NITROGEN SUPPLY FOR ORGANIC CROPS

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DECLARATION

I, Elizabeth Anne Stockdale, declare that this thesis was composed by myself, and the work described was carried out by myself, except for the instances detailed in the text and acknowledgements.

Elizabeth Stockdale

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ABSTRACT

An integrated series of field, laboratory and pot experiments was carried out between 1990 and 1993 to study the release of nitrogen from organic manures and its subsequent recovery by crops. The aim was to increase understanding of the soil processes controlling N release from manures and therefore enable N supply to be more closely matched to crop demand in organic cropping systems.

The study of N release from manures is handicapped by the lack of appropriate methods to measure rates of mineralisation (both net and gross) in the field. The use of isotope dilution techniques under field conditions was found to be difficult due to the slow diffusion of ammonium ions in soils. The release of N from manures was therefore studied indirectly by monitoring plant uptake and changes in the soil mineral N pool. Indices, used to predict N release, were not found to be applicable where additions of manure had been made.

Various management strategies aimed at maximising N supply for organic crops were studied. The N released from manures in the first year was shown to be derived mainly from the pool of mineral N added in the manure. The availability of this pool was controlled by the supply of soluble carbon also added in manures, which stimulates the growth of the microbial biomass and therefore leads to immobilisation of the mineral N. The availability of any immobilised N for crop growth is not clear, though some evidence suggested that it was completely recovered by a spring barley crop. The organic N pool of the manure did not seem to be important in supplying N for crop growth in the first year.

The use of ^{15}N-labelled manures enabled the separation of the N taken up by plants into that derived from the soil and that derived from the manure. Manures were labelled non-uniformly by incubation with ^{15}N salts for a short period before application. Where the assumption could not be made that the manure was uniformly labelled, a simple model was developed based on isotope dilution theory, to calculate the percentage of plant N uptake from the manure. ^{15}N was also used to determine the source of the N extracted by a number of methods, used to assess potential N availability.

The structure of a simple model was described, which could be used to select a manurial strategy, which maximised crop yields and estimated the potential N losses for an organic farm. It was suggested that this could be constructed using an expert system approach, which was able to refer to databases and simple models to provide the final output. The full development of such a model is not yet possible, as the availability of the N applied in organic manures is only partly understood. However, our understanding of the complex processes controlling the release of N from manures has been increased as a result of the work described.

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1.0 INTRODUCTION

The extent and rate of supply of nitrogen is one of the principal factors limiting organic crop production. As well as supplying nitrogen, an organic system (like a conventional one) must provide available soil nitrogen at a time which matches crop demand. Physiological, as well as economic, considerations demand that crops are managed differently in terms of planting dates, irrigation, mulching etc. The timing of maximum nitrogen demand and the pattern of nitrogen uptake through the season will vary significantly between crops. Where nitrogen supply and crop demand can be closely matched, maximal yields will be obtained with minimum losses of nitrogen to the environment.

In organic farming, nitrogen is supplied to crops from a range of materials: predominantly animal manures, green manures and crop residues. The release of nitrogen, in mineral forms available for crop uptake, from these manures is effected by soil micro-organisms, whose activity is limited by a wide range of environmental factors: temperature, water availability, supply of other nutrients etc.

Although the main paths and mechanisms for nitrogen transformation are known in soils, most of the work on maximising crop uptake and minimising losses has been carried out in conventional systems. The dynamics of additions of large pools of nitrogen in mineral forms are therefore reasonably well understood. However, large additions of mineral nitrogen are rarely possible in organic systems and, even where they do occur, they are accompanied by large additions of nitrogen in organic forms. Quantitative data on the relative efficiency of different organic manures to crops and the timing of nitrogen release from different pools of nitrogen in manures are limited. Greater understanding of the interacting processes of the soil nitrogen cycle is required, with respect to nitrogen release from manures, before these processes can be manipulated to the advantage of the cropping system.

The use of manures labelled with ^{15}N (both uniformly and non-uniformly) in field trials and pot experiments, enables the separation of nitrogen taken up by the plant into that derived from a number of nitrogen pools. The use of ^{15}N

also enables the transformations of nitrogen between pools to be studied in terms of gross, rather than net, rates and effects.

Field trials enable hypotheses generated from pot experiments and earlier work to be tested on a large scale. However, the use of field trials increases the variability in the data obtained, which may mask important differences between treatments. The integration of results from a series of field trials, pot and laboratory experiments and literature-derived data is necessary to draw any conclusions.

1.1 Aim and objectives

The project aimed to increase understanding of the soil nitrogen cycle, especially with respect to the release of nitrogen from added organic nitrogen sources, therefore enabling nitrogen supply to be more closely matched to crop demand in organic cropping systems.

The objectives of the project were therefore to:
1) Describe empirically the timing of nitrogen release, and the efficiency of use of this nitrogen from a range of manures, including animal manures, green manures and ploughed-out leys.

2) Predict N availability from soil and manures, using appropriate availability indices, soil and manure properties and simple models.

3) Relate N release characteristics to the N uptake pattern of crops, likely to be used in organic crop production in Scotland.

4) Increase understanding of the interacting processes of the soil nitrogen cycle, with respect to nitrogen release from organic manures.

5) Produce a reliable data set to allow development of a semi-quantitative model enabling N supply to be matched to crop demand in organic farming systems, and for farmers using a high proportion of organic wastes as fertilisers, thus minimising losses of N to the environment.

A series of independent and integrated field, pot and laboratory experiments, using ^{15}N as appropriate, was carried out, backed up by literature research, so that these aims might be achieved.

1.2 Organic Farming

1.2.1 Definition

Organic farming is a term, which is difficult to define precisely (Vine and Bateman, 1981). The term biological farming is preferred in other European countries, and various other names have been used to describe these systems and variants of them: 'natural', 'ecological', 'alternative', 'sustainable' (Dixon and Holmes, 1987). Definitions of organic farming usually focus on the basic characteristics of the system: the avoidance of soluble chemical fertilisers; the prohibition of agro-chemical pesticides, hormones and antibiotics; the extensive use of manures and the adoption of varied crop rotations. However, the basis of organic farming lies in the awareness that plant and animal health are closely linked to each other and to the soil fertility by biological cycles, which are disrupted or broken by conventional farming methods (Sykes, 1959). The basis of organic farming is the development of the natural biological cycles involving "the lowly earthworm, the bacteria, the fungi and the other myriads of micro-organisms of a benevolent character whose habitat is the soil" (Sykes, 1946) as well as plants and animals, where the emphasis is not only on maximising yield, but also on feeding the soil to feed the crops of the future (Balfour, 1945). Goldstein (1976) puts it eloquently:
"It takes only a very few words to describe what organic means.. . Begin with the soil, get into the compost heap, natural cycles, the need to return garbage, sludge and wastes back to the land, the hazards of pesticides and artificial fertilizers to the environment, and the personal health benefits that result from eating organic food."

The International Federation of Organic Agriculture Movements (IFOAM), which operates world-wide and lays down minimum standards to which members operate, sets out the following aims in its literature (Lampkin, 1990):

* to produce food of a high nutritional quality in sufficient quantity;
* to work with natural systems rather than seeking to dominate them;
* to encourage and enhance biological cycles within the farming system, involving micro-organisms, soil flora and fauna, plants and animals;

* to maintain and increase the long-term fertility of soils;
* to use as far as possible renewable resources in locally organised agricultural systems;
* to work as much as possible within a closed system with regard to organic matter and nutrient elements;
* to give all livestock conditions of life that allow them to perform all aspects of their innate behaviour;
* to avoid all forms of pollution that may result from agricultural techniques;
* to maintain the genetic diversity of the agricultural system and its surroundings, including the protection of plant and wildlife habitats;
* to allow agricultural producers an adequate return and satisfaction from their work, including a safe working environment;
* to consider the wider social and ecological impact of the farming system.

These are minimum standards and allow a great breadth of farming practice to be described as organic. For marketing purposes within the UK, separate bodies have drawn up standards defining the materials and techniques which are acceptable in the growth of organic crops and the rearing of livestock, (UKROFS, Soil Association (1990)) to allow organic produce to be clearly labelled.

It is clear from all the above definitions that organic farming does not simply mean a substitution of organic inputs for agro-chemical ones. Misuses of organic materials, either by excess, or inappropriate timing, or both, will also short circuit the development and working of natural biological cycles, just as would agro-chemicals (Lampkin, 1990). Much of the basis and practices of organic agriculture are very similar to the best of pre-1940's agriculture in the UK. Organic farming is not, however, a return to the systems of the past. It is forward looking; it is concerned with how agriculture can be sustained on a secure footing with both the methods which have proved reliable in the past and a host of others, which have yet to be developed (Body, 1987).

1.2.2 History of the organic movement in the UK

The history of the sustainable agriculture movement around the world was reviewed by Harwood (1990). Organic agriculture was established in the UK as an alternative to mainstream agriculture during the post-war agricultural

reconstruction in the late 1940's. Liebig's ground breaking work in 1840 ('Chemistry in its application to agriculture and physiology') had brought chemistry to agriculture. As agriculture was rebuilt following a long period of depression, caused by a range of economic, social and agricultural factors (Hall, 1941), production intensified and agriculturalists looked to chemists, increasing their use of simple soluble fertilisers, crop protection chemicals and animal drugs.

A number of voices called for a restriction in the use of chemical fertilisers and a return to a sensible maintenance of soil fertility using animal and plant manures and composts. Such methods had been developed in India (Howard, 1940) and were already in use in England (Balfour, 1945; Sykes 1946, 1959). Coupling techniques such as subsoiling and the use of medium-term deep rooting leys, they were able to match yields from contemporary conventional agriculture, even in the absence of farmyard manure applications (Sykes, 1946). They warned from long experience in farming that a depletion in soil fertility would result from the dependence of the agricultural industry on chemical fertilisers, especially nitrogen, which while stimulating the crop also inflicted noticeable damage to the soil through the destruction of worms and other micro-organisms (Sykes, 1946). The loss of organic matter would also lead to increased erosion losses. As yields in conventional systems continued to increase rapidly, there were those who continued to point out that production increases were not equivalent to increases in fertility, and that increased production for human use is usually secured by reducing natural soil fertility (Balfour, 1945).

In 1946 the Soil Association was inaugurated, inviting membership from all who were working towards "a positive concept of health based, through wholesome nutrition, on biologically fertile soil" (Jenks, 1959). Organic producers continued to operate very much in the minority and largely unsupported by technical advice or research (Goldstein, 1976), while conventional farming increased its dependence on chemical fertilisers and other agro-chemicals. Organic produce was consumed on the farm of origin or marketed through normal channels with little or no differentiation from conventional products (Daw *et al.*, 1991).

1.2.3 Current position of the organic farming sector in the UK

Once dismissed as cranks, organic farmers are gaining credibility. Organic farms occur in all areas of the UK: ranging from all grassland to all arable systems; from almost self-sustaining units to those importing large quantities of nutrients from outside the farm gate; from farms which are completely organic to those which have just a few acres of organic cereals or fruit (Vine and Bateman, 1981). The number of organic farmers in the UK had risen from less than 100 in 1980 to over 700 in 1989 (Lampkin, 1990). During the period from 1986-1991 the area of farmland in Scotland under organic systems or in conversion is estimated to have doubled. Organic cereal production now occupies approximately 1,100 ha and organic vegetable production 350 ha (Daw *et al.*, 1991).

The increased production of organic agriculture has largely been consumer led. The consumer's view of the quality and freshness of the food on offer, including attitudes to herbicides, pesticides, hormones and drugs of all kinds (Spedding, 1983; Halley and Soffe, 1988), has encouraged some significant recent interest in and willingness to pay for organic food (Daw *et al.*, 1991). However, this increase in demand was somewhat slowed by the recession of the early 1990's, though it is predicted that it will turn upwards once more following economic recovery.

Heightened awareness of food surpluses within the EEC and an increased environmental focus, in addition to the consumer demand for organic food led by health concerns, brought positive media attention to organic agriculture. During the 1980's a number of critical reviews of conventional farming practice (Hodges, 1981; Body, 1987) appeared and there was a realisation of the problems, as well as benefits, that conventional agriculture had brought. There was an re-awakening of interest in the organic farming sector with studies of organic farming being carried out in a number of countries: France in 1975; Holland in 1977; USA in 1980; England and Wales in 1981 (Vine and Bateman, 1981). The US Department of Agriculture report concluded that organic farming is energy efficient, environmentally sound, productive and stable and that it tends towards long-term sustainability (Reganold *et al.*, 1990). During the late 1970's and 1980's, academic interest around the world was also awakened

and in the UK, the Elm Farm Research Centre was set up, specifically to carry out research for organic farmers.

During the 1980's, a significant number of conventional producers made the conversion to organic production on all or a part of their land in response to the strong consumer demand (Dixon and Holmes, 1987). While in 1981, the majority of organic farmers in England and Wales indicated that they had chosen to farm an organic-type system to maintain the land 'in good heart' (Vine and Bateman, 1981), economic considerations had become more significant by 1990, as prices for organic produce were at record levels (Daw *et al.*, 1991). Although small in terms of acreage and production (less than 0.5% of UK output is organic), organic production looks set to expand further under the influence of high price premiums, the encouragement of conversion to organics through extensification and set-aside schemes, as well as heavy media coverage.

A study of over 200 organic farms in Northern Europe and the US by Agrow (Lampkin, 1990) found that on average there was a yield reduction of only 14%, for crops grown without chemical pesticides and fertilisers, a much smaller differential than had been expected. Comparison of old organic and conventional farms neighbouring one another in the US (Reganold, 1989), showed that the soil on the organic farm contained significantly more organic matter, nitrogen and biologically available potassium. The soil had a greater cation exchange capacity, a larger and more active biomass and a greater polysaccharide content. The soil under organic production had better structure and tilth and significant losses of topsoil by erosion had been prevented. The economic comparison between conventional and organic agriculture is difficult (Vine and Bateman, 1981). Conversion periods are costly, with 4-5 years of poor financial performance usually expected before original profit levels can be attained (Daw *et al.*, 1991). Premium prices are necessary for organic agriculture to match the economic performance of conventional systems and these are unreliable. Dixon and Holmes (1987) summed up their economic assessments saying:

"There is no doubt that a living can be made from organic farming, but those considering conversion should realise the changeable nature of the market and the possible difficulties with aspects of the husbandry. Organic farming is no 'easy option'."

1.2.4 Organic farming systems and practices - an overview.

Detailed reviews of organic farming systems and practices in the UK were presented by Vine and Bateman (1981) and Lampkin (1990). Reviews of sustainable agricultural systems around the world, including the tropics, are contained in the proceedings of the International Conference on Sustainable Agricultural Systems (Edwards *et al.*, 1990).

The output from an organic farm is controlled by the management of the whole system, rather than the simple management of individual enterprises. Efficient cycling of nutrients between farm enterprises, in particular with rotations and appropriate manure management and cultivations, minimises the need for external inputs and they are used only to supplement this efficient management of internal features (Lampkin, 1990). Organic cropping systems fall broadly into two categories: alternate ley-arable husbandry and predominantly arable systems (Vine and Bateman, 1981). Both systems rely heavily on rotations, including short- or medium-term leys, to control weeds and maintain soil structure and nutrient levels. Where substantial quantities of nutrients are sold off the farm, especially in the predominantly arable and horticultural systems, nutrients may be imported from outside the farm, in organically acceptable forms (Vine and Bateman, 1981).

Nutrients may be supplied to crops from a number of sources. Farm wastes are widely recycled to land as nutrient sources: farmyard manure, slurry and farm-made composts. Cereal straws may also be chopped behind the combine harvester and incorporated to the soil directly after harvest. Fodder crops may be grazed *in situ*, eg. stubble turnips, and this leads to a nutrient return to the field via dung and urine. In alternate ley-arable systems and in predominantly arable systems to a lesser extent, the ploughed out leys form a major supplier of nutrients to the crop rotation (Vine and Bateman, 1981). Crops may be grown specifically to be incorporated to the soil, as in the case of green manures. In Scotland, however, these are often grazed before turning in (Dixon and Holmes, 1987) as farmers are reluctant to 'waste' such growth.

External sources of nutrients have to conform to organic standards and may include: wastes from other farms or processors, manures, slurries, spent mushroom compost, sugar beet lime etc.; abattoir wastes, hoof and horn;

fishmeal manures; seaweeds and refined/manufactured organic fertilisers. Such nutrient sources can be much more costly than conventional fertilisers, and their use tends to be restricted as much as possible to high value crops (Lampkin, 1990). Indirect nutrient sources such as animal feed may also be imported.

Organic farmers aim to maintain the natural structure of the soil, as far as possible. Ploughing is usually shallow (10 cm) and the chisel rather than mouldboard plough is preferred where possible (Vine and Bateman, 1981). Deeper ploughing may be needed in some cases to break up leys before the cultivation of arable crops. Subsoiling to improve soil aeration and break up cultivation pans is also carried regularly in most organic systems (Sykes, 1959).

Weed and pest control is achieved mostly by the choice of suitable rotations, using break crops, to prevent the build up of weed populations. Varietal mixtures have also been used in some cases to minimise any disease impact on output (Lampkin, 1990), and the majority of organic farmers select varieties carefully for disease resistance. The biggest disease problem cited by organic farmers was potato blight (Vine and Bateman, 1981), with mechanical destruction of haulms the most common remedy. Seedling competitiveness against weeds is maximised by careful seed-bed preparation, and for horticultural crops, stale seed-bed and mulching techniques may be used. Organic farming systems often have more frequent and intensive cultivations compared to conventional systems, due to the need for physical, rather than chemical, removal of weeds. Cereals may be vigorously harrowed and rolled in early spring and in precision drilled crops mechanical hoeing between rows is common. Fodders and root crops are cultivated in ridges to facilitate weed removal with minimum disturbance to the crop and in some cases, hand roguing is also carried out (Lampkin, 1990).

There are a broad range of crops grown organically in Scotland (Dixon and Holmes, 1987). Forage and conserved grassland and oats were the most important crops in terms of area of production, with wheat, swedes, barley, ware potatoes, brassicae and carrots also occupying significant areas. Other organic crops included rye, seed potatoes, leeks, onions, lettuce, courgettes, strawberries and raspberries. Organic oats formed approximately 1% of the total oat production in Scotland (Dixon and Holmes, 1987). Most organic farmers in

Scotland operate small-scale mixed farming systems, including livestock, though there are a number of intensive horticultural holdings.

1.3 Crop Nitrogen Uptake

1.3.1 N - an essential nutrient

Nitrogen had been established as a constituent of all plant proteins as early as 1785 by Berthelot (Glass, 1989). The bulk of N in plants occurs in proteins, nucleic acids and amino acids. N is also an important building block in the structure of nucleic acids, chlorophyll, adenine, a constituent of both ATP and NAD^+, and a number of the plant growth regulators. With increases in nitrogen supply, there are increases in both soluble amino compounds and proteins; the additional protein allows the leaves to grow larger and hence increases the photosynthetic area. Nitrogen deficiency results in plants with yellow or pale green leaves with small cells and thick walls, and the leaves may be harsh and fibrous (Wild and Jones, 1988). Nitrogen is usually taken up by plants as the simple inorganic ions: nitrate (NO_3^-) and ammonium (NH_4^+). However, some plants are able to use dinitrogen (N_2) through symbiotic associations with nitrogen fixing bacteria. This is considered further in Section 1.3.5. There is also some evidence that plants are also able to use simple organic N compounds taken up through the roots (Alexander, 1983).

Individual species differ qualitatively and quantitatively with respect to their demands for inorganic nutrients (Wild and Jones, 1988). Early fertiliser trials demonstrated clear-cut examples of differential responses to appliations of N, P and K, with brassicae giving a much stronger N response than potatoes or cereals (Glass, 1989). Yield response to applied nutrients is often shown as a parabolic curve, which may be described by the Mitscherlich equation, or as a graph composed of two straight lines, which are used to establish the optimum application rate of N (Cooke, 1982; Neeteson and Wadman, 1987). However, such response curves show large variability between sites and alternative approaches to the analysis of such data have been suggested (van Keulen and Stol, 1991). The different yield responses to nitrogen of different crops may arise from differences in rates of absorption, patterns of allocation within the plant and the efficiency by which absorbed N is used in metabolic processes.

1.3.2 Nitrogen uptake

Rapidly transpiring plants draw a substantial bulk of soil solution to the root. However, where this mass flow of nutrients is insufficient to supply the demand, then diffusion will supplement the supply. Soil supply processes will be discussed further in section 1.4.

Active transport, a process in which energy is expended in moving ions from a zone of lower to higher electrochemical potential, is necessary for ion uptake into the root cell (Russell, 1977), since the composition of plant cells is typically very different both quantitatively and qualitatively from the surrounding soil solution. Active transport is under the control of the intrinsic proteins in the plasma membrane (Glass, 1989). Cations and anions show different initial uptake rates by plants, associated with the equilibriation of the ions in the apoplasmic free space in the root (contained largely within cell walls). Cations are subject to ion exchange reactions in this space, similar to those seen with clay minerals (Russell, 1977). However, after this equilibriation has occurred then net uptake rates of ions are similar and are controlled by the active-transport systems. Absorption of ammonium and nitrate is via systems with the same characteristics as those which operate for other ions. In the case of ammonium and potassium, however, the kinetics of absorption are so similar that the carrier mechanisms are probably identical (Epstein, 1972).

1.3.3 Roots

Rooting systems of different species have very different characteristics with the biggest differences occuring between monocotyledons (dominantly cereals and grasses) and dicotyledons (Figure 1). The general habit of cereals is to establish a well developed superficial rooting system penetrating the whole of the first 25-40 cm of soil and extending laterally for up to half a metre (Hector, 1937a). A number of the seminal roots also penetrate deeply, on average to 1.5 - 2 m. The differentiation into a shallow dense upper region and a deeper penetrating lower region is usually very marked. There are differences between the cereal crops: wheat, barley and oats; linked partly to their sowing date, so that winter cereals have deeper rooting systems; but genotypic variations also occur. Differences in the root morphology of the dicotyledons are much more marked.

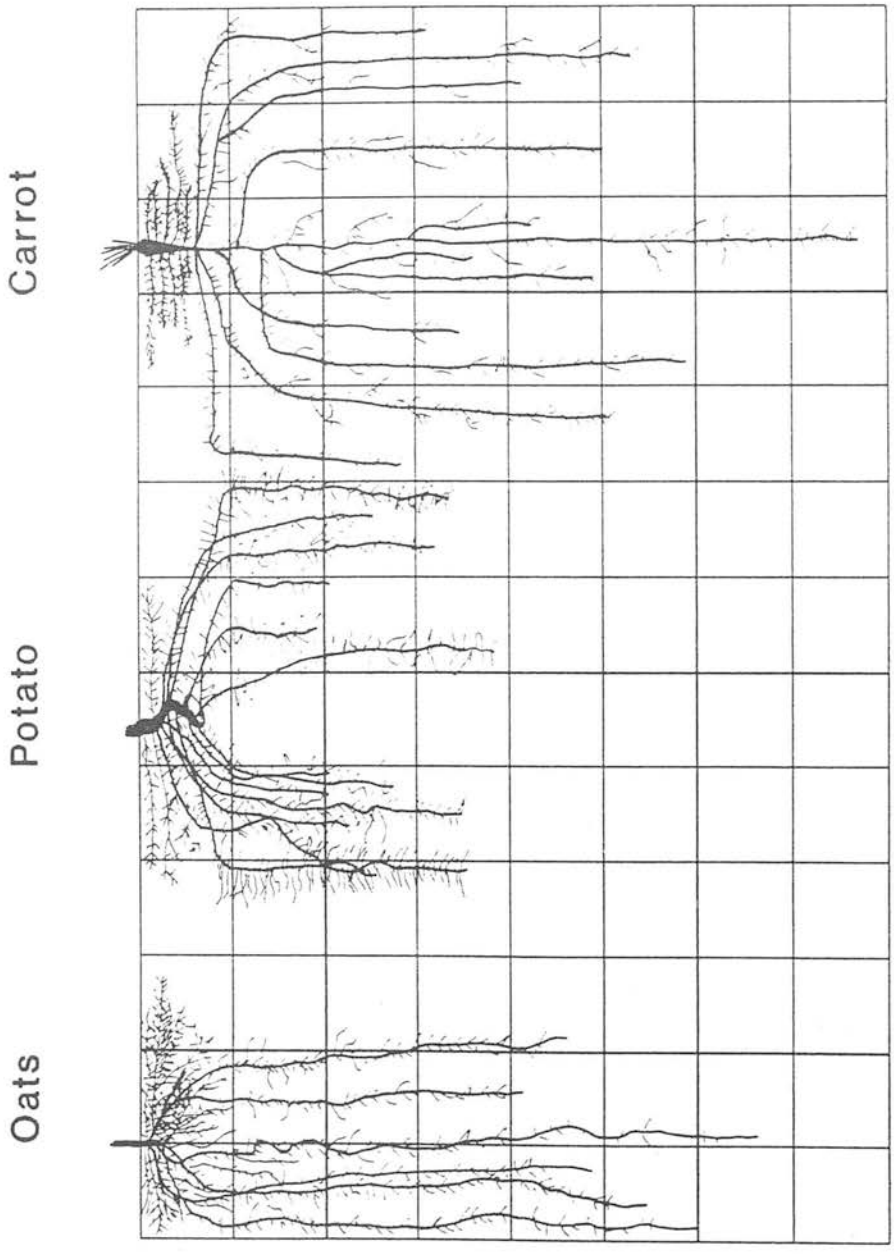


Figure 1. Root systems of crops: spring oats, potato, carrot. Adapted from Hector (1937a and 1937b). Each square represents an area 30 x 30 cm.

Root growth begins from a primary or tap root, all subsequent roots are adventitious. The tap root may penetrate deeply (carrot and winter cabbage) or remain very short (potato). Rooting is also largely concentrated in the upper horizons of the soil with a network of highly branched fibrous adventitious roots developing (Hector, 1937b).

Root length and rooting density are very important in controlling the surface area over which nutrient absorption can occur. However, the efficiency of different parts of the root system of cereals has been shown to be more closely related to root volume than surface area or length (Russell, 1977). Root age may be another important factor (Barber, 1984). The morphological plasticity of the rooting system is very important, so that the rooting density will increase very markedly in regions of higher nutrient concentration (Cooke, 1954; Drew and Saker, 1975). For a single plant of winter rye (Dittmer, 1937) root length was shown to be in excess of 500 km and the surface area greater than 200 m^2. The root length of vegetable crops at harvest ranges from 1.8 km m^{-2} in onion to 15.0 km m^{-2} in turnip (Greenwood *et al.*, 1989). However, the volume of the rooting system of any plant is seldom more than 5% of the surrounding soil (Russell, 1977).

The root-to-shoot biomass ratio is not a constant throughout the life of the plant. During rapid vegetative growth, the weight of roots and shoots commonly increases in parallel. However, there may be a marked divergence in the reproductive stage when nutrient uptake reduces and the death of old roots may not be fully compensated for by the growth of new roots (Russell, 1977). A plasticity of the root-to-shoot biomass ratio is also seen in response to stress. Under conditions of low light but adequate nutrients the ratio tends to increase. However, under conditions of nutrient deficiency the root-to-shoot biomass ratio tends to increase. In barley the ratio is usually 0.25, but values of up to 1.0 have been recorded (Glass, 1989).

1.3.4 Nitrogen fixation

Atmospheric N_2 is introduced into the soil-plant system by the action of micro-organisms living either under symbiotic conditions (eg. Rhizobia) or freely in the soil (eg. *Azotobacter*) (Postgate, 1982). Nitrogen-fixing micro-organisms are found in most habitats, although for agricultural purposes, the most important

ones in terms of N fixed per annum are those in nodules on plant roots (Nap and Bisseling, 1990). Bacteria of the genus *Rhizobium* invade the root cells of different leguminous plants, resulting in nodule formation and eventually nitrogen fixation. This symbiosis exhibits specificity, particular legumes only being infected by a limited range of rhizobial strains or species (Postgate, 1982). The mutual advantages of the symbiosis are that the plants receive a supply of nitrogen and the micro-organisms are in a protected environment inside the nodule cells and are supplied with photosynthates to satisfy their demands for carbon and energy. Leguminous plants in symbiosis with *Rhizobium* can fix N at rates in the range 52 - 300 kg ha^{-1} $year^{-1}$, with annual crops fixing much less than perennials (Phillips, 1980). The association between legumes and their corresponding *Rhizobium* species has been the focus of intensive investigation (eg. Yates, 1980; Robertson and Farden, 1980; Pueppke, 1986).

Nodule establishment and nitrogen fixation may be affected by a wide range of factors. Legumes are very intolerant of water stress and some temperate legumes may also be adversely affected by waterlogging (Postgate, 1982). Legumes also seem to have greater requirements for phosphorus, potassium and molybdenum than non-legumes (Sprent, 1980) and may be more sensitve to low pH. The presence of large amounts of mineral nitrogen in the soil significantly inhibits rhizobial infection, nodule development and nitrogen fixation (Postgate, 1982). Where significant amounts of mineral nitrogen are present then the legume crop behaves in a very similar way to non-legumes and little N is fixed.

1.3.5 Yield and N uptake

In annual crops, early in the growth period the need for N is low, this increases during the period of most rapid growth and then tapers off at maturity. This leads to the typical sigmoidal curve seen when N uptake through the season is plotted (Broadbent, 1984), with the 3 parts of the curve roughly corresponding to: establishment and seedling growth; rapid growth, stem elongation and flowering; maturation and senesence (Harper, 1983). Substantial differences are seen between the relative growth rate of crops during the early season (Gregory, 1988) and curves occupy different times and scales related to differences in crop sowing date and adaptability to cool spring and early summer temperatures (Harper, 1983) (Figure 2).

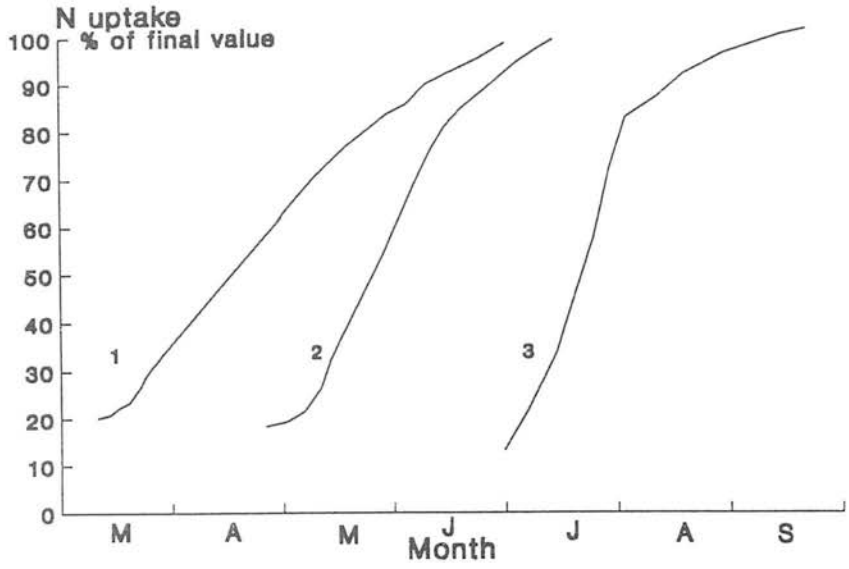


Figure 2. The N uptake pattern of several crops shown schematically with N uptake shown as a proportion of total uptake. 1, Winter barley; 2, Spring Barley; 3, Swede.

The attainment of maximal growth rates by a plant is highly dependent on the expansion of leaf area to intercept maximal radiation to drive photosynthesis. The duration of the period of leaf expansion is controlled by temperature and photoperiod, but the number of leaves, their expansion rate and final size is controlled by the temperature, plant water stress and nutrient supply. The maximum rate of uptake of nutrients usually precedes the period of maximum growth rate (Wild and Jones, 1988). Nitrogen is one of the most important factors controlling leaf expansion (van Keulen and Stol, 1991), since N supply controls protein production and N is also a component of chlorophyll. In the potato crop, leaf area duration is also an important factor in controlling yield, and increased N applications were shown to increase the leaf area duration (Dyson and Watson, 1971), not only increasing the number of leaves produced but also delaying senescence. Crop yield and nutrient uptake are then largely controlled by photosynthetic rates, and where nutrient and water supplies are not limiting, yields are closely related to radiation intercepted (Gregory, 1988).

The nitrogen demand of the storage organs most often harvested by agriculture (seeds, roots, tubers) is partly met by translocation from vegetative organs and partly by continued nitrogen uptake. Contributions from both sources usually make up about half of the grain nitrogen of a wheat crop (Wild and Jones, 1988). Photosynthesis during maturation in potatoes was reduced by translocation of N from the leaves to the tubers, where N supply was restricted (Dyson and Watson, 1971), which caused early senesence in the leaves. Continued uptake of N is not always seen in field experiments and has been discounted in some models (van Keulen and Seligman, 1987).

1.3.6 Climatic effects

Temperature and photoperiod are the main controls on the development of a crop and crops may be excluded from certain climatic regimes because the full development cycle would not be able to be completed. Growth determining factors, such as light, CO_2, water and nutrients, are much more indirectly related to climate. At temperatures less than about 5 °C, no development and little growth is seen in the common agricultural crop species. However, this base temperature varies greatly between crops and may also vary in the same crop according to sowing date (Gregory, 1988). Changing photoperiod seems to act as an important trigger to changes in the development cycle of crops eg. the onset of flowering. Photoperiod and temperature may also act together eg. in controlling leaf appearance in barley and wheat (Gregory, 1988). It is not clear whether N supply has any effect on the development of crops (van Keulen and Stol, 1991) with conflicting evidence presented, which may in part be a result of interaction with other factors.

Scotland can be regarded as having an adverse climate for crop production (Speirs, 1990). The 5 °C base temperature is reached in mid-late March in the southern half of Britain, but it is not attained in Edinburgh until April 5th and Aberdeen until April 15th on average. Summer temperatures are also lower on average and the growing season is restricted to 225 days at sea level in Scotland and N.E. England, compared to 270 days in southern England. The late start and lower summer temperatures lead to later harvests in Scotland, which may not allow the establishment of cover crops after harvest.

