

**Nitrogen mineralisation and availability
under kikuyu dairy pastures
on the mid-north coast of NSW**

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Literature review for

Dairy Australia and Hunter Local Land Services

Project C100001006

Fertiliser use efficiency in high input dairy pastures

Final Version – March 6 2015

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1. Executive summary

Dairy farming makes a significant contribution to the economy of the mid-north coast of New South Wales with a farm gate value of \$60 million in 2007. The majority of dairy farms are situated on alluvial soils on the lower flood plains with others based on creek flats in the middle catchments and on the fertile ferrosols of the Comboyne Plateau. The region is located in the transition between the tropical and temperate climatic zones and experiences wet, hot summers, wet autumns and relatively dry, mild winters. In some years flooding leaves paddocks waterlogged for significant periods, especially on the lower flood plains.

Kikuyu (*Pennisetum clandestinum*) forms the main pasture base for dairy farms on the mid-north coast. It is usually oversown with ryegrasses (*Lolium spp.*) in autumn to maintain pasture productivity and whole milk production during the cooler months when kikuyu becomes dormant. An autumn feed gap is created when kikuyu paddocks are taken out of rotation for the establishment of ryegrass. Nitrogen (N) supply is integral to pasture establishment and growth. On the mid-north coast the majority of N fertiliser is applied to forage ryegrass with little or no application on kikuyu due to inconsistent responses from variable rainfall and growth exceeding demand when conditions are favourable. A yield response to N fertiliser in autumn comes at the expense of successful ryegrass establishment.

Not applying N to kikuyu creates another set of problems. Rapid kikuyu growth leads to the build up of large amounts of organic matter, and thus organic carbon (C), under the sward with depleting levels of N. As decomposition of kikuyu residue proceeds, applied N fertiliser can become temporarily unavailable to establishing ryegrass. Unless fertiliser recommendations are developed from sufficient local trials that accurately simulate the grazed kikuyu-ryegrass system they may not allow for this 'tie-up' of N.

Despite the dominance of kikuyu-ryegrass pastures on dairy farms on the mid-north coast of NSW and the importance of the soil N cycle underpinning growth, little research has been published on N transformations, interactions and availability between the two species. This literature review will investigate the soil N cycle, factors affecting the growth of kikuyu, the fate of N under the sward and its availability to establishing ryegrass pastures.

Soil nitrogen

Nitrogen is often the most limiting factor for plant growth. There is a complex N cycle with transformations between the atmosphere, soil, plants, animals and microorganisms and N must be 'fixed' into available forms for plant uptake. Nitrogen fixation by the *Rhizobium* bacteria, living in a symbiotic relationship with legumes, can potentially supply more than 300 kg N/ha/year to Australian pastures. However, the median content of legumes in dairy pastures is less than 10% with variable yields and there is an increasing problem with persistence. Dairy farmers are reducing their reliance on legumes and biological fixation has not been included as an input into the soil N cycle for this review.

Total N is the sum of all nitrogenous molecules in the soil. Organic matter from decomposing plants, animals and the soil biomass contributes 95 to 99% of total N in soils although more than 50% of this N is bound into humic molecules and is not readily available to plants. Grazing animals return a significant amount of organic matter and

nitrogenous compounds to the soil in manure and urine. A herd of 200 cows could potentially leave 56 kg N on their allocated strip of pasture each day.

Pastures return approximately the same amount of herbage to the soil as is removed by grazing or harvesting. Nitrogen is recycled both from the remobilisation and translocation of N from mature to new growth and from the senescence and decomposition of plant material. Roots make up more than half the dry matter yield of pastures, with increasing grazing pressure reducing the life span of individual roots from years to months. Higher N fertiliser rates and grazing intensities reduce the accumulation of dead material but damage from trampling and dung increase senescence.

Total N is one of the more common soil N tests. Soil samples from more than 100 paddocks across 12 dairy farms on the mid-north coast of NSW showed a mean total N concentration of 0.37% (0.19-0.77%). These soils contained 2470 to 10,010 kg N/ha in the top 10 cm. However, only 1 to 2% of this N will be mineralised to the plant available ions, ammonium (NH_4^+) and nitrate (NO_3^-), during a growing season. Unlike other major plant nutrients these N ions cannot be stored in the soil as they are both prone to leaching and NH_4^+ can be adsorbed onto clay minerals or bound into humic complexes.

Plants take up both NH_4^+ and NO_3^- , the relative amounts of each depending on the species and soil and environmental factors. Nitrate uptake is more common in well-aerated and acidic soils and results in a rise in soil pH. Ammonium uptake is more abundant in anaerobic soil conditions and lowers soil pH. Growth rates are maximised when both forms of mineral N are taken up by plants. The rate of N uptake by grasses varies widely depending on the time of year, stage of growth, temperature, moisture supply, fluctuations in N supply and grazing regime. It is greatest during the vegetative phase of growth.

The N content of most plant tissue ranges from 1 to 5%. Green herbage has a higher concentration of N than stems, roots and residue. Tropical C_4 grasses have lower levels of the photosynthetic enzyme rubisco, and hence N, in their leaves than temperate C_3 species. Nitrogen supply affects both the rate of tiller production in grasses and leaf size, the area of the canopy increasing with N supply until a critical point is reached and the maximum growth rate has been attained. The strategic use of N can increase the number of grazings in a rotational system.

The soil nitrogen cycle

Plants rely on transformation cycles within the soil to provide a continual supply of available N. Plant litter and other residues are first decomposed by a complex web of soil organisms. The rate of decomposition depends on the component parts with the degradation of water soluble compounds occurring within months whereas 70 to 80% of lignin can be retained after the first year of decomposition.

Following decomposition, nitrogenous compounds are converted into inorganic forms. Heterotrophic soil organisms produce an array of enzymes that break down macromolecules into simpler compounds. The functional amine groups ($-\text{NH}_2$) are then removed and converted to NH_4^+ in the process of ammonification. Nitrification occurs when NH_4^+ is oxidised to nitrite (NO_2^-) mostly by the chemoautotrophic *Nitrosomonas* bacteria. Further oxidation of NO_2^- to NO_3^- by *Nitrobacter spp.* completes the nitrification process. For this review the term N mineralisation will be used to encompass both ammonification and nitrification.

Heterotrophic soil organisms obtain their energy from decomposing residues with populations increasing rapidly as sources of organic C become available. They compete very effectively with plants for available N. Nitrogen is assimilated and immobilised into organic compounds in the biomass and becomes temporarily unavailable to plants until the death and decomposition of the microorganisms themselves.

Both mineralisation and immobilisation occur simultaneously in the soil with the net rate of each determined by the relative supply of C and N. In general, net mineralisation will occur if the C:N ratio of the decomposing substrate is less than 20:1 to 25:1, equivalent to an N content greater than 2.0 to 2.5% N. If the substrate has an N content less than 1.5%, the C:N ratio is greater than 30:1 and net immobilisation occurs. Between 20:1 and 30:1 there is an equilibrium between mineralisation and immobilisation.

The C:N ratios of plants vary depending on the species, stage of growth, plant part and N supply. The higher the protein content, the lower the C:N ratio. Pastures receiving little or no N fertiliser average C:N ratios of 25:1 in their shoots to 60:1 in their roots leading to immobilisation during the decomposition of their residues. As decomposition proceeds, the C:N ratio of the substrate narrows and eventually proceeds to net mineralisation. Adding high rates of N fertiliser during growth can also reduce these ratios although a proportion of this fertiliser could also be immobilised until the N requirement of the soil biomass is met.

The C:N ratio of existing soil organic matter must also be taken into consideration. Dairy paddocks on the mid-north coast of NSW have C:N ratios ranging from 8.8 to 15.4. Those with ratios below 11.0, the C:N ratio of humus, would be in a state of net mineralisation. Given suitable soil temperature and moisture and assuming no significant additions of decomposing substrate, these soils could produce from 50 to 200 kg mineral N each year. In contrast, net immobilisation would be occurring in those paddocks with wider C:N ratios and additions of N fertiliser would be first utilised by the microbial biomass before plants could access the remainder.

The rate of net mineralisation is determined by several factors:

Temperature: The optimum temperature and ranges of each step in the mineralisation of N depend largely on the microorganisms involved. An array of decomposing organisms is common in soils in the mesophilic temperature range of 0°C to 45°C. The rate of decay increases with temperature unless moisture becomes limiting. Ammonification also has a wide temperature range with increasing rates measured from 0°C to an optimum of 50°C before ceasing at 60°C. Nitrification has a much narrower range with optimum temperatures ranging from 25°C to 35°C, although nitrifying organisms may acclimatise to local conditions outside this range. Rapid temperature fluctuations can result in the death of large populations of soil microorganisms and an accompanying flush of mineral N.

Soil Moisture: Moisture stress can occur either when microbial growth is inhibited by a lack of water or too much water in the soil pores restricts aeration. At temperatures above 5°C the rate of N mineralisation increases from wilting point to field capacity. The activity of ammonifiers continues below wilting point, though that of nitrifiers is inhibited. Nitrifying bacteria can, however, survive in dry soils in an inactive form.

Oxygen availability: As soil moisture rises above field capacity, aerobic decomposition is gradually replaced by the fermentation of organic matter and the activity of soil microorganisms is depressed. Both aerobes and anaerobes carry out ammonification, which can proceed in waterlogged soils. However, nitrifiers are obligate aerobes, requiring an adequate supply of oxygen to oxidise NH_4^+ to NO_3^- . Hence, severe and prolonged

waterlogging results in soils with high levels of organic C and a build up of NH_4^+ whereas NO_3^- is the dominant form of mineral N in well-aerated soils.

Wetting and drying cycles: Alternate wetting and drying cycles, typically experienced in tropical and sub-tropical climates, enhance N mineralisation. The repeated wetting and drying of the soil breaks down water soluble aggregates and exposes new sections of substrate and soil to microbial attack. Drying partially sterilises the soil, releasing N compounds from the dead biomass. Drying also converts organic N compounds to more soluble forms that are readily utilised by the remaining soil microbes.

Soil pH: The diverse range of soil organisms responsible for decomposition and ammonification tolerate a wide range of soil pH conditions, although the rate of decomposition is greater in neutral than acid soils. Nitrification can take place in soils from $\text{pH}_{(w)}$ 4.5 to 10, but rates are restricted outside the optimum range of $\text{pH}_{(w)}$ 7 to 9. Given that other factors are not limiting, NH_4^+ tends to accumulate in acid soils. Nitrate becomes the dominant form of mineral N in neutral and alkaline soils though some nitrifiers may have adapted to acid conditions. The application of lime can increase mineralisation rates by 1 to 2%.

Nutrient deficiencies and toxicities: Nitrogen is usually the most limiting nutrient for the growth of soil organisms. Phosphorus can also become limiting but this does not often occur in dairy pastures. Increasing the concentration of mineral salts in the soil solution can increase the rate of ammonification and hence net mineralisation. Micronutrients can also have a stimulatory effect on decomposition and the N cycle when applied in trace amounts. However, in larger quantities a number of the trace elements can be toxic and inhibit N mineralisation, especially in acid soils or those low in clay and organic matter. The nitrifying bacteria are very sensitive to pesticides applied to the soil with a flush of N mineralisation can occur following the death of a portion of the biomass.

Soil type: Nitrogen mineralisation is more rapid in soils with a low clay and/or organic matter content than in heavier textured, high organic C soils. Clay minerals sorb organic compounds and form complexes with humic molecules. These microaggregates protect N compounds from the enzyme attack of the soil biomass.

Soil fauna and plants: Soil macrofauna accelerate the net rate of N mineralisation by increasing the surface area to volume ratio of litter and redistributing it through the soil profile, allowing for greater microbial attack. The excreta of earthworms and dung beetles also has a lower C:N ratio than the substrate consumed. Root exudates supply microorganisms with a ready supply of energy and may leave the rhizosphere in a state of net immobilisation as biomass populations increase until N becomes the limiting factor.

Kikuyu

Kikuyu was introduced to Australian pastures from the highland plateaux of central and east Africa in the 1920s. It is a perennial, warm-season, C_4 grass producing both rhizomes and stolons that elongate continuously. Leafy aerial tillers develop from these runners, which form a thick intertwining mat, with old tillers lodging and accumulating in the soil surface under the thatch. Common kikuyu and the seeded variety Whittet are the most widespread types. This review will focus on factors that affect kikuyu growth and N mineralisation and availability under the sward.

Soils and nutrients: Kikuyu developed in the fertile, volcanic African highlands and growth is limited on shallow, infertile soils. Preferring a well-drained soil it can tolerate some waterlogging and neither high salinity nor acidity affect yield. Substantial inputs of phosphorus are needed to establish new kikuyu pastures. Annual N fertiliser rates of 300-

500 kg N/ha in split dressings of 50-100 kg N/ha are recommended for maximum dry matter production.

Photosynthesis: The Kranz leaf anatomy, typical of C₄ grasses, provides kikuyu with an efficient photosynthetic process. Growth rates in full sunlight and warm conditions can be twice that of C₃ grasses. However, kikuyu is only moderately shade tolerant and the C₄ advantage is lost with decreasing light intensity. Lower levels of the enzyme rubisco and rapid translocation of sucrose away from the leaf also leave kikuyu leaves with lower protein and soluble carbohydrate contents than C₃ grasses.

Water use efficiency: High photosynthetic rates due to the C₄ mechanism give tropical grasses good water use efficiency. However, kikuyu is more sensitive to water stress than other C₄ species, especially under high evaporative demand. It requires a minimum annual rainfall of 900 mm unless irrigated. Drying surface soils reduce photosynthesis and slow shoot growth while assimilate is redirected to root growth in wetter parts of the soil.

Temperature: With its high plateau origins, kikuyu has a lower optimum temperature and narrower range than other tropical grasses. Maximum dry matter production occurs between 21° and 30°C, growth outside this range being restricted and ceasing below 8°C to 10°C. Light frosts desiccate exposed herbage with sustained frosts killing kikuyu.

Given temperatures in the optimal range and adequate soil water, N is often the most limiting factor for the growth of kikuyu. Yields greater than 30 t/ha/year have been measured in dryland pasture trials on the north coast of NSW where N supply was unlimited. Similarly, irrigated kikuyu maintained at field capacity averaged 25 t DM/ha/year at Camden although the yield was depressed when N was applied at 80% of removal. Other trials averaged 12 t DM/ha/year with 500 kg N/ha and more than 1000 mm of water. Production of unfertilised kikuyu without irrigation is more modest.

Utilisation of kikuyu pastures, under field trial conditions, can be as high as 75%. However, actual utilisation of dairy pastures can be as low as 3 to 4 t/ha. The higher net dry matter production and lower net utilisation of tropical versus temperate pastures results in more residues being left on the paddock after grazing. Any pasture that is not consumed by the dairy herd or conserved as feed is available for decomposition and mineralisation in the soil and is a potential source of N for microbial and plant uptake.

As with all grasses, the concentration of N in kikuyu is normally greater in the leaves than stems and the shoots than roots. Forage quality is maximised when kikuyu is grazed at 4.5 leaves per tiller, the stage at which the both the protein content and leaf:stem ratio are highest. The N concentration of the various plant parts can be measured from tissue samples or derived from feed quality data giving results for kikuyu leaf ranging from 2.7 to 4.1% N and an average N content in stems of 1.4%.

Nitrates accumulate, mainly in the stem tissue of kikuyu, when uptake exceeds the requirements for protein synthesis. This can result from an excessive supply of N and/or when growth slows. Conditions leading to NO₃⁻ accumulation, such as moisture stress, low light levels and low temperatures, often coincide with the autumn feed deficit. High NO₃⁻ levels reduce the palatability of grazed kikuyu and can lead to nitrate and nitrite poisoning, particularly when cattle are forced to eat pastures down to the ground. Nitrates represent a potential source of readily available N when recycled in ungrazed or under-grazed pastures. Unfortunately, balancing NO₃⁻ availability between plants and livestock can be a minor consideration during the autumn feed gap when dairy farmers are striving to maintain dry matter intake from limited pasture availability.

Few studies into the effect of fertiliser on N in kikuyu have been conducted in Australia. One such study at Wollongbar on the north coast of NSW showed that the N concentration of all plant parts rose with increasing rates of N fertiliser and ranged from 0.8% N in the roots with no applied N to 4.0% N in the leaves of pasture receiving 672 kg N/ha. Higher stocking rates also increased N in the shoots at the expense of the rhizomes. Nitrogen return to and cycling within the paddock was accelerated with increased utilisation but left the residue with a reduced content of N.

Information about the C content of kikuyu is not recorded in the literature. Extrapolating from studies of other species, a concentration of 40% C has been set for this review. This allows C:N ratios to be estimated for the various above and below ground organs and the state of net mineralisation versus immobilisation to be determined for decomposing kikuyu. Net mineralisation would occur in decomposing leaf of kikuyu, especially from pastures receiving N fertiliser. However, 336 kg N/ha is required for stem decomposition to lead to net mineralisation and decaying root and rhizomes result in immobilisation regardless of the amount of N applied.

This has consequences for dairy pastures on the mid-north coast of NSW where the low levels of N fertiliser applied to kikuyu might result in relatively high C:N ratios of the decomposing substrate. Net immobilisation of N into the biomass in autumn could reduce its availability to establishing ryegrass, both from that being mineralised in the soil and N applied as fertiliser. Compounding the problem is the management of kikuyu thatch. Strategies that remove the leaf, such as heavy grazing or mowing for silage increase the C:N ratio of the residue and thus the potential for net immobilisation.

Assessment of soil nitrogen availability

Nitrogenous fertilisers are the largest and most expensive input to Australian dairy pastures. An accurate assessment of mineralisation rates and quantities of plant avail N in the soil is essential to determine the timing and rates of strategic fertiliser N and to optimise N use efficiency. However, the soil N cycle is extremely complex and dynamic and rates of N mineralisation cannot be measured by simply analysing routine soil samples taken at a single point in time.

Where sufficient local trials have been conducted to provide accurate fertiliser recommendations, mineral N supplied from the soil, though not directly measured, has been accounted for in the recommended rates. Alternatively, mineralisation and accumulation rates can be determined from field assessments of available soil N, laboratory incubations, chemical methods, N tracing studies and the development of indices and models. Each method relies on assumptions that, by necessity, simplify aspects of the soil N cycle. Data collected still only represents points in time and space and may miss rapid N transformations in the soil. Thus, any extrapolation should be treated with appropriate caution.

