**
	60	**<0.001**	**<0.001**	**0.010**
Cumulative rate of NNM	0	**<0.001**	**<0.001**	**0.002**
	30	**<0.001**	**<0.001**	**<0.001**
	60	0.385	**<0.001**	0.078

The effect of air-drying on mineral N availability in litter varied with the time of sampling. At day 0 air-drying significantly increased the amount mineral N (Figure 7.7). In contrast by day 30 air-drying significantly ($p<0.001$) decreased the amount of N in fertilised litter, and by day 60 there was no difference.

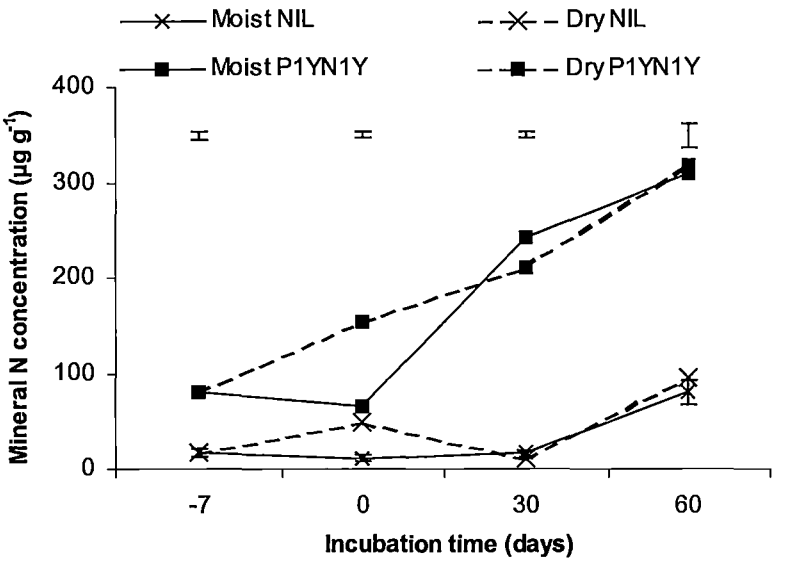


Figure 7.7 Mineral N concentrations (µg g^{-1}) measured in *E. regnans* litter at days –7 to 60. Bars indicate LSD (p = 0.05) for comparison of fertiliser-by-drying interaction means

As a result the difference between rates of NNM in fertilised and unfertilised litter was thirty-fold, when moist, and two-fold, when dried (Figure 7.8) ($p<0.05$). Rates of NNM at both 30 and 60 days where significantly ($p<0.001$) decreased in air-dried litter due to the large variation in the initial mineral N measured, rather than changing mineral availability during the incubation time (Table 7.8).

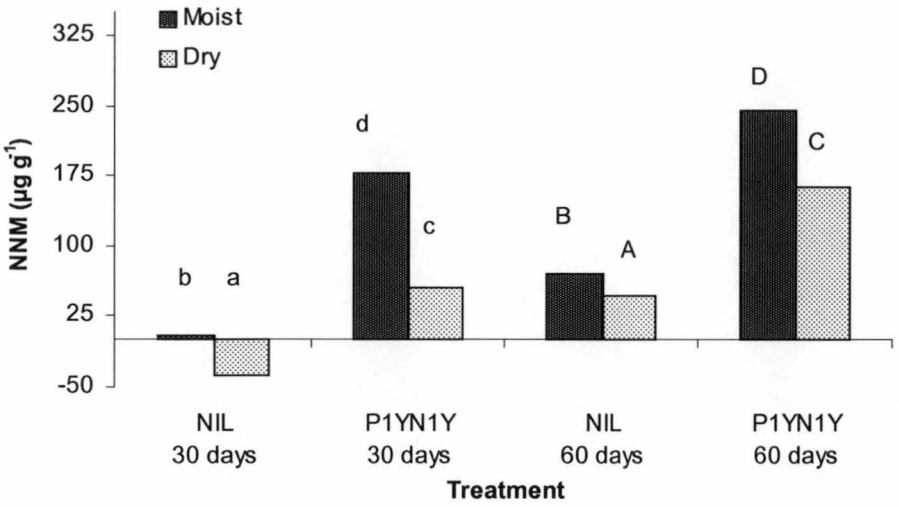


Figure 7.8 NNM (µg g^{-1}) in *E. regnans* litter after 30 and 60 day incubations. Letters indicate significant differences between fertiliser and drying treatment. Lower case at 30 days, and upper case at 60 days.

Table 7.8 MANOVA summery table of treatment (fertiliser and air-drying) effects on *E. regnans* litter (O2) NNM, significant differences between means indicated by p values in bold.

	Day	Fertiliser treatment	Drying treatment	Interaction
Daily rate NNM	30	**<0.001**	**<0.001**	**<0.001**
	60	**<0.001**	**<0.001**	**0.005**
Cumulative rate of NNM	0	**<0.001**	**<0.001**	**0.01**
	30	**<0.001**	**<0.001**	**0.01**
	60	**<0.001**	0.190	0.615

Low concentrations of nitrate were measured throughout the laboratory study, remaining below 1 µg g^{-1} in both litter and soil horizons. Air-drying had a varied effect on nitrate production generally increasing it in the soil (by up to 3-fold), but decreasing it in some litter samples (Table 7.9, $p<0.05$).

Table 7.9 Nitrate concentrations (μg g^{-1}) measured in air-dried and moist samples. Star (*) indicates LSD (p = 0.05) for comparison of fertiliser-by-drying interaction means.

	Horizon	Treatment	Day	Moist	Dry
Ferrosol	Soil	P1YN1Y	0	0	0.01
		NIL		0.01	0.01
		P1YN1Y	30	0.04	0.02
		NIL		0.01	0.05
		P1YN1Y	60	0.00	**0.03***
		NIL		0.02	**0.03**
E. regnans	Litter	P1YN1Y	0	**0.02***	0.07
		NIL		**0***	0.09
		P1YN1Y	30	0.07	0.13
		NIL		0.05	0.05
		P1YN1Y	60	0.11	0.15
		NIL		0.08	0.03
Kurosol	Soil	(P)N1Y	0	0	0
		NIL		0	0
		(P)N1Y	30	0.00	0.01
		NIL		0.01	0.03
		(P)N1Y	60	0.00	**0.01***
		NIL		0.00	**0.00**
P. radiata	Litter	(P)N1Y	0	**0.02***	0.07
		NIL		**0.01**	0.02
		(P)N1Y	30	0.13	0.08
		NIL		0.11	0.06
		(P)N1Y	60	0.12	0.12
		NIL		0.17	0.10

7.3.2. Experiment 2. Effects of laboratory incubation period on NNM in both litter and topsoils.

7.3.2.1 Kurosol topsoil and litter under *P. radiata*

A small non-significant effect of disturbance, in this case mixing, on NNM in samples was apparent in the fertilised Kurosol topsoil measured at day three (0.3 μg g^{-1} day^{-1}). Daily rates were calculated as $T_{3,\ 7,\ 15,\ 30\ or\ 60}$ minus initial N concentrations T_0 divided by the number of incubation days (3, 7, 15, 30 or 60). During the entire incubation period of sixty days, daily rates of NNM were extremely low; rates between day 7 and 60 rates were around 0.1 μg g^{-1} day^{-1} (Figure 7.9). Long-term annual fertilisation had no significant effect on daily rates of NNM. Results suggest that average rates of NNM in Kurosol topsoils would be less than 50 μg g^{-1} yr^{-1}.

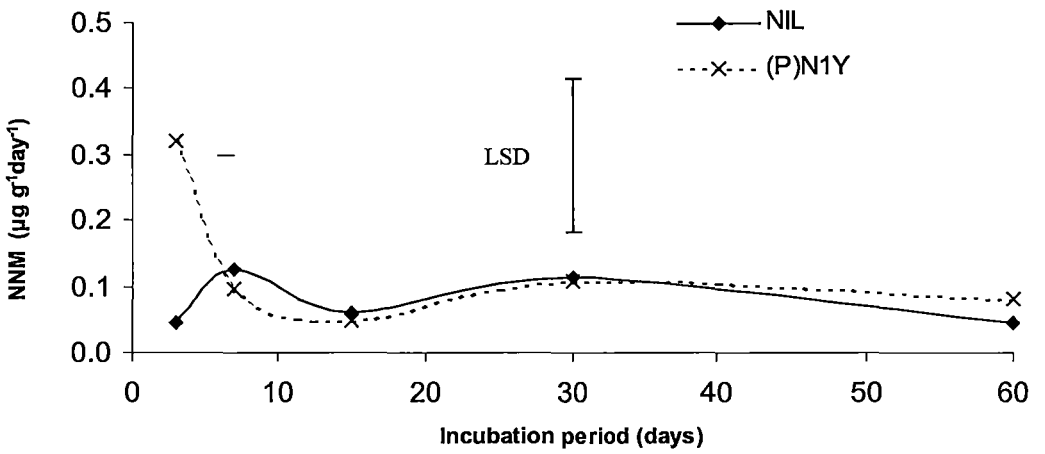


Figure 7.9 Rates of NNM (µg g^{-1} day^{-1}) in Kurosol topsoil. Bar indicates LSD for comparing interaction means of fertiliser and incubation period.

Cumulative rates of NNM during 60 days show a significant ($p<0.05$) increase in NNM in fertilised compared to unfertilised topsoil (Figure 7.10). However, significant differences occurred only by 60 days, with a distinct divergence in NNM rates occurring after day 30. Net N immobilisation occurred in unfertilised litter from day 30 to 60, while from day 15 onwards, cumulative NNM increased significantly with fertilised topsoil.

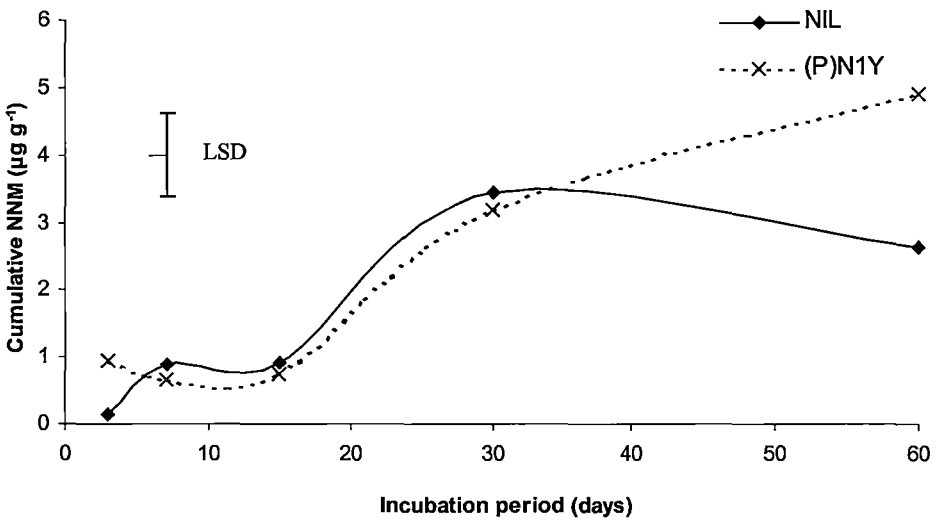


Figure 7.10 Cumulative rates of NNM (µg g^{-1}) in Kurosol topsoil. Bar indicates LSD for comparing interaction means of fertiliser and incubation period.

Daily NNM rates in *P. radiata* litter were generally ten times greater than that in Kurosol topsoil (Figure 7.11). Fertilised litter had significantly ($p<0.05$) higher rates of NNM than that unfertilised and showed a NNM flush at day 7, possibly in response to disturbance during sample preparation. Extrapolation of NNM rates between days 3 and 60 resulted in estimated annual NNM rates of 68 to 1282 μg g^{-1} yr^{-1} in fertilised litter, and 135 to 576 μg g^{-1} yr^{-1} in unfertilised litter. Average annual NNM from fertilised litter was significantly higher than the unfertilised litter at 702 and 287 μg g^{-1} yr^{-1}, respectively.

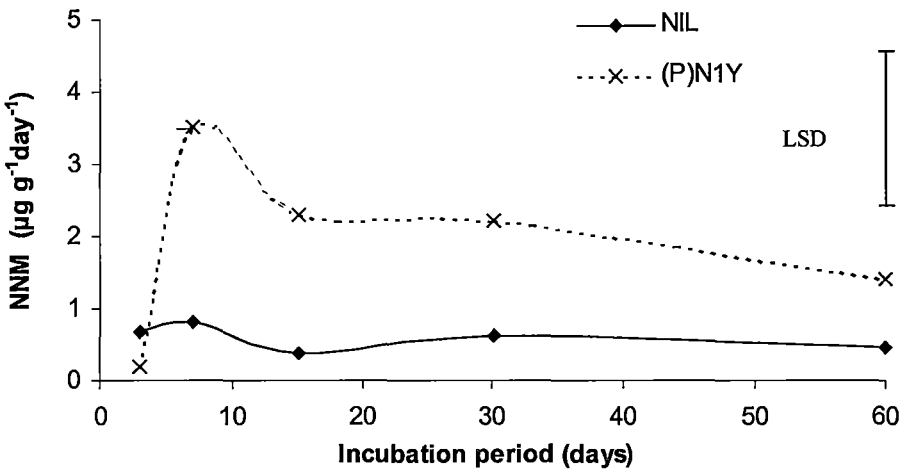


Figure 7.11 Rates of NNM (μg g^{-1} day^{-1}) in *P. radiata* litter. Bar indicates LSD for comparing interaction means of fertiliser and incubation period. Fertiliser affect was significant but there was no significant interaction.

In fertilised litter, cumulative rates of NNM increased significant ($p<0.001$) between day 3 and 7, and again between day 15 and 30 (Figure 7.12). Rates of NNM decreased slightly over time in unfertilised litter (Figure 7.11), which led to a significant cumulative effect by day 60.

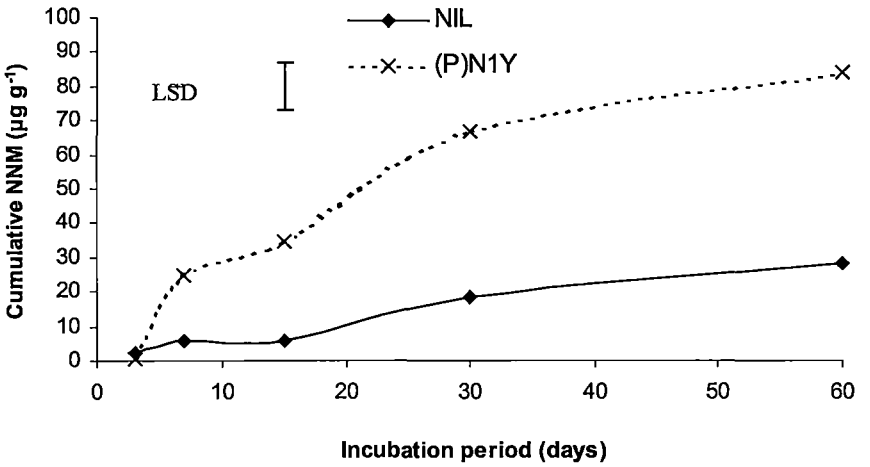


Figure 7.12 Cumulative rates of NNM (µg g^{-1}) in the *P. radiata* litter. Bar indicates LSD for comparing interaction means of fertiliser and incubation period.

Nitrate concentrations were measured throughout the 60-day incubation in both Kurosol topsoil and *P. radiata* litter. Data was not graphed, as the maximum amount of nitrate measured was only 0.16 µg g^{-1} in the NIL litter after 60 days. In both horizons daily rates of net nitrification were not greater than 0.01 µg g^{-1} day^{-1}.

7.3.2.2 Ferrosol topsoil and litter under *E. regnans*

Daily rates of NNM in Ferrosol topsoil were significantly ($p<0.001$) lower in fertilised topsoil at day 3 than unfertilised topsoil (Figure 7.13). From day 7 onwards, NNM in fertilised topsoil increased slightly, but was not significantly higher than those unfertilised. During the 60 day incubations, the average daily rates in fertiliser treatments were not significantly different, resulting in annual rates of NNM less than 14 µg g^{-1} in both fertilised and unfertilised topsoils.

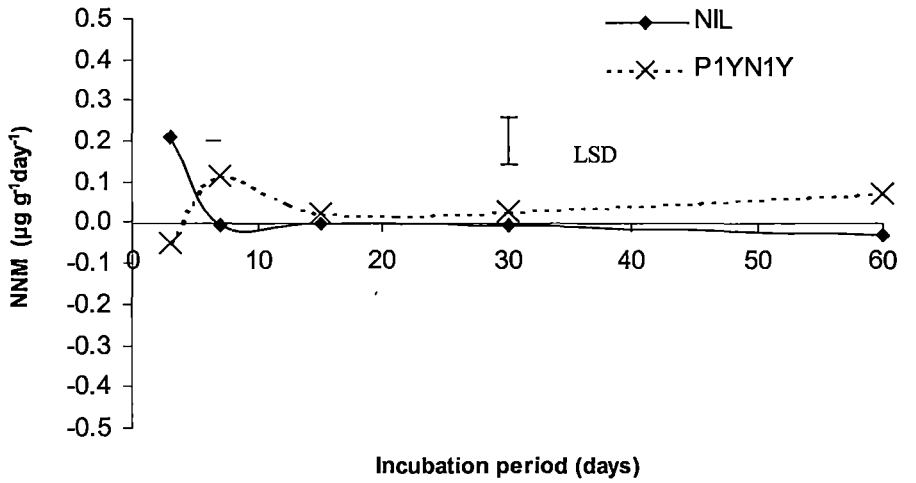


Figure 7.13 Rates of NNM (µg g^{-1} day^{-1}) in Ferrosol topsoil. Bar indicates LSD for comparing interaction means of fertiliser and incubation period.

Cumulative rates of NNM were significantly ($p<0.001$) higher in fertilised topsoil at day 60 only (Figure 7.14). Prior to this time, rates of NNM were similar between fertiliser treatments.

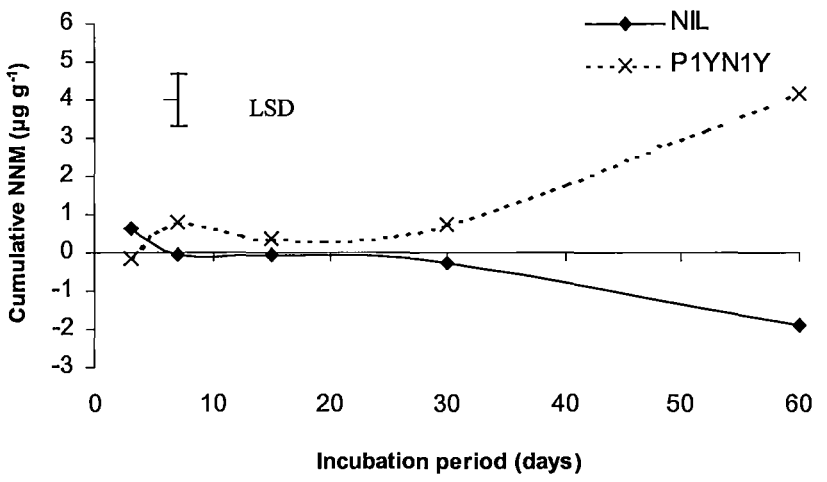


Figure 7.14. Cumulative rates of NNM (µg g^{-1}) in Ferrosol topsoil. Bar indicates LSD for comparing interaction means of fertiliser and incubation period.

Daily rates of NNM in fertilised *E. regnans* litter increased significantly ($p<0.001$) during the 60 day incubation (Figure 7.15). In contrast, unfertilised litters mineralised less between days 7 and 30, than at day 3. This resulted in fertilised litter having 3 to 30 times greater mineralisation rates then those unfertilised, dependent on the period

of incubation. Rates of NNM per annum, calculated from each date, varied in fertilised litters between 271 and 2820 μg g^{-1} yr^{-1}, and in unfertilised litters between 68 and 837 μg g^{-1} yr^{-1}. During the entire incubation period, fertilised litter had an NNM rate four times higher than that in unfertilised litter, i.e. 1810 and 317 μg g^{-1} yr^{-1}, respectively.

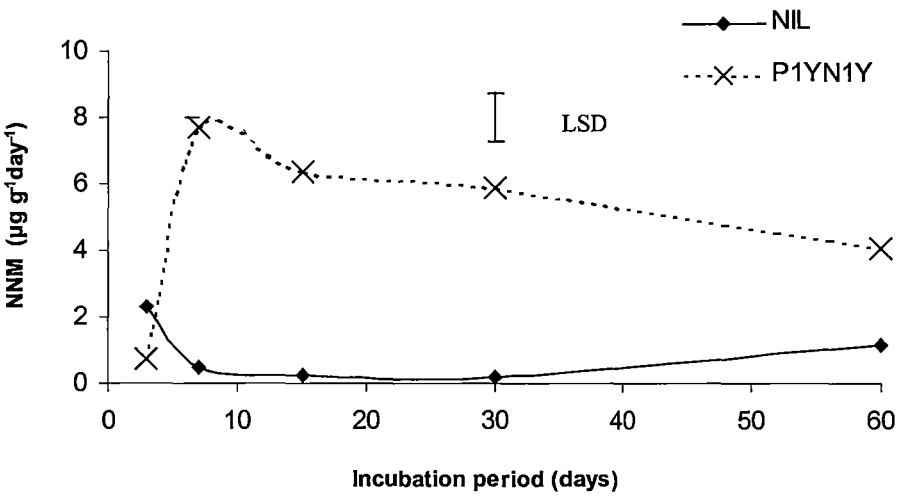


Figure 7.15. Rates of NNM (μg g^{-1} day^{-1}) in *E. regnans* litter. Bar indicates LSD for comparing interaction means of fertiliser and incubation period.

Cumulative rates of NNM in fertilised litters increased significantly ($p<0.001$) during all incubation periods (Figure 7.16). In contrast, cumulative rates of NNM in unfertilised litter remained around zero up to day 30, and then increased ten-fold from day 30 to 60. Comparisons between the rates of mineralisation in fertilised and unfertilised litter were similar between this period, both mineralising around 60 μg g^{-1} during this period.

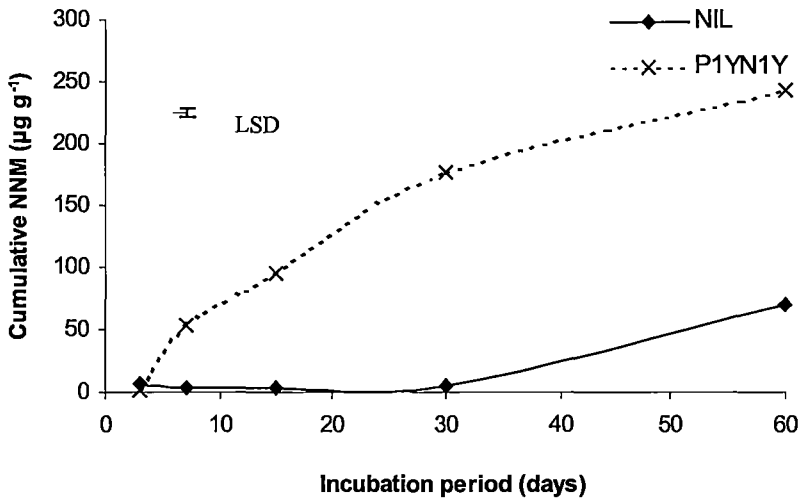


Figure 7.16 Cumulative rates of NNM (µg g^{-1}) in *E. regnans* litter. Bar indicates LSD for comparing interaction means of fertiliser and incubation period.

Nitrate concentrations were measured throughout the 60-day incubation in both Ferrosol topsoil and *E. regnans* litter. Data was not graphed, as the maximum amount of nitrate measured was only 0.09 µg g^{-1} in the P1YN1Y litter after 60 days. In both horizons daily rates of net nitrification were not greater than 0.01 µg g^{-1} day^{-1}.

7.3.3 Experiment 3. Testing the sensitivity of in vitro *NNM to various long-term N and P fertilisation.*

7.3.3.1 Kurosol topsoil and litter under *P. radiata*

Initial mineral N concentrations were extremely low, with the highest concentration measured in the (P)N1Y treatment (3 µg g^{-1}). Other treatments contained mineral N concentrations less than 1 µg g^{-1} (Figure 7.17).

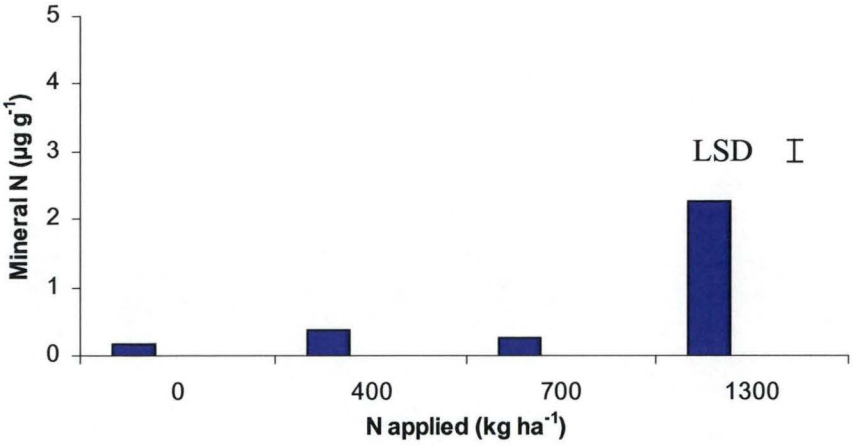


Figure 7.17 Initial mineral N concentrations (µg g^{-1}) in Kurosol topsoil. Bar indicates least significant differences (LSD) for comparing treatment means ($p<0.001$).

Rates of NNM at 20 °C increased significantly ($p<0.05$) between N input increases from 0 (NIL), to intermediate (700 kg N ha^{-1} (P)N2Y and 400 kg N ha^{-1} (P)N4Y) and 1300 ((P)N1Y) kg N ha^{-1} (Figure 7.18). The highest rate of NNM observed in (P)N1Y topsoil, was still low, resulting in less than 5 µg g^{-1} during the entire incubation period of 60 days. Reducing the incubation temperature to 10 °C significantly reduced NNM rates in all topsoils. Declines in NNM rates were proportionally greater in (P)N1Y topsoil, decreasing to almost one third of that measured at 20 °C. Subsequently, at 10 °C the highest rate of NNM occurred in (P)N4Y topsoil.

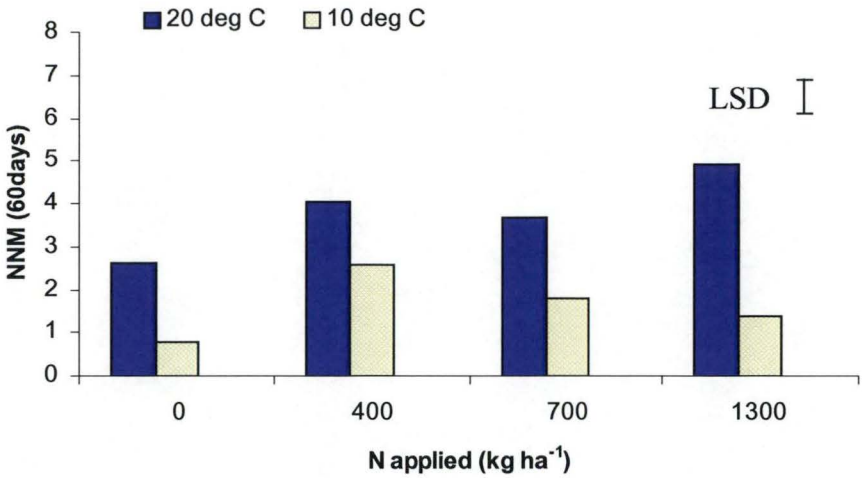


Figure 7.18 Rates of NNM in (μg g^{-1} 60 d^{-1}) Kurosol topsoil after 60 days incubations at 10 and 20 °C. Bar indicates LSD for comparing interaction means of fertiliser rate by incubation temperature ($p<0.05$).

These rates of NNM, projected as the proportion of total N mineralised annually, was less than 5 % per year when incubated at 20°C (Table 7.10) and lower when incubated at 10 °C .

Table 7.10 Projected percentage of total N mineralised annually (based on 60 d incubations) in topsoil and litter. Means with the same letters within a site and temperature combination are not significantly different ($p < 0.05$).

	Total N ug g^{-1}	20°C		10°C	
Kurosol topsoil					
NIL	553	2.9	a	0.9	a
(P)N4Y	584	4.2	a	2.7	b
(P)N2Y	783	2.8	a	1.4	a
(P)N1Y	964	3.1	a	0.9	a
***Pinus radiata* litter**					
NIL	8251	2.1	a	0.9	bc
(P)N4Y	9939	4.0	b	0.4	ab
(P)N2Y	9315	1.7	a	0.3	a
(P)N1Y	9933	5.1	b	1.4	c

Fertiliser significantly ($p<0.001$) increased initial mineral N concentrations in *P. radiata* litter. The highest concentration was observed in the (P)N1Y treatment, which was six times that measured in the unfertilised (Figure 7.19). In contrast to the rate of

fertilisation the initial concentrations were higher in the bulked sample of the (P)N4Y (400 kg N ha^{-1}) than (P)N2Y (700 kg N ha^{-1})

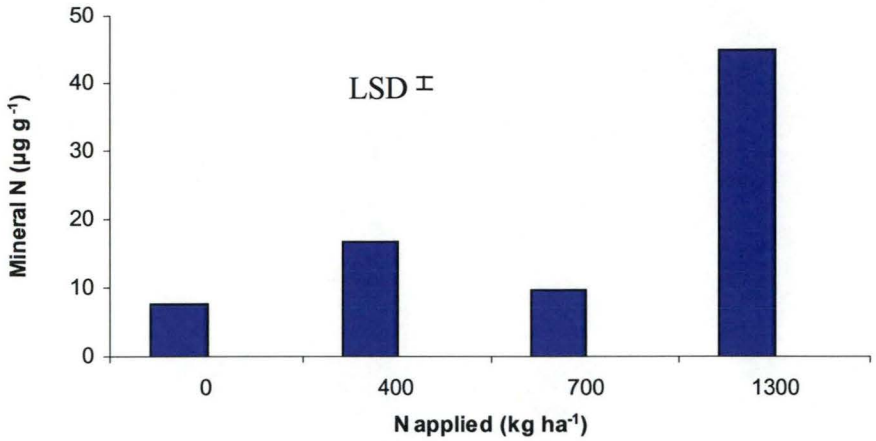


Figure 7.19 Initial mineral N concentrations (µg g^{-1}) in *P. radiata* litter. Bar indicates LSD for treatments ($p<0.001$).

Trends in NNM rates between treatments incubated at 20 °C reflected initial N concentrations, (P)N4Y mineralising double that of (P)N2Y, while (P)N1Y was significantly ($p<0.001$) higher than all other treatments (Figure 7.20). Reducing the temperature to 10 °C significantly reduced NNM in all litters, and decreased the variation between (P)N1Y and unfertilised litters by five-fold. At 10 °C, NNM was similar between treatments in all but the highest fertiliser treatment.

These rates of NNM in the litter, projected as the proportion of total N mineralised annually, was less than 5 % per year when incubated at 20°C (Table 7.10) and lower when incubated at 10 °C. At 10 °C the proportion mineralised decreased in treatments between 50 and 90 percent.

Initial N concentrations and NNM rates at both 10 °C and 20 °C in soils and litter were fitted with linear and quadratic regression. No significant relationships were observed.

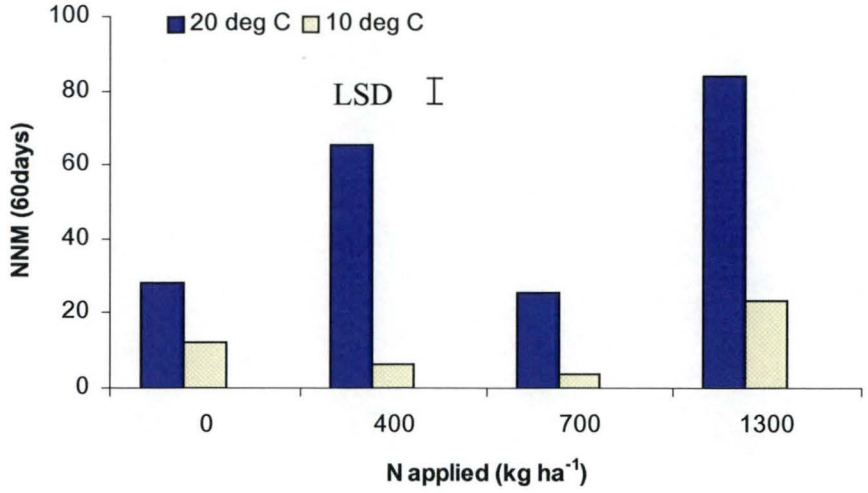


Figure 7.20 Rates of NNM in (μg g^{-1} 60 d^{-1}) in *P. radiata* litter at 10 and 20 °C. Bar indicates LSD for comparing interaction means of fertiliser rate by incubation temperature ($p<0.001$).