Scotland is also significantly wetter than much of southern England. Rainfall in Fife is on average 30 % greater than that measured at Bedford. However, when evapotranspiration is considered, excess winter rain may be more than double that in southern England (Speirs, 1990). Soil moisture deficits are not so great in Scotland throughout the summer and this may reduce the need for irrigation for some crops eg. potatoes. Grass growth is favoured due to the lower summer soil moisture deficit and longer daylight hours in the late season. However, cereals, which have a high nitrogen demand early in spring when temperatures are still low, may be at a disadvantage unless fertiliser nitrogen is judiciously applied.

The effects of climate are much better understood than the effects of weather during the growing season. The effects of weather are confounded by indirect effects, eg. pest and diseases, management practices, and the effect of weather may itself be indirect eg. changing soil moisture status.

1.4 Soil N supply

1.4.1 Movement of NO_3^- and NH_4^+ through soils.

As water moves through the soil in response to potential gradients (Hillel, 1982), NO_3^- and NH_4^+ will also be moved through the soil, since they are found in the soil solution. This movement of solutes is known as mass flow. Mass flow may be caused by plant water absorption, excess winter rain or irrigation moving down through the profile or lateral flow of water due to topography. The rate at which NO_3^- and NH_4^+ are moved by mass flow can be obtained by multiplying the water flux and the concentration of the ions in the soil solution. However, estimates of water flux and ion concentration are spatial averages over a wide ensemble of soil pores (Nielsen *et al.*, 1982) and as a result only very coarse estimates of the movement of NO_3^- and NH_4^+ can be made by this method. The net charge density of the surfaces of soil particles influences the distribution of solutes within each pore: anions tend to be excluded from a small volume close to the soil particle surfaces, while cations are attracted to the surfaces and may not move through the soil at the same velocity as the soil water (Nielsen *et al.*, 1982).

In addition to movement by mass flow, NO_3^- and NH_4^+ may also diffuse through the soil, where concentration gradients occur (Wild, 1981), eg. in zones around the roots. Diffusive movement of nutrients to the roots is usually

restricted to a zone 0.1 to 15 mm from the root surface (Barber, 1984). Cation exchange on soil surfaces leads to a much smaller effective diffusion coefficient for NH_4^+ in soils (1.9×10^{-8} cm^2 s^{-1}) than for nitrate (2.5×10^{-6} cm^2 s^{-1}) (Barber, 1984). The tortuosity of soil pores also restricts diffusion in soils and this increases as the soil water content decreases (Wild, 1981).

Ammonium may also occur in soils as 'fixed' ammonium within the lattices of clay minerals, especially hydrous mica and vermiculite (Wild, 1988). This ammonium is in a slow equilibrium with ammonium held on clay mineral surfaces by cation exchange (exchangeable ammonium) and that in solution, but is not directly accessible to nitrifying bacteria or plant roots. Ammonium fixation is reviewed in detail by Nommik and Vahtras (1982).

The supply of NO_3^- and NH_4^+ from soils to plants is heavily dependent on the concentrations of these ions occurring in the soil solution. In the absence of large additions of NO_3^- and NH_4^+ as soluble fertilisers, these concentrations are the result of a large number of reactions, which both produce and consume NO_3^- and NH_4^+, and are discussed in the remainder of this section.

1.4.2 The soil N cycle part of a global cycle.

Nitrogen occurs in the four recognized spheres of the earth: the lithosphere, atmosphere, hydrosphere and biosphere. The bulk of the N is found in the lithosphere (98%) and most of the remainder occurs in the atmosphere, where dinitrogen (N_2) comprises about 78% of the gases. However, unlike in the other spheres the N in the biosphere is in a constant state of flux, which is commonly described as the N cycle (Figure 3). This links the nitrogen in all four spheres. It should be noted that a 'N cycle' as such does not occur in nature, but that any given N atom moves from one form to the other in a completely irregular fashion (Stevenson, 1982a). Plant available forms of nitrogen usually only occur at a very low concentration in soil (Woodmansee *et al.*, 1981). However, the total amount of N in temperate arable soils is considerable, often exceeding 4000 kg N ha^{-1} to plough depth (Stevenson, 1982a). The amounts of NO_3^- and NH_4^+ in the profile are controlled by a number of complex, simultaneous and often opposing processes occurring in soils, dominantly mediated by micro-organisms, and known as the soil nitrogen cycle (Figure 4).

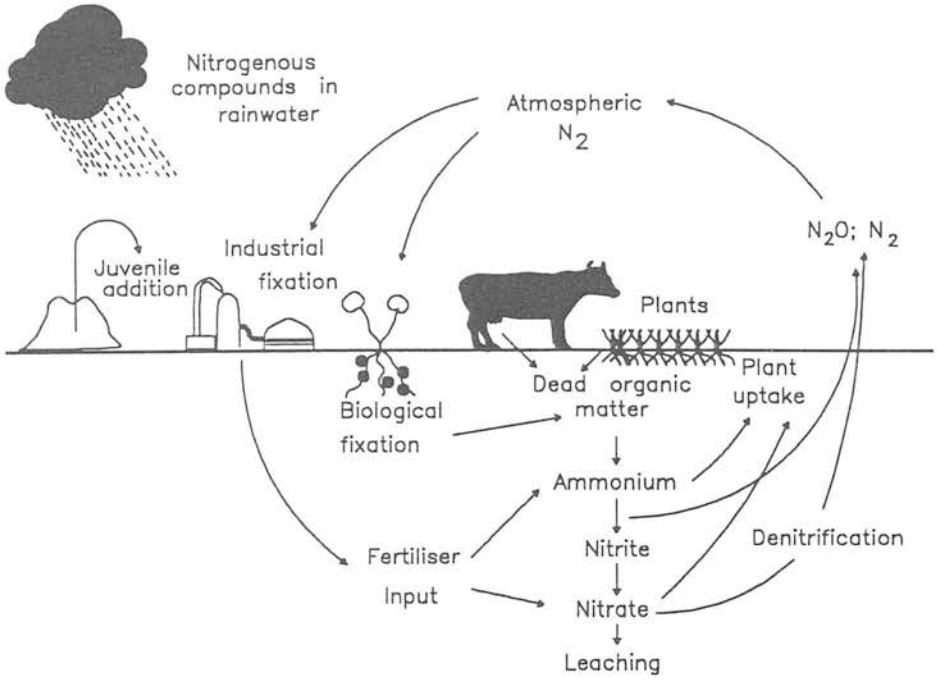


Figure 3. The global N cycle, based on Haynes (1986)

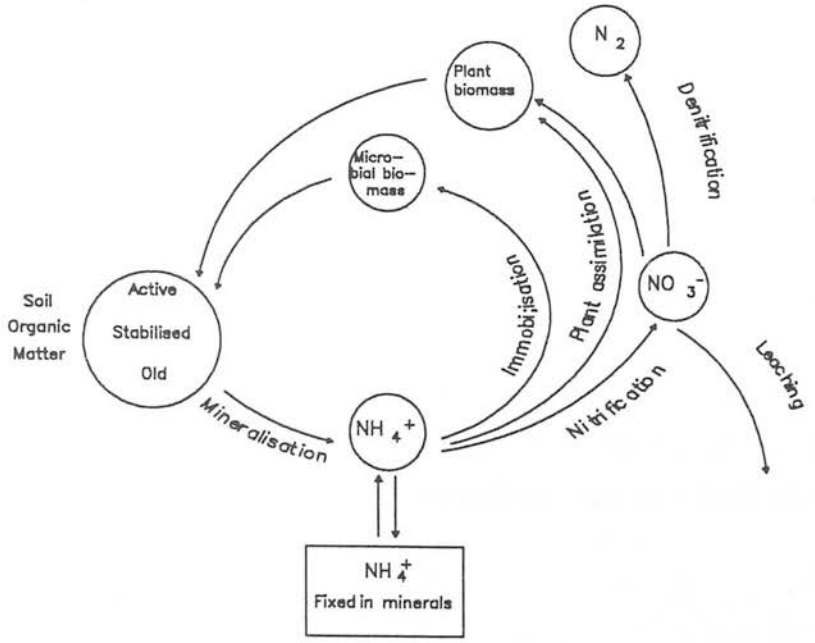


Figure 4. The soil N cycle, adapted from Jansson and Persson (1982) and Paul and Juma (1981).

The net magnitude, general microbiology and abiotic controls (moisture, temperature, aeration, residue composition etc.) of many of the processes regulating N availability in soils are reasonably well understood (Paul, 1984), and will be discussed in the following sections.

1.4.3 Soil organic matter - a nitrogen reservoir.

In undisturbed soils, organic matter attains a steady state level governed by the soil forming factors and their interaction: climate, topography, parent material, vegetation and organisms and time (Jenny, 1941). Where soils are disturbed by man and brought into agricultural production then this equilibrium is disturbed and organic matter tends to decline to a new equilibrium, dependent on the cropping and soil management. Clay type and aggregation affect soil organic matter dynamics. Allophane reduces degradation of added carbon sources (Paul, 1984) and clay minerals may protect bacteria and microbial metabolites (Hassink, 1992).

The organic matter has long been split conceptually into active and stable fractions (Jansson, 1958), but recently the active soil organic matter has been further divided into biomass and active non-biomass pools, whose sizes can be estimated (Paul and Juma, 1981). The size and turnover rate of the active fraction of soil organic matter is related to agricultural practices, soil and vegetation type (Paul, 1984). Recently added organic residues are about 7 times more decomposable than native organic matter (Shen *et al.*, 1989). However, mineralisation is dominated by native organic matter due to its very large pool size, despite the fact that it is inherently less decomposable than recently added residues. The passive phase is heterogeneous with respect to origin, composition and age (Jansson and Persson, 1982) and the elucidation of the chemical structure of this fraction has kept humus chemists busy (Stevenson, 1982b). However, no one fractionation technique has been found, which adequately separates soil organic matter into biologically meaningful fractions (Paul, 1984).

1.4.4 Soil biomass - source, sink and power-house.

The soil biomass is both the agent of change in the soil, mediating many of the processes of the soil nitrogen cycle, and a repository of considerable quantities of N and other nutrients in a form more labile than in the majority of organic matter (Shen *et al.*, 1989). It is incomplete to view the biomass simply as a 'driving variable' of the process of decomposition (Swift *et al.*, 1979).

The soil biomass is composed of almost every class/order of invertebrate, hundreds of fungal species and a wide range of bacterial types. Plant detritus is broken down by the combined action of all the soil biomass. These organisms feed on the detritus and utilise the energy, C and other nutrients for their own growth (Swift *et al.*, 1979). Eventually decomposers die or are consumed and their carcases are further decomposed. Divisions of the soil biomass are often made with respect to size or functionality of the organisms. There is a high degree of correlation between these two approaches as the largest animals are dominantly saprotrophic and the meso- and micro- fauna are microtrophic (Swift *et al.*, 1979). However, neither approach is complete, as many organisms demonstrate functional flexibility.

Soil invertebrates contribute both directly and indirectly to N fluxes in soils (Anderson, 1988). They mobilise N through trophic transfers in food webs and turnover of tissue production (Figure 5). Indirectly they may alter physiochemical environments and may provide feedbacks controlling populations at lower trophic levels. The role of microbivorous fauna and the whole soil biomass food web in nutrient cycling is currently receiving considerable attention (Hunt *et al.*, 1984; De Ruiter *et al.*, 1993). The influence of protozoa may be considerable. In simulations (Robinson *et al.*, 1989) mineralisation occurs over a much wider range of protozoa/bacteria combinations than if bacteria only are considered. Predator/prey relationships between organisms may be important in controlling N availability since the organisms involved have different C:N ratios: bacteria 3:1; fungi 10:1 and protozoa 7:1 (Robinson *et al.*, 1989). The net outcome of predation is difficult to predict. However, it has been confirmed that predators increase the productivity of their prey with respect to nutrient supply, by recycling limiting nutrients (Hunt *et al.*, 1984). Soil macrobes (eg. earthworms) either stimulate microbial decomposition or are correlated with it. In the presence of earthworms, CO_2

evolution is increased and the size of the microbial biomass is decreased, while mineralisation of nitrogen is increased (Ruz Jerez *et al.*, 1988).

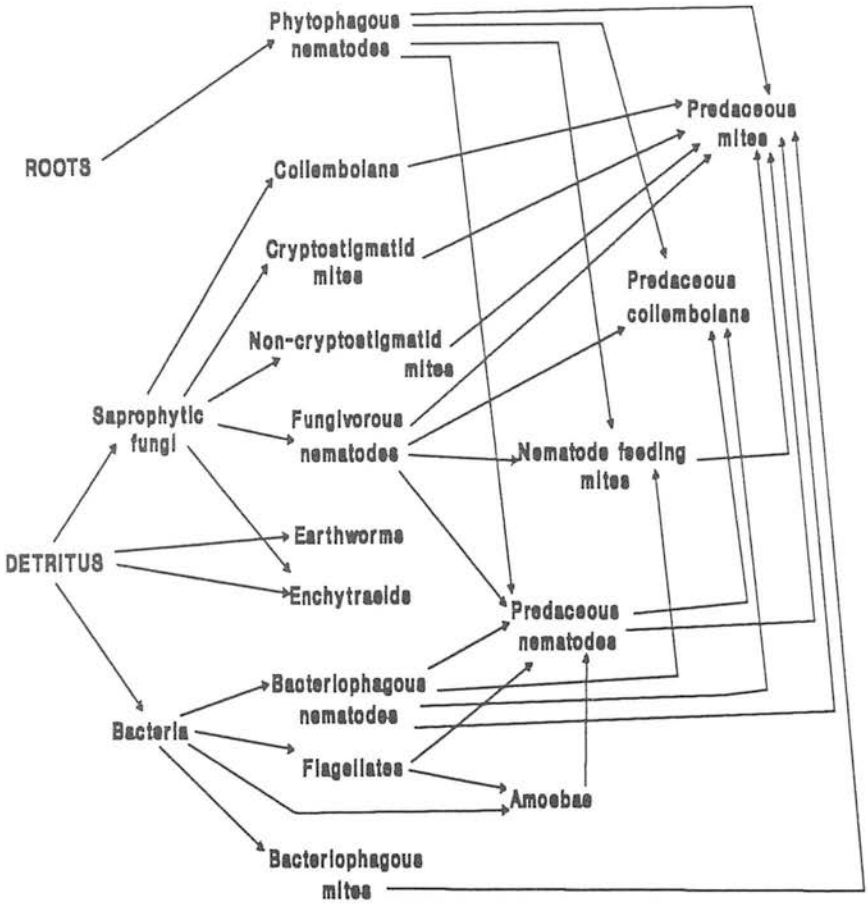


Figure 5 A typical agricultural soil biomass food web (De Ruiter *et al.*, 1993).

The size of the biomass compartment is related to soil type and soil management (Chaussod *et al.*, 1988). Tillage, pesticides and fertilisers used to maintain production in conventional cropping systems not only override faunal processes but also further reduce and obscure their contribution to soil fertility (Anderson, 1988). Seasonal changes in the size of the microbial biomass seem to be related to rhizosphere development and the production of root exudates, with modifications due to soil temperature and moisture (Carter and Rennie, 1984). Bacterial and fungal feeding nematodes will also respond rapidly to such growth (Anderson, 1988) and their populations might be expected to track the

microbial population. Widely varying temperature and moisture conditions at the soil surface under field conditions induce a variable half-life for the microbial biomass and may cause them to switch between being a sink and source of labile C and N during the season (Carter and Rennie, 1984).

The existence of two biomass compartments was suggested by Carter and Rennie (1984) that part which is stable, occurring in micro-sites throughout the soil and that part which is increased due to plant root exudation. This more active part is more sensitive to levels of available energy, it probably has a shorter half life and may act as the primary sink for added N. This population may be a random fraction of the original biomass, which just happens to be in micro-sites intercepted by plant roots, or else may be a, as yet undefined, niche-adapted population. Two compartments for the biomass have also been suggested by Nicolardot (1988) and van Veen *et al.* (1984). In this case the stable microbial biomass is suggested to be that part of the population, which is protected by colloidal soil particles, especially clay.

Microbial biomass forms a key compartment in models of the soil N cycle, where the rates of transformation of N are closely linked to the C availability (Paul and Juma, 1981; van Veen *et al.*, 1984; Chaussod *et al.*, 1988).

1.4.5 The processes of the soil N cycle.

Energy flows through the N cycle, beginning with the absorption of solar energy by plant and some micro-organisms in photosynthesis (Jansson and Persson, 1982), and there are strong links between this flow of energy and the cycling of N. Since most processes of the soil N cycle are microbially mediated, the soil N cycle is heavily dependent on the C flow through the soil and the interaction between the two cycles often controls the availability of N for plants.

1.4.5.1 Mineralisation - immobilisation

Micro-organisms in the soil mediate the decomposition of organic matter to supply their demands for C , N and other nutrients for tissue building. Where there is insufficient organic N available, they will assimilate and transform inorganic N from the soil. This process is known as immobilisation. Immobilisation has been shown to occur predominantly from the pool of NH_4^+

rather than from the NO_3^- pool (Recous *et al.*, 1988). Where N is supplied to micro-organisms in excess of their immediate metabolic requirements, this will be excreted. The release of N, mediated by micro-organisms, from soil organic matter as inorganic ions is known as mineralisation. Both these processes are continually occurring in soils and as a consequence the availability of NH_4^+ for plant uptake and nitrification is commonly described as a net effect: a net rate of mineralisation or immobilisation. However, the simple monitoring of pool sizes with time and net rates and effects is not sufficient to describe the soil processes. A low net rate of mineralisation may reflect either a low gross rate of mineralisation or a high rate of gross mineralisation balanced by a high rate of gross immobilisation, the dynamics of the soil biomass will be very different in the two cases (Jansson and Persson, 1982).

Net mineralisation rates increase with temperature (Stanford *et al.*, 1973) and variability between net mineralisation rates in samples of the same soil is also reduced as temperature is increased (Addiscott, 1983). Stanford and Epstein (1974) observed optimum net mineralisation rates between 0.33 and 0.1 bar, where water occupied 80 - 90 % of the pore space. There was a steep decline in net mineralisation rates at moisture contents less than 0.33 bar. There is a significant interactive effect of soil moisture and temperature on net mineralisation rates. Cassman and Munns (1980) generated a response surface for net mineralisation rates dependent on temperature and moisture from data obtained in the laboratory. The absolute amounts of moisture in the soil are not so important as moisture distribution in controlling net rates of mineralisation (Cassman and Munns, 1980). Gross mineralisation rates also seem to track average temperatures through the season, but with a marked dependence of the rate also on moisture (Nishio and Fujimoto, 1989).

Mineralisation may be stimulated in the rhizosphere due to release of carbon from roots by cell death and/or exudation of soluble C compounds. However, increases in N supply to the plant may only be significant if the bacteria are grazed (Robinson *et al.*, 1989). Although mineralisation rates in soil decrease with depth, correlating with the decrease in organic matter and micro-organisms, a substantial proportion of N released may be mineralised at up to a metre depth (Cassman and Munns, 1980).

Cumulative gross mineralisation through the season greatly exceeds the uptake of N by a maize crop (Nishio and Fujimoto, 1989). This is due to competition for the NH_4^+ produced between micro-organisms and roots. Jansson (1958) postulated that there is a continuous mineralisation-immobilisation turnover in soils, as a result of the constant turnover of the microbial biomass (Jenkinson and Ladd, 1981; Juma and Paul, 1984). This simple model has formed the basis of many models of the soil nitrogen cycle (Paul and Juma, 1981; van Veen *et al.*, 1984). However, the soil internal nitrogen cycle model proposed by Jansson (1958) does not always fit all the observed results (Myrold and Tiedje, 1986; Drury *et al.*, 1991). A modification of the model has been proposed by Drury *et al.* (1991), where N transformations may also be limited by the spatial separation of N pools in the soil. In modelling mineralisation processes, it is important to relate the N cycle to the C cycle (Addiscott, 1983), since the balance between available energy and N will largely control the rate of release of N from added crop residues and manures. This is discussed further in Section 1.5.6.

1.4.5.2 Nitrification

This is defined as the process by which NH_4^+ is oxidised to NO_3^- and occurs world-wide in terrestrial, aquatic and sedimentary ecosystems (Schmidt, 1982). The process is strictly biological and is mainly mediated by chemoautotrophic bacteria (Nishio and Fujimoto, 1990). The elucidation of the mechanism and micro-organisms involved began in the middle of the 19th century, an excellent review of the history of the developments in nitrification theory and microbiology was presented by Macdonald (1986). The process occurs in two stages, mediated by two separate groups of microorganism: the oxidation of NH_4^+ to NO_2^- (nitrite); then the further oxidation of NO_2^- to NO_3^-. Nitrite rarely accumulates in soils (Nishio and Fujimoto, 1990). Heterotrophic nitrification, mediated by fungi, may also be important especially in acid soils, such as those under coniferous forests (Killham, 1986).

Nitrification is controlled by the supply of NH_4^+ and HCO_3^- from the soil solution, temperature, moisture, pH and aeration (Schmidt, 1982). Nitrification was rapid in field soils even at temperatures as low as 3.5 °C (Recous *et al.*, 1988). Under laboratory conditions, nitrification rate increases with

temperature over the range 2.5 °C to 20 °C, but below 5 °C the temperature sensitivity of the process may increase in some systems (Addiscott, 1983).

The number of nitrifiers is almost constant, when soil NH_4^+ concentrations are low and Michaelis-Menten kinetics can be used to model the conversion (Nishio and Fujimoto, 1990). When concentrations of NH_4^+-N are very high (exceeding 300 $\mu g\ ml^{-1}$), inhibition of the growth of nitrifier populations has been observed (Nishio and Fujimoto, 1990). However, such concentrations are only likely to occur in soils close to fertiliser granules.

1.4.5.3 Denitrification

Denitrification is an irreversible process and as such represents a loss of nitrogen from the biosphere to the atmosphere. N_2O, one of the gaseous products of the process, may cause depletion of ozone in the troposphere and may also contribute to the greenhouse effect (Firestone, 1982). N_2O may also be produced during nitrification (Smith and Arah, 1990).

Denitrification occurs in anaerobic sites in the soil, where a number of microbial species use NO_3^- in place of O_2 as the terminal electron acceptor in respiration (Powlson, 1988). The bacteria responsible are ubiquitous in soils and represent a significant fraction of the normal heterotrophic microbial biomass (Focht, 1982). Fungi have also been shown to be capable of reducing NO_2^- and NO_3^- anaerobically (Shoun *et al.*, 1992).

NO_3^- is sequentially reduced and both N_2 and N_2O are released as gaseous products, with the relative amounts of each gas depending on environmental conditions especially pH (Focht, 1982). Organic C forms the main electron donor for the process, which will only occur in the absence of oxygen and in the presence of NO_3^-. Significant denitrification fluxes in the field may be observed, where high concentrations of NO_3^- occur and where the system is not C limited (Smith and Arah, 1990) and in response to rainfall events (Recous *et al.*, 1988).

1.4.5.4 Leaching

Leaching is defined simply as the removal of materials in solution from soil (Brady, 1974). Since both ammonium and nitrate ions are found in the soil

solution, they are at risk to losses from the soil by leaching, during periods when drainage exceeds evapotranspiration (excess rain). However, ammonium ions are held in the soil by cation exchange with soil colloids and are therefore less vulnerable to leaching. The main losses of nitrogen from the profile by leaching are therefore losses of nitrate (Rosswall *et al.*, 1990).

Loss of nitrate in water may include water movement downward through soils (drainage), surface run-off and lateral flow (Powlson, 1988). The texture and structure of soils, which control the size and number of transmission pores, have a marked effect on the soil water balance and hence leaching losses, with sandy soils losing nitrate more readily than loams and clays (Wild, 1988). Nitrate at risk to leaching during winter is mainly derived from organic N mineralised in the late summer and autumn (Powlson, 1988) and any excess fertiliser remaining in the soil after harvest. The practice of applying N in the autumn to winter cereals has now been almost completely abandoned, since this N was mainly lost by leaching (SAC, 1990). Soil and crop management affects the amounts of nitrogen lost by leaching. Wild (1988) noted that under cut grass leaching losses of 0.5 - 6 kg N ha^{-1} were observed during the period of excess winter rain, while following cultivation of leys for cereals a leaching loss of 200 kg N ha^{-1} was observed over two years (Cameron and Wild, 1984).

1.4.6 Predicting soil N supply

Potential soil nitrogen supply by mineralisation to plants during the growing season is very difficult to predict. There have been many attempts to obtain a direct measurement of the likely release of nitrogen from a given soil by a wide variety of methods. However, to date no methods have been widely incorporated into recommendation systems for UK farmers because of either the time and effort required to obtain the necessary data, or the unreliability of the results.

Biological incubation methods, involving the monitoring of mineral N before and after a period of incubation under different conditions, have been used (Keeney and Bremner, 1967) and have been shown to correlate well with nitrogen uptake by a crop (Ryan *et al.*, 1971; Baerug *et al.*, 1973). Stanford and Smith (1972) devised an incubation procedure, which with some modifications has become widely accepted (Smith *et al.*, 1980; Skjemstad *et al.*, 1988). The

nitrogen mineralisation potential of the soil (N_0) was estimated from the cumulative amount of nitrogen mineralised over the incubation period, based on the assumption that the nitrogen mineralised obeys first-order kinetics. First-order kinetics are not always observed (Tabatabai and Al-Khafaji, 1980; Macduff and White, 1985) and more complex two component models have also been proposed to describe the release of N from the soil (Deans *et al.*, 1986).

Anaerobic incubations have also been carried out as they are much simpler than aerobic incubations (Keeney, 1982). Such incubations are based on a technique (Waring and Bremner, 1964) modified by Keeney and Bremner (1966). This has shown to be highly correlated with nitrogen uptake by plants and has been used widely especially in forestry (Geist, 1977; Stalk and Clapp, 1980). The nitrogen released on anaerobic incubation has been shown to be released dominantly from the biomass (Myrold, 1989).

Chemical extraction techniques have been devised to estimate potentially available N. Although they do not simulate the microbial activity which leads to release of nitrogen from organic matter, they are much more convenient and rapid than biological incubations. Mild chemical extractants have shown the most promise (Stanford, 1982) and are believed to derive much of their nitrogen from the newly immobilised N in soil (Jenkinson, 1968; Kelley and Stevenson, 1985). Techniques using KCl solutions at differing concentrations and temperatures over various periods of time have been developed more recently (Oien and Selmer-Olsen, 1980; Whitehead, 1981; Gianello and Bremner, 1986), since extracts in KCl can be analysed using the same procedures as for assessing mineral N. McTaggart and Smith (1993) showed that nitrogen released by a method based on that of Gianello and Bremner (1986) correlated very well with soil nitrogen uptake in malting barley in Scotland.

Many models have been developed, which aim to predict the soil supply of nitrogen to crops and therefore aid in the prediction of fertiliser requirement and of potential leaching losses of nitrate from soils (Addiscott and Whitmore, 1987; Addisott *et al.*, 1991; de Willingen, 1991; Vereecken *et al.*, 1991). However, these have met with only limited success. Van Keulen and Stol (1991) concluded that insufficient quantitative knowledge of the basic processes involved in soil N turnover and crop uptake is currently available to allow the development of an accurate model. Soil processes, especially those which are

biologically driven, pose the most difficult problems for the modellers (de Willingen, 1991). However, as our basic understanding of these processes continues to increase, accurate and useful models will almost certainly be developed.

1.5 Organic sources of N.

1.5.1 Animal manures

Animals only use part of the N they are fed for production. The N compounds not absorbed during digestion will be egested in faeces. The proportion of undigested N will vary in different diets. Not all the protein digested by an animal will be metabolized and much will be excreted from the blood to urine (McDonald *et al.*, 1988). Total manure production varies widely from species to species, reflecting differences in metabolism and also in characteristic diets (Cooke, 1982; Spedding *et al.*, 1981). There is also variability within species, depending on age and productivity. Milking cows will retain about 80 % of feed N, whereas store cattle will only retain 1 - 5 % of feed N (Bouldin *et al.*, 1982). The main nitrogen compounds contained in animal manures are undigested and partly digested proteins in faeces and urea or uric acid in urine, depending on the animal species. Urine and faeces also contain a small amount of NH_4^+. However, since urea and uric acid are rapidly hydrolysed in soil, they are usually included in the determined mineral N content of an animal manure.

Animal manures may be managed on the farm in a number of ways. Historically, the excreta from housed animals was mixed with straw and stored before application to the land, to form farmyard manure (Hall, 1921). In organic farming systems, the composting of such manures, by controlled aerobic decomposition, was encouraged (Sykes, 1946). Composting attempts to recreate the conditions which would occur in an undisturbed ecosystem, where organic matter builds up on the soil surface and is not incorporated, as in agricultural ecosystems (Lampkin, 1990). Liquid manures and slurries are now more common, where wastes from housed animals are collected, mixed with water and stored in tanks or pits on the farm (Spedding, 1983).

The main loss of nitrogen during storage and composting of manures and residues is as ammonia gases (Kirchmann, 1985). If energy-rich substances are available to micro-organisms in large quantities, however, volatilisation does not

occur as ammonia is immobilised as microbial N (Kirchmann, 1985). The loss and retention of N during decomposition in the absence of soil is dependent on the C sources present (Kirchmann, 1985). During composting, the total amount of N in the manure declines with time, although more slowly than C, so that the N concentration of the compost increases (Kirchmann, 1985). Leaching losses from unprotected heaps of farmyard manure or compost may also be considerable (Lampkin, 1990). Animal manures are very variable in nutrient content, which is controlled mainly by the animal species, the production system including diet and the collection and storage system used on the farm.

1.5.2 Green manures

Green manuring is the practice by which a green crop is turned under for the enrichment of the soil (Pieters, 1927). The crops are used to supply nitrogen or to increase the organic matter content of the soil. The first recorded use of green manures is in China more than 3,000 years ago (Mahler and Hemamda, 1993). The practice of green manuring is recorded through history (Pieters, 1927), and green manures have continued to be used widely in Europe, especially Germany, to the present day. The ploughing-out of leys, to supply N to following crops, has also been widely practiced in mixed farming systems (Spedding, 1983). The expansion of use of inorganic fertilisers after the Second World War, reduced use of green manures in Britain, but in response to environmental concern about nitrate leaching over winter, the use of cover crops is increasing.

Where the aim of green manuring is to supply nitrogen to a following crop, then the crop should be ploughed under when in an immature succulent stage (Lampkin, 1990). Legumes are commonly used as green manure crops, as in this way nitrogen may be added to the system by nitrogen fixation. Cover crops are ploughed under in late winter/early spring, thus forming a green manure. Catch crops are used to temporarily conserve N in the soil-plant system and prevent losses of nitrogen (Jensen, 1991), and will then be used for stock feeding or incorporated as green manures to the soil.

The efficient use of green manures requires knowledge of the plant growth and decomposition dynamics of appropriate species (Marstorp and Kirchmann, 1991). Barney (1987) investigated the suitability of several legume species for

use as green manures in southern Britain. He showed dry matter yields of 1 - 2 tonnes ha^{-1} with N uptake ranging from 20 kg N ha^{-1} in fallow plots with weeds to 42.9 kg N ha^{-1} with white clover. Non-leguminous species were found to be more efficient in removing residual N from the profile in autumn than legumes (Atallah and Lopez-Real, 1991), because they show rapid initial growth and N uptake. Green manure crops may also be undersown with the previous crop, where yields are not reduced.

1.5.3 Crop residues

Crop residues are used in a number of ways. Above ground residues (eg. straw and haulms) may be cut and incorporated directly following harvest or composted on the farm for incorporation later. In the USA, 363 million tonnes of crop residues are estimated to be produced annually (Smith and Peterson, 1982), containing approximately 4 million tonnes of N per annum. The input of organic matter and nitrogen through root residues remaining in the soil after harvest may also be quite considerable. Olson and Kurtz (1982) estimated that after a wheat crop of 5.5 tonnes ha^{-1}, between 2 and 5 tonnes ha^{-1} of roots remain in the soil, containing approximately 18 kg N ha^{-1}. Barley roots remaining in the soil after harvest had a C:N ratio of about 20 and contained approximately 1.5 % N (Rutherford and Juma, 1989) equivalent to about 10 kg N ha^{-1} . Crop residues will vary widely in N concentration and contents of other chemicals eg. tannins (Smith and Peterson, 1982).

1.5.4 Amounts of nitrogen released

The decomposition of a range of plant materials in different soils under different environmental conditions has been comprehensively reviewed by Dickinson and Pugh (1979).

The addition of farmyard manure is often believed to increase crop yield through the supply of nitrogen for crop growth (Beauchamp, 1986). Bunting (1963), in a large number of field trials with a wide range of crops, showed that although farmyard manure and straw-sludge composts did supply some nitrogen, other effects of these manures were more important in increasing crop yields. Garner (1966) also showed this for potatoes, and attributed the effect of farmyard manure mainly to its ability to supply potassium. The use of organic

materials, especially composts, may also provide unexpected yield increases not simply related to nutrient content (Lampkin, 1990) and perhaps attributable to the prevention of crop disease or the supply of growth hormones, which have formed or been released during composting (Welty *et al.*, 1988). These effects may only become apparent when N is not limiting crop growth (Welty *et al.*, 1988). The complex nature of all organic materials added to the soil means that their effects may be difficult to attribute to only one process occurring in the soil-plant system and the following consideration of only the nitrogen supply from manures is a over-simplification of the potential effects of manures on plants and soil.

In field experiments, Pomares-Garcia and Pratt (1978) showed that for the first crop (barley) 4.5 % and 17 % of the nitrogen applied in farmyard manure and sewage sludge respectively was mineralised. This increased to 17.2 % and 40 % over a 10 month period. In a first crop, 1 % of excreted N was recovered in plants, where poultry manure had been composted, and 27 % where it was applied fresh (Kirchmann, 1990). Long-term use of any manure will increase the apparent efficiency of N uptake, due to cumulative effects (Kirchmann, 1985).