Modelling, in particular, is a tool that could potentially be used to predict mineralisation rates and the accumulation of available N under pastures. However, models are only as good as the underpinning knowledge of the processes involved, the accuracy of the data included and the relevance of the assumptions made. Modelling a system as complex as the soil N cycle compounds the problem. Not all pools and fluxes within the soil N cycle can be accurately evaluated. A small estimation error at one point in the cycle, for example mineralisation rate, can introduce a much larger error at another point, such as the amount of mineral N available to the pasture.

Conclusion and recommendations

The soil N cycle under kikuyu can be likened to a very large and complex jigsaw puzzle for which most pieces are currently missing. Although we know, to a certain extent, what is happening under the kikuyu sward we are not yet able to quantify the processes involved. Apart from a small number of trials conducted almost 30 years ago, most of the information about kikuyu growth and management on the mid-north coast is anecdotal or observational in origin or is extrapolated from other regions.

Water and N supply are the two main limiting factors affecting kikuyu growth, given the limitations of seasonal temperature variations, and are variables that can be manipulated by farmers. Nitrogen uptake also affects the N concentration of various plant parts and ultimately the C:N ratio of decomposing litter. However, no tissue C tests have been reported for kikuyu. Nitrogen transformations under kikuyu have not been investigated. Until valid data is available, strategic N use through the kikuyu growing season that will improve N availability for ryegrass cannot be determined.

Trial data often does not accurately represent the conditions under which the kikuyu/ryegrass rotation is managed on the farm. Current recommendations for N fertiliser application for ryegrass are made from field trials with a prepared seedbed and harvested by the cut and carry method. Although more difficult to conduct, a trial where the ryegrass is oversown into kikuyu and the harvest method better approximates grazing, could provide more information to farmers and allow a benefit-cost analysis of various N rates applied to kikuyu as well as strategic N use on ryegrass.

These recommendations will focus on surveys and field trials on dairy farms that will fill the information gap for kikuyu pastures on the mid-north coast of NSW:

1. On-farm surveys and sampling to collect data relating to the growth and management of kikuyu pastures on the mid-north coast of NSW.
2. A series of field trials to establish regional data for kikuyu growth and N availability under the sward:
 - a. Kikuyu growth trials to determine growth rates, soil N and tissue N concentrations of a pure kikuyu sward grown under a range of N fertilisation rates and moisture regimes.
 - b. Mineralisation trials to determine N transformations and mineralisation activity in the soil under kikuyu.
 - c. Kikuyu/ryegrass trials to determine strategic N use in kikuyu that will improve N availability for ryegrass establishment and growth, focussing on the transition period from kikuyu to ryegrass.

2. Introduction

Dairy farming is a significant industry on the mid-north coast of New South Wales. In 2007 there were more than 150 dairies operating in the MidCoast Dairy Advancement Group's area, milking approximately 20,000 cows and producing 130 million litres of milk at a farm gate value of \$60 million (DAGs 2007). MidCoast DAGs represents the local dairy industry from the Great Lakes in the south, west to Gloucester and north to the Hastings River Valley. Included in this region, which straddles the North Coast and Hunter Local Land Services, are the Manning and Camden Haven Valleys and the Comboyne and Bulga Plateaux. Dairy farms are located at elevations ranging from less than 1 metre above sea level on the lower river flood plains to approximately 650 m on the Comboyne Plateau.

The mid-north coast region of NSW is in the transition zone between the tropical climate of the north and the southern coastal temperate zone (Chan and McCoy 2010). This subtropical region experiences wet, hot summers, wet autumns and relatively dry, mild winters and springs (Fulkerson and Doyle 2001). Mean monthly temperatures at Taree (Figure 1) on the lower Manning River range from a minimum of 5.9°C in July to a maximum of 29°C in January (BOM 2014). On average, only 8 days of frost are recorded each year at Taree with the number of frosts increasing further west in the hinterland and with elevation. Maximum daily temperatures above 40°C at Taree have been recorded from October through to March.

Taree receives a mean annual rainfall of 1178 mm compared with 982 mm at Gloucester, 50 km to the south west, and 1999 mm on the Comboyne Plateau, 30 km to the north (BOM 2014). Mean monthly evaporation exceeds rainfall from late winter through to mid summer (Figure 2). Rainfall is dominant during the summer and early autumn months but is also highly variable. Figure 3 shows monthly rainfall for six years from 2008 to 2013. April rainfall varied almost 400% from the mean and ranged from 504 mm in 2008 to 16.8 mm in 2010.

The region is also subject to periodical flooding, mainly during autumn and winter. The Manning River has had more than 30 significant floods, at 3.4 m or higher above the Australian Height Datum (Gow and Gow 2005), since European settlement with numerous moderate to minor floods. These floods can leave paddocks waterlogged for significant lengths of time, especially when there is repeated flooding, as occurred in 1963 and 2009. The lower river flood plains, which support a number of dairy farms, are particularly prone to flooding and water logging.

Soils in the region are largely sedimentary in origin, with shales and other fine-grained sedimentary rocks underlying the Manning and Hastings basins (Bale 1992). Alluvial soils (kandosols) are common along the river and creek flats, the loams and silt loams being easy to work and having good physical conditions for plant growth (Elliott 1979). They are mildly acidic and there are large areas of potential and actual acid sulphate soils on the lower flood plains (Lines-Kelly 2000). The majority of dairy farms in the region are based on these alluvial soils which were originally covered with extensive rainforests (Bale 1992).

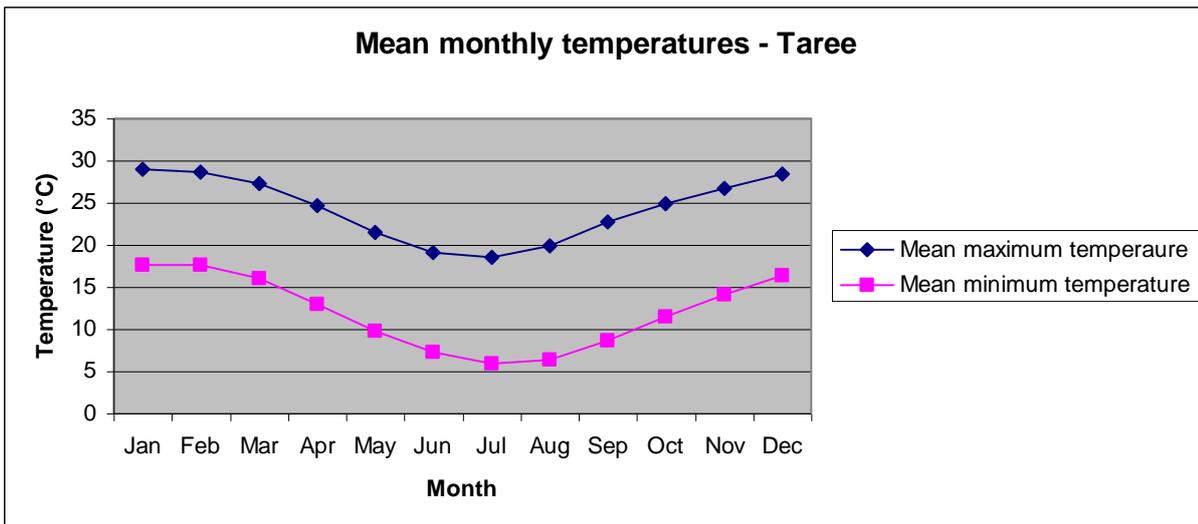


Figure 1 - Mean monthly temperatures at Taree, NSW [Source: BOM 2014]

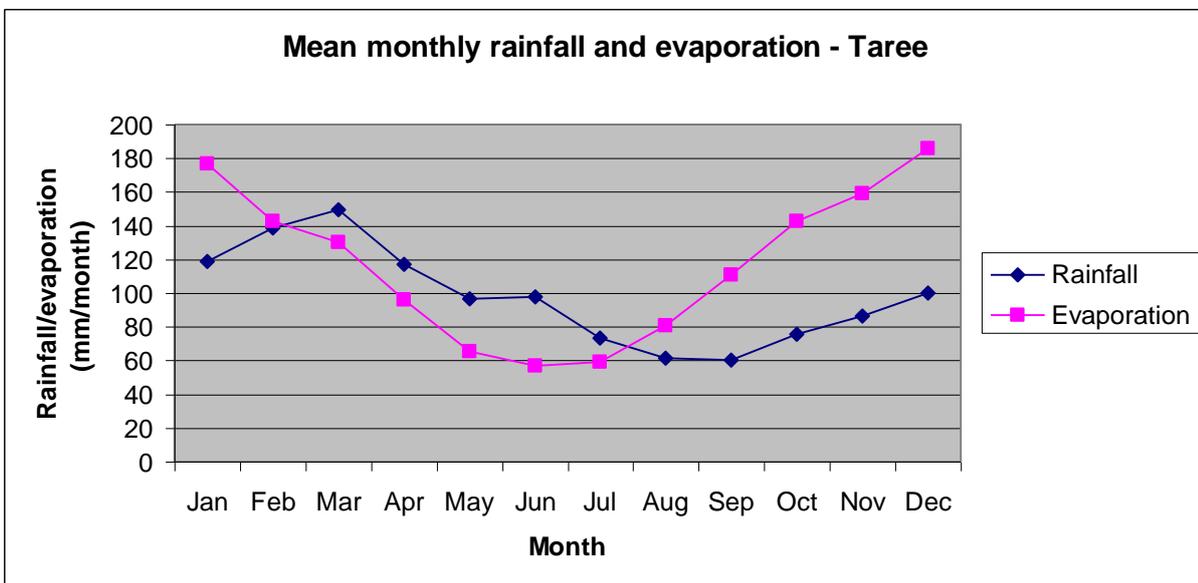


Figure 2 - Mean monthly rainfall and evaporation - Taree NSW [Source: BOM 2014]

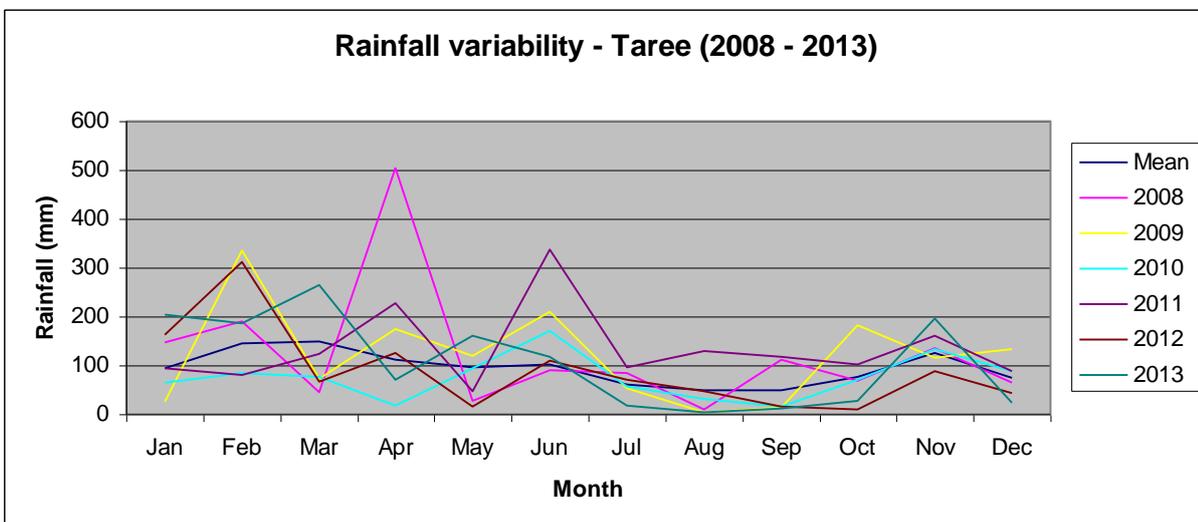


Figure 3 - Rainfall variability - Taree (2008 - 2013) [Source: BOM 2014]

Kurosols dominate the middle catchments, yellow podzolics being the most common soil type with some areas of red, brown and lateritic podzolics (Launders 2009). These hard-setting topsoils with strongly acidic subsoils and a tendency for dispersion and erosion developed under sclerophyll forests (Bale 1992) and are suitable for beef grazing and timber production (Lines-Kelly 2000). However, some dairy farms based on creek flats in the middle catchments have paddocks on surrounding hills and associated gullies that have been successfully pasture improved. The lithosols (tenosols) on shallow, stony ridges do not support pasture production for dairy farms.

The Comboyne Plateaux is volcanic in origin, with Kraznozem soils (ferrosols) having developed on the underlying basalts in a high rainfall area. These soils are strongly weathered and leached and typically acid with average pH values of 4.0 (Elliott 1979). The original massive stands of 'brush' timber species, including red cedar, rosewood and beech as well as numerous hardwoods (Bale 1992) were cleared in the late 19th and early 20th centuries for dairy farming which has continued to the present day, both on the plateaux and in the surrounding valleys. The original native fertility of the soil has been maintained by good pasture and fertiliser management.

Until the 1970s temperate grasses, such as perennial ryegrass (*Lolium perenne*), were most commonly used as the base species in permanent pastures on the mid-north coast (Kemp 1975). However, poor growth over summer and invasion from volunteer tropical grasses, especially kikuyu grass (*Pennisetum clandestinum*) and carpet grass (*Axonopus affinis*), resulted in poor persistence of the temperate species. Kemp (1975) began a series of field trials to determine yields and seasonal growth curves of tropical grasses grown in the region. His results showed that kikuyu could form an effective permanent pasture base at sites where it was intensively grazed with applied nitrogen (N) fertiliser.

C₄ tropical grasses, especially kikuyu, now form the main pasture base on dairy farms on the mid-north coast of NSW (Fulkerson and Doyle 2001). Declining autumn temperatures reduce the growth of kikuyu until it is virtually dormant in winter (Kemp 1975). To maintain productivity and whole milk supply through the cooler months, kikuyu pastures are oversown with temperate C₃ species, particularly a number of cultivars of annual and Italian forage ryegrasses (*Lolium multiflorum*) bred for a range of characteristics to fill the winter/spring feed gap (Launders *et al.* 2010).

This pasture-based production system, with its relatively low costs, provides the local dairy industry with a key competitive advantage (Garcia *et al.* 2014). However productivity, and therefore profitability, is largely a function of the quantity and quality of pasture grown and converted into milk. To supply whole milk, stocking rates and average milk production per cow must be kept at a relatively constant level throughout the year. Without supplements and/or conserved feed, production is limited to that supplied by the pasture at its minimum growth rate.

An autumn feed gap is created when kikuyu paddocks are taken out of rotation for the establishment of ryegrass. Figure 4 shows the estimated growth rates of kikuyu and ryegrass in the mid-north coast on NSW relative to the requirements of dairy cows stocked at 1.5 cows/ha on the milking area. The dry matter intake assumes a herd of 600 kg Friesian cows, each consuming 20 kg DM/day of pasture throughout the year (Freer *et al.* 2007). The pasture yield curves also assume average rainfall and temperature conditions.

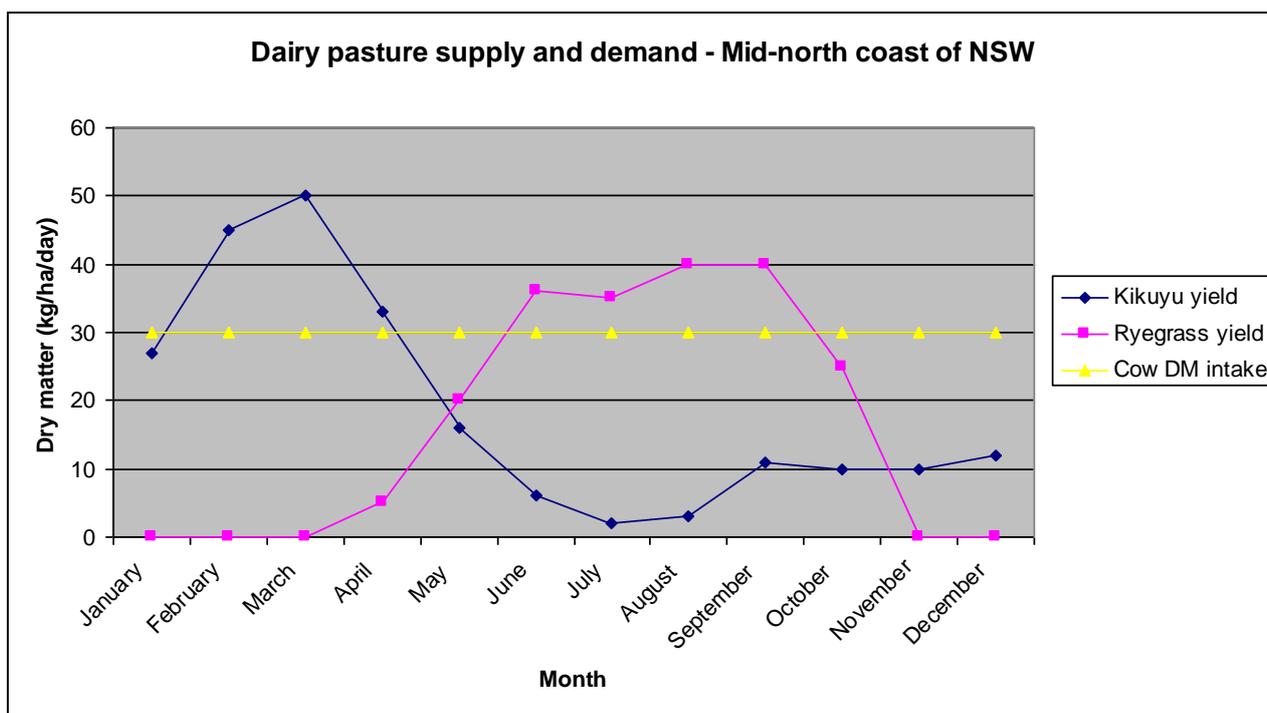


Figure 4 - Pasture supply and demand for a dairy farm on the mid north coast of NSW stocked at 1.5 cows/ha [Estimated yield data sourced from McDonald (2004)]

Some farmers plant a large proportion of the milking area to ryegrass at one time, relying on silage until the ryegrass is ready for its first grazing. Others adopt a rotational system with, ideally, a ryegrass paddock being ready to graze for every kikuyu paddock that is taken out of production. In practice, and especially at establishment, this is not always the case. Kikuyu growth needs to be suppressed, especially during warm, wet autumns, to reduce competition for the germinating ryegrass. This is attempted with a variety of methods including mulching, heavy grazing, or mowing and ensiling the kikuyu (Fulkerson and Doyle 2001). Herbicides at low rates are also used to retard kikuyu growth. The need to resow one or more paddocks of ryegrass later in autumn is not uncommon if establishment has been unsuccessful. Annual ryegrasses are typically spun and mulched or sod-seeded into the remaining kikuyu sward at seeding rates of 20 kg/ha for small seeded diploid varieties and 30 kg/ha for the larger seeds of tetraploids (Beale, personal communication). Higher seeding rates of 30-40 kg/ha are not uncommon (Fulkerson and Doyle 2001).