7.3.3.2 Ferrosol topsoil and litter under *E. regnans*

Intermediate fertilisation amounts, 400 kg N ha^{-1} (P4YN4Y) and 700 kg N ha^{-1} (P2YN2Y) had similar initial N content to unfertilised (NIL) topsoil, and ten times the amount measured in the 1300 kg N ha^{-1} (P1YN1Y) treatment (Figure 7.21). With the exception of P1YN1Y, mineral N concentrations were initially higher when N and P were applied in combination, than when either N or P was applied alone (N2Y and P2Y, respectively).

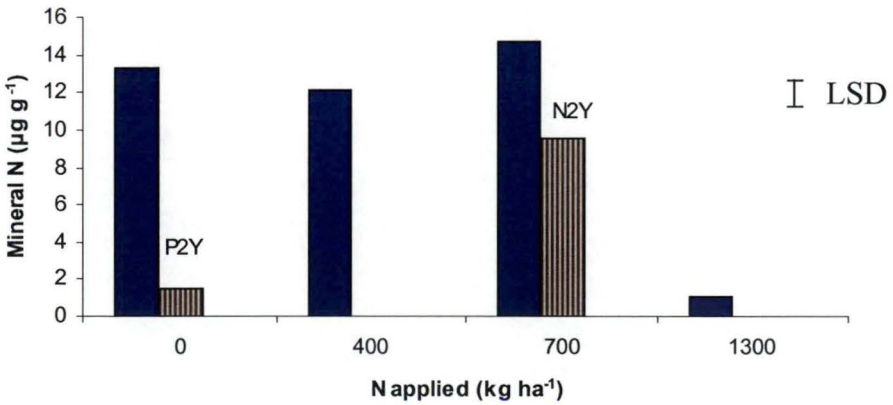
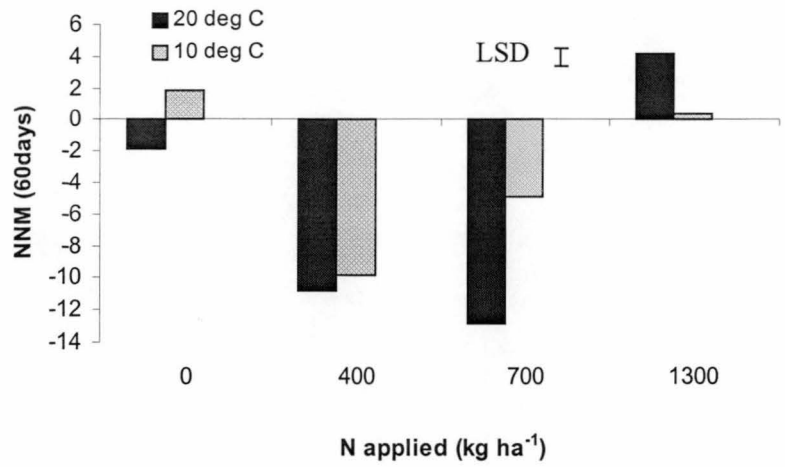


Figure 7.21 Initial mineral N concentrations (μg g^{-1}) in Ferrosol topsoil. Bar indicates LSD for treatments ($p<0.001$).

a.



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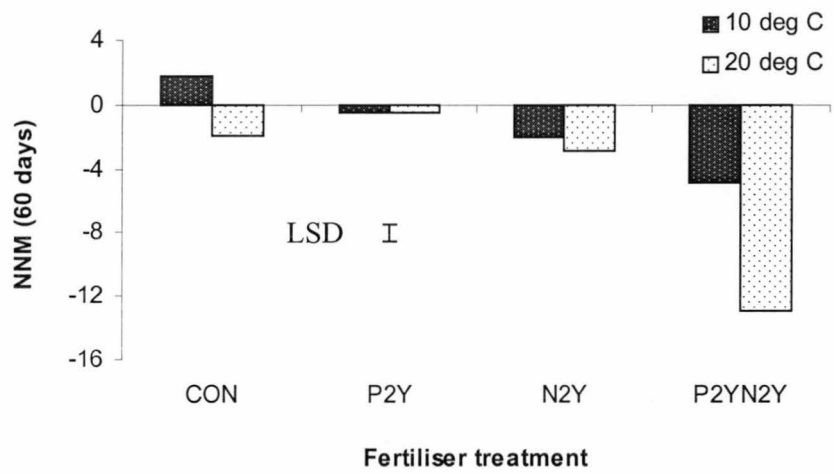


Figure 7.22 Rates of NNM in Ferrosol topsoil (μg g^{-1} 60 d^{-1}) at 10 and 20 °C (a) N level trial, (b) N by P factorial trial. Bar indicates LSD for comparing interaction means of fertiliser rate by incubation temperature ($p<0.001$).

In contrast, positive NNM (60 days at 20 °C) occurred only in P1YN1Y topsoil, which was significantly ($p<0.001$) higher than all other treatments (Figure 7.22a). Intermediate levels of N and P applications in combination or alone resulted in net immobilisation. Application of both N and P alone had significantly lower rates of N immobilisation than when the fertilisers were applied in combination (Figure 7.22b). Reducing the incubation temperature to 10 °C resulted in marginal increases in NNM from unfertilised and P4YN4Y treatments and a significant increase in P2YN2Y. In contrast, NNM rates in P1YN1Y topsoil declined when the incubation temperature was reduced. As a result, the differences between rates of NNM topsoil in the highest

application of N (P1YN1Y) and the NIL were reversed at the different incubation temperatures.

The proportion of N mineralised in Ferrosol topsoil was effected by incubation temperature. At 20 °C, P1YN1Y topsoil mineralisation was the highest at just under 2 %, while at 10 °C, unfertilised topsoil mineralised the highest at just less than 1 % (Table 7.11).

Table 7.11 Projected percentage of total N mineralised annually (based on 60 d incubations) in topsoil and litter. Means with the same letters within a site and temperature combination are not significantly different ($p < 0.05$).

	Total N ug g^{-1}	20°C		10°C	
Ferrosol topsoil					
NIL	1806	-0.6	b	0.6	e
P2YN2Y	1563	-4.2	a	-3.8	a
P4YN4Y	1618	-4.8	a	-1.8	b
P1YN1Y	1344	1.9	c	0.2	d
N2Y	1990	-0.9	b	-0.6	c
P2Y	1620	-0.2	b	-0.2	d
***Eucalyptus regnans* litter**					
NIL	8836	4.8	a	0.15	ab
P2YN2Y	9499	7.8	b	-0.03	a
P4YN4Y	9060	7.1	b	-0.19	a
P1YN1Y	12159	12.2	d	8.68	d
N2Y	10068	9.0	c	2.33	c
P2Y	10009	11.4	d	0.55	b

Fertiliser treatment significantly ($p<0.001$) effected initial mineral N concentration in *E. regnans* litter (Figure 7.23). Increases in mineral N concentrations were unrelated to the rate of N application, that is double the rate of N application P2YN2Y compared to and P1YN1Y, increased mineral N concentrations three-fold, whilst almost doubling the application of N between P4YN4Y and P2YN2Y, resulted in a less than 50 % increase. Application of N alone significantly increased initial N above that of N and P fertiliser combined.

Initial mineral N concentration in the P1YN1Y litter was significantly higher than other fertiliser treatments. In contrast, in the topsoil initial mineral N concentration was much lower, resulting in a sixty-fold difference between the horizons. N2Y litter also had initial mineral N concentrations were four times greater than the topsoil.

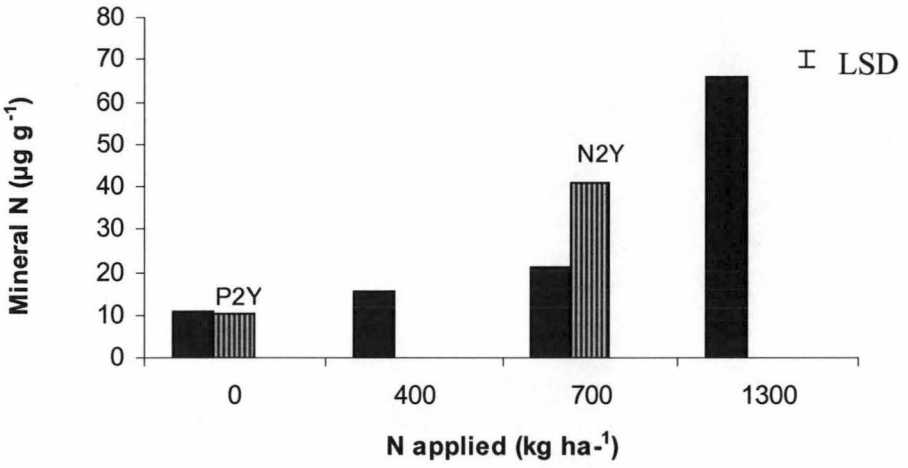
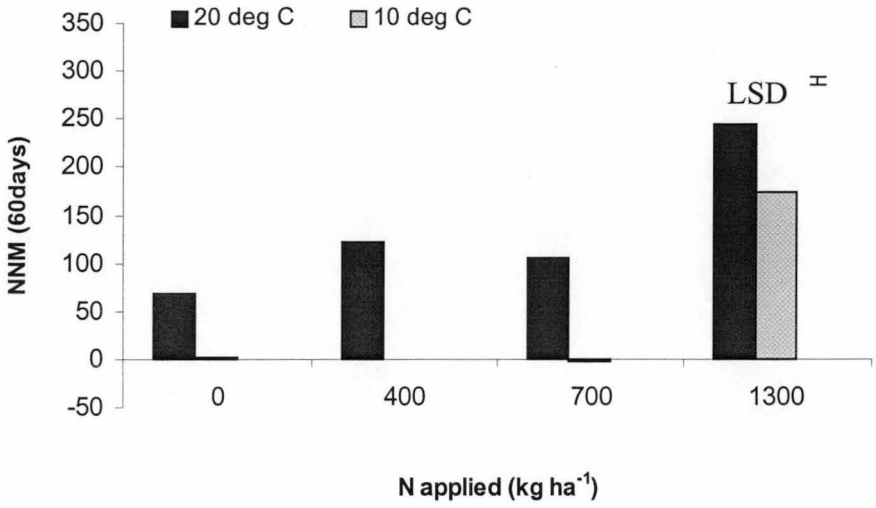


Figure 7.23 Initial N concentrations (µg g^{-1}) in *E. regnans* litter. Bar indicates LSD for treatments ($p<0.001$).

Rates of NNM in litter at 60 days and 20 °C increased significantly with fertilisation (Figure 7.24a). NNM rates were three-fold higher in P1YN1Y than unfertilised litter and doubled that measured in P2YN2Y and P4YN4Y. In contrast to initial N values, NNM rates were higher in P4YN4Y than P2YN2Y treatments. Application of P and N alone also significantly increased NNM. Phosphorus application actually increased rates of NNM more than when applied in combination with N, or when N was applied alone (Figure 7.24b).

Decreasing the incubation temperature to 10 °C significantly reduced NNM in all litters, by up to 178 µg g^{-1}. At 10 °C, NNM was positive only in P1YN1Y and N2Y litters, and there was no effect of P on NNM. Differences between the intermediate rates of N fertilisation declined at 10 °C, and were generally around zero.

a.



b.

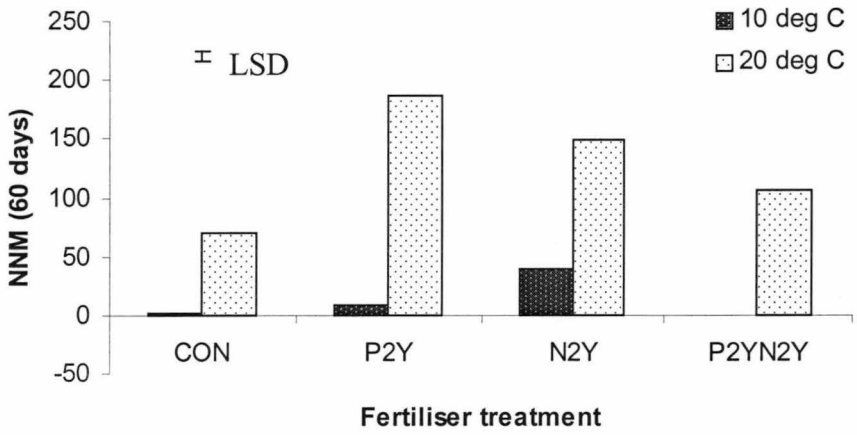


Figure 7.24 Rates of NNM in *E. regnans* litter (μg g^{-1} 60 d^{-1}) at 10 and 20 °C (a) N level trial, (b) N by P factorial trial. Bar indicates LSD for comparing interaction means of fertiliser rate by incubation temperature ($p < 0.001$).

In comparison to the topsoils, at 20 °C, *E. regnans* litter the proportion of N mineralised was between twelve and five percent of the total N available (Table 7.11). At both temperatures the highest efficiency of conversion occurred in P1YN1Y, which declined by one quarter when the temperature was reduced. However, in comparison to other treatments the proportion of total N mineralised in the P1YN1Y litter was the least effected by temperature.

Initial N concentrations and NNM rates at both 10 °C and 20 °C in soils and litter were fitted with linear and quadratic regression. No significant relationships were observed.

7.3.4 Correlations between initial mineral N and NNM to nutrient content

Initial mineral N and NNM rates of litter and topsoil are examined by scatter plot and correlation analysis for relationships with nutrient concentrations measured in Chapter 3 (Table 3.8). Each site is examined separately first for correlations between nutrients and NNM. Although this provides insight into the relationship between the nutrient content and NNM, the strength of this analysis is limited due to a single measurement of nutrient content for each three replicates of NNM. Therefore, a combined analysis that uses each site, soil and litter nutrient content, as the replicate is given in Section 7.3.4.3.

7.3.4.1 Kurosol topsoil and litter under *P. radiata*

Although initial mineral N concentrations measured in the Kurosol topsoil were significantly linearly related to N (positive, +ve), Ca (negative, -ve) and Mg (-ve) concentrations and pH (-ve), these relationships were strongly dependent on the highest fertilised treatment (P)N1Y (Table 7.12). Such that when the data from the ((P)N1Y) treatment was removed, the relationship was no longer significant. The same trend was observed in the *P. radiata* litter. In the litter, Ca and Mg concentrations and pH where significantly negatively (linearly) related. However, even for Mg ($r^2 = 0.94$), the relationship was dependent on the highest fertiliser treatment ((P)N1Y) (Figure 7.25). When the data from this treatment were removed, the relationship was not significant.

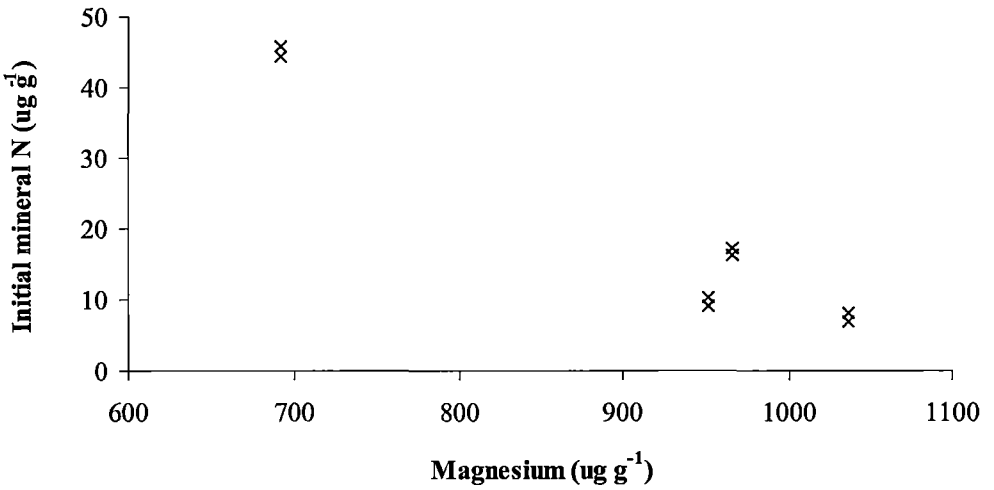


Figure 7.25 Initial N compared to magnesium concentrations (μg g^{-1}) in Kurosol topsoil

NNM rates in Kurosol topsoil incubated at 10 °C (Table 7.12) were linearly related to P (+ve) concentrations only, and at 20 °C, rates were linearly related to total N (+ve), P (+ve) and Mg (-ve) concentrations and soil pH (-ve) at 1:5 soil: water (Figure 7.26). Unlike initial mineral N concentrations, these relationships were not dominated by the data from the highest rate of N fertilisation ((P)N1Y). Although not significant, the N concentration was almost double at 0.1 % in the annually fertilised than the unfertilised and (P)N4Y topsoil (0.06 %). NNM rates were unrelated to organic C concentration and C: N ratios. Organic carbon in the unfertilised and annually fertilised topsoil was around 1.3 and 2.5, respectively. However, organic C concentrations were highly variable in the (P)N1Y topsoil and ranged from 1.5 to 2.6 % resulting in no significant differences between treatments.

Pinus radiata litter incubated at 10 °C showed a strong negative (linear) relationship with Ca concentration, however, this was not significant at 20 °C. Magnesium was linearly related (-ve) at both temperatures, while N (+ve) concentration and pH (-ve) were linearly related to NNM only at 20 °C.

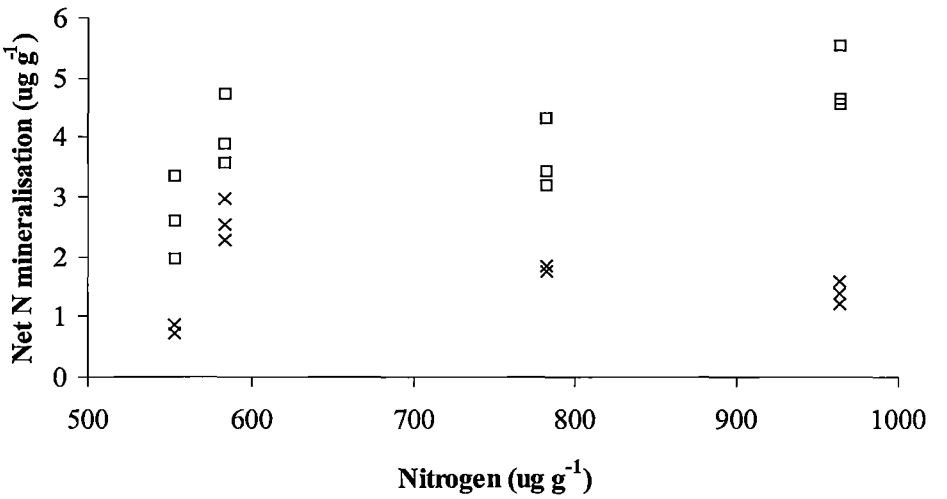


Figure 7.26 Kurosol topsoil Net N mineralisation compared to total nitrogen concentrations (μg g^{-1}), crosses 10°C and squares 20°C

Table 7.12 Correlations (%) between initial N and NNM (10°C and 20°C) and major substrate nutrients (r squared statistic). Significant correlations indicated with * ($p < 0.05$) and **($p < 0.001$).

Nutrient	initial N		NNM at 10°C		NNM at 20°C	
	P. radiata	Kurosol	*P. radiata*	Kurosol	*P. radiata*	Kurosol
N	32	66*	<0	<0	56*	38*
P	13	<0	<0	75**	18	30*
Mg	94**	51*	51*	<0	58*	38*
Ca	50*	75*	85**	<0	24	17
pH	54*	43*	5	11	35*	64*
S	<0	14				

7.3.4.2 Ferrosol topsoil and litter under *E. regnans*

Relationships between initial N concentrations and concentrations of nutrients in Ferrosol topsoil depended on the number of fertiliser treatments included in the regression analysis. When all treatments were included in scatter graphs and regression analysis, Ca (-ve), S (+ve), and Mg (-ve) concentrations and pH (+ve) of the Ferrosol topsoil were linearly related to initial mineral N (Table 7.13a). However, like those measured in the Kurosol topsoil the relationship depends on the highest fertiliser treatment P1YN1Y, such that when the data from this treatment was removed, the relationship was not significant. In *E. regnans* litter S (+ve) concentration and pH (-ve), was significantly linearly related to mineral N concentrations (pH $r^2 = 0.85$) when all treatments were included.

Examination of the N level treatments in *E. regnans* litter showed initial mineral N concentration in the litter to be linearly related to N, Mg, Ca, P and S concentrations and pH (Table 7.13b). Magnesium and mineral N concentrations were strongly positively correlated ($r^2 = 0.93$ and 0.95) within these treatments. Other relationships between nutrients and mineral N in the litter were again dependent on data from the P1YN1Y treatment. In the four N level treatment Ferrosol topsoils N, Mg, concentrations and pH were significantly related to mineral N concentrations, but the relationships were weak compared to the litters.

Table 7.13 Correlations (%) between initial N and NNM (10°C and 20°C) and major substrate nutrients (r squared statistic), (a) includes all six treatments data (b) includes N level data only. Significant correlations indicated with * ($p < 0.05$) and **($p < 0.001$).

(a)

Nutrient	initial N		NNM at 10°C		NNM at 20°C	
	E. regnans	Ferrosol	*E. regnans*	Ferrosol	*E. regnans*	Ferrosol
N	<0	20	9	<0	<0	7
P	<0	20	<0	<0	13	<0
Mg	<0	52*	<0	53**	1	74**
Ca	9	28*	22*	23*	<0	15
pH	85**	28*	93**	<0	49**	9
S	31*	48*				

(b)

Nutrient	initial N		NNM at 10°C		NNM at 20°C	
	E. regnans	Ferrosol	*E. regnans*	Ferrosol	*E. regnans*	Ferrosol
N	95**	66*	96**	<0	97**	5
P	52*	<0	37*	<0	68**	<0
Mg	91**	53*	80**	65**	95**	98**
Ca	93**	21	87**	76**	99**	51*
pH	88**	51*	94**	<0	72**	7

Like the initial mineral N, linear correlations between NNM and nutrients analysed in litter and topsoil were dependent on the number of fertiliser treatment included in the regression. When N2Y and P2Y treatment data were included in regression analysis of *E. regnans* litter, pH (-ve) at both temperatures, and Ca (+ve) at 10 °C showed a significant linear relationship with NNM (Table 7.13a). At 10 °C, the relationship between pH and NNM had an $r^2 = 0.93$, but was significant only when P1YN1Y treatment data was included.

Removal of N2Y and P2Y data from regressions resulted in significant linear positive relationships between NNM in Ferrosol topsoils for only Mg and Ca ($r^2 = 0.98$ at 20 °C) concentrations, at both temperatures. In the litter total N (+ve), P (+ve), Ca (+ve) and Mg (-ve) concentrations and pH (-ve) were linearly related to NNM at both temperatures. Both Ca and Mg showed strong linear relationships with NNM, at $r^2 = 0.99$ and $r^2 = 0.96$, respectively (Figure 7.27).

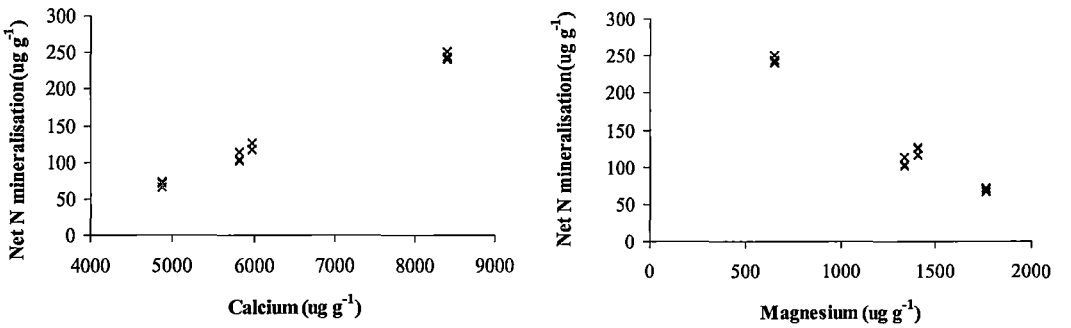


Figure 7.27 Net N mineralisation compared to (a) calcium and (b) magnesium concentrations (µg g^{-1}) in *E. regnans* litter incubated at 20°C

7.3.4.3 Combined sites

Combined topsoil data (using Kurosol and Ferrosol topsoils as replicates) indicated that only the initial mineral N concentration was correlated to NNM at 20 °C ($r^2 = 75.5$ $p = 0.003$) (Table 7.14). No other nutrient content was correlated for NNM across both soil types. Combined litter data (using *P. radiata* and *E. regnans* as replicates) NNM was correlated with mineral initial N, total N, P and Ca at 20 °C and initial N and total N at 10 °C (Figure 7.28).

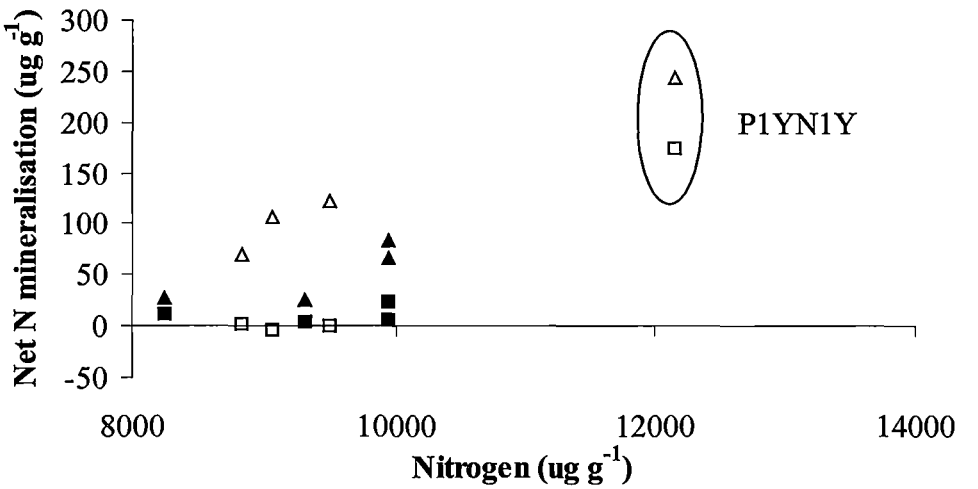


Figure 7.28 *Pinus radiata* (solid)and *E. regnans* (open) litter Net N mineralisation compared to nitrogen concentrations (μg g^{-1}), triangles 10 °C and squares 20 °C.

Table 7.14 Correlations between NNM (10°C and 20°C) and major substrate nutrients (r squared statistic), includes combined data from N level experiments at both sites. Significant correlations indicated with * ($p < 0.05$) and **($p < 0.001$).

Nutrient	NNM at 10°C		NNM at 20°C	
	Litter	Topsoil	Litter	Topsoil
N	74.4*	15.8	69.4*	38.7
P	20.7	23.3	72.0*	38.8
Mg	35.1	<0	65.5*	1.1
Ca	19.8	<0	<0	<0
pH	<0	15.3	<0	39.5
S	19.3	6.0	15.6	11.2
initial N	70.0*	31.7	64.7*	75.5*

Predicted annual rates of NNM using 60 day incubations in NIL and (P)N1Y Kurosol topsoils were 8 kg N ha^{-1} yr^{-1} and 14 kg N ha^{-1} yr^{-1}, respectively at 10 °C, and at 20 °C this increased to 26 kg N ha^{-1} yr^{-1} and 49 kg N ha^{-1} yr^{-1}, respectively (Table 7.15). Although temperature significantly increased NNM in both treated topsoils, the difference between rates of NNM in fertilised and unfertilised topsoils was always around double. Although temperature significantly increased NNM in both treated topsoils, the difference between rates of NNM in fertilised and unfertilised topsoils was always around double. In Ferrosol topsoils predicted annual rates of NNM using

60 day incubations in NIL and P1YN1Y were 14 kg N ha^{-1} yr^{-1} and 3 kg N ha^{-1} yr^{-1}, respectively at 10 °C, and at 20 °C NIL treatment declined to -14 kg N ha^{-1} yr^{-1} while P1YN1Y treatment increased to 32 kg N ha^{-1} yr^{-1} (Table 7.15).

Table 7.15 Summary of results from Experiments 1, 2 and 3. Annual NNM ha^{-1} yr^{-1} in each horizon for each laboratory treatment and fertiliser treatment.

Horizon	Fertiliser treatment	Laboratory treatment		Time				
		Moisture	Temperature	3	7	15	30	60
Ferrosol (0-10 cm)	NIL	Dry	20 °C				428	115
		Moist	20 °C	96	-4	-2	-4	-14
		Moist	10 °C					14
	P1YN1Y	Dry	20 °C				323	230
		Moist	20 °C	-23	52	11	12	32
		Moist	10 °C					3
	P2YN2Y	Moist	20 °C					-99
		Moist	10 °C					-37
	P4YN4Y	Moist	20 °C					-83
		Moist	10 °C					-76
	N2Y	Moist	20 °C					-22
		Moist	10 °C					-16
	P2Y	Moist	20 °C					-4
		Moist	10 °C					-4
E. regnans (O2) litter	NIL	Dry	20 °C				-6	4
		Moist	20 °C	11	2	1	1	6
		Moist	10 °C					0
	P1YN1Y	Dry	20 °C				53	77
		Moist	20 °C	21	219	180	167	115
		Moist	10 °C					82
	P2YN2Y	Moist	20 °C					13
		Moist	10 °C					0
	P4YN4Y	Moist	20 °C					7
		Moist	10 °C					0
	N2Y	Moist	20 °C					9
		Moist	10 °C					0
	P2Y	Moist	20 °C					22
		Moist	10 °C					6
Kurosol (0-10cm)	NIL	Dry	20 °C				144	75
		Moist	20 °C	27	75	36	68	26
		Moist	10 °C					8
	(P)N1Y	Dry	20 °C				114	114
		Moist	20 °C	190	56	29	63	49
		Moist	10 °C					14
	(P)N2Y	Moist	20 °C					36
		Moist	10 °C					39
	(P)N4Y	Moist	20 °C					40
		Moist	10 °C					61
P. radiata (O2) litter	NIL	Dry	20 °C				8	6
		Moist	20 °C	10	12	5	9	7
		Moist	10 °C					3
	(P)N1Y	Dry	20 °C				47	30
		Moist	20 °C	5	92	60	58	36
		Moist	10 °C					10
	(P)N2Y	Moist	20 °C					8
		Moist	10 °C					1
	(P)N4Y	Moist	20 °C					10
		Moist	10 °C					0

7.4 Discussion

7.4.1 Kurosol topsoil and litter formed under *P. radiata*

Net N mineralisation was almost double in the (P)N1Y Kurosol topsoil than the NIL at 49 kg N ha^{-1} yr^{-1} and 26 kg N ha^{-1} yr^{-1}, respectively. However, significant differences were observed between treatments only when incubated for a period of 60 days. The low rates of NNM in Kurosol topsoil observed in this study at both temperatures concur with previous field and laboratory observations (13 kg N ha^{-1} yr^{-1} for NIL and 52 kg N $ha^{-1}yr^{-1}$ for (P)N1Y), Chapter 4). In general, soil micro-organisms grow at N starvation levels with four percent N in their biomass (Rosswall, 1982). The efficiency of N conversion can be examined by determining the amount of total N that was converted to mineral N in excess of microbial requirements, i.e. net N mineralisation divided by total N. In this study, laboratory incubations of both litter and topsoil at 20 °C, resulted in less than five percent of the total N being mineralised annually. This was a similar amount to that observed at the lower end of N mineralisation in agricultural soils (Tabatabai and Al-Khafaji, 1980). Hence, even after receiving large applications of N and P and incubating soils at the higher end of field temperatures (20 °C), N mineralisation rates were low.

Reducing the incubation temperature from 20 °C to 10 °C significantly reduced NNM in all Kurosol topsoils, and the efficiency of N conversion declined to less than 1 % in both annually fertilised and unfertilised topsoils. Many researches have reported different temperature sensitivities when comparing temperature responses in various systems, such as soil vs litter vs horizon types (Kirschbaum, 1995). In this study the effects of temperature were more pronounced in topsoils where high rates of fertiliser had been applied i.e. Kurosol topsoil (P)N1Y incubated at 10 °C did not have significantly higher NNM rates than the unfertilised topsoil. Both Powers (1990) and MacDonald *et al.* (1995) observed that increasing temperatures often had an equal or greater influence than site on NNM rates. This study indicated that at a given site the amount of substrate N could influence the effect of temperature on NNM. However, such variations in response to temperature were only found when there were large changes in N inputs. At 20°C NNM rates in (P)N4Y and (P)2Y treatments were similar to at 40 kg N ha^{-1} $yr^{-1}$and 36 kg N ha^{-1} yr^{-1}, respectively.