Legume residues were shown to increase crop yields to 123 - 255 % of the unfertilised yield (Welty *et al.*, 1988), but the recovery of legume N was heavily dependent on the environmental conditions under which the following crop was grown. In general recovery of green legume residues in the field is between 11 and 28 % of applied N (Ladd *et al.*, 1983; Ladd and Amato, 1986; Bremer and van Kessel, 1992; Rees *et al.*, 1993a). Higher recoveries have been reported by some workers (Hesterman *et al.*, 1987; Mahler and Hemamda, 1993). Recovery of applied N is reduced, when the period of fallow before sowing of a following crop is long (Ladd *et al.*, 1983; Ladd and Amato, 1986). Differences in recovery of N from legume species is also recorded (Yaacob and Blair, 1980; Hesterman *et al.*, 1987; Mahler and Hemamda, 1993).

Powlson *et al.* (1985) showed that 12 % of N applied in straw was recovered in the following winter wheat crop, though yields were slightly depressed, and 78% of the straw N remained in the soil. Following additions of straw, microbial biomass increased, with 30 % of straw N recovered from the biomass within 5 days (Ocio *et al.*, 1991). Similar data was obtained for the legume *Sesbania aculeata*, with crop recoveries of only around 5 % of N applied, and much of the

N immobilised in biomass and humic acids (Azam *et al.*, 1985). The release of nitrogen from wheat and lentil straw is very similar (7 % of N applied), with slightly higher N losses recorded from where lentil straw was incorporated compared to wheat straw (Bremer and van Kessel, 1992).

In the field, lower N recoveries from manures and residues are measured than in pot experiments or laboratory incubations (Rees *et al.*, 1993a), since environmental effects can be excluded, the volume of soil available for root exploration is limited and losses by leaching are reduced. In a 16 week incubation study with a range of manures, Serna and Pomares (1991) showed 0 - 39 % of the nitrogen applied was mineralised. With the same manures, the nitrogen taken up by maize in growth chambers was only 3.7 - 9.4 % of nitrogen added.

The amount of nitrogen released during incubations is very variable (Douglas and Magdoff, 1991). Nitrogen released from sewage sludges has been shown to be close to 50 % by some workers (Epstein *et al.*, 1978; Magdoff and Amadon, 1980) and much lower by others depending on the sludge pre-treatment (Epstein *et al.*, 1978; Parker and Sommers, 1983). Chescheir *et al.* (1986) found the highest mineralisation rate constant (k) for poultry manure and the lowest for a low ammonium containing sludge. In incubations, Marstorp and Kirchmann (1991) showed that 17 - 35 % of the nitrogen from legume residues was mineralised after 115 days, depending on legume species.

1.5.5 Timing of nitrogen release

The decomposition of plant and animal residues in soil is believed to be a two-stage process. Decay is initially rapid, this process is then followed by a process of much slower decay, since the humic substances synthesised during the first stage of decomposition of plant material are much more stable to decay than the original plant components (Jenkinson, 1981). The length of the first stage is controlled by the residue quality, environmental conditions and soil factors. These will be considered in detail in Section 1.5.6. Once the second stage is reached the residues are considered to have joined the 'passive' pools of soil organic matter.

The net accumulation of nitrate and ammonium from soil-waste mixtures has been shown to follow simple first-order kinetics by some workers: Parker and Sommers (1983) for a range of sewage sludges; Chescheir *et al.* (1986) for a wide range of animal manures (dairy, poultry, swine) and a domestic sewage sludge; Frankenberger and Abdelmagid (1985) for legume green manures. Amounts of potentially mineralisable nitrogen in manures have therefore been estimated by N_0, where accumulation of mineral nitrogen is described by the first order equation (Stanford and Smith, 1972):

$$N_{min} = N_0 (1 - e^{-kt})$$

N_{min} = accumulated mineral nitrogen
N_0 = potentially mineralisable nitrogen
k = rate constant for mineralisation
t = time.

Following studies of the release of available nitrogen from poultry manure (Hadas *et al.*, 1983; Sims, 1986; Bitzer and Sims, 1988; Serna and Pomares, 1991), a two component first-order model was used to describe mineralisation of nitrogen from poultry manure. The presence of two organic nitrogen fractions in the manure, rapidly and slowly mineralisable, was therefore suggested (Sims, 1986; Bitzer and Sims, 1988; Serna and Pomares, 1991). Hadas *et al.* (1983), however, suggested that the initial rapid release of ammonium was due to simple chemical hydrolysis of uric acid type compounds rather than microbially mediated mineralisation. This was followed by slow mineralisation of organic substrates by microbial action. Hydrolysis of urea and uric acid in soil may also be carried out by the enzyme, urease. It is unclear whether release of N in this way would be defined as mineralisation.

Lindemann and Cardenas (1984) also suggested that the release of nitrogen from a sewage sludge at various rates is best represented by a two component first-order model. Simple first order models have also been used (Hsieh *et al.*, 1981; Chescheir *et al.*, 1986). Maximum rates of mineral N accumulation were observed in the first week (Lindemann and Cardenas, 1984).

Lags for onset of nitrification have been observed from some manures (Castellanos and Pratt, 1981; Hsieh *et al.*, 1981; Chae and Tabatabai, 1986). Ryan *et al.* (1973) found a lag in the onset of nitrification at high rates of anaerobically incubated sewage sludge, but after the first week, rates of

nitrification in these treatments equalled or exceeded the rates in the other treatments. Reddy *et al.* (1980) observed a delay in nitrate accumulation with poultry and beef manures, but not with swine manures incubated under the same conditions. A lag in the onset of mineralisation of nitrogen from a swine manure was noted by Serna and Pomares (1991), but not with the sheep, poultry or cattle manures used in the study.

Chae and Tabatabai (1986) studying the release of nitrogen from animal manures, sewage sludges and plant materials could not fit first order accumulation kinetics in all the cases. They, therefore, suggested four common patterns of nitrogen release from manures:
1) Initial immobilisation, followed by mineralisation. In some cases, the immobilisation phase lasts the duration of the incubation.
2) Rate of release decreasing with time, (simple or double component first-order kinetics).
3) Steady linear release, (zero order kinetics).
4) Rapid initial release, followed by slow linear release.

All the studies described above have measured net releases of nitrogen from manures and are unable to separate nitrogen released from the soil organic matter and that released from the manure, except by difference. Kirchmann (1990) used uniformly labelled chicken excrement to produce two contrasting manures and observed no differences in net nitrogen mineralized from composted and dried-fresh manures. However, the labelled nitrogen uptake from the composted manure (composed of a single pool of stabilized nitrogen) was linear with time (zero-order), while the labelled nitrogen uptake from the dried-fresh manure (composed of a pool of labelled excrement nitrogen and an unlabelled pool of straw nitrogen) followed a curvilinear course (approximating first-order).

The release of nitrogen by mineralisation is the result of a number of simultaneous and sequential processes and it is not surprising that as a result, the kinetics should be indeterminate, especially where the release of nitrogen from heterogeneous materials, such as manures and wastes.

1.5.6 Factors controlling nitrogen release.

On a field scale the amount of available N in soils is dependent on a number of interacting factors: soil characteristics (texture, structure, organic matter etc.), rainfall and temperature patterns, and agronomic management practices such as tillage and rotations. The quantity of residue or manure N that will be available to crops is influenced by application rate, timing and method of application, as well as residue characteristics (Douglas and Magdoff 1991). Results of laboratory experiments, where incubation conditions are varied, are usually just as difficult to interpret, due to interactions between soil micro-organisms, the potentially mineralisable manure substrate, temperature and moisture effects (Sims 1986).

1.5.6.1 Environmental factors

Sims (1986) observed some mineralisation of poultry manure, even at 0 °C, but nitrification was severely inhibited at this temperature, with a lag of 90 days before any nitrate was produced. Over the temperature range 10 - 40 °C the Q_{10} for decomposition is usually found in the range 2 - 3 (Jenkinson, 1981). Increasing temperature increased mineralisation, but nitrification was delayed at 35 °C, though not permanently inhibited (Hadas *et al.*, 1983). It has been shown that the pattern of decomposition is not changed by increasing temperature, only the rate constants are changed (Jenkinson, 1981). When comparisons between manures are made, then temperature effects are small compared to the differences between manures (Pratt *et al.*, 1973).

Mineralisation is reduced by sub-optimal levels of moisture (Sims, 1986). Hsieh *et al.* (1981) observed that there was no significant effect of moisture levels between 0.06 bar and 0.33 bar. In waterlogged soils, anaerobic conditions lead to the proliferation of an altered microbial population and the decomposition pathway is altered (Jenkinson, 1981). Organic acids may accumulate and gases such as ethylene and methane may be released.

1.5.6.2 Soil factors

Soil type does not seem to be the most important factor in controlling the amount of mineral nitrogen released from manures (Wagger *et al.*, 1985). CO_2

release during decomposition was largely shown to be independent of soil properties (Miller, 1974). However, decomposition is inhibited in strongly acid soils (Jenkinson, 1981), probably due to a changed and less active soil flora and fauna. Barbarika *et al.* (1985) observed that as soil total nitrogen increased, mineralisation increased and as soil C:N increased, mineralisation decreased.

Working with two soil types, Chescheir *et al.* (1986) observed that net available nitrogen released from manures was lower on the heavier soil, although mineralisation of native soil organic matter was greater on the heavier soil. The nitrogen unaccounted for after an incubation was higher in a heavier soil, and the soil texture seems to be a major factor in controlling nitrogen losses (Lindemann and Cardenas, 1984). There was a moisture/texture interaction on net N accumulation (Chescheir *et al.*, 1986), which may be attributable to increased losses of N by denitrification. Reduced recoveries of nitrogen applied are seen in waterlogged and compacted soils due to poor root system development and increased denitrification (Redman *et al.*, 1989).

1.5.6.3 Management factors

Jenkinson (1977) showed that the decomposition of organic materials in soils under aerobic conditions was substantially independent of the amount added, except in the case of short term incubations with low N-containing residues. The mineralisation of manures and residues have been thought to be largely independent of the rate of application (Ladd *et al.*, 1983; Rees *et al.*, 1993a), but decreased amounts of mineralisation are seen at increased rates of sludge application (Pomares-Garcia and Pratt, 1978). Epstein *et al.* (1978) found no effect of increasing the rate of sludge addition on the percentage of the N in the residue mineralised. However, Lindemann and Cardenas (1984) observed that rates of mineralisation were affected by the rate of sludge addition, net sludge mineralisation as a percentage of nitrogen added was also lower at higher rates of addition (Ryan *et al.*, 1973; Lindemann and Cardenas, 1984). Recovery of added inorganic nitrogen added also decreased with increased rate of anaerobic sewage sludge addition (Ryan *et al.*, 1973). At very high rates of manure and residue addition, the recovery of N in crops may be reduced due to a reduction in mineralisation (Rees *et al.*, 1993a). This is thought to be linked to the development of anaerobic sites, due to high oxygen demand.

The effectiveness of manures for following crops is controlled by the timing of application. Recoveries of manure N decrease with increased time between application and crop demand (Bouldin *et al.*, 1982), due to the increased risk of losses of mineralising N. Francis *et al.* (1992) showed that the earlier in the autumn/winter a leguminous pasture is broken up, the more N is lost by leaching. Delaying cultivation of pasture until late autumn or winter succeeded in minimising N loss (Francis *et al.*, 1992). Increasing N uptake by spring wheat was also seen with decreasing amount of time under fallow.

Nitrogen release is very significantly reduced by composting sludges and manures (Epstein *et al.*, 1978; Parker and Sommers, 1983; Douglas and Magdoff, 1991), as highly stable N compounds are formed during composting. Drying of slurries before use also reduced the nitrogen mineralised (Laura and Idnani, 1972), since the slurries tended to flocculate on drying, increasing the stability of nitrogen containing colloids.

The recovery of green manures was not affected by the previous crop rotations of the soil and was only slightly affected by the crop following the green manure (Janzen and Radder, 1989).

1.5.6.4 Manure factors

Patterns of mineralisation were shown to be a function of sludge pretreatment (Parker and Sommers, 1983). However, there can be large variability within any pre-determined residue type (King, 1984). The quantity of N supplied by a legume to a following crop varies with the cultivar, growth stage and environment (Welty *et al.*, 1988). Residue quality has an effect in determining the rate at which the initial stages of decomposition will proceed (Jenkinson, 1977). However, after this initial phase roughly similar amounts of resistant material will persist.

The total nitrogen content of manures and nitrogen mineralised has been shown to be strongly correlated in some experiments (Castellanos and Pratt, 1981; Douglas and Magdoff, 1991). There has also been shown to be significant correlations between the organic nitrogen concentration in manures and nitrogen released during incubations (Parker and Sommers, 1983; Douglas and Magdoff, 1991). For a range of crop residues, there was also a significant

relationship between total nitrogen of the residues and nitrogen released (Iritani and Arnold, 1960; Frankenberger and Abdelmagid, 1985). This relationship was improved when water-soluble nitrogen was also included as a factor, and it was suggested that the pool of water-soluble nitrogen was approximately twice as effective as the insoluble nitrogen pool in releasing nitrogen during these incubations (Iritani and Arnold, 1960). The critical breakpoint separating net immobilisation from net mineralisation for plant residues has been suggested as: 1.5 % N (Smith and Peterson; 1982) or 1.73 % N (Frankenberger and Abdelmagid, 1985).

The carbon:nitrogen ratio of manures was not significantly correlated with the amount of nitrogen released from manures by some workers (Castellanos and Pratt, 1981; Douglas and Magdoff, 1991), but it did correlate significantly with the fraction of organic nitrogen released by the residues (Douglas and Magdoff, 1991). However, Barbarika *et al.* (1985) found that as the C:N ratio of sewage sludge increased, mineralisation decreased linearly and the C:N ratio was the only sludge property included in a stepwise regression procedure. A curvilinear relationship has been found between the C:N of a range of plant materials and the total nitrogen released (Iritani and Arnold, 1960; Frankenberger and Abdelmagid, 1985), with the dividing line between net mineralisation and net immobilisation at a C:N ratio of 19 - 20. The turning point between net immobilisation and net mineralisation has also been determined as a C:N ratio of 15 (Castellanos and Pratt, 1981; Kirchmann, 1985; Beauchamp, 1986; Marstorp and Kirchmann, 1991). Kirchmann (1990) showed that while the net mineralisation of N from manures with different C:N ratios may not be different, they may affect the gross processes of the soil N cycle differently. Initial immobilisation, seen with some manures, is usually attributed to a high C:N ratio of the manure or plant residue (Castellanos and Pratt, 1981; Chae and Tabatabai, 1986). The increased recovery of N from Siratro (a leaf legume) compared to soybean was attributed to its lower C:N ratio of 16 compared with a C:N of 28.4 in soybean residues (Yaacob and Blair, 1980). The use of simple C:N ratios has been criticised since the C and N is not all equally available to micro-organisms, and it is the available C: available N which is the crucial factor (Reinertsen *et al.*, 1984; Frankenberger and Abdelmagid, 1985).

Beauchamp (1986) observed yield responses of maize to three different manures, and demonstrated that they were closely correlated to the amounts of

ammonium applied. Douglas and Magdoff (1991), also showed that the amount of mineral nitrogen present in the manure initially was related to the percentage of organic N, which was mineralised from a wide range of organic manures. The nitrogen release from poultry manure in the field resembled a single application of inorganic nutrients (Bitzer and Sims, 1988), suggesting that for a first crop the inorganic and easily hydrolysed N in the manure might be the main source of N.

Douglas and Magdoff (1991) found that the nitrogen release for a wide range of organic materials was significantly correlated with organic carbon. However, for a range of crop residues, Iritani and Arnold (1960) could find no relationship between total carbon or lignin content and amount of nitrogen released. The overall decomposition rate of wheat straw in the early stages was found to be controlled by the size of the soluble C pool in the straw and the availability of a second C pool, which is not water-soluble (Reinertsen *et al.*, 1984).

1.5.7 Losses of applied nitrogen

The main losses of N from manures added to the soil occur within the first two weeks of incubations (Chescheir *et al.*, 1986). These are often attributed to denitrification and ammonia volatilisation, which are irrecoverable to the system and immobilisation, which may be recoverable later in incubations due to biomass turnover.

Disappearance of NO_3 from the system does not necessarily indicate denitrification is occurring (Epstein *et al.,* 1978), a substantial proportion of the NO_3 may be recycled through organic matter. However, the addition of large quantities of manure will create, at least initially, conditions in the soil favouring denitrification. Waterlogging was increased in plots, which had received high rates of manure application (Cooper *et al.*, 1984). High levels of active carbon are commonly found in residues, and it has been postulated that this leads to the development of aerobic-anaerobic micro-environments, where simultaneous or alternate nitrification and denitrification are able to occur. (Epstein *et al.,* 1978; Hsieh *et al.*, 1981). Unaccounted for nitrogen, comparing organic nitrogen lost to mineral nitrogen recovered, is generally shown to increase with increasing application rate of manure (Cooper *et al.*, 1984) and on heavier soils (Lindemann and Cardenas, 1984; Chescheir *et al.*, 1986).

Ammonia volatilisation losses can be significant from manures, even after application to the field. Where manure is incorporated rapidly, losses can be minimised, thus only 5 - 10 % of N applied was lost by ammonia volatilisation in studies by Pratt *et al.* (1976) and Beauchamp (1986). Ammonia volatilisation losses are largely controlled by the amounts of NH_4^+-N added to soils, the soil's water content and the pH of the soil. Only 2 - 4 % of applied N was lost from aerobically-composted pig manure over 9 days were, while 14 % of applied N was lost from anaerobically-composted pig manures (Bernal and Kirchmann, 1992), reflecting differences in the NH_4^+ concentrations of the manures.

Flowers and Arnold (1983) observed losses from the ammonium pool of a soil incubated with a high NH_4^+ pig slurry, however they only measured low losses by volatilisation, and the losses were attributed to immobilisation. There was no evidence in this experiment that immobilised N was released, even after prolonged incubation (Flowers and Arnold, 1983). Two years after a sequence of annual applications of manure, Cooper *et al.* (1984) observed that much of the nitrogen applied (14 - 45 %) remained in the 0 - 6 m profile in organic forms. The amount of nitrogen remaining in the profile was greatest at the higher application rates. Additions of cereal straw have also been seen to cause significant increases in the soil microbial biomass (Allison, 1987).

Increased losses of nitrate by leaching will not necessarily follow immediately after additions of manures, but residual effects may be seen in the soil profile. Two years after an annual sequence of manure applications and NO_3^- concentration was still significantly higher at 6 m in the manured compared to the control plots (Cooper *et al.*, 1984). Where pea crop residues were incorporated at harvest, the leaching losses overwinter under a following winter wheat crop were between 40 and 60 kg N ha^{-1}, where no haulm and haulm had been incorporated respectively (Rees *et al*, 1993a). After incorporation of a lentil green manure, 24 % of the N was lost, and this was attributed to leaching of nitrate below the root zone, since it coincided with a period of high precipitation (Bremer and van Kessel, 1992).

1.5.8 Predicting availability of applied nitrogen.

Many of the chemical extraction techniques used to predict the potential availability of N contained in manures or manure-soil mixtures are based on those used on soils alone. However, losses of N from the soil system can be large and therefore even where a good index is found it will only be able to predict potential availability (Chescheir *et al.*, 1986). Many indices are able to rank nitrogen availability from a range of manures (Serna and Pomares, 1991). Parker and Sommers (1983) found that incubations of soil-manure mixtures resulted in a much greater range in the amounts of nitrogen released than extraction procedures, indicating that the use of chemical indices may oversimplify the complex process of N release from manures.

Extractions with acidified and alkaline permanganate gave significant correlations with N released from manures in aerobic incubations (Castellanos and Pratt, 1981; Parker and Sommers, 1983; Serna and Pomares, 1991). The ammonium released on autoclaving also gave a significant correlations (Parker and Sommers, 1983; Douglas and Magdoff, 1991; Serna and Pomares, 1991). An extraction with 6 M HCl did not show a good correlation with N release when considered alone (Serna and Pomares, 1991), but it was the best of the chemical indices in predicting maize nitrogen uptake, when it was included in a multiple linear regression model, with C:N and total nitrogen content. Extractions with acid permanganate, strong and weak acids gave predictions only slightly greater than the ammonium content of the manures (Chescheir *et al.*, 1986). The pepsin digestion method adapted from nutrition studies gave a good correlation with N release from manures (Castellanos and Pratt, 1981; Serna and Pomares, 1991). Pepsin digestion predicted a large release of organic nitrogen (Chescheir *et al.*, 1986). The amount of ammonium present in a Walkley-Black digest (Douglas and Magdoff, 1991) has also been suggested as a chemical predictor for the nitrogen released by a wide range of organic residues.

The carbon dioxide evolved during a short period of incubation has been shown to be a good index of nitrogen availability (Castellanos and Pratt, 1981). This may be because the amount of biomass produced during the utilization of the initial available C fraction significantly influences the overall rate of decomposition in the initial stages (Reinertsen *et al.*, 1984).

There have been many attempts to use models to aid in the prediction of N release (actual and potential) from soil-manure systems. Simple empirical approaches, include the decay series approach (Pratt *et al.*, 1973; Powers *et al.*, 1975) and the calculation of efficiency indices and fertiliser equivalents for manure fractions (Sluijsmans and Kolenbrander, 1977; Beauchamp and Paul, 1989). Such models are limited in their application, since although their general principles are rarely disputed, decay series and efficiency indices have only been determined for a limited range of manures in a limited range of environmental conditions. A statistical-empirical approach was taken by Barbarika *et al.* (1985), where several sets of laboratory data were analysed and multiple regressions carried out on soil, manure and environmental factors. Models derived in this way from laboratory data have only limited application in the field, but may help to increase understanding of the processes controlling N release from manures. Computer simulation models of the nitrogen cycle in soils, including the incorporation of crop residues and manures, have also been developed (Bhat *et al.*, 1980; Hsieh *et al.*, 1981; Jenkinson and Parry, 1989) with reasonable fits to laboratory (Hsieh *et al.*, 1981) and field data (Bhat *et al.*, 1980; Jenkinson and Parry, 1989).

1.6 The use of ^{15}N in soil-plant studies.

The use of an N tracer, increases the sensitivity with which a N input to the system can be followed through the soil nitrogen cycle (Saffigna, 1988). There is no radioactive isotope for N, which can be used as a tracer, equivalent to ^{14}C for carbon, since the half lives of the radioactive isotopes of N are very short: for ^{13}N 10 minutes, for ^{16}N 7 seconds and for ^{17}N 4 seconds (Jansson, 1966). However, a stable isotope of N occurs naturally, where atmospheric N_2 is composed of approximately 99.6 % ^{14}N and 0.4 % ^{15}N.

To enable ^{15}N to be used as a tracer for N, it must be possible to concentrate or deplete the concentration of ^{15}N in natural N. Chemical exchange reactions are commonly used to fractionate the isotopes, eg. the exchange reaction between ammonia gas and ammonium nitrate solution (Jansson, 1966). However, it is also necessary to be able to assume that where ^{15}N is used as a tracer, chemical and biological processes will not differentiate ^{14}N and ^{15}N. Slight differences in the behaviour of ^{14}N and ^{15}N in biological systems have been observed (Cheng *et al.*, 1964; Heaton, 1986). Such differences are largely a result of the difference

in mass between the isotopes and hence their different physical properties (Hauck and Bremner, 1976). As a result of this slight fractionation, background ^{15}N atom % in the soil usually exceeds the 0.3663 atom % ^{15}N enrichment found in inorganic salts and the atmosphere. However, it rarely naturally exceeds 0.38 atom % (Bremner *et al.*, 1966). Where high enrichments of ^{15}N are added to systems and the atom % enrichment does not approach background, then the assumption that ^{14}N and ^{15}N are used indistinguishably in the processes of the soil N cycle probably holds.

The use of ^{15}N in agricultural research was pioneered by Rittenberg and others in the late 1940's and early 1950's, it is now much more widely used as the cost of obtaining ^{15}N compounds in a range of enrichments has reduced substantially and determinations of ^{15}N enrichment are also more routine (Hauck and Bremner, 1976). The determination of the isotopic enrichment of N in samples is mostly commonly measured by mass spectrometry, after all the N in a sample has been converted to N_2. This is discussed further in section 2.6. Mass spectrometric analysis of samples had a relatively high demand for total N (up to 1 mg). Large soil samples were therefore be used, especially where $^{15}NO_3^-$ or $^{15}NH_4^+$ were to be determined, and experimental designs had to be carefully planned to deliver enough N to the mass spectrometer at an enrichment distinguishable from background (Jansson, 1966). More recently optical emission spectroscopy methods have been developed to measure ^{15}N enrichment and modern mass spectrometers have a much smaller demand for total N: 10 - 50 μg in a sample (Bremner and Hauck, 1982).

It was hoped that the development of ^{15}N methodology would provide new and exact methods to evaluate the use of fertilisers and untangle the soil N cycle (Jansson, 1966). Although the use of ^{15}N has revealed more obviously the complex nature of the dynamic and complicated chemical and biological system of the soil, ^{15}N methods have also taken us further in our understanding of the soil N cycle than would otherwise have been possible.

1.6.1 Use of ^{15}N to follow the fate of added N.

^{15}N may be introduced to the soil in a number of ways. Most commonly, experiments are carried out to follow the fate of fertiliser N, and ^{15}N may be added as $^{15}NO_3^-$ or $^{15}NH_4^+$. The fate of added crop residues or green

manures can be followed using ^{15}N, where the crops have been grown, using ^{15}N labelled nutrient solutions or fertilisers. Labelled organic manures have also been used, where stock, usually poultry, have been feed with a labelled feed and the dung and/or urine collected (Kirchmann, 1990). $^{15}N_2$ may also be used in growth chambers in investigations of N_2 fixation, by legumes and free-living N fixers. Use may also be made of natural variations in isotope abundance to trace fertiliser N transformations and the fate of legume material in the soil system (Shearer and Kohl, 1977; Karamanos and Rennie, 1980).

Such experiments allow the identification of pathways of N movement (Nason and Myrold, 1991) and the fate of N can also be quantified, since soil N can be separated from N added in fertiliser. Proportions of fertiliser applied taken up by crops, immobilised in biomass and lost from the profile by denitrification or leaching can be determined. However, the construction of full N balances using ^{15}N has proved difficult, since there are problems in obtaining quantitative recoveries of ^{15}N, especially in field experiments (Saffigna, 1988). Pruden *et al.* (1985) showed that the coefficient of variability between plots, when ^{15}N enrichment and the total N content of plant samples were measured, was very much larger than the coefficient of variability within plots. The field heterogeneity contributed a large degree of measurement error to the data. The main sources of error in trying to construct a full N balance for any system are linked to the sampling procedures for soil and plant material. This problem was reviewed by Saffigna (1988).

Most commonly experiments have been used to quantify the efficiency of N fertilisers in different forms to different crops, with different crop management strategies under different environmental conditions. The interpretation of data gained in such experiments is usually interpreted with reference to a static dilution equation (Nason and Myrold, 1991), where the proportion of N derived from the fertiliser/crop residue is obtained from the equation.

$$\frac{(\text{atom \% } ^{15}N_1 - \text{atom \% } ^{15}N_0)}{(\text{atom \% } ^{15}N_f - \text{atom \% } ^{15}N_0)} \times \frac{\text{total N in crop}}{N_f}$$

The measurements required are the ^{15}N enrichment in the fertilised (N_1) and unfertilised (N_0) crops, the total nitrogen in the crop, which received an amount of fertiliser nitrogen (N_f), and the ^{15}N enrichment in the fertiliser. The

assumption has to be made that the fertiliser nitrogen is used in addition to soil N, and the pools do not interact. This assumption, however, rarely holds.

Interaction of added N with the processes occurring in the soil can lead to misinterpretation of the results of experiments involving ^{15}N. Displacement of 'bound' unlabelled N by added ^{15}N, eg. in fixation equilibria, will cause a pool dilution effect in the added ^{15}N and lead to an overestimate of soil-derived N uptake. More commonly, pool substitution effects are seen, where labelled N added to a pool takes the place of unlabelled N that would have otherwise have been used from that pool (Jenkinson *et al.*, 1985). This is can be caused by mineralisation-immobilisation turnover processes in the soil (Jensen, 1987) and/or denitrification (Jenkinson *et al.*, 1985). Where pool substitution is significant, fertiliser recovery will be underestimated.

It is often also assumed that the ^{15}N enrichment at the beginning of an experiment can be calculated from the concentration and enrichment of the tracer added to the soil and the size of the pool to which it is being added (Hauck and Bremner, 1976). However, N losses shortly after fertilisation may affect the added fertiliser preferentially eg. volatilisation of ammonia from the soil surface, heavy rainfall stimulating losses by denitrification. It is therefore important to know whether any significant losses of N might have occurred before the fertiliser and soil N pools reach equilibrium (Harmsen and Moraghan, 1988). When fertiliser is added where the crop is already established, it is also very important to follow the spraying of a labelled solution with the addition of a large volume of water to rinse the ^{15}N added from the leaves of the crop (Barraclough, 1991). Where this is not carried out effectively artificially high N recoveries may be calculated for the plants at the following harvest (McTaggart, 1992).

The use of ^{15}N to determine fertiliser efficiencies does not therefore supersede the use of unfertilised control plots to determine fertiliser recovery (Harmsen and Moraghan, 1988). Both methods have limitations and problems in their use, especially where recovery by plant roots is not included explicitly (Rao *et al.*, 1992). The comparison of the two methods used in conjunction is able to provide more information about the underlying processes occurring in the soil (Jenkinson *et al.*, 1985; Rao *et al.*, 1992).

1.6.2 Use of ^{15}N to allow gross rates of soil processes to be estimated.

Where labelled ammonium is added to the ammonium pool in the soil and comes rapidly into equilibrium with it, the decline in the ^{15}N enrichment of the pool, as ammonium at natural abundance is introduced by mineralisation of soil organic nitrogen, can be used to calculate the gross mineralisation rate. In the same way addition of labelled nitrate can allow measurement of the gross nitrification rate. In this case, small additions of ^{15}N are usually made, where the aim is not to change the size of, but only to label, the pool of soil N (Nason and Myrold, 1991). These systems are then assumed to continue to function as in the absence of the tracer.

It is necessary to assume that the added ^{15}N equilibrates rapidly with the pool to which it has been added creating a single homogenous pool. When experiments are carried out in the laboratory with sieved and well mixed soils or litters (eg. Bjarnason, 1988; Wessel and Tietema; 1992) it is relatively easy to achieve uniform addition of ^{15}N, however, in the field it is almost impossible (Barraclough, 1991: Davidson *et al.*, 1991). Spatial variability of the soil mineral nitrogen pool and rates of transformation processes (Drury *et al.*, 1991), even in well mixed samples, will lead to spatial variability of the ^{15}N enrichment of the ammonium pool even where the application is uniform. However, Davidson *et al.* (1991) demonstrated, by simulation, that where more than 70 % of sites received ^{15}N then the calculation of gross mineralisation rates is not significantly affected. The heterogeneity of soils in the field, and especially the spatial variability of mineral nitrogen, is a serious problem for the development of *in situ* applications of isotope dilution since rate estimates are calculated using differences, which amplify errors (Myrold and Tiedje, 1986).

To simplify calculations, mineralisation and immobilisation rates are usually assumed to be constant (Kirkham and Bartholomew, 1954; Blackburn, 1979) or varying according to some known relationship (Nason and Myrold, 1991) between measurements of pool size and enrichment. The change in the ^{15}N enrichment of the ammonium pool as mineralisation and consumption processes proceed is complex and simple averages of the ^{15}N enrichment between measurements (Shen *et al.*, 1984; Guiraud *et al.*, 1989) can only give approximations of gross rates. ^{15}N enrichment only declines linearly for very

short periods of time even where mineralisation and immobilisation are proceeding at constant rates (Bjarnason, 1988).

A formal mathematical treatment to allow the calculation of gross mineralisation and consumption rates has existed since 1954 (Kirkham and Bartholomew, 1954), where the changes in pool sizes are described by differential equations, and solved analytically. Remineralisation of immobilised mineral nitrogen is disregarded and the change in amount of ^{15}N in the ammonium pool is derived only from the consumption process. The symbols are defined below.

$$\frac{d\,AL}{d\,t} = -\,c\,\frac{d\,AL}{d\,AT}$$

$$\text{Hence} \quad m = \frac{(AT_2 - AT_1)}{t}\,\frac{\log\,(AL_1AT_2 / AL_2AT_1)}{\log\,(AT_2 / AT_1)}$$

Where:

AT_1 = Total size of ammonium pool g g^{-1} at time 1
AT_2 = Total size of ammonium pool g g^{-1} at time 2
AL_1 = Size of labelled ammonium pool g g^{-1} at time 1
AL_2 = Size of labelled ammonium pool g g^{-1} at time 2

t = Time between measurements, days
@ = Natural ^{15}N enrichment of mineralising ammonium

m = Rate of mineralisation / production of ammonium, g g^{-1} day^{-1}
c = Rate of ammonium consumption, g g^{-1} day^{-1}

This framework is only valid where ^{15}N addition to the soil is high and where the ^{15}N enrichment of the ammonium pool does not approach background by the end of the incubation period. However, it is still widely used for the calculation of gross mineralisation rates (eg. Davidson *et al.*, 1991; Ambus *et al.*, 1992). This model was extended to allow for nitrogen mineralising at natural or any fixed ^{15}N abundance from the organic nitrogen pool for anoxic sediments (Blackburn, 1979) and for aerobic soils (Nishio *et al.*, 1985), deriving the decline in ^{15}N enrichment from the consumption and mineralisation processes, but still not accounting for remineralisation. Symbols as for previous equation.

$$\frac{d\,AL}{d\,t} = @\,m - c\,\frac{d\,AL}{d\,AT}$$

$$\text{Hence } m = \frac{(AT_2 - AT_1)}{t}\,\frac{\log((AL_2/AT_2) - @\,/(AL_1/AT_1) - @)}{\log(AT_2 / AT_1)}$$

These equations can be applied to $^{15}NO_3^-$ as well as $^{15}NH_4^+$ additions, when the size and enrichment of the nitrate rather than ammonium pool is measured (Schimel *et al.*, 1989) allowing calculation of both gross mineralisation and nitrification rates. A calculation method allowing calculation of gross nitrification rates where only $^{15}NH_4^+$ is added has also been developed (Wessel and Tietema, 1992).