Nitrogen supply is integral to the establishment and growth of ryegrass as it is to overall productivity on a dairy farm. The Accounting for Nutrients project conducted by Gourley *et al.* (2012), which studied whole-farm nutrient balances on 41 dairy farms across Australia, found a positive correlation between total N inputs and milk production ($r^2 = 0.37$). Inorganic fertiliser, mostly urea, accounted for 43% of N brought onto the dairy farms with another 40% of N imported as feed. At a median value of 104.5 kg N/ha, the purchase of nitrogenous fertilisers is the largest and most expensive input to pasture production (Gourley *et al.* 2012).

On the mid-north coast of NSW most N fertiliser is applied to forage ryegrass. Current recommendations are 20-40 kg N at sowing for broadcast operations and 20 kg N when direct drilling (Griffiths *et al.* 2011). Topdressing with 30-60 kg N/ha after every second grazing is also recommended when conditions are favourable for growth. In contrast, kikuyu on dairy farms may receive from zero to 200 kg N/ha throughout the entire growing

season (J. Neal, personal communication). This is despite recommendations to apply 120-240 kg N/ha/year to intensively managed pastures (Mathews *et al.* 2004) in split dressings of 45 kg N/ha before rain or irrigation to promote growth (Griffiths *et al.* 2011).

Kikuyu poses a challenge to pasture management on dairy farms in late summer and early autumn, even in an average season. To maintain quality, kikuyu should be grazed at the 4.5 leaf stage (Reeves *et al.* 1996). Warm, moist conditions lead to rapid growth rates that quickly pass this optimum. Forage supply exceeds the flat demand of herds supplying whole milk and the farmer is left with an excess of standing feed of deteriorating quality (Figure 4). There is considerable expense in both labour and fuel to 'top' kikuyu paddocks that cannot be conserved as silage or fed off to stock other than the milking herd.

Applying N fertiliser to kikuyu increases growth rates and therefore the magnitude of the oversupply problem. Local environmental constraints also limit the effectiveness of N application to kikuyu (J. Neal, personal communication). The highly variable rainfall in summer and autumn reduces consistent N responses in dryland conditions. Irrigation could provide a regular moisture supply but pumping is often restricted on the local unregulated rivers during low flows and peak summer demand. Increased kikuyu growth rates from the strategic use of N in autumn would increase competition with establishing ryegrass.

Cost-benefit considerations also discourage the use of nitrogenous fertilisers on kikuyu. Launders (personal communication) considered that there was little benefit of applying N in spring, when low rainfall would lead to poor growth responses or in summer when the additional growth would not be converted into higher milk production. A yield response in autumn could be of benefit but would come at the expense of ryegrass growth. This is in contrast to the N fertilisation of ryegrass with urea despite fluctuating fertiliser prices. When urea cost \$425/t, the first 30 kg N/ha applied after each grazing resulted in a net profit from milk produced, whereas the dairy farmer only broke even when using an additional 30 kg N/ha (Launders, unpublished data). The return from milk produced remained the same when urea rose to \$750/t but a further price rise to \$1100/t of urea resulted in the farmer losing money if the second 30 kg N/ha was applied. Comparative cost-benefit calculations have not been carried out for N fertiliser applied to kikuyu.

There is a frequent observation that kikuyu pastures do not respond to N fertiliser (Miles 1998). However, not applying N to kikuyu creates its own set of problems. In the two-pasture dairy system summer growth largely relies on N remaining from fertilised ryegrass. Rapid kikuyu growth leads to the build up of large amounts of organic matter under the sward. As available N is depleted from the soil and organic carbon levels rise, N in the decomposing kikuyu litter and any applied N can be 'trapped' in the organic matter and become unavailable to growing ryegrass pastures. During decades conducting ryegrass trials in the Manning Valley, Launders (personal communication) frequently observed that fertiliser N was rapidly exhausted from the soil between harvests. The tendency to withdraw N fertilisation from kikuyu to 'force' both it and the following ryegrass to utilise available reserves simply doesn't work. Nitrogen can only be made available by depleting reserves of carbon and increasing those of N (Miles 1998).

Nitrogen fertiliser recommendations for ryegrass pastures are largely based on field trials conducted under conditions that ensure the success of the trial but, by necessity, simplify the complexity of the plant-soil ecosystem. Due to competition during establishment in the kikuyu sward, ryegrass is usually sown into a prepared seedbed with conditions optimised to promote germination. Fertiliser application rates are often determined on a simple

arithmetic basis without strategic additions to investigate responses at particular growth stages. Unless a water use x fertiliser rate interaction is being investigated, plots are watered if the season turns dry. The harvest method is usually cut-and-carry that does not accurately simulate the grazing situation.

Kikuyu-ryegrass rotations have been the dominant pasture type on dairy farms on the mid-north coast of NSW for 40 years. However, little research has been published on the interactions between the two species and, especially, N transformations in the soil and N availability to the pasture. A knowledge gap exists relating to such questions as:

- How much N is retained in the soil at the end of the kikuyu season?
- How much N becomes available to establish and grow ryegrass for winter/spring feed?
- What factors can be manipulated on-farm that will increase the amount and availability of N for emerging ryegrass pastures?

This literature review will investigate soil N and the soil N cycle, especially mineralisation, immobilisation and the availability of N to plants. Factors affecting the growth of kikuyu, the fate of N in kikuyu pastures and N transformations under the sward will also be examined. Where it is available, local and regional data will be used to begin the task of quantifying N in its different forms for both kikuyu and the various stages in the soil N cycle. Recommendations for further research and field trials will be made based on the findings of the review.

3. Soil nitrogen

Nitrogen is essential for all life forms. As a component of amino and nucleic acids it is required for protein synthesis and the production of genetic material (Whitehead 1995). Plant and animal metabolism is driven by enzymes, most of which are proteins. After carbon (C), hydrogen (H) and oxygen (O), the building blocks of organic matter, N is the nutrient required by plants in the greatest amounts. It is often the most limiting factor for plant growth, especially in intensive farming systems. Nitrogen gas (N₂) comprises approximately 78% of the earth's atmosphere by volume but cannot be converted directly into organic molecules by most life forms (Haynes 1986c). Instead it must be 'fixed' into forms that can be taken up by plants.

Nitrogen transformations between the atmosphere, soil, plants and animals occur as a complex cycle. There is also an internal N cycle in the soil involving a number of microorganisms (Stevenson and Cole 1999). The movement of any single atom of N through these cycles is random. The cycling of other nutrients, especially phosphorus (P) and sulphur (S), is closely linked to the biochemical transformations of N.

In summary, atmospheric N is fixed by microorganisms, electrical discharges and industrial processes to provide N ions that can be absorbed and assimilated by plants. Plants convert the N ions into proteins and nucleic material which are ingested, digested and incorporated into animal tissues. Plant residues, urine, manure and dead remains of animals return N to the soil in organic matter. Following decomposition, some of the N is mineralised into plant available forms. Nitrogenous gases are lost from the soil, returning to the atmosphere through the processes of denitrification and volatilisation. Losses also occur via runoff and leaching.

The N cycle and its impact on pastures has been thoroughly reviewed in the literature, including work by Haynes *et al.* (1993), Whitehead (1995), Stevenson and Cole (1999), Bellows (2001) and Henry (2012). This report will focus on: the forms and quantities of N in the soil; the processes of N mineralisation and immobilisation; and factors affecting N availability under kikuyu.

Biological fixation of nitrogen

Biological fixation of atmospheric N is the major natural pathway by which N enters the soil-plant cycle. Nitrogen fixation by the *Rhizobium* bacteria, living in a symbiotic relationship in root nodules on agricultural legumes, accounts for one quarter of all biological fixation (Haynes 1986c). The literature on biological fixation is extensive as summarised in general by Stevenson and Cole (1999) and reviewed more recently by Unkovich (2012) for the Australian dairy industry.

Legumes have been an important component of improved pastures in Australia since the 1930s, for their capacity to fix nitrogen and improve soil fertility and structure (Nichols *et al.* 2007) and supply protein and minerals to grazing animals in a more digestible form than grasses (Whitehead 1995). More than 300 kg N/ha/year can potentially be fixed by legumes growing in Australian pastures (Peoples *et al.* 2012). Despite this, in a survey of 41 Australian dairy farms, Gourley *et al.* (2012) found that the median legume content of dairy pastures was only 6% DM with 71% of the paddocks assessed having less than 10%

legume content. Average annual N₂ fixation by legumes in dairy pastures was less than 50 kg N/ha and accounted for only 16% of total N inputs to the farm (Gourley *et al.* 2012).

White clover (*Trifolium repens*) is the most common legume sown into pastures on the mid-north coast of NSW. Although it has naturalised across much of the region, there are serious problems with persistence. Yield is variable from year to year (Unkovich 2012) and, in the sub-tropical climate of the region, white clover is essentially an annual rather than a perennial legume and must be resown if a sufficient self-sown seed bank is not available in the soil (T Launders, personal communication). Compounding the problem is the poor viability of rhizobia on pre-coated clover seed and the lack of availability of uncoated seed and fresh inoculant (Gemell *et al.* 2005).

Overall dry matter production is reduced when dairy pastures are managed to maintain an adequate clover percentage, affecting milk yields. Dairy farmers are gradually reducing their reliance on legumes, preferring to source nitrogen from a bag and protein from supplements. Unkovich (2012) considered that while the clover component of dairy pastures remained so low, measurement of N₂ fixation would be of little quantitative value. For these reasons the biological fixation of N will not be considered as an input into the soil N cycle for this review.

Forms of soil nitrogen

The sum of all nitrogenous molecules in the soil is known as total N. Organic matter provides the largest pool of N, contributing from 95 to 99% of total N in most soils (Goh and Haynes 1986; Whitehead 1995). However, more than 50% of the organic N in soil exists as structural components of humic substances and is not readily available to plants (Stevenson and He 1990). Nitrogen can also be fixed into clay minerals in relatively unavailable forms. Some topsoils can retain from 3 to 10% and subsoils more than 50% of total soil N as NH₄⁺ fixed to clay particles (Whitehead 1995).

The soil biomass also accumulates nitrogen. Haynes (1986a) reviewed studies that calculated microbial N and found, for a range of agricultural soils, that the biomass held from 0.5 to 15% of total soil N. Microorganisms act as both a source and sink for mineral N, which is available to plants in the form of nitrates (NO₃⁻) and ammonium (NH₄⁺). These ions, including the intermediate form nitrites (NO₂⁻), make up only 2 to 5% of total soil N (Tisdale *et al.* 1993).

Total nitrogen

The range of total N varies from less than 0.02% in subsoils to more than 2.5% in peats (Tisdale *et al.* 1993). Long-term grassland soils can accumulate up to 15,000 kg N/ha following decades of organic matter inputs from plant residues and animal excreta accompanied by relatively slow rates of decomposition (Whitehead 1995). Total N in cultivated soils is much less, ranging from 2000 to 4000 kg N/ha.

In general, returning cropping land to pasture will increase total N stored over time whilst ploughing up pasture will reduce total N (Whitehead 1995). In each circumstance a new equilibrium can be reached given sufficient time, in decades for pasture converted to cropping land and possibly centuries for arable land returned to pasture. However, changing management practices leave the total N status of agricultural soils in a state of flux. Whitehead (1995) reviewed studies measuring the accumulation of total N under

pasture. Depending on the soil type, climate, botanical composition and fertiliser regime, total N increased at rates from 52 to 112 kg N/year.

Beale (unpublished data) soil tested 111 paddocks across 12 dairy farms on the mid-north coast of NSW. Total N ranged from 0.19 to 0.77% with a mean of 0.37 % N on a dry matter basis. Assuming an average bulk density of 1.3 g/cm³ for soils in the region (P. Beale, personal communication), the dairy paddocks tested contained, on average, 4810 kg N/ha in the top 10 cm with a range of 2470 to 10,010 kg N/ha. The majority of the pastures were kikuyu based and were tested following a late summer/early autumn break of 130 mm (BOM 2014). The preceding eight months had been some of the driest on record with Taree receiving 361.4 mm of rain from July 2013 to February 2014, only 50% of average for the period. That summer saw only 30% of average seasonal rainfall with an accompanying reduction in growth of all but irrigated pastures.

Neal *et al.* (2013) measured total N under a range of forage crops that had been established three years previously on pastures at Camden, NSW. Results ranged from 0.30 % N under forage rape to 0.38% N under kikuyu. Topsoils underlying perennial and Italian ryegrasses stored 0.31% N and 0.33% N respectively. Total N stored under kikuyu equated to 5282 kg N/ha and under perennial ryegrass, 4309 kg N/ha.

Organic nitrogen

The concentration of N in organic matter is approximately 5% of dry matter (Whitehead 1995). Factors that affect the accumulation or loss of organic matter from soil will also determine organic N content. Vegetation, climate and soil type are major factors in all soils. Pasture and cropping management are important in agricultural soils. Changes in total N are highly correlated to additions to or losses of organic C from the soil. Figure 5 shows the relationship between total N and organic C ($r^2 = 0.89$) in the top 10 cm of 111 dairy paddocks on the mid-north coast of NSW (Beale, unpublished data). Organic C in the paddocks ranged from 2.1 to 9% with a mean of 4.1% OC. Although sampling to 10 cm depth does not account for the total N stored in the whole soil profile, it can be a useful indicator of N availability for establishing ryegrass.

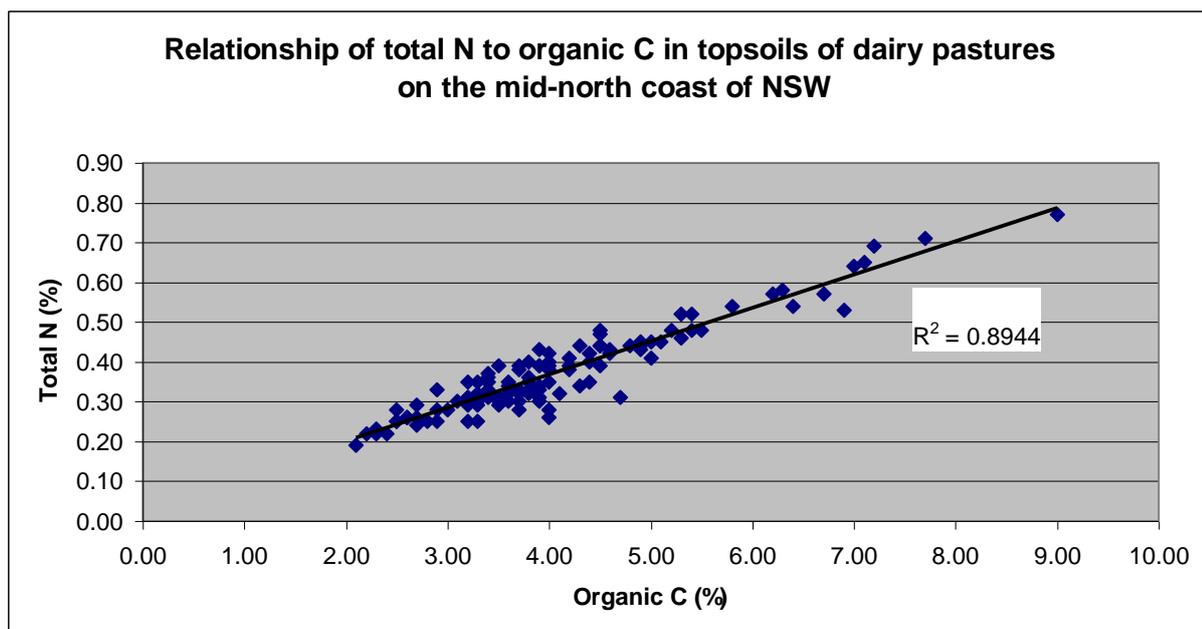


Figure 5 - Relationship between total N and organic C in topsoils (0-10 cm) of dairy pastures on the mid-north coast of NSW [Source: Beale, unpublished data]

Initially, the source of soil organic matter and organic N are the dead remains of plants and animals and excreta from grazing animals. It is difficult to measure the amount of herbage that senesces, dies and decomposes. Some is consumed by soil fauna and decomposition is a continual process. However, physiological measurements show that, in grazed or harvested pasture, approximately the same dry weight of herbage is returned to the soil as is removed (Whitehead 1995). Increasing the time between defoliations increases the amount available for decomposition. Stubble retained after cutting at a height of 5 cm in various grassland scenarios can range from 1500 to 4000 kg/ha (Whitehead 1995).

Roots make up more than half the dry matter yield of pastures (Whitehead 1995). Newly sown pastures can produce 3000 kg DM/ha of root material in the first six months and 10,000 kg DM/ha by the third or fourth year. Permanent pastures may contain more than 20,000 kg DM/ha in living and dead root material. Whitehead's (1995) review of long-term pastures found root growth rates from 2800 kg DM/ha/year to 6400 kg DM/ha /year.

Studies into the length of life of grass roots are difficult to undertake (Whitehead 1995). Apart from the obvious complications of removing roots intact from the soil and separating live from dead material, the central vascular stele of mature roots can continue conducting water and nutrients long after the outer cortex has decomposed. The roots of permanent pastures may live from two to five years with light grazing pressure (Whitehead 1995). Increasing the frequency of defoliation reduces the life span to less than six months in intensively managed pastures.

The N content of plants depends on the species, plant part, stage of growth and N supply and can range from 0.1 N to 6% N (Haynes 1986a) with most plant tissues containing 1 to 5% N (Whitehead 1995). On a dry matter basis the concentration of N in stubble and litter is approximately 50 to 75% of that of the green herbage with a range of less than 1% N (Haynes 1986a) to as much as 3% N (Whitehead 1995). Maximum root growth requires a lower N supply than maximum shoot growth and the concentration of N in roots is considerably less than the shoots of the same plant.

Grazing animals return a significant amount of organic matter and nitrogenous compounds to the soil in urine and dung. The proportion of total N excreted depends on the type, weight and physiological state of the animal, dry matter intake and the concentration of N in the feed (Haynes *et al.* 1993). The urea in urine is rapidly hydrolysed to ammonia (NH₃), some of which is lost to the atmosphere via volatilisation (Whitehead 1995). The remainder moves through the soil N cycle. Manure is decomposed, first by earthworms, dung beetles and other macrofauna, and then it too enters the soil N cycle.