A pronounced temperature effect on NNM was also observed in litter horizons in this study. Increasing the incubation temperatures of litter horizons, to 20 °C, generally increased the variation in NNM between different fertiliser treatments. However, this was not linearly related to N application, as 400 kg N ha^{-1} ((P)N4Y) treatment, often had significantly higher rates of NNM than the 700 kg N ha^{-1} ((P)N2Y) treatment. It was interesting to note that the total N concentration in the (P)N4Y treatment was similar to the (P)N1Y treatment in the litters from this site (Table 7.10). However, NNM was significantly higher only at the higher incubation temperature in this treatment ((P)N4Y). This suggested that at the higher temperature some other factor became important in microbial growth, which remained limited at lower temperatures. In soils with adequate moisture availability, at higher temperatures increased metabolic rate and access to substrates in microbial communities have been observed (Ellert and Bettany, 1992; MacDonald *et al.*, 1995; Beier and Eckerersten, 1998). An increasing rate of litter decomposition in (P)N4Y treatment due to the increase in temperature, compared to the other layers, would explain such a flux in NNM. However, it is generally accepted that organic matter decomposition is limited not only by N, but also by soluble C (McLaughlin *et al.*, 2000). The increase in soluble C availability due to disturbance and temperature changes in samples such as those taken from the (P)N4Y treatment, may have increased litter decomposition and therefore rates of NNM. In both horizons (O2 and A1) the response of N mineralising microbes to temperature was stronger in annually fertilised ((P)N1Y) than the NIL treatment.

Incubation length had a significant effect on the overall rate of NNM, response depended on fertiliser treatment and horizon being studied. In short incubation periods, effects of disturbance (mixing) were particularly evident in fertilised *P. radiata* litter, with a depression in daily rates at day 3 compared to day 7, and settling to an intermediate rate by day 15. In contrast, fertilised Kurosol topsoil showed a reversed trend, with daily NNM rates enhanced at day 3, and then rates of NNM remaining constant for the duration of the incubation.

Patterns of N mineralisation under field conditions are influenced by the organisation of the physical, chemical, and biological components of the soil matrix on a micro

scale (Strong *et al.*, 1998a). Disruption of these aggregates due to mixing soils increases N mineralisation, the amount of increase depends on the degree of physical protection, and consequently the size and stability of the aggregates prior to disturbance (Sollins *et al.*, 1984). The lack of aggregation in the Kurosol topsoil could explain limited changes in NNM due to disturbance of this soil. The percent of water stable aggregates (>0.25 mm) in the Kurosol topsoil were low ranging from 10 to 23 % (Chapter 3 Table 3.11). In addition to release of organic matter from microsites, Sierra (1992) noted disturbance could produce a more homogenous soil aeration, by rupturing anaerobic soil microsites, and increasing sites accessibility for microbial colonisation. In poorly aggregated soil such as the Kurosol topsoil, limited increases in microsite nutrient or oxygen availability would occur due to physical disruption. This difference in the 'physical protection of organic matter' may explain why the Kurosol topsoil reacted differently, by releasing a peak of mineral N at day 3 due to the initial disturbance, causing limited disruption of aggregates compared to the Ferrosol , discussed in Section 7.4.2.

Lower rates of daily NNM at day 15, after a period of higher activity, agree with the theory put forward by Sierra (1992), who determined that the rate of mineralisation during one period would change the subsequent N mineralisation rate. Low rates of NNM after disturbance in horizons where aggregation was present, may therefore correspond to increases in microbial populations as they occupy the newly available sites. Hart *et al.* (1994) observed an increase in microbial biomass during the first seven days of incubation and associated decreases in mineral N production. Once the population has increased, NNM rates also increased and then stabilised. This relationship was seen in the unfertilised Kurosol. This study indicated stabilisation had occurred by around 15 days. Therefore the period between day three and day seven was a critical period for either population expansion, or species divergence, due to the mixing disturbances before incubation.

In the Kurosol day 3 had the highest rate of NNM of any of the incubation periods but this was only in the (P)N1Y treatment. Calculation of annual NNM at this time would have over estimated the rate at around four times greater than any other time

measured (at 190 kg N $ha^{-1}yr^{-1}$). These results agree with many authors who have observed short-term increases in C and N mineralisation during laboratory incubations (Van Gestel *et al.*, 1993; Murphy *et al.*, 1998b; Paul *et al.*, 1999; Pulleman and Tietema, 1999). In contrast, use of the NNM rate at Day 3 would have under estimated annual NNM in the (P)N1Y *P. radiata* litter by around one tenth (at 5 kg N $ha^{-1}yr^{-1}$). Such trends emphasise the importance of incubations long enough to go beyond the initial period of disturbance, to a length of at least 15 days, a period also noted as critical by Robertson (1988). Beyond this point, microbes are operating more in equilibrium (or steady state) and are more likely to represent the N turnover that naturally occurs in these horizons in the field. The use of short-term experiments to predict field NNM rates may overestimate the results (Sierra, 1992). The impact of mechanisms which influence NNM such as changing microbial populations, accessibility of N and oxygen supplies, and fluxes between mineralisation and immobilisation process, will all vary with the period of incubation.

Although both the fertilised and unfertilised pre-dried topsoil and pre-dried litter had significantly higher initial mineral N concentrations there was no significant effect on the overall rate of NNM. Lower enhancement of mineral N concentrations due to drying in Kurosol topsoil may be due to these soils undergoing similar large moisture fluctuations in the field. The lowest incubation moisture content in the pre-drying study, just above permanent wilting point, was not uncommonly measured in field (Chapter 4). Previous research has indicated that soil biota response to drying and re-wetting depend on prevailing site conditions (Birch, 1958; Lund and Goksoyr, 1980; Van Gestel *et al.*, 1993). West *et al.* (1988) demonstrated, using soils from a climosequence, that soil biota biomass from the lowest rainfall regions were more resistant to imposed gradual drying treatments.

7.4.2 *Eucalyptus regnans* litter on Ferrosol topsoil

Rates of NNM were very low at all times in the Ferrosol topsoils incubated moist regardless of fertiliser treatments. Immobilisation was observed to be a dominant process, with only topsoil fertilised at the highest rate, receiving a total of 1300 kg N ha^{-1}, actively mineralised N in excess of microbial requirements during incubations at

20 °C (32 kg N $ha^{-1}yr^{-1}$). The exception occurring when calculating NNM at Day 3. P1YN1Y topsoil was the most efficient mineraliser (the efficiency of N conversion NNM/total N is explained in Section 7.4.1) converting around two percent of the available N. In contrast, at 10 °C the unfertilised topsoil converted the most N, however, this was less than one percent of total N. Low rates of N mineralisation are common in forest soil (Stump and Binkley, 1993) and immobilisation of N is well recognised as a major component of the N cycle in eucalypt forest soil in Australia (Adams and Attiwill, 1986). Since I only measured NNM, I could not determine whether low gross rate of NM or high rate of N immobilisation occurred. However, the large amounts of immobilisation in the intermediate fertiliser treatments observed in this study indicate that high rates of immobilisation could occur, particularly in the Ferrosol soils. The results from this study also concur with the low rates of NNM observed in the field and laboratory studies presented in Chapters 4 and 6.

Changing the temperature reversed the treatment effects on NNM in the Ferrosol topsoil. Total N in Ferrosol topsoil was one third higher in the NIL treatment than the P1YN1Y (Table 3.12), which suggested higher total N content resulted in stronger temperature effects on N immobilisation. Comparisons between concentration were not significant, however it can be noted that the concentration of N was also higher in the unfertilised than annually fertilised topsoil at 0.18 and 0.13 %, respectively. In contrast, organic C concentration was lower (not significantly) at 3.8 and 4.5 %, respectively. Such trends suggest that there is a build up on recalcitrant organic matter as in this soils as suggested by (Mathers *et al.*, 2000), which could be derived from the microbial biomass (DeMontigny *et al.*, 1993).

Increased N immobilisation with higher incubation temperatures was also noted by Bonde and Rosswall (1987), who suggested that increasing the temperature could increase the metabolic rate, size or number of species of microbes resulting in greater net N immobilisation. In the field in Tasmania soil temperatures these temperatures were rare and thus the marked change in population dynamics in Ferrosol topsoil may not occur under field conditions. This suggests that laboratory incubation parameters have vastly accelerated NNM the processes.

In the field *in situ* rates of NNM, calculated using closed cores, were higher (not significantly) in unfertilised Ferrosol topsoil at 23 kg N ha^{-1} yr^{-1} than the fertilised (P1YN1Y) topsoil, at 16 kg N ha^{-1} yr^{-1} (Chapter 4). This supports the observations from topsoils incubated at 10 °C in the laboratory. In laboratory incubations held at 10 °C, unfertilised topsoil, mineralised 14 kg N ha^{-1} yr^{-1}, a similar rate to that observed in the field, while P1YN1Y topsoil mineralised less than 4 kg N ha^{-1} yr^{-1} (Table 7.15). However, in laboratory incubations held at 20 °C, unfertilised topsoil immobilised 14 kg N ha^{-1} yr^{-1}, but P1YN1Y topsoil mineralised 32 kg N ha^{-1} yr^{-1}. Although not consistent with field observations, these results were in agreement with the laboratory study on moisture and temperature where NNM often doubled in P1YN1Y topsoil when the temperature increased from 10 °C to 22 °C, but showed inconsistent variations in NNM due to temperature changes in unfertilised topsoil, (Chapter 6).

Application of N and P alone, every second year, immobilised significantly less N than when N and P were applied together. A reduction in mineralisation from P application contrasts to results observed by Falkiner *et al.* (1993) in mixed forests and *P. radiata* plantations. Under these forests rates of NNM increased after P application (200 to 500 kg P ha^{-1}) for at least 2 years, and were readily availably for plant uptake. However, in the current study as no increase in volume growth occurred due to P application and P was not thought to be limiting at this site (Chapter 3). When P is not limiting due to the site history such as ex-pastures (Aggangan *et al.*, 1998) or naturally high P concentrations (Johnson *et al.*, 1980; McLaughlin *et al.*, 2000), addition of P fertiliser may have little effect on NNM. Net N mineralisation may actually be reduced after P application as a result of increased organic inputs and associated microbial activity (Adams and Attiwill, 1991; McLaughlin *et al.*, 2000). Such behaviour was observed in the present study. However when P is limiting, low rates of P application, 100 kg P ha^{-1}, can cause dramatic increases in total N accumulation and wood production, with increased N accumulation for up to 30 years after fertilisation (Falkiner *et al.*, 1993). Results similar to those observed by Falkiner *et al.* (1993) would be expected in the P-poor Kurosol under the *P. radiata*.

In comparison to the topsoil, the largest total N pool was measured in P1YN1Y *E. regnans* litter, which also corresponded to the most effective mineralising substrate, converting twelve percent of the N annually. This emphasises the importance of the litter horizon in supplying N for tree growth at this site. In a review of litter quality and annual NNM across a range of sites in northern America, Scott and Blinkley (1997) observed that both litter N concentrations and N content correlated poorly with NNM. In contrast, annual rate of NNM measured in *E. regnans* litter in this study correlated well with the total N measured at the time of sampling.

Decreasing the incubation temperature of litters decreased the differences between NNM rates due to fertiliser treatments. At the lower incubation temperature, NNM was similar between unfertilised and intermediate fertilisation. The exception occurred when N was applied alone to *E. regnans* litter. Unlike the topsoil, no field rates of NNM were measured in litter horizons at either site. However, field examination of mineral N in Chapter 4 and 5, clearly showed that large long-term differences in mineral N content of litter horizons developed due to fertilisation.

Laboratory incubations have often been used to compare N mineralisation or nitrification potential of soils of different regions, ecosystem types, topographic positions, or ages (Nadelhoffer, 1990). However, comparisons between field and laboratory rates of NNM using air-dried soil are not always clear. In agreement with this study, Connell *et al.* (1995) found a poor correlation ($r^2 = 0.20$) between *in situ* rates of mineralisation and laboratory rates, which were of up to ten times greater. In this study drying increased rates of NNM by up to twenty-fold, which could have a large effect on the calculated rate of annual NNM. For example calculating annual NNM from 60-day incubations of P1YN1Y treated topsoils air-drying pre-treatments resulted in 230 kg N $ha^{-1}yr^{-1}$ compared to 32 kg N $ha^{-1}yr^{-1}$ in those maintained moist (Table 7.15). Previous research has shown that pre-drying soil can result in substantial microbial death. The soil micro-organisms which survive, respond to re-wetting by temporarily entering a state of high metabolic activity, resulting from the cannibalisation of the dead microbial cells and a burst of humus decomposition (Lund and Goksoyr, 1980). Substantial increases in NNM rates of the Ferrosol topsoil

suggest that drying this soil significantly changed the natural metabolic rate and therefore mineralisation rates of the microbial communities.

In addition, drying changed the difference in NNM rates between fertiliser treatments, and this effect was dependent on the incubation length. In their study Lund and Goksoyr (1980) noted that changes in microbial activity in a soil after drying and re-wetting proceeded in waves, due to bacterial and fungal populations having different growth and activity patterns. From the field study in Chapter 4, there was no indication that the Ferrosol topsoil reached moisture content as low as air-dried laboratory topsoil. The field study also did not measure mineral N content above 20 $\mu g\ g^{-1}$. Both of these factors indicated that air-drying, to the level used in this study, was not a very likely natural occurrence at this site, and therefore air-drying prior to incubation probably resulted in a significant overestimation of the amount of N available for tree growth.

In agricultural soils, both Cabrera and Kissel (1988b) and Van Gestel *et al.* (1993) observed significant increases in NH_4^+ and NNM after drying and re-wetting. Cabrera and Kissel (1988b) observed drying and sieving resulted in an over prediction of the amounts N mineralised by, between 60 and 340 percent, compared to rates measured in the field. In agreement with the current study and the previous laboratory study (Chapter 6) Cabrera and Kissel (1988b) noted that drying soils enhanced nitrate production. This contrasted to the increase in ammonifying organisms and ammonification from drying observed by Stevenson (1956) and Van Gestel *et al.* (1993). Although the amount of nitrate increase was small, (less than 1 $\mu g\ g^{-1}\ NO_3^{-1}$) it was up to a five-fold increase. Nitrate was not found in field samples (Chapter 4 and 5) or laboratory samples maintained moist. Ammonification was the dominant N mineralisation process in many forest soil (Adams and Attiwill, 1986), and was consistently observed as the dominant process in both the forest soils examined here. Hence, drying soil prior to laboratory incubations could over predict the amount of nitrate present and lead to erroneous conclusions about N leaching.

Low nitrification rates were observed at both sites throughout the 60-day incubation. The maximum net nitrification measured was only 0.16 $\mu g\ g^{-1}$ at day 60 in the NIL *P.*

radiata litter. At both sites, rates on net nitrification remained below 0.01 μg g^{-1} day^{-1}. In a study of N mineralisation and nitrification in soils at 17 sites, including 10 coniferous stands, Vitousek *et al.* (1982) observed that litter horizons had a threshold for N between 60 and 90 μg g^{-1} above which net nitrification occurred. In this study, cumulative rates of mineral N reached a maximum in the *E. regnans* litter at 244 μg g^{-1}, however rates of net nitrification remained low at 0.09 μg g^{-1}.suggesting that some other mechanism other than the availability of ammonium was limiting net nitrification. Vitousek *et al.* (1982) observed in mineral soil (0-15 cm) the pattern was more linear with high mean mineral N concentrations in the field associated with high net nitrate production. Again this was not observed in this study. Both studies week conducted as aerobic laboratory studies over an eight period, and in this study the rate of nitrification did not increase over the incubation period, which suggest that the length of the incubation period was not the limiting factor.

Previously I have discussed that the low pH at these sites was probably the factor limiting net nitrification. However soils with low pH can have mineral N dominated by nitrate (Bauhus and Khanna, 1994). In a study of drying and wetting on two acid forest soils in south-east Australia, Bauhus and Khanna (1994) observed that nitrification was more constrained by low soil moisture than ammonification. However, results from this study indicated that samples incubated at near field capacity did not have significant net nitrification. Net nitrification was only evident in these soils after air-drying (Chapter 6).

Large differences in clay and organic matter content could also influence NNM response to drying prior to laboratory incubation. Disruption of soil physical structure, substrate desorption from soil surfaces during soil desiccation, and re-wetting can release organic compounds available for subsequent mineralisation (Degens and Sparling, 1995). Bartlett and James (1980) showed drying soil increases the solubility of organic matter, seen by the increased yellow colour in solution extracts. Such colour variations were observed during the analysis on mineral N extracts during this study. However, these variations were not characterised. The relative amounts of organic N released have been associated with the previous aggregation state (Van Gestel *et al.*, 1991) and clay content of the soil (Wetselaar, 1968; Cabrera and Kissel, 1988a). Strong *et al.* (1999a) not only observed that clay content often had a

significant influence on N mineralisation and nitrification response to disturbance, but also observed that when soils were maintained moist the relationship between clay content and N mineralisation and nitrification was negative, while drying and re-wetting resulted in a positive relationship.

Disturbance effects were also observed in short incubation periods. In short incubation period effects due to disturbance (mixing) were particularly evident in fertilised litter and topsoil, observed as a depression in daily rates at day 3 compared to day 7, settling to an intermediate rate by day 15. In contrast, unfertilised topsoils had increased rates of fertilisation at day 3. This large disturbance affect was associated with the relatively high aggregate stability. The percent of water stable aggregates (>0.25 mm) was between 69 and 84 % in the Ferrosol topsoil (Table 3.11).

The large variation in clay and organic matter content between the Kurosol (loamy sand, A1 with approximately 4 % clay) and Ferrosol (clay loam to loam, A1 with approximately 50 % clay) was also considered a factor in the differences in disturbance response between the soils. Cabrera and Kissel (1988a) observed that N mineralisation over-prediction, using disturbed samples, was related to the clay and total N concentration within soils. The higher the clay to total N ratio, the larger the physical disruption due to disturbance such as sieving, the greater the change in accessibility to organic matter, and the higher the over prediction of NNM. Even during long incubations, 224 days, if the period of greatest variation in NNM was included, the first 28 days, N production was over-predicted in disturbed samples (Cabrera and Kissel, 1988a). To prevent this the incubation of intact cores has been suggested to better reflect field conditions (Adams and Attiwill, 1986; Raison *et al.*, 1987). However, results in this study indicated that an extremely high number of intact soil-core replicates would be required to show this in a statistically consistent manner.

Changes in NNM rates due to drying may also reflect major shifts in the activity of soil populations such as a change in the microbial population base to a more fungal dominated one that is better adapted to low moisture environments. Bauhus and Khanna (1994) observed an increase in the C: N ratio of microbial biomass, which indicated an increase in the proportion of fungi due to drying and wetting soils.

Moisture content and pore size distribution will determine the ability of different microbial structures to move through water filled pathways. Many bacterial activities in soil have been shown to decrease sharply as matric potential in the soil falls to between -50 kPa and -30 kPa. As filamentous fungi do not suffer from the limitation of movement imposed by unicellular structure, the relative competitive advantage of bacteria and fungi in soil changes markedly as moisture content declines (Harris, 1981). Such response to drying alone was also evident in the Ferrosol topsoil from the first laboratory study (Chapter 6). Changing populations emphasise the importance of selecting an appropriate incubation time to determine the rate of NNM.

Air-drying the *E. regnans* litter layers resulted in an increase of mineral N prior to re-wetting, by up to five-fold over the litter kept moist and cool. However, after re-wetting rates of NNM decreased. This contrasts to NNM enhancement due to drying in the Ferrosol. Clein and Schimal (1993) and Pulleman and Tietema (1999) also observed that the effect of drying on N mineralisation and microbial activity differed in the litter and soil. In the field, litter horizons are more exposed and are therefore naturally more prone to drying and re-wetting than the soil. Under these conditions there would be selection pressures for desiccation-resistant microbial populations. Both Clein and Schimal (1993) and Pulleman and Tietema (1999) observed increased microbial activity within a few hours of re-wetting, and the effect of air-drying on overall N mineralisation was found to be dependent on the severity and length of drying. The length and severity of the drying event in the litter affected the overall reduction in microbial species diversity and subsequent re-colonisation time (Clein and Schimal, 1993).

The decreased impact of drying and re-wetting on N mineralisation in the litter horizons would also result from a reduction in the severity of physical change due to drying. The litter has larger poor spaces and limited aggregation, compared to highly structured soils, such as the Ferrosol topsoil. Consequently, there would be limited fractionation of the particles resulting in a reduction of newly available surfaces of organic matter for mineralisation.

In addition to changes in N availability discussed in this study and previous research, other studies, for example Bartlett and James (1980) noted that drying and re-wetting soils affected the solubility of exchangeable Mn, K, P, Ca and Mg and soil flocculation. This affects the nutrients available to the soil biota, and subsequently the size of the mineralisation flush due to re-wetting.

Results from this study suggest that removal of the first seven days of cumulative N mineralisation would remove the initial flush of N mineralisation caused by disturbance. However, they would not remove the effects of air-drying in Ferrosol topsoils, which are still evident at days 30 and 60. Cumulative rates of NNM in topsoil also showed a distinct divergence between fertilised and unfertilised horizons between days 30 and 60. At this point, it was proposed that in unfertilised samples there was depletion in available organic N for mineralisation or a build up of waste products. However, depletion of organic N availability was more likely, as fertilised samples continued actively mineralising N. A similar divergence trend at day 48 was observed by Johnson *et al.* (1980) in their 100-day laboratory study, of N or N plus P composite mineral soil and litter horizons. In the present study, calculation of NNM between days 30 and 60 produced N mineralisation rates two to three times higher in fertilised litters than unfertilised. Another explanation for the general reduction in the rate of NNM in unfertilised horizons between days 30 and 60 could be a change in soil biota populations to a fungal dominated one. Bonde and Rosswall (1987) hypothesised that in long-term incubations decomposition of a large part of the microbial biomass would occur within the first few weeks, resulting in a favouring of fungal populations in the later part. This change in decomposer communities may also reflect a change in the amount and type of available C. These results indicated for the sites examined in this study incubation length of 60 days were adequate to measure changes in NNM from fertiliser treatments.

Changes in NNM during longer incubation times suggest that the isolation of small amounts of soil may impose unnatural restrictions to N processes. However, any removal of samples from the forest will change the natural cycling of N, due to the absence of litter inputs and leaching. In a forest situation leaching of mineralised N from the litter horizon and uptake by mycorrhizal fungi and tree roots result in an

internal dynamic between the litter and soil horizons. The concentration of tree feeder roots at the litter and soil horizon interface indicates the importance of this process on tree growth.

Combining the sites as replicates indicated that rates of NNM in topsoil could not be predicted by total N, P, Ca, Mg or S concentrations or by soil pH. In an examination of 38 podzolized sands (0-15 cm) under similar laboratory conditions (56 day aerobic incubation) Carlyle *et al.* (1990) also observed no correlation between NNM and total C, total N or C/N ratio. This was despite the soils being of similar texture, parent material and weak aggregation, i.e., similar to Kurosol topsoil. Correlations were observed between total N and NNM when the soils were split into highly and weakly nitrifying soils. In this study low rates of nitrate were observed in both soil types. In contrast, Carlyle *et al.* (1990) observed a significant correlation between NNM and total P in the soil. However it was difficult to determine relationship with nutrients in the soil as this study was complicated by additions of various nutrients added together in the fertilisers (N, P, Ca and S) and depletion of nutrients by increased tree growth (Mg). For example, the correlation of NNM with Ca and Mg was negative for the Kurosol and positive for the Ferrosol topsoil. Calcium differences relate to the large amounts of Ca added with superphosphate at the Ferrosol topsoil, while concentrations declined with uptake in the Kurosol topsoil. Magnesium concentrations declined in both topsoils however increased immobilisation during incubations of intermediate fertiliser applications (every second and fourth year) resulted in a positive correlation in the Ferrosol topsoils. Such comparisons indicate that negative correlations between NNM and nutrients measured in this study would not reflect growth limitations as a result of restrictions in N cycling.

NNM rates from the combined litter data were significantly correlated to total N, P and Mg for incubations at 20 °C. Although relationships were strongly influenced by NNM rate of the P1YN1Y treatment. However, if this treatment was excluded from the regression retains a significant relationship. This was in agreement with the analysis of sites separately, where litter often had stronger correlations than the underlying soil. .

7.5 Conclusions

The influence of fertilisation on N cycling in litter produced clear results, i.e. daily rates of mineralisation were higher in annually fertilised than unfertilised litters, at both sites if incubations went for 7 days or longer. In contrast, low mineralisation rates in both topsoils often produced similar daily rates of NNM regardless of fertiliser treatments. Increased replication of samples in laboratory experiments did increase the sensitivity of NNM measurements and significant differences between fertiliser treatments could be measured. However, comparisons between the effects of intermediate fertiliser treatments on NNM rates were dependent on incubation temperature.

In topsoils from both sites, low and often negative rates of NNM during 60 days may have indicated that this period of incubation might not have been enough to overcome the effects of disturbance at intermediate rates of N fertilisation. However, results from experiment 2 showed that shorter incubation periods of 3 to 7 days generally resulted in enhanced rates of NNM compared to those measured in longer incubations. Longer periods of incubation lead to depletion in available substrate or a build up of products that negatively feeds back on microbial activity. However, in both topsoils, 60 days was required to produce a significant divergence in cumulative NNM rates between the fertiliser treatments during the later stage of the incubation.

Overall, soil type had a much larger affect than temperature on NNM trends, i.e. at both temperatures, the Kurosol topsoils were net N mineralisers, while the Ferrosol topsoils were net N immobilisers. However, temperature influenced changes in NNM due to the amount of fertiliser supplied. For example, decreasing the incubation temperature from 20 to 10 °C significantly decreased the rate of NNM, in all except the unfertilised Ferrosol topsoil. Decreasing the temperature also reduced the strength of correlations between NNM and measured chemical parameters. However, incubation temperatures of 10 °C, in this study, were considered to more closely reflect the natural environment for microbial mineralisation in the field. As the effect of temperature was not linear or consistent across the horizons or sites, the incubation

temperature of 10°C was considered the most appropriate temperature to indicate actual field rates of NNM.

This study indicated that enhanced NNM rates in topsoils would occur if soils were not maintained moist prior to and during incubations. Samples maintained moist more closely reflected natural changes in the litter and soil quality and quantity, rather than changes caused by air-drying. Drying events occur in field environments and are a natural process that causes flushing of NNM. However, the length and severity of the drying event could determine microbial population species diversity and activity. As a result, pre-drying both the topsoil and litter horizons in laboratory incubations can produce water contents not previously experienced by microbes and could lead to NNM rates outside the sites natural range. Results indicated that there was a greater change in NNM in both litter and topsoil horizons due to air-drying in the inherently wetter Ferrosol site than the inherently drier Kurosol site. In unfertilised Ferrosol topsoil air-drying increased initially negative NNM rates (immobilisation) to NNM rates above 100 kg N ha^{-1} yr^{-1} at 30 or 60 days. In contrast, pre-drying increased NNM rates in unfertilised Kurosol topsoil from between 26 to 75 kg N ha^{-1} yr^{-1}. These results are not inconsistent with the hypothesis of microbial pre-adaptation to moisture stress in the sandier Kurosol topsoil and *P. radiata* litter at the drier site.

Examination of the relationship between substrate nutrient content and NNM was limited by the low replication of treatments at both sites and confounding of the addition of some nutrients during fertilisation, i.e. N, P, Ca and S. Rates of NNM in litter substrates (O2, combined *P. radiata* and *E. regnans*) were significantly correlated with total N, P and Mg for incubations at 20°C. In contrast, in the combined Ferrosol and Kurosol topsoil dataset, NNM rates were not related to total N, P, Ca, Mg or S concentrations or soil pH. It can therefore be concluded that, from the sites and conditions studied in these experiments, NNM was more sensitive to the nutrient content of litter than to that of soil. Concentrations of N and P in litter were positively correlated with NNM, probably because fertilisation improved litter quality. In contrast, Mg concentrations were negatively correlated with NNM, probably because N fertilisation decreased Mg availability in soil and Mg uptake, while Mg concentrations in the range studied did not limit NNM of litter (Chapter 3).

Chapter 8. Simulation of nitrogen dynamics and *Pinus radiata* growth in response to fertilisation.

8.1 Introduction

Nutritional management of plantations requires information on soil and site characteristics, and clear guidelines on what and where fertilisers are required. While information of this type is available, much of it is restricted and not reliable across sites and regions. Site availability and uniformity are often a major restriction to experiments on nutritional management of plantations. The effects of site restrictions were highlighted in this study as results were often limited by the replication established at the beginning of the project in the early 1980s. For example, examination of the relationship between substrate nutrient content and NNM was restricted by the low replication of treatments at both sites. At the *P. radiata* site, most of the variability in results, particularly NNM, was observed within the treatment plot replication. Further complication arises when trying to assess long-term change independent of short-term variations, such as the periodic low rainfall (Chapter 4) and insect attack (Chapter 3).

One tool increasingly used to inform silvicultural practices is process-based modelling. Models can summarise the results of many experiments by incorporating hypotheses and conclusions into a quantitative framework. These models predict tree growth in response to the environment and silviculture and they can assist in identifying factors which limit growth (McMurtrie and Landsberg, 1992; Battaglia and Sands, 1998). In addition, they allow the assessment of risk involved both in terms of changing climate effects on economics (i.e. drought) and possible offsite movement of fertilisers.

Process-based models such as CABALA (Carbon Balance) are hypotheses of the cycles and interactions between C, N and water within a forest (Battaglia *et al.*, 2004). As part of a silvicultural decision support system for forest managers, CABALA draws on and combines existing concepts and sub-models of tree and stand growth, light interception, canopy growth, water use and C and N cycling. The N mineralisation sub-model of CABALA is that of the CERES model, which has been

used to model N dynamics and growth in several agricultural ecosystems (Goodwin and Jones, 1991). Once validated for a given plantation system, CABALA can be used to develop a range of hypotheses about stand and ecosystem functioning. In addition, comparisons between the two (model and observed) outcomes may highlight areas where experimental data is not available or where observed results lie outside standard outcomes. For example, with N mineralisation tuning, CABALA successfully simulated some aspects of N dynamics in a *E. nitens* plantation in Tasmania (Smethurst *et al.*, 2004a), but predictions beyond 2.8 years were biased mostly by parameters for allocation and plant N concentration that were suited only to much younger plantations. As N dynamics in the soil and litter had not been measured at the site these aspects of CABALA required further validation.

A limitation for this thesis is that the model had not previously been validated for *P. radiata* or *E. regnans*. Results presented in earlier chapters of this thesis therefore could be used to address this need, and physiological parameters suitable to run CABALA for *P. radiata* have recently become available.

The objectives of this chapter, therefore, were to compare observed attributes of the *P. radiata* site with those predicted by CABALA; including key components of the N cycle with and without N fertilisation, and thereby determine the strengths and weaknesses of CABALA for this application. If the validation was satisfactory, there would then be the opportunity to predict aspects of system behaviour that were not measured, including the extent to which low water availability limited tree growth.

8.2 Model description and methods

A full description of CABALA is given by Battaglia *et al.* (2004). The model works on a daily time step to calculate net daily production of stand biomass. Stand biomass, a set of identical trees (with the exception of edge trees), is divided into separate compartments where net daily production is allocated (foliage, roots fine and coarse, stem sapwood and heartwood, branches and sapwood of branches and tree bark). Biomass losses from the stand occur through litter fall and biomass turnover. A one-dimensional water balance model allocates tree water use from a number of overlapping compartments. Nitrogen inputs in the stand include fertiliser additions,

atmospheric inputs and N mineralisation. Atmospheric inputs at this site were set at 2.5 kg N ha^{-1} yr^{-1}, while fertiliser inputs are described in Chapter 3. Nitrogen mineralisation is calculated by a N turnover sub-model in CABALA, the CERES model (Goodwin and Jones, 1991). In CERES, N mineralisation in humus fraction is calculated as,

$$RHMIN = NHUM\ (L) \times DMINR \times TF \times MF \times DMOD$$

RHMIN = N mineralised in the humus
NHUM (L) = N associated with a stable humus fraction
DMINR = humic fraction decay rate (1/ days)
TF = temperature factor, which approximates the soil temperature effects on ammonification.
MF = moisture factor, which approximates the soil moisture effects on ammonification.
DMOD = the amount of chemical or physical protection that organic matter has at a given site. Used to adjust the mineralisation rate on atypical soils (<1 equates to slow mineralising sites and >1 relates to sites where mineralisation occurs faster than expected).

CABALA requires daily weather inputs, climate data used to drive the model in this study was simulated data based on the interpolation of local meteorological stations and calculated within a range of 5 km of the site. Other inputs including silvicultural practices such as tree spacing, thinning, pruning and fertilising, as well as the site parameters such as soil pH and N concentration, are described in Chapter 3. Simulations were initiated at planting age with identical values of initiation and parameterisation for unfertilised and fertilised trees. The parameterisation of CABALA for *Pinus radiata* was derived from "the biology of forest growth experiment" data (Benson *et al.*, 1992a) and provided by Dr Barrie May CSIRO pers. comm (Commercial in confidence). A summary of the site details at the beginning of the simulation is given in Table 8.1.