Kirkham and Bartholomew (1955) developed a second mathematical framework allowing for nitrogen mineralisation at natural abundance and possible remineralisation of added labelled nitrogen, by estimation of the interacting organic nitrogen pool. However, this model was developed for a simple system of two pools, with mass conservation assumed, and cannot be corrected for losses to the ammonium pool other than by immobilisation to organic nitrogen.

Numerical solutions of the differential equations have also been developed, where numerical simulation by use of non-linear curve fitting, uses the measured ^{15}N abundances in the mineral N pool to fit the gross transformation rates and the size of the initial organic nitrogen pool involved (Myrold and Tiedje, 1986; Barraclough and Smith, 1987; Bjarnason, 1988). The advantages of numerical solutions is that they can be applied to any set of differential equations and the solution procedure remains the same, irrespective of the chosen set of rates, pools and other conditions (Wessel and Tietema, 1992). However, a high degree of replication is required to fit a solution with any degree of certainty and analytical models offer a quick way to calculate gross rates, so long as their assumptions have not been violated.

2.0 SITES, MATERIALS AND METHODS

2.1 Field trials

Many of the field trials were carried out at Jamesfield Farm, Abernethy, Fife (O.S. grid ref. no. 205 183). During the period Autumn 1989 - Autumn 1991, part of the farm formed the field station for the Scottish Agricultural College's Organic Farming Centre. The farm held Soil Association certified organic status. The soils are developed on terrace and raised beach deposits at the edge of the River Tay and are dominantly a complex of Carpow and Carey soil series, with some Stirling series close to the river (Speirs, 1989). The spatial heterogeneity of the soils was large, and caused problems in the selection of areas for field trials. Trials carried out in the seasons of 1990 and 1991 at Jamesfield were monitored by different groups for different purposes: for pests and diseases, the effect of plants and manures on soil processes; and many of the trials were intensively monitored by a number of groups.

Table 1 Topsoil properties (approx. 0 - 25 cm) and previous cropping in the trials at Jamesfield Farm, Abernethy.

Soil analysis		Previous cropping	
pH	6.6	1986	Oats
P status	High*	1987	Grass
K status	Moderate*	1988	Cabbage/barley
Mg status	Moderate*	1989	Oats
Available Mn	Very low*		
Available Cu	Moderate*		
Available B	Low*		
Organic matter	3 %		
Texture	Sandy loam		

* Classified according to SAC (1990)

2.1.1 Small plot trials with potato and spring barley, Jamesfield, 1990. (Full method details in Paper I).

Two field experiments were carried out with spring barley (cv. Atem) and maincrop potatoes (cv. Cara) at Jamesfield farm during 1990. The study was undertaken to compare the N uptake patterns and the efficiency of use of heap-

composted cattle manure by the two crops, at the same site, in the same season and with the same N application. The hypothesis tested was that potato (and other late-sown crops) are more suited to organic production than barley, as barley has a high N demand early in the season, when little mineral N is available.

The whole field was treated with farmyard manure (30 t ha^{-1}) in the autumn of 1989. Spring barley was drilled on the 18th April 1990 in plots 10 x 10 m. The experimental treatments were: plus or minus farmyard manure, applied at a rate of 20 t ha^{-1} at the beginning of April. The treatments were replicated four times and randomised in each of four blocks. The potato trial incorporated a second treatment: a biodynamic field spray consisting of preparations 500 and 501. These are made in a carefully specified manner from cow manure and quartz and sprayed on the crops in a highly diluted form at rates of 300 g and 4 g ha^{-1} respectively (Lampkin, 1990). Each treatment was replicated four times and the trial was laid out as a Latin Square design with plots of 16 x 16 m.

Microplots (2.5 x 3 m) of ^{15}N labelled manure were laid out within the manured plots. The labelling was carried out by incubating a portion of manure with a small amount of high enrichment (99.2 atom %) $(^{15}NH_4)_2SO_4$ for four weeks. The incubation did not significantly change the overall N content of the manure (on average 2.2 % N on a dry weight basis), but resulted in an enrichment of the manure nitrogen to c. 1 atom % ^{15}N abundance. The enrichment of the manure occurred preferentially in the active N pools of the manure, due to the method of labelling.

Above-ground plant material was harvested from plots and microplots regularly throughout the season, and a sample of tubers was also taken at lifting in late September. Soil samples were taken from the topsoil before sowing and in conjunction with the plant harvests. Soil cores were also taken in the microplots to 1 m depth after harvest. The plots were also monitored by other groups for pests and diseases, fallow areas were also maintained to study the effect of plants on soil processes.

2.1.2 Rates and form of manures for spring barley, Jamefield, 1991. (Full method details in Paper IV).

A large trial was carried out to investigate the effect of different manure types and rates on the yield and N uptake pattern of spring barley. The efficiency of use of the inorganic N pool added in the manure was also estimated using ^{15}N. The hypothesis tested was that poultry manure is a more effective N source for organic barley than heap composted manure. At the same rate of total N application, poultry manure gives higher grain yields than farmyard manure, since poultry manure contains more NH_4^+-N, which is immediately available to plants.

The trial followed a commercial potato crop, and the site had therefore been managed homogeneously in the previous season. The experimental treatments were no manure and 3 rates of heap-composted farmyard manure and poultry manure (Table 2). The treatments were replicated four times and laid out in a Youden Square design (Cochran and Cox, 1957) with four blocks of seven plots of 18 x 6 m. Manure was applied by hand to the plot surface on the 9th April and this was incorporated by Rotaspike. The trial was drilled with spring barley (cv. Sherpa) on the 18th April 1992.

Table 2 Rates of poultry manure and heap-composted farmyard manure applied to spring barley in 1992 trial at Jamesfield Farm.

Treatment	Manure rate (wet) t ha^{-1}	Total N kg ha^{-1}	NH_4^+-N kg ha^{-1}
Control	0.0	0.0	0.0
Low FYM	6.5	40.2	1.7
Medium FYM	27.8	172.2	7.2
High FYM	56.5	350.2	14.7
Low PM	2.8	27.8	3.8
Medium PM	13.0	129.6	17.9
High PM	20.4	203.7	28.2

Microplots (2 x 1.5 m) were included in each manured plot, in which the inorganic N pool of the manure was labelled with ^{15}N. A solution of $(^{15}NH_4)_2SO_4$ was applied to the plot surface with a fine mist sprayer, just after manure application, to give ^{15}N enrichments in the inorganic N pool of approximately 13 atom% for the poultry manure and 8 atom% for the farmyard manure.

Soil reference samples were taken before manure application (4th April) and following drilling (23rd April). Soil samples taken in the microplots after drilling were used to assess the actual ^{15}N enrichment in the mineral N in the soil as seen by the crop. Two N availability indices (anaerobic incubation and an extraction with hot KCl) were also used on these soils, to determine if N uptake from the soil and manure could be predicted (See Section 2.5.3). Soil and plant samples in the plots and microplots were taken in conjunction at intervals throughout the season and a plot combine was also used to assess the grain yield at the final harvest. After harvest, the ^{15}N enrichment of the soil mineral N pool was also measured. The plots were also monitored by other groups for pests and diseases, fallow areas were also maintained to study the effect of plants on soil processes.

On one treatment at harvest (the high rate of poultry manure), a grid was laid out around the microplot (Figure 6) and plant harvests were made to determine the extent of any edge effect on the ^{15}N enrichment of plants in and around the microplot.

2.1.3 Pre-planting mulching trial, Jamesfield, 1991. (Full details of methods are given in Appendix I)

Trials were carried out to examine the use of pre-planting black polythene mulches for three timings of calabrese, leek and carrot crops in 1991. Some soil parameters were also monitored, while the mulches were in place, to test the hypothesis that a black polythene mulch increases mineralisation of N in the soil, by changing temperature or moisture status, and therefore increases the supply of N to a following crop.

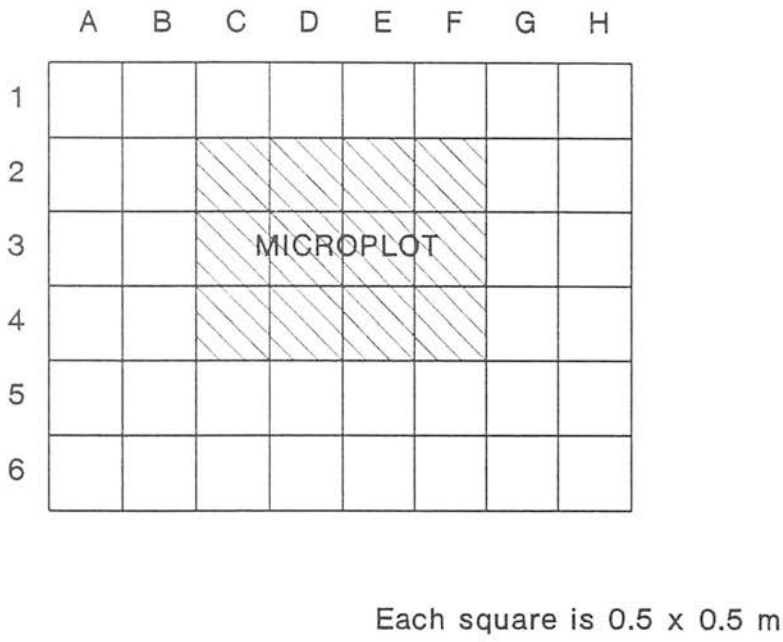


Figure 6 Grid laid out at harvest around the microplots in the plots treated with the high rate of poultry manure. A plant sample was taken from the centre of each square and the ^{15}N enrichment of the head and straw determined.

Small plot (2 m x 3 m) trials of complete randomised block design, with four replications of four pre-planting treatments were carried out. The plots were maintained as stale seedbeds (ie. all plants were removed weekly) by hand-weeding until they were covered by black polythene either 8, 4 or 2 weeks before planting. Four replicate plots remained as stale seedbeds until planting. All polythene was removed before planting and the ground was cleared of any weeds.

Separate trials were used for each of the crops or crop timings (three timings of calabrese, one timing of leek and carrot). Weed ground cover was assessed during crop growth and crop vigour was assessed according to a qualitative scale on several occasions through the season. The yield of the marketable crop and total yields were measured at harvest.

Since the trials were located in a very small area of the field, it was decided to only monitor soil parameters on one of the trials. Soil measurements were therefore only made on the carrot trial. After the ground was prepared on April 22nd, soil was sampled randomly across the trial, four replicate soil samples were then taken in every plot weekly for the first fortnight and then fortnightly until the mulch was removed. Soil temperature probes (Grant Instruments) were inserted at a depth of approximately 10 cm in all the plots at the beginning of the experiment and connected to a paper roll recorder.

2.1.4 Cover crops trial, Jamesfield, 1991-1992. (Full method details in Paper IV)

The trial was sited on the edge of the river terrace and had been under spring barley during 1991, which had been harvested in late August to leave a stubble with weeds. The cropping history of the field was uniform and the area chosen for the trial had shown no observable yield differences in the previous barley crop. Four overwinter management strategies were used, and their efficiency at using residual mineral N in the profile measured. In the spring of 1992, oats were drilled and the N uptake pattern of the crop and the efficiency of use of the N contained in the cover crops was also measured. the hypothesis tested was that maintaining soil cover overwinter reduces the risk of NO_3^- leaching and supplies N to a following cereal crop after it has been incorporated.

The overwinter treatments were: autumn ploughed and fallow; autumn ploughed with rye grass sown; autumn ploughed with a rye grass - red clover mix sown and stubble left overwinter. Four replicate plots (20 x 6 m) of each treatment were laid out in a Latin Square design in October 1991. Microplots (2.5 x 2 m) were laid out in each treatment. $(^{15}NH_4)_2SO_4$ (99.2 atom %) was applied to the microplots with a fine mist sprayer, at a rate of 1.66 kg N ha^{-1} to label the residual profile mineral N. Soil samples were taken after application of the ^{15}N, to allow initial ^{15}N enrichments of the profile residual N to be calculated.

The cover crops were ploughed out on the 11th March 1992, after a sample had been taken to determine the yield and ^{15}N enrichment of the cover crop in each treatment. The trial was drilled to oats in early April. Soil and plant samples in the plots and microplots were taken in conjunction at intervals throughout the season and a plot combine was also used to assess the grain yield at the final harvest.

2.1.5 Ploughed-out leys, Bush, 1992. (Full method details in Paper IV)

This trial was carried out at Boghall Farm on the Bush estate, about 15 km south of Edinburgh. Old ley trial plots were ploughed out and the yield and N uptake of a following spring barley crop was measured. Porous cup samplers were also installed in the plots in the autumn, to allow leaching losses over the following winter to be assessed (Grossman and Udluft, 1991). The hypothesis tested was that larger cereal yields are obtained following the ploughing out of leys, due to increased N supply. However, the ploughing out of leys for cereal production also leads to increased leaching losses of NO_3^- over the following winter.

Three old trial plots were ploughed out in late February: a 5 year grass-clover ley; a one year grass-clover ley; a 5 year rye-grass ley and these were compared with a plot under continued arable cultivation. Spring barley was drilled on 25th March and soil and plant samples in the plots and microplots were taken in conjunction at intervals throughout the season. After harvest, soil sampling continued and porous cup samplers were installed at the plots on 20th October, with samplers also installed in the continued grass-clover guard area to provide a control.

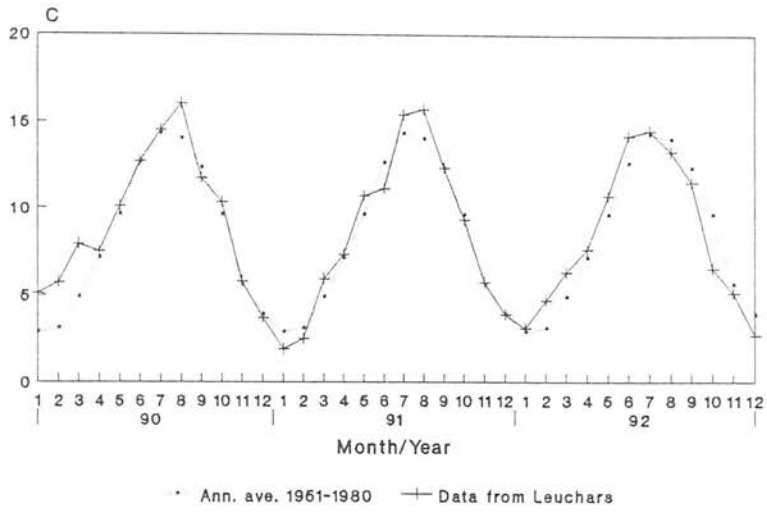
2.2 Weather at the sites 1990-1993.

Monthly rainfall and temperature data was obtained for the Jamesfield site from Meteorological Office Climatological Station 1577, at Leuchars (O.S. Grid Ref. no. 462 208), for the whole period January 1990 to December 1992 (Figure 7), and for the trial carried out at Boghall Farm in 1992, from the weather station at Bush for January 1992 to April 1993.

Table 3 Monthly rainfall and temperature data for January 1992 until April 1993, obtained from the Bush House weather station.

	Total Monthly Rainfall mm	Mean daily temperature °C
1992		
January	83.3	3.53
February	67.2	4.41
March	100.3	5.78
April	66.5	6.94
May	34.3	10.87
June	36.3	13.53
July	51.3	13.65
August	128.4	12.58
September	129.3	10.89
October	58.3	6.00
November	109.0	5.33
December	49.6	3.76
1993		
January	165.6	4.34
February	10.6	5.38
March	49.9	5.51
April	86.3	7.05

a)



b)

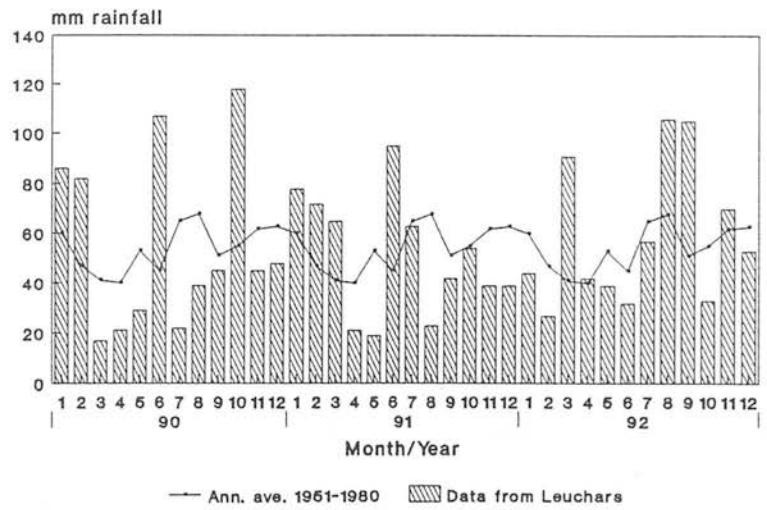


Figure 7 a) Monthly average temperatures for January 1990 until December 1992, obtained from Leuchars Meteorological Station (Met. Office Station No. 1577). Annual averages for the period 1951 - 1980 also shown.

b) Total monthly rainfall for January 1990 until December 1992, obtained from Leuchars Meteorological Station, plotted as bars. Annual average monthly rainfall shown by the line.

2.3 Laboratory and greenhouse experiments

2.3.1 Measurement of gross mineralisation rates (Full details of methods are given in Paper II)

A study was carried out on three soil types (Table 4) to investigate the potential problems in using intact soil cores to measure gross mineralisation rates under field conditions as outlined by Davidson *et al.* (1991).

The cores from the Beechgrove site were incubated at a range of temperatures (4, 10, 14 °C) for a number of incubation periods (1, 2, 4, 7, 11, 14 and 18 days) in sand tanks placed in constant temperature rooms, to determine an appropriate length of field incubation under Scottish conditions. The cores from the Glencorse and No. 3 sites were incubated at constant temperature at a range of matric suctions (300, 100, 10 and 1 kPa) for three periods of time (7, 18, and 40 days) to investigate the effect of soil moisture potential on mineralisation. Intact soil cores were injected with 5 ml of 0.17 g l^{-1} $(^{15}NH_4)_2SO_4$ solution of 99.2 atom % ^{15}N enrichment, containing 190.06 μg ^{15}N, using 1 ml syringes at 5 points around the core, following a preliminary study to determine the most suitable method of injection.

Table 4 Properties of the soils (0-20cm) used in the experiment to determine the suitability of the method of Davidson *et al.* (1991) to measure gross mineralisation rates in the field.

Site	Glencorse	No. 3 field	Beechgrove
Soil texture	Clay loam	Sandy loam	Sandy loam
O.M. %	5.0	4.0	4.8
pH	6.4	6.3	6.0
Crop	Cereals	Cereals	Pasture
Soil series	Winton	Macmerry	Winton

The size and ^{15}N enrichment of the ammonium pool was measured at the end of each incubation period and the analytical solution developed by Blackburn (1979) was used to calculate the gross mineralisation and consumption rates. Net mineralisation rates were also calculated.

2.3.2 Relationships between N availability indices (Full details of methods and soil characteristics are given in Paper III)

A large laboratory study was carried out over two winters (1991 and 1992) to investigate the source of the nitrogen extracted by a number of methods, used to assess potential N availability. The amount of microbial biomass has been suggested as an estimate of the potentially plant-available nutrient pool in soil (Marumoto *et al.*, 1982) and so biomass N was also measured to ascertain if this was the major source of the N extracted by the availability methods.

Soils from three sites widely varying in texture (Quixwood, Kettle and Jamesfield), were sampled in January 1991 and labelled with ^{15}N by incubation with high enrichment (99.2 atom %) $(NH_4)_2SO_4$, in the absence of a crop, for seven days, to allow the biomass to become labelled with ^{15}N during mineralisation-immobilisation turnover (Jansson, 1958). A further set of soil samples were taken from seventeen sites in southern Scotland, including those previously sampled in 1991, in February 1992.

A number of methods were used to assess N availability and the size of the biomass N pool in all the soils. The fumigation-extraction technique was attempted but there were great problems in digesting the extracts with sufficient gentleness to avoid hot concentrated sulphuric acid spitting out of the digestion tubes and so fumigation-incubation was used. Measurements in each case were made on three replicate subsamples taken from each of the samples of soil. Available mineral N (section 2.5.2) was determined for all the soils. Biomass N was determined by a fumigation-incubation method (Voroney and Paul, 1984), and the flush of N released, when a dried soil is rewetted (Marumoto *et al.*, 1982) was also measured. Two N availability indices, anaerobic incubation and a chemical extraction with hot 2M KCl, were also used on all the soils (Section 2.5.3). A crop of rye-grass was also grown in the greenhouse using the three labelled soils, so that the source of the N taken up by the rye-grass could be identified.

The amounts of nitrogen released by each of the techniques was measured, and where appropriate the ^{15}N enrichment of the extracts was also measured.

2.3.3 N release from a range of organic manures (Full details of methods, manure and soil characteristics are given in Paper V)

A large pot experiment was carried out in the winter of 1992/3 to compare the N release from the manures used in the field experiments and allow a more complete study of N transformations than was possible in the field. A range of characteristics of the soils and manures was also measured, and two N availability indices (Section 2.5.3) used. The hypothesis tested was that manures have a characteristic N release pattern and that this can be predicted from the measured manure and soil properties.

Two soils of contrasting textures (Quixwood and Kettle) were sampled in autumn 1992 (0 - 30 cm). The animal manures and sewage sludge collected locally were aerobically incubated with ^{15}N salt solution, made up from unused fertiliser, for four weeks . Turf was cut from a grass-clover sward at the Beechgrove site, and peas and winter cabbage were grown from seed in peat-sand mixtures. These were placed in the greenhouse and watered with ^{15}N labelled nutrient solution once a week. Straw was collected from a labelled microplot in the field after harvest, dried and chopped into 1 - 2 cm lengths.

All the pots received approximately 100 mg N kg^{-1} soil as manure, except in the case of the sewage sludge, where the N content was too low to be determined, which was therefore applied at 50 g sludge wet mass kg^{-1} of soil. The manures were mixed with the 2 kg portions of the soils by hand, with the cabbage residues being applied only to the Quixwood soil.

Three replicate pots were filled with 2 kg of the soil-manure mixtures and sown with rye-grass (1 g seed pot^{-1}). Three further replicate samples of the soil-manure mixtures were placed in plastic buckets (21 cm diameter, 18 cm high) with air-tight lids, to be incubated alongside the sown pots, but remaining unsown throughout the experiment. The Quixwood soil-manure mixtures (500 g) were also placed in Kilner Jars and incubated in the laboratory so that the N_2O production over the first five days could be studied in greater detail.

Incubations were carried out for 16 weeks. Soil samples were taken from the unsown pots at the beginning of the experiment and after 1, 2, 4, 6, 8, 10, 12, 14 and 16 weeks. Soil samples were also taken from the sown pots after 16 weeks. Plant samples (cut with a scalpel to approximately 0.5 cm above the soil surface) were taken after 2, 4, 8, 12 and 16 weeks.

Soils sampled at the beginning of the experiment were characterised by two N availability indices (Section 2.5.3) and Walkley-Black wet oxidation (Allison, 1965). Soils sampled after 16 weeks were characterised again by anaerobic incubation, and routine analysis was also carried out. Mineral N extracted from the soil samples after the anaerobic incubations and also from the soils sampled from the unsown pots after 16 weeks incubation, was analysed for ^{15}N enrichment, after steam distillation (Hauck ,1982).

Ammonia volatilisation was monitored for two consecutive periods of 5 days at the beginning of the experiment by absorbing any NH_3 produced in vials containing 50 ml 0.05 M H_2SO_4. Nitrous oxide (N_2O) production from the soils was also monitored. The buckets used for incubations had been modified to give a sampling port in the lids, and after one hour a gas sample was withdrawn for gas chromatographic analysis (Smith and Arah, 1991). Gas samples from the three treatment replicates were bulked in one syringe. Gas samples were taken immediately after mixing and after 1, 2, 4, 10 and 16 weeks. N_2O production from the Quixwood soil-manure mixtures was measured twice daily for the first five days after mixing in Kilner Jars.

2.4 Plant sampling, preparation and analysis

Plant samples from the different trials (1990 - 1992) were taken on up to five occasions throughout the season in the field (Sections 2.1.1, 2.1.2, 2.1.4). On each occasion an area 1 m^2 in each plot was cut to within a few mm of ground level for dry matter yield determination. Simultaneously, samples of two 0.5 m rows were taken from the microplot, where appropriate. At Bush in 1992 (section 2.1.5), four replicate 1 m^2 samples were taken from each treatment. Plant sampling also occurred in the pot experiments with rye-grass (sections 2.3.2, 2.3.3), where each pot was harvested with a scalpel.

All plant samples were oven dried at 100 °C. Dry matter yields were recorded and where microplot samples were present, dry matter samples were discarded. In plots without microplots, a subsample of the dry matter sample was taken and processed further alongside the microplot samples. Microplot and pot samples were milled in a hammer mill and then sub-samples (3 - 5 g) were finely ground in an agate ball mill to produce a very fine flour-like consistency. This was necessary to achieve adequate homogeneity in the very small samples taken for ^{15}N analysis (Robinson and Smith, 1991). Samples were analysed for total N content and the ^{15}N enrichment in a single determination (section 2.6).

N uptake from manures was calculated from the static isotope dilution equation (section 1.5.1) and by the difference method in comparison with the unmanured (control) plots incorporated in the field trials and pot experiment.

2.5 Soil sampling, preparation and analysis.

2.5.1 Sampling and routine analysis

Soil samples were taken at intervals during the growing season in all the field trials (1990 - 1993). The soil was sampled in the 1 m^2 area from which the plants had been removed for estimation of dry matter. Three cores from this area were taken to a depth of 30 - 35 cm and sealed in plastic bags to prevent moisture loss. Separate samples of three bulked soil cores were taken in the microplots to ensure that sufficient soil was present for the ^{15}N enrichment of the mineral N pool to be determined.

Samples were stored at 5 °C overnight, where analysis was to occur the next day. Otherwise soil samples were stored frozen (-15 °C) until analysis could be carried out. Soil texture, pH and organic matter determinations were carried out on samples of the soils from the field trial sites at the beginning of the season and soils used in the laboratory and field experiments were fully characterised before use. Soil texture was determined by particle size analysis (Gee and Bauder, 1986). Soil pH was determined in water (McLean, 1982). Soil organic matter was determined by the Walkley-Black method (Allison, 1965). Extractable P was determined by colorimetry, extractable K by flame photometry and extractable Mg by atomic absorption spectrophotometry (MAFF, 1986).

2.5.2 Determination of mineral N

Soil samples were first sieved and a subsample was taken to determine the soil moisture content, by drying in an oven at 105 °C for 24 hours. Approximately 20 g of fresh soil were accurately weighed into 250 ml conical flasks and shaken with 100 ml of 1M KCl extracting solution for one hour. The extractant was then filtered (Whatman No. 42 filter papers) and NH_4^+-N and (NO_3^- and NO_2^-)-N determined by continuous flow analysis (Crooke and Simpson, 1971; Best, 1976) in a Chemlab autoanalyser.

Where the ^{15}N enrichment of the mineral N pool was to be determined, samples of 200 g of soil were shaken with 1 l of 1M KCl for an hour. A small vial of the extract (approximately 10 ml) was taken for determination of the NH_4^+-N and (NO_3^- and NO_2^-)-N concentrations, and the remaining extract was slightly acidified and evaporated on a sand bath to approximately 300 ml. The mineral N was concentrated in boric acid indicator solution, following steam distillation with MgO and Devarda's alloy (Hauck, 1982). The indicator solution was made slightly acidic to prevent ammonia volatilisation and the solution was evaporated to dryness, in which state it was ready for ^{15}N analysis.

2.5.3 N availability indices used in the field and pot experiments.

Anaerobic incubations were carried out, after manures had been applied, on soils from the plots of the cereal trial at Jamesfield in 1991 (section 2.1.2) and on the soils from the pot experiment, 1992/93 (section 2.3.3),using a modification of the technique of Keeney and Bremner (1966). Soil samples were made into slurries by the addition of distilled water at a ratio of 1:2.5 dry soil to water in flasks. The neck was sealed and the soil slurries were incubated at 40 °C for 7 days. At the end of the incubation, ammonium was extracted from the soils by the addition of a volume of 2M KCl, equivalent to the volume of distilled water added initially. The N availability was calculated as the difference between the ammonium pool extracted after anaerobic incubation and that present in the samples extracted for mineral nitrogen.

A mild chemical extraction techique was also used on the same soils. This was based on a technique proposed by Gianello and Bremner (1986) and modified by McTaggart and Smith (1993). Samples of moist soil were weighed into

conical flasks, 2M KCl was added to the soil at a ratio of 1:5 moist soil to 2M KCl and the soil slurry was gently refluxed for 4 hours. Once the extracts had cooled, a volume of distilled water, equivalent to the volume of 2M KCl used, was added and the solution was filtered through Whatman 42 filter paper. The N availability was calculated as the difference between the ammonium pool extracted after the extraction and that present in the samples extracted for mineral N.

2.6 Mass spectrometry

The technique of mass spectrometry determines the isotopic compostion of a sample by separating charged ions on the basis of their mass to charge ratio and determining their relative proportions (Robinson and Smith, 1991). In the case of nitrogen analysis this involves the conversion of N in the sample to N_2. The N_2 molecules are ionised and then passed through a magnetic field which separates the N in to three components: $^{14}N_2$, $^{14}N^{15}N$ and $^{15}N_2$, with masses of 28, 29 and 30 respectively. The ^{15}N enrichment of the sample can be determined from the relative amounts of each component, or more commonly from the ratio of the peaks due to $^{14}N_2$ and $^{14}N^{15}N$ (Hauck, 1982).

$$\text{atom\% } ^{15}\text{N enrichment} = \frac{100}{2R + 1}$$

Where R = the ratio of the peaks due to $^{14}N_2$ and $^{14}N^{15}N$.

For more detailed reviews on mass spectrometry see Hauck (1982) and Robinson and Smith (1991).

Plant and soil extracts were analysed for ^{15}N content using a VG Isogas MM622 mass spectrometer linked to a Carlo-Erba 1400 automatic N analyser (Figure 8), which converts nitrogen compounds to N_2 by the Dumas oxidation-reduction procedure (Robinson and Smith, 1991). Subsamples of the prepared plant material (section 2.4) and the crystallised soil extracts (Section 2.5.2) were accurately weighed out into small tin cups and sealed for analysis. In this system at least 100 μg of N is required to give an accurate reading. For young plant samples with N contents greater than 1.5 %, a sample of 10 mg is sufficient. More mature plant samples, including straw, and the soil extracts normally

require subsamples of 20 - 30 mg to ensure that there is sufficient N present for analysis. Reference values are obtained from standards of known N and ^{15}N content in the same batch as the samples. These standards allow the mass spectrometer computer software to calibarate the ion beam currents and current ratios obtained for the samples and calculate values for total percentage N and ^{15}N enrichment.

Checks carried out have shown that the variability between replicate 5 g subsamples of plant material selected for grinding in the agate ball mill, and that between replicate 10 mg portions of the subsequently ground material were very much less than is commonly observed between replicate field plots (Robinson and Smith, 1991). As a further check, any samples whose replicate ^{15}N enrichment values differed by greater than 0.003 atom% were repeated.

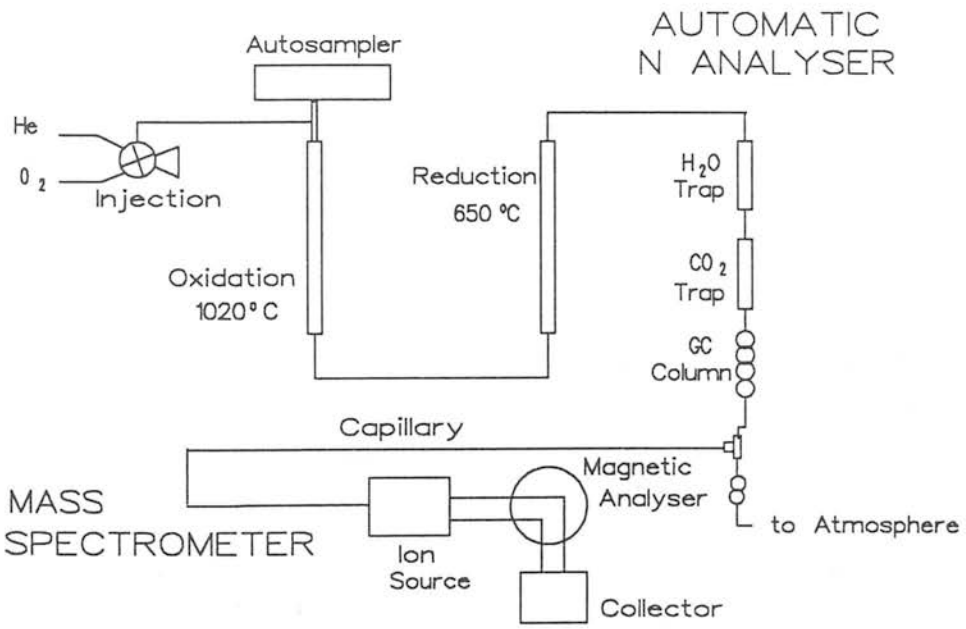


Figure 8 Mass spectrometer, linked to automatic N analyser, as used in the determination of the ^{15}N enrichment of plant and manure materials and soil extracts.

2.7 Statistical methods

The data from each experiment were first tabulated in MINITAB and treatment means with accompanying standard errors calculated. Secondary measurements (eg. crop N uptake, % recovery of manure in the plant) were then calculated for each replicate from the measured data, and these were also tabulated and means and standard errors calculated.

Analyses of variance were carried out for each experiment, where the exact form of the effects and interactions varied according to the experiment. Analysis was carried out either in MINITAB, if the design was simple, or in GENSTAT V, if missing values needed to be incorporated into an analysis of variance. In each case the null hypothesis was tested:
"The differences seen here are no greater than those, which would have been seen, if all the plots had been treated identically" (Dyke, 1974).

This enabled the identification of treatments which had increased N uptake/ yield etc. significantly from the control treatments and perhaps from other treatments applied. These effects were examined with respect to the original hypothesis tested by the experiment, and this original hypothesis was either accepted or rejected.