Dairy cows have a protein requirement varying from 10% crude protein (CP) for dry cows to 18% CP for those in early lactation (Moran 2005). They excrete 75 to 80% of the N consumed, at a relatively constant rate in manure (0.8 g N/100 g DM intake) equivalent to 6% CP, with the remainder being reflected in urinary output (Whitehead 1995). Birchall (2008) calculated that a 600 kg pasture fed cow on a Victorian dairy farm would produce between 265 and 394 g N/day in urine and faeces depending on the type of concentrates fed in the bales. Assuming cows spend 15% of their time out of the paddock, in laneways and dairies (Birchall *et al.* 2008), a herd of 200 cows could potentially leave 56 kg N on their allocated strip of pasture each day. Although time spent on a feed pad would reduce this amount, an effective effluent collection and recycling system from dairies, yards and feed pads would return more N to the paddock.

Although organic N in soils originates from plant and animal material, when it is analysed the major forms are found to be amino acids and amino sugars deriving largely from bacteria and fungi (Haynes 1986a). The amounts and types of each amino group vary widely but are more prevalent in the rhizosphere, either as compounds secreted into the soil solution by microorganisms or as products of decay of the biomass. The processes of mineralisation and immobilisation that lead to this transfer of N into the microbial biomass will be discussed in the section on the soil N cycle.

Plant available nitrogen

Plants take up N as the NO_3^- anion and/or the NH_4^+ cation largely by root absorption. These 'available' forms of nitrogen can be supplied as fertiliser or from the mineralisation of organic N in the soil, hence the term 'mineral N'. During a growing season 1 to 2% of total N is mineralised to inorganic forms in temperate soils (Stevenson & Cole 1999). Unlike other major plant nutrients, N cannot be stored in the soil profile in these plant available forms (Miles 1998). Nitrate, an anion, is not adsorbed onto clay or organic matter colloidal surfaces and is extremely soluble and prone to leaching from the profile. Ammonium, a monovalent cation, is only held loosely on exchange sites in the soil and is readily substituted by aluminium (Al), calcium (Ca) and magnesium (Mg) ions. Once in solution this exchangeable NH_4^+ is also subject to leaching.

Ammonium fixation

Both NH_4^+ and NH_3 present in soil pores can be fixed in relatively unavailable forms (Stevenson and Cole 1999). Ammonia undergoes a chemical reaction with soil organic matter known as NH_3 fixation. Simple nitrogenous organic compounds, such as amino acids, can also be adsorbed onto clay minerals or bound into humic molecules protecting them from mineralisation.

The expandable clay minerals vermiculite, illite and, to a lesser extent montmorillonite, chemically fix NH_4^+ in their clay lattices. The amount of NH_4^+ fixed depends on the concentration of ions and the potassium (K) status of the soil. Potassium ions can block the fixation of NH_4^+ . Soil moisture and pH also have an effect. Wetting and drying cycles and increasing pH enhance fixation whereas little occurs in acid soils ($\text{pH}_{(w)} < 5.5$) (Stevenson and Cole 1999). This provides a slowly available pool of N over time with fixed NH_4^+ replacing exchangeable NH_4^+ as the latter is removed from the soil solution (Tisdale *et al.* 1993).

The majority of soils on the mid-north coast of NSW are not self-mulching, having kaolinite as the dominant clay type (Elliott 1979). In addition, the acid nature of the soils reduces the possibility of NH_4^+ fixation occurring. Of the 111 dairy paddocks tested by Beale (unpublished data) the mean pH value was 4.96 and 90% of the paddocks had a pH less than 5.4. The same paddocks had a medium to high organic C status, with 50% being greater than 3.8% OC to a maximum of 9.0% OC, which could predispose them to NH_3 fixation. However, this is more likely to occur in neutral to alkaline soils (Stevenson and Cole 1999).

Plant uptake of nitrogen

Both forms of mineral N are taken up by plants. The varying amounts of NO_3^- and NH_4^+ taken up depends on the species as well as soil and environmental factors (Whitehead 1995). Nitrate uptake is more common in well-aerated and acidic soils whereas NH_4^+ is more abundant in the anaerobic conditions of poorly drained or flooded soils (Mathews *et*

al. 2004). The rate of NH_4^+ uptake increases as pH rises. Temperature also has an effect with NO_3^- uptake in perennial ryegrass increasing with temperature from 5°C to 35°C whilst that of NH_4^+ decreases (Whitehead 1995). Other species show less effect of temperature on relative N ion uptake.

Nitrate absorption is accompanied by that of calcium (Ca^{2+}), magnesium (Mg^{2+}) and potassium (K^+) cations. Bicarbonate (HCO_3^-) and hydroxide (OH^-) anions are exuded into the soil solution or protons (H^+) are absorbed into the plant to maintain the electrical balance in the roots and the surrounding rhizosphere becomes more alkaline. Conversely, phosphate (H_2PO_4^-), sulphate (SO_4^{2-}) and chloride (Cl^-) anions are absorbed with NH_4^+ . Protons are exuded by roots, again to maintain electroneutrality. The resulting acidification of the surrounding soil solution can be as much as two pH units and affect both nutrient availability and microbial activity in the rhizosphere (Haynes 1986d).

A mixture of both ions maximises growth rates, the relative proportions of each depending on the stage of development. Ammonium uptake is more efficient as NO_3^- must be reduced to NH_4^+ before incorporation into plant tissue, imposing an energy cost (Mathews *et al.* 2004). The ammonium ions are rapidly assimilated into low molecular weight organic compounds such as amino acids and translocated to shoots via the xylem. However, high rates of NH_4^+ uptake will suppress the absorption of cations, especially K^+ , inducing a deficiency and retarding growth (Tisdale *et al.* 1993). Nitrates, on the other hand can be taken up in luxury amounts and stored in plant tissues with little effect on growth.

The rate of N uptake by grasses varies widely depending on the time of year, stage of growth, temperature, moisture supply, fluctuations in N supply and grazing regime (Whitehead 1995). If N is not limiting, growth and N content of the herbage depend largely on water supply to the pasture. Whitehead (1995) reviewed studies of ryegrasses finding an average uptake rate of 1 to 3 kg N/ha/day with a maximum recorded uptake by an Italian ryegrass of 7.5 kg N/ha/day after the application of 140 kg N/ha two weeks previously. In pastures that are not defoliated, the vegetative stage has the greatest rate of N uptake reaching a peak just before ear emergence and well before maximum yield is attained.

Defoliation can temporarily reduce the rate of N uptake (Whitehead 1995). Temperate pastures under intensive management, with several defoliations during the season, can produce from 8000 to 15,000 kg DM/ha/year containing 200-550 kg N/ha. Whitehead (1995) estimated the same pasture to produce approximately 6000-12,000 kg DM/ha/year in stubble and roots with an N content of 100-250 kg N/ha. Thus a temperate pasture could store between 300 and 700 kg N/ha/year.

Nitrogen in pastures

Most plants contain from 1 to 5% N in their tissues on a dry matter basis (Whitehead 1995). Chlorophyll and proteins, especially enzymes, make up a large proportion of plant N. The photosynthetic enzyme rubisco accounts for approximately 50% of enzymatic material in C_3 shoots (Lawlor 1991). Tropical C_4 grasses have lower levels of rubisco (ribulose biphosphate carboxylase-oxygenase) in their leaves, resulting in lower protein and total leaf N than C_3 grasses (Mathews *et al.* 2004).

The quantity of N required, consumed and stored in plants varies greatly depending on the species and genotype of the plant, growth stage and environmental conditions (Stevenson

and Cole 1999). Uptake is greatest during the rapid phase of vegetative growth and in general, more N is stored in the shoots than in the roots. As plants mature the proportion of cell wall material increases with a corresponding decrease in N compounds stored in the cytoplasm, such as enzymes, chlorophyll and nucleic acids (Whitehead 1995). The concentration of N in herbage decreases after ear emergence. In a study of seven pairs of annual-perennial congeneric temperate grass species, Garnier and Vancaeyzeele (1994) found a significant difference in the allocation of N to various plant parts between annuals and perennials. Annuals allocated more N to leaves than perennials (52.1% N versus 49.8% N; $P < 0.001$) whereas perennials allocated more plant N to roots (31.5% N) compared to annuals (29.8% N; $P < 0.05$).

Nitrogen supply affects the rate of tiller production in grasses (Whitehead 1995). Each leaf has a bud in its axil which can potentially form a new tiller. An N deficiency in a pasture inhibits the development of tillers. Increasing the rate of N fertiliser in the sward will increase the number of tillers until competition reduces the life span of individual tillers. In contrast, N has little effect on the number of leaves produced per tiller, which is largely a function of temperature (Whitehead 1995). Temperate C_3 grasses, such as ryegrass and cocksfoot (*Dactylis glomerata*), support three leaves per tiller at any one time. If they are not grazed or harvested old leaves will senesce and die at the same rate as the new leaves emerge. Kikuyu, a tropical C_4 grass, produces four to five new leaves before the first begins to senesce (Garcia *et al.* 2014).

The greatest influence of N on pasture production is due to its effect on leaf size, both in terms of leaf area and weight (Whitehead 1995). Assuming no other limiting factors, leaf extension and size increase with N supply. However, the law of diminishing returns applies and a critical point is reached when the pasture attains its maximum growth rate. Further additions of N will not significantly raise the yield. However, strategic use of N fertiliser in a rotational system can increase herbage yield by increasing the number of grazings and/or harvests during the growing season (Whitehead 1995).

Nitrogen is recycled within grasses, especially when the supply is limited and as plants age (Whitehead 1995). Proteins and nucleic acids are broken down, translocated to new growing points and reconstructed on a regular basis. The process of photorespiration also releases ammonia within the leaf which can be reabsorbed and assimilated into amino acids, although losses can occur via the stomata. Low rubisco levels in C_4 grasses reduce this recycling and loss pathway (Mathews *et al.* 2004).

In a case of severe deficiency, most of the N is concentrated in structural components, such as cell walls, as well as in nucleic acids which provide the blue print for future growth (Whitehead 1995). There is little metabolic activity. As N supply rises, photosynthesis and growth resume and increase to the point where N supply is greater than short-term requirements and the remainder is stored as nitrates or amides. The critical concentration of N, as determined by tissue analysis, is 5 to 10% less than that required for maximum yield and decreases with the age of the pasture. Whitehead (1995) reviewed studies of grasses cut at a height of 2.5 cm. The critical N concentration for 4-week-old regrowth was 3.2 to 3.5% N, decreasing to 2.5% N at six weeks after harvest.

There is a continual turnover of leaf, stem and root material in grasses. The timing and rate of senescence are determined by light intensity as well as nutrient and water supply (Whitehead 1995). Fertiliser N increases the longevity of tillers in the first 2-3 weeks after application but hastens senescence in the weeks following. As leaves age, the activity of enzymes involved in assimilation decreases, while those responsible for degradation

increase. There is a net breakdown and partial remobilisation of proteins and chlorophyll, N being translocated in the phloem to other parts of the plant. Membrane disintegration follows, restricting N translocation. The concentration of N in pasture leaf residue is approximately half that of green leaves (Whitehead 1995). However, if rapid senescence takes place, for example during water stress, the residue can have a higher N concentration due to limited translocation.

Any herbage in pastures that is not grazed or harvested will undergo senescence. Therefore, the amount of litter left on the soil surface depends on management practices. In a review of pasture senescence studies, Whitehead (1995) found a large difference in the percentage of leaf lamina left as residue. Senescence accounted for 60% of the leaf lamina material in a low N input (60 kg N/ha), low stocking rate system (Laidlaw and Steen 1989) whereas only 30% of the leaf material senesced in a high N input (360 kg N/ha), high stocking rate system. Thus, higher N fertiliser rates and grazing intensities reduced the accumulation of dead material. However, grazing increased senescence due to damage from trampling and dung.

Fertiliser application before senescence will increase the N concentration of litter though Whitehead (1995) found that both fertilised and unfertilised pastures returned N to the soil from decomposing litter at rates ranging from 0.06 kg N/ha/day to 0.42 kg N/ha/day. Changing temperature and light conditions at the end of each growing season rapidly increase senescence. Decreasing temperatures and light availability in autumn result in the more rapid senescence of summer species. Conversely, winter species senesce more rapidly in spring as both daylength and temperature increase.

Roots are a vital source of carbohydrates, N compounds and other nutrients for pasture regrowth after defoliation. Studies into the growth and senescence of roots under different conditions are difficult to undertake, especially in the field. The results are not conclusive but indicate that grasses allocate more photosynthate to root growth and exploration when N is deficient (Whitehead 1995). However, with increasing N supply the relative proportion of root material decreases as plant resources are channelled into herbage production. Fertiliser N increases the N concentration of roots from less than 1% N in pastures receiving no fertiliser to 1 to 2% N in fertilised grass pastures and more than 2% N in legume swards. Increasing the N concentration of roots accelerates the rate of decomposition.

4. The soil nitrogen cycle

Plants rely on transformation cycles within the soil to provide a continual supply of available N. Organic matter is first decomposed, then mineralised to provide NH_4^+ and NO_3^- ions for plant uptake. In the process, a proportion of the N released from organic matter is assimilated and immobilised into the microbial biomass.

Decomposition

Decomposition is the process by which plant and animal residues are broken down in the soil. There are various pools of organic matter in soil that decompose at different rates and are grouped as 'active' or 'stable' (Stevenson 1994).

The active pool, which undergoes relatively rapid decomposition, includes litter, the light fraction, water soluble organics and enzymes (Stevenson 1994). Litter is the macroorganic matter visible on the soil surface and is important for the cycling of N and other nutrients, especially in natural grasslands and pastures. Litter in the various stages of decomposition is known as the light fraction. Plant roots exude water soluble organic compounds, such as amino acids, sugars and organic acids into the rhizosphere. Microorganisms secrete an array of extracellular enzymes to catabolise organic compounds in the soil. In turn, the enzymes themselves become denatured and break down. When plant roots and soil flora and fauna die, the cell walls and membranes rupture and the contents are released into the soil solution adding to the active pool for rapid decomposition followed by the more resistant structural cell components.

The mean residence time of the active pool can be as little as days or weeks. In comparison the stable pool, commonly termed humus, has already undergone substantial chemical change in the soil and is more resistant to further decomposition. The stable humus fraction may resist decomposition from decades to as much as 1000 years (Stevenson and Cole 1999).

Most of the C from organic compounds is ultimately returned to the atmosphere as CO_2 and nutrients are recycled in plant available forms (Haynes 1986a). The rate of decomposition of litter is largely a function of its organic component parts (Haynes 1986a). Water soluble compounds are highly degradable, losing 80 to 90% of their C in the first three to six months. In comparison, 70 to 80% of lignin, the most resistant of the organic compounds, can be retained in the first year of decomposition. Crop residues lose 60 to 70% of their C as CO_2 in the first year after harvest (Haynes 1986a). During the next six to nine years another 20% is lost. A proportion of the C, N and other nutrients is assimilated into the microbial biomass or incorporated into resistant humic molecules that comprise 70 to 80% of organic C in soils (Piccolo 2001).

There is a complex food web in the soil. Each trophic or feeding level within the web provides the substrate for the next, which in turn regulates the population size and activity of the former (Haynes 1986a). The macrofauna begin the process of decomposition by ingesting litter and other soil organisms. Earthworms feed on dead plant material and the collembola mainly on fungi (Haynes 1986a). Mites feed on both, adding bacteria to their diet while some adult insects consume woody detritus and manure. Soil nematodes are diverse feeders, with species that are herbivores, carnivores or omnivores.

The macrofauna fragment the litter, redistributing and incorporating it into the soil by bioturbation (Orgill 2008). Earthworms, ants and dung beetles contribute greatly to this process increasing the surface area of litter for colonisation and attack by microorganisms (Haynes 1986a). Digestion of litter by soil fauna also accelerates nutrient cycling. Generation times are generally short and, as C is lost through respiration, the bulk of ingested N is excreted back into the soil as urea, amino acids and NH_4^+ .

Fungi are the primary decomposers in soils (Haynes 1986a). They spread a mycelium or network of thread-like hyphae through the soil litter and light fraction, secreting extracellular enzymes that breakdown more complex organic compounds, such as cellulose and lignin, into simpler forms (Stirling 2001). Actinomycetes act in a similar manner, decomposing chitin and humic substances as well as cellulose.

As secondary decomposers, bacteria colonise particulate detritus that has a very high surface area to volume ratio and complete the decomposition process producing simple inorganic compounds (Haynes 1986a). Some mycolytic bacteria also colonise and break down fungal hyphae, controlling fungal populations. In turn, protozoa move through the soil solution feeding on bacteria.

Many factors affect the activity of and interactions between microbial populations and hence the decomposition of organic material. They include plant growth, additions of organic residues and substrate quality, moisture, temperature, soil pH, inorganic nutrients, physical characteristics of the soil and management practices, especially cultivation and the use of pesticides (Haynes 1986a). As these factors also determine mineralisation rates they will be dealt with in detail in that section of this report.

Nitrogen leaching

Plant residues release water-soluble C & N compounds shortly after litter fall without microbial action (Haynes 1986a). Approximately 10% of total N content of the initial litter can be lost from the substrate via leaching. The extent and rate of leaching is dependent on the amount of water that percolates through the litter, being greater in regions with a high precipitation rate or during high rainfall events. Nitrogen leaching can become a serious problem for livestock grazing dry standing pasture due to the reduction in quality of the feed (Curnow and Butler 2014).

Nitrogen mineralisation

Nitrogen mineralisation is the process by which the N in organic compounds is converted to inorganic or mineralised forms. A number of heterotrophic bacteria, fungi, actinomycetes and some soil macrofauna are involved in the formation of NH_4^+ (Stevenson and Cole 1999). As heterotrophs they all acquire their energy source from organic compounds in the soil solution. Mineralisation is not greatly affected by soil pH, given that the different organisms involved operate under a range of pH conditions. However, the population and activity of bacteria decline at pH values less than 4.5 with some increase in fungal growth (Whitehead 1995).

The microorganisms involved in mineralisation produce a large array of enzymes, each of which acts on a particular organic compound (Stevenson and Cole 1999). Some enzymes also derive from plant and animal origins (Haynes 1986a). The macromolecules in

decomposing material are broken down into their building blocks – proteins to peptides and then amino acids by proteases and peptidases in the process of amination. Nucleic acids are degraded ultimately to purine and pyrimidine bases and pentose sugars by another set of enzymes and urea in the soil is hydrolysed to CO₂ and NH₃ by the action of urease (Stevenson and Cole 1999). Many of these enzymes become stabilised in the soil, persisting and acting long after the original source has itself become degraded (Haynes 1986a).