To predict growth responses of NIL treated trees at this site the predicted rate of N mineralisation needed to be lowered. That is, the degree of humus protection, DMOD, was adjusted to 0.7, using the mineralisation rates indicated in the field study discussed in Chapter 4. Once this was completed the predicted volume growth responses were similar to those observed during the fifteen years of annual measurements. No further parameterisation changes were required to fit simulated

data to those observed. Leaf area index (LAI) was measured at age 36 years using both a visual guide (Cherry *et al.*, 2002) and the LICOR LAI2000 (Cherry *et al.*, 1998). Other observations are described in previous chapters.

Table 8.1 Summary of site details at the beginning of the simulation

Description	Site and Initial Plant Variables		
Latitude			-41.67
Species			*Pinus radiata*
Seedling Description	Seedling height (cm)		15
	Leaf area (cm^2)		200
	Foliar N conc (g/g)		2.5
interrow spacing (m)			3.0
intrarow spacing (m)			2.2
row direction			340
Thickness of Horizon (cm)			60
Initial plant available water (mm)			600
Depth permanent water table (cm)			1000
Drainage			0
Salinity			0
Hard pan			0
Depth (cm)	**0 - 10**	**10 - 20**	**20 - 50**
Organic C	1.3	1.4	0.7
C:N ratio	23	30	16
Bulk Density	1.6	1.7	1.8
soil pH	3.8	3.8	3.8
DMOD (scalar 0-1)*	0.7	0.7	0.7

*** degree of organic matter protection**

8.3 Results of Simulations

The performance of CABALA for predicting volume growth was evaluated by comparing predicted and observed stand volume of fertilised and unfertilised trees during the fifteen years of annual measurements (Figure 8.1). The large drop in predicted volume at age 20 years was due to thinning that occurred at establishment of the fertiliser experiment. Trees were waste thinned and provided a predicted 36 t ha^{-1} of slash. As the version of CABALA used in this study assumes that P was not limited, both the NIL and P only fertilised treatments were compared to the predicted volume growth of unfertilised trees. Phosphorus was applied at ages 20 and 26 years and was observed to significantly increase *P. radiata* growth (Chapter 3). By the third application of N fertiliser, predicted volume growth of N fertilised trees was 30 % higher that that observed. By age 34 years, predicted stand volume was within 15 % of that observed. Figure 8.2 a and b shows that the quality of fit for predicted *vs*

observed values is very similar for (P)N1Y stand volume, but declines in the NIL and (P) stand volumes. The quality of fit was also tested for the first application of P only, this decreased the slope slightly ($r^2 = 0.99$).

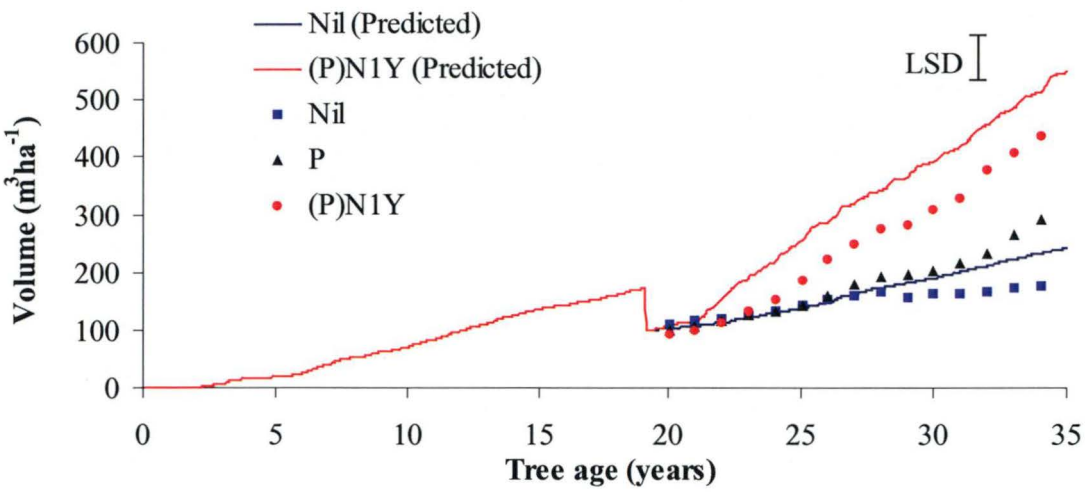
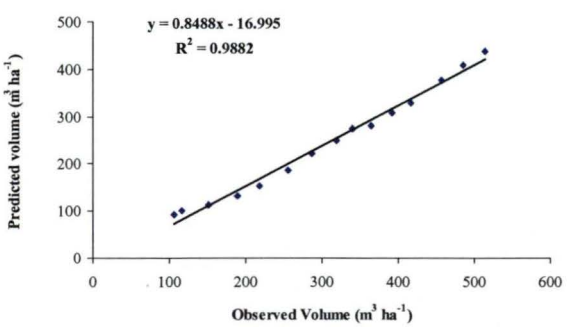


Figure 8.1 Volume growth in three treatments at the Kurosol site. Lines indicate predicted growth using CABALA. Points indicate observed growth as described in Chapter 3. Bar indicates LSD of observed values.

a.



b.

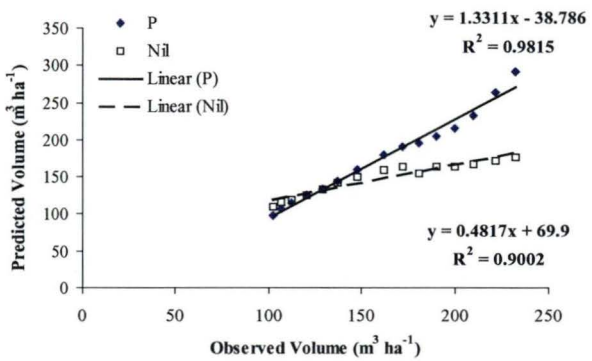


Figure 8.2. Comparison of observed and predicted stand volumes in the (a) (P)N1Y treatment, and (b) NIL and P treatments.

CABALA simulates the movement of N throughout the forest system and predicts N mineralisation and uptake separately in three layers; layer 1 (0-10 cm), layer 2 (10-20

cm) and layer 3 (20-50 cm). It is assumed that mineral N below 50 cm is unavailable to the tree. In all three layers of mineral soil CABALA predicted low rates of monthly nitrification, resulting in nitrate content generally below 1 kg N ha^{-1}, in both fertilised and unfertilised treatments (Figure 8.3). Observed NO_3^- contents at the end of the fertiliser experiment (age 34 years) were also low or below detectable limits. In contrast to NO_3^-, predicted NH_4^+ concentrations increased substantially in the top 20 cm of soil after each annual fertiliser application (Figure 8.4). Predicted NH_4^+ increases occurred for a period of six to twelve months after fertilisation. In subsoils (20 – 50 cm), CABALA predicted a large long-term increase in the concentration of NH_4^+ due to fertilisation.

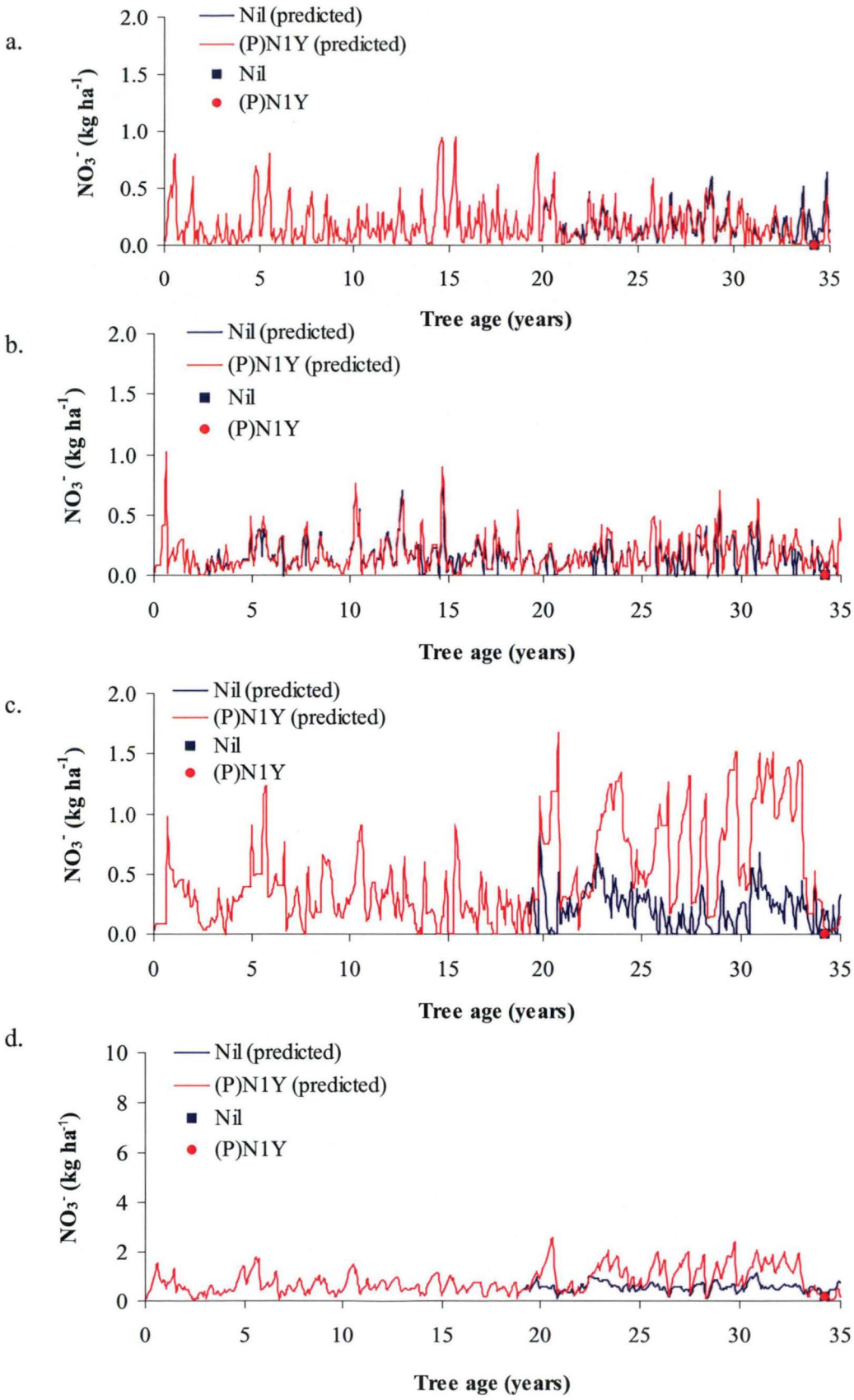


Figure 8.3 Predicted and observed nitrate content in fertilised and unfertilised Kurosol profiles at depths (a) 0-10 cm, (b) 10-20 cm, (c) 20-50 cm and (d) 0-50 cm.

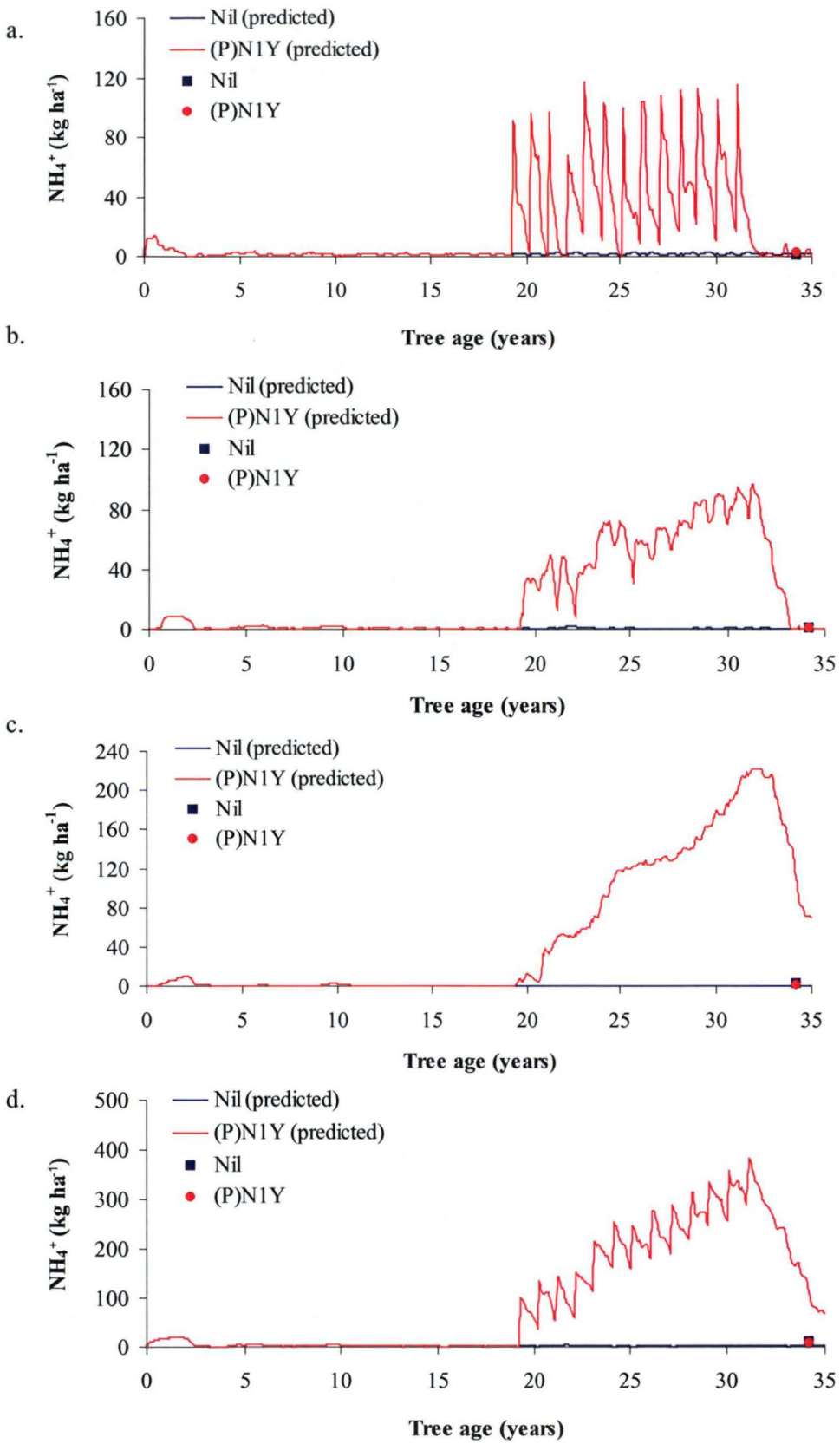


Figure 8.4 Observed and predicted ammonium content in fertilised and unfertilised Kurosol profiles at depths (a) 0-10 cm, (b) 10-20 cm, (c) 20-50 cm, and 0-50 cm. (LSD of observed ammonium content was 1.8 at age 34 years)

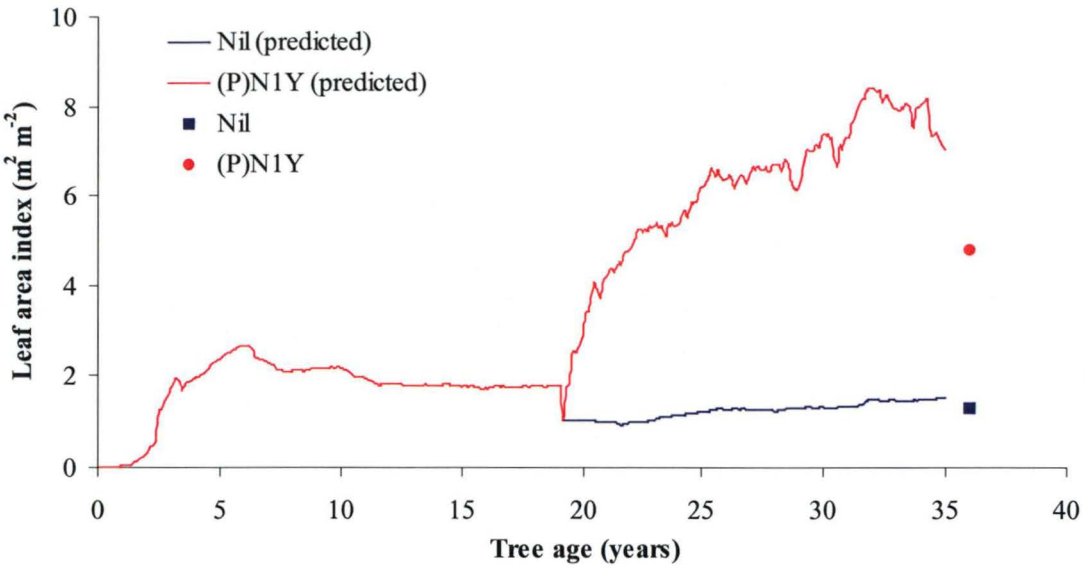


Figure 8.5 Observed and predicted Leaf area index of *P. radiata* fertilised and unfertilised trees.

Predicted leaf area index (LAI) of fertilised tress increased significantly following the commencement of fertilisation at age 20 years and reached a maximum of 8.4 at age 32 years. By age 35 years LAI had declined to a value of 7 (Figure 8.5). Field measurements at age 36 years indicated that the LAI of fertilised trees was approximately 4.8. In contrast, observed and predicted values for unfertilised trees were similar at 1.3 and 1.5, respectively. Simulated crown volume prior to thinning was 27816 m^3 ha^{-1}, which was thinned to 16149 m^3 ha^{-1}. Simulating crown volumes during the fertiliser experiment indicated that, without fertilisation, trees were only able to reach their pre-thinning volume, while fertilised trees had doubled the volume, at 53274 m^3 ha^{-1}.

Monthly N mineralisation predicted during the experiment was similar between fertilised and unfertilised topsoil (0-10 cm layer) (Figure 8.6). This resulted in predicted annual N mineralisation during 2000 of 6.0 and 8.6 kg N ha^{-1} in unfertilised and fertilised topsoils, respectively. The predicted range of monthly N mineralisation in the fertilised topsoil ranged from –0.4 and 1.6 kg N ha^{-1} and in unfertilised topsoil between 0.3 and 0.9 kg N ha^{-1} (Figure 8.7). Observed in situ NNM was extremely

variable and often higher than that predicted.

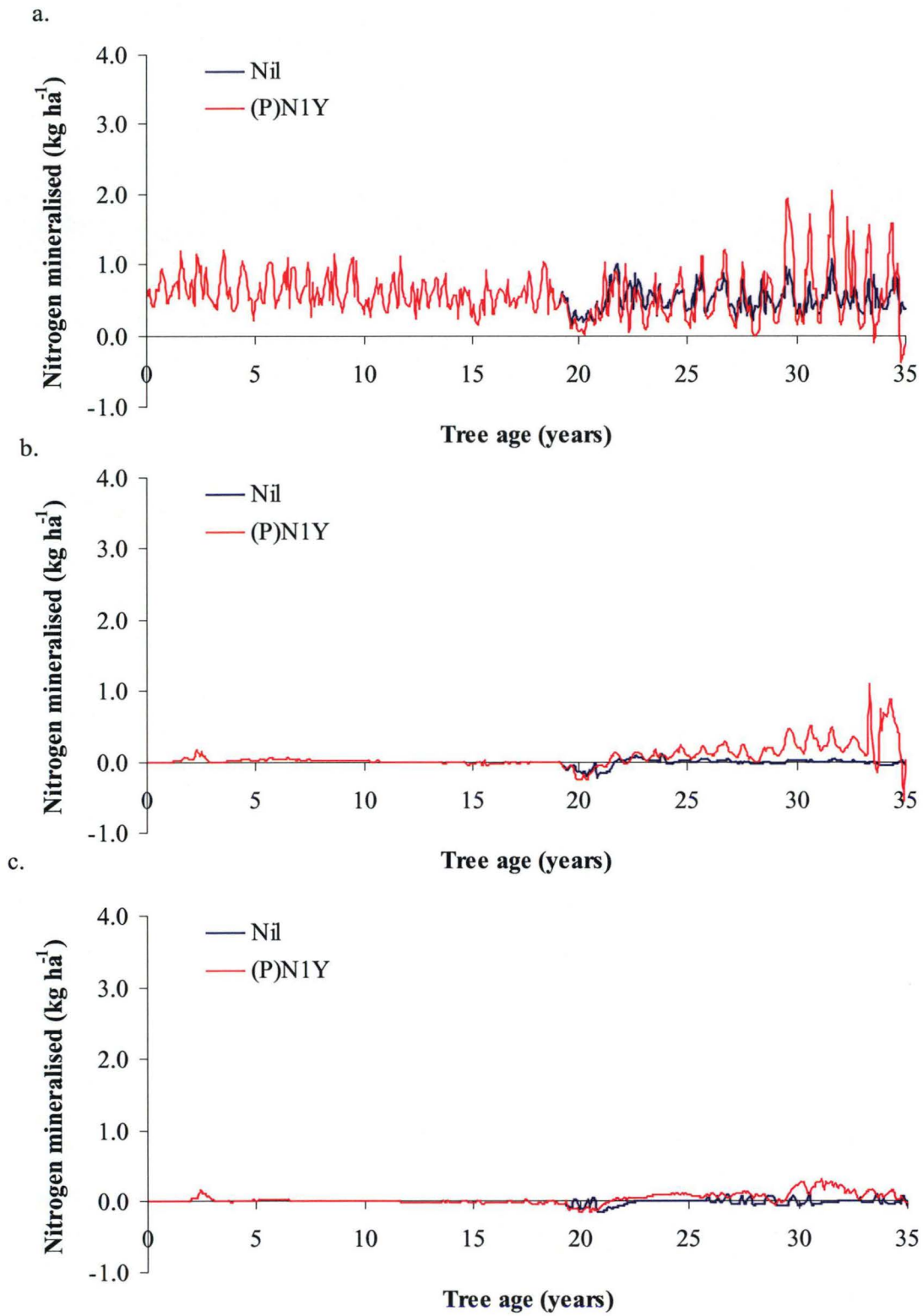


Figure 8.6 Predicted monthly N mineralisation (kg ha^{-1}) in the (a) 0-10 cm (b) 10-20 cm and (c) 20-50 cm depths of the fertilised and unfertilised Kurosols.

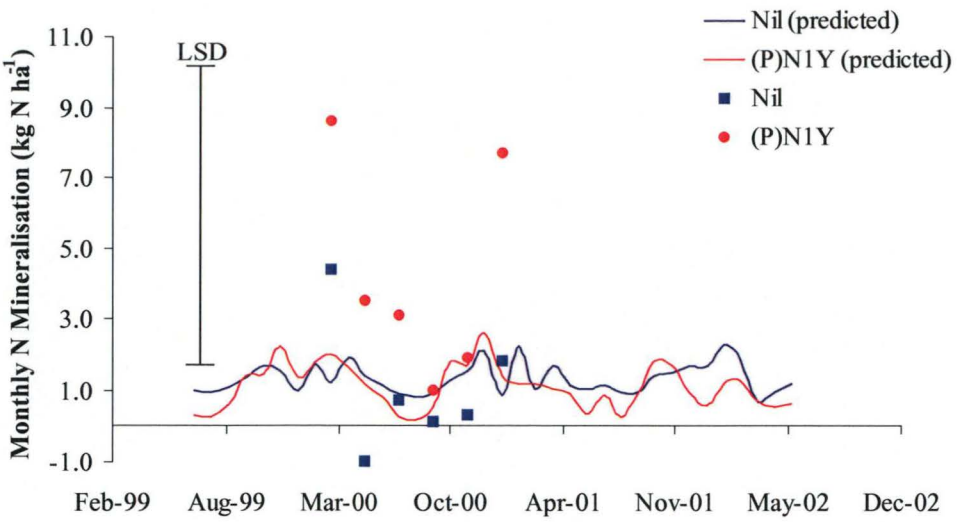


Figure 8.7 Observed and predicted monthly N mineralisation (kg ha^{-1}) in the 0-10 cm depth during period of the *in situ* sampling at fertilised and unfertilised sites.

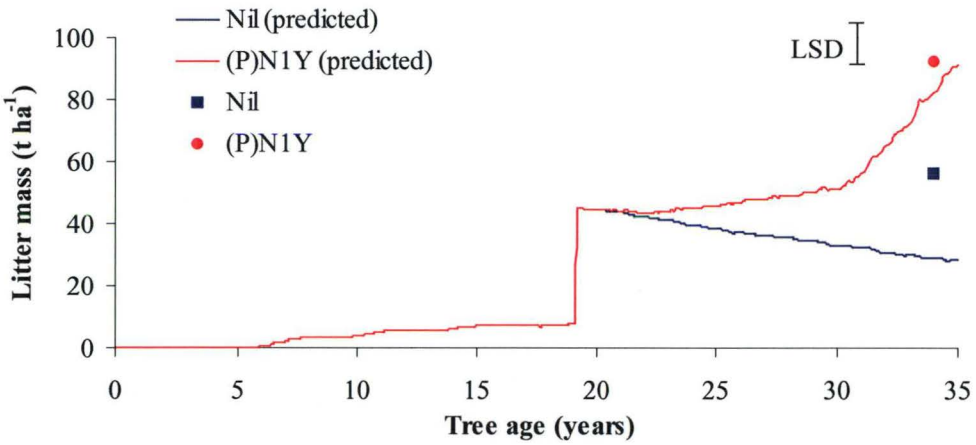


Figure 8.8 Observed and predicted leaf litter mass (kg ha^{-1}) includes foliage, branch, bark, stemwood and duff at fertilised and unfertilised sites.

Predicted litter amounts were slightly lower at 29 and 83 t ha^{-1} for unfertilised and fertilised treatments at age 34 years, than those measured in the field at 56 and 92 t ha^{-1}, respectively. (Figure 8.8), Increased litter inputs were also predicted between the ages of 32 and 35 years (Figure 8.9).

The temporal patterns of predicted NNM in the litter (O1 horizon) and duff (O2 horizon) are shown in Figure 8.10. Predicted annual rates of NNM in litter and duff were 1.7 and 4.9 kg N ha^{-1} yr^{-1} in unfertilised and fertilised treatments, respectively.

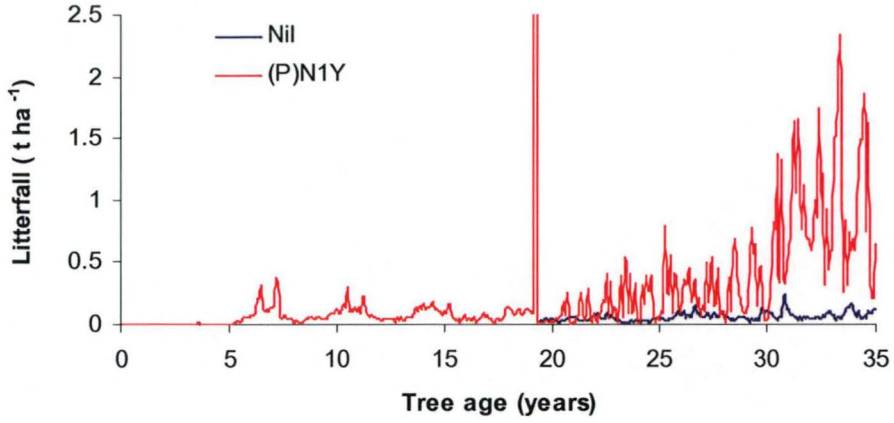


Figure 8.9 Predicted litter fall (t ha^{-1}) at fertilised and unfertilised sites.

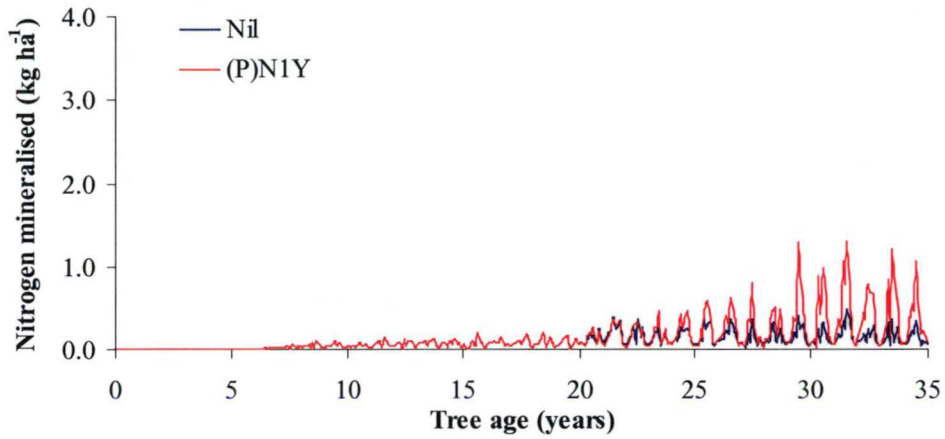


Figure 8.10 Predicted monthly N mineralisation (kg N ha^{-1}) in litter and duff at fertilised and unfertilised sites.

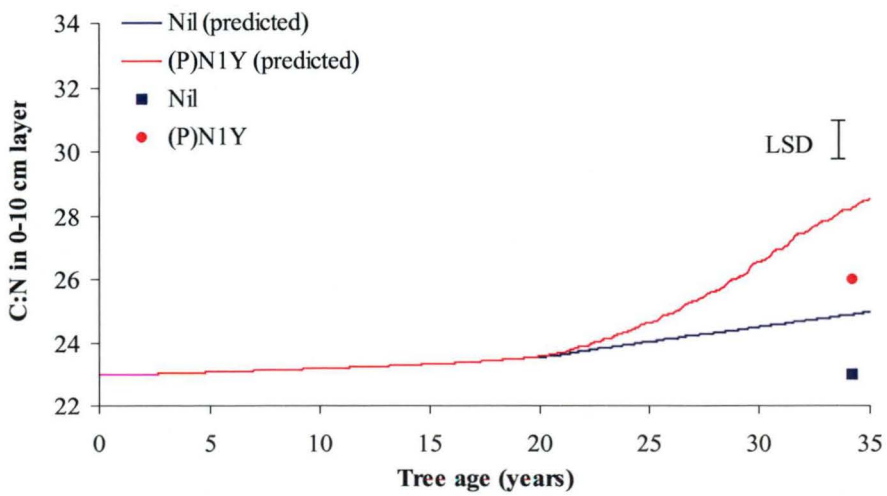


Figure 8.11 Observed and predicted C/N ratios in fertilised and unfertilised Kurosol topsoil (0-10 cm).

Increased C in the forest was predicted with total soil C mass alone increasing from 50 t ha^{-1} to 92 t ha^{-1} after fertilisation, while the above ground mass of trees doubled (120 to 287 t ha^{-1}). Increased predicted soil C resulted in C/N ratios increasing from 25 in unfertilised topsoils to 28 in fertilised topsoils (Figure 8.11).

8.4 Simulation of stand biomass allocation and N budget

In addition to comparisons between predicted and measured values, CABALA can be used to compare parameters not measured in this study. This is particularly useful when examining parameters that are labour intensive or expensive to measure, such as root structure and water movement. A partial N budget could be estimated using the data presented in Chapter 3 (Table 8.2). However, numerous values were not measured during this analysis and CABALA provided an opportunity to do a total N budget for each treatment and examine N retention within each stand (Table 8.3).

Table 8.2 Nitrogen budget (t ha^{-1}) calculated from data presented in Chapter 3, Table 3.8.

	Nil	(P)N1Y	Difference
Total Soil N (0-50 cm)	4.01	4.95	0.95
Total Stand N [a]	0.14	0.36	0.23
Total O2 N [b]	0.33	0.72	0.39
Total Site N	4.47	6.04	1.57
Total N Loss (in runoff and deep drainage) [c]	~	~	~
Partial site N Budget	**4.47**	**6.04**	**1.57**

[a] Total Stand N biomass of *P. radiata* trees was estimated using an equation derived from Neilsen and Lynch (1988) (Table 3.6).
[b] Total N in the Litter Layer (O1) was not measured.
[c] Total N Loss was not calculated.

Table 8.3 Predicted N budget (t ha^{-1}) for fertilised and unfertilised stands

	Nil	(P)N1Y	Difference
Total Soil N (0-50 cm)	3.93	4.31	0.38
Total Stand N	0.15	0.72	0.57
Total Litter plus Duff N	0.02	0.11	0.09
Total Site N	4.10	5.15	1.04
Total N Loss (in runoff and deep drainage)	0.05	0.11	0.06
Site N Budget	**4.15**	**5.26**	**1.10**

The predicted difference in N budgets between the fertilised and unfertilised sites was 1.1 t ha^{-1} of N, which is close to the total amount of N supplied during the thirteen annual applications (1.3 t ha^{-1}). The model simulates that a large proportion of applied N is retained within the stand at age 34 years, while the proportion of total N that was lost (2 % of total N budget) at the fertilised site was twice as large as the unfertilised site (1 %). Monthly N losses were predicted to be <1 kg N ha^{-1} $month^{-1}$ in the unfertilised Kurosol and averaged around 2 kg N ha^{-1} $month^{-1}$ after fertilisation (Figure 8.12). Assuming the normal loss of N in this forest is represented by the unfertilised rate (0.05 t N ha^{-1}, total at age 34 years) of the 1.3 t of N supplied, less than 5 % was lost from the stand. Nitrogen immobilisation increased four-fold in fertilised compared to the unfertilised Kurosol (Figure 8.13). Resulting in the amount of N immobilised throughout fertiliser application increasing from 181 kg N ha^{-1} to 637 kg N ha^{-1} (Table 8.4).