Where appropriate, further data analysis was carried out in an attempt to explain the effects of the treatments applied, considering the effects on soil properties, any losses which might have occurred etc. Linear regression, multiple linear regression and stepwise linear regression procedures were used where appropriate. Results of experiments were also compared and new hypotheses to be tested in future work were drawn from the results of the integrated series of experiments.

3.0 RESULTS AND DISCUSSION

3.1 N supply from manures

3.1.1 Measuring N supply from manure

Historically, measurement of the amount of N released by a manure, entailed measurement of the accumulation of N in mineral forms released when a manure-soil or manure-sand mixture was incubated aerobically in the absence of plants, compared to that from the soil or sand alone (eg. Potter, 1917). Where manures are labelled with ^{15}N, change in the ^{15}N enrichment of the mineral N pool with time, gives direct evidence that the N accumulating is derived from the manure. A variety of laboratory methods have been used to study the release of N from manures. These have usually involved leached (Epstein *et al.*, 1978; Douglas and Magdoff, 1991) and unleached incubations (Chescheir *et al.*, 1986; Bernal and Kirchmann, 1992; Paper V, p. 5). These methods were compared by Parker and Sommers (1983) and there was found to be little difference between the results obtained. Such methods are useful for comparative purposes, and to increase understanding of processes occurring in the soils, where conditions can be carefully controlled.

Where the N released from unsown pots and the N uptake by rye-grass was compared on soils treated similarly (Paper V, p.17), results were very significantly correlated (r^2 = 51 %, $p < 0.01$). However, the recovery of N by the rye-grass was not as high as might have been predicted by the recovery of N as NO_3^- from the unsown pots. Serna and Pomares (1991) also showed that 0 - 39 % of N was released from manures in the absence of plants, while N recovery in a maize crop was in the range 3.5 - 9.5 %. Huntjens (1971) observed that the net effect of plant roots was to stimulate N immobilisation. However, the presence of plants is more commonly shown to increase the apparent net mineralisation occurring in soil (Rosswall and Paustian, 1984). The conflict between these observations is only superficial, since the N immobilisation due to the microbial growth fuelled by the root exudates may exceed the potential stimulation of net N mineralisation from soil organic matter (Breland and Bakken, 1989).

The effect of roots on N mineralisation can be explained in part by competition between plant roots and soil micro-organisms for mineral N. The success of roots in this competition often reduces immobilisation (Wang and Bakken, 1989). This was seen where straw was added to soils and plant N uptake was not affected (Paper V, Table 3, p. 8), whereas in unsown pots, mineral N levels fell due to immobilisation (Paper V, Fig. 1d, p. 11). Griffiths *et al.* (1993), monitoring two of the field trials (Sections 2.1.1 and 2.1.2), observed no significant effect of plants on the bacterial-feeding nematode population in the presence or absence of manure and suggested that this may indicate that microbial activity is not significantly affected by the presence of plants.

Laboratory incubations are of little value in quantifying N release from manures under field conditions, as such incubations, usually carried out under controlled and ideal conditions, are far removed from the 'real' situation of the field. The absence of plants in such incubations also causes difficulties for extrapolation of results to the field, since the interest in N release from manure is mainly associated with the supply of N for crop growth.

In situ incubations of soil cores in the field have been used for many years to make measurements of net changes in the mineral N pool (eg. Raison *et al.*, 1987; Adams *et al.*, 1989; Redman *et al.*, 1989). However, such methods involve the severing of plant roots, which are able to supply the microbial population with significant quantities of carbon. This can lead to significant immobilisation of N released within the cores (Rees, 1989). Denitrification losses from field incubated soil cores also may rise as the concentration of NO_3^- rises in the presence of increased levels of C (Rees *et al.*, 1993b).

An investigation was carried out into the suitability of a field method to determine gross rates of mineralisation (Paper II), in this case defined as the release of N as NH_4^+ from organic matter. The intention had been to use this method to compare treatments and soils. However, the method seemed to have more problems than benefits for use in the field (Paper II, p. 23-25).

In laboratory studies it was difficult to achieve a uniform distribution of label in the core quickly, so that very large drops were seen in the $^{15}NH_4^+$ pool in the first 24 hours after injection. Only 5 - 35 % of the added $^{15}NH_4^+$ was recovered after 24 hours depending on the soil used (Paper II, Table 3, p. 13). The added $^{15}NH_4^+$ and pre-existing NH_4^+ pools may have been subject to different consumption rates in this period, as slow equilibriation of NH_4^+ occurred, leading to overestimates of the gross rates of mineralisation. Evidence of remineralisation of immobilised ^{15}N was seen after seven days at 14 °C, as the $^{15}NH_4^+$ pool size began to increase. However, some remineralisation was likely to have been occurring in cores earlier, decreasing the rate of decline of ^{15}N enrichment in the NH_4^+ pool and leading to underestimates of the gross rate of mineralisation. Numerical, rather than analytical, solutions are needed to estimate gross mineralisation rates where turnover rates of N through microbial biomass are high (Wessel and Tietema, 1992).

All methods attempting to measure changes in the mineral N pool in the soil are hampered by the high degree of spatial variability seen in the mineral N concentration in the soil, with 40 or more replicates required to make a good estimate of the population mean (Paper II, p. 17). This problem is increased by the addition of manures to the soil, which increase the spatial heterogeneity of the system. In circumstances where N supply from manures (or soil) was to be studied under a growing crop, it was decided that the best approach was to determine plant N uptake, at the same time as monitoring changes in the soil mineral N pool (Rees *et al.*, 1993b). Losses of N by leaching, denitrification and volatilisation were also monitored in the trials where possible, so that the N released from the manures could be estimated.

3.1.2 The importance of the mineral N added

The simplest division of N supply from manures is into that from two pre-existing pools: mineral and organic N (Beauchamp and Paul, 1989). The N in the mineral N pool is the most readily available and has been considered to be as available for plant uptake as that from conventional fertilisers (Beauchamp and Paul, 1989).

Beauchamp (1986) demonstrated that yields of corn were controlled by the amount of NH_4^+ added, regardless of the source of the NH_4^+. Cumulative N uptake by rye-grass in a pot experiment (Paper V, p. 17) and N uptake by spring barley supplied with poultry or farmyard manure (Paper IV, Fig. 3, p. 14) were shown to be highly significantly correlated with the initial NO_3^- concentration of the soil. When the data for all the cereal trials were coalesced (Figure 9), N uptake at harvest was very highly significantly correlated (r^2 = 53.5 % $p < 0.001$) with initial mineral N in the topsoil. When plant recoveries of poultry and farmyard manure (16 and 7.5 % of applied N respectively) were calculated by difference (Paper IV, p. 8), they were found to be only a little greater than the percentage of manure N present as mineral N (14 and 4 % respectively). Mineral N supplied in the manure seems to be the most important fraction of N for crop uptake, at least in the first year.

3.1.3 Factors controlling the availability of the mineral N

The immediate availability of mineral N added is controlled by the amounts of soluble C present. Where levels of soluble C are increased by the addition of a manure, microbial growth is rapidly stimulated (Jensen, 1931) and as a result mineral N is immobilised in the bodies of the developing micro-organisms (Bernal and Kirchmann, 1992; Figure 10). Rapid microbial growth seems to be stimulated by the presence of exogenous nutrients, leached from the residues (Forbes, 1979) but the population rapidly stabilises, when competition and antibiosis become limiting factors (Forbes, 1979).

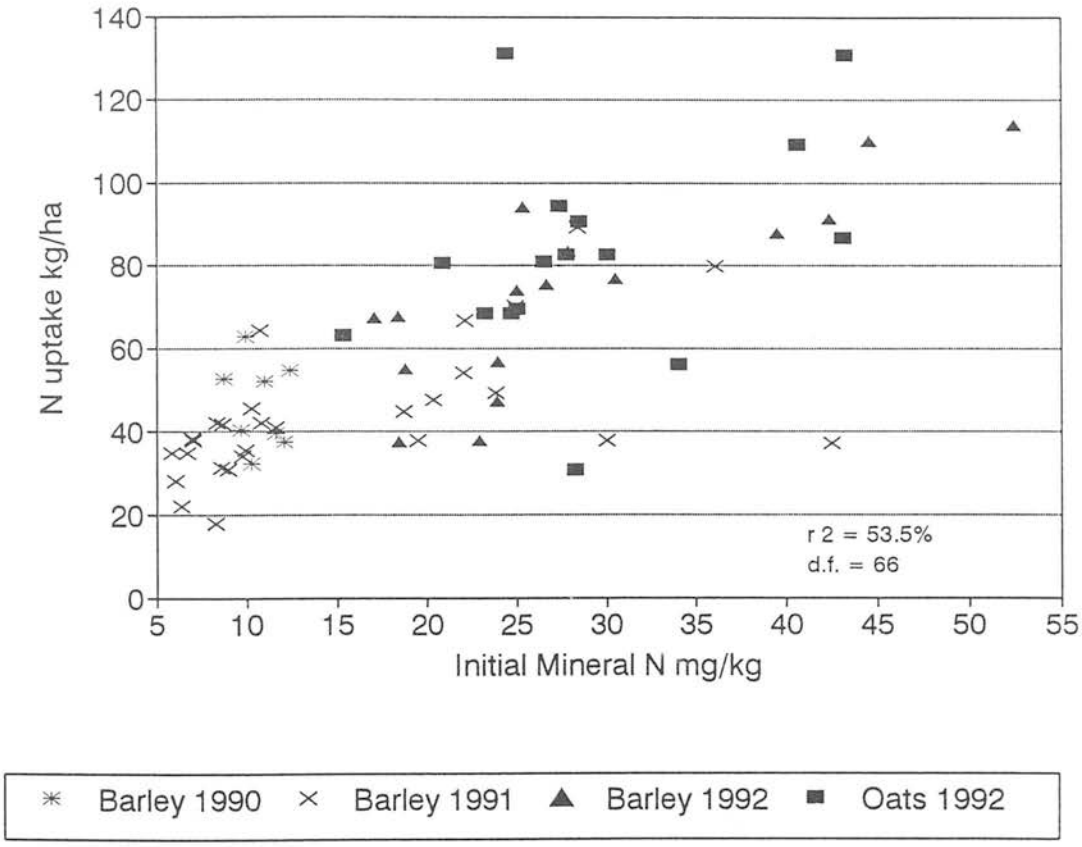


Figure 9 N uptake at harvest by a number of cereal crops (Barley 1990, Jamesfield, farmyard manure and control treatments; Barley 1991, Jamesfield, three rates of poultry manure, farmyard manure and control treatments; Barley 1992, Bush, ploughed out leys and control treatments; Oats 1992, Jamesfield, following different overwinter management practices) plotted against mineral N concentation in the topsoil (0 - 30 cm) between 5 and 10 days after manure had been applied

Net immobilisation results, therefore, where manures with high levels of soluble C, but low levels of mineral N are added to soil (Jenkinson, 1981), and the mineral N pool of the soil is utilised by the micro-organisms to meet their growth requirements (Figure 10b). This was believed to be the case when a rye-grass cover crop was incorporated (Paper IV, p. 17) and mineral N in the soil was significantly reduced relative to the control plots. Where added manures contain soluble C and N, this N is rapidly immobilised because of the spatial proximity to the soluble C (Figure 10c). This was seen for the straw-amended soils (Paper V, p. 12) and was believed to be the case for the poultry manure used in 1991 (Paper IV, p. 11). As a result mineral N added with most manures will not behave in an identical fashion to conventional fertiliser applications (Bernal and Kirchmann, 1992). Reinersten *et al.* (1984) showed that the the amount of biomass produced during the utilisation of the soluble C fraction had a significant influence on the overall rate of decomposition of cereal straw. Soluble C declines rapidly in the first few weeks after addition of manures (Reddy *et al.*, 1980) and any N immobilised may be released as the microbial population declines (Jensen, 1931) or is grazed by predators (Robinson *et al.*, 1989). The mineral N applied to the soil with poultry manure was 100 % efficiently recovered by the spring barley crop (Paper IV, p. 10), which suggests that when the immobilised N is released, it may be almost 100 % available to plants. Manures containing little soluble C will not cause this effect and any mineral N that they contain will be as available as conventional fertiliser additions (Figure 10a).

The availability of mineral N is also controlled by losses. Denitrification fluxes have been linked with the supply of soluble C (Burford and Bremner, 1975; Jarvis *et al.*, 1991), but such losses are usually restricted to anaerobic microsites where the supply of O_2 is restricted (Smith and Arah, 1990). The development of such microsites is dependent on O_2 consumption rates and soil aeration. Denitrification will be significant if soil becomes wet soon after manure application or where large amounts of moisture are added with the manure (Epstein *et al.*, 1978). Significant losses of N as N_2O were seen from soils treated with sewage sludge (Paper V, p. 13) immediately after application, which were linked to denitrification. In such a case, where the soil is close to neutral pH and relatively warm, N_2O would be expected to form only a small part of the gaseous loss of N by denitrification with the remainder of N lost as N_2.

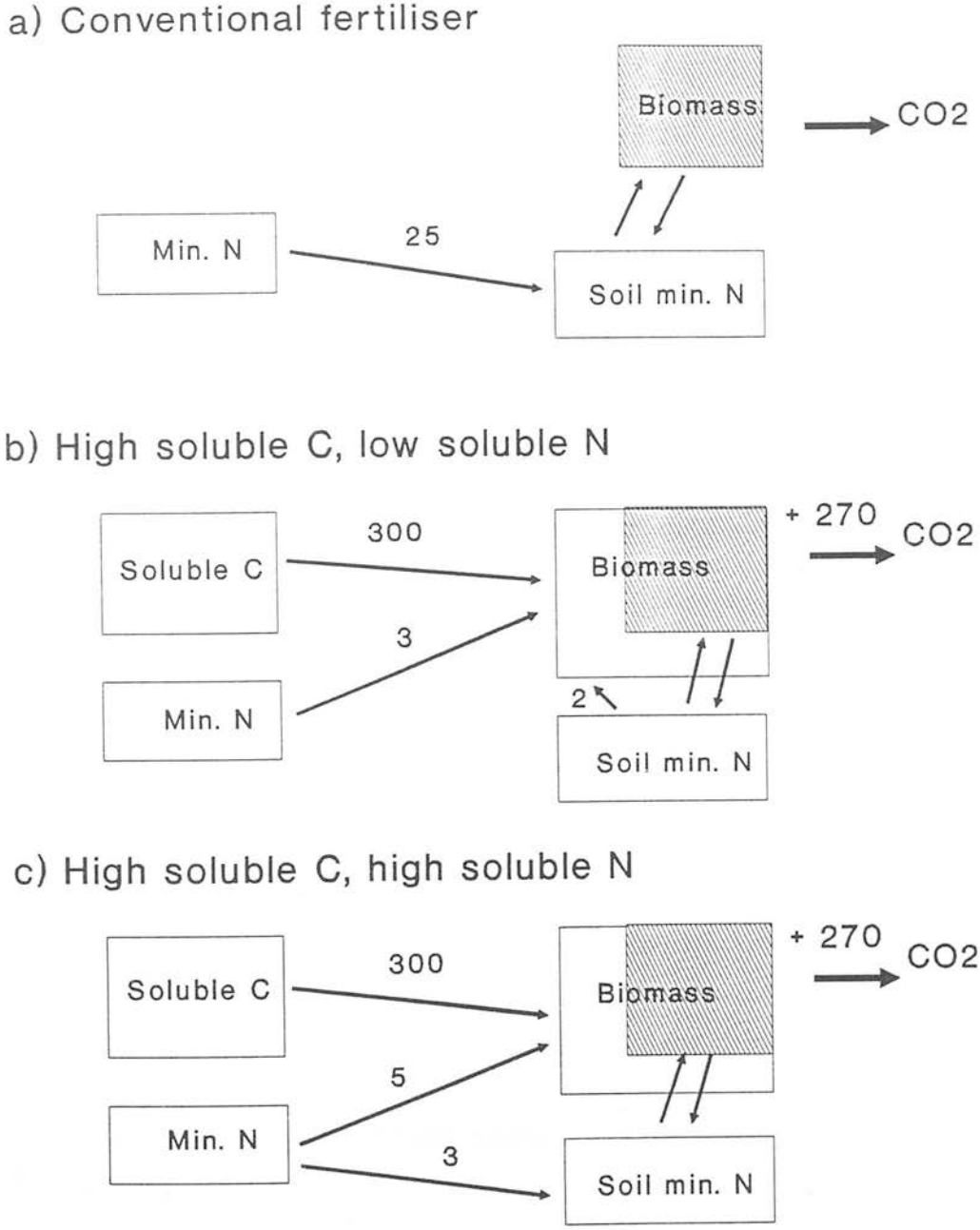


Figure 10 The effect of the amount of soluble C added in manures on the availability of the mineral N in the short term. The soluble C is used in respiration and biosynthesis and N is drawn from the added and soil mineral N to meet the demand. The micro-organisms are assumed to have a C:N ratio of 6 (Robinson *et al.*; 1989) and an assimilation efficiency of 10 % for C. The long term availability of the immobilised N is not known. The pool size of the microbial biomass is represented by the area of the box, the shaded area represents the size of the original biomass pool.

Losses of N as N_2O were also seen from the straw amended soils (Paper IV, p. 13), but it was not clear whether these resulted from nitrification or denitrification. In dry and well aerated soils, a large proportion of the N_2O and NO released to the atmosphere is released during nitrification, with a high NO:N_2O ratio observed for nitrification compared to denitrification (Skiba *et al.*, 1992). Significant losses of NO were seen following the incorporation of a mustard green manure (Bergevoet and Vos, 1993), however, the N_2O flux was not measured.

Losses of N through volatilisation of NH_4^+ may be high where delay occurs between manure spreading and incorporation, since soils for crop production are usually maintained close to neutral pH (Pratt *et al.*, 1976). Volatilisation is only significant where manures contain large concentrations of NH_4^+ or compounds such as ammonium carbonates and uric acids, which release NH_4^+ rapidly. Only very amounts of N were trapped in acid in the pot experiment (Paper V, p. 13). This was probably because the manures were well-mixed with the soils and not applied to the surface (Beauchamp and Paul, 1989).

3.1.4 The role of the added organic N

The amount of N added in organic forms in the manure does not correlate well with the amount of N released from the manure (Paper IV, p. 11; Paper V, p.19). It seemed that after the leaching of the soluble fractions of the manure, the added N could be considered to be as available as N from the soil organic matter. The organic N added in manure to the soil would then be released slowly as the turnover processes of the soil nitrogen cycle continued. The amounts of N released from the added organic matter would be proportional to the amount of N added to each of the pools of N in the soil.

3.1.5 Predicting the supply of N from manure

3.1.5.1 Indices

Two indices of N availability, anaerobic incubation and an extraction with hot KCl were used on the manure treated soils in a number of the trials to ascertain whether these indices were useful in predicting N availability from soil-manure mixtures.

Soils sampled after spring barley had been drilled, approximately a fortnight after manure application, from the spring barley trial at Jamesfield in 1991 (Paper IV, p. 7) showed an increase in the mineral N pool of the soil well correlated with the NH_4^+ applied in the manure (r^2 = 63 % $p < 0.01$). The coefficient of variability of the measurement increased markedly on plots where manure had been applied (Table 5), and increased further where higher rates of NH_4^+ had been applied, as the difficulty of taking a representative soil sample in the plots increased. Such heteroscedastic data cannot be analysed using conventional analysis of variance techniques, since the assumption of homogeneity of variance does not hold (Mead and Curnow, 1983). However, this effect reduced with time so that conventional analysis of variance could be used on the plant N uptake data.

N availability determined by anaerobic incubation was increased slightly, but not significantly at the higher rates of manure application, although a marked increase in the variability of the measurement was seen on plots to which manure had been applied.The N availability determined by extraction with hot KCl was not significantly different between treatments and the highest mean value was given by the control plots (Table 5). The variability of the measurement was also not changed on the manure treated plots. The highest mean N availability determined by hot KCl extraction was also measured on the untreated soils in the pot experiment, with neither N availability determined by anaerobic incubation nor extraction with hot KCl showing significant differences between treatments (Paper V, p. 19).

N release from the manures was not correlated significantly with the N availability determined by extraction with hot KCl in any of the experiments carried out. However, N availability predicted by anaerobic incubation was significantly correlated with N release from manures in the pot experiment (r^2 = 21 % $p < 0.05$) and with N uptake in the field trial (r^2 = 33 % $p < 0.05$).

Table 5 Mineral N, N availability determined by anaerobic incubation and N availability determined by extraction with hot KCl (all expressed as mg N kg^{-1} soil) on topsoil (0 -30 cm) sampled from the plots of the Spring Barley trial at Jamesfield in 1991. Means, standard errors of the means (S.E.) and coefficients of variability (CV) given for each treatment, which are listed in order of increasing application of NH_4^+ in the manure.

	Mineral N	mg N kg^{-1}	
Treatment	Mean	S.E.	CV %
Control	8.08	0.33	8
Low rate FYM	8.50	1.18	27
Low rate PM	8.80	0.72	16
Medium rate FYM	11.81	2.64	45
High rate FYM	20.35	4.00	39
Medium rate PM	23.18	4.69	40
High rate PM	27.65	4.83	35
	Anaerobic incubation mg N kg^{-1}		
Treatment	Mean	S.E.	CV %
Control	26.85	0.40	3
Low rate FYM	24.13	1.77	15
Low rate PM	25.79	3.26	25
Medium rate FYM	24.19	4.30	36
High rate FYM	31.69	8.04	51
Medium rate PM	35.45	2.43	14
High rate PM	35.95	5.21	29
	Hot KCl extraction mg N kg^{-1}		
Treatment	Mean	S.E.	CV %
Control	19.32	2.57	27
Low rate FYM	16.36	0.98	12
Low rate PM	16.66	1.04	13
Medium rate FYM	18.10	2.16	24
High rate FYM	16.37	3.15	38
Medium rate PM	15.34	1.92	25
High rate PM	16.16	3.53	44

Detailed studies were carried out to ascertain whether the N extracted by these availability indices was derived from the microbial biomass (Paper III). The N extracted after anaerobic incubation was found to derive from the soil microbial biomass or a pool closely related to it. This hypothesis was supported by the earlier work of Myrold (1989). However, the N extracted with hot KCl appeared to have been extracted from a different pool or pools, than were the source of the N extracted after anaerobic incubation or the biomass N (Paper III, p. 16). It is possible that this technique extracted N dominantly from the stabilised and active non-biomass fractions of organic matter, since it failed to predict changes in N availability that result from recent additions of organic manures (Table 5). This was also reported by Fox and Piekielek (1984). The success of this index for predicting soil N uptake for malting barley (McTaggart and Smith, 1993) may lie in the relatively stable management of index 0 sites (East of Scotland College of Agriculture (ESCA), 1985). Where sites receive the same residue input annually, the differences in the amount of N released by mineralisation are attributable to the differences in the basal rate of mineralisation from the stabilised and active non-biomass pools of N in soil.

Although much work has been done in trying to find a chemical index for N availability from soil-manure systems (Parker and Sommers, 1983; Chescheir *et al.*, 1986; Serna and Pomares, 1991), there has only been limited success. Many indices, including anaerobic incubation used here, are able to rank manures for N availability. However, perhaps not surprisingly, no index has been found which is able to describe the complex processes leading to N supply from manures.

3.1.5.2 Models

The N release from manures is often modelled by a statistical-empirical approach, where data is analysed and multiple regressions and curve fitting used (King, 1984; Barbarika *et al.*, 1985). Such approaches often find regression equations which fit the measured data very well, but which fail to describe the data generated by other workers using different soils or manures (King, 1984). This approach was used in an attempt to model the N released from a wide range of manures in a pot experiment (Paper V, p. 17). However, models were never fitted with a correlation coefficient of greater than 75 %, which though highly significant was felt to be inadequate for use in routine prediction of N

release. Stepwise regression identified initial NO_3^- levels in the soil-manure mixtures as the most important factor for predicting either cumulative N uptake by rye-grass or NO_3^- accumulation in the unsown pots. The manure C:N ratio, N concentration of the manure and a variable formed by the mutiplication of initial NO_3^- with manure C:N entered the regression at lower levels.

Mechanistic modelling of the release of N from manures is difficult, since the processes involved are not fully understood. Models where conceptual divisions of the added manure into a number of differently available fractions (Sluijsmans and Kolenbrander, 1975; Bhat *et al.*, 1980) demand coefficients for N supply from these fractions that can only be estimates and best guesses, and may be fitted to the data set by numerical simulation. This can lead to both complex and simple models explaining the data set adequately, but where the pools of organic matter are not measurable neither is adequately verifiable.

Manure N added to soils is composed of a broad spectrum of N containing compounds, which are acted on by a broad spectrum of micro-organisms occurring in the soil, to release nutrients to meet their own growth requirements. Only where N is supplied in quantities surplus to the requirements of the micro-organisms will it be released in forms which can be extracted from soils (NH_4^+ and NO_3^-), though plant roots are able to compete with micro-organisms for any mineral N present. De Willingen (1991) reviewing a large number of models describing N dynamics in the soil-plant system, concluded that our ability to model the whole system was handicapped by our failure to understand and model the biological processes occurring in the soil. While the qualitative controls on manure N release are already largely known and relatively easily verified, the complex system to which manure N is added, and which its addition further complicates, makes quantitative prediction of N release from manures very difficult.

3.2. The use of ^{15}N

3.2.1 Uniform and non-uniform labelling of manures

The production of uniformly labelled plant residues for use in studying N release is not difficult. Plants are grown hydroponically from seed with ^{15}N labelled nutrient solutions used (Azam *et al.*, 1985; Janzen and Radder, 1989; Rees *et al.*, 1993a). The cabbage and pea residues used in the pot experiment (Paper V, p. 3) were labelled in this way. Crop residues from microplots treated with ^{15}N fertiliser at the beginning of crop growth are often also assumed to produce plants with approximately uniform labelling of ^{15}N (Wagger *et al.*, 1985; Ocio *et al.*, 1991) The straw used in the pot experiment (Paper V, p. 3) was obtained in this way. The assumption that plants labelled in this way are uniformly labelled was also made in calculating the efficiency of use of N from cover crops grown overwinter, where the profile residual N was labelled (Paper IV, Trial 2, p. 5). The process may be time consuming eg. in growing up a labelled turf from seed and in such cases the labelling procedure might be altered by the cutting of turf from the field and growing this for a number of weeks, applying ^{15}N salt regularly to allow it to be incorporated into the plant material (procedure as applied to turf, Paper V, p. 3). Although such a procedure will almost certainly not produce a uniformly labelled residue, with younger parts of the turf more highly enriched, where the turf is mixed well with the soil the assumption of uniform labelling can be used without too much error.

The production of uniformly labelled animal manures is difficult, if not almost impossible. Large quantities of ^{15}N enriched feedstuff would be needed, and even where this was fed the enrichment of the urine and dung would be different, depending on the balance of digestible to indigestible proteins in the feed. Uniformly labelled excrement is easiest to produce and collect from small animals such as poultry. Poultry were fed with ^{15}N labelled barley grain and uniformly labelled excrement collected (Kirchmann, 1990).

However, animal manures are rarely simply the excrement of the animals, but are mixed with the bedding material, with both occurring in a semi-decomposed state. If full decomposition was allowed to occur before the material was used the ^{15}N enrichment would be approaching uniformity throughout the pools. However, this is rarely desirable for studies of N release from manures. Studies have been carried out (Kirchmann, 1989; Kirchmann, 1990) using uniformly labelled poultry excrement mixed with varying amounts of straw and composted under aerobic or anaerobic conditions, to study the release of N from the excrement after such pretreatments.

Heap-composted farmyard manure was incubated with small amounts of high enrichment $(^{15}NH_4)_2SO_4$. A rapid decline in the ^{15}N enrichment of the mineral N pool of the manure during the first three weeks of the incubation is observed (Table 6), with the ^{15}N enrichment of the organic N pool of the manure increased significantly above background to approximately 1 atom % ^{15}N enrichment. It was thought that the ^{15}N became incorporated into the microbial biomass active in the manure and thence into the organic N pool of the manure. When non-uniformly labelled manures are used the calculation of percentage plant N uptake derived from the manure is slightly different from the static isotope dilution equation (see Section 1.6.1).

Table 6 The ^{15}N enrichment of the mineral N pool of two replicates of a heap-composted farmyard manure (A and B) during incubation with 0.64 g 99 atom % $(^{15}NH_4)_2SO_4$ kg^{-1} manure (wet weight), which had been applied to the manure with a fine-mist spray. Means given with S.E. of the mean in brackets. (Rees, unpublished data).

	atom % ^{15}N	
Length of incubation	A	B
1 week	28.815 (7.565)	40.135 (5.135)
3 weeks	3.815 (0.925)	12.660 (3.080)
4 weeks	9.150 (4.950)	7.275 (4.215)
8 weeks	4.200 (2.093)	13.643 (4.119)

Where the assumption can be made that all the pools of N in the manure have the same ^{15}N enrichment or are equally available for plant uptake, then the standard static isotope dilution equation can be used, with the substitution of the average ^{15}N enrichment of the manure for the fertiliser ^{15}N enrichment.

$$NDFM = \frac{e_p - e_s}{e_m - e_s} \times 100$$

Where NDFM = Percentage of plant N uptake derived from manure

e_p = ^{15}N enrichment of plant sample

e_s = ^{15}N enrichment of soil N (background)

e_m = ^{15}N enrichment of whole manure

However, the assumptions made for this model to be used do not hold where the manure is not uniformly labelled. As a result a simple model was developed, based on static isotope dilution theory, considering two pools of N applied in manure: a small pool of mineral N whose size (N_i) and ^{15}N abundance (e_i) are measured and a larger pool of organic N with a different but known ^{15}N abundance (e_o). The ^{15}N abundance of the manure organic N pool (e_o) can be calculated where the ^{15}N enrichment of whole manure (e_m) is corrected using the size (N_i) and ^{15}N abundance (e_i) of the small mineral N pool. Nitrogen is also supplied to the crop from a pool of soil N with background ^{15}N abundance (e_s). It is assumed that 100 % of the mineral N from the manure is taken up by the plant before any of the organic N from the manure is used. This would lead to underestimates of the value of the organic N pool, where the manure N is used less than 100 % efficiently. A calculation is first made to ascertain whether all the ^{15}N taken up by the plant at any harvest (N_p) could have been derived from the mineral N pool of the manure (N_i).

If:

$$\frac{e_p - e_s}{e_i - e_s} \times N_p < N_i$$

Then

$$NDFM = \frac{e_p - e_s}{e_i - e_s} \times 100$$

More commonly:

$$\frac{e_p - e_s}{e_i - e_s} \times N_p > N_i$$

And

$$NDFM = \frac{e_p N_p - e_s N_p - (e_i - e_o) N_i}{(e_o - e_s) N_p} \times 100$$

Where the NDFM is calculated from the N uptake data of the spring barley and potato trials carried out in 1990 (Paper I, p. 4), using both the average ^{15}N abundance of the manure and the simple model outlined above, higher recoveries are predicted by the use of the average ^{15}N abundance of the manure (Table 7). This occured because the added mineral N was more highly enriched than the average ^{15}N abundance predicted, and when this was accounted for, smaller amounts of N were needed to meet the measured ^{15}N enrichment of the plant material. The difference between these two estimates reduced as growth occurred, since the uptake from the small mineral N pool of the manure became proportionally less of the total crop N uptake as growth occurred.

Table 7 The mean nitrogen contents (kg ha^{-1}), with standard errors of the means, and estimates of the percentage of plant N uptake supplied by the manure derived by the difference method (difference) and two static isotope dilution equations. One model used the average ^{15}N abundance of the manure (average) and the other divided the manure into two pools, using a simple model outlined in the text (model). The data comes from the trials using heap composted farmyard manure for spring barley and potatoes carried out in 1990.

	Total N uptake kg ha^{-1}	% of plant uptake derived from manure		
		Difference	Average	Model
BARLEY				
17th May	10.4 (1.2)	-ve	47	38
4th June	25.7 (13.0)	24	28	26
5th July	49.0 (12.5)	25	20	18
28th August	51.9 (8.6)	21	13	11
POTATO HAULMS				
4th June	4.0 (0.48)	30	22	18
19th June	40.2 (4.85)	34	30	29
10th July	54.5 (6.50)	17	16	15

Manures are also non-uniformly labelled, where studies of the dynamics of the mineral N pool of the manure are made and inorganic ^{15}N salts are added to the manure as the manure is applied to label the mineral N pool (Paper I, Trial 1, p. 4).

3.2.2 Border effects on ^{15}N microplots

Where physical barriers are not used to separate the microplot from the surrounding soil, plants grown inside the microplot area are able to use unlabelled N from outside the microplots and plants grown outside the microplot may become labelled with ^{15}N. Although barriers eliminate the possiblity of such lateral movement of ^{15}N occurring, they may also introduce artifacts caused by disruption of the soil or prevention of root growth and distribution (Sanchez *et al.*, 1987).

The ^{15}N enrichment of barley head and straw was determined within the microplots of the high rate poultry manure treatment in the field trial with spring barley in 1991 (Section 2.1.3). The pattern of ^{15}N enrichments seen within the microplot was similar for both head and straw (Figure 11). Samples taken in the 0.5 m just outside the microplot to the north (Row 1) had average ^{15}N enrichments of 0.536 atom % in the straw. Since the label was applied as a labelling spray to the manure, which was then incorporated, the increased ^{15}N enrichments in Row 4 (Figure 11) reflected the movement of the manure in the direction of tillage, and were not surprising. Despite the effect of the direction of tillage, samples taken in the centre of the microplots (3D and 3E) seemed to be representative. The minimum plot size recommended by Follett *et al.* (1991) for winter wheat (1.5 x 1.5 m) would leave only an area at the centre of the microplots of 0.5 x 0.5 m, where representative cuts could be made. A slightly larger microplot than this is required, where microplot harvests are to be made sequentially throughout the season, and the microplots used in this experiment of 2 x 1.5 m would seem to be adequate to allow such a sampling regime.

a) Head

	C	D	E	F
1				
2	1.571	1.660	1.649	1.805
3	2.067	3.829	2.766	1.777
4	2.560	2.767	3.027	1.776

Direction of cultivation

b) Straw

	C	D	E	F
1	0.461	0.756	0.543	0.573
2	1.617	1.720	1.771	1.953
3	2.127	3.983	2.891	1.863
4	2.748	2.961	3.205	1.865

Direction of cultivation

Figure 11 ^{15}N enrichment of samples of a) head and b) straw of spring barley sampled, where a high rate of poultry manure had been applied, in 1991. The microplot consists of Rows 2, 3 and 4, where each square represents a sampling area of 0.5 x 0.5 m, from which the central plants were removed. Each value is the mean of 4 similar samplings taken in the four microplots under the same treatment.