Ammonification completes the mineralisation of organic compounds to NH₄⁺ (Stevenson and Cole 1999). It is carried out by enzymes released by other groups of bacteria and fungi that remove the functional amine (-NH₂) from the simpler compounds. The addition of protons (H⁺) to produce NH₄⁺ completes ammonification. The activity of these enzymes depends on simultaneous microbial growth (Haynes 1986a).

A further transformation called nitrification rapidly oxidises the NH₄⁺ released during mineralisation to NO₃⁻. Two specific chemoautotrophic bacteria are largely responsible for this process. In the first step *Nitrosomonas spp.* and some other members of the Nitrobacteraceae convert NH₄⁺ to NO₂⁻, sourcing their energy from the oxidation process and their C from CO₂ (Haynes 1986b). The NO₂⁻ is further oxidised to NO₃⁻ by *Nitrobacter spp.* Other autotrophic bacteria and some heterotrophs have been found that can nitrify NH₄⁺. They may make an important contribution in acid soils and where soil temperatures are high.

Nitrites are toxic to plant roots but do not readily accumulate in well-drained soils due to differences in the rates of the chemical reactions involved (Tisdale *et al.* 1993). That is, the conversion of NH₄⁺ to NO₂⁻ by *Nitrosomonas* proceeds more slowly than that of NO₂⁻ to NO₃⁻ by *Nitrobacter*. In the first reaction H⁺ is released, lowering the pH of the soil solution. Soil acidification is a natural function of N mineralisation. However, the addition of N fertilisers containing NH₄⁺ can increase problems of acidification due to higher rates of nitrification.

In the literature mineralisation is sometimes restricted to the processes of amination and ammonification with the resultant production of NH₄⁺. Nitrification to NO₃⁻ is considered to be a separate process (Whitehead 1995). However, the object of this study is to determine the availability of mineral N under kikuyu. For convenience therefore, mineralisation will be defined as the conversion of organic N compounds to both NH₄⁺ and NO₃⁻, encompassing both ammonification and nitrification in the one term.

Nitrogen immobilisation

The soil biomass is in continual competition with plants for available resources. Each gram of soil can contain from 10⁶ to 10⁹ bacteria and 10⁴ to 10⁶ fungi and actinomycetes alone (Stirling 2001). Although accounting for a relatively small fraction at 1 to 8% of total organic C, the biomass in a fertile temperate soil can be greater than 20 t/ha (Pankhurst and Lynch 1994).

Most of the species are heterotrophs, sourcing their energy requirements from organic C sources in the soil. They also need a regular supply of nutrients including N to build proteins and nucleic acids. As organic matter is decomposed and mineralised, microorganisms take up available NH₄⁺ and NO₃⁻, competing very effectively with plants and incorporating and immobilising N into organic compounds in the biomass (Stevenson

and Cole 1999). Carbon is lost from the soil system as CO₂ through the process of respiration.

In essence, immobilisation is the reverse of mineralisation although the enzymes and biochemical pathways involved in each process are different (Stevenson and Cole 1999). At any point in time and space in the soil, both mineralisation and immobilisation are taking place. If inorganic N ions are being produced at a faster rate than they are being taken up by microorganisms, net mineralisation is said to be occurring. Conversely, when NH₄⁺ and NO₃⁻ are being incorporated into organic N molecules in the biomass more rapidly than they are being mineralised, then there is a state of net immobilisation.

Immobilisation renders inorganic N unavailable to plants until the death and decomposition of the microorganisms, resulting in a temporary deficit of available N for plant uptake. If the decomposing substrate cannot supply the microbes with sufficient N they will take up any available N from the surrounding soil (Haynes 1986a). Nitrogen fertiliser may need to be applied both to compensate for the immobilisation and to meet the requirements of the crop or pasture (Tisdale *et al.* 1993). However, except for a short duration after application, N fertilisers are subject to the same processes in the soil as NH₄⁺ and NO₃⁻ mineralised from organic matter and can also be immobilised (Haynes 1986a).

C:N ratio

Microorganisms can respond rapidly to changing soil conditions, such as temperature, moisture and nutrients, with populations rising or falling within days (Reid and Wong 2005). An input of organic matter will result in an increase in microbial numbers as decomposition proceeds. The soil biomass has a C:N ratio between 8:1 and 10:1. The ratio of C to N in bacteria ranges between 3:1 and 5:1. Cellulose is a component of cell walls in fungi, resulting in a wider fungal C:N ratio of 15:1 (Whitehead 1995). Carbon and available N from the decomposing substrate and surrounding soil is utilised first by these rapidly growing populations for the synthesis of microbial cells. Only mineral N surplus to these requirements will be available for plant uptake.

Whether mineralisation or immobilisation of N occurs depends largely on the relative amounts of C and N in the substrate. In general, net mineralisation will occur if the organic matter is high in N relative to C, that is a C:N ratio less than 20:1 to 25:1 (Stevenson and Cole 1999; Whitehead 1995). This is equivalent to an N content greater than 2.0 to 2.5%. If the substrate has an N content less than 1.5%, the C:N ratio is greater than 30:1 and net immobilisation occurs. Between 20:1 and 30:1 there is an equilibrium between mineralisation and immobilisation. Microbial demand for N is equivalent to the amount released and available N levels in the soil do not change. This rule of thumb assumes that the compounds containing both C and N have similar decomposition rates.

As N is incorporated into the biomass and C is lost from the system as CO₂, the C:N ratio of the decomposing material narrows (Stevenson and Cole 1999). The rate of growth of the decomposer population slows with the reduction in available substrate. The biomass itself dies and is decomposed and mineralised. Thus, over time, a situation of net immobilisation will reach equilibrium and then proceed to net mineralisation.

Organic materials vary in their C:N ratios. Legume residues range from 13:1 to 25:1 whereas cereal residues have C:N ratios of 60:1 to 80:1 (Stevenson and Cole 1999). Animal manures are relatively low at 9-25:1 and woody wastes can be as high as 500:1. If

crop or pasture residues with a high C:N ratio are left on the soil surface or incorporated into the topsoil before planting, the germinating seedlings can be starved of N unless adequate fertiliser N is applied (Stevenson and Cole 1999).

Plants also differ in their C:N ratios depending on the species, stage of growth, plant part and N supply. Pastures that have received little or no N fertiliser average C:N ratios from 25:1 to 40:1 in their shoots and between 40:1 and 60:1 in their roots and litter (Whitehead 1995). Adding N fertiliser at high rates of 350-450 kg N/ha/year can reduce herbage ratios to less than 25:1 and root ratios to 25-30:1.

During decomposition organic molecules decay at different rates depending on the resistance of the substrate (Stevenson and Cole 1999). Proteins break down more rapidly than carbohydrates, followed by cellulose and hemicellulose, with lignin being the most resistant. Therefore, the accumulation or loss of plant available N ions depends not only on the C:N ratio but the chemical composition of the substrate. For example, a decaying woody waste with a very high C:N ratio should immobilise a high proportion of N. However, the relatively slow decomposition leads to lower immobilisation rates than would be expected on the basis of the C:N ratio alone (Whitehead 1995). Mineralisation of manure is slower than that of the plants consumed because grazing animals digest and utilise the fractions that are more readily broken down and excrete those that are more resistant.

Apart from the relative C and N proportions in decomposing substrate and that in the microbial biomass, the C:N ratio of soil organic matter itself needs to be considered. All three sources contribute to the turnover of N in the soils. Undisturbed topsoils in equilibrium with their environment have a C:N ratio of 10:1 to 12:1 (Tisdale *et al.* 1993). The C:N ratio of soil humus is also between 10:1 and 12:1 but the rate of turnover of N is five times slower than in the biomass due to the resistant nature of humic molecules (Stevenson and Cole 1999). The mineralisation of freshly added plant and animal material occurs more rapidly and produces more mineral N than that of humified organic matter despite the wider C:N ratio of most forms of litter.

Total C:N ratios were calculated for 111 dairy paddocks soil tested on the mid-north coast of NSW (Beale, unpublished data). Values ranged from 8.79 to 15.38 with a mean of 10.91. Fifty percent of the paddocks tested had a C:N ratio below 11.0. Given the C:N ratio of humus is approximately 11:1, those soils with an equal or lower ratio would have largely met the N requirement of their biomass. Therefore, with adequate soil moisture and temperature and assuming no significant additions of substrate, they could be in a state of net mineralisation. Given the range of total N in the top 10 cm of 2470 to 10,010 kg N/ha and assuming a net mineralisation rate of 2% (Stevenson and Cole 1999), these paddocks could produce from 50 to 200 kg mineral N each year. Additions of inorganic N fertiliser should be readily available to plants.

As the C:N ratio widens, the soils move to a state of net immobilisation and additions of fertiliser N would be first utilised by the microbial biomass to satisfy its own C:N requirements before plants could access the remaining N. Those soils from the mid-north coast with high C:N ratios may have had large inputs of residues from pastures with a low N status and, thus a high C:N ratio (D. Herridge, personal communication).

Mineralisation rate

Factors that affect the activity of soil microbes in turn determine the rate of mineralisation (Stevenson and Cole 1999). They include temperature, soil factors such as soil type, moisture content, aeration and pH, supply of substrate and populations of soil fauna and microorganisms. The narrow species diversity of nitrifying bacteria compared with those involved in decomposition and ammonification results in nitrification being influenced by environmental factors to a greater extent than the other processes (Haynes 1986a). Therefore, nitrification can become the rate limiting step in the overall process of N mineralisation.

Temperature

Temperature is a major factor affecting N cycling in soils. The different organisms responsible for the various stages of N mineralisation function within different temperature ranges (Haynes 1986a). For example, decomposing fungi, actinomycetes and bacteria are common in soils in the mesophilic temperature range of 0°C to 45°C, whereas the nitrifying bacteria have a much narrower temperature optima of 25°C to 35°C.

Variations in temperature do not affect the progress of decomposition but change the rate of tissue breakdown and the evolution of CO₂ (Haynes 1986a). The higher the temperature, the faster the rate of decay except in dry conditions when soil moisture becomes the limiting factor. Therefore decomposition proceeds more rapidly in tropical regions during the wet season than in more temperate climates.

Most research into mineralisation rates has been carried out under temperate conditions with the general conclusion that, assuming other factors are not limiting, a rise in soil temperatures between 5°C and 30°C increases N mineralisation (Whitehead 1995). Within this range an increase of 10°C will double the mineralisation rate (Tisdale *et al.* 1993). Studies with soils from hotter climates indicate that optimum temperatures for N mineralisation may have a wider range than shown by the temperate studies (Hoyle *et al.* 2006; Myers 1975).

Ammonification has a higher and wider optimum temperature range than nitrification, with the lower limit of the former process being at freezing point (Haynes 1986a). Myers (1975) incubated a clay loam soil from the Northern Territory at temperatures ranging from 20°C to 60°C. The rate of ammonification peaked at 50°C whilst the optimum temperature for nitrification was closer to 35°C. Soil temperatures at a depth of 2.5 cm at Katherine can reach 52°C on hot cloudless days in summer but never exceed 40°C at 20 cm depth (Myers 1975). Therefore NH₄⁺ ions could accumulate in the top layer of soil on hot days, to be nitrified rapidly at night when soil surface temperatures cool.

Hoyle *et al.* (2006) measured ammonification, nitrification and immobilisation rates in a red-brown earth from Western Australia with a clay content of 26%. The soil came from an established wheat:legume cropping rotation and was incubated at temperatures ranging from 5°C to 40°C. Immobilisation rates were equivalent to ammonification rates in the 5°C to 20°C range, suggesting that at low temperatures NH₄⁺ is rapidly consumed by the microbial biomass. Above 20°C nitrification increased linearly with temperature and dominated in the consumption of NH₄⁺ over immobilisation. Thus, NO₃⁻ may accumulate at higher temperatures due to the greater activity of nitrifying organisms compared to those involved in immobilisation (Hoyle *et al.* 2006).

There is some evidence that nitrifying bacteria can acclimatise to local conditions. Optimum activity of nitrifiers in soil from the Northern Territory occurred at 35°C with activity continuing up to 50°C. (Myers 1975). In comparison, Canadian soils reach maximum nitrification rates at 20°C with activity almost ceasing at 30°C (Haynes 1986b). There is conflicting evidence as to whether rapid temperature fluctuations of more than 10°C can affect N mineralisation. However, the death of large populations of soil microorganisms following a rapid drop in temperatures provides a readily mineralisable substrate with a low C:N ratio. Flushes of mineral N regularly occur with the first cold spell of autumn and/or a late frost in spring (Haynes 1986a).

Soil moisture

All metabolic reactions must occur in an aqueous solution and soil microorganisms are no exception to the rule. An adequate, though not excessive, supply of water is required for the biomass to function effectively (Haynes 1986b). Moisture stress can occur when too little water inhibits microbial growth or too much water in the soil pores restricts aeration and again limits the activity of microorganisms. In addition, denitrification of NO_3^- to N_2O and N_2 occurs under anaerobic conditions, reducing the net rate of mineralisation (Stevenson & Cole 1999)

Generally, if the soil temperature is above 5°C the rate of N mineralisation will increase as soil water status rises from permanent wilting point to field capacity (Whitehead 1995). Moisture potentials above and below these points will reduce activity. Fungi, actinomycetes and bacteria responsible for N mineralisation are relatively tolerant of dry soils whereas other bacteria can become inactive at 50% of field capacity (Haynes 1986a). Nitrifying bacteria can survive in dry soils in an inactive form (Whitehead 1995). At -700 kPa all available NH_4^+ will nitrify to NO_3^- in 21 days, dropping to half that amount at wilting point (Tisdale *et al.* 1993). Below wilting point the activity of ammonifiers continues whilst that of nitrifiers is inhibited.

Oxygen availability

Most soil fungi, actinomycetes and many of the bacteria are aerobes and require an adequate supply of oxygen to function (Haynes 1986a). As soil moisture rises above field capacity, more than 90% of the soil pore space is filled with water, oxygen becomes limiting and aerobic decomposition slows. Where severe and prolonged waterlogging occurs, anaerobic degradation of organic matter replaces aerobic decomposition (Stevenson and Cole 1999). Fermenting organic matter provides less energy for the biosynthesis of microbes whose activity is depressed resulting in soils with a high level of organic C.

Ammonification is carried out by both aerobes and anaerobes. In contrast, nitrifying bacteria are obligate aerobes and an adequate supply of oxygen (O_2) is required for the oxidation of NH_4^+ in the soil (Stevenson and Cole 1999). Therefore, in well-aerated soils NO_3^- is the predominant form of inorganic N. Ammonium ions accumulate in flooded or waterlogged soils where O_2 is limited, leading to a concurrent reduction in NO_3^- . Reduced decomposition also slows the turnover of N in waterlogged soils. Mineralisation is incomplete and less N is immobilised than in well-drained and aerated soils.

Soil structure and texture also play an important role in aeration (Tisdale *et al.* 1993). Clay soils can have a lower level of gas exchange than sandy soils, especially if compaction has damaged the structure and reduced pore size and connectivity. Organic matter

amendments and a healthy population of macrofauna can improve soil structure and aeration.

Wetting and drying cycles

The effect of drying and wetting cycles on decomposition has not yet been ascertained. Some studies indicate no effect whereas others have shown increased decomposition rates (Haynes 1986a). Nitrogen mineralisation is, however, enhanced when soils undergo alternate periods of wetting and drying (Stevenson and Cole 1999). The flush of mineral N decreases with successive cycles. Weather patterns that produce repeated wetting and drying are more common in tropical and sub-tropical climates than in temperate zones where soils can remain moist for extended periods (Birch 1964).

Stevenson and Cole (1999) suggested three reasons for increased N mineralisation. Repeated wet and dry soil cycles break down water soluble aggregates, exposing new sections of substrate and soil to microbial attack. Freezing and thawing have a similar effect. Organic N compounds are converted to soluble forms during drying, which can be more readily utilised by microbes. A third explanation is that drying partially sterilises the soil and N compounds are released as microbial cells undergo autolysis.

Reviewing studies of wetting and drying cycles, Haynes (1986a) found that dead microbial cells contributed 76% of a flush of N mineralisation when a dry soil was remoistened. The rate of decomposition of the dead biomass was five times greater than that of mobile soil organic N. In the four weeks after a dry/wet cycle mineralisation of microbial cells increased mineral N in the topsoil by 40 kg N/ha.

Birch (1964) found no evidence of decomposition of the microbial population in a study comparing the N mineralisation of kikuyu, meadow hay (scientific name not provided) and Rhonfa grass (*Phalaris tuberosa*). Three kikuyu samples with a total N content (air-dry basis) each of 1.55% N, 2.14% N and 2.49% N were ground and mixed with water and subsoil of a very low N status (0.045% N). Meadow hay (1.01 % N) and phalaris (2.90% N) were also ground and mixed with the subsoil. Eight repeats of each sample and a water/subsoil control were initially incubated for 15 days until a steady rate of decomposition was achieved. The samples were incubated for a further 33 days interrupted by one, two or three air-dry periods and subsequent remoistening.

With initial and final C:N ratios of 38:1 and 21:1 respectively, the meadow grass produced no net mineral N, the sample remaining in a state of declining immobilisation throughout the incubation (Birch 1964). As decomposition proceeded the kikuyu samples progressed from net immobilisation to net mineralisation. Nitrogen mineralisation increased both with the N content of the kikuyu and the number of wet and dry cycles to a maximum of 340 mg N/100 g kikuyu. The longer the period of drying, the more N that accumulated. The phalaris sample had an initial C:N ratio of 12:1 and accumulated mineral N throughout the treatment to a maximum of 742 mg N/100 g phalaris.

Birch (1964) concluded that plant material with an N content above 1.5% N (air dry), subject to wetting and drying cycles, will supply more mineral N than substrate that is maintained under constant moisture conditions. Increasing the frequency and length of the drying period can both reduce N immobilisation and increase the rate of mineralisation. Enhanced decomposition of the substrate following drying was given as the mechanism for improved mineralisation.

Soil pH

The activity of decomposers is greater in neutral than in acid soils (Haynes 1986a). There is a wide variation in tolerance between individual species. Earthworms avoid acid soils below pH 4.5 and numbers in a South Australian study doubled when soil pH was raised from 4.1 to 6.7 (Lines-Kelly 2004). In general as pH declines, increasingly acid conditions favour bacteria, then actinomycetes with fungi being the most acid tolerant of the soil microorganisms (Haynes 1986a).