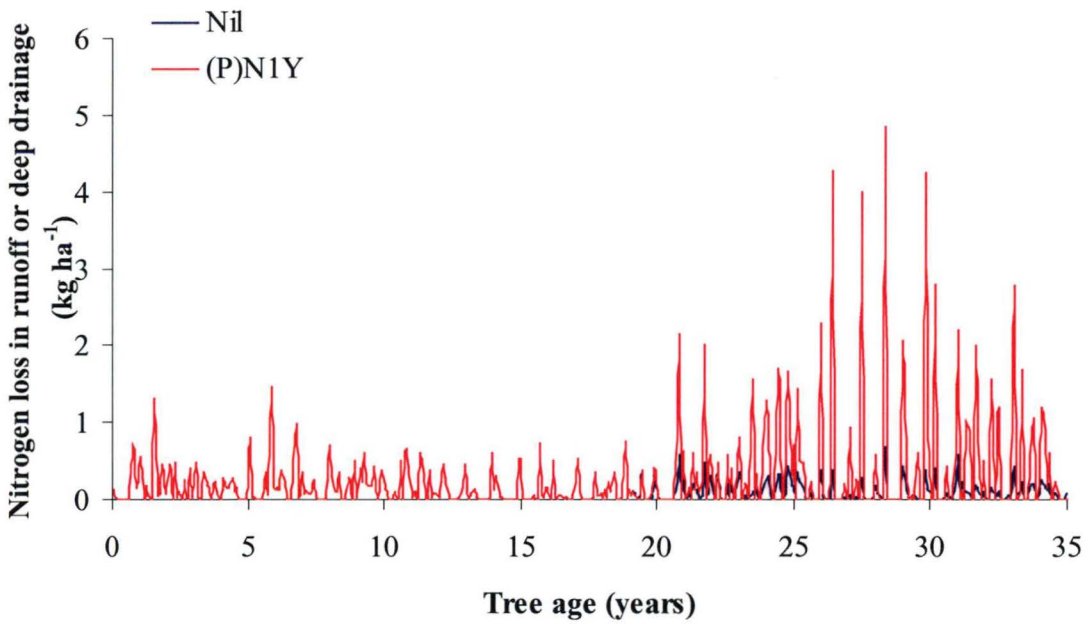


Figure 8.12 N loss in run off and or deep drainage (kg ha^{-1}) in unfertilised and fertilised Kurosol profiles.

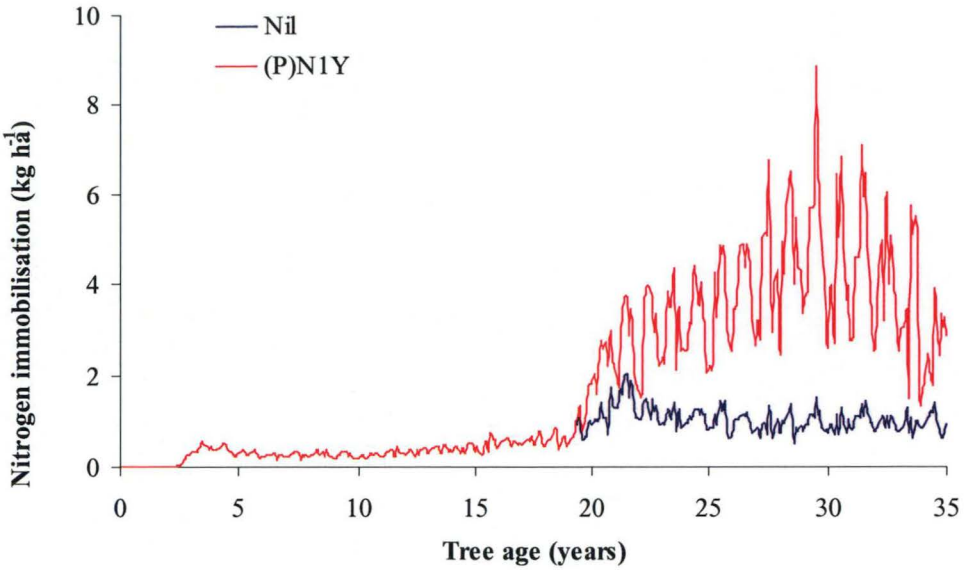


Figure 8.13 Nitrogen immobilisation (kg ha^{-1}) in unfertilised and fertilised Kurosol profiles.

Table 8.4 Total N mineralisation and immobilisation in the unfertilised and fertilised stand up to the age of 34 years.

N Mineralised	Nil	(P)N1Y	Difference
Litter and Duff	25.9	46.0	20.2
0 -10 cm	87.4	93.3	5.9
10 - 20 cm	-3.2	23.4	26.6
20 - 50 cm	-1.7	14.2	15.8
Total	108.4	176.9	68.5
N immobilised			
Total	180.8	637.2	456.4

Increased growth in fertilised stands resulted in a large reduction in water available within the profile (Figure 8.14). As a result, fertilised trees were predicted to be drought-stressed for a period of over 1000 days up to age 34 years, compared to only 30 days in unfertilised trees (Figure 8.15).

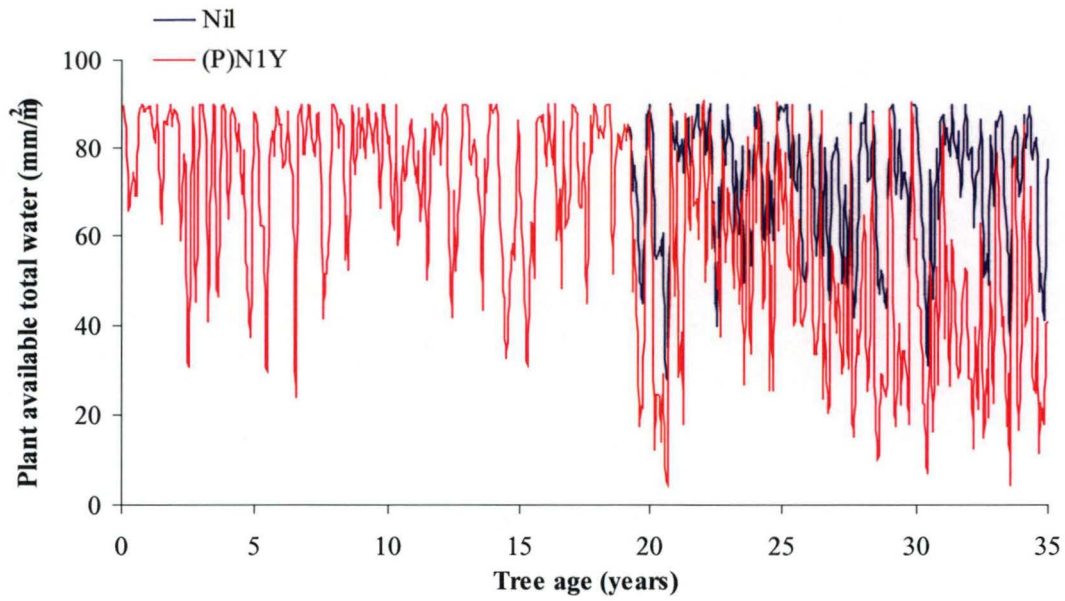


Figure 8.14 Plant available total water in the fertilised and unfertilised Kurosol profiles.

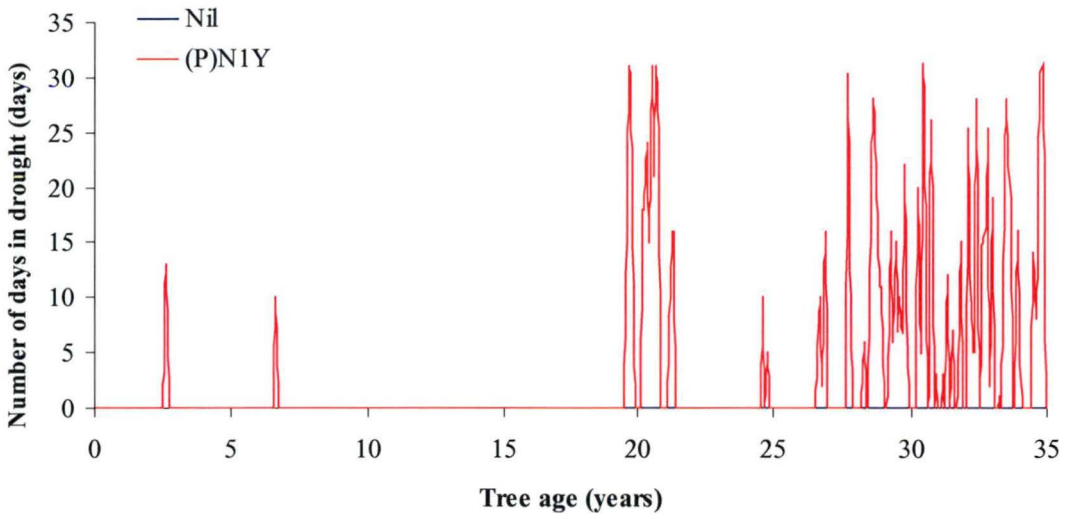


Figure 8.15 Number of days *P. radiata* trees were predicted to be in drought in fertilised and unfertilised Kurosol profiles.

8.5 Discussion

Predicted volume growth responses were generally well simulated. Particularly in the fertilised ((P)N1Y) treatment, where the slope of observed *vs* predicted was close to 1. The version of CABALA used in this study assumes that P was not limited. As previously indicated P applied at age 20 and 26 significantly increased growth. By the

cessation of fertilisation, incremental growth in the P fertilised trees was triple of those unfertilised. The projected growth of unfertilised trees was similar to that observed in (P) fertilised trees prior to the second application of P. Projected growth was also higher during this period for fertilised trees ((P)N1Y). Following the third N application predicted growth was 30 % higher than that observed, however by the cessation of fertilisation differences had declined to 15 %. Decreased observed responses to the initial applications of fertiliser may be associated with low P availability limiting root growth and therefore restricted N acquisition. Site and land use factors determine the overall ability for trees to compete for nutrients and in *P. radiata* plantations, Smethurst and Nambiar (1995) found trees were unable to take up N due to limited root development from weed competition. In this study fine root mass was predicted to increase 10-fold from 0.14 t ha^{-1} to 1.6 t ha^{-1} following fertilisation. However, decreased root vitality has been associated with ammonium sulphate application in Norway spruce (Persson *et al.*, 1995). The linking of CABALA with PCATS, a P uptake model, successfully predicted fertiliser responses in a *Eucalyptus nitens* plantation (Smethurst *et al.*, 2004b). Incorporation of the PCATS model might resolve suspected P limitations in the current simulations.

At the age sampled, mineral N concentrations were generally well simulated by CABALA. Predicted concentrations of NO_3^- directly after fertilisation were generally less than 1% of total mineral N, which was in agreement with low rates of nitrification measured in both field (Chapter 4 and 5) and laboratory (Chapters 6 and 7) incubations. At similar rates of fertilisation up of 1440 kg N ha^{-1} (from three annual applications), Nohrstedt (1990) also found that nitrate was generally below detection limits in soil water. Elevated mineral N concentrations after fertilisation were predicted to last six to twelve months, which was slightly longer than field measurements of four to five months. Following the cessation of fertilisation at age 32 years, predicted NH_4^+ concentration in fertilised topsoil declined to levels similar to those measured in the NIL treatment, which was in agreement with observations in the field at this age (Chapter 3). In subsoils (20 – 50 cm), a large long-term increase in the concentration of NH_4^+ due to fertilisation was predicted; such increases were not evident in soils measured at the end of the experiment (Chapter 4). Increased immobilisation immediately after fertilisation and increased annual mineralisation

(Chapter 4) suggests that at this site, ammonia was probably retained in surface soil and turned over relatively quickly, rather than being leached down the soil profile as predicted. Increases in subsoil mineral N concentrations following high rates of fertilisation are not uncommon, but the effect lasts for only a year or two unless applications are repeated (Khanna *et al.*, 1992; Smethurst *et al.*, 2001).

Simulated rates of NNM in the Kurosol topsoil were low and less than 10 kg N ha^{-1} was mineralised annually in 2000 (age 32 years). Both monthly and annual rates of NNM were only slightly higher in fertilised soils. In contrast, laboratory rates of NNM in fertilised soils were significantly higher at 49 kg N ha^{-1} yr^{-1} compared to 26 kg N ha^{-1} yr^{-1} when unfertilised (Chapter 6). Although not significant, *in situ* rates were also four-fold higher in (P) N1Y than NIL soils, at 12.6 and 51.7 kg N ha^{-1}, respectively (Chapter 4). In CERES, N mineralisation in the humus fraction is calculated as, RHMIN = NHUM (L) x DMINR x TF x MF x DMOD. Results from this study suggest that the decay rate constant (DMINR) increases with large N inputs and may need to be modified to reflect these changes. Further study would be required to determine how this function should be modified. Organic matter may be physically protected from mineralisation in the soil through adsorption to negatively charged clay surfaces or within micro-aggregate small pores (Tiessen and Stewart, 1983). In a study of grassland soils Tiessen and Stewart (1983) indicated that particle-size fractionation of soils yields organic matter fractions with distinctly different properties, which undergo different transformations during organic matter turnover. Strong *et al.* (1999a) also observed a significant negative relationship between N mineralisation and clay content. However, the clay content of the Kurosol topsoil (loamy sand) was 4 % clay and therefore was unlikely to limit NNM in these soils. These soil properties require further investigation for forest soils, which don't undergo constant cultivation as occurs in the agricultural soils for which the CERES model was developed.

This study also suggests that low P availability limited tree growth even after the initial application of P fertiliser and that it might have restricted the rate of N release from decomposition (which is controlled by the DMINR function in the N turnover sub-model CERES). There was evidence of higher rates of litter decomposition due to P additions at the *P. radiata* site, because the P2YN2Y treatment had significantly

less litter than the (P)N2Y treatment, i.e. 29 t ha^{-1} compared to 64 t ha^{-1}, respectively (Chapter 3). An increase in N uptake by enhancing soil N mineralisation has been observed in a range of forest soils, including *P. radiata,* following P application (Waring, 1969; Khanna *et al.*, 1992; Falkiner *et al.*, 1993). At this site, NNM rates measured in topsoils during laboratory incubations were linearly correlated with P concentrations. Carlyle *et al.* (1990) also observed a significant correlation between NNM and total P in the soil under *P. radiata*. However, potential P effects on N mineralisation through the rate of N release (DMINR) or protection of organic N (DMOD) are not taken into account in the N turnover predicted in the CERES submodel.

The protection of organic N, resulting in a DMOD value less than one may also be a function of the species studies. In a comparison under *P. radiata* and *E. regnans* plantations, Jurgensen *et al.* (1986) observed although C: N ratios in the litter were higher under the eucalypts ranging from 38 to56 compared to 23 to 29 under the pines, decomposition rates were clearly lower under *P. radiata*. Skene (1997) related this to the protection of inorganic matrices. Protection depended on the quality of the substrate, in high quality substrates (such as straw) physical protection was the limiting factor on decomposition, whereas chemical protection was the limiting factor for low quality substrates such as Eucalyptus litter. Reduced bacterial numbers have been observed under pines and between first and second rotation *P. radiata* (Theodorou and Bowen, 1981; Theodorou and Bowen, 1983b; Upadhyay and Singh, 1985). Further investigation is required to take into account the species dependent chemical protection of organic matter in the CERES submodel.

In subsoils predicted mineralisation rates also remained low, i.e. less than 4 kg N $ha^{-1}yr^{-1}$ during the last ten years of simulation, which is consistent with subsoil microbial populations having a severely limited ability to conduct NNM (Page *et al.*, 2003).

The mineralisation of N in litter was determined as an important N source at both sites (Chapter 9) and can be expected to ultimately affect site productivity (Nadelhoffer *et al.*, 1982). Predicted litter amounts were slightly lower at 29 and 83 t ha^{-1} for unfertilised and fertilised treatments at age 34 years than those measured in the field at 56 and 92 t ha^{-1}, respectively (Chapter 3). Observed litter masses were in the range

expected in temperate forests, which can accumulate to between 10 and 100 t ha^{-1} (Fisher and Binkley, 2000). In a previous study, Neilsen and Lynch (1998) observed a litter mass of almost 50 t ha^{-1} under a 34-year-old *P. radiata* plantation that had been fertilised with 429 kg N ha^{-1}, while unfertilised trees had an underlying litter mass of almost 40 t ha^{-1}. These results generally agree with the current study and suggest that large amounts of litter accumulate in *P. radiata* plantations.

Annual predicted N mineralisation in litter and duff was less than 5 kg N ha^{-1} yr^{-1} in both treatments. Although these rates are similar to NNM rates measured in the unfertilised O2 horizon during laboratory incubations (up to 4 kg ha^{-1} yr^{-1}), they contrast to those measured in the fertilised O2 horizon, which had NNM rates up to 106 kg ha^{-1} yr^{-1} (Chapter 6). Hence, laboratory incubations in this study suggested that fertilised litter was capable of mineralising up to 20 times more quickly than that unfertilised. However, this high rate of NNM also suggests that, with an N concentration of 1 %, over 10 t ha^{-1} of litter was decomposed each year, which contrasts to the large litter mass measured on site. Predicted litter fall inputs (including leaves, branches, bark and stemwood) at age 34 years in fertilised and unfertilised sites, were a total 61 t ha^{-1} and 120 t ha^{-1}, respectively, suggesting low rates of decomposition overall. Annual litter fall after thinning was around 1 t $ha^{-1}yr^{-1}$ when unfertilised and between 1 and 10 t $ha^{-1}yr^{-1}$ when fertilised. This result indicates that laboratory rates on NNM in fertilised litters would result in the decomposition of more litter than that which is actually available. The laboratory temperatures used (10 and 20 °C), were well within the expected field range and not expected to greatly overpredict NNM at this site. However, mineralisation would have been increased due to the mechanical disturbance that occurred during laboratory preparation. Therefore, and as others have found, rates of NNM measured in litter during laboratory incubations could not be assumed to be occurring in the field. Theodorou and Bowen (1990) noted that even though fertilisation (806 kg N ha^{-1} respectively) increased N return in by more than 100 % annually in 12-year-old *P. radiata*, forest floor N content, and rates of decomposition and NNM were similar to those unfertilised. Also in agreement with simulated data, Piatek and Allen (2001) suggested under loblolly pine the decomposing litter (O2) horizon contributed negligible N during the growing season and in fact could be viewed as a N sink. Aber and Melillo (1980) reviewed

many studies demonstrating fresh litter is a mineral N sink, rather than a source. In addition, Smethurst and Nambiar (1990b) observed low rates of NNM of 8.1 kg N ha^{-1} yr^{-1} in the litter of a 37 year-old *P. radiata* plantation in south-eastern Australia. Because rates of NNM in litter could not be reliably extrapolated from the laboratory to the field, the current study would have benefited from *in situ* measurements of litter turnover.

Observed LAI was lower than the predicted value at 4.8 compared to 7, at age 35 years. Higher predicted LAI in fertilised trees may be due to the assumption of adequate P during the experiment. However, even with fertiliser, the P concentration in foliage remained marginal throughout the experiment, i.e. between 0.08 and 0.10 % (Chapter 3). In contrast, observed and predicted values for unfertilised trees were similar at 1.3 and 1.5, respectively. The extremely low LAI in unfertilised trees was associated with the high mortality and a high proportion of dead tops. The percentage of dead tops in unfertilised trees was over 70 % by age 26 years, and this number continued to increase with increasing age (Chapter 3). Thinning and dead tops would have also affected the measurement of LAI in these plantations (Cherry *et al.*, 1998) and might have contributed to an underestimation of LAI.

The model simulated that a large proportion of applied N was retained within the stand at age 34 years, of the 1.3 t of N supplied, only 5 % was lost from the stand. These values are extremely low compared to other field studies, where N losses are in the range 20-50% (Nohrstedt, 1990; Raison *et al.*, 1990; Neilsen and Lynch, 1998; Fisher and Binkley, 2000; Smethurst *et al.*, 2001). One reason for limited loss of applied N could be the predicted increase in immobilisation, which was often four times higher in fertilised than unfertilised Kurosol soil. Significant increases in immobilisation of N were observed for the first six months after fertilisation (Chapter 4). Another reason could be that LAI was over-predicted in fertilised trees and leading to a higher than actual accumulation of N above-ground. A similar problem was encountered by Smethurst *et al.* (2004) for *E. nitens* simulations, i.e. canopy N content was over-predicted. In both cases, if less N had been simulated to accumulate above-ground, it is likely that more N would have been lost by leaching.

Pinus radiata plantations grown in summer drought areas often have growth limited by water availability (Crane and Banks, 1992; Snowdon and Benson, 1992). In a 10-year-old plantation of *P. radiata*, Crane and Banks (1992) observed that irrigation increased growth by 25 % above fertiliser applied alone. Simulation in this study also suggested that *P. radiata* growth could be substantially boosted if fertilisation was combined with irrigation.

8.6 Conclusions

After tuning the model for only the degree of N limitation, there was broad quantitative agreement between simulated and observed data of several key variables in the unfertilised and fertilised treatments. These simulations were for a period of 36 years of tree growth, and included one thinning. Stem volume growth, nitrate and ammonium concentrations, LAI, NNM, litter mass, and the overall N budget were adequately simulated, which is a very pleasing result considering the difficulties often encountered in such long-term, complex simulations. However, simulations of the fertilised treatment over-predicted ammonium accumulation in the 20-50 cm depth, and the LAI and N content of the canopy. Litter mass of the unfertilised treatment was under-predicted. Further improvements to the CABALA model should therefore consider the way these parameters are calculated, and particularly the sensitivity of decomposition and N mineralisation to fertiliser inputs.

A partial N budget based on field measurements indicated that N losses due to leaching were low. Simulations using CABALA were consistent with this result, with losses at less than 5 % of N applied, but this N flux may have been under-estimated due to a simulated over-accumulation of N in the canopy.

This model also provided an opportunity to compare the water status of fertilised and unfertilised stands, which suggested that, even though rainfall at this site is considered high by Australian standards, further growth increases would probably require a combination of irrigation and fertilisation.

Chapter 9. General Discussion and Conclusions

9.1 Introduction

Increased demand for plantation wood production, on a base of decreasing land availability, has resulted in the progression of plantations onto soils of reasonable physical structure but low nutrient status. This trend is combined with increasing demands for faster growth and greater product utilisation from forest sites, intensifying nutrient demand and removal. Many Australian soils require N and P fertilisers at planting and during the early stages of tree establishment to achieve rapid early growth and high survival rates (Gentle *et al.*, 1965; Judd *et al.*, 1996; Neilsen, 1996). Results from this study support the hypothesis that long-term fertilisation of *E. regnans* and *P. radiata* plantations significantly increase stem volume and change the distribution of nutrients in these systems. In both plantations, fertilisation at the highest rate (totalling 1300 kg N ha^{-1}) resulted in a significant increase in growth, doubling wood volume in both *P. radiata* and *E. regnans* plantations.

Long-term fertilisation resulted also resulted in the accumulation of nutrients in the mass of the lower litter (O2 horizon), which formed a substantial nutrient and carbon pool. Under *E. regnans*, at the highest rate of fertilisation, the O2 horizon contained over seven times the N content of the NIL treatment, equivalent to about 60 % of the N applied. Phosphorus, S and Ca also accumulated within the O2 horizon, showing the importance of this layer as a nutrient reserve. However, changes in the rate of N cycling, as indicated by *in situ* NNM, were not significantly effected by fertilisation in either the Kurosol or Ferrosol topsoils. At both sites, *in situ* net N mineralisation (NNM) rates were low, ranging between 13 and 52 ha^{-1} yr^{-1}.

During laboratory incubations, NNM rates in both Kurosol and Ferrosol topsoils could be significantly increased due to fertilisation, however, this depends on the temperature and particularly the moisture content of the soil during incubations. Changes in moisture content and the incubation temperature altered NNM rates in topsoils by up to five-hundred-fold. In contrast, NNM rates on a unit per mass basis, were up to 50 times higher in the O2 horizon than those in the topsoil and at both sites fertilisation significantly increased NNM.

9.2 Plantation growth health and foliar nutrient concentrations

Limitations of plantation growth due to low N availability have been frequently reported in the literature (Cromer *et al.*, 1975; Menge *et al.*, 1977; Neilsen *et al.*, 1984; Schönau and Herbert, 1989; Raison *et al.*, 1990; Raison and Connell, 1992). Low rates of N mineralisation in cool temperate forests (Ellis, 1974; Adams *et al.*, 1989a; Foster, 1989; Polglase *et al.*, 1992b) means that high growth rates can only be achieved with applications of fertilisers (Waring, 1969; Hunter and Hoy, 1983; Raison and Connell, 1992; Neilsen and Lynch, 1998; Jacobson and Pettersson, 2001). In this study, substantial responses to N fertiliser, applied as ammonium sulphate, were observed in both plantations. Volume growth of both stands doubled with annual applications of N fertiliser at 100 kg N ha^{-1} yr^{-1}. Although applications every second and fourth year produced less response, these treatments had better fertiliser-use efficiency. This is in agreement with large volume increase in *P. radiata* due to annual and periodic N application observed in a number of other studies (Hunter and Hoy, 1983; Raison *et al.*, 1990; Fife and Nambiar, 1997; Neilsen and Lynch, 1998).

Two applications of P fertiliser, as superphosphate, totalling 144 kg ha^{-1} produced substantial increases in *P. radiata* growth. In contrast, application of N and P every second year together to *E. regnans* resulted in no difference from N applied alone. Responses of eucalypt plantations to later age N and P addition are variable and site specific (Cromer *et al.*, 1981; Weston *et al.*, 1991). Ward *et al.* (1985) only found responses to N and P in combination, which showed a positive interaction on height growth, but not to P applied alone. However, single applications of P fertiliser have produced continued responses for many years in a number of forest crops (Ballard, 1978; Flinn *et al.*, 1979b; Turner and Lambert, 1986; Turner *et al.*, 2002).

Foliar nutrient levels are good indicators of tree health (Woolons and Will, 1975; Dell *et al.*, 2002). However there are limitations of foliage analysis in predicting growth responses. In an intensive study of 12 year old *P. radiata* effected by N and water supply, Benson *et al.*(1992b) observed that there was little similarity between patterns of stem growth and that of N concentration in foliage. In Eucalyptus species it has also been recognised that it is difficult to correlate growth and foliar nutrient concentrations beyond two years of age (Judd *et al.*, 1996).

Foliar nutrient levels are effected by many factors including seasonal differences, soil moisture availability and time since fertilising (Nason *et al.*, 1990). In both species in this study, foliar nutrient levels varied between years, independent of fertiliser application. The periodic foliage sampling within this trial made it difficult to assess pulses in nutrients from fertiliser application and uptake and subsequent re-translocation within the tree. In *P. radiata,* deficiency symptoms were associated with low nutrient levels and P levels reflected the increase in P availability when P was applied. Phosphorus application alone resulted in dead tops recovering, but other symptoms were only relieved when N was also added. Enhanced overall stand health and needle biomass observations have been common after N application to *P. radiata* previously showing severe nutrient deficiencies (Hunter and Hoy, 1983; Neilsen *et al.*, 1984; Neilsen *et al.*, 1992). Even though rainfall at this site was considered high by Australian standards, further enhancement of stand health and growth would probably require a combination of irrigation and fertilisation. CABALA predicted that fertilised trees were drought-stressed for a period of over 1000 days up to age 34 years, compared to only 30 days in unfertilised trees (Chapter 8).

In contrast to the *P. radiata* plantation, accumulation of P within the *E. regnans* foliage suggested P was not the limiting growth at this site. When limited by other factors trees have been capable of accumulating P, as inorganic P, beyond the immediate tree requirement (Bennett *et al.*, 1997). Evidence of such responses was also observed in the accumulation of P up to 300 percent in young *E. globulus* plantations by Hooda and Weston (1999).

9.3 Changes in nutrient distribution and soil acidity

Various authors have addressed the effect of repeated fertiliser applications on growth and sustainability issues (Nohrstedt, 1990; Tamm and Popovic, 1995; Nohrstedt *et al.*, 2000). In this study, significant reductions in soil pH, of about 1 unit, were associated with the highest rates of fertilising and reductions in pH occurred after the application of both nitrogenous and phosphatic fertilisers. Fertiliser treatments significantly reduced pH by up to 0.6 of a unit in the 0 – 50 cm soil depth at both sites and by 0.3

and 0.6 of a unit in the *P. radiata* and *E. regnans* litter. In the Ferrosol the greatest reduction was at the surface, while the greatest reduction occurred at 20 to 50 cm in the Kurosol.

High rates of N application significantly reduced exchangeable Mg by half to the depth of 50 cm in the Kurosol, and by over one third in the top 10 cm of the Ferrosol. Leaching of Mg from surface soils due to fertilisation with ammonium sulphate have been observed (Khanna *et al.*, 1992). Combined with the potentially large losses of nutrients in the order of 221 kg Ca ha^{-1}, 285 kg K ha^{-1} and 66 Mg ha^{-1} from the removal of *P. radiata* sawlogs (Webber and Madgwick, 1983), it indicates a high risk of base cation decline in production forests. Base cation losses due to acid deposition and harvesting have been linked to declines in forest growth and health (Watmough and Dillon, 2003). Although leaching per say was not measured at this site, a partial N budget based on field measurements and simulated data using CABALA indicated that N losses due to leaching were low. Less than 5 % of the N applied was predicted to be lost from the site. In addition, both observed and predicted concentrations of nitrate were low, generally less than 1 % of the total mineral N.

9.4 Nitrogen Fertilisation effects on N availability and NNM

9.4.1. Soil

Field measurements of NNM were assessed using *in situ* cores, both open and covered from rainfall, as reviewed by Raison *et al.* (1987), Adams *et al.* (1989b) and Smethurst and Nambiar (1989b). At both sites, rates of NNM were low, ranging between 13 and 52 kg N ha^{-1} yr^{-1}, and were not significantly effected by long-term fertilisation. These rates are within the range measured in eucalyptus forests in Tasmania, between 13 and 188 kg N ha^{-1} (Adams and Attiwill, 1988; Wang *et al.*, 1998; Moroni *et al.*, 2002) and *P. radiata* plantations of southern Australia, between 16 and 74 kg N ha^{-1} (Carlyle *et al.*, 1998a).

In the Ferrosol topsoil, long-term fertilisation had no significant effect on annual NNM (both below 25 kg N ha^{-1} yr^{-1}), N leaching or N uptake. This is not surprising as the total N content was actually significantly higher in the unfertilised (NIL) than the

annually fertilised (P1YN1Y) treatment. Long-term N fertilisation also had no statistically significant effect on total N, NNM, N uptake or N leaching, in the Kurosol topsoil. However, all three measurements (NNM, N uptake and N leaching) had average annual rates at least four-fold greater in the annually fertilised ((P)N1Y) topsoil than the NIL. Average annual NNM increased from 13 kg N ha^{-1} yr^{-1} to 52 kg N $ha^{-1}yr^{-1}$ in unfertilised and annually fertilised ((P)N1Y) topsoil, respectively ($p=0.3$). In comparison, Raison *et al.* (1992) observed significant increases of two to three-fold four years after fertilisation in the Biology Growth Experiment. Such results highlight that the interpretation of the long-term effects of fertilisation on NNM in this experiment was constrained by the low treatment replication at the sites studied.

To reduce variability, and examine whether recent climate variability temporally restricted field rates of NNM, rates of NNM were measured in the laboratory (Chapter 6). Under controlled temperatures and moisture long-term fertilisation significantly increased NNM in the Kurosol topsoil, from 4 kg N ha^{-1} yr^{-1} to 60 kg N ha^{-1} yr^{-1} (incubated at field capacity moisture and 16 °C). Ferrosol topsoils also had significantly higher NNM due to fertilisation but only when incubated at a water content close to field capacity and in this soil long-term fertilisation had no significant effect overall. Although this experiment demonstrated that under controlled laboratory incubations, significant differences in NNM due to fertilisation could be observed, it also demonstrated that this occurred only under particular conditions. In addition, as observed with *in situ* measurements, rates of NNM in the laboratory still varied considerably between plot replicates of fertiliser treatments.

To reduce site variability further, soils from replicate plots of each treatment were collected again and homogenised (pooling) (Chapters 7). During this second laboratory study, although rates of NNM in the fertilised Kurosol were higher than the unfertilised, fertilised every second ((P)N2Y) or every fourth year ((P)N4Y) had no significant effect. In contrast, Ferrosol topsoil fertilised every second (P2YN2Y) and fourth year (P4YN4Y), immobilised significantly more than the unfertilised and annual fertilisation (P1YN1Y) treatments. The efficiency of conversion (the rate of NNM compared to the amount of total N available, NNM/total N) in both topsoils

was low compared to agricultural soils (Tabatabai and Al-Khafaji, 1980), at less than four percent and two percent per annum in the Kurosol and Ferrosol, respectively. All three studies indicated that the soils studied in this experiment required large changes in N inputs to occur before significant changes in soil NNM rates could be measured. This may also indicate that the methods used were not sensitive enough to measure change at such low rates of NNM.