3.3 N uptake from manure by crops

Where ^{15}N is used in conjunction with control plots there are two ways of determining the amount of N recovered in the plant from the manure (section 1.6.1). In the pot experiment (Paper V, p. 14) differences in the percentage recovery of N applied in the manures resulting from the treatments were greater than those related to the method of determination of that recovery. In general, the recovery of N determined by the difference method gave slightly lower values than the percentage recovery determined using a static dilution equation.

An added nitrogen interaction (ANI) may be seen when ^{15}N labelled fertilisers are used. This is defined as any increase or decrease in the quantity of soil-derived N taken up by the plant caused by the added N (Jenkinson *et al.*, 1985). In the pot experiment, soil N uptake, calculated as the difference between total N uptake and the N derived from the labelled pool, in the manured soils was generally lower than that measured directly from the control soils, a negative ANI (Paper V, p. 17). With mineral fertilisers positive ANIs are more commonly seen (Hart *et al.*, 1986). Such effects may be caused by pool substitution, where labelled N takes the place of unlabelled N that would otherwise have been used from that pool (Jenkinson *et al.*, 1985). In the pot experiment, the observed negative ANI increased as the C:N ratio of the manure increased (Paper V, p. 17). This is consistent with the results of Powlson *et al.* (1985) and Rees *et al.* (1993a), and is probably due to the increased immobilisation of soil mineral N by the soil microorganisms where the C supply is abundant. The reverse would be expected to occur with increased mineral fertiliser additions (Hart *et al.*, 1986).

In the field trials, the two methods for determining the percentage recovery of N applied were only compared directly in the trials with spring barley and potatoes in 1990 and in the cover crops trial in 1992. Except in the case of the spring barley in 1990, higher recoveries were estimated using the static isotope dilution equation (Table 8). The higher estimated recovery by the difference method for spring barley in 1990 may indicate that recovery from unlabelled fractions of the manure was important for the barley.

However, although estimates of recovery by difference are similar for farmyard manure at the medium rate in 1991 (Table 8) and in the pot experiment (14 % on average), estimated recoveries determined using the static isotope dilution equation were higher in the pot experiment (19.6 % on average), and the odd result for the field trial in 1990 may have been caused by non-representative sampling of plots and within plot variability.

Immobilisation occurred following ploughing in of the rye-grass and rye/clover cover crops and all the N uptake from the manure measured by the static isotope dilution equation (Table 8), therefore occurred by substitution of manure N for soil N during soil turnover processes. Similar recoveries were seen for incorporated straw (Powlson *et al.*, 1985) with negative recoveries determined by difference and N uptake from the straw measured by the static isotope dilution equation of 12 % of the added N. Plant roots are also able to compete with micro-organisms for mineral N (Wang and Bakken, 1989), and therefore reduce immobilisation. The recovery of N estimated from the static isotope dilution equation for the grass/clover turf in the pot experiment (18 % on average) was similar to that determined for the cover crops in the field, but net mineralisation was seen and the recovery estimated by difference was 10 % on average.

The comparison between the barley and potato crops in 1990 demonstrated than potato crops were able to use more of the N released by manure than barley (Table 8), since mineralisation continues to occur after barley has ceased actively taking N up (Paper I, p. 9). This extra uptake of manure N seemed to occur by substitution for soil N, as little yield benefit seemed was gained over the control plots. Differences between treatments in both the trials were small probably due to the high residual fertility of the control plots after an autumn application of manure.

Table 8 Percentage recovery of N applied in manure determined in field trials 1990-1992 by two methods: by comparison with the control plot (difference) and using the ^{15}N enrichment of the plant samples and a static isotope dilution equation (^{15}N).

Trial and treatment	% recovery of N applied in manure	
	Difference	^{15}N
Barley 1990		
Farmyard manure	11	5
Potatoes 1990		
Farmyard manure	8	23
Barley 1991		
Low rate poultry manure	18	89[a]
Medium rate poultry manure	21	113[a]
High rate poultry manure	10	104[a]
Low rate farmyard manure	5	88[a]
Medium rate farmyard manure	12	26[a]
High rate farmyard manure	5	32[a]
Oats 1992		
Stubble fallow	0	15
Rye-grass cover crop	-ve	26
Rye/clover cover crop	-ve	16

a For this trial the mineral N pool of the manure was labelled, and so these recovery values represent the recovery of the mineral N pool of the manure in the crop.

3.3.3 Factors controlling the efficiency of recovery of N by crops

The maximum efficiency of use of manure N by crops is controlled by their growing season and management. Crops with short and early growing seasons are handicapped by the slow rates of mineralisation in the early part of the season. Under Dutch conditions, Sluijsmans and Kolenbrander (1977) suggested that cereals were able to take up 50 % of the N released from manure at the utmost, potatoes and beets about 70 %, whereas permanent grassland may utilise 90 % of the N released. These efficiencies are largely controlled by the sowing and harvesting date of crops.

For late-sown crops prevention of losses of N accumulating in the profile before planting due to mineralisation can be important, and this may be why pre-planting mulching techniques conferred unexpected yield benefits on crops (Appendix I). However, management practices which increase efficiency of use of manure N by crops will only enter farming practice where they bring economic benefit to the farmer. Practices, such as pre-planting mulches, which may be shown to be suitable for high value late-sown vegetable crops, will not necessarily be transferable to crops of lower value and larger acreages (eg. potatoes), even where planting dates are similar. Farmers are also unlikely to be willing to take a cut in yield following the use of overwinter cover crops (Paper IV, p. 17), despite the advantages in minimising N lost by leaching. In fact, increases in yield may be necessary to convince farmers that the extra expenditure (seed and labour costs) are in fact worthwhile. In nitrate sensitive areas (Archer, 1992), yield reducing practices may be imposed in order to minimise the risk of nitrate leaching. However, in such circumstances compensation payments will be made to the farmer.

3.4 Leaching of NO_3^- after harvest

Leaching losses can be high overwinter where a green manure crop is incorporated in the spring (Bremer and van Kessel, 1992; Rees *et al.*, 1993a). Leaching losses were only measured directly following the ploughing out of leys for cereal cropping, but the effectiveness of cover crops were also monitored. The importance of maintaining a soil cover overwinter was emphasised, though an unploughed stubble and weeds acted as effectively as a sown rye grass cover crop in conserving residual profile mineral N overwinter (Paper IV, p. 15). On average, the plant material harvested from the plots with an overwinter cover recovered 24 % of the residual profile mineral N following spring barley, while the bare fallow only retained 2 % of the residual mineral N (Paper IV, p. 15). Leaching losses from ploughed out leys were lower than expected, due to strong regrowth beneath the spring barley crop, which provided a substantial sink for nitrate in the autumn (Paper IV, p. 15). Leachate concentrations were also below the EEC maximum acceptable concentration of 50 mg NO_3 per litre (Tunney, 1992) throughout the sampling period.

3.5 Other nutrients supplied by manures

The addition of manures and residues to soils will also alter the status of other soil nutrients. In the pot experiment treatment differences were observed for P, K and pH in soils sampled after the 16 week incubation (Table 9). The differences between the soils were very highly significant ($p < 0.001$). The Kettle soil had a higher pH, higher phosphorus availability and lower potassium and magnesium availability than the Quixwood soil. All the treatments increased the exttractable phosphorus levels slightly, with very highly significant increases ($p < 0.001$) after straw, farmyard manure, swine manure, slurry and poultry manure applications. The addition of straw, farmyard manure, swine manure and slurry also significantly increased available potassium ($p < 0.05$). pH was significantly increased when pea residues, farmyard manure and sewage sludge were added to the soils ($p < 0.05$), while the addition of grass-clover turf significantly reduced pH ($p < 0.05$).

Table 9 Available phosphorus, potassium and pH levels determined on two soils (Quixwood and Kettle) mixed with different manures and sown to rye-grass after 16 weeks growth of rye-grass. Means given for P and K, standard deviations indicated in brackets.

Treatment	P mg l^{-1} soil	K mg l^{-1} soil	pH
Quixwood soil			
Control	11 (1.3)	179 (20.6)	6.6
Pea	14 (0.0)	261 (5.7)	7.0
Turf	11 (0.0)	175 (7.8)	6.5
Cabbage	11 (1.3)	202 (18.2)	6.7
Straw	15 (2.6)	382 (35.0)	6.8
Farmyard manure	15 (0.0)	278 (26.5)	6.9
Swine manure	13 (0.6)	281 (28.9)	7.0
Sewage sludge	13 (0.0)	195 (11.3)	6.6
Slurry	15 (1.5)	286 (32.8)	6.7
Poultry manure	16 (1.5)	232 (30.7)	6.6
Kettle soil			
Control	17 (1.7)	112 (10.0)	7.0
Pea	18 (1.0)	126 (19.0)	7.0
Turf	22 (1.2)	122 (6.1)	6.8
Straw	27 (0.6)	334 (20.1)	6.9
Farmyard manure	25 (2.1)	176 (14.8)	7.0
Swine manure	31 (1.5)	242 (25.7)	6.9
Sewage sludge	21 (0.6)	103 (10.5)	7.0
Slurry	29 (1.7)	214 (12.3)	7.0
Poultry manure	28 (1.0)	113 (8.6)	7.0

4.0 APPRAISAL AND SUGGESTIONS FOR FUTURE WORK

Laboratory methods to study the release of N from manures are well developed and used carefuly are able to increase our understanding of the processes occurring in soils, which control N release. Isotope dilution techniques also work well, where it they are used in the laboratory on well-mixed soils (Bjarnason, 1988; Wessel and Tietema, 1992). Field methods are far less developed and are complicated by the presence of plants or excised roots, as well as fluctuating and uncontrollable environmental conditions (Rees *et al.*, 1993b). The use of isotope dilution techniques in the field is complicated by the slow attainment of equilibrium of added $^{15}NH_4^+$ with native NH_4^+ in structured soils, so that remineralisation effects also need to be taken into account in the calculation of gross rates of mineralisation (NH_4^+ release). Better injection techniques and greater understanding of how much the soil internal N cycle is limited by diffusion are needed before such techniques can give meaningful answers in the field. The release of N from manures is currently best estimated indirectly, with reference to plant uptake and losses of N. The use of ^{15}N enables the original source of the N to be known, although such N may have become involved in large number of soil turnover processes before it is taken up by a plant or lost from the system by leaching, volatilisation or denitrification.

The release of N from manures in the first year has been shown to be largely due to the supply of N added in the manure in mineral forms. However, the availability of this pool is affected by the complementary addition of soluble carbon to the soil, a system which is dominantly carbon and not N limited. The supply of soluble carbon drives the expansion of the population of micro-organisms (van Veen *et al.*, 1984), which utilise any available N for tissue production until C, N or some other nutrient becomes limiting. This rapid immobilisation of the added mineral N into the biomass N pool means that the mineral N added with manure does not behave in a similar fashion to additions of conventional mineral fertilisers. Immobilisation of fertiliser N was shown to occur only very slowly, unless soluble C compounds were added to the soil (Recous *et al.*, 1990). The immobilisation of mineral N immediately after the addition of manure seems to be controlled by the size of the pool of soluble C added.

The use of ^{15}N to label the mineral N pool of the manure, when it is added to soil, is crucial in determining the fate of the added mineral N in the short and long-term, in the presence and absence of soluble C. The field trial in 1991 (Paper IV, p. 10) suggested that N immobilised as a result of the expansion of the microbial population was recovered almost completely by a spring barley crop. Other workers (Flowers and Arnold, 1983; Bernal and Kirchmann, 1992) have suggested that such immobilised N would not be released. The careful use of ^{15}N to label the mineral N pool of the manure, in combination with different levels of soluble C, would enable us to investigate the controlling factors of this immobilisation and enable us to follow the fate of the added ^{15}N through the turnover processes of the soil. It might also be advantageous to use labelled C in conjunction with ^{15}N to investigate the efficiency with which the biomass is able to utilise the soluble C from the manure and to study how the C and N transformations interact. The release of immobilised N is probably controlled by the rates of biomass turnover and the activity of their predators (Robinson *et al.*, 1989). Therefore observations of the size and activity of predatory populations (eg. nematodes, flagellates and amoebae) and biomass turnover rates, subsequent to manure additions, would also help to increase understanding of how mineral N immobilised from manures might be released.

The role of the organic N added in manures is unclear, as a result of the trials carried out during this project. It seems that in most cases the availability of this organic N falls within the range of the pre-existing organic matter, and it is only where manures are added over a long time period that their residual effect becomes important by increasing the size of the pool of organic N in the soil. The use of non-uniformly labelled manures in conjunction with manures, where only the mineral N pool is labelled, may help to elucidate the complementary roles of the mineral and organic N pools in the manure for crop supply in the first year. The efficiency of use of the added mineral N pool can be determined where only this pool is labelled. Careful use of static dilution theory then enables the contribution of non-uniformly labelled manures to be divided into that from the mineral and organic N pools, where their size, ^{15}N enrichment and the efficiency of use of the mineral N pool is known. The long-term increase in soil organic matter where plots are treated with farmyard manure is well known (Jenkinson and Raynor, 1977) but it is difficult to determine whether this added organic matter is of higher quality than native organic matter.

The addition of manure to soils was clearly shown to increase the heterogeneity of the already heterogeneous soil system, with heteroskedasticity exhibited in the mineral N content of the plots in the 1991 field trial (Section 3.1.5.1). Great care needs to be taken therefore before conventional analysis of variance techniques are applied to data where different rates of manures and unmanured plots are compared, as for such techniques homogeneity of variance needs to be assumed (Mead and Curnow, 1983).

The efficiency with which crops are able to recover the N added in manures is highly dependent both on the crop and the manure, as well as management and climatic factors. The approach of Sluijsmans and Kolenbrander (1977) in assigning efficiency indices to crops based on the length of their period of active N uptake, could be very useful. However, in the trial carried out in 1990 there was no evidence of any yield benefit of a greater manure recovery to the crops. Management practices will also modify these efficency indices. This approach appears to be potentially useful for incorporation into a simple semi-quantitiative model. However, more validation would need to be carried out, using literature-derived data as well as more directly comparative field trials.

The selection of a manurial strategy for an organic farm which maximises crop yields, while estimating the potential N losses, could be achieved by the use of a semi-quantitative model. A possible structure for such a model is laid out in Figure 12, although no testing has been carried out to test the validity of such a structure or to assess an appropriate technique to model such a system. An expert system approach may be possible, including database tables or simple models outside the system to which the system is able to refer (Ignizio, 1991).

The structure (Figure 12) shows the flow of information through the suggested model structure, with user inputs compared to databases and combined in sub-routines to produce new inputs for later routines. The final output of the model is the allocation of a limited amount of organic manures to supply the N requirements of the farm crops, maximising the crop value to the farmer and predicting the potential losses of N from the system. Such an output would allow a farmer to compare different cropping systems in terms of farm output and pollution risk.

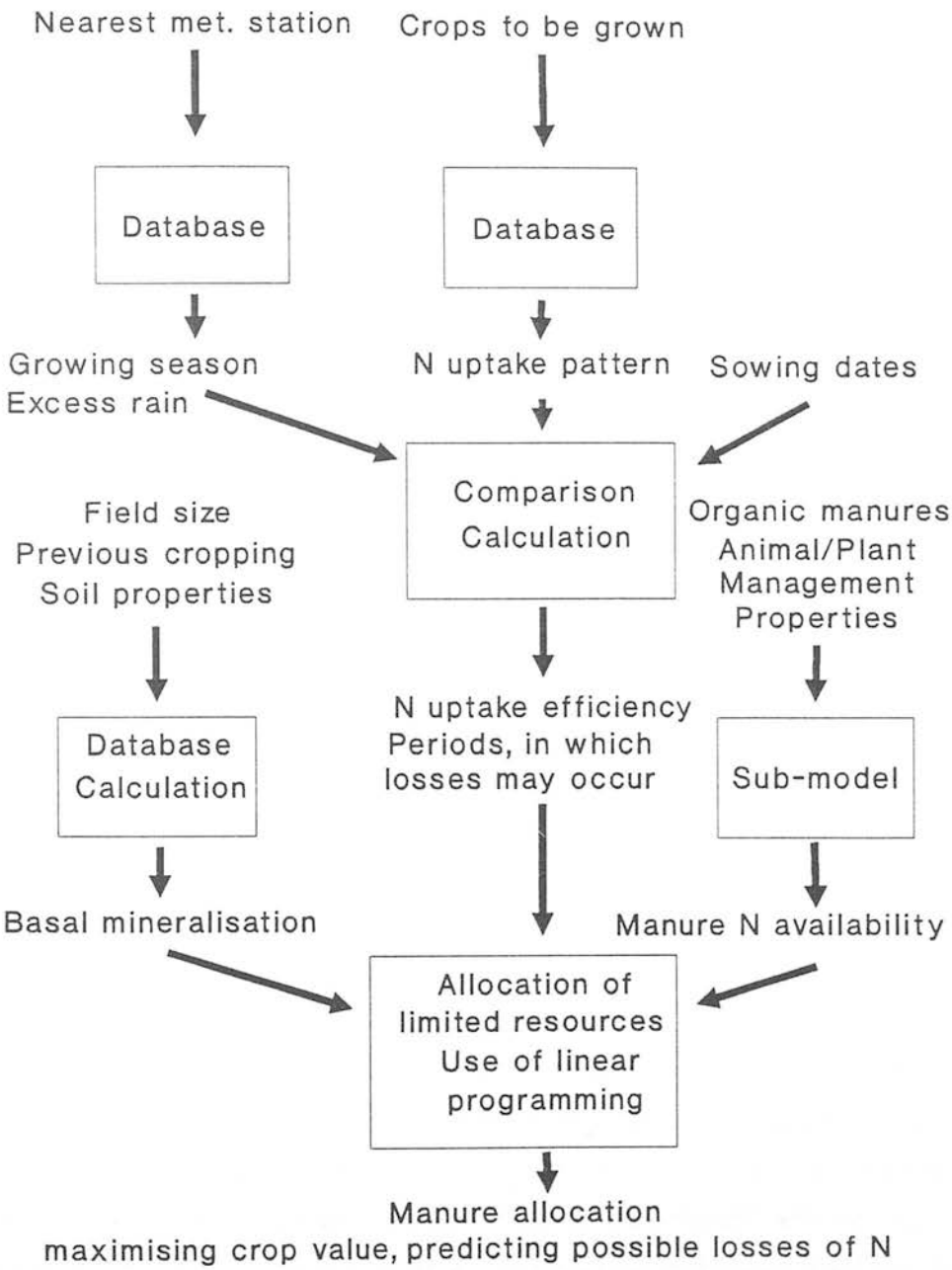


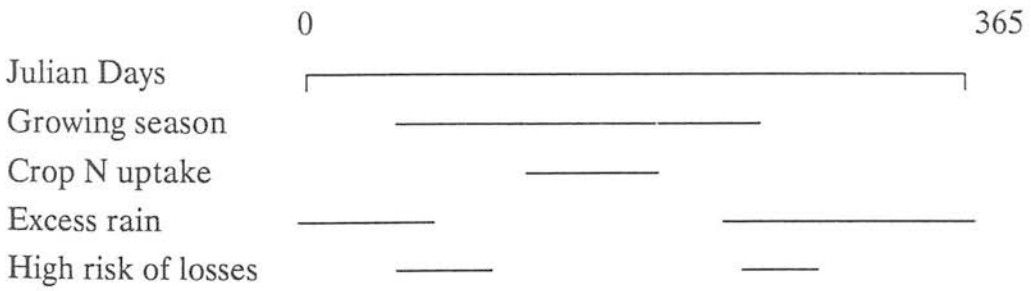
Figure 12 Structure for semi-quantitative model to optimise manure applications for a cropping strategy, given limited supplies of manure, to maximise crop value and predict the risk of N losses

The early sub-routines combine crop and climate data to estimate the N uptake efficiency of various crops and highlight the periods in which leaching losses may occur. The length and timing of the growing season, based on the period in which the base temperature of 5 °C is exceeded, and the length and timing of the period of excess rain, based on the period in which received rainfall exceeds potential evapotranspiration, can be derived from the climatic data available for the Meteorological Office stations around the country. The period in which crop N uptake occurs can be obtained from the combination of graphs of non-limited N uptake against time from a range of trials, by including only those periods in which the gradient of the graph exceeds a critical value. By combining these data, with the expected sowing dates of the crops, the N uptake efficiency could be calculated simply as:

$$\frac{\text{Length of N uptake period of crop}}{\text{Length of growing season}}$$

The periods where losses of N may occur may also be highlighted, as it is commonly assumed that few losses occur where a crop is rapidly growing, unless N is supplied to a large excess.

eg. In this case crop N uptake efficiency is 35.4 %



The other two sub-routines which input into the final allocation of manure are less simple. The estimation of the basal rates of mineralisation based on soil properties, especially organic matter, and supplemented by N supplied by the residues of any previous cropping is largely understood qualitatively but not quantitatively. Further testing of the hot KCl extraction technique used to estimate soil N uptake for spring barley (McTaggart and Smith, 1993) may confirm the hypothesis that this is an index for the rate of basal mineralisation. The modifications to fertiliser recommendations based on the soil index system (ESCA, 1985) would then provide an estimate of the N supplied according to

the previous cropping of the field, which would supplement this basal mineralisation.

The estimation of the availability of the N supplied in manures for crop uptake in the first year is the least complete part of the model. The results obtained during the project have suggested that the potential availability of manure N is controlled by the size of the soluble C and soluble N pools added to the soil. However, the efficiency with which the N immobilised by the biomass can be used by the crop is not fully understood. Although, it is known that the availability of N for crop uptake decreases as the length of time between incorporation of manures and the sowing of a crop increases (Ladd *et al.*, 1983; Francis *et al.*, 1992), the effects of manure timings and application methods on crop N uptake were not studied during this project and therefore the modification of the potentially available pool of manure N by losses after application can only be approximated.

The proposed structure (Figure 12) highlights some of the data needed for the model not obtained as a result of this project. The prediction of the potential N supply from a manure using chemical indices was shown to be of little value. Therefore, the prediction of N supply from manures to crops was focused on assessing the availability of the mineral N and the organic N added in the manure. Although some progress has been made in understanding the processes controlling nitrogen release from manures, more work needs to be carried out with a range of organic materials before the N supply potential of a material can be predicted with any confidence.

The development of a semi-quantitative model which enables the N supply to be matched to crop demand in organic farming systems and therefore minimises the potential for N losses, still lies over the horizon. However, our understanding of the interacting processes of the soil nitrogen cycle has increased, especially with respect to N release from manures and this has led to the formulation of a simple model structure, which should be further developed, and after validation tested in the field.

4.1 Were the original objectives achieved?

4.1.1 Objective 1:

Describe empirically the timing of nitrogen release, and the efficiency of use of this nitrogen from a range of manures, including animal manures, green manures and ploughed-out leys.

For all the manures studied, whether in field or pot experiments, the timing of N release was monitored by measuring the mineral N concentration of the soil sequentially and the efficiency of uptake of this N was monitored by measuring crop yield and N concentration through the growing season. As a result, Objective 1 was achieved though the data are not always presented in this form in the text.

4.1.2 Objective 2:

Predict N availability from soil and manures, using appropriate availability indices, soil and manure properties and simple models.

As a part of the experiments carried out, two availabilty indices were tested. These proved to be inadequate to predict N release from manures. No combination of soil and manure properties used alone or in conjunction with the indices was able to satisfactorily predict N release from manures. As a result of the experiments carried out, Objective 2 was not achievable.

4.1.3 Objective 3:

Relate N release characteristics to the N uptake pattern of crops, likely to be used in organic crop production in Scotland.

Only a small range of crops were used in the experiments and so the relationship between N release characteristics of manures and crop uptake was only studied for cereals, dominantly spring barley. The 1990 trial, however, showed that sowing date was one of the most important factors affecting the efficiency with which manure N could be recovered by different crops (Paper I, p. 6). For Objective 3 to have been fully achieved a wider range of crops should have been studied in the experiments.

4.1.4 Objective 4:

Increase understanding of the interacting processes of the soil nitrogen cycle, with respect to nitrogen release from organic manures.

The use of ^{15}N revealed the complex nature of the processes of the soil nitrogen cycle , with respect to the release of N from manures. I feel that my understanding of the soil N cycle has increased as a result of this work. However, I doubt that this objective will ever be completely achieved.

4.1.5 Objective 5:

Produce a reliable data set to allow development of a semi-quantitative model enabling N supply to be matched to crop demand in organic farming systems, and for farmers using a high proportion of organic wastes as fertilisers, thus minimising losses of N to the environment.

Objective 5 was not achieved, since the data set obtained was not large enough to allow such a model to be developed. A possible structure for such a model was outlined which highlighted the areas in which more data need to be collected so that the model can be more fully developed and validated.

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PAPER I

SOIL NITROGEN MANAGEMENT AND INTERACTION WITH CROP PESTS AND DISEASES IN ORGANIC FARMING

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INTRODUCTION

The supply of nitrogen is one of the principal factors limiting organic crop production. As well as providing adequate quantities of nitrogen, an organic farming system (like a conventional one) must supply available soil nitrogen at a time which matches crop demand. The timing of maximum demand and the pattern of nitrogen uptake through the season will vary significantly between crops. Where nitrogen supply and crop demand can be closely matched, maximal yields will be obtained with minimal losses of nitrogen to the environment (Powlson, 1988). The slow release of nitrogen from some manures may be a limiting factor to the growth of some organic crops, especially those with a high nitrogen demand in early spring when rates of mineralisation are low (Redman et al., 1989).

This study was undertaken to compare the nitrogen uptake patterns and the efficiency of use of nitrogen from heap-composted cattle manure by two crops, spring barley and maincrop potatoes. The incidence of pests and diseases on the potatoes and barley was monitored to test the hypothesis that manure applied in increasing quantities to plants will lead to increased pest and disease problems.

MATERIALS AND METHODS

Two field experiments were carried out with spring barley (cv. Atem) and potato (cv. Cara) at Jamesfield Farm, Abernethy during 1990. The farm held Soil Association certified organic status and the experiments were situated on soils described as a complex of Carpow and Carey series, dominated by the imperfectly drained Carey series (Speirs, 1989).

The field was treated with farmyard manure (30 t/ha) in the autumn of 1989. The spring barley was drilled on 18.4.90 and potatoes were planted on 2.5.90 in four blocks. Farmyard manure (20 t/ha) was applied to half the plots at the beginning of April, while the other plots were left untreated , with the treatment randomised within the blocks.

Microplots of N-15 labelled manure were laid out within the manured plots. The labelling was carried out by incubating a portion of manure with a small amount of high enrichment N-15 ammonium sulphate (99 atm%) for four

weeks. The addition of the inorganic salt did not significantly change the overall N content of the manure (ave. 2.2% N dwt) but resulted in an enrichment of the manure to c. 1 atm% N-15 abundance. The enrichment of the manure occurred preferentially in the microbial and inorganic pools of nitrogen due to the method of labelling.

Nitrogen uptake was monitored in the experiments by successive harvests of above ground plant material in all treatments; a sample of potato tubers was taken once only at the end of September. the plant samples were dried and milled before analysis for their isotope ratio and total N content by a Carlo-Erba total N analyser linked to a VG Isogas mass spectrometer. Soil samples (0 - 30 cm) were taken at the plant harvests. Available nitrate and ammonium were extracted from fresh soil with 1 M KCl and then determined by continuous flow analysis. Pests and diseases were assessed on the foliage of both potatoes and barley throughout the growing season; the incidence of diseases on harvested tubers was also assessed.

RESULTS

Both the barley (above ground material) and the potato haulms showed a sigmoid curve for increase of dry matter (DM) and nitrogen uptake with time (Fig. 1). Tuber samples were taken once only (at lifting) and so no curve could be produced for tuber DM increase and nitrogen uptake. However, unlike barley roots, the tubers form a significant 'sink' for nitrogen, with plant increase in nitrogen occurring mainly in the tuber from six weeks after emergence (Dyson and Watson, 1971). This means that the observed decline in nitrogen uptake by the haulms would be more than compensated for by tuber uptake, and for the potato plant as a whole nitrogen uptake would have continued to increase through July and early August only levelling out as the tubers reached maturity (Moorby, 1978)

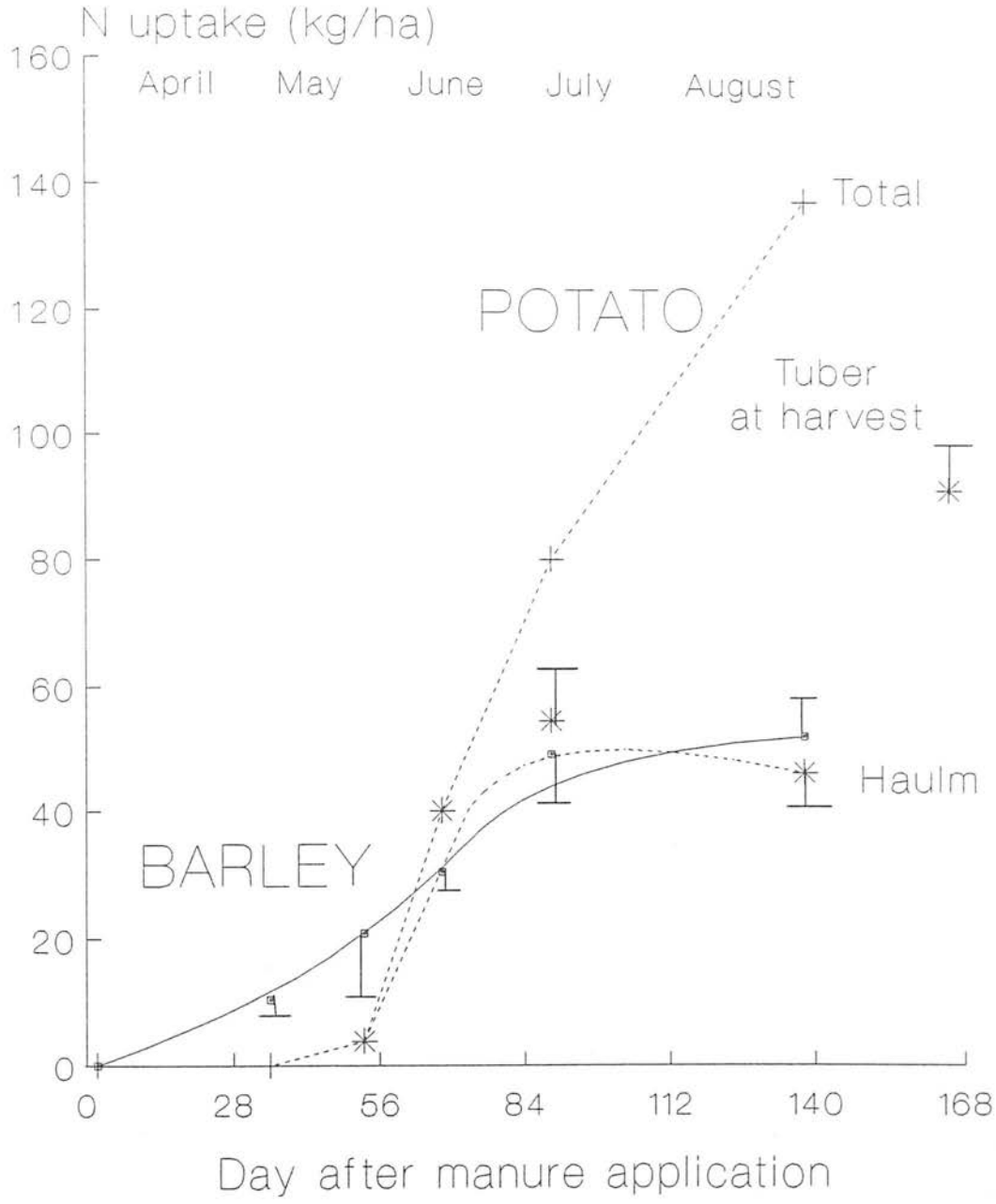

Figure 1 Cumulative N uptake in potato (---) and spring barley (---) through the growing season at Jamesfield 1990. Values for total N uptake in potatoes (+) are derived from a model using values of measured N in haulms through the season (*) and N measured in tubers in September (x). SE of mean values indicated.

At harvest in the barley, the manured plots had significantly higher yields ($P < 0.05$) of both grain and straw than the control plots. However, at all the earlier cuts, there were no significant differences between treatments, although manured plots showed consistently greater mean values. In the potato crop, the manured plots had very significantly higher yields during the period of rapid stem elongation ($P < 0.01$), but the differences between the treatments reduced as the season continued. The residual effects of the autumn manure application and selective grazing by rabbits appear to have reduced the differences between treatments.

The potato plants increased in nitrogen much more rapidly than the barley plants, after their emergence around the beginning of June. This may be linked to the larger pool of mineral nitrogen available in the soil, when the potatoes were planted (Fig. 2). Net mineralisation of 0.9 kg N/ ha/ day was observed assuming no losses for leaching and denitrification in the potato plots for the month between manure application and planting. under the barley crop for the same period, taking crop uptake in to account, net mineralisation was observed as 0.5 kg N/ ha/ day.

The changes in the mineral nitrogen pool in the soil through the season seemed to mirror crop demand. A decline of the mineral nitrogen pool under barley occurred as the barley took up nitrogen from April to June, but under potatoes mineral nitrogen initially increased until the potatoes began to rapidly take up nitrogen towards the end of May. Mineral nitrogen levels remained low under the potato crop during July and August, as the tuber demand for nitrogen remained high. However, under the barley crop, nitrogen uptake approached zero during August and an increase in the amount of mineral nitrogen in the soil was observed.