A diverse variety of microorganisms are responsible for ammonification resulting in a low sensitivity of that process to soil pH (Haynes 1986a). Nitrification can take place in soils from $\text{pH}_{(w)}$ 4.5 to 10 (Tisdale *et al.* 1993), but rates are restricted outside the optimum range of $\text{pH}_{(w)}$ 7 to 9 (Haynes 1986b). Where other factors are not limiting, low pH values can reduce nitrification leading to an accumulation of NH_4^+ in acid soils whereas NO_3^- becomes the dominant form of mineral N in neutral and alkaline soils (Haynes 1986a). *Nitrobacter* are inhibited more so than *Nitrosomonas* in alkaline soils above pH 8, which can lead to an accumulation of NO_2^- (Whitehead 1995).

The effects of aluminium (Al) toxicity on nitrifying bacteria are thought to be the main problem in acid soils (Haynes 1986b). However, in an incubation of 40 acid soils, Nyborg and Hoyt (1978) found no correlation between nitrification rates and the concentrations of iron (Fe), manganese (Mn) or Al. Instead they attributed depressed nitrification to the high concentration and activity of protons (H^+) in acid soils.

There is some evidence that nitrification may occur in acid soils due to the presence of a variety of heterotrophic nitrifiers that are less sensitive to pH (Haynes 1986b). *Nitrobacter* may be more prolific and active in microsites in the soil that are less acid than bulk soil testing across a paddock would indicate. There are also species and strains of nitrifying bacteria that have adapted to acid conditions such as *Nitrosospira spp.* that is active down to pH 4.1 (Haynes 1986b).

The mid-north coast region of NSW has predominately acid soils. Of the 111 dairy paddocks tested by Beale (unpublished data), 50% were below $\text{pH}_{(\text{CaCl}_2)}$ 4.90 with a range from 4.20 to 6.40. Liming acid soils stimulates nitrification more so than ammonification (Haynes 1986b) as well as providing a vital source of calcium for earthworms and bacteria and making other nutrients more available in the soil solution (Lines-Kelly 2004). In a laboratory incubation, Nyborg and Hoyt (1978) doubled the amount of N mineralised in 40 acid soils ($\text{pH}_{(w)} = 4.0\text{-}5.6$) when they raised the pH to 6.7 with lime. However, they found that the effect of liming on N mineralisation in the field, measured as N uptake by oats (*Avena sativa*), was only temporary. Increases in mineralisation rates of 1 to 2% were significant ($P < 0.05\%$) in the first 2 years after liming but decreased rapidly in the third year.

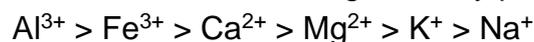
Edmeades *et al.* (1986) applied lime at 7.5 t lime/ha on a ryegrass-white clover pasture with a soil $\text{pH}_{(w)}$ of 5.7 in New Zealand. Over a two year period total uptake of soil N by the ryegrass was increased by 50 kg N/ha although the uptake of N by clover was unchanged. The researchers concluded that the lime application increased the rate of N mineralisation in the soil and that the effect would be more pronounced in more acid soils.

Nutrient deficiencies and toxicities

Soil organisms gain most of their nutrients from decomposing substrate or the soil solution. Nitrogen is required in the greatest amounts and is, therefore, usually the most limiting nutrient factor (Haynes 1986a). Phosphorus can also become limiting although this does

not often occur in dairy pastures. Relatively high use of phosphate fertilisers, effluent reuse and the spreading of poultry manure in regions where both enterprises are common can lead to high levels of P in soils. Beale (unpublished data) tested 111 dairy paddocks for Colwell P on the mid-north coast of NSW. Most were in the high to very high categories for P Cowell (Fert\$mart 2013) with only 10 paddocks being adequate or marginal for P.

Increasing the concentration of mineral salts in the soil solution can increase the rate of ammonification and hence net mineralisation. Haynes (1986a) provided three theories for this increase in a review of the general salt effect. High salt concentrations are toxic to microorganisms and plasmolysis of microbial cells would release organic compounds that could be readily mineralised by the surviving population. Osmotic effects on litter would also extract organic compounds from intact plant cells, with chemical reactions splitting $\text{NH}_4^+\text{-N}$ from carbon complexes. In addition, the capacity of cations to stimulate N mineralisation is in the same order as their exchange on clay particles. That is:



Thus, as NH_4^+ is removed from nitrogenous compounds, it could be replaced from cations in the soil solution (Haynes 1986a).

Micronutrients can have a stimulatory effect on decomposition and the N cycle when applied in trace amounts. However, in larger quantities a number of the trace elements can be toxic and inhibit N mineralisation (Haynes 1986b). There is a deleterious effect on all stages of mineralisation with *Nitrobacter* being the most sensitive organisms. Acid soils and those low in clay and organic matter are most affected as the majority of trace elements are more available at low pH but are adsorbed by clays and humic molecules, reducing their toxicity (Haynes 1986a). Pastures that have been amended with biosolids or municipal solid waste are most at risk as are those on which rock phosphate, with an unknown heavy metal analysis, has been applied.

The nitrifying bacteria are also very sensitive to pesticides applied to the soil (Haynes 1986b). At normal application rates there is little effect from most pesticides although those in the urea, carbamate, thiocarbamate and aminotriazole groups are most likely to inhibit nitrification. Other pesticides suppress the activity of *Nitrobacter* leading to a build up of NO_2^- in soils. Application of some pesticides can increase net mineralisation (Haynes 1986a). This occurs when a portion of the biomass is killed by the pesticide and becomes a source of readily available organic N for mineralisation by surviving populations.

Soil type

Mineralisation proceeds more rapidly in sandy than in loam or clay loam soils due to different C:N ratios and activity of the microbial biomass in soils of different textures (Whitehead 1995). Clay minerals sorb organic compounds, such as amino acids, proteins and nucleic acids, reducing their access to microbial attack and slowing the rate of decomposition and mineralisation (Haynes 1986a). Humic molecules also form complexes with clay minerals forming microaggregates that also limit physical accessibility of organic compounds to the microbial biomass. The disruption of these aggregates, for example following wetting and drying cycles in the soil, exposes new surfaces to biodegradation and increases N mineralisation.

Soil fauna and plants

Macrofauna, such as earthworms and dung beetles, accelerate the net mineralisation of N (Whitehead 1995). They consume plant litter and manure, utilising the organic compounds for their own metabolic needs and releasing CO_2 via respiration. Their own excreta, for example worm casts, have a lower C:N ratio than the original substrate and a higher

proportion of NH_4^+ and NO_3^- . Macrofauna also increase the surface area to volume ratio of litter and the light fraction for microbial attack, redistributing the substrate through the soil profile.

In a review of studies of earthworms and N mineralisation, Whitehead (1995) found a yield increase of 10% when earthworms were introduced to newly reclaimed Dutch polders and a sustained increase in grass yield of 25 to 30% when earthworms were added to a dense mat of peaty organic matter. New Zealand pastures benefited from 10 earthworms/kg soil that increased mineral N by 50% in 11 weeks. The rate of turnover of N is also enhanced by fungi and nematodes that feed on bacteria, using some of the C and N and excreting N in soluble forms (Whitehead 1995).

The roots of growing plants excrete exudates into the rhizosphere and continuously slough off older root tissue (Haynes 1986a) giving microorganisms a ready supply of energy and promoting their replication and growth until N becomes the limiting factor. Thus a state of net immobilisation could occur in the rhizosphere, restricting the supply of available N for plant uptake.

5. Kikuyu

Kikuyu is native to forest margins of the highland plateaux in central and east Africa where it grows at elevations of 1950 to 2700 m and a mean annual rainfall of 1000 to 1600 mm (Mears 1970). Seed was originally introduced to Australia from the Congo in 1919. Following germination at the Sydney Royal Botanic gardens, the kikuyu plants were grown at the Hawkesbury Agriculture College from where cuttings were distributed throughout the mainland states of Australia from late 1920. Later introductions of kikuyu were made from Kenya (Mears 1970).

Kikuyu is a perennial grass producing both stolons and rhizomes (Hanna *et al.* 2004). The stolons are long, prostrate and multi-branched bearing numerous short culms and rooting strongly at the nodes. Leaf blades are alternate and linear, up to 15 cm in length. The basic repeating unit is the phytometer, consisting of the leaf blade and sheath, the internode and the node with its associated axillary bud (Moore *et al.* 2004). Phytometers are aggregated into tillers which arise from a single crown. Whereas the stems of other grasses only elongate during the reproductive stage, kikuyu stolons elongate continuously (Marais *et al.* 1987). The above-ground stolons, or runners, form a thick intertwining mat from which the leafy aerial tillers develop. Old tillers lodge and dead leaves accumulate with the rhizomes in the surface layers of the soil under the thatch.

Kikuyu is a tetraploid, apomictic grass ($2n = 4x = 36$) (Hanna *et al.* 2004). Seed is produced from spikelets hidden in the leaf sheaths of fertile shoots. Apomictic reproduction, the formation of viable seed without fertilisation, makes breeding difficult due to its instability and variation (Marais 2001). Little work has been done on the selection and breeding of kikuyu in Australia with only four cultivars being available – Whittet, Breakwell, Noonan and Crofts. Seed from the latter three may be very difficult to obtain in commercial quantities (Moore *et al.* 2006; NSW_DPI 2004). Vegetative propagation of cuttings from stolons is very successful under adequate soil conditions. Although common kikuyu is thought to only reproduce vegetatively (NSW_DPI 2004), its spread across suitable habitats in Australia and other countries may have been due to the germination of viable seed in manure (Hanna *et al.* 2004).

The advantages and disadvantages of kikuyu as a dairy pasture base have been reviewed in Australia by Mears (1970) and Garcia (2014) and elsewhere by Marais (2001) and Hanna (2004). This summary will focus on those aspects of kikuyu growth, production and management that have a bearing on its response to nitrogen (N) and factors that may affect N mineralisation and availability under the sward.

Factors affecting growth

Soils and nutrients

Kikuyu grows naturally on the deep, lateritic red loams of the African highland plateaux, which originated from volcanoes, and adapts well to similar soils in other countries (Mears 1970). Marais (2001) summarised the soil requirements for kikuyu to thrive. It prefers a well-drained soil though can tolerate some water-logging. Both high salinity and high acidity levels do not affect yield. However, calcium uptake is impaired as pH falls. Kikuyu growth is limited on shallow, infertile soils, especially those low in P, K, Mg, S, Fe, Mn and copper (Cu).

Phosphorus use efficiency is high but substantial amounts are needed to establish new kikuyu pastures (Miles 1998). Potassium can be taken up in luxury amounts with tissue concentrations up to 5% of dry matter. As K levels increase, NO_3^- uptake is stimulated. Miles (1998) recommended annual N fertiliser rates of 300-500 kg N/ha for maximum dry matter production with split dressings of 50-100 kg N/ha applied throughout the growing season to increase N use efficiency.

Photosynthesis

Kikuyu is a perennial warm-season grass with the C_4 photosynthetic anatomy and pathways typical of tropical grasses (Moore *et al.* 2004). The unique Kranz anatomy in the leaves of C_4 grasses enables atmospheric CO_2 to be first fixed into 4-carbon (4-C) intermediates in the mesophyll cells that encircle the bundle sheath. These 4-C acids are then transported into the bundle sheath where the photosynthetic reaction is completed. Sucrose is efficiently transferred out of the bundle sheath to the phloem and translocated to leaf sheaths, growing points and internode stalk tissues. Up to 70% of the sucrose produced can be translocated away from the leaf in one to two hours (Moore *et al.* 2004). C_4 grasses store less starch than C_3 dicots.

The steep CO_2 gradient between the atmosphere and bundle sheath, operating in conjunction with efficient photosynthetic enzymes, enables C_4 plants to overcome the negative impact of photorespiration common to C_3 species (Moore *et al.* 2004). Photorespiration occurs when the enzyme rubisco fixes oxygen and releases CO_2 , losing up to 25% of carbon (C) plus N as ammonia (NH_3) into the atmosphere (Leegood 2007). Relatively low levels of rubisco (Mathews *et al.* 2004) and elevated CO_2 levels in the bundle sheaths of C_4 grasses (Moore *et al.* 2004) overcome this problem. Kikuyu and other tropical grasses have greatly increased growth rates in full sunlight and warm conditions, up to twice that of C_3 plants. The C_4 advantage diminishes with decreasing light intensity. Kikuyu, which is only moderately shade tolerant, also loses this advantage as shading of the canopy increases (Hanna *et al.* 2004).

Water use efficiency

High photosynthetic rates due to the C_4 mechanism also give tropical grasses good water use efficiency (WUE). Water use efficiency for C_4 grasses can be twice that of their C_3 counterparts, despite similar transpiration rates and daily water use for whole canopies (Moore *et al.* 2004). A decrease in photosynthetic rate caused by low light intensity, high or low temperatures resulting in plant stress, low N status or low specific leaf weight, for example after grazing, will all reduce WUE. Tropical C_4 plants are generally thought to have better drought tolerance than C_3 species (Marais 2001). However, given the similar daily evapotranspiration rates, Moore (2004) questions this assumption.

Despite rooting to a depth of more than two metres (Garcia *et al.* 2014), kikuyu is more sensitive to water stress than other tropical grasses, especially under high evaporative demand (Murtagh 1988). It requires a minimum annual rainfall of 900 mm unless irrigated (Hanna *et al.* 2004). In a field trial at Wollongbar, northern NSW, Murtagh (1988) measured the growth of a dense monoculture of kikuyu under different soil water contents and rates of evapotranspiration. On a wet soil with a medium to high evaporative demand of 5 mm/day, growth was reduced by 61% compared to plots subjected to no water stress. As evapotranspiration decreased to 2 mm/day, growth was not affected until total available water was reduced below 50% of field capacity.

Moore (2004) reviewed the mechanisms underlying plant responses to water stress. Drying surface soils will result in reduced photosynthesis and slower growth despite good

turgor pressure in above ground parts and adequate water supplies in deeper soil horizons. This change is triggered by the production of abscisic acid (ABA) in roots exposed to a water deficit. Stomatal conductance and leaf area are reduced and assimilate is shifted to root growth in wetter parts of the soil, where water can be exploited more efficiently. These changes may continue for one to two days after rainfall or irrigation, until all of the ABA has been metabolised. The osmotic potential at which turgor in shoots begins to decline depends on the species, light level and acclimatisation to stress. Other C₄ drought mechanisms are: leaf rolling to avoid direct solar radiation and reduce leaf area; leaf abscission; and regrowth from buds in crowns, stolon and rhizomes when adequate water returns to the soil profile (Moore *et al.* 2004).

Temperature

Temperature affects many aspects of plant development and growth. Given other factors are non-limiting, photosynthesis and vegetative growth increase as temperatures increase from the cardinal base to optimum level (Moore *et al.* 2004). Moore (2004) reviewed studies of base, optimum and maximum critical temperatures for several C₄ grasses. Optimum daytime temperatures ranged between 30°C and 38°C with night temperatures from 25-28°C, some 10°C hotter than for C₃ grasses. The rate of C₄ grass growth slows appreciably below 10°C.

Plant maturation is also accelerated with increasing temperatures, shortening the time to flowering (Moore *et al.* 2004). However, the progressively aging tissues have a lower nutritive value. Increasing deposition of cellulose, lignin and silica all reduce digestibility more rapidly in leaves than in stems (Marais 2001). There is little effect on the concentration of non-structural carbohydrates which accumulate in tropical species at a lower rate than in temperate grasses.

Originating in the high plateau country of Africa, where mean temperatures range from 2-22°C (Mears 1970), kikuyu has a lower optimum temperature and narrower range than other tropical grasses. Growth ceases below 8-10°C (Marais 2001) and is restricted when maximum temperatures drop below 21°C (Colman and O'Neill 1978) or rise regularly above 30°C (Hanna *et al.* 2004). Temperatures above or below the optimum will slow growth and increase digestibility (Marais 2001). Light frosts (< 3°C) will desiccate exposed herbage (Mears 1970) and moderate frosts (-3°C to -5°C) kill top growth, leaving the stolons unaffected (Marais 2001). Sustained winter frosts will kill kikuyu (Mears 1970).

Pasture growth and utilisation

Production from kikuyu pastures depends on temperature, moisture supply and reserves of soil nutrients (Miles 1998). Given temperatures in the optimal range and adequate soil water, N is often the most limiting factor for growth. Other factors, such as solar radiation and differences between cultivars can also explain variations in yields (Garcia *et al.* 2014).

Kikuyu yields between 25-30 t/ha have been reported from trial sites in New South Wales (Garcia *et al.* 2014). At Wollongbar on the north coast, Colman and O'Neill (1978) measured an annual yield of 31.4 t/ha of kikuyu from successive harvests of a dryland trial fertilised with 1345 kg N/ha/year. Neal *et al.* (2013) compared irrigation regimes for kikuyu grown at Camden, west of Sydney. The trial pastures received 100 kg N/ha at sowing plus 80% of N removed in harvested herbage. At field capacity an average of 25 t DM/ha/year was harvested compared with 17 t DM/ha/year from kikuyu irrigated to only 33% of optimal soil water status. By the third year of the trial, replacing N at 80% of removal had

depressed the kikuyu yield by 4.4 t DM/ha. The yield was restored when the N fertiliser rate was increased by 50% in the fourth year.

Garcia *et al.* (2014) summarised annual statistics from other published work on kikuyu trials. Mean yields were 12.2 t DM/ha/year with more than 1000 mm of water applied from rainfall and irrigation and fertilised with approximately 500 kg N/ha. However, production of unfertilised kikuyu without irrigation is more modest. Mears and Humphries (1974) measured daily growth rates of kikuyu at Wollongbar on the north coast of NSW. Growth rates ranged from 30 kg/ha/day where no N fertiliser was applied to 103 kg/ha/day where sufficient N (686 kg N/ha) had been applied so as to be non-limiting.

Estimated yields from daily growth rates on the north coast are 8.4 t DM/ha/year and on the mid-north coast, 6.8 t DM/ha (McDonald 2004). Figure 6 shows the estimated daily pasture growth rates of kikuyu for the two regions. Growth rates peak in February at 54 kg DM/day in the north and at 50 kg DM/day in March on the mid-north coast. Growth virtually ceases during the winter months. Seasonal variability can result in kikuyu production as much as 114% higher than the average in a good summer and 80% below average in a poor autumn on the mid-north coast (McDonald 2004).