Low and often negative rates of NNM during 60 days may have indicated that this period of incubation might not have been enough to overcome the effects of disturbance at intermediate rates of N fertilisation. However, 60 days was required to produce a significant cumulative effect, resulting from a divergence in NNM rates between the fertiliser treatments during the later stage of the incubation. Results from this study also indicate that longer periods of incubation may underestimate NNM due to depletion in available organic N for mineralisation or a build up of waste products in the small samples used in laboratory incubations. In both litter and topsoil horizons, the effect of laboratory method on NNM could mask smaller changes due to changing N inputs. As a result when small, < 10 $\mu g\ g^{-1}$, changes in NNM occurred due to N inputs, these become swamped by large changes that occurred due to, air-drying, water availability, temperature, the time of sampling and the physical disturbance of the samples during preparation. As cool temperate mature eucalypt forests had NNM rates <10 $ha^{-1}\ yr^{-1}$ (Adams *et al.*, 1989a), the use of some laboratory methods, for example pre-drying samples to predict field NNM rates in soils is questionable.

9.4.2 Litter

Fertilisation significantly increased NNM in the litter layer (O2) of both sites and corresponded to large increases in total N mass (Chapter 6 and 7). Increases in litter N mass and NNM were not always observed in underlying soil, indicating that the litter layer may represent a clearer picture of long-term changes in N cycling at a site, than the soil. Previous investigations have also noted large increases in litter fall N, following N fertilisation (Hunter and Hoy, 1983; Nohrstedt, 1990; Fife and Nambiar, 1997; Neilsen and Lynch, 1998), but few investigations noted significant changes in soil. Both *E. regnans* and *P. radiata* litter had NNM rates ten times greater than that measured in topsoil. Annual rates of NNM were up to 106 kg $ha^{-1}\ yr^{-1}$ and 320 kg ha^{-1}

yr^{-1}, in fertilised *P. radiata* litter and *E. regnans* litter, respectively. The increased N cycling in litter due to fertilisation highlights the importance of estimating N inputs in litter horizons, to avoid underestimation of the overall site productivity. Hendrickson and Robinson (1984) also observed rates of NNM were higher in litter than mineral soils and declined, moving downward through the profile. However, previous studies have indicated that the litter layer is a N sink rather than a source (Aber and Melillo, 1980). For Example, Piatek and Allen (2001) observed that *in situ* NNM rates in the decomposing litter (O2) horizon under loblolly pine were low and often negative and suggested this layer contributed negligible N during the growing season. CABALA also predicted low rates of N mineralisation in litter and O2 horizon at less than 5 kg N ha^{-1} yr^{-1} in both treatments (Chapter 8).

Annual litter fall after thinning was predicted at around 1 t $ha^{-1}yr^{-1}$ when unfertilised and between 1 and 10 t $ha^{-1}yr^{-1}$ when fertilised. With a N concentration of around 1 % in both litter layers, the high rates of litter decomposition measured in the first laboratory study would suggest that over 10 t ha^{-1} of litter was decomposed each year, which contrasts to the large litter mass measured on site. Rates of mineralisation in the second laboratory study at 10 °C are considered to reflect field rates of NNM more realistically. These rates also concur with those measured in a 37 year-old *P. radiata* plantation in south-eastern Australia by Smethurst and Nambiar (1990b).

There were large differences in the efficiency of N conversion between the litter horizons at both sites. Annually fertilised *E. regnans* litter (O2) had higher N concentration (1.2%) and mineralised nine times more N than *P. radiata* litter (N, 1.0%). In contrast, even though N concentrations from unfertilised litter from both sites were under 0.9 % (Chapter 3), annual rates of NNM were ten-fold higher in *P. radiata* than *E. regnans* litter. In addition, the litter layer under *P. radiata* generally had twice the mass of N, P, S, Ca and Mg of that present under the *E. regnans* plantation. Jurgensen *et al.* (1986) also observed these differences in *E. regnans* and *P. radiata* plantations as young as four-years-old. Higher nutrient contents in the *P. radiata* litter compared to the *E. regnans* litter contrasts to the hypothesis proposed by Lamb (1976), that the poor-quality litter inputs characteristic of *P. radiata* plantation

result in site degradation due to the cyclic effect of slow litter degradation and low N mineralisation.

9.5 Moisture and temperature effects on NNM during field and laboratory incubations

The effect of moisture and temperature on NNM and N availability was strongly influenced by the site studied. Overall, the influence of incubation temperature and moisture on NNM was limited in topsoil and litter of the drier lower fertility site under *P. radiata*, both in the field and in the laboratory. This contrasted to the significant effects of both moisture and temperature on NNM in the topsoil and litter from the wetter more fertile, *E. regnans* site.

Moisture content had a significant effect on *in situ* NNM in the Ferrosol topsoil, where rainfall restriction (in caped cores used to estimate N leaching) could reduce rates of NNM by up to 200 % (Chapter 4). In the laboratory the effect of moisture was also significant and in particular the procedure of air-drying samples prior to incubation. This occurs in many laboratories (Richards *et al.*, 1985; Bonde and Rosswall, 1987; Robertson *et al.*, 1988; Ross *et al.*, 1995) and the data presented here show this could change N mineralisation from negative (immobilisation) to NNM rates above 100 kg N ha^{-1} yr^{-1}. In contrast, air-drying and re-wetting *E. regnans* litter decreased NNM significantly.

The limited response of NNM rates in the Kurosol topsoil to moisture changes was consistent with the idea that local soil biota may have adapted to extreme soil water fluctuations (Birch, 1958; West *et al.*, 1988; Van Gestel *et al.*, 1993). In comparison to the Ferrosol site, the Kurosol topsoil experienced large moisture fluctuations in the field this may lead to micro-organisms capably of facilitating NMN over a wide moisture range. Like the mineral Kurosol topsoil, NNM rates in the *P. radiata* litter (O2) were generally unaffected by moisture, even after air-drying and re-wetting.

Site characteristics such as clay and organic matter content of the soil also determine the relative disruption of the physical structure of the soil during moisture changes and subsequently the rate of newly available organic matter release (MacKay and

Carefoot, 1981; Van Gestel *et al.*, 1991; Strong *et al.*, 1999a). The soils studied in this experiment had almost a ten-fold difference in clay content, with 4 % and 50 % in the Kurosol and Ferrosol topsoils, respectively. This would have contributed significantly to differences in NNM due to disturbance effects. Cabrera and Kissel (1988a) noted the higher the clay to total N ratio, the larger the physical disruption due to disturbance and the higher the over prediction of NNM.

The overall effect of temperature in field experiments at both sites was limited, and often dependent on moisture availability (Chapter 4). Seasonal trends were not significantly difficult during the time frame of the study, but both sites had a six-fold increase in rates of NNM, from the lowest measured in May and the highest in March, which corresponded to a period of high temperatures and rainfall. Adams and Attiwill (1986) also observed that the highest rates of NNM occurred in summer when both high temperature and moisture levels were present, while rates of other seasons were generally similar.

In the laboratory, the effect of temperature on NNM was also moisture dependent. However, some temperature effects were evident in the laboratory in both experiments (Chapters 6 and 7). In both laboratory experiments, Kurosol topsoils mineralised more N at the higher temperatures (between 16 and 22 °C, compared to 3 to 10 °C) and there was a slight increase in the effects of temperature on NNM in long-term fertilised topsoil. In contrast, temperature effects on NNM in Ferrosol topsoil were different between the two laboratory experiments. In the first study, when positive NNM occurred, rates generally increased with increasing temperature and this was more pronounced in annually fertilised than unfertilised topsoil. In the second study when soils were homogenised, increasing the temperature suppressed net mineralisation in all except the annually fertilised treatment.

Temperature also generally increased NNM in *E. regnans* litter in both laboratory experiments and effects were more pronounced in annually fertilised treatments. For example, in the first laboratory study, decreasing the temperature from 16 to 10 degrees in the *P. radiata* litter decreased NNM by just under 10 $\mu g\ g^{-1}$. However, as the unfertilised litter had an initial N concentration of 4 $\mu g\ g^{-1}$ only, and the annually

fertilised initial N concentration was 150 μg g^{-1}, reducing the temperature had little effect on NNM in the fertilised but resulted in immobilisation in the unfertilised litter. Net N mineralisation also declined when the temperature was reduced in the second laboratory experiment, but all rates remained positive.

The time of sampling was found to be critical when measuring laboratory rates in NNM at both sites and horizons. Polglase *et al.* (1992b) also noted that considerable variation in *in situ* NNM rates occurred between years. Although both litter and topsoil for each laboratory study were taken at the same time of year (July), they were taken one year apart (Chapter 6 compared to Chapters 7). When comparing the relative parameters for each horizon, the second laboratory study generally resulted in NNM rates lower than the first. These reductions could be large, particularly at 10 °C. For example, NNM in unfertilised *E. regnans* litter was 63.5 μg g^{-1} in the first laboratory study but only 2.2 μg g^{-1} in the second. Although samples from replicate plots were homogenised in the second study, a thirty-fold reduction was not explained by physical disturbance, as all samples were sieved prior to incubation in both laboratory experiments. It was also noted that reductions in NNM were not consistent across all samples, as ten-fold increases in NNM were also measured.

Differences in rates of N mineralisation between the laboratory studies may have been associated with changes in substrate quality, that is, variations in litter fall, fine root turnover and changes in soil organic matter and microbial biomass pools related to climatic conditions in the interval between sampling. The delay of one year between sampling for each laboratory experiment was not expected to produce the changes in NNM measured between the experiments. However, Connell *et al.* (1995) demonstrated that pools of mineralisable N in forest soils can show significant temporal variation and observed dramatic changes in rates of laboratory NNM over a four year period (11.5 to 3.1 kg N ha^{-1} 30 $days^{-1}$). Raison *et al.* (1990) also observed a decline in NNM from 38 to 7 kg N ha^{-1} yr^{-1} (0-40cm , A1, Yellow podzol) over four years, due to a gradual decline in soil organic N supplies.

The marked temperature effect on NNM rates in the Ferrosol topsoil, i.e. at 10 °C the unfertilised topsoil mineralised more, while at 20 °C the annually fertilised topsoil mineralised more, has important implications when using laboratory measurements to predict field rates of NNM.

There are a number of different models available that can be used to predict N mineralisation in forest soils (Ellert and Bettany, 1988; Goodwin and Jones, 1991; Goncalves and Carlyle, 1994; Mary *et al.*, 1999). Many models require a large number of input values, which although they may be good research tools, limit their use for forest managers. One model is SNAP, Soil N Availability Predictor, which is an empirical model developed to predict N mineralisation across a wide range of sites (Paul *et al.*, 2002). Using sub-models to predict the soil moisture and temperature changes in the field, the model predicts field N mineralisation from N mineralisation basal rates (k) determined during aerobic laboratory incubations. Inputs required include site data, daily climatic data, LAI, estimated depth of water, litter mass and height, and the proportion of soil surface covered by weed, canopy and litter. This model was used in Chapter 4 to predict temperatures in the Ferrosol topsoil.

Sensitivity analysis of the SNAP model (Paul *et al.*, 2002) showed that the model was more sensitive to soil temperature changes than relative water contents (RWC). This was because N mineralisation rapidly increased with water contents only at RWC between 0.2 and 0.6, whereas mineralisation was found to increase exponentially with increasing temperatures between 0-40 °C. In the current study, such trends were not observed, as temperature did not consistently result in an exponential increase in N mineralisation rates within the laboratory. In addition, water was often found to have a larger inflence than temperature on Ferrosol topsoil NNM rates in the field and laboratory (Chapters 4 and 6).

Preliminary observations in SNAP also indicate that there was no effect of disturbance such as sieving on the calculation of k, regardless of soil type (Paul *et al.*, 2002). The current study indicated that disturbances such as sieving and drying could have a large effect on overall mineralisation rates depending on the soil type. In addition, in the current study the litter layer could be a significant source of mineral N, which is not calculated in SNAP. Using topsoil only to predicted increased NNM

at this site could result in a substantial under prediction of volume growth increases due to fertilisation. In addition, given that the rate of NNM in the field from the Ferrosol topsoil was less than 23 kg N ha^{-1}, the error in predicting annual NNM rates using SNAP model of 23 kg N ha^{-1} (due to variations in forest management, soil and climate types) was relatively high.

Reduced rates of NNM and increased N immobilisation in the Ferrosol soil in the second laboratory study compared to the first also highlight the difficulty of interpreting data from higher fertility, well structured, topsoil when the structure is highly modified prior to incubation. Even though relatively long periods of time (two weeks) were used to allow soils to equilibrate, in an attempt to reduce the effects of soil homogenisation, a significant impact of disturbance on NNM still occurred. It was therefore concluded from this study that the litter (O2) horizon provided a better indication of the N cycling occurring in temperate Tasmanian forest. In contrast, the topsoil was less effected by long-term fertilisation and more effected by incubation parameters. This was a particular problem when soil had higher fertility and good physical structure, such as the Ferrosol topsoil studied in these experiments. Generally, rates of NNM in topsoil during laboratory incubations varied greatly, suggesting that with the methods used to measure NNM currently, produced non-repeatable results. The source of variation includes the time of sampling and the laboratory parameters used. Incubation of intact soil-cores has been suggested to better reflect field conditions due to limited disturbance effects, (Adams and Attiwill, 1986; Raison *et al.*, 1987). However, previous results in this study indicated that an extremely high number of intact soil-core replicates would be required.

9.6 Implications of long-term fertilisation for site productivity, soil conditions and management

The amount of nutrients removed during harvesting depends on harvesting practises and rotation length. Nutrient removal in wood is much higher in short rotation plantations due to the high percentage of sapwood relative to heartwood in harvested logs (Birk and Turner, 1992). Large proportions of N are removed in wood, for example from the above ground biomass of 29-year-old *P. radiata* plantation tree,

approximately one third of the N content, occurred within the stem, 434 kg N ha^{-1}, and around one quarter was in the needles, 117 kg N ha^{-1} (Switzer and Nelson, 1972). In the current study, over half of the N applied, as fertiliser, could be retained in the litter horizon alone. Management and harvesting techniques are therefore key regulators affecting long-term site N availability (Switzer and Nelson, 1972; Keeney, 1980; Webber and Madgwick, 1983). Of particular importance is the amount of fine material that is moved off-site in the form of branches, twigs, leaves, litter and critically topsoil (Smethurst and Nambiar, 1990a; Smethurst and Nambiar, 1990b; Smith *et al.*, 1994). As a result, it is well recognised in Australia that slash and litter removal during intensive site preparation could decrease long-term productivity in plantations (Nzila *et al.*, 2002).

Following canopy development, internal redistribution and nutrient return from decomposition become critical processes in supplying nutrients for new growth (Weston, 2001). The litter layer formed during forest development plays a major role in mineral cycling and retention in forest ecosystems, and is strongly linked to the cycling of N, supply of base cations and nutrient buffering in the rhizosphere (Tamm and Popovic, 1995; Neilsen and Lynch, 1998). The release of N from litter is a basic process in the cycling of N in forests. Virtually all the N taken up by plants is inorganic and the continual replenishment of this pool requires that inorganic N be released from litter during decomposition (Carlyle, 1986).

As evident in this study and previous research, fertilisation results in a significant increase in the litter total biomass and N mass (Hunter and Hoy, 1983; Nohrstedt, 1990; Theodorou and Bowen, 1990; Fife and Nambiar, 1997; Neilsen and Lynch, 1998; Maier and Kress, 2000). At both sites mass of litter O1 horizons increased slightly, while O2 horizons increased significantly due to large applications of N fertiliser, increasing two-fold under *P. radiata* and seven-fold under *E. regnans*. Although concentrations of N were not significantly different between fertilised and unfertilised O2 horizons, the significant increases in litter mass resulted in large proportions of N being held in the litter system. The heaviest fertiliser treatments resulted in a significant increase in the mass of the O2 horizon in both the *P. radiata*

stand and the *E. regnans* stand with over 70 t ha^{-1} for the annual fertilised treatment at both sites compared to 40 t ha^{-1} for NIL in the *P. radiata* stand and 13 t ha^{-1} for NIL in the *E. regnans* stand. At the *E. regnans* site the highest rate of fertilisation resulted in significant increases in N, P, S and Ca within the O2 horizon, with over 60 percent of the N fertiliser applied was accounted for in this horizon alone two years after the final fertiliser application (Chapter 3). Fenn *et al.* (1998) also noted that in many labelled N experiments, the majority of N was retained in the litter and topsoil horizons.

This large increase in litter mass also has positive implications for carbon sequestration in plantation forests. The amount and rate of litter decomposition influences the amount of C retained in the litter and transferred to the soil and effects the ability of a forest site to store C. This has important implications for carbon accounting and potential C trading schemes (Paul and Polglase, 2004a). Although C was not measured in the litter the increased litter mass at both sites from 56 to 92 t ha^{-1} and 28 to 102 t ha^{-1} under *P. radiata* and *E. regnans* indicates a substantial increase in C at these sites due to fertilisation. In addition, under *P. radiata* soil C content in the top 10 cm doubled from 21 t ha^{-1} in NIL to 41 t ha^{-1} in (P)N1Y (Table 3.8). The total C budget for the fertilised site was predicted (CABALA) to be almost double of that unfertilised, 50 to 92 t ha^{-1}, respectively. Combined with the fact that cooler climates generally have greater soil organic matter in a less advanced stage of decomposition than soils in warmer climates (Anderson, 1992). The results from this study indicate that with careful site management during harvesting and site preparation the addition of fertilisers to increase wood production may have secondary benefits of C sequestration.

Subsequent to large increases in N retained in the litter, this study also highlighted the importance of litter horizons in the cycling of N in the forest ecosystem, with significantly higher rates of NNM in litter after annual fertilisation at both sites. Annual rates of N mineralised from litter were estimated using laboratory incubations at 10 °C (Chapter 7), this lower temperature being closer to the average temperature

observed in the field. Comparisons between observed and predicted rates of NNM in *P. radiata* confirm this assumption, as discussed above (Section 9.2.2).

Under *E. regnans,* the effect of litter horizons in the long-term increase of site productivity due to fertilisation was more pronounced. Unfertilised litter mineralised very little N during the life of the plantation (less than 0.2 kg N ha^{-1} yr^{-1}, at 10°C Chapter 7, Table 7.15). However, annual fertilisation increased NNM rates over 400-fold to 82 kg N ha^{-1} yr^{-1}. These litter layers were collected three years after the final fertilisation, indicating a natural cycling in eucalypt forests large enough to sustain enhance growth during this period.

Under *P. radiata,* mineralisation in litter increased three-fold due to large applications of N fertiliser, from 2.9 kg N ha^{-1} yr^{-1} to 10 kg N ha^{-1} yr^{-1} (incubated at 10 °C, Chapter 7 Table 7.15). Such increases would not be large enough to maintain growth. Even when NNM rates in topsoil were measured around 20 kg N ha^{-1} yr^{-1}, growth would only be maintained by internal recycling in the tree and repeated fertilisation.

The large nutrient value of the litter layer in the eucalypt system indicates the need for conservation of the litter layer during harvesting. In addition to the litter horizons supplying a direct nutrient source, litter horizons also provide a buffering capacity during fertilisation. For example in Chapter 5, litter horizons were observed to delay the movement of mineral N through the profile, retaining relatively large quantities of mineral N, and re-mineralising N at a later date when moisture and temperature conditions were more favourable. Litter horizons provide environmental protection for the large portion of feeder roots acting directly underneath. Such root protection prevents N movement through the profile, as seen in Chapter 5, often being absorbed by tree roots at the litter and soil interface. In addition, retained litter can increase moisture availability and growth in subsequent rotations (Flinn *et al.*, 1980; Hendrickson, 1985).

The effect of management practices on soil N and organic C concentration is often inconsistent. For example, some researchers have demonstrated a large effect on mineral soil C and N in coniferous forests due to residue management (Burger and Pritchett, 1984; Smethurst and Nambiar, 1990a), but a literature review and meta analysis concluded that, on average, forest harvesting had little or no effect on soil C and N (Johnson and Curtis, 2001). However, as highlighted in this study it is often difficult to observe statistical differences in soil C and N concentration due to large spatial variation in forest systems. Consequently, conflicting effects of organic matter removal on extractable N and N mineralisation have also been reported, with no effects observed by Smith *et al.*(1994), Vitousek and Matson (1985), Piatek and Allen (1999) and Li *et al.* (2003), reductions in N availability observed by Burger and Pritchett (1984), Vitousek and Matson (1985), Smethurst and Nambiar (1990b), (1995) and Piatek and Allen (1999), and increases in NNM for periods between 7 and 17 years observed by Frazer *et al.*(1990) and Matson and Vitousek (1981). Large increases have also been observed on plantation growth due to harvesting treatments. For example, early growth in second rotation *P. radiata* plantations was found to be markedly better on sites where residue (litter and logging residue) was retained (Flinn *et al.*, 1980). Results from this study suggest that long-term rates of NNM at a site will decline significantly if surface litter and soil horizons are removed during harvesting practises. In addition, this study indicates perturbations, such as increased environmental exposure, will increase rates of NNM in surface horizons. This was particularly evident when comparing predicted (CABALA) rates of NNM in litter, around 5 kg N ha^{-1} yr^{-1} compared to those measured in the laboratory after disturbance of up to106 kg N ha^{-1} yr^{-1}.

Changes due to harvesting and site preparation for plantation establishment is therefore important when assessing site organic C and N availability. Site management needs to minimise N removal and disturbance to maintain future productivity. Removal of the litter horizons at the *E. regnans* site after long-term fertiliser application during harvesting could effectively remove a potential 80 kg N ha^{-1} input for the first few years of second rotation planting. Considering the topsoil at this site mineralised only around 20 kg N ha^{-1} yr^{-1}, site productivity in terms of N alone would be reduced to one fifth of its potential if the litter horizons were removed.

Also of importance, as was indicated at both sites, excluding roots, little N was added to the mineral soil component of a forest system during fertilisation. Most of the N added was distributed between the tree and litter components.

As fertilising also changes the allocation of resources in the tree, increasing allocation of dry matter to branch biomass and decreasing stem dry matter allocation (Messina, 1992), site management such as pruning and biofuel resources also need to be managed to take into account the possible increased nutrients retained in finer tree components. Pruning regimes will occur earlier in the rotation and resupply nutrients for further growth. However, removal of fine fuels as biofuels after harvesting may decrease long-term site productivity.

At a cost of $150 per hectare for the addition of 100 kg N alone, as urea, by air (pers. comm. P. Adams), the overall management of N resources becomes essential for economic returns. As seen in this study, litter horizons, alone, supply between 10 and 80 kg of N ha^{-1} after large amounts of N have been applied. Under the eucalypt plantation, this study indicates that removal of the litter layer during site preparations could result in an economic cost of $120 ha^{-1} for a period of at least three years. That is the cost of resupplying the N alone. Under *P. radiata* although the rates of N mineralisation in the litter were substantially lower, almost 40 percent of the total Ca and 70 percent of the exchangeable Ca for the total profile depth, was tied up in the O2 horizon. Although urea additions may be adequate to amend reductions in site productivity caused by forest floor removal, fertilising will aggravated shortages and reduced uptake of other elements such as P and boron (Smith *et al.*, 1994).

9.7 Conclusions

As the demand for wood resources increase and the period of rotation length decreases, the amount of fertiliser required for wood production on a global scale will also increase. This study highlighted that large volume growth increases in both *P. radiata* and *E. regnans* plantations occurred due to N fertilisation. However, the efficiency of fertilisation utilisation varied between the sites. In *P. radiata,* although for the initial seven years after the fertilisation commencement the annual application had an advantage above application every second and fourth years, beyond this time all the N treatments showed similar volume growth response. In *E. regnans,* applications every second year were sufficient to maintain significant growth response at the same rate as annual applications, once 300 kg of N ha^{-1} had been applied to stimulate initial growth.

Annual N fertilisation (100 kg N ha^{-1}) for a period of thirteen years (in addition to the P fertiliser) increased *P. radiata* growth from 192 m^3 ha^{-1} to 344 m^3 ha^{-1}, at the age of 34 years. Annual N (plus P) fertilisation for thirteen years, doubled *E. regnans* growth from 125 to 281 m^3 ha^{-1}, at the age nineteen years. On the P deficient Kurosol, two single applications of P alone, totalling 144 kg P ha^{-1}, increased *P. radiata* volume growth from 78 m^3 ha^{-1} to 192 m^3 ha^{-1}. In contrast, application of P alone or in combination with N to the Ferrosol, up to a total of 598 P ha^{-1} had no effect on *E. regnans* growth. After fifteen years of measurements, nitrogen fertilisation significantly increased volume growth at both sites.

Concerns over long-term fertiliser effects on forest sustainability through changes in the soil chemistry were confirmed in this study. Substantial soil pH changes of 0.5 to 1.0 units throughout the soil profile could pose long-term consequences for productivity directly and through reductions in exchangeable bases. The vulnerability of some forest soils was highlighted in the Kurosol profile with its initial low pH and low buffering capacity. At both sites the reduction in active acidity was matched by reductions reserve acidity as indicated by lower base saturation in high N treatments. Although the effects of acidification could have been reduced through careful

fertiliser management, this study indicated that there was little remediation of pH decline over time.

The current study also highlighted that to examine the long-term effect of fertilisation on site changes the whole soil profile plus litter layers need to be examined. Long-term changes may not be observed when measuring the topsoil alone because of mineral cycling this soil horizon. For example, application of N alone in the Ferrosol halved the total Ca content throughout the entire depth of the profile, compared to the unfertilised. In the Kurosol, high rates of N application significantly reduced exchangeable Mg by half throughout the entire profile. In the Ferrosol topsoil, substantial reduction in exchangeable Mg concentration, from 258 to 71 mg kg^{-1}, was also measured.

Long-term fertilisation resulted in a net transfer of nutrients for the soil into the tree and litter horizons. As a result, a large proportion of sites nutrient capital was tied up in the components of the system most effected by the eventual harvesting of the trees and site preparation of the next rotation. Under *E. regnans*, at the highest rate of fertilisation, N, P, S and Ca all accumulated in the O2 horizon. The O2 horizon of the most heavily fertilised treatment had over seven times the N content of unfertilised treatment, equivalent to about 60 % of the N applied. However, soil to a depth of 50 cm contained less N in the fertilised treatment, suggesting a transfer of soil N to the O2 horizon nutrient pool from the mineral soil. The litter might provide a buffer for future fertiliser additions where N can be immobilised and released during a period of time under favourable climatic conditions. In addition, large increase in litter mass has positive implications for carbon sequestration in plantation forests. At the *P. radiata* site the predicted total C budget for the fertilised site double from 50 to 92 t ha^{-1} due to fertilisation.

To measure the effect of long-term fertilisation on N cycling, N mineralisation was examined in the field and laboratory for both sites. This study indicated that:

- Low rates of *in situ* N mineralisation occurred in topsoil at both sites, and neither soil was significantly effected by long-term fertilisation (Chapter 4). Net N mineralisation ranged between 13 and 52 kg N ha^{-1} yr^{-1} across both soil types and fertiliser treatments.
- Large site variability made it difficult to assess N mineralisation with the methods applied using the field *in situ* incubations. Although not significant, average rates of *in situ* NNM in Kurosol topsoil increased four-fold due to long-term fertilisation. In the laboratory, under controlled conditions, the effect of fertilisation on NNM in Kurosol topsoil increased to ten times that measured in the unfertilised topsoil, and was significant. Generally, rates of NNM in the laboratory between fertilised and unfertilised Ferrosol topsoil remained similar.
- This study highlighted the strong effect of laboratory parameters, moisture and temperature, on topsoil NNM, which were often larger than the effect of long-term fertilisation during 30-60 d incubations.
- It was concluded from this study that the litter was more sensitive than soil to changes in N cycling. The litter horizons had higher rates of NNM than the soil due to increases in both NNM per unit mass and litter mass. These rates were also less effected by laboratory parameters than the topsoil.
- In both litter and topsoil, when there were smaller changes in N inputs (i.e. application every second or fourth year) the effects on NNM could be masked by the effects of laboratory parameters. That is, the change in rates of NNM due to incubating samples at moisture and temperatures outside the range observed in the field is greater than that between the fertiliser treatments. As cool temperate forests often have low rates of NNM, the use of laboratory measurements of NNM to predict field rates is questionable, when using the current methods applied in this study. To prevent this the incubation of intact cores has been suggested to better reflect field conditions (Adams and Attiwill, 1986; Raison *et al.*, 1987). However, results in this study indicated that an extremely high number of intact soil-core replicates would be required to show this in a statistically consistent manner.

Process-based models, such as CABALA, can be used to identify likely limitations to plant growth, define growth potential and predict overall site changes from fertilisation. In this study, CABALA adequately simulated growth and N dynamics in the *P. radiata* plantation for approximately one rotation. Predicted growth increases due to annual fertilisation were within 15 % of the actual growth at age 34 years. In agreement with field measurements, low rates of nitrification and little offsite movement was also predicted, with less 5 % of the N applied as fertiliser predicted to be was lost from the system. Predicted data in this study also indicated that despite the rainfall at the *P. radiata* site considered to be high by Australian standards, further growth increases would probably require a combination of irrigation and fertilisation.

9.8 Future research

The ability to predict growth and several other site characteristics due to intensive silvicultural management of plantations are provided by a number of models, such as the CABALA model used in this study. However, this study also indicated that there are still a number of knowledge gaps that need to be quantified to allow improved understanding of the changes that occur in the forest. Predictions using the CABALA model need to be tested across multiple rotations, and the N sub-module might benefit from more explicit definition of N mineralisation in the litter layer, and clarification of the role of C: N ratio and physical protection in controlling NNM. Aspects of leaching, subsoil retention, and canopy storage of N in CABALA also require attention.

Of particular importance is further investigation into the large removal of cations from throughout the soil profile and substantial changes in pH, which were observed in this study. How these changes progress during successive rotations, in particular short intensive rotations, will determine the long-term effects of the current silvicultural practices. Changes in available cations, such as Ca and Mg, will need to be closely monitored to maintain optimum plantation productivity. To measure long-term changes in forests, monitoring will need to include subsoil as well as topsoil.

Further investigation of the role of litter in N turnover is required for its application to current N mineralisation predictive models such as SNAP.

This study indicated that due to the low rates of NNM in these temperate forests, and high site variability, the current methodology used to measure NNM may not be sensitive enough to predict changes due to large N inputs in these systems. In this study, changes in NNM due to fertilisation were often swamped by the short-term changes that occurred due to temperature, moisture and soil disturbance. Further investigation of relationship between soil structure (such as aggregate stability and clay content) and NNM would enable the development of more sensitive methods for measuring NNM and be incorporated into predictive models.

References

Aarnio, T. and P. J. Martikainen (1995). "Mineralization of C and N and nitrification in Scots pine forest soil treated with nitrogen fertilizers containing different proportions of urea and its slow-releasing derivative, ureaformaldehyde." Soil Biology and Biochemistry **27**(10): 1325-1331.

Aarnio, T., K. McCullough, *et al.* (1996). "Fate of Urea and Ureaformaldehyde nitrogen in a one-year laboratory incubation with Douglas-fir forest floor." Soil Biology and Biochemistry **28**(10/11): 1407-1415.

Aber, J. D. and J. M. Melillo (1980). "Litter decomposition: measuring relative contributions of organic matter and nitrogen to forest soils." Canadian Journal of Botany **58**(4): 416-421.

Aber, J. D. and C. A. Federer (1992). "A generalized, lumped-parameter model photosynthese, evapotranspiration and net primary production in temperate and boreal forest ecosystems." Oecologia **92**: 463-474.

Adams, M. A. and P. M. Attiwill (1983). "Patterns of nitrogen mineralisation in 23-year old pine forests following N fertilising." Forest Ecology and Management **7**: 241-248.

Adams, M. A. and J. B. Martin (1984). Liming effects on nitrogen use and efficiency. Nitrogen in Crop Production. R. D. Hauck. Madison, American Society of Agronomy**:** 417-426.

Adams, M. A. and P. M. Attiwill (1986). "Nutrient cycling and N mineralisation in Eucalypt forests of S.E Australia: II indices of N mineralisation." Plant and Soil **92**: 341-362.

Adams, M. A. and P. M. Attiwill (1988). Nutrient cycling in forest of N.E Tasmania, Tasmanian Forests Research Council inc.

Adams, M. A., P. M. Attiwill, *et al.* (1989a). "Availability of nitrogen and phosphorous in soils in northeastern Tasmania." Biology Fertility of Soils **8**: 212-218.

Adams, M. A., P. J. Polglase, *et al.* (1989b). "*In situ* studies of nitrogen mineralization and uptake in forest soils; some comments on methodology." Soil Biology and Biochemistry **21**(3): 423-429.

Adams, M. A. and P. M. Attiwill (1991). "Nutrient balance in forest of northern Tasmania. 2. Alteration of nutrient availability and soil-water chemistry as a

result of logging, slash burning and fertilizer application." Forest Ecology and Management **44**: 115-131.