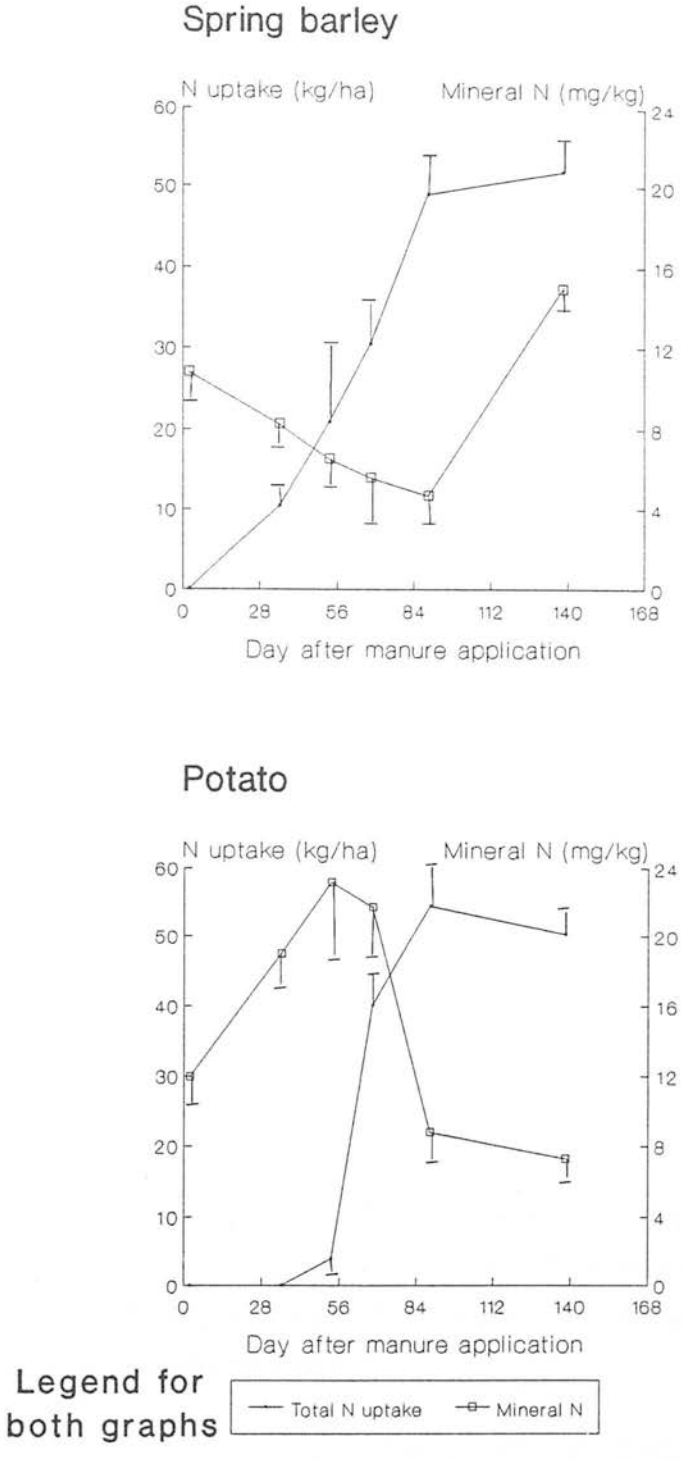

Figure 2 Changes in the mineral N pool in the soil and N uptake through the season in barley and potato haulms at Jamesfield 1990. SE of mean measured values indicated.

In both crops, the main accumulation of N-15 in the plant occurred early in the season (Fig. 3). This was probably due to the preferential labelling of labile pools of nitrogen in the manure. In the potato crop, although there was a general decline in the proportion of plant nitrogen derived from the labelled pools of nitrogen in the manure through the growing season, there was a large uptake of labelled nitrogen during the early part of stem extension. This was probably linked to the rapid growth of the stolons below ground and the onset of tuber development. While the stems were still expanding there seemed to be a transfer of nitrogen from the haulms to the newly initiated tubers, showing as a marked drop in the amount of labelled nitrogen in the haulms. The barley crop recovered 4.9% of the nitrogen from the manure; this does not include non-labelled nitrogen recovery and is therefore an underestimate. The potato crop recovered 23% of the nitrogen from the manure, but this is also an underestimate.

The incidence of diseases on the potatoes was low to non-existent [eg. blackleg (*Erwinia carotovora* var. *atroseptica*), blight (*Phytophora infestans*), black scurf (*Rhizoctonia solani*), powdery scab (*Spongospora subterranea*)] and as a result, treatment differences were not apparent. The effects of the treatments on potato aphid numbers are shown in Table 1. The manure treatment affected potato aphid numbers only on 14 June when it significantly ($P < 0.05$) increased total numbers of *M. euphorbiae* over untreated plots.

The incidence of diseases on the barley was very low [eg. powdery mildew (*Erysiphe graminis*), leaf blotch (*Rhynchosporium secalis*)] and as a result treatment differences were not apparent. aphids were assessed approximately every 10 days from 29 May until 7 August. Three aphid species were assessed: rose-grain aphid (*Metopolophium dirhodum*), grain aphid (*Sitobion avenae*) and bird cherry aphid (*Rhopalosiphum padi*). The effects of the treatments on the rose-grain aphid, numerically the most dominant species during 1990, are shown in Table 2. The aphid numbers peaked on or around 11 July, when significantly ($P < 0.05$) more were found on manure treated plots than on untreated plots, both total aphid and wingless nymphs.

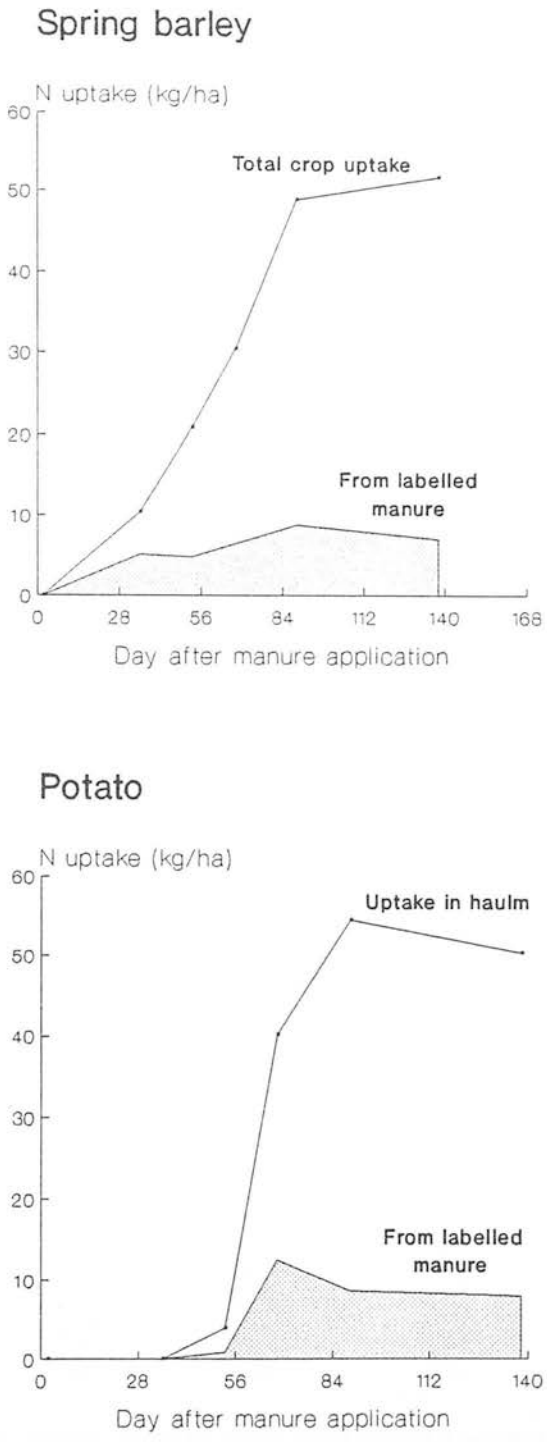

Figure 3. Cumulative uptake of N in barley and potato haulms through the growing season - partitioned into N uptake from labelled sources (manure) and unlabelled sources (soil and some manure fractions).

Table 1 Pests and diseases x nitrogen on potatoes 1990; total aphid numbers [means (transformed by √x + 0.5) and least significant differences]
a) potato aphid (*Macrosiphum euphorbiae*)
and b) peach potato aphid (*Myzus persicae*).

Sampling dates	Treatments		LSD (P = 0.05)
	Manure	Untreated	
(a)			
14 June	2.5	1.9	0.59
20 June	1.4	1.4	0.84
4 July	3.9	3.2	1.85
11 July	4.6	5.0	1.36
25 July	9.5	9.3	3.07
7 August	3.6	3.5	1.08
21 August	1.9	1.7	0.82
(b)			
14 June	0.8	0.8	0.24
20 June	1.3	1.1	0.51
4 July	1.7	1.2	1.15
11 July	2.2	2.6	0.67
25 July	5.0	4.9	2.04
7 August	1.9	1.8	0.75
21 August	0.8	1.3	0.67

Table 2 Pests and diseases x nitrogen on spring barley 1990; total numbers [means (transformed by √x + 0.5) and least significant differences] of rose-grain aphids (*Metopolophium dirhodum*).

Sampling dates	Treatments		LSD (P = 0.05)
	Manure	Untreated	
29 May	1.3	1.2	1.1
6 June	1.5	1.1	0.7
14 June	1.1	1.5	1.5
20 June	1.9	2.5	3.9
4 July	5.8	6.1	0.8
11 July	10.6	9.0	1.1
25 July	8.7	8.1	5.6
7 August	0.7	0.8	0.4

DISCUSSION

The manure increased aphid numbers early in the season on potatoes and by the middle of the season on barley. some evidence exists to support the hypothesis that manure applied in increasing quantities to plants will lead to increased pest (in this case, aphid) problems. The different times at which aphid numbers increased on the crops may be linked with the different rates of mineralisation of nitrogen in the soil under the two crops: aphid numbers were increased by manure in the middle of June when net mineralisation was of the order of 0.9 kg N/ ha/ day; and aphid numbers on barley were increased by manure in the middle of July when the net mineralisation was of the order of 0.5 kg N/ ha/ day.

These results indicate that the potato crop had a higher demand for nitrogen, and was more efficient in terms of its uptake of applied N-15. The increased utilisation of the labelled nitrogen sources by the potato crop compared to the barley crop was linked to the later sowing and harvesting of potatoes, so that the potato crop was still actively taking up nitrogen throughout the warm summer months. the barley crop had a high demand for nitrogen early in the growing season, when temperatures were lower, and did not use the labelled nitrogen sources as efficiently as potato. This relationship also held for unlabelled manure and soil sources of nitrogen.

Early sown crops, such as spring barley, may profit form the use of an organic manure which has a high proportion of mineral nitrogen eg. poultry manure, slurries. Whereas potatoes and other field vegetables are able to utilise the nitrogen released from more 'organic' manures.

In planning the nitrogen strategy for a farm, it is important to consider nitrogen uptake patterns of crops and use available nitrogen sources to maximise nitrogen uptake and yield by the crop and also to minimise nitrogen losses to the environment.

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PAPER II

DEVELOPMENT OF A SUITABLE METHOD FOR MEASURING GROSS MINERALIZATION RATES *IN SITU* UNDER SCOTTISH CLIMATIC CONDITIONS.

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ABSTRACT

Intact soil cores from 3 sites were injected with a small amount of 99 atm% enriched ammonium sulphate and incubated at a range of temperatures or moisture potentials for between 1 and 40 days in the laboratory. The experiments were were aimed at developing a suitable methodology for measuring field rates of gross mineralization in intact cores, following work done by Davidson et al. (1991).

Very large drops in the $^{15}NH_4^+$ pool were seen in the first 24 hours after injection, which were not characteristic of the whole incubation period. This rapid fall was extended to 48 hours at the lowest temperature and may be caused by slow equilibriation of NH_4^+ between the fixed and exchangeable pools. The added $^{15}NH_4^+$ and pre-existing NH_4^+ pools may also be subject to different consumption rates during this period. Recycling of ^{15}N was indicated in our experiments by an increase in the amount of $^{15}NH_4^+$. This was seen after seven days at 14°C, and took longer to occur at the lower temperatures.

Use of pool dilution methods must be accompanied by a check that the underlying assumptions are valid and experimental procedures should be chosen to minimise error. Spatial variability of mineral nitrogen in the field can lead to inaccuracies in calculated rates. Where the method is used with care, however, it will lead to a greater understanding of the processes of the soil nitrogen cycle.

INTRODUCTION

The soil nitrogen cycle involves a large number of identifiable pools of nitrogen linked by complex, often simultaneous and opposing, processes (Jansson, 1958; Paul and Juma, 1981). Simple observations mapping the sizes of the nitrogen pools involved over time are therefore not adequate to describe the full dynamics of the system (Jansson and Persson, 1982). Use of ^{15}N has provided a powerful tool, where direct tracer methods determine the location of ^{15}N after a period of exposure and pool dilution methods estimate flow rates through a given nitrogen pool (Nason and Myrold, 1991). Where labelled NH_4^+ is added to the NH_4^+ pool in the soil and comes rapidly into equilibrium with it, the decline in the ^{15}N enrichment of the pool, as NH_4^+ at natural abundance is introduced by mineralization of soil organic nitrogen, can be used to calculate the gross mineralization rate. In the same way addition of labelled NO_3^- can allow measurement of gross nitrification rate.

As in all isotopic work, it is necessary to assume that ^{14}N and ^{15}N are not discriminated by processes occurring in soils and that the added ^{15}N equilibrates rapidly with the pool to which it has been added, creating a single homogeneous pool. Whilst some microbial transformation processes do discriminate between ^{14}N and ^{15}N (Cheng et al., 1964; Heaton, 1986), the assumption probably holds for incubations of enriched samples over a short time period. The heterogeneity of ^{15}N in the organic nitrogen pool has not been studied and it is assumed that the ^{15}N abundance is naturally at background levels (Wessel and Tietema, 1992). There are some indications from plant uptake studies (McTaggart, 1992) that the

^{15}N natural abundance of the active pool of soil organic nitrogen is higher (0.370 - 0.377 atm%) than the value of 0.3663 atm%, and where possible the ^{15}N natural abundance should be measured at the study site. When experiments are carried out in the laboratory with sieved and well mixed soils or litters (eg. Bjarnason, 1988; Wessel and Tietema; 1992) it is relatively easy to achieve uniform addition of ^{15}N. However, in the field it is almost impossible to achieve such uniform applications (Barraclough, 1991: Davidson et al., 1991). The heterogeneity of soils in the field , particularly the spatial variability of mineral nitrogen, is a serious problem for the development of *in situ* applications of isotope dilution since rate estimates are calculated using differences, which amplify errors (Myrold and Tiedje, 1986).

To simplify calculations, mineralization and immobilization rates are usually assumed to be constant (Kirkham and Bartholomew, 1954; Blackburn, 1979) or to vary according to some known relationship (Nason and Myrold, 1991), between measurements of pool size and enrichment. The change in the ^{15}N enrichment of the NH_4^+ pool as mineralization and consumption processes proceed is complex and simple averages of the ^{15}N enrichment between measurements (Shen et al., 1984; Guiraud et al., 1989) can only give approximations of gross rates. The enrichment of ^{15}N only declines linearly for very short periods of time even where mineralization and immobilization are proceeding at constant rate (Bjarnason, 1988).

A formal mathematical treatment to allow the calculation of gross mineralization and consumption rates has existed since 1954 (Kirkham and Bartholomew, 1954),

where the changes in pool sizes are described by differential equations, and solved analytically. Remineralization of immobilized mineral nitrogen is disregarded and the change in amount of ^{15}N in the NH_4^+ pool is derived only from the consumption process. Symbols are defined in Table 1.

$$\frac{d\,AL}{d\,t} = -c\,\frac{d\,AL}{d\,AT}$$

$$\text{Hence} \quad m = \frac{(AT_2 - AT_1)}{t}\,\frac{\log(AL_1AT_2 / AL_2AT_1)}{\log(AT_2 / AT_1)}$$

This framework is only valid where ^{15}N addition to the soil is high and where the ^{15}N enrichment of the NH_4^+ pool does not approach background by the end of the incubation period. However, it is still widely used for the calculation of gross mineralization rates (eg. Davidson et al., 1991; Ambus et al., 1992).

Table 1 List of symbols used in the equations

AT_1	Total size of NH_4^+ pool, $\mu g\ N\ g^{-1}$, at time 1.
AT_2	Total size of NH_4^+ pool, $\mu g\ N\ g^{-1}$, at time 2.
AL_1	Size of labelled NH_4^+ pool, $\mu g\ N\ g^{-1}$, at time 1.
AL_2	Size of labelled NH_4^+ pool, $\mu g\ N\ g^{-1}$, at time 2.
t	Time between measurements, days.
@	Natural ^{15}N enrichment of mineralising NH_4^+.
m	Rate of mineralization / production of NH_4^+, $\mu g\ N\ g^{-1}\ day^{-1}$.
c	Rate of NH_4^+ consumption, $\mu g\ N\ g^{-1}\ day^{-1}$.

The model was extended to allow for nitrogen mineralising at natural or any fixed ^{15}N abundance from the organic nitrogen pool for anoxic sediments (Blackburn, 1979) and for aerobic soils (Nishio et al., 1985), deriving the decline in ^{15}N enrichment from the consumption and mineralization processes, but still not accounting for remineralization. Symbols are defined in Table 1.

$$\frac{d\,AL}{d\,t} = @\,m - c\,\frac{d\,AL}{d\,AT}$$

$$\text{Hence } m = \frac{(AT_2 - AT_1)}{t}\,\frac{\log((AL_2/AT_2) - @\,/(AL_1/AT_1) - @)}{\log(AT_2 / AT_1)}$$

These equations can be applied to $^{15}NO_3^-$ as well as $^{15}NH_4^+$ additions (Schimel *et al.*, 1989), allowing calculation of both gross mineralization and nitrification rates. A calculation method allowing calculation of gross nitrification rates where only $^{15}NH_4^+$ is added has also been developed (Wessel and Tietema, 1992).

Kirkham and Bartholomew (1955) developed a second mathematical framework allowing for nitrogen mineralizing at natural abundance and possible remineralization of added labelled nitrogen, by estimation of the interacting organic nitrogen pool. However, this model was developed for a simple system of two pools with mass conservation assumed and it cannot be corrected for losses to the NH_4^+ pool other than by immobilization to organic nitrogen.
Numerical solutions of the differential equations have also been developed, where numerical simulation by use of non-linear curve fitting, uses the measured ^{15}N abundances in the mineral N pool to fit the gross transformation rates and the size

of the initial organic nitrogen pool involved (Myrold and Tiedje, 1986; Barraclough and Smith, 1987; Bjarnason, 1988). The advantages of numerical solutions are that they can be applied to any set of differential equations and the solution procedure remains the same, irrespective of the chosen set of rates, pools and other conditions (Wessel and Tietema, 1992). However, a high degree of replication is required to fit a solution with any degree of certainty and analytical models offer a quick way to calculate gross rates, so long as their assumptions have not been violated.

A study was carried out on three soil types to investigate the potential problems in applying the method used to measure gross mineralization rates described by Davidson et al. (1991). Concentrations of NO_3^- and NH_4^+ in injected and uninjected cores were compared to establish if the addition of $^{15}NH_4^+$ solution provided a stimulus to mineralization. Incubations were carried out for increasing periods to determine an appropriate length of field incubation under Scottish temperature regimes. Observations were made of remineralization on one soil type, whilst the effect of soil moisture potential on mineralization was observed in intact cores of the other soils. Some preliminary work was carried out to establish an appropriate injection methodology with minimum disturbance to the core, and to determine the variability of immediate NH_4^+ fixation within a soil type. A further experiment attempted to assess the replication necessary to allow estimates of the mean mineral nitrogen pool size.

Core sampling and core preparation

Intact soil cores, 54 mm in diameter and 20 cm deep, were sampled inside P.V.C. sleeves using a specially designed corer at three sites on the Bush Estate, 15 km south of Edinburgh, in winter 1991-2. Properties of the soils used in the study are presented in Table 2. The cores from the Beechgrove site were roughly crumbled and the sward torn into pieces in an attempt to simulate ploughing. The 'ploughed' soil was then packed back into the P.V.C. sleeve with the sward distributed randomly throughout. The core was pushed out of the liner into a polyester sock to enable good moisture equilibriation during incubation. The cores from the Glencorse and No. 3 sites were not prepared in any way but left intact for the incubations. A further 100 samples were taken from the Beechgrove site, three days after spring ploughing, using an Dutch auger, to allow assessment of the spatial variability of mineral nitrogen at the site.

Table 2 Properties of the soils used (0-20cm).

Site	Glencorse	No. 3 field	Beechgrove
Soil texture	Clay loam	Sandy loam	Sandy loam
O.M. %	5.0	4.0	4.8
pH	6.4	6.3	6.0
Crop	Cereals	Cereals	Pasture
Soil series	Winton	Macmerry	Winton

Core Injection

Several preliminary tests were carried out using an iodine-green dye solution to assess the most suitable method of injection and appropriate points of injection into the core. Preliminary extractions were carried out on twenty cores from the Beechgrove site to establish how much $^{15}NH_4^+$ should be added to each core to give an NH_4^+ pool with an enrichment of approximately 25 atm%. Five ml (measured gravimetrically) of 0.17 g l^{-1} $(^{15}NH_4)_2SO_4$ solution of 99.2 atm% enrichment, containing 190.06 μg ^{15}N, were injected into the core using 1 ml syringes at 5 points. As the solution was injected the needle was slowly withdrawn to aid an even distribution of solution through the core.

Core incubation

The cores from the Beechgrove site were incubated in sand tanks at a range of constant temperatures (4, 10, 14°C), chosen to reflect the seasonal range of temperatures in Scotland, for a number of incubation periods (1, 2, 4, 7, 11, 14 and 18 days). The tanks were filled with 32 cm depth of coarse sand, which had been saturated and left to drain to give a water table 1 cm above the base of the tank and 11 cm below the base of the cores. This water level was maintained using a simple gravity fed constant head device, and enabled the cores to be held at constant moisture potential through the incubation period. The cores were randomly divided into three groups and allowed to equilibrate for four days at each temperature before any cores were injected.

Cores of the Beechgrove soil were placed into the sand tanks into auger holes of an appropriate size and the sand was tamped to ensure a good core-sand contact. For

each temperature and each incubation period, three cores were injected with $(^{15}NH_4)_2SO_4$ solution and four replicate cores were used for assessment of net mineralization rates. The position of the cores was randomized in the tank, with cores 4 cm apart. Core temperatures were measured on each sampling date, using a temperature probe inserted through the centre of each core, prior to its removal from the tank. Gravimetric moisture contents were calculated for each core after removal, by oven drying at 105°C for 24 hours.

The cores from the Glencorse and No. 3 sites were incubated at approximately 18°C at a range of matric suctions (300, 100, 10 and 1 kPa) for three periods of time (7, 18, and 40 days). The 300 kPa and 100 kPa matric suctions were obtained using a pressure membrane apparatus. The 10 kPa matric suction was set up using a tension tank and this apparatus was modified to provide the 1 kPa suction. Four replicate cores for each matric tension and time period were injected with $(^{15}NH_4)_2SO_4$ solution, after equilibriation at the appropriate moisture potential. The gravimetric moisture contents of the cores were measured at the end of each time period.

Nitrogen analysis

Available NH_4^+ and NO_3^- were determined by extracting soil using 1 M KCl in a 1:5 soil:solution ratio. Extracts were filtered through Whatman 42 filter paper and NO_3^--N and NH_4^+-N determined by continuous flow analysis (Best, 1976; Crooke and Simpson, 1970). Four cores from each temperature or moisture potential were extracted at the beginning of the incubations. Analysis of injected cores for $^{15}NH_4^+$ enrichment was carried out by the steam distillation followed by mass

spectrometric determination as described by Hauck (1982). Where NH_4^+-N concentrations in the extracts were < 500 g, a carrier solution containing 1 mg N as NH_4^+ was added prior to steam distillation to ensure that enough N was present to allow determination of ^{15}N abundance.

To allow for fixation of the NH_4^+ in the soil, a recovery factor (defined in the results section) was calculated by extracting the NH_4^+ pool of the soil 15 minutes after injection and determining its ^{15}N enrichment (Davidson et al., 1991). This was carried out for the Beechgrove and Glencorse soils. A recovery factor was also calculated for the No. 3 and Glencorse soils where extractions took place within twenty four hours of injection.

Mathematical models

The analytical model developed by Blackburn (1979) was used to calculate the gross mineralization and consumption rates for the system, where remineralization could be neglected. As no value for the natural ^{15}N enrichment of the active organic pool was available, a natural abundance of 0.3663 atm% was used. This may have led to slight underestimates of the rates of gross mineralization.

Statistical methods

Differences between treatments were compared using oneway Anova procedures and injected and uninjected cores compared using t-tests. All analyses were carrried out using the MINITAB statistics package.

Recovery

After 15 minutes, only 61.4% on average of the $^{15}NH_4^+$ injected into the Beechgrove soil was recovered in a KCl extract ie. the mean recovery factor was 0.614 (S.E. 0.0366, 12 replicates). In the Glencorse soil, the corresponding recovery factor was 0.42. The processes leading to this disappearance did not seem to be highly spatially variable for any one soil series, but recovery was significantly different between soils ($p < 0.05$). The recovery factor was significantly ($p < 0.001$) affected by soil moisture (Fig. 1), with the values decreasing in the drier soils ($r = 0.854$; $df = 22$). The relationship seemed to hold over a relatively wide moisture range, irrespective of soil texture. Injected solution was subject to greater soil aggregate suction in the drier soils, hence in these soils the solution may have penetrated further into the soil aggregates and possibly encountered more fixation/consumption sites.

A separate set of recovery measurements were made for No. 3 field and the Glencorse site. However, in this case extractions were made within 24 hours of injection, rather than 15 minutes (Tab. 3). The recovery in No. 3 was higher than that in Glencorse, and the decline in the $^{15}NH_4^+$ over the first 24 hours in the experiment with the Beechgrove soil results in very low 24 hour recoveries, which were not significantly different between incubation temperatures (Drury and Beauchamp, 1991). The rapid disappearance of $^{15}NH_4^+$ seems to be extended to 48 hours at the lowest temperature. Compared to the data obtained for later in the experiment this period appeared exceptional.

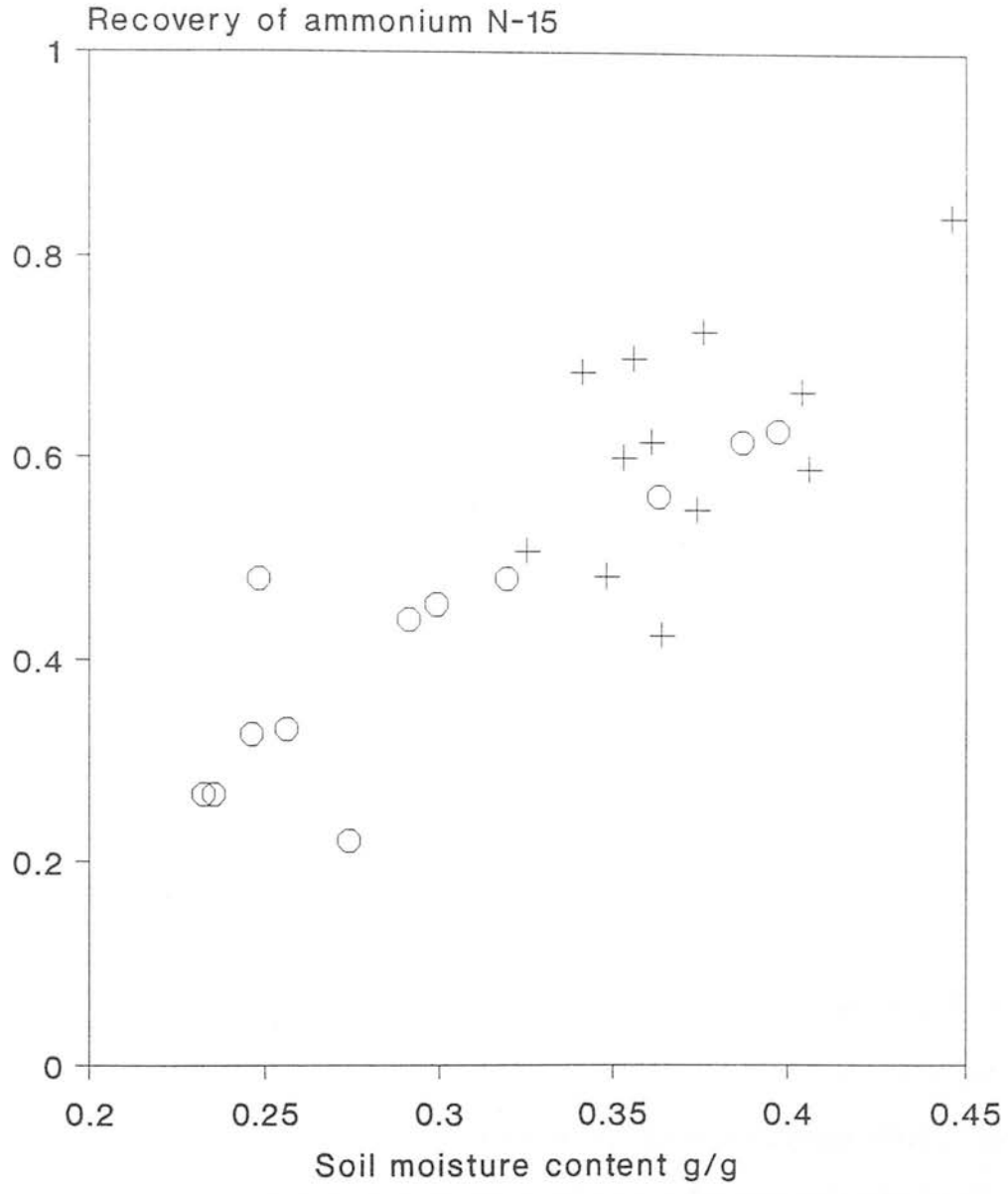

Figure 1 The recovery factor 15 minutes after injection plotted against soil moisture content (g g^{-1}) for two sites: Glencorse (clay loam; graph symbol, o) and Beechgrove (sandy loam; graph symbol, +). r = 0.854 df = 22.

Table 3 Mean recovery factors for $^{15}NH_4^+$ in the three soils, measured after 15 minutes and 24 hours.
N.D. = not determined.

	Recovery factor				
Time	15 minutes		24 hours		
Temperature	18°C	4°C	10°C	14°C	18°C
Site					
Beechgrove	0.61	0.076	0.054	0.13	N.D.
Glencorse	0.37	N.D.	N.D.	N.D.	0.17
No. 3	N.D.	N.D.	N.D.	N.D.	0.34

Fixation of NH_4^+ by clays (Drury and Beauchamp, 1991) or organic matter (Foster et al., 1985) are possible mechanisms to explain the rapid removal of $^{15}NH_4^+$ from the extractable pool after addition. Davidson et al. (1991) did not observe any significant effect of sterilization on the process and suggested that fixation by vermiculite and other 2:1 clays is the cause. Shen et al. (1984) observed that immediately after addition of $^{15}NH_4^+$, 1 - 5% of the labelled N was found in the "fixed fraction", extracted by the hypobromite method.

It is assumed that after adjustment of fixed NH_4^+, equilibriation of the added and exchangeable NH_4^+ rapidly occurs to give a homogeneous exchangeable NH_4^+ pool, which can be simply described in terms of its size and ^{15}N enrichment. The injection methodology should therefore be optimised to give as close to uniform distribution of added ^{15}N as is possible. However, any injection procedure will introduce liquid preferentially into the macropores. Thus the injected NH_4^+ is spatially separated from much of the natural solution, exchangeable and fixed NH_4^+. Given the low effective diffusion coefficient for NH_4^+ in soils (Barber, 1984), it has been calculated that on average in a moist soil NH_4^+ or K^+ will

diffuse about 0.13 cm in a day (Wild, 1981). The complex spatial distribution of production, consumption and fixation sites for NH_4^+ (Drury et al., 1991) means that $^{15}NH_4^+$ may well not reach many of the microsites during the initial incubation period and hence it would be liable to a different consumption rate than the natural NH_4^+. If preferential consumption of $^{15}NH_4^+$ is occurring shortly after injection, then overestimates of gross mineralization rates will be made, if that period is included in the calculations.

Davidson et al. (1991) observed that there was no difference in the amount of ^{15}N extracted from the sterilized soils at 15 minutes or 24 hours, and therefore suggested that the abiotic reaction is completed very quickly. However, this contrary to the results of Drury and Beauchamp (1991), who observed that fixation of $^{15}NH_4^+$ continued for at least 3 days. Schimel et al. (1989) measured similar consumption rates in intact and mixed cores and suggest that this indicates that the distribution of $^{15}NH_4^+$ was adequately uniform in the intact cores. Although it is not clear what processes are contributing to the rapid fall in the extractable pool of $^{15}NH_4^+$, it would seem more appropriate to allow at least 24 hours for the soil and added NH_4^+ to come into equilibrium before an initial measurement of the ^{15}N content of the NH_4^+ pool is made (Barraclough, pers. comm.). However, other workers only incubate cores for a 24 - 26 hour period to measure mineralization rates (Davidson et al., 1991; Ambus et al., 1992). Further work needs to be done to assess the speed with which an injected solution will diffuse throughout the soil pore system in soils of different structures and textures, using fluorescent dyes and applying diffusion models to assess how rapidly equilibrium between pools of NH_4^+ may be attained.

Remineralization

One of the assumptions necessary for the use of most analytical solutions of pool dilution measurements is that enriched NH_4^+ immobilized in microbial tissue during the incubation is not remineralized (Kirkham and Bartholomew, 1954; Nishio et al., 1985). An increase in the size of the ^{15}N content of the NH_4^+ pool strongly suggests that remineralization is occurring (Bjarnason, 1988) and if this is not taken into account in the mathematical framework used, negative gross immobilization rates may result (McTaggart, 1992). In the incubations with the cores of Beechgrove soil (Fig. 2), the ^{15}N content of the NH_4^+ pool began to increase after seven days at 14°C, whilst at the lower temperatures, where slower turnover might be expected, evidence of remineralization appeared later in the incubations. Incubations should not therefore be carried out for longer than a week (Bristow et al., 1987; Bjarnason, 1988). Remineralization is likely to be occurring before it is indicated by the increase in size of the $^{15}NH_4^+$ pool and will therefore decrease the decline in ^{15}N abundance and lead to underestimates of the gross rate of mineralization (Wessel and Tietema, 1992). Only by using models which take account of remineralization (mostly numerical) throughout the incubations (Myrold and Tiedje, 1986; Bjarnason, 1988) can gross rates of mineralization be found, where turnover rates are high and remineralization is significant from early in the incubations.