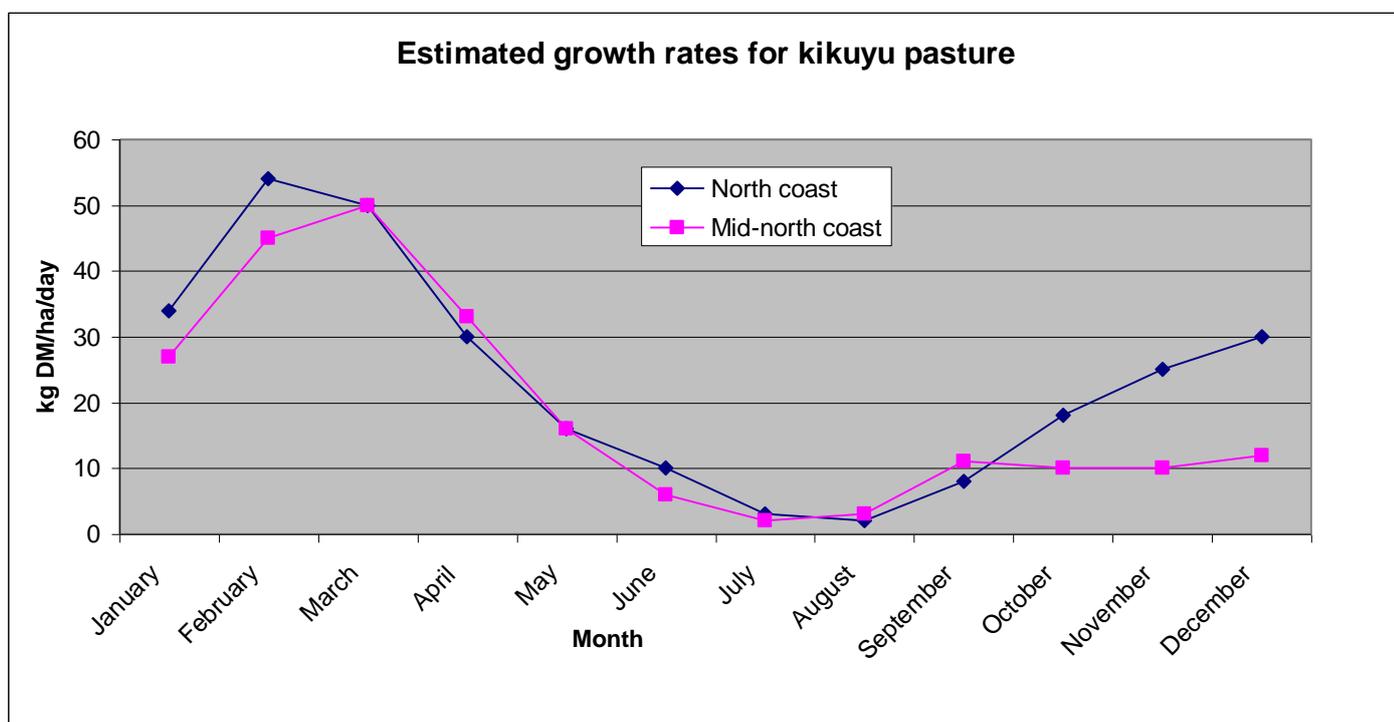


Figure 6 - Estimated pasture growth rates for unfertilised kikuyu on the north and mid-north coasts of NSW (Source Macdonald 2004)

Pearson *et al.* (1985) measured growth rates of three varieties of kikuyu fertilised with three levels of N at Bega, Camden and Taree on the NSW coast. Nitrogen was applied at 0, 40 and 120 kg N/ha 10-14 days after sowing and again following each harvest. Yield decreased with latitude, averaging 14.33, 12.99 and 5.58 t DM/ha/year at Taree, Camden and Bega respectively over the two year period of the trial and averaged across all treatments. The mean kikuyu yield at Taree was more than double that estimated by McDonald (2004). This highlights the problem of the dearth of regional field data relating to kikuyu.

The cultivar Crofts consistently produced the highest yield, with a maximum productivity of 18.46 t DM/ha/year, followed by Common and then Whittet kikuyu (Pearson *et al.* 1985).

Growth rates were also highest for Crofts, peaking at 140 kg DM/ha/day at the highest N rate for all locations and averaging 92 kg DM/ha/day across the three N treatments. There was a linear relationship between fertiliser N and kikuyu yield, although growth responses were markedly seasonal, reaching a peak in late January/early February at 0.75 kg DM/ha/day per kg N applied.

Under field trial conditions, utilisation of kikuyu pastures can be as high as 75% (Garcia *et al.* 2014). However, pasture yields and utilisation in plots are usually measured under conditions where water and nutrients are either not limiting or controlled as independent variables. Garcia (2014) summarised actual pasture utilisation across Australia. Utilisation of ryegrass pastures across Victoria averaged 7t DM/ha/year compared with 3 to 4 t/ha for home grown feed in Queensland. The higher net dry matter production and lower net utilisation of tropical versus temperate pastures results in more residues being left on the paddock after grazing. Any pasture that is not consumed by the dairy herd or conserved as feed is available for decomposition and mineralisation in the soil and is a potential source of N for microbial and plant uptake.

Nitrogen in kikuyu

Under normal growing conditions the concentration of N is greater in leaves than in stems and in shoots than in roots (Whitehead 1995). The ratio of leaf to stem tissue is critical for the chemical composition, nutritive value and dry matter production of grasses and also for the total N content and N concentration of the shoots. Stems are less digestible than leaves as the xylem becomes lignified and sclerenchyma cells increase in concentration in the vascular bundles (Marais 2001) to provide support.

Dairy farmers are advised to graze kikuyu when there are 4.5 leaves/tiller, the stage at which forage quality is optimised (Reeves *et al.* 1996). Once the fifth and sixth leaves emerge the leaf:stem ratio begins to decline along with the crude protein content of all the leaves. With increasing maturity there is less leaf tissue and the proportion of both stem and dead material increases.

Marais (2001) analysed the results of 20 samples of plant fractions from studies into the nutrient composition of kikuyu. The samples included both leaf and stem and were taken at various growth stages and plant heights, manually plucked, clipped or retrieved from the oesophagus of grazing animals. Total N ranged from 13.6 g N/kg DM (1.4% N) for stem at 51-87 day growth to 41.1 g N/kg DM (4.1% N) for leaf at 28 days of growth. This has implications both for feed quality and the C:N ratio of residues left on the paddock. As leaf is consumed by dairy cows and stems age the C:N ratio of the remaining thatch increases.

Nitrogen concentrations can also be derived from feed quality data. To simulate intake by milking cows at Wollongbar on the north coast of NSW, Reeves (1996) plucked kikuyu samples from pasture trials fertilised monthly with 100 kg urea/ha. An average crude protein content of 20.1% was measured. Using the general equation for crude protein [CP = total N x 6.25] (Freer *et al.* 2007), the average total N content of the plucked samples can be calculated as 3.2% N.

Garcia (2014) summarised results from 49 studies into the nutritive quality of kikuyu-based pastures. The studies had been conducted across a number of countries and regions with different cultivars, treatments and plant materials. The mean value for crude protein (CP) was 17.8% with a range of 12.1% CP at the 25% quartile to 22.8% CP at the 75% quartile.

Using the same equation as above the mean total N of the kikuyu samples analysed was 2.7% N.

Few studies into the N concentration of kikuyu have been undertaken in Australia. In a two year field trial at Wollongbar, Mears and Humphries (1974) studied the effect of N fertiliser rate and stocking rate on the growth and N concentration of kikuyu pastures. Angus weaners were stocked at rates ranging from 2.2 head/ha up to 16.6 head/ha on the paddocks receiving high N fertiliser regimes. Ammonium nitrate was applied at 0 (N₀), 134 (N₁₃₄), 336 (N₃₃₆) and 672 (N₆₇₂) kg N/ha. There were two replicates of each treatment and additional fertiliser was applied at 350 kg N/ha to one half of a caged quadrat in each of the paddocks.

The N concentration of whole tops ranged from 1.13% at the lowest stocking rate and nil N fertiliser to 3.15% at 7.4 head/ha and N₁₃₄ receiving additional N. Increasing the N rate raised the N concentration in all plant parts in the following order:

leaf > stem > rhizome > litter > roots.

This gradient supports findings by Moore (2004) that N compounds in C₄ grasses are rapidly translocated to the leaves from underground organs. Leaf N at the high stocking rate increased from 1.6% (N₀) to 4.0% (N₆₇₂) whereas root N rose from 0.8 to 1.4% for the same treatments and litter from approximately 1.0 to 2.3% N. This was equivalent to a rise in the amount of N stored in litter from 15 kg N/ha (N₀) to 75 kg N/ha (N₆₇₂).

Mears and Humphries (1974) took root sample cores and estimated that the underground organs had a collective dry matter yield of 5000-10,000 kg/ha in the top 10 cm. The effects of both N and stocking rates were variable and not significant on yield of roots and rhizomes. At an average N concentration of 1.2%, these underground parts could store 60-120 kg N/ha.

Stocking rate did have a significant effect on the relative concentrations of N in the leaf, stem and rhizomes of kikuyu over a 20 month period (Mears and Humphreys 1974). As stocking rate increased, so did the N concentration of leaves and stems, whereas it fell in the rhizomes. At a high stocking rate (11.1 head/ha), 78% of shoot N was allocated to the leaves compared with 68% at the low stocking rate (4.9 head/ha). The higher intensity of grazing increased both the rate of defoliation and the rate of N return to and cycling within the paddock. The normal pattern of N translocation from roots to shoots was accelerated with an equivalent reduction in the N content of the litter.

This has implications for strip grazing on dairy pastures. Applying very high stocking pressures to small areas for only a few hours may increase the N concentration of kikuyu leaf which is grazed, but leave stems and litter with a reduced content of N. However, in the Mears and Humphries study (1974), dry matter yield averaged over 20 months at the low stocking rate for all N treatments had a mean value of 3720 kg DM/ha, double that of the high stocking rate (1865 kg DM/ha). Gains in N concentration may be at the expense of overall yield but improve pasture utilisation.

The relative proportions of green matter and litter were affected significantly by an interaction between stocking rate and N level (Mears and Humphreys 1974). At the lower stocking rate of 4.9 head/ha, litter accounted for 62% of total above-ground yield for the low N applications (N₀ and N₁₃₄). The proportion of litter fell to 48.5% at both higher N rates of N₃₃₆ and N₆₇₂ for the same stocking rate. There was little change in the leaf:litter ratio when kikuyu with no N fertiliser was grazed at the higher stocking rate of 11.1 head/ha. However, the other three N treatments had an average litter proportion of 53.6%.

Mears and Humphries (1974) also compared the uptake of N by kikuyu in N-limited and non-limiting conditions in the same field trials. Where no additional N was supplied N uptake was greatest in August at 1.6 kg N/ha/day, more than double the N uptake during summer (0.7 kg N/ha/day). The addition of 350 kg N/ha not only increased the rate of N uptake, but also the response during summer with 3.7, 4.8 and 9.0 kg N/ha/day being taken up in August, November and January respectively. No effect of stocking rate on N uptake by kikuyu was found over the two year period.

Pearson *et al.* (1985) measured the total N concentration of kikuyu (cv. Crofts) on a monthly basis in a field trial at Taree. Figure 7 shows the results averaged across three N fertiliser treatments of 0, 40 and 120 kg N/ha. Nitrogen concentration peaked in April when growth was beginning to slow due to declining temperatures. Nitrogen content of the shoots increased with applied N resulting in mean concentrations of 2.03, 2.83 and 3.75% N in shoots respectively for the three fertiliser treatments.

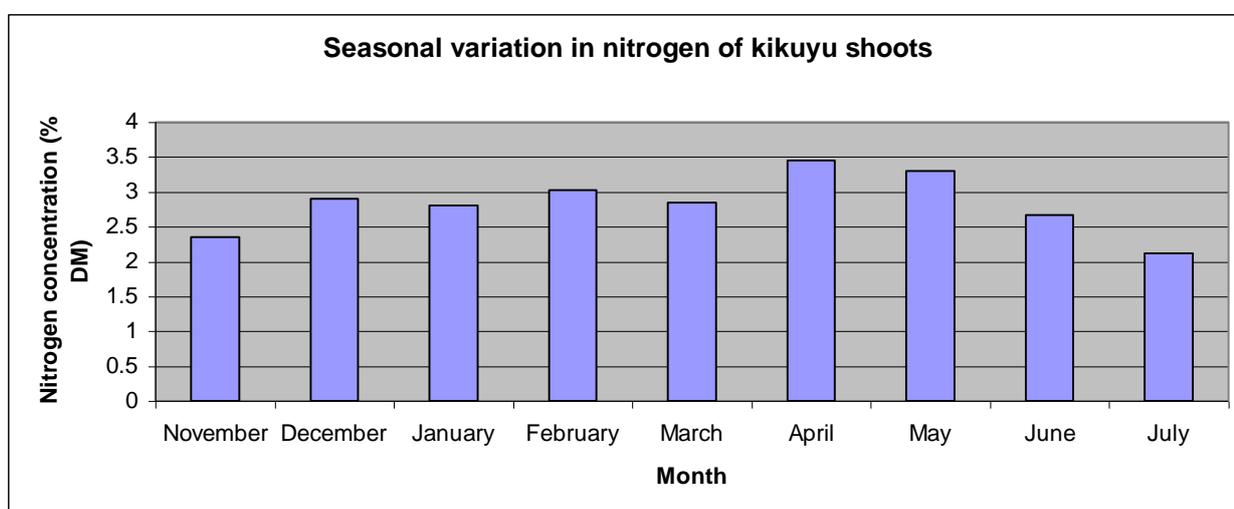


Figure 7 - Seasonal variation in the N concentration of kikuyu grown at Taree [Source Pearson *et al.* (1985) Table 4, pg 115]

C:N ratio of kikuyu

Carbon to nitrogen ratios can be calculated once the total C and N contents of the substrate or soil are known. Ideally, data relating to 'readily available' fractions, such as dissolved organic C and N, would be more useful than total C and N but the former are less easily measured (Whitehead 1995). Also, information about the C content of kikuyu is not recorded in the literature and, until suitable studies are undertaken, extrapolations need to be made from existing research.

Plants contain 40 to 50% C by dry matter weight (Haynes 1986a). This range has been narrowed for grasses by Garnier and Vancaeyzeele (1994) who measured the C and N content of seven pairs of annual-perennial temperate grass species. Annual species had a mean C concentration of 38.6%, slightly higher than the mean for perennials of 37.9%. The range in the perennials was greater than for the annuals, from *Poa pratensis* with 35.9% C to *Bromus erectus* with a C concentration of 39.4%. Both annuals and perennials allocated similar proportions of C to their various plant parts, 48.5% of C to leaves, 18.4% to sheaths and 33.1% to roots. A carbon content of 37% can be calculated for one kikuyu

sample for which the N concentration was measured (1.55% N) and a C:N ratio of 24:1 given by Birch (1964). However, no information was provided relating to the derivation of the C content in the C:N ratio.

Most C₄ grasses, including kikuyu, have a lower digestibility than temperate species (Garcia *et al.* 2014). Rapid growth and the development of structural cell components lead to relatively high proportions of cellulose and hemicellulose, both long chain carbon molecules. Therefore, for the purposes of calculation of C:N ratios in this study, the concentration of C in kikuyu will be set at 40%, slightly higher than for the temperate species measured by Garnier and Vancaeyzeele (1994) and that calculated from Birch (1964).

Table 1 shows C:N ratios for the litter and above and below ground organs of kikuyu harvested from field trials where the kikuyu was grown at different N rates. They were derived from N contents measured by Mears and Humphries (1974) with a C concentration of 40% assumed for all plant parts. These ratios indicate that only decomposition of fresh leaf from the three N fertiliser treatments and stem and litter at the highest N rate would lead to net mineralisation of the kikuyu substrate. The remainder of the plant parts at various N rates would, at best be in equilibrium during decomposition with the higher C:N ratios (> 30:1) leading to immobilisation.

This has consequences for dairy pastures on the mid-north coast of NSW where the low levels of N fertiliser applied to kikuyu might result in relatively high C:N ratios of the decomposing substrate. Net immobilisation of N into the biomass in autumn could reduce its availability to establishing ryegrass, both from that being mineralised in the soil and N applied as fertiliser. Compounding the problem is the management of kikuyu thatch. Strategies that remove the leaf, such as heavy grazing or mowing for silage increase the C:N ratio of the residue and thus the potential for net immobilisation.

By definition, where fresh leaf is left on the paddock, it must die and become litter for decomposition to occur leading to a widening of the C:N ratio. Nitrogen compounds are translocated away from the leaf during the early stages of senescence (Whitehead 1995). However, membrane disintegration restricts translocation in the phloem, nitrogenous compounds are leached from the leaf and autolysis releases cell contents into the soil. This could lead to a rapid flush of mineral N in the early stages of decomposition as soluble nitrogenous compounds are mineralised. Whether that N would be available for plant uptake or would be immobilised into the biomass depends on the relative contents of C and N in the surrounding substrate, its resistance to decomposition and the C:N ratio of the soil itself.

Table 1 - C:N ratios for kikuyu at various N fertiliser rates

[Derived from Mears and Humphries (1974)- Figure 5, pg 461]

	Applied N (kg/ha)			
	0	134	336	672
Leaf	25	19	16	11
Stem	40	33	24	15
Litter	40	33	24	17
Rhizome	50	40	31	24
Root	50	36	36	29

Nitrates in kikuyu

Kikuyu accumulates NO_3^- when uptake exceeds requirements for protein synthesis. This can occur under conditions of high N availability, either from excessive additions of N fertiliser or the recycling of nitrogenous compounds from manure and urine deposits of grazing animals (Marais *et al.* 1987). Factors that reduce growth rates, such as water stress, low light levels and low temperatures can stimulate NO_3^- accumulation as can high K levels in the soil.

Marais *et al.* (1987) measured NO_3^- levels in a long-established kikuyu pasture fertilised with 250 kg N/ha at the beginning of the growing season. On a dry matter basis the main stems contained 1.58% NO_3^- -N compared with the newest leaves at 0.39% NO_3^- -N. Nitrate levels rose as the leaf matured to the 3 and 4-leaf stage (0.59% NO_3^- -N) then declined to 0.51% NO_3^- -N in older leaves. Kikuyu stems contained 44% of the total NO_3^- present in the kikuyu tillers whilst only constituting 22% of the total dry matter. Complete tillers of kikuyu with a high N status contained 0.88% NO_3^- -N compared with 0.06% NO_3^- -N in low N status tillers.