Aggangan, R. T., A. M. O'Connell, *et al.* (1998). "Fertiliser and previous land use effects on C and N mineralisation in soils from *Eucalyptus globulus* plantations." Soil Biology and Biochemistry **13**: 1791-1798.

Aggangan, R. T., A. M. O'Connell, *et al.* (1999). "The effects of *Eucalyptus globulus.* Labill. leaf litter on C and N mineralisation in soils from pasture and native forests." Soil Biology and Biochemistry **31**: 1481-1487.

Anderson, J. M. (1992). "Responses of soil to climate change." Advances in Ecological Research **22**: 163-210.

Armson, K. A. (1979). Forest soils: properties and processes.

Attiwill, P. M., H. B. Guthrie, *et al.* (1978). "Nutrient cycling in *Eucalyptus obliqua* (L'Herit.) forest. I Litter production and nutrient return." Australian Journal of Botany **26**: 79-91.

Attiwill, P. M. (1980). "Nutrient cycling in a *Eucalyptus obliqua* (L'Herit.) forest. IV Nutrient uptake and nutrient return." Australian Journal of Botany **28**: 199-222.

Attiwill, P. M. (1983). Phosphorus in Australian Forests. Phosphorus in Australia. A. B. Costin and C. H. Williams. Canberra, Centre for Resource an Environmental Studies, Australian National University.

Attiwill, P. M. and G. W. Leeper (1987). Forest Soils and Nutrient Cycles. Melbourne, Melbourne University Press.

Attiwill, P. M. (1996). Nutrition of Eucalypts.

Baker, T. G. and P. M. Attiwill (1985). "Above-ground nutrient distribution and cycling in *Pinus radiata* D.Don and *Eucalyptus obliua* L'Herit. Forests in Southeastern Australia." Forest Ecology and Management **13**: 41-52.

Baker, T. G., G. R. Oliver, *et al.* (1986). "Distribution and cycling of nutrients in *Pinus radiata* as affected by past lupin growth and fertilizer." Forest Ecology and Management **17**: 169-187.

Ballard, R. (1978). "Effect of first rotation phosphorus applications on fertilizer requirements of second rotation radiata pine." New Zealand Journal of Forestry Science **8**(1): 135-145.

Bartlett, R. and B. James (1980). "Studying dried, stored soil samples- some pitfalls." Soil Science Society of America Journal **44**: 721-724.

Battaglia, M. and P. Sands (1997). "Modelling Site Productivity of Eucalyptus globulus in Response to Climatic and Site Factors." Australian Journal of Plant Physiology **24**: 831-850.

Battaglia, M. and P. Sands (1998). "Application of sensitivity analysis to a model of Eucalyptus globulus plantation productivity." Ecological Modelling **111**(3): 237-259.

Battaglia, M., P. Sands, *et al.* (2004). "CABALA: a linked carbon, water and nitrogen model of forest growth for silvicultural decision support." Forest Ecology and Management **193**: 251-282.

Bauhus, J. and P. K. Khanna (1994). "Carbon and nitrogen turnover in two acid forest soils of southeast Australia as affected by phosphorus addition and drying and rewetting cycles." Biology Fertility of Soils **17**: 212-218.

Bauhus, J. (1996). "C and N mineralization in an acid forest soil along a gap-stand gradient." Soil Biology and Biochemistry **28**(7): 923-932.

Beier, C. and H. Eckerersten (1998). "Modelling the effects of nitrogen addition on soil nitrogen status and nitrogen uptake in a Norway Spruce stand in Denmark." Environmental Pollution **102**(51): 409-414.

Bennett, L. T., C. J. Weston, *et al.* (1997). "Biomass, nutrient and growth responses to fertilisers of six-year-old *Eucalyptus globulus* plantations at three contrasting sites in Gippsland, Victoria." Australian Journal of Botany **45**: 103-121.

Benson, M. L., J. J. Landsberg, *et al.* (1992a). "The biology of forest growth experiment: an introduction." Forest Ecology and Management **52**: 1-16.

Benson, M. L., B. J. Myers, *et al.* (1992b). "Dynamics of stem growth of *Pinus radiata* as affected by water and nitrogen supply." Forest Ecology and Management **52**: 117-137.

Berg, B. and G. Ekbohm (1983). "Nitrogen mineralization in decomposing needle litter at variable carbon: nitrogen ratios." Ecology **64**(1): 63-67.

Berg, B. and C. O. Tamm (1991). "Decomposition and nutrient dynamics of litter in long-term optimum nutrition experiments. 1. Organic matter decomposition in *Picea abies* needle litter." Scandinavian Journal of Forestry Research **6**(3): 305-321.

Berg, B. (2000). "Litter decomposition and organic matter turnover in northern forest soils." Forest Ecology and Management **133**: 13-22.

Binkley, D., C. T. Driscoll, *et al.* (1988). Acidic deposition and forest soils. New York, Springer-Verlag.

Binkley, D. and S. C. Hart (1989). "The components of nitrogen availability assessments in forest soils." Advances in Soil Science **10**: 55-112.

Birch, H. F. (1958). "The effect of soil drying on humus decomposition and nitrogen availability." Plant and Soil **10**(1): 9-31.

Birk, E. M. (1979). "Overstorey and understorey litter fall in a Eucalyptus forest: Spacial and temperal variability." Australian Journal of Botany **27**: 145-156.

Birk, E. M. (1991). "Stem and branch form of 20-year-old radiata pine in relation to previous land use." Australian Forestry **54**: 30-39.

Birk, E. M. (1992). "Nitrogen availability in radiata pine plantations on former pasture sites in southern New South Wales." Plant and Soil **143**: 115-125.

Birk, E. M. and J. Turner (1992). "Response of flooded gum (*E. grandis*) to intensive cultural treatments: biomass and nutrient content of eucalypt plantations and native forests." Forest Ecology and Management **47**: 1-28.

Bonde, T. A. and T. Rosswall (1987). "Seasonal variation of potentially mineralizable nitrogen in four cropping systems." Soil Science Society of America Journal **51**: 1508-1514.

Boone, R. D. (1992). "Influence of sampling date and substrate on nitrogen mineralization: comparison of laboratory-incubation and buried-bag methods for two Massachuetts forest soils." Canadian Journal of Forest Research **22**: 1895-1900.

Bromfield, S. M., R. W. Cumming, *et al.* (1983). "Changes in soil pH, manganses and aluminium under subterranean clover pasture." Australian Journal of Experimental Agriculture and Animal Husbandry **23**: 181-191.

Burger, J. A. and W. L. Pritchett (1984). "Effects of clearfelling and site preparation on nitrogen mineralization in a southern pine stand." Soil Science Society of America Journal **48**: 1432-1437.

Cabrera, M. L. and D. E. Kissel (1988a). "Potentially mineralizable nitrogen in disturbed and undisturbed soil samples." Soil Science Society of America Journal **52**: 1010-1015.

Cabrera, M. L. and D. E. Kissel (1988b). "Evaluation of a method to predict nitrogen mineralized from soil organic matter under field conditions." Soil Science Society of America Journal **52**(4): 1027-1031.

Cabrera, M. L. and D. E. Kissel (1988c). "Length of incubation time effects the parameter values of the double exponential model of nitrogen mineralization." Soil Science Society of America Journal **52**(4): 1186-1187.

Campbell, I. C. and T. J. Doeg (1989). "Impact of timber harvesting and production on streams: a review." Australian Journal of Marine and Freshwater Research **40**: 519-539.

Canary, J. D., R. B. Harrison, *et al.* (2000). "Additional carbon sequestration following repeated urea fertilization of second-growth Douglas-fir stands in western Washington." Forest Ecology and Management **138**: 225-232.

Carlyle, J. C. (1986). "Nitrogen Cycling in Forested Ecosystems." Forestry Abstracts **47**(5): 307-336.

Carlyle, J. C., J. R. Lowther, *et al.* (1990). "Influence of Chemical Properties of Nitrogen Mineralization and Nitrification in Podzolized Sands. Implications for Forest Management." Australian Journal of Soil Research **28**: 981-1000.

Carlyle, J. C., M. W. Bligh, *et al.* (1998a). "Woody residue management to reduce nitrogen and phosphorus leaching from sandy soil after clear-felling *Pinus radiata* plantations." Canadian Journal of Forest Research **28**: 1222-1232.

Carlyle, J. C., E. K. Nambiar, *et al.* (1998b). "The use of laboratory measurements to predict nitrogen mineralisation and nitrification in *Pinus radiata* plantations after harvesting." Canadian Journal of Forest Research **28**: 1213-1221.

Cassman, K. G. and D. N. Munns (1980). "Nitrogen mineralization as affected by soil moisture, temperature and depth." Soil Science Society of America Journal **44**(6): 1233-1237.

Cherry, M. L., A. Hingston, *et al.* (1998). "Calibrating the LI-COR LAI-2000 fore estimating leaf area index in eucalypt plantations." Tasforests **10**: 75-82.

Cherry, M. L., C. MacFarlane, *et al.* (2002). Visual guide to leaf area index of eucalypt plantations. C. R. C. f. S. P. Forestry. Hobart: 22.

Clein, J. S. and P. Schimal (1993). "Reduction in microbial activity in Birch litter due to drying and rewetting events." Soil Biology and Biochemistry **26**: 403-406.

Cole, D. W. and M. Rapp (1981). Elemental cycling in forest ecosystems. Dynamic properties of forest ecosystems. D. E. Reichle. Cambridge, Cambridge University Press: 205-265.

Cole, D. W. (1995). "Soil nutrient supply in natural and managed forests." Plant and Soil **168-169**: 43-53.

Comerford, N. B., M. McLeod, *et al.* (2002). "Phosphorous form and availability in the pine rotation following fertilization P fertilization influences P form and potential bioavailability to pine in the subsequent rotation." Forest Ecology and Management **169**: 203-211.

Comins, H. N. and R. E. McMurtrie (1992). "Long-term responses of nutrient-limited forests to CO_2 enrichment; Equilibrium behaviour of plant-soil models." Ecological Applications **3**(4): 666-681.

Connell, M. J., R. J. Raison, *et al.* (1995). "Nitrogen mineralization in relation to site history and soil properties for a range of Australian forest soils." Biology Fertility of Soils **20**: 213-220.

Corbeels, M. (2001). Plant litter and decomposition: General concepts and model approaches. Net Ecosystem Exchange, Canberra, CRC for Greenhouse Accounting.

Crane, W. J. B. (1978). "Phosphorus stability in Eucalypt forests." Australian Forestry **41**(2): 118-126.

Crane, W. J. B. and J. C. G. Banks (1992). "Accumulation and retranslocation of foliar nitrogen in fertilised and irrigated *Pinus radiata*." Forest Ecology and Management **52**: 201-223.

Cresser, M., K. Killham, *et al.* (1993). Soil chemistry and its applications. Cambridge, Cambridge University Press.

Cromer, R. N., M. Raupach, *et al.* (1975). "Eucalypt plantations in Australia- The potential for intensive production and utilization." Appita **29**(3): 165-172.

Cromer, R. N., D. M. Cameron, *et al.* (1981). "Response of eucalypt species to fertiliser applied soon after planting at several sites." Australian Forestry **44**(1): 3-13.

Cromer, R. N., D. M. Cameron, *et al.* (1993). "Response to nutrients in *Eucalyptus grandis*. 2. Nitrogen accumulation." Forest Ecology and Management **62**: 231-243.

Crow, T. R. (1971). Estimation of biomass in an even-aged stand-regression and 'mean tree' techniques. Working Group on Forest Biomass Studies. Section 25, Growth and Yield. International Union of Forest Research Organizations. XVth IUFRO Congress, University of Florida, Gainesville, Florida, USA.

Davidson, E. A., S. C. Hart, *et al.* (1992). "Internal cycling of nitrate in soils of a mature coniferous forest." Ecology **73**(4): 1148-1156.

Degens, B. P. and G. P. Sparling (1995). "Repeated wet-dry cycles do not accelerate the mineralization of organic C involved in the macro-aggregation of a sandy loam soil." Plant and Soil **175**: 197-203.

Dell, B., N. Malajczuk, *et al.* (2002). Nutrient disorders in plantation eucalypts. Canberra.

DeMontigny, L. E., C. M. Preston, *et al.* (1993). "Comparison of humus horizons from two ecosystem phases on northern Vancouver Island using ^{13}C CPMAS NMR spectroscopy and CuO oxidation." Canadian Journal of Soil Science **73**: 9-25.

Dyck, W. J., J. R. Gosz, *et al.* (1983). "Nitrate losses from disturbed ecosystems in New Zealand- a comparative analysis." New Zealand Journal of Forestry Science **13**(1): 14-24.

Edmonds, R. L. (1980). "Litter decomposition and nutrient release in Douglas-fir, red alder, western hemlock, and Pacific silver fir ecosystems in western Washington." Canadian Journal of Forest Research **10**: 327-337.

Ellert, B. H. and J. R. Bettany (1988). "Comparison of kinetic models for describing net sulfure and nitrogen mineralization." Soil Science Society of America Journal **52**: 1692-1702.

Ellert, B. H. and J. R. Bettany (1992). "Temperature dependence of net nitrogen and sulfur mineralization." Soil Science Society of America Journal **56**: 1133-1141.

Elliott, H. J., R. Bashford, *et al.* (1993). "Effects of defoliation by the leaf beetle, *Chrysophtharta bimaculata*, on growth of *Eucalytpus regnans* plantations in Tasmania." Australian Forestry **56**(1): 22-26.

Elliott, H. J. and B. S. Hodgson (2004). "Water sampling by Forestry Tasmania to determine presence of pesticides and fertiliser nutrients, 1993-2003." Tasforests **15**: 29-42.

Ellis, R. C. (1974). "The seasonal pattern of nitrogen and carbon mineralization in forest and pasture soils in Southern Ontario." Canadian Journal of Forest Research **54**: 15-28.

Ellis, R. C. and A. M. Graley (1987). "Soil Chemical properties as related to forest succession in a highland area in north-eastern Tasmania." Australian Journal of Ecology **12**: 307-317.

Ellis, R. C. and P. I. Pennington (1989). "Nitrification in soils of secondary vegetational successions from Eucalyptus forest and grassland to cool temperate rainforest in Tasmania." Plant and Soil **115**: 59-73.

Evans, C. A., E. K. Miller, *et al.* (1998). "Nitrogen mineralization associated with birch and fir under different soil moisture regimes." Canadian Journal of Forest Research **28**: 1890-1898.

Fagerstrom, T. and U. Lohm (1977). "Growth of Scots pine (*Pinus silvestris* L.) mechanism of response to nitrogen." Oecologia **26**: 305-315.

Falkiner, R. A., P. K. Khanna, *et al.* (1993). "Effects of Superphosphate Addition on N Mineralization in some Australian Forest Soils." Australian Journal of Soil Research **31**: 285-296.

Federer, C. A. (1983). "Nitrogen Mineralization and Nitrification: Depth variation in 4 New England forest soils." Soil Science Society of America Journal **44**: 1314-1320.

Feigin, A., D. H. Kohl, *et al.* (1974). "Variation in the natural nitrogen-15 abundance in nitrate mineralized during incubation of several Illinios soils." Soil Science Society of America Proc. **38**: 90-95.

Feller, M. C. (1978). "Nutrient movement into soils beneath eucalypt and exotic conifer forests in southern central Victoria." Australian Journal of Ecology **3**(4): 357-372.

Fenn, M. E., M. A. Poth, *et al.* (1998). "Nitrogen excess in North America ecosystems: Predisposing factors, ecosystem responses, and management strategies." Ecological Applications **8**(3): 706-733.

Fife, D. N. and E. K. Nambiar (1997). "Changes in the canopy and the growth of *Pinus radiata* in response to nitrogen supply." Forest Ecology and Management **93**: 137-152.

Fisher, D. S., J. L. Steiner, *et al.* (2000). "The relationship of land use practices to surface water quality in the Upper Oconee Watershed of Georgia." Forest Ecology and Management **128**: 39-48.

Fisher, R. F. and W. S. Garbett (1980). "Response of semimature Slash and Loblolly pine plantations to fertilization with nitrogen and phosphorus." Soil Science Society of America Journal **44**: 850-854.

Fisher, R. F. and D. Binkley (2000). Ecology and Management of Forest Soils. New York, John Wiley & Sons, Inc.

Flinn, D. W., P. Hopmans, *et al.* (1979a). "Response of radiata pine to fertilisers containing N and P applied at planting." Australian Forestry **42**(4): 125-131.

Flinn, D. W., I. M. Moller, *et al.* (1979b). "Sustained growth responses to superphosphate applied to established stands of *Pinus radiata*." New Zealand Journal of Forestry Science **9**(2): 201-211.

Flinn, D. W., R. O. Squire, *et al.* (1980). "The role of organic matter in the maintenance of site productivity on sandy soils." New Zealand Journal of Forestry **25**: 229-236.

Florence, R. G. and D. Lamb (1974). "Influence of stand and site on Radiata Pine litter in South Australia." New Zealand Journal of Forestry Science **1974**(4): 502-510.

Foster, N. W. (1989). "Influence of seasonal temperature on nitrogen and sulfur mineralization/immobilization in a Maple-Birch forest floor in Central Ontario." Canadian Journal of Forest Research **69**: 501-514.

Frazer, D. W., J. G. McColl, *et al.* (1990). "Soil nitrogen mineralization in a clearcutting chronosequence in a northern California conifer forest." Soil Science Society of America Journal **54**: 1145-1152.

Frederick, D. J., H. A. I. Madgwick, *et al.* (1985). "Dry matter content and nutrient distribution in an age series of *Eucalyptus regnans* plantations in New Zealand." New Zealand Journal of Forestry Science **15**(2): 158-179.

Fyles, I. H. and W. B. McGill (1987). "Nitrogen mineralization in forest soil profiles from central Alberta." Canadian Journal of Forest Research **17**: 242-249.

Fyles, J. W., I. H. Fyles, *et al.* (1990). "Comparison of nitrogen mineralization in forest floor materials using aerobic and anaerobic incubation and bioassay techniques." Canadian Journal of Soil Science **70**: 73-81.

Gallardo, A. and J. Merino (1992). "Nitrogen immobilization in leaf litter at two Mediterranean ecosystems of SW Spain." Biogeochemistry **15**: 213-228.

Genstat 5 Committee (1988). Reference Manual. Oxford, Oxford Science Publication.

Gentle, S. W., F. R. Humphreys, *et al.* (1965). "An examination of a *Pinus radiata* phosphate fertilizer trial fifteen years after treatment." Forest Science **11**(3): 315-324.

Gentle, S. W., F. R. Humphreys, *et al.* (1986). "Continuing responses of *Pinus radiata* to phosphatic fertilizers over two rotations." Forest Science **32**(3): 822-829.

Goncalves, J. L. M. and J. C. Carlyle (1994). "Modelling the influence of moisture and temperature on net nitrogen mineralization in a forested sandy soil." Soil Biology and Biochemistry **26**(11): 1557-1574.

Goodwin, D. C. and C. A. Jones (1991). Nitrogen dynamics in soil plant systems. Modeling Plant and Soil Systems. J. Hanks and J. T. Ritchie. Madison, Wisconsin USA, American Society of Agronomy, Inc**:** 187-321.

Gower, S. T. and Y. Son (1992). "Differences in soil and leaf litter nitrogen dynamics for five forest plantations." Soil Science Society of America Journal **56**: 1959-1966.

Grant, J. C., M. D. Laffan*, et al.* (1995). Forest Soils of Tasmania. A Handbook for Identification and Management. Launceston, Sprinta Print Pty Ltd.

Grier, C. C., K. A. Vogt*, et al.* (1981). "Biomass distribution and above- and below-ground production in young and mature *Abies amabilis* zone ecosystem of the Washington Cascades." Canadian Journal of Forest Research **11**: 155-167.

Griffin, D. M. (1981). Water potential as a selective factor in the microbial ecology of soils. Water Potential Relations in Soil Microbiology. J. F. Parr, W. R. Gardener and L. F. Elliot. Madison, Soil Science Society of America.

Hamilton, C. D. (1965). "Changes in the soil under *Pinus radiata*." Australian Forestry **29**(4): 275-289.

Haque, I. and D. Warmsley (1972). "Incubation studies on mineralization of organic sulfure and organic nitrogen." Plant and Soil **37**: 255-264.

Harris, M. M. and S. J. Riha (1991). "Carbon and nitrogen dynamics in forest floor during short-term laboratory incubations." Soil Biology and Biochemistry **23**(11): 1035-1041.

Harris, R. F. (1981). Effects of Water Potential on Microbial Growth and Activity. Water Potential Relations in Soil Microbiology. J. F. Parr, W. R. Gardener and L. F. Elliot. Wisconsin, Soil Science Society of America. **9:** 23-96.

Hart, S. C., G. E. Nason*, et al.* (1994). "Dynamics of gross nitrogen transformations in an old-growth forest: The carbon connection." Ecology **75**(4): 880-891.

Hart, S. C. and P. Sollins (1998). "Soil carbon and nitrogen pools and processes in an old growth conifer forest 13 years after trenching." Canadian Journal of Forest Research **28**: 1261-1265.

Heal, O. W. (1979). Decomposition and nutrient release in even-aged plantations. The ecology or even aged forest plantations, Edinburgh, IUFRO Division 1.

Heal, O. W., J. M. Anderson, *et al.* (1997). Plant litter quality and decomposition: An historical overview. Driven by Nature: Plant Litter Quality and Decomposition. G. Cadisch and K. E. Giller. Cambridge, CAN INTERNATIONAL: 3-30.

Heilman, P. (1974). "Effect of urea fertilization on nitrification in forest soils of the Pacific Northwest." Soil Science Society of America Journal **38**: 664-667.

Heilman, P. E., T. H. Dao, *et al.* (1982). "Comparison of fall and spring application of ^{15}N-labeled urea to Douglas-fir: II. Fertilizer nitrogen recovery in trees and soil after 2 years." Soil Science Society of America Journal **46**: 1300-1304.

Hendrickson, O. Q. and J. B. Robinson (1984). "Effects of roots and litter on mineralization processes in forest soil." Plant and Soil **80**: 391-405.

Hendrickson, O. Q. (1985). "Variation in the C: N ratio of substrate mineralized during forest humus decomposition." Soil Biology and Biochemistry **17**(4): 435-440.

Herbert, A. H., M. Laffan, *et al.* (1995). Soils: Technical report No. 2. Laboratory procedures for soil analysis and preparation of plant materials. F. Tasmania. Hobart: 17.

Herbert, M. A. (1990). "Fertilizer/Site interactions on the growth and foliar nutrient levels of *Eucalyptus grandis*." Forest Ecology and Management **30**: 247-257.

Hingston, F. J. (1984). Application of agricultural methodology to forest soils. Australian Forestry Council Workshop Group 3 Meeting, Launceston.

Hingston, F. J. and M. S. Jones (1985). "Changes in the composition on soil solutions resulting from application of fertilizer to Jarrah forest in south-western Australia." Australian Forest Research **15**: 293-308.

Homann, P. S., B. A. Caldwell, *et al.* (2001). "Douglas-fir soil C and N properties a decade after termination of urea fertilization." Canadian Journal of Forest Research **31**: 2225-2236.

Hooda, N. and C. J. Weston (1999). "Influence of site and fertilizer additions on nutrient cycling in *Eucalyptus globulus* plantations in Gippsland, South-eastern Australia. I. Foliage and litter quality." Australian Journal of Botany **47**: 187-206.

Hopmans, P., D. W. Flinn, *et al.* (1987). "Nutrient dynamics of forested catchments in Southeastern Australia and changes in water quality and nutrient exports following clearing." Forest Ecology and Management **20**: 209-231.

Hopmans, P. and D. W. Flinn (1998). Long-term responses of radiata pine to phosphate fertiliser on a strongly P-fixing soil. Environmental Forest Science, IUFRO Division 8 Conference, Kyoto, Japan, Kluwer Academic Publishers.

Hunter, I. R. and G. F. Hoy (1983). "Growth and nutrition of *Pinus radiata* on a recent coastal sand as affected by nitrogen fertiliser." New Zealand Journal of Forestry Science **13**(1): 3-13.

Hunter, I. R., H. M. Hunter, *et al.* (1987). "Pinus radiata stem volume increment and its relationship to needle mass, foliar and soil nutrients, and fertiliser inputs." New Zealand Journal of Forestry Science **17**(1): 67-75.

Isbell, R. F. (1996). The Australian Soil Classification. Collingwood, CSIRO Publishing.

Jacobson, S. and F. Pettersson (2001). "Growth responses following nitrogen and N-P-K-Mg additions to previously N-fertilized Scots pine and Norway spruce stands on mineral soils in Sweden." Canadian Journal of Forest Research **31**: 899-909.

Javid, Z. and R. F. Fisher (1990). "Fertilzation influences nitrogen dynamics in soils under *Dalbergia sissoo*." Plant and Soil **123**: 67-71.

Johnson, D. W., N. T. Edwards, *et al.* (1980). "Nitrogen mineralization, immobilization and nitrification following Urea fertilization of a forest soil under field and laboratory conditions." Soil Science Society of America Journal **44**: 610-616.

Johnson, D. W. (1995). Role of carbon in the cycling of other nutrients in forested ecosystems. Carbon Forms and Functions in Forest Soils. W. W. McFee and J. M. Kelly. Madison, Soil Science Society of America, Inc: 299-328.

Johnson, D. W. and P. S. Curtis (2001). "Effects of forest management on soil C and N storage: meta analysis." Forest Ecology and Management **140**: 227-238.

Judd, T. S., L. T. Bennett, *et al.* (1996). "The response of growth and foliar nutrients to fertilizers in young *Eucalyptus globulus* (Labill.) plantations in Gippsland, southeastern Australia." Forest Ecology and Management **82**: 87-101.

Jurgensen, M. F., D. J. Frederick, *et al.* (1986). "Soil development under *Pinus radiata* and *Eucalyptus regnans* plantations." New Zealand Journal of Forestry Science **16**(1): 69-77.

Keeney, D. R. (1980). "Prediction of soil nitrogen availability in forest ecosystems: A literature review." Forest Science **26**(1): 159-171.

Keeves, A. (1966). "Some evidence of loss of productivity with successive rotations of Pinus radiata in the south-east of South Australia." Australian Forestry **30**: 51-63.

Kelly, J. M. and G. S. Henderson (1978). "Effects of nitrogen and phosphorous additions on deciduous litter decomposition." Soil Science Society of America Journal **42**: 972-976.

Khanna, P. K., R. J. Raison, *et al.* (1992). "Effects of N P K fertilisation on the chemistry of a yellow podzolic soil under *Pinus radiata*." Forest Ecology and Management **52**: 65-85.

Kirschbaum, M. U. F. (1995). "The temperature dependence of soil organic matter decomposition and the effect of global warming on soil organic C storage." Soil Biology and Biochemistry **27**(6): 753-760.

Kirschbaum, M. U. F. (1999). "Cen W, a forest growth model with linked carbon, energy, nutrient and water cycles." Ecological Modelling **118**: 17-59.

Kirschbaum, M. U. F. (2000). "Will changes in soil organic carbon act as a positive or negative feedback on global warming." Biogeochemistry **48**: 21-51.

Kushla, J. D. and R. F. Fisher (1980). "Predicting slash pine response to nitrogen and phosphorous fertilization." Soil Science Society of America Journal **44**: 1303-1306.

Lamb, D. and R. G. Florence (1975). "Influence of soil type on the nitrogen and phosphorus content of radiata pine litter." New Zealand Journal of Forestry Science **5**(2): 143-151.

Lamb, D. (1976). "Decomposition of organic matter on the forest floor of *Pinus radiata* plantations." Journal of Soil Science **27**: 206-217.

Landsberg, J. J. and R. H. Waring (1997). "A generalised model of forest productivity using simplified concepts of radiation-use efficiency, carbon balance and partitioning." Forest Ecology and Management **95**: 209-228.

Leon, A. (1989). "The Tasmanian Eucalypt leaf beetle, *Chrysoptharta bimaculata*: an overview of the problem and current control methods." Tasforests **1**: 33-37.

Li, Q.-C., H. L. Allen, *et al.* (2003). "Nitrogen mineralization dynamics following the establishment of a loblolly pine plantation." Canadian Journal of Forest Research **33**: 364-374.

Lim, M. T. and J. E. Cousens (1986). "The internal transfer of nutrients in a Scots Pine stand 2. The patterns of transfer and the effects of nitrogen availability." Forestry **59**: 17-27.

Linn, D. M. and J. W. Doran (1984). "Effect of water-filled pore space on carbon dioxide and nitrous oxide production in tilled and nontilled soils." Soil Science Society of America Journal **48**: 1267-1272.

Lowther, J. R. (1980). "Use of single acid-hydrogen peroxide digest for analysis of *Pinus radiata* needles." Communications in Soil Science and Plant Analysis **11**: 175-188.

Lund, V. and J. Goksoyr (1980). "Effects of water fluctuations on microbial mass and activity in soil." Microbiolical Ecology **6**: 115-123.

MacDonald, N. W., D. R. Zak, *et al.* (1995). "Temperature effects on kinetics of microbial respiration and net nitrogen and sulfur mineralization." Soil Science Society of America Journal **59**: 233-240.

MacKay, D. C. and J. M. Carefoot (1981). "Control of water content in laboratory determination of mineralizable nitrogen in soils." Soil Science Society of America Journal **45**: 444.

Maier, C. A. and L. W. Kress (2000). "Soil CO_2 evolution and root respiration in 11 year-old loblolly pine (*Pinus taeda*) plantations as affected by moisture and nutrient availability." Canadian Journal of Forest Research **30**: 347-359.

Manoharan, V., P. Loganathan, *et al.* (1995). Effects of long-term application of phosphate fertilisers on soil acidity under pasture in New Zealand. Third International Symposium on Plant-Soil Interactions at Low pH, Brisbane, Kluwer Academic Publishers.

Mary, B., N. Beaudoin, *et al.* (1999). "Calculation of nitrogen mineralization and leaching in fallow soil using simple dynamic model." European Journal of Soil Science **50**: 549-566.

Mathers, N. J., X. A. Mao, *et al.* (2000). "Recent advances in the application of ^{13}C and ^{15}N NMR spectroscopy in soil organic matter studies." Australian Journal of Soil Research **38**: 769-87.

Matson, P. A. and P. M. Vitousek (1981). "Nitrogen mineralization and nitrification potentials following clearcutting in the Hoosier National Forest, Indiana." Forest Science **27**(4): 781-791.

Matson, P. A., K. A. Lohse, *et al.* (2002). "The globalisation of nitrogen deposition: Consequences for terrestrial ecosystems." Ambio **31**(2): 113-119.

McClaugherty, C. A., J. D. Aber, *et al.* (1982). "The role of fine roots in the organic matter and nitrogen budgets of two forested ecosystems." Ecology **63**(5): 1481-1490.

McDonald, R. C., R. F. Isbell, *et al.* (1990). Australian soil and land survey field handbook. Second Edition. Melbourne, Inkata Press.

McGarity, J. W. and R. R. Storrier (1986). Fertilizers. Australian Soils The Human Impact. J. S. Russell and R. F. Isbell. St Lucia, University of Queensland Press: 305-333.

McIntosh, R. (1982). "Effects of different forms and rates of nitrogen fertilizer on the growth of Lodgepole pine." Forestry **55**(1): 61-68.

McLaren, R. G. and K. C. Cameron (1996). Soil Science: Sustainable production and environmental protection. Auckland, Oxford University Press.

McLaughlin, J. W., M. R. Gale, *et al.* (2000). "Soil organic matter and nitrogen cycling in response to harvesting, mechanical site preparation and fertilisation in a wetland with a mineral substrate." Forest Ecology and Management **129**: 7-23.

McMurtie, D. A. Rook, *et al.* (1990). "Modelling the yield of *Pinus radiata* on a site limited by water and nitrogen." Forest Ecology and Management **30**: 381-413.

McMurtrie and J. J. Landsberg (1992). "Using a simulation model to evaluation the effects of water and nutrients on the growth and carbon partitioning of *Pinus radiata*." Forest Ecology and Management **52**: 243-260.

Mead, D. J. and W. L. Pritchett (1975a). "Fertilizer movement in a slash pine ecosystem." Plant and Soil **43**: 451-465.

Mead, D. J. and W. L. Pritchett (1975b). "Fertilizer movement in a slash pine ecosystem. II. N distribution after two growing seasons." Plant and Soil **43**(2): 467-478.

Melillo, J. M., J. D. Aber, *et al.* (1982). "Nitrogen and lignin control of hardwood leaf litter decomposition dynamics." Ecology **63**(3): 621-626.

Melillo, J. M., J. D. Aber, *et al.* (1989). "Carbon and nitrogen dynamics along the decay continuum: Plant litter to soil organic matter." Plant and Soil **115**: 189-198.

Mendham, D. S., P. J. Smethurst, *et al.* (2002). "Soil analyses as indicators of phophorus response in young eucalypt plantations." Soil Science Society of America Journal **66**: 959-968.

Menge, J. A., L. F. Grand, *et al.* (1977). "The effect of fertilization on growth and mycorrhizae numbers in 11-year-old Loblolly pine plantations." Forest Science **23**(1): 37-44.

Messina, M. G. (1992). "Response of *Eucalyptus regnans* F. Muell. to thinning and urea fertilization in New Zealand." Forest Ecology and Management **51**: 269-283.

Metson, A. J. (1974). Methods of chemical analysis for soil survey samples. New Zealand Department of Scientific and Industrial Research, Bulletin 12. Wellington, Government Print: 207p.

Miller, H. G., J. M. Cooper, *et al.* (1979). "Nutrient cycling in pines and their adaptation to poor soils." Canadian Journal of Forest Research **9**: 19-26.

Miller, H. G. (1981). "Forest fertilization: Some guiding concepts." Forestry **54**(2): 157-168.

Moroni, M. T., P. J. Smethurst, *et al.* (2002). "Nitrogen fluxes in surface soils of young *Eucalyptus nitens* plantations in Tasmania." Australian Journal of Soil Research **40**: 543-553.

Morrison, I. K. and N. W. Foster (1977). "Fate of urea fertilizer added to a boreal forest *Pinus banksiana* Lamb. Stand." Soil Science Society of America Journal **41**: 441-448.

Murphy, D. V., G. P. Sparling, *et al.* (1998a). "Stratification of microbial biomass C and N and gross N mineralization with soil depth in two contrasting Western Australian agricultural soils." Australian Journal of Forest Research **36**: 45-55.

Murphy, D. V., G. P. Sparling, *et al.* (1998b). "Mineralisation of soil organic nitrogen and microbial respiration after simulated summer rainfall events in an agricultural soil." Australian Journal of Forest Research **36**: 231-246.

Murphy, J. and J. P. Riley (1962). "A modified single solution method for the determination of phosphate in natural waters." Analytica Chimica Acta **27**: 31-36.

Nadelhoffer, K. J., J. D. Aber, *et al.* (1982). "Leaf-litter production and soil organic matter dynamics along a nitrogen-availability gradient in Southern Wisconsin (U.S.A)." Canadian Journal of Forest Research **13**: 12-21.

Nadelhoffer, K. J., J. D. Aber, *et al.* (1983). "Leaf production and soil organic matter dynamics along a nitrogen availability gradient in south Wisconsin (USA)." Canadian Journal of Forest Research **13**: 12-21.

Nadelhoffer, K. J. and J. D. Aber (1984). "Seasonal patterns of ammonium and nitrate uptake in nine temperate forest ecosystems." Plant and Soil **80**: 321-335.

Nadelhoffer, K. J. (1990). "Microlysimeter for measuring nitrogen mineralization and microbial respiration in aerobic soil incubations." Soil Science Society of America Journal **54**(2): 411-415.

Nadelhoffer, K. J., A. E. Giblin, *et al.* (1991). "Effects of temperature and substance quality on element mineralization in six artic soils." Ecology **72**(1): 242-253.

Nason, G. E., D. J. Pluth, *et al.* (1990). "Dynamics of foliar N in Douglas-fir after spring and fall applications of ammonium nitrate and urea." Canadian Journal of Forest Research **20**: 1515-1523.

Neilsen, W. A. (1983). Use of fertilisers in establishing and growing coniferous plantations in Australia. Proceedings, Sept. 1982. pp 79-92. Workshop Meeting on Establishment of Coniferous Plantations, Mt.Gambier, South Australia.

Neilsen, W. A. and G. R. Davis (1984). Environmental Impact Statement on Tasmanian Woodchip Exports Beyond 1988. Tasmanian Forest Soils Working Paper 4. Tasmanian Soils. Hobart: 39.

Neilsen, W. A., G. R. Davis, *et al.* (1984). "Growth and nutrient uptake of *Pinus radiata* seedlings over the first 3 years following treatment with nitrogen and phosphorus fertilizers." Australian Forest Research **14**: 1-10.

Neilsen, W. A., W. Pataczek, *et al.* (1992). "Growth response of *Pinus radiata* to multiple applications of nitrogen fertilizer and evaluation of the quality of added nitrogen remaining in the forest system." Plant and Soil **144**: 207-217.

Neilsen, W. A. (1996). "Response of *Eucalyptus nitens* and *Eucalyptus regnans* seedlings to application of various fertilizers at planting and soon after." New Zealand Journal of Forestry Science **26**: 355-369.

Neilsen, W. A. and T. Lynch (1998). "Implications of pre- and post-fertilizing changes in growth and nitrogen pools following multiple applications of nitrogen fertilizer to *Pinus radiata* stand over 12 years." Plant and Soil **202**: 295-307.

Neilsen, W. A. and P. D. Dredge (1999). Magnesium deficiency in forestry Tasmania plantations. Forest Research Working Group 3, Rotorua, New Zealand.

Neilsen, W. A., C. Ringrose, *et al.* (2002). Profile Descriptions. Hobart, Forestry Tasmania: 1-21.

Nohrstedt, H.-O. (1990). "Effects of repeated nitrogen fertilization with different doses on soil properties in a *Pinus sylvesteris* stand." Scandinavian Journal of Forestry Research **5**: 3-15.

Nohrstedt, H.-O., S. Jacobson, *et al.* (2000). "Effects of repeated Urea doses on soil chemistry and nutrient pools in a Norway Spruce stand." Forest Ecology and Management **130**: 47-56.

Nzila, J. D., J.-P. Bouillet, *et al.* (2002). "The effects of slash management on nutrient cycling and tree growth in *Eucalyptus* plantations in the Congo." Forest Ecology and Management **171**: 209-221.

O'Connell, A. M., T. S. Grove, *et al.* (1978). "Nutrients in the Jarrah forest soils." Australian Journal of Ecology **3**: 253-260.

O'Connell, A. M. and P. M. A. Menage (1982). "Litter fall and nutrient cycling in karri (*Eucalyptus diversicolor* F. Muell.) forest in relation to stand age." Australian Journal of Ecology **7**: 49-62.

O'Connell, A. M. and T. S. Grove (1993). "Influence of nitrogen and phosphorous fertilizers on amount and nutrient content of litterfall in a regrowth eucalypt forest." New Forests **7**: 33-47.

O'Connell, A. M. and S. J. Rance (1999). "Predicting nitrogen supply in plantation eucalypt forests." Soil Biology and Biochemistry **31**: 1943-1951.

Page, K. L., R. C. Dalal, *et al.* (2003). "Subsoil nitrogen mineralization and its potential to contribute to NH_4 accumulation in a Vertosol." Australian Journal of Soil Research **41**: 119-126.

Parfitt, R. L., D. J. Ross, *et al.* (1998). "Nitrogen and Phosphorus mineralization in *Pinus radiata* harvest residue samples from coastal sand." New Zealand Journal of Forestry Science **28**(3): 347-360.

Parton, W. J., D. S. Schimel, *et al.* (1987). "Analysis of factors controlling soil organic matter levels in Great Plains grasslands." Soil Science Society of America Journal **51**: 1173-1179.

Pastor, J., J. D. Aber, *et al.* (1984). "Aboveground production and N and P cycling along a nitrogen mineralization gradient on Blackhawk Island, Wisconsin." Ecology **65**(1): 256-268.

Pastor, J., R. H. Gardner, *et al.* (1987). "Successional changes in nitrogen availability as a potential factor contributing to spruce declines in boreal North America." Canadian Journal of Forest Research **17**(10-12): 1394-1400.

Paul, E. A. and N. G. Juma (1981). "Mineralization and immobilization of soil nitrogen by micro-organisms." Ecological Bull. **33**: 179-195.

Paul, K. I., A. S. Black, *et al.* (1999). "Influence of moist-dry cycles on pH changes in surface soils." Australian Journal of Soil Research **37**: 1057-1072.

Paul, K. I. (2001). Temperature and moisture effects on decomposition. Net Ecosystem Exchange, Canberra, CRC for Greenhouse Accounting.

Paul, K. I., P. J. Polglase, *et al.* (2002). "Soil nitrogen availability predictor (SNAP): a simple model for predicting mineralization of nitrogen in forest soils." Australian Journal of Soil Research **40**: 1011-1026.

Paul, K. I., A. S. Black, *et al.* (2003a). "Development of acidic subsurface layers of soil under various management systems." Advances in Agronomy **78**: 187-214.

Paul, K. I., P. J. Polglase, *et al.* (2003b). "Defining the relation between soil water content and net nitrogen mineralisation." European Journal of Soil Science **54**: 39-47.

Paul, K. I. and P. J. Polglase (2004a). "Prediction of decomposition of litter under eucalypts and pines using the FullCAM model." Forest Ecology and Management **191**: 73-92.

Paul, K. I. and P. J. Polglase (2004b). "Calibration of the RothC model to turnover of soil carbon under eucalypts and pines." Australian Journal of Soil Research **42**: 883-895.

Paustain, K., G. I. Agren, *et al.* (1997). Modelling litter quality effects on decomposition and soil organic matter dynamics. Driven by Nature: Plant Litter Quality and Decomposition. G. Cadisch and K. E. Giller. Cambridge, CAN INTERNATIONAL: 313-336.

Persson, H., Y. Von Fircks, *et al.* (1995). "Root distribution in a Norway spruce (Picea abies (L.) Karst.) stand subjected to drought and ammonium-sulphate application." Plant and Soil **168-169**: 161-165.

Piatek, K. B. and H. L. Allen (1999). "Nitrogen mineralisation in a pine plantation fifteen years after harvesting and site preparation." Soil Science Society of America Journal **63**: 990-998.

Piatek, K. B. and H. L. Allen (2001). "Are forest floors in mid-rotation stands of loblolly pine (*Pinus taeda*) a sink for nitrogen and phosphorus?" Canadian Journal of Forest Research **31**: 1164-1174.

Plymale, A. E., R. E. J. Boerner, *et al.* (1987). "Relative nitrogen mineralization and nitrification in soils of two contrasting hardwood forests: Effects of site microclimate and initial soil chemistry." Forest Ecology and Management **21**: 21-36.

Polglase, P. J., P. M. Attiwill, *et al.* (1992a). "Nitrogen and phosphorus cycling in relation to stand age of *Eucalyptus regnans* F. Muell II. N mineralization and nitrification." Plant and Soil **142**: 167-176.

Polglase, P. J., N. B. Comerford, *et al.* (1992b). "Mineralization of nitrogen and phosphorous from soil organic matter in southern pine plantations." Soil Science Society of America Journal **56**: 921-927.

Pook, M. J. and W. F. Budd (2002). An evaluation of cloud seeding operations carried out in Tasmania by Hydro Tasmania. An Independent Report to Hydro Tasmania, Department of Primary Industries, Water and Environment and The Tasmanian Farms and Graziers Association. Hobart: 1-39.

Porter, W. M., C. D. A. McLay, *et al.* (1995). Rates and sources of acidification in agricultural systems of southern Australia. Third International Symposium on Plant-Soil Interactions at Low pH, Brisbane, Kluwer Academic Publishers.

Powers, R. F. (1990). "Nitrogen mineralization along an altitudinal gradient: Interactions of soil temperature, moisture and substrate quality." Forest Ecology and Management **30**: 19-29.

Prasolova, N. V., Z. H. Xu, *et al.* (2000). "Spatial-temporal variability of soil moisture, nitrogen availability indices and other chemical properties in hoop pine (*Araucaria cunninghamii*) plantations of subtropical Australia." Forest Ecology and Management **136**: 1-10.

Prescott, C. E., M. A. McDonald, *et al.* (1993). "Availability of N and P in the forest floors of adjacent stands of western red cedar-western hemlock and western hemlock-ambilis fir on northern Vancouver Island." Canadian Journal of Forest Research **23**: 605-610.

Prescott, C. E. and C. M. Preston (1994). "Nitrogen mineralization and decomposition in forest floors in adjacent plantations of western red cedar, western hemlock and Douglas-fir." Canadian Journal of Forest Research **24**: 2424-2431.

Prescott, C. E., L. Vesterdal, *et al.* (2000). "Nutrient concentrations and nitrogen mineralization in forest floors of single species conifer plantations in coastal British Columbia." Canadian Journal of Forest Research **30**: 1341-1352.

Pritchett, W. L. and N. B. Comerford (1982). "Long-term response to phosphorus fertilization on selected southeastern coastal plain soils." Soil Science Society of America Journal **46**: 640-644.

Pulleman, M. and A. Tietema (1999). "Microbial C and N transformations during drying and rewetting of coniferous forest floor material." Soil Biology and Biochemistry **31**: 275-285.

Raison, R. J. (1980). "Possible forest site deterioration associated with slash-burning." Search **11**(3): 68-72.

Raison, R. J., M. J. Connell, *et al.* (1987). "Methodology for studying fluxes of soil mineral-N *in situ*." Soil Biology and Biochemistry **19**(5): 521-530.

Raison, R. J., P. K. Khanna, *et al.* (1990). "Effects of water availability and fertilization on nitrogen cycling in a stand of *Pinus radiata*." Forest Ecology and Management **30**: 31-43.

Raison, R. J. and M. J. Connell (1992). Collaborative thinning and fertilization research in east Gippsland: background and description of studies established in 1992 under national estate funding. Victoria, CSIRO Division of Forestry.

Raison, R. J., M. J. Connell, *et al.* (1992). "Effects of irrigation and nitrogen fertilisation on fluxes of soil mineral nitrogen in a stand of *Pinus radiata*." Forest Ecology and Management **52**: 43-64.

Raison, R. J. and B. J. Myers (1992). "The Biology of forest growth experiment: linking water and nitrogen availability to the growth of *Pinus radiata*." Forest Ecology and Management **52**: 279-308.

Rayment, G. E. and F. R. Higginson (1992). Australian Laboratory Handbook of Soil and Water Chemical Methods. Melbourne, Inkata Press.

Reuter, D. J. and J. B. Robinson (1997). Plant analysis: an interpretation manual. Collingwood, Victoria, CSIRO Publishing.

Ribeiro, C., M. Madeira, *et al.* (2002). "Decomposition and nutrient release from leaf litter of Eucalyptus globulus grown different water and nutrient regimes." Forest Ecology and Management **171**: 31-41.

Richards, B. N., J. E. N. Smith, *et al.* (1985). "Mineralization of soil nitrogen in three forest communities from New England region of New South Wales." Australian Journal of Ecology **10**(4): 429-441.

Robertson, K., J. Schnurer, *et al.* (1988). "Microbial biomass in relation to C and N mineralization during laboratory incubations." Soil Biology and Biochemistry **20**(3): 281-286.

Ross, D. J., G. P. Sparling, *et al.* (1995). "Microbial biomass C and N, and mineralizable-N, in litter and mineral soil under Ion a coastal sand: Influence of stand age and harvest management." Plant and Soil **175**: 167-177.

Ross, D. J., N. A. Scott, *et al.* (2001). "Root effects on soil carbon and nitrogen cycling in a Pinus radiata D.Don plantation on a coastal sand." Australian Journal of Soil Research **39**: 1027-1039.

Rosswall, T. (1982). "Microbial regulation of the biogeochemical nitrogen cycle." Plant and Soil **67**: 15-34.

Running, S. W. and J. Coughlan (1988). "A general model of forest ecosystem processes for regional applications. 1. Hydrologic balance, canopy gas exchange and primary production processes." Ecological Modelling **42**: 125-154.

Ryan, P. J., N. J. McKenzie, *et al.* (2000). "Intergrating forest soils information across scales: spatial prediction of soil properties under Australian forests." Forest Ecology and Management **138**: 139-157.

Schlesinger, W. H. and M. M. Hasey (1981). "Decomposition of Chaparral shrub foliage: Losses of organic and inorganic constituents from deciduous and evergreen leaves." Ecology **62**(3): 762-774.

Schoenholtz, S. H., H. Van Miegroet, *et al.* (2001). "A review of chemical and physical properties as indicators of forest soil quality: challenges and opportunities." Forest Ecology and Management **138**: 335-356.

Schönau, A. P. G. and M. A. Herbert (1989). "Fertilizing eucalypts at plantation establishment." Forest Ecology and Management **29**: 221-244.

Scott, N. A. and D. Binkley (1997). "Foliage litter quality and annual net N mineralization: comparison across North American forest sites." Oecologia **111**: 151-159.

Seneviratne, R. (1985). "Effect of mild drying on the mineralization of soil nitrogen." Plant and Soil **84**: 175-179.

Sierra, J. (1992). "Relationship between microbial N content and N mineralization rate in disturbed and undisturbed soil samples incubated under field and laboratory conditions." Australian Journal of Forest Research **30**: 477-492.

Skene, T. M., J. O. Skemstad, *et al.* (1997). "The influence of inorganic matrices on the decomposition of *Eucalyptus* litter." Australian Journal of Soil Research **35**: 73-87.

Skinner, M. F. and P. M. Attiwill (1981). "The productivity of pine plantations in relation to previous land use. I. Growth responses in agricultural and forest soils." Plant and Soil **60**: 161-176.

Smethurst, P. J. and E. K. Nambiar (1989a). "Role of weeds in the management of N in a young Pinus radiata plantation." New Forests **3**: 203-224.

Smethurst, P. J. and E. K. S. Nambiar (1989b). "An appraisal of the *in situ* soil-core technique for measuring nitrogen uptake by a young *Pinus radiata* plantation." Soil Biology and Biochemistry **21**(7): 939-942.

Smethurst, P. J. and E. K. S. Nambiar (1990a). "Effects of slash and litter management on fluxes of N and tree growth in a young *Pinus radiata* plantation." Canadian Journal of Forest Research **20**: 1489-1506.

Smethurst, P. J. and E. K. S. Nambiar (1990b). "Distribution of carbon and nutrients and fluxes of mineral nitrogen after clear-felling a *Pinus radiata* plantation." Canadian Journal of Forest Research **20**: 1490-1497.

Smethurst, P. J. and E. K. S. Nambiar (1995). "Changes in soil C and N during the establishment of a second crop of *Pinus radiata*." Forest Ecology and Management **73**: 145-155.

Smethurst, P. J., M. A. Line, *et al.* (1998). "Soil microbial biomass and activity in two eucalypt plantation soils after fertilisation." Tasforests **10**: 69-73.

Smethurst, P. J., A. M. Herbert, *et al.* (2001). "Fertilization effects on soil chemistry in three eucalypt plantations." Soil Science Society of America Journal **65**: 795-804.

Smethurst, P. J., C. Baillie, *et al.* (2003). "Fertilizer effects on LAI and growth of four *Eucalyptus nitens* plantations." Forest Ecology and Management **176**: 631-542.

Smethurst, P. J., G. K. Holz, *et al.* (2004a). "Nitrogen management in *Eucalyptus nitens* plantations." Forest Ecology and Management **19**: 63-80.

Smethurst, P. J., D. S. Mendham, *et al.* (2004b). Simultaneous prediction of nitrogen and phosphorus dynamics in a Eucalyptus nitens plantation using linked CABALA and PCATS models. Eucalyptus in a changing world. International IUFRO Conference, Aveiro.

Smith, C. T., W. J. Dyck, *et al.* (1994). "Nutrition and productivity of *Pinus radiata* following harvest disturbance and fertilization of coastal sand dunes." Forest Ecology and Management **66**: 5-38.

Smith, C. T., A. T. Lowe, *et al.* (2000). "Response of radiata pine forests to residue management and fertilisation across a fertility gradient in New Zealand." Forest Ecology and Management **138**: 203-233.

Smolander, A., O. Priha, *et al.* (1998). "Nitrogen and carbon transformations before and after clear-cutting in repeatedly N-fertilised and limed forest soils." Soil Biology and Biochemistry **30**(4): 477-490.

Smolander, A., L. Paavolainen, *et al.* (2000). "C and N transformations in forest soil after mounding for regeneration." Forest Ecology and Management **134**: 17-28.

Snowdon, P. and M. L. Benson (1992). "Effects of combinations of irrigation and fertilisation on the growth and above-ground biomass production of *Pinus radiata*." Forest Ecology and Management **52**: 87-116.

Snowdon, P. (2002). "Modeling Type 1 and Type 2 growth responses in plantations after application of fertilizer or other silvicultural treatments." Forest Ecology and Management **163**: 229-244.

Sollins, P., G. Spycher, *et al.* (1984). "Net nitrogen mineralization from light- and heavy-fraction forest soil organic matter." Soil Biology and Biochemistry **16**(1): 31-37.

Sommers, L. E., C. M. Gilmour, *et al.* (1981). The Effect of Water Potential on Decomposition Processes in Soils. Water Potential Relations in Soil Microbiology. J. F. Parr, W. R. Gardener and L. F. Elliot. Wisconsin, Soil Science Society of America. **9:** 97-118.

Soulides, D. A. and F. E. Allison (1961). "Effect of drying and freezing soils on carbon dioxide production, available mineral nutrients, aggregation, and bacterial population." Soil Science **91**(5): 291-298.

Spurr, S. H. and B. V. Barnes (1980). Forest Ecology. New York, John Wiley & sons.

Squire, R. O. and D. W. Flinn (1981). Site disturbance and nutrient economy of plantations with special reference to radiata pine on sands. Australia, Australian Forestry Council: Proceedings, Australian Forest Nutrition Workshop. Productivity in perpetuity. Canberra, Australia, CSIRO Division of Forest Research.

Stace, H. C. T., G. D. Hubble, *et al.* (1968). A handbook of Australian soils. Glenside, South Australia, Rellim Technical Publications.

Stanford, G. and S. J. Smith (1972). "Nitrogen mineralization potentials of soils." Soil Science Society of America Proc. **36**: 465-472.

Stevenson, F. J. (1982). Humus Chemistry; Genisis, Composition, Reactions. New York, John Wiley & Sons.

Stevenson, I. L. (1956). "Some observations on the microbial activity in remoistened air-dried soils." Plant and Soil **8**(2): 170-182.

Strader, R. H. and D. Binkley (1989). "Mineralization and immobilization of soil nitrogen in two Douglas-fir stands 15 and 22 years after nitrogen fertilization." Canadian Journal of Forest Research **19**: 798-801.

Strong, D. T., P. W. G. Sale, *et al.* (1998a). "The influence of the soil matrix on nitrogen mineralisation and nitrification. I. Spatial variation and a hierarchy of soil properties." Australian Journal of Soil Research **36**(3): 429-447.

Strong, D. T., P. W. G. Sale, *et al.* (1998b). "The influence of the soil matrix on nitrogen mineralisation and nitrification. II. The pore system as a framework for mapping the organisation of the soil matrix." Australian Journal of Soil Research **36**: 855-872.

Strong, D. T., P. W. G. Sale, *et al.* (1999a). "The influence of the soil matrix on nitrogen mineralization and nitrification V. Microporosity and manganese." Australian Journal of Soil Research **37**: 345-355.

Strong, D. T., P. W. G. Sale, *et al.* (1999b). "The influence of the soil matrix on nitrogen mineralisation and nitrification. III. Predictive utility of traditional variables and process location within the pore system." Australian Journal of Soil Research **37**: 137-49.

Stump, L. M. and D. Binkley (1993). "Relationships between litter quality and nitrogen availability in Rock Mountain forests." Canadian Journal of Forest Research **23**: 492-502.

Switzer, G. L. and L. E. Nelson (1972). "Nutrient accumulation and cycling in lobby pine (*pinus taeda* L.) Plantation ecosystems: The first 20 years." Soil Science Society of America Proc. **36**.

Tabatabai, M. A. and A. A. Al-Khafaji (1980). "Comparison of nitrogen and sulfur mineralization in soils." Soil Science Society of America Journal **44**: 1000-1006.

Tamm, C. O. and B. Popovic (1995). "Long-term field experiments simulating increased decomposition of sulphur and nitrogen to forest plots." Ecological Bull. **44**: 301-321.

Taylor, B. R., D. Parkinson, *et al.* (1989). "Nitrogen and lignin content as predictors of litter decay rates: A microcosm test." Ecology **70**(1): 97-104.

Theodorou, C. and G. D. Bowen (1981). "Microbial Studies of First- and Second-Rotation Pine Forest Soils in South Australia." Australian Forest Research **11**: 129-39.

Theodorou, C. and G. D. Bowen (1983a). "Effects of Temperature, Moisture and Litter on Nitrogen Mineralization in *Pinus radiata* Forest Soils." Australian Forest Research **13**: 113-119.

Theodorou, C. and G. D. Bowen (1983b). "Nitrogen Transformations in First- and Second-rotation *Pinus radiata* Forest Soils." Australian Forest Research **13**: 103-112.

Theodorou, C. (1990). "Nitrogen transformations in particle size fractions from a second rotation pine forest soils." Communications in Soil Science and Plant Analysis **21**(5&6): 407-413.

Theodorou, C. and G. D. Bowen (1990). "Effects of fertilizer on litterfall and N and P release from decomposing litter in a *Pinus radiata* plantation." Forest Ecology and Management **32**: 87-102.

Thomas, K. D. and C. E. Prescott (2000). "Nitrogen availability in forest floors of three tree species on the same site: the role of litter quality." Canadian Journal of Forest Research **30**: 1698-1706.

Thomas, R. C. and D. J. Mead (1992a). "Uptake of nitrogen by *Pinus radiata* and retention within the soil after applying ^{15} N-labelled Urea at different

frequencies. 1. Growth response and nitrogen budgets." Forest Ecology and Management **53**: 131-151.

Thomas, R. C. and D. J. Mead (1992b). "Uptake of nitrogen by *Pinus radiata* and retention within the soil after applying ^{15} N-labelled Urea at different frequencies. 2. Seasonal trends and processes." Forest Ecology and Management **53**: 153-174.

Tiessen, H. and J. W. B. Stewart (1983). "Particle-size fractions and their use in studies of soil organic matter: II Cultivation effects on organis matter compostion in size fractions." Soil Science Society of America Journal **47**: 509-514.

Tietema, A., B. Warmerdam, *et al.* (1992). "Abiotic factors regulating nitrogen transformations in the organic layer of acid forest soils: Moisture and pH." Plant and Soil **147**: 69-78.

Trinsoutrot, S., C. Recous, *et al.* (2000). "Biochemical quantity of crop residues and carbon and nitrogen mineralization kinetics under non-limiting nitrogen conditions." Soil Biology and Biochemistry **64**: 918-926.

Turner, J. and M. J. Lambert (1983). "Nutrient cycling within a 27-year-old *Eucalyptus grandis* plantation in New South Wales." Forest Ecology and Management **6**: 155-168.

Turner, J. and M. J. Lambert (1986). "Fate of applied nutrients in a long-term superphosphate trial in *Pinus radiata*." Plant and Soil **93**: 373-382.

Turner, J., M. Lambert, *et al.* (2002). "Continuing growth responses to phosphate fertilizers by a *Pinus radiata* plantation over fifty years." Forest Science **48**(3): 556-568.

Turvey, N. D. and P. J. Smethurst (1994). "Nutrient concentrations in foliage, litter and soil in relation to wood production of 7- to 15-year-old Pinus radiata in Victoria, Australia." Australian Forestry **57**(4): 157-164.

Upadhyay, V. P. and B. R. Singh (1985). "Nitrogen dynamics of decomposing hardwood leaf litter in a central Himalayan forest." Soil Biology and Biochemistry **17**(6): 827-830.

Usman, S., P. H. Singh, *et al.* (2000). "Fine root decomposition and nitrogen mineralisation patterns in *Quercus leucotrichophora* and *Pinus roxburghii* forests in central Himalaya." Forest Ecology and Management **131**: 191-199.

Van Cleve, K., R. Barney, *et al.* (1981). "Evidence of temperature control of production and nutrient cycling in two interior Alaska black spruce ecosystems." Canadian Journal of Forest Research **11**: 258-273.

Van Cleve, K., O. W. Heal, *et al.* (1986). "Bioassay of forest floor nitrogen supply for plant growth." Canadian Journal of Forest Research **16**: 1320-1326.

Van Gestel, M., J. N. Ladd, *et al.* (1991). "Carbon and nitrogen mineralization from two soils of contrasting texture and microaggregate stability: Influence of sequential fumigation, drying and storage." Soil Biology and Biochemistry **23**(4): 313-322.

Van Gestel, M., R. Merckx, *et al.* (1993). "Microbial biomass responses to soil drying and rewetting: The fate of fast- and slow-growing micro-organisms in soils from different climates." Soil Biology and Biochemistry **25**(1): 109-123.

Verburg, P. S. J. and N. Van Breemen (2000). "Nitrogen transformations in a forest catchment in Southern Norway subjected to elevated temperature and CO_2." Forest Ecology and Management **129**: 31-39.

Vestgarden, L. S., G. Abrahamsen, *et al.* (2001). "Soil solution response to nitrogen and magnesium application in a Scots pine forest." Soil Science Society of America Journal **65**: 1812-1823.

Vitousek, P. M., J. R. Gosz, *et al.* (1982). "A comparative analysis of potential nitrification and nitrate mobility in forest ecosystems." Ecological Monographs **52**(2): 155-177.

Vitousek, P. M. and P. A. Matson (1985). "Disturbance, nitrogen availability, and nitrogen losses in an intensively managed loblolly pine plantation." Ecology **66**(4): 1360-1367.

Wang, B., P. J. Smethurst, *et al.* (1998). "Nitrogen fluxes in surface soils of 1-2 year old eucalypt plantations in Tasmania." Australian Journal of Soil Research **36**: 17-29.

Ward, S. C., G. E. Pickersgill, *et al.* (1985). "Responses to factorial combinations of nitrogen, phosphorous and potassium fertilizers by saplings of *Eucalyptus saligna* Sm., and the prediction of the responses by DRIS Indices." Australian Forest Research **15**: 27-32.

Waring, H. D. (1969). "The role of nitrogen in the maintenance of productivity in conifer plantations." Commonwealth Forestry Review **48**(135): 226-236.

Waring, H. D. (1972). *Pinus radiata* and the nitrogen-phosphorus interaction. Australian Forestry and Tree Nutrition, Canberra, Forestry and Timer Bureau.

Waring, H. D. (1981). Forest fertilisation in Australia: early and late. Australia, Australian Forestry Council: Proceedings, Australian Forest Nutrition Workshop. Productivity in perpetuity, Canberra, Australia, Canberra, Australia: CSIRO Division of Forest Research.

Waring, R. H. and W. H. Schlesinger (1985). Forest Ecosystems Concepts and Management. Orlando, Florida, Academic Press, Inc

Harcourt Brace Jovanovich, Publishers.

Watmough, S. A. and P. J. Dillon (2003). "Base cation and nitrogen budgets for seven forested catchments in central Ontario, 1983-1999." Forest Ecology and Management **177**: 155-177.

Webber, B. and H. A. I. Madgwick (1983). "Biomass and nutrient content of a 29-year-old *Pinus radiata* stand." New Zealand Journal of Forestry Science **13**(2): 222-228.

Weinstein, D. A., R. M. Beloin, *et al.* (1991). "Modeling changes in red spruce carbon balance and allocation in response to interacting ozone and nutrient stresses." Tree Physiology **9**: 127-146.

West, A. W., G. P. Sparling, *et al.* (1988). "Dynamics of microbial C, N-flush and ATP, and enzyme activities of gradually dried soils from a climosequence." Australian Journal of Soil Research **26**: 519-530.

Weston, C. J., P. M. Attiwill, *et al.* (1991). Growth and nutrition of eucalypt plantations in relation to soil type and former land use in Gippsland, Victoria. IUFRO Symposium on Intensive Forestry: The role of Eucalypts. A. P. G. Schönau. Durban, South Africa, Southern African Institute of Forestry. **1:** 480-491.

Weston, C. J. (2001). An overview of recent nutrition research for the rapidly expanding *Eucalyptus* plantation industry in Australia. Creswick, University of Melbourne, School of Forestry**:** 1-31.

Wetselaar, R. (1968). "Soil organic nitrogen mineralization as affected by low water potentials." Plant and Soil **29**(1): 9-17.

Whynot, T. W. and G. F. Weetman (1991). "Repeated fertilization effects on nitrogen fluxes measured by sequential coring." Soil Science Society of America Journal **55**: 1101-1111.

Wilkinson, G. R. and W. A. Neilsen (1995). "Implications of early browsing damage on the long term productivity of eucalypt forests. For. Ecol. and Management 74, 117-24." Forest Ecology and Management **74**: 117-124.

Williams, B. L. (1972). "Nitrogen mineralization and organic matter decomposition in Scots pine humus." Forestry **45**(2): 177-188.

Wise, P. K. and M. G. Pitman (1981). "Nutrient removal and replacement associated with short-rotation eucalypt plantations." Australian Forestry **44**(3): 142-152.

Woolons, R. C. and G. M. Will (1975). "Increasing growth in high production radiata pine stands by nitrogenous fertilisers." New Zealand Journal of Forestry **20**: 243-253.

Yin, X. (1992). "Empirical relationships between temperature and nitrogen availability across North American forests." Canadian Journal of Forest Research **22**: 707-712.

Zak, D. R., W. E. Holmes, *et al.* (1999). "Soil temperature, matric potential, and the kinetics of microbial respiration and nitrogen mineralisation." Soil Science Society of America Journal **63**: 575-584.