Large vacuoles in the stem tissue are the main storage pool for NO_3^- in grasses and once stored, it is not readily translocated for assimilation into nitrogenous organic compounds (Marais 1987). It is either consumed by grazing animals or released into the soil during senescence and decomposition. Marais (2001) reviewed studies of NO_3^- levels in kikuyu finding a range of 0.03 to 0.8% NO_3^- -N. Garcia *et al.* (2014) summarised five kikuyu studies, calculating a mean NO_3^- -N concentration of 0.7% of dry matter. Nitrates stored in kikuyu represent a potential source of readily available N, especially in ungrazed or under-grazed pastures. A kikuyu pasture on the mid-north coast of NSW producing 8 t DM/ha/year with 0.7% NO_3^- -N contains 56 kg NO_3^- /ha. This does not account for N stored in organic forms.

High NO_3^- levels reduce the palatability of grazed kikuyu (Marais *et al.* 1987) and can be dangerous, especially for hungry livestock (Robson 2007). Nitrate poisoning occurs when feed containing a high concentration of NO_3^- is consumed. Diarrhoea, salivation and abdominal pain result from the caustic action of NO_3^- on the gut lining (Robson 2007).

Far more serious in ruminants is nitrite poisoning. Microorganisms in the rumen reduce NO_3^- to NO_2^- that, when absorbed into the blood stream, lowers the capacity of haemoglobin to carry oxygen. Severe nitrite poisoning leads to cyanosis and death. Nitrate concentrations greater than 1.5% NO_3^- -N are considered to be dangerous (Halpin and Hides 2008). Rumen flora are able to adapt to high concentrations of NO_3^- and NO_2^- over time, especially if supplied with a source of soluble carbohydrates (Robson 2007). They convert the NO_2^- to NH_3 that is then assimilated into microbial protein and ultimately made available to the ruminant via digestion in the abomasum and small intestine.

Dairy cows offered kikuyu pastures with a high leaf:stem ratio are less likely to be affected by nitrate and nitrite poisoning (Marais 2001). However, when cattle are forced to eat pastures down to the ground and consume a high proportion of kikuyu runners the risk increases. Conditions leading to NO_3^- accumulation, such as moisture stress, low light levels and low temperatures, often coincide with a feed deficit. The period immediately following the breaking of a drought poses the greatest risk, especially in autumn when kikuyu growth rates slow. Reduced plant uptake and lower levels of leaching during the drought result in the accumulation of NO_3^- in the soil (Robson 2007). In the first week

following rain plant uptake is rapid and NO_3^- accumulates if growth does not match NO_3^- supply to plant tissues.

When conditions favour the accumulation of NO_3^- dairy farmers might make best use of affected pastures by giving them a light graze and returning the remainder to the soil through heavy mulching. This would reduce the kikuyu thatch, utilise the available N in pasture regrowth or to establish ryegrass and potentially reduce fertiliser-N applications. Unfortunately, balancing NO_3^- availability between plants and livestock can be a minor consideration during the autumn feed gap when dairy farmers are striving to maintain dry matter intake from limited pasture availability.

6. Assessment of soil nitrogen availability

Nitrogenous fertilisers are the largest and most expensive input to Australian dairy pastures (Gourley *et al.* 2012). Their efficient use is vital, both for the productivity and profitability of the business and also the sustainability of the farm and surrounding environment. Dairy pastures on the mid-north coast of NSW contain, on average, 5 t total N/ha in the topsoil, providing a potentially valuable source of N. However, at any point in time, less than 0.1% or 5 kg/ha of N is available in a mineral form (Stevenson and Cole 1999). An accurate assessment of mineralisation rates and quantities of plant available N is essential to determine the timing and rates of strategic fertiliser N application and to optimise N use efficiency.

The plant-soil ecosystem is extremely complex and dynamic. Rates of N mineralisation in a soil cannot be measured simply by analysing routine samples, taken at a single point in time (Stevenson and Cole 1999). Multiple transformations of N can occur simultaneously in any portion of the soil. Ammonium ions are preferred by microbes as a source of N and may be immobilised as they are produced. In well-aerated soils any remaining NH_4^+ is rapidly nitrified to NO_3^- which in turn can be taken up by plants or lost by leaching (Tisdale *et al.* 1993). Denitrification can occur in waterlogged soils. Ammonium ions can also be fixed to clay particles in cracking clays. Volatilisation of NH_3 can occur in alkaline soils, although losses of mineralised NH_4^+ via this pathway are minor compared with volatilisation from urine, manure and some N fertilisers.

In general, net mineralisation leads to an increase of mineral N levels in the soil whilst immobilisation results in an increase in organic N levels (Whitehead 1995). However, in pastures where total inputs of N are greater than its removal in plant or animal product or transfer out by grazing animals, net mineralisation can occur with an accumulation of organic N. Elevated levels of mineral N may be relatively transient due to the reasons given above and a reduction in mineral N may not necessarily indicate net immobilisation.

Several methods are employed to determine mineralisation rates and the accumulation of mineral N in pasture soils. They include the assessment of available soil N in the field, laboratory incubations, chemical methods and the development of indices and models. However, it should be remembered that where sufficient field trials have been conducted to provide accurate local N fertiliser recommendations for a particular crop or pasture, mineral N supplied from the soil, though not directly measured, has been accounted for in the recommended rates.

Field assessments of available soil nitrogen

Plant available N can be determined from plots containing only grass species with no legume content or added N fertiliser (Whitehead 1995). Dry matter yields and the N content of tissue samples are measured from regular harvests through the growing season and N uptake determined. A summary of field trials using this method in the United Kingdom by Whitehead (1995) showed that available soil N under different cropping and pasture management systems could vary from 6-255 kg N/ha/year. Long term grasslands usually mineralised more than 100 kg N/ha/year.

The above method accounts for both mineral N present in the soil at the beginning of the trial as well as that mineralised during the season. It assumes that grasses take up the entire stock of available N in the soil and does not allow for N losses from leaching or denitrification. Also assumed is no net transfer of N from roots and stubble, that is, that the

amount of N in underground organs, stems and litter is the same at the end of the season as at the beginning. However, there is considerable movement of nitrogenous compounds throughout the plant via both the phloem and xylem (Haynes 1986d). N losses from leaching and denitrification, especially during periods of wet weather would also reduce the reliability of this method to accurately quantify mineral N availability in the soil.

Residual mineral N in the rooting zone of cropped paddocks can be measured to determine potential crop responses to applied fertiliser N (Goh and Haynes 1986). The number of samples taken in a paddock, time of sampling and depth of the profile sampled are all critical to provide an accurate assessment of residual mineral N in the soil. Both forms of mineral N, NH_4^+ and NO_3^- , should be measured. This method is most practical in arid and semi-arid environments with low rainfall and assumes that neither leaching nor denitrification are significant N loss pathways from the soil (Stevenson and Cole 1999). Spatial variability across a paddock is a major problem, reducing the usefulness of this method in grazed pastures due to urine and manure patches (Goh and Haynes 1986).

Incubation methods

Soil incubations provide biological indices as a measure of the potential of soils to provide mineral N for plant uptake (Goh and Haynes 1986). Short-term incubations are usually conducted over a period of 1-6 weeks with mineral N being measured before and after incubation. In long-term studies lasting up to 30 weeks, mineral N is removed and measured at regular intervals. Aerobic incubation measuring NO_3^- production in the soil requires a constant moisture content, a difficult variable to control. Waterlogging the soil for anaerobic incubation eliminates this problem, leaving only NH_4^+ to be measured (Stevenson and Cole 1999).

Any disturbance of soil samples before incubation can alter mineralisation rates, especially sieving, drying, grinding or mixing (Whitehead 1995). Incubations in the field can be carried out with soil samples placed in containers in the topsoil, though NO_3^- losses can occur via denitrification unless the samples are treated with acetylene to inhibit nitrification. Another *in situ* method involves the placement of tubes into the soil, some of which are covered to prevent leaching and including tubes with and without growing plants to estimate plant uptake of N (Whitehead 1995). Adequate replication is essential, especially in grazed pastures due to large variations across a paddock. Net mineralisation may be occurring at one point under a urine patch while another point, only centimetres away under a slashed windrow, may be in a state of net immobilisation.

Ghani *et al.* (2007) used the tube incubation method to determine the mineral N content of a grazed pasture soil in New Zealand over a period of 14 months. Lengths of PVC pipe (40 mm diameter, 150 mm length) were inserted into the soil, all herbage was removed and the upper end of each pipe was closed with 25 mm of cotton wool and capped with aluminium foil. Four pipes (replicates) were inserted into the soil for each incubation period of six weeks. Cattle were rotationally grazed for two days between each incubation period, at the end of which the soil cores were removed and analysed for mineral N using K_2SO_4 .

Quantities of mineral N in the soil, measured by the tube incubation method for each 6-week period, showed an initial peak of 55 kg N/ha in March followed by a relatively flat response of 15-20 kg N/ha from July to January (Ghani *et al.* 2007). However, a rapid acceleration of mineralisation through January resulted in a second peak of 130 kg N/ha in mid February. This rapid increase in mineralisation occurred after rain re-wet soil that had experienced a long drying period during a summer drought.

Another 93 soils from various agricultural and forestry land uses on the North and South Islands of New Zealand were also sampled by Ghani *et al.* (2007). They were incubated under anaerobic conditions for 7 days at 40°C before NH₄⁺ was measured in a flow injection analyser. Dairy pasture soils had an average mineralisable-N (NH₄⁺) content of 183 mg N/kg soil with a range of 107-324 mg N/kg soil.

Incubation methods are expensive and time consuming and, therefore, are not suitable for routine soil N tests (Goh and Haynes 1986). If results are to be reliably calibrated to the N requirement of a particular crop or pasture, samples must be collected for a given soil type, in a particular climatic environment and at the same time during the growing season (Stevenson and Cole 1999).

Chemical indices and predictive models

A number of chemical extraction methods have been developed to analyse the various forms of soil N. They range from mild procedures, such as hot water and hot salt extractions, to strong extractants based on concentrated acid or alkaline solutions (Goh and Haynes 1986). No single or simple method of chemical analysis can provide a reliable prediction of plant available N in pastures due to the multitude of variable factors affecting decomposition and mineralisation, such as rainfall and the heterogeneous nature of paddocks (Whitehead 1995).

Although both biological and chemical indices have shown good correlations with N availability in glasshouse experiments where variables can be controlled, they have not been replicated in the field (Goh and Haynes 1986). Recent studies have attempted to correlate one or more indices to actual mineralisation rates. For example, Ros *et al.* (2011) analysed over 2000 soil test results for extractable organic N (EON) and, although they found a positive relationship between EON and mineralisable N ($r^2 = 0.47$), it was no better a predictor than total N.

Schomberg *et al.* (2009) ran multiple regressions with a number of chemical indices for southern USA soils. None of the indices were good predictors of mineralisation rate, although the combination of four indices, total N, flush of CO₂, NaOH-N and cold-N, in a multiple regression was the best predictor of potentially mineralisable N ($r^2 = 0.94$). They concluded that the results of their study could be useful in modelling but would need to be tested with other soils of different clay types and with varying organic matter contents.

Gilmour and Mauromoustakos (2011) suggested that the problems associated with the accurate prediction of N mineralisation stemmed from the practice of combining soil organic matter (SOM) that is decomposing in the same pool as that which is being simultaneously mineralised. They separated the two pools and built a sequential model based on first order kinetics. That is, Pool I contained SOM from recent additions of organic matter to the soil (Gilmour 2011). Pool II was SOM that had been in the soil for some length of time and had already undergone a certain level of transformation. Total N was the sum of N in Pools I and II. The theory was that all of the N in Pool I would be mineralised before the decomposition of Pool II would commence.

The sequential model was tested on four long term studies with 108 soils from the USA, Israel and Australia, each having different cropping systems, tillage practices, organic amendments and soil depths (Gilmour and Mauromoustakos 2011). It was found to accurately model N mineralisation rates across the range of soils if the model was corrected for local temperature and moisture conditions (Gilmour 2011). However, simple,

reliable and repeatable indices are needed to move the method from the laboratory to routine testing in the field.

Data have also been collected and mathematical formulas derived based on ^{15}N tracing studies in an attempt to quantify mineralisation and immobilisation rates (Stevenson and Cole 1999). However, the calculations and models give an approximation only as they are often based on assumptions that may not be valid in the field. Inputs with ^{15}N only trace the fate of the labelled nitrogen molecules but do not provide information about over-all gains and losses for the whole soil system, especially losses from leaching, volatilisation and denitrification. Any results obtained from ^{15}N tracing are specific to the particular soil-pasture system being studied and its characteristic N balance sheet which may not apply to a different soil or pasture (Stevenson and Cole 1999).

In Australia, the biophysical simulation DairyMod is used as a research tool to model pasture growth, management and utilisation as well as other aspects of farm production (Johnson *et al.* 2008). It incorporates nutrient uptake and N fixation, the allocation of photosynthates to various plant parts, growth and senescence and the effect of trampling from grazing animals. Both C_3 and C_4 grasses as well as legumes can be included in the pasture composition of each paddock which can be grazed or subjected to a field trial. Rainfall, infiltration and drainage can be simulated, nutrients applied, and the turnover of organic matter followed from fast to slow and then inert pools.

DairyMod also models some aspects of the soil N cycle including nitrification and denitrification (Johnson *et al.* 2008). Its usefulness in the current research into N mineralisation under kikuyu is yet to be tested. However, it must be remembered that all models are only as good as the underpinning knowledge of the physical, chemical and biological processes involved, the accuracy of the data included and the relevance of the assumptions made. Modelling a system as complex as the soil N cycle compounds the problem. Not all pools and fluxes within the soil N cycle can be accurately evaluated (Stevenson and Cole 1999). A small estimation error at one point in the cycle, for example mineralisation rate, can introduce a much larger error at another point, such as the amount of mineral N available to the pasture.

6. Conclusion and recommendations

The soil N cycle under kikuyu can be likened to a very large and complex jigsaw puzzle for which most pieces are currently missing. Although we know, to a certain extent, what is happening under the kikuyu sward we are not yet able to quantify the processes involved. Theoretically, if the appropriate data were available, net mineralisation and immobilisation rates could be determined and the availability of N estimated. However, most of the required research has not been conducted, published and/or replicated. Therefore, these recommendations will focus on surveys and field trials on dairy farms that will fill the information gap for kikuyu pastures on the mid-north coast of NSW. They assume that budgets and resources are not limiting.

1. *On-farm surveys and sampling*

Aim: To collect on-farm data relating to the growth and management of kikuyu pastures on the mid-north coast of NSW.

Rationale: Apart from a small number of trials conducted almost 30 years ago, most of the information about kikuyu growth and management on the mid-north coast is anecdotal or observational in origin or is extrapolated from other regions. Trial data often does not accurately represent the conditions under which kikuyu is managed on the farm. Information from a survey of management techniques and data collected will provide a baseline that will inform the field trials.

Sample population: Survey and sampling to be conducted on a minimum of 12 dairy farms across the region selected to represent the range of geographical locations, microclimates and soils (E.g. lower catchment, middle catchment and plateaux) and management strategies. Initial sampling could form the basis of an ongoing regional survey.

Data collection: Soil tests including total N; kikuyu tissue tests; paddock histories including previous soil tests; local rainfall and temperature records; soil fauna; pasture management strategies, especially fertiliser application rates and timing, N management and kikuyu thatch management.

2. *Field trials*

A series of field trials is recommended to establish regional data for kikuyu growth and N availability under the sward.

a. Kikuyu growth trials

Aim: To determine growth rates, soil N and tissue N concentrations of a pure kikuyu sward grown under a range of N fertilisation rates and moisture regimes.

Rationale: No recent growth trials have been conducted for kikuyu on the mid-north coast. Water and N supply are the two main limiting factors affecting kikuyu growth, given the limitations of seasonal temperature variations, and are variables that can be manipulated by farmers. Nitrogen uptake also affects the N concentration of various plant parts and ultimately the C:N ratio of decomposing litter. No tissue C tests have been reported for kikuyu.

Site selection: Three dairy farms, one selected from each of the three main geographical locations surveyed in Recommendation 1.

Trial design: Fully randomised, replicated block design conducted over a period of two years investigating the interaction between N fertiliser rates and timing, single vs split applications and soil water regimes.

Note – number of replicates and plot size need to be sufficient to allow a split plot design to be implemented for the kikuyu/ryegrass trial in the second year.

Data collection: Pre and post soil tests; strategic total and mineral N tests; dry matter yields of leaf, stubble and litter; tissue C & N tests of leaf, stem, litter and roots.

b. Mineralisation trials

Aim: To determine N transformations and mineralisation activity in the soil under kikuyu.

Rationale: Nitrogen transformations under kikuyu have not been investigated. Until valid data is available, strategic N use through the kikuyu growing season that will improve N availability for ryegrass cannot be determined.

Site selection: Plots from the kikuyu growth trials that cover a range of soil and substrate C:N ratios, including those that will be allocated to the kikuyu/ryegrass trial.

Trial design: *In situ* incubation study after the method of Ghani *et al.* (2007).

Data collection: Total soil C and N; microbial biomass C and N; mineralisable N; dissolved organic C and N.

c. Kikuyu/ryegrass trials

Aim: To determine strategic N use in kikuyu that will improve N availability for ryegrass establishment and growth, focussing on the transition period from kikuyu to ryegrass.

Rationale: Current recommendations for N fertiliser application for ryegrass are made from field trials with a prepared seedbed and harvested by the cut and carry method. Although more difficult to conduct, a trial where the ryegrass is oversown into kikuyu and the harvest method better approximates grazing, could provide more information to farmers and allow a benefit cost analysis of various N rates applied to kikuyu as well as strategic N use on ryegrass.

Site selection: As for the kikuyu growth trials.

Trial design: Split plot design established in the second year using one-half of each replicate from the kikuyu growth trials. Strategic N application for ryegrass is to be determined from the results of the mineralisation trial. A proportion of the harvested kikuyu should be returned to the plots.

Data collection: Soil tests including total and mineral N; ryegrass establishment; ryegrass and kikuyu dry matter yields; cost-benefit analysis.

Other factors affecting N mineralisation that could be investigated in field trials include:

- Kikuyu thatch management for ryegrass establishment
- Wetting and drying cycles
- Addition of lime to raise soil pH

It is also recommended that the field trials are designed to collect data required to calibrate DairyMod for kikuyu and that that data is made available to DairyMod.

8. References

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