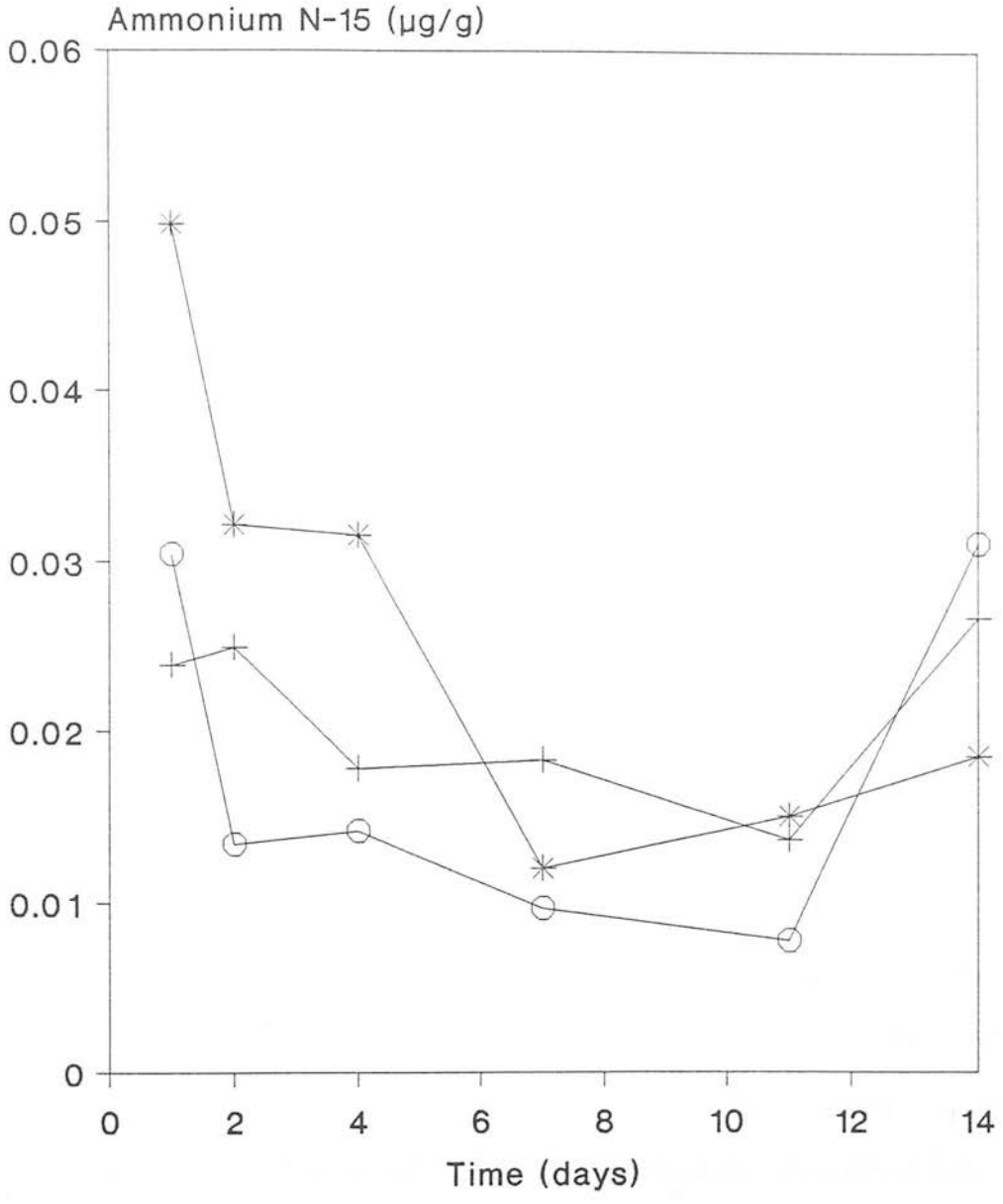

Figure 2 The change in $^{15}NH_4^+$ pool size ($\mu g\ g^{-1}$) with length of incubation using soil from the Beechgrove site at three incubation temperatures: 4 (o), 10 (+), and 14°C (*).

Release of $^{15}NH_4^+$, which has disappeared early in the experiment, can also lead to underestimates of the gross rates of mineralization. Fixed NH_4^+ is only released very slowly and when the solution activity of NH_4^+ is very low (Pasricha, 1976). Shen et al. (1984) observed that $^{15}NH_4^+$ fixed immediately after addition was released slowly during an incubation of 20 days with unfumigated soils, but remained constant or increased slightly when previously fumigated soils were incubated. In short term experiments release of NH_4^+ from sites, where it has been selectively fixed, is unlikely to significantly influence the change in ^{15}N enrichment of the NH_4^+ pool.

Spatial variability of soil mineral nitrogen

Shortly after ploughing at the Beechgrove site, NO_3^- levels were very low and at the bottom of the detectable range. NH_4^+ levels were higher, ranging from 1 - 7.5 μg N g^{-1} and showing a log-normal distribution (Macduff and White, 1984; Fig. 3). The geometric mean was 2.52 μg N g^{-1} and the coefficient of variability was 60%. Random groups of ten cores selected from the population indicated that the mean was estimated within its true 95% confidence interval on 6 out of 10 occasions (Fig. 4). The problem of spatial variability of the inorganic nitrogen pool is well known and large samples need to be taken to allow the population mean to be accurately estimated, in this case 40 cores. Wessel and Tietema (1992) carried out a separate experiment with a larger number of replicates to follow the changes in the mineral nitrogen pool as a part of their pool dilution experiments. The sampling of a large number of cores at the beginning and end of the field incubations would allow better estimation of the size of the mineral nitrogen pool, though if cores were covered to prevent leaching the sampling area would also have to be covered.

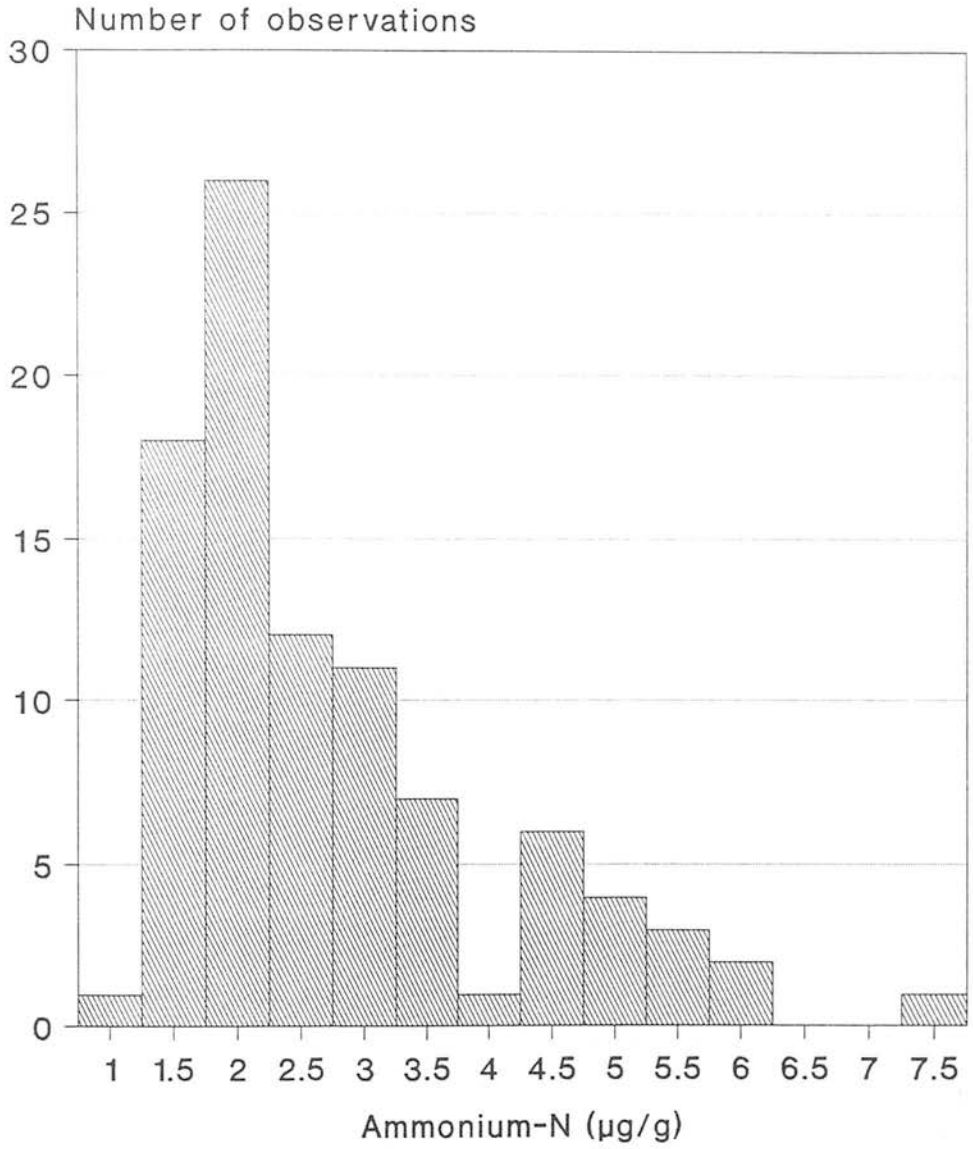

Figure 3 The frequency distribution for NH_4^+ concentration ($\mu g\ N\ g^{-1}$) determined for 100 cores from the Beechgrove site after spring ploughing.

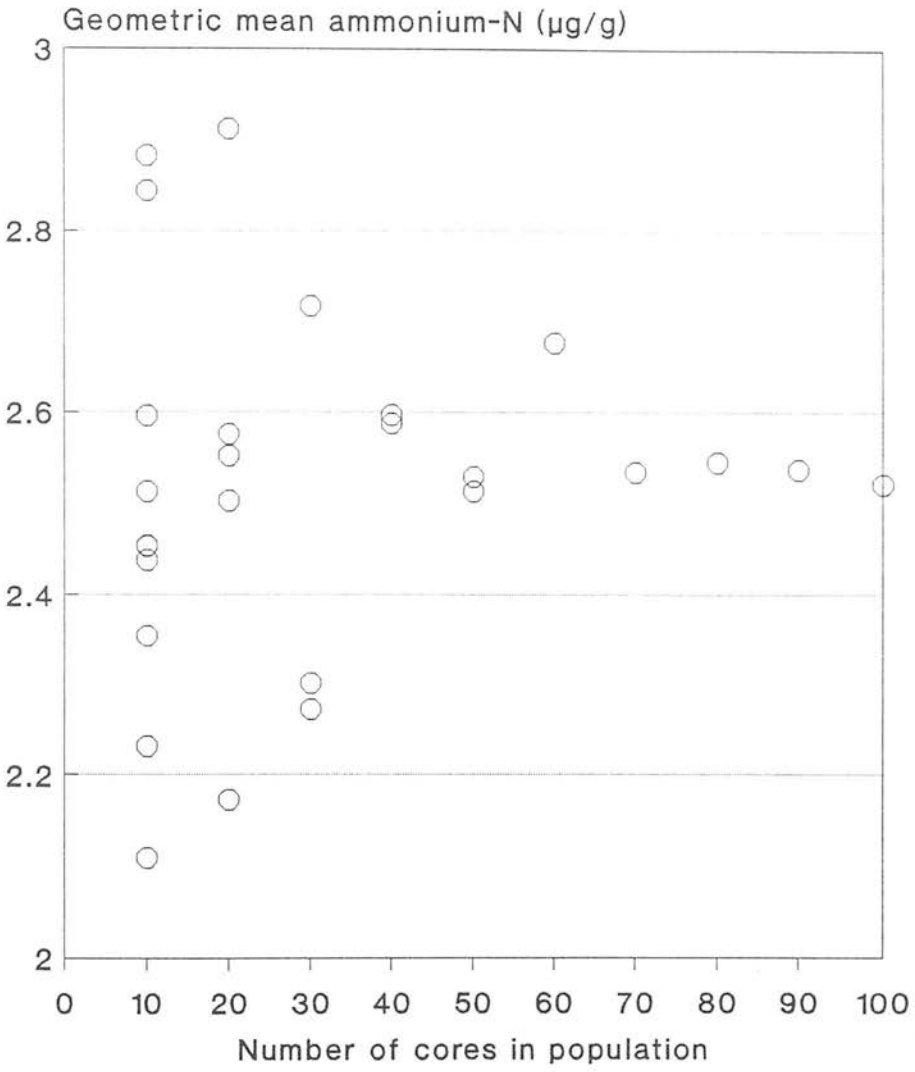

Figure 4 The geometric means for populations of different numbers of cores selected randomly from the 100 cores sampled at the Beechgrove site after spring ploughing against number of cores in the population.

Spatial variability of the soil mineral nitrogen pool and rates of transformation processes (Drury et al., 1991) will lead to spatial variability of the ^{15}N enrichment of the NH_4^+ pool, even where the application is uniform (Davidson et al., 1991). Where such variability is random, small but non-significant errors are introduced to the mineralization rates calculated, though spatially biased distribution of injected $^{15}NH_4^+$, eg. with respect to depth, should be avoided. Davidson et al. (1991) showed by simulation that where less than 70% of mineralizing-immobilizing microsites received ^{15}N as a result of the injection, mineralization rates would be significantly underestimated.

Net mineralization rates

There were no significant differences in NO_3^- and NH_4^+ concentrations in injected and uninjected cores, indicating that injections of small amounts of high enrichment NH_4^+ solution in the experiment did not seem to affect rates of net mineralization. This is in accordance with previous studies showing that net mineralization was unaffected or reduced following additions of inorganic nitrogen (Shen et al., 1984). It was considered by Jenkinson et al. (1985) that only in exceptional circumstances (eg. following recent additions of high C:N ratio residues or when pH is affected by fertilization) would addition of nitrogen affect net mineralization. In the Beechgrove soil, seven replicate cores were therefore used to give the NO_3^- and NH_4^+ concentrations for each temperature, at the end of each incubation period. NH_4^+ and NO_3^- concentrations were widely different between cores and even with seven replicates the standard error of the mean was large (Tab. 4). In experiments with all the soils, the variability of the NH_4^+ and NO_3^- concentrations within treatments was often more significant than the difference

between treatments and this affected the calculation of the net mineralization rates. The net rates of mineralization varied between 3.48 μg N g^{-1} day^{-1} and -1.23 μg N g^{-1} day^{-1}. No significant effects of increasing temperature or moisture on rates of net mineralization were seen, but the No. 3 soil showed more rapid rates of net mineralization (0.25 μg N g^{-1} day^{-1}) than Glencorse (0.13 μg N g^{-1} day^{-1}).

Table 4 The concentrations of NO_3^- and NH_4^+ in the cores from the Beechgrove site at the end of each incubation period. Values are means of seven replicates with standard errors of the means given in brackets.

Incubation temperature	Time (days)	NH_4^+ (μg N g^{-1})	NO_3^- (μg N g^{-1})
4°C	0	0.96 (0.19)	7.05 (1.23)
	1	1.85 (0.26)	9.64 (1.09)
	2	2.01 (0.45)	8.99 (0.89)
	4	3.04 (1.07)	9.34 (0.76)
	7	2.04 (0.27)	10.09 (1.06)
	11	1.81 (0.12)	12.81 (1.00)
	14	5.61 (1.11)	12.45 (1.68)
	18	3.00 (1.04)	11.12 (1.53)
10°C	0	1.75 (0.18)	5.78 (3.12)
	1	2.60 (0.39)	3.70 (0.53)
	2	2.92 (0.41)	4.59 (1.80)
	4	3.32 (0.71)	3.91 (0.55)
	7	5.14 (0.63)	3.30 (0.48)
	11	3.51 (0.85)	3.46 (0.75)
	14	3.93 (1.45)	10.58 (1.78)
	18	2.62 (0.77)	6.87 (1.85)
14°C	0	3.51 (1.12)	3.84 (0.78)
	1	4.72 (1.23)	4.24 (1.63)
	2	4.01 (0.93)	5.51 (2.58)
	4	4.35 (1.45)	7.32 (2.78)
	7	2.89 (0.96)	5.00 (0.89)
	11	2.44 (0.68)	4.02 (1.00)
	14	3.25 (1.00)	7.75 (1.57)
	18	2.70 (0.83)	18.38 (3.84)

Since estimates of NH_4^+ pool sizes are needed for the calculation of gross mineralization rates, such variability introduces a large degree of uncertainty into the calculation of gross, as well as net, mineralization rates.

Gross mineralization and consumption rates

The estimates of gross mineralization rates in all the soils are similar to those observed by Davidson et al. (1991) in grassland soil. Estimates made in forest floor litters give much higher mineralization and consumption rates (Davidson et al., 1991; Wessel and Tietema, 1992). There was no significant effect of increasing moisture potential in the No. 3 soil, with mineralization and consumption rates being estimated at 0.9 μg N g^{-1} day^{-1}. In the Glencorse soil, no significant effect of moisture potential was seen for consumption rates (0.75 μg N g^{-1} day^{-1}). However, mineralization rates increased significantly with decreasing moisture potential from 0.9 μg N g^{-1} day^{-1} at 300 kPa to 1.8 μg g^{-1} N day^{-1} at 10 kPa. In the Beechgrove soil, estimates of gross mineralization rates were made with reference both to day 0 (with a correction for 15 minute recovery) and to day 1 (Tab. 5). Decreasing rates were seen with increasing incubation period if day 0 was used. However, this trend almost disappeared if concentrations after 1 day were used as the initial conditions. This seems to indicate that it may not be remineralization leading to the fall in gross mineralization rates with time (Wessel and Tietema, 1992) but that some preferential consumption of $^{15}NH_4^+$ is occurring before equilibrium is established. Mineralization and NH4+ consumption rates determined in intact cores indicate a very rapid turnover of the active fraction of the soil organic nitrogen (Ambus et al., 1992).

Table 5 Gross mineralization and consumption rates for the Beechgrove soil for the first seven days calculated with t=0 and t=1 as the initial values.

Incubation temperature	Day	m (μg N g^{-1} day^{-1})		c (μg N g^{-1} day^{-1})	
		To t=0	To t=1	To t=0	To t=1
4°C	1	4.35		3.67	
	2	3.42	2.91	3.00	2.74
	4	2.82	2.28	2.33	1.85
	7	1.26	0.86	1.13	3.56
10°C	1	7.30		6.62	
	2	4.01	0.37	3.50	14.87
	4	2.89	1.23	2.53	0.98
	7	2.20	1.20	1.87	0.93
14°C	1	9.69		8.62	
	2	5.40	2.10	5.22	1.41
	4	2.98	1.01	2.80	1.14
	7	2.52	1.90	2.63	2.20

CONCLUSIONS

The use of pool dilution experiments seems to provide a method for measurement of the gross rates of some of the simultaneous and often opposing processes occurring in soils. However, the soil internal nitrogen cycle model proposed by Jansson (1958) does not always fit all the observed results (Myrold and Tiedje, 1986; Drury et al., 1991). Pool dilution experiments may well pose as many questions as they answer about the interlocking processes occurring in the soil. Use of the method must be accompanied by a check that the underlying assumptions are valid and experiments must be designed to minimise error multiplication factors (Wessel and Tietema, 1992). Most information may be obtained, where the fate of ^{15}N added is also determined at the end of the incubation period.

Optimising the injection procedure can help to ensure that a homogeneous NH_4^+ pool is achieved as rapidly as possible. However, it is recommended that the injection procedures are checked for each soil to be studied using dyes and/or modelling. Even where uniform addition of $^{15}NH_4^+$ can be achieved, slow equilibriation of NH_4^+ between the fixed and exchangeable pools, limited by diffusion, will also lead to a pool dilution effect and added $^{15}NH_4^+$ and natural NH_4^+ may be subject to different consumption rates initially. While this problem is exacerbated by non-uniform additions in intact cores, it may also be a problem in laboratory incubations. Short term recovery analysis to correct for the amount of $^{15}NH_4^+$ becoming irrecoverable is necessary (Davidson et al., 1991). However, from our results it would seem more appropriate to determine the initial NH_4^+ pool size and enrichment 24 - 48 hours after injection.

Davidson et al. (1991) suggest that estimates of rates are most accurate where drops in the enrichment of the pool are large over the incubation period, but the final ^{15}N enrichment should not approach too closely to background (Wessel and Tietema, 1992). Larger additions of $^{15}NH_4^+$ would help to achieve these aims, but care must be taken as the system may be dramatically altered if subtrate is added to a substrate-limited process.

Application of analytical solutions to calculate gross rates means that incubations should be completed before recycling becomes significant. Recycling was observed after seven days in the experiment with the Beechgrove soil. However, remineralization may become significant much earlier. The length of incubations should therefore be limited to approximately one week after injection. The use of

numerical solutions may remove this restriction, however numerical models may need to be expanded to take NH_4^+ fixation into account.

The measurement of gross mineralization rates can however only produce estimates of these rates especially where measurements are made *in situ*. The spatial variability of mineral nitrogen in soils leads to a measurement of initial and final mineral nitrogen pools with significant errors attached, which are multiplied in the calculation of gross mineralization rates. The use of a parallel experiment using a large number of uninjected cores over the same or a longer time period than the injected cores, allows an increase in the accuracy with which pool sizes can be measured.

The use of pool dilution techniques may on many occasions seem to have more problems than advantages. However, the continued sensible use and continual reassessment of these procedures can lead us further in our understanding of the processes controlling the supply of mineral nitrogen from soils in forms available to plants.

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PAPER III

RELATIONSHIPS BETWEEN BIOMASS NITROGEN AND NITROGEN EXTRACTED BY OTHER NITROGEN AVAILABILITY INDICES

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ABSTRACT

Experiments were carried out to investigate relationships between a number of nitrogen availability indices and biomass nitrogen, in a wide range of Scottish soils, and to establish the source of the nitrogen released. In one experiment, three soils were incubated for a week with high enrichment (99.2 atom %) $(^{15}NH_4)_2SO_4$ to label the soil biomass, and extracted as follows: 1 M KCl (Available Mineral-N; MIN-N); by fumigation-incubation (Microbial Biomass-N; BIO-N); by drying at 70°C, rewetting, incubating and then measuring mineral-N released (DRY-N); by anaerobic incubation, followed by measurement of NH_4^+ (Anaerobic-N; AN-N); by refluxing soils with 2 M KCl for 4 hours and measuring the NH_4^+ released (Hydrolysable-N; CHEM-N); by measuring labelled N uptake by ryegrass (Plant-N Uptake; RYE-N); and by Kjeldahl digestion (TOTAL-N). The isotopic enrichment of the N released by each of the above methods was determined. In 1992, the N contained in 17 soils from a wide range of sites was extracted using the above techniques but without selective labelling.

The amounts of N extracted increased in the order of MIN-N < CHEM-N < AN-N < BIO-N < DRY-N < < TOTAL-N. Biomass-N was found to be well correlated with the N extracted by DRY-N ($r = 0.82$; $P < 0.001$), AN-N ($r = 0.75$; $P < 0.001$) and CHEM-N ($r = 0.54$; $P < 0.01$). The results of the ^{15}N labelling experiment demonstrated that the different techniques resulted in the extraction of different N pools. The correlation between the ^{15}N enrichment of the N extracted in the microbial biomass and that in the CHEM-N was very low (0.06), whereas the corresponding values for AN-N, DRY-N, MIN-N and RYE-N were 0.91, 0.97, 0.85 and 0.93. The biological extraction techniques seem to have extracted N at least partly from the microbial biomass pool. The chemical extraction technique would appear to have extracted nitrogen from a quite different pool, or pools, much more closely related to the total soil nitrogen pool ($r = 0.95$; $P < 0.001$).

INTRODUCTION

Many methods have been used to assess the amount of N that is potentially available for crop uptake, a number of which are related explicitly or implicitly to the size and activity of the microbial biomass. Potentially available-nitrogen is released throughout the cropping season by mineralization of organic compounds in the soil, carried out by the action of microorganisms. Microbiological processes now form the core of many models of the agricultural and terrestrial cycle (eg Van Veen et al, 1984; Jenkinson and Parry, 1989) and the biomass itself may be a major source of N for mineralization (Bonde et al, 1988; Paul and Juma, 1981).

It has been proposed that soil organic N occurs in a number of pools. Paul and Juma (1981) defined four; the biomass, active non-biomass (recent residues), stabilised N and old organic matter. The turnover of the biomass has been shown to be up to five times faster than other fractions of soil organic matter (Amato and Ladd, 1980). However, during a twelve week incubation in a loam soil (Paul and Juma, 1981) the biomass, active and stabilised pools were found to contribute equally to the total N mineralized. The faster turnover times of the biomass and active pools were offset by the fact that they only represented a small fraction of the total pool of soil N.

Potentially mineralizable N as determined by the aerobic incubation technique of Stanford and Smith (1972) has been correlated with the microbial biomass in a wide range of soils by Bonde et al (1988), Carter and Macleod (1977), and Carter and Rennie (1982). Myrold (1987) related microbial biomass-N and the N released during an anaerobic incubation and showed that the origin of the N mineralized was the aerobic microorganisms killed as a result of flooding. Anaerobic incubation is recommended by the American Society of Agronomy as a biological index of N-availability (Keeney, 1982) and has been widely related to N uptake, especially in forest soils (Geist, 1977; Keeney and Bremner, 1966). This seems to support the suggestion that the microbial biomass contains a pool of easily available nutrients for crop uptake.

The enhancement of N mineralization following a dry-wet cycle has long been recognized as an important phenomenon in the process of N turnover (Birch 1964), although underlying mechanisms are not well understood. The enhanced mineralization is thought to result from the increased availability of organic

substrates due either to chemical changes induced by the dry-rewetting or to the death of microbial cells during drying. Marumoto et al (1982) found that, over a range of soil types, on average 77% of the mineral-N flush, after a wet-dry cycle, was derived from the biomass pool. It has been proposed that the microbial biomass is also a major source of the N released by mild chemical extractants (Jenkinson, 1968; Kelley and Stevenson, 1985). Ammonium released by reflux with 2M KCl has been shown to be well correlated with soil N uptake in spring barley (McTaggart and Smith, 1993), though the identity of the N pool from which it originates is not known.

If microbial biomass is the source of much plant available-N, then reliable indices of N-availability should depend on methods which preferentially extract N from the biomass pool. However, such methods may extract other N pools or combinations of pools. The object of these studies was to relate a number of N release techniques, including well known N availability methods, to biomass-N in a wide range of soils. Three soils were also incubated with a highly enriched ^{15}N-labelled mineral N compound to carry out a more detailed study.

MATERIALS AND METHODS

Three replicate samples of soils (0-30 cm), widely varying in texture, from three sites in southern Scotland (Quixwood, Kettle and Jamesfield) were collected in January 1991 (Table 1) and a further set were collected from seventeen sites in southern Scotland, including those previously sampled in 1991, in February 1992 (Table 2). The soils were sieved to pass a 6 mm mesh and stored in sealed plastic bags at $4^{o}C$ before use. The following soil properties were determined: texture, pH (using a glass/calomel electrode), extractable P (by colorimetry), K (by flame photometry), Mg (by atomic absorption spectrophotometry), organic C (by wet oxidation) and total soil N (by a micro-Kjeldahl procedure).

TABLE 1. Some characteristics of soils (0-30 cm) from 3 sites in S.E. Scotland sampled in January 1991.

Site	Quixwood	Kettle	Jamesfield
pH	6.3	6.7	6.8
P mg l^{-1}	43	56	39
K mg l^{-1}	145	72	146
Mg mg l^{-1}	346	126	186
% organic-C	2.9	1.6	1.3
Total N mg kg^{-1}	1900	1200	1418
% clay	20.9	8.4	15.9
% silt	44.5	12.1	63.2
% fine sand	20.5	48.9	10.6
% coarse sand	14.1	30.6	10.3
Moisture content at sampling g g^{-1}	0.24	0.24	0.17
Moisture content at 8 KPa suction	0.32	0.26	0.28

Soil labelling

High-enrichment (99.2 atom %) $(NH_4)_2SO_4$ was added to the soils sampled in 1991, with a fine mist sprayer. Three replicate samples of each soil were thinly spread on a plastic sheet, and N was added at a rate of about 1 mg ^{15}N kg^{-1} dry soil. The soils were then incubated at room temperature (20-23^{o}C) in sealed plastic bags for seven days, to allow the biomass to become labelled with ^{15}N during mineralization-immobilization turnover (Jansson, 1958). This was the optimal period found by Kelley and Stevenson (1985) to allow incorporation of the added N into the biomass.

Nitrogen availability methods

A number of methods were used to assess N-availability and the size of the biomass-N pool in all the soils collected in 1990 and 1991 as described below. Measurements in each case were made on three replicate subsamples taken from each of the three soil samples.

TABLE 2. Some properties of soils (0-30 cm) from a wide range of sites, sampled in the Borders, Fife and Lothian in February 1992. Mean values given, with SE of the mean in brackets.

	Total Organic-C g kg^{-1}	Total N mg kg-1	Coarse sand %	Fine sand %	Silt %	Clay %
Bush (Cowloan)	42.9	2615	23.4	24.9	29.9	21.7
	(1.45)	(64.3)	(0.38)	(0.37)	(0.50)	(0.68)
Tranent	37.7	1645	32.0	38.6	13.3	16.1
	(0.4)	(15.3)	(2.59)	(2.21)	(0.58)	(0.25)
Quixwood	33.1	2578	15.4	21.5	38.7	24.4
	(0.52)	(52.8)	(0.23)	(0.21)	(0.64)	(0.48)
Treaton	33.1	2534	38.4	32.7	17.7	11.3
	(0.52)	(48.0)	(0.63)	(0.76)	(0.50)	(0.42)
WM East Bank	31.3	2425	24.1	30.2	29.6	16.1
	(1.16)	(55.3)	(1.60)	(0.95)	(1.48)	(0.70)
Bush (Farmers	26.1	1697	26.9	27.4	24.5	21.1
holding)	(0.99)	(16.0)	(0.39)	(0.70)	(0.64)	(0.36)
Bush (No. 3	23.2	1713	47.8	23.8	15.7	12.7
field)	(0.70)	(73.9)	(1.68)	(0.38)	(1.37)	(0.85)
Glencorse	22.0	1649	20.5	21.9	35.0	22.5
	(0.58)	(42.6)	(0.34)	(0.29)	(0.45)	(0.35)
Lintlaw	20.9	1431	16.4	53.6	18.9	11.2
	(1.33)	(79.4)	(1.68)	(2.51)	(0.44)	(0.47)
Greenlaw	18.0	1431	15.9	32.0	31.8	20.3
	(1.51)	(26.6)	(3.54)	(2.32)	(1.02)	(0.98)
Greenknowe	18.0	1247	16.8	36.4	29.5	17.3
	(1.51)	(70.3)	(2.3)	(3.1)	(1.9)	(0.75)
Panlathy	18.0	911	33.0	25.7	27.6	13.7
	(0.70)	(15.3)	(3.31)	(2.72)	(0.85)	(0.54)
Upper Cairnie	16.8	1171	33.4	23.9	24.2	16.7
	(1.22)	(55.0)	(0.68)	(0.90)	(1.65)	(0.72)
Manorhill	16.2	1186	28.7	29.8	27.0	24.4
	(0.52)	(40.3)	(3.11)	(2.51)	(0.52)	(0.48)
Balmano A	14.5	1079	4.9	17.6	58.3	19.2
	(0.41)	(15.3)	(0.15)	(1.99)	(1.78)	(0.23)
Jamesfield	14.5	834	10.2	22.4	50.3	17.1
	(0.17)	(2.0)	(0.76)	(1.73)	(1.89)	(0.36)
Kettle	13.9	834	34.0	44.4	11.9	9.6
	(0.87)	(45.7)	(1.34)	(1.44)	(0.56)	(0.49)

The isotopic enrichment of N released by the different methods in the labelling experiment (1990) was determined by a VG Isogas MM622 mass spectrometer. Samples for mass spectrometry were prepared, after the volume of the extracts had been reduced to 200-300 ml, using a steam distillation method (Hauck, 1982). Where extracts were analysed for ^{15}N enrichment and contained less than 500 lg N, a carrier solution of NH_4Cl (0.3663 atom % ^{15}N) containing 1 mg NH_4^+-N was added to each sample to provide sufficient N for mass spectrometry.

Available mineral nitrogen (MIN-N)

The soils were extracted with 1 M KCl with a wet soil to solution ratio of approximately 1:5. The soils were shaken vigorously for an hour and the extract was then filtered through Whatman 42 filter paper. The concentrations of NH_4^+ and NO_3^- were determined by continuous flow analysis (Best, 1976; Crooke and Simpson, 1970). Extracts for determination of ^{15}N enrichment were distilled for NO_3^- and NH_4^+ in a single distillation.

Microbial biomass nitrogen (BIO-N)

Microbial biomass N was determined using the chloroform fumigation-incubation method (Voroney and Paul 1984). Soil samples were brought to a moisture content equivalent to 8 kPa suction, placed in flasks and exposed to ethanol-free $CHCl_3$ vapour for 24 hours at room temperature in a vacuum oven. After removal of the $CHCl_3$, the flasks were sealed with gas-tight Suba-seals and transferred to an incubator at 25°C. Non-fumigated controls were not included. At the end of the incubation a sample of the headspace gas was analysed for CO_2 by gas chromotography (with labelled soils only), and the soil was extracted with 1 M KCl. The extracts used for the determination of ^{15}N enrichment were distilled for NH_4^+ only, since concentrations of NO_3^- were very low. The N-flush (BIO-N) was calculated as the difference between the mineral-N pool extracted after fumigation-incubation and that present in the unfumigated samples. The CO_2-C flush was calculated on a similar basis. The k_n values were then modified according to the ratio between the C and N flushes, as described by Voroney and Paul (1984) to allow for N immobilization during the incubation.

N released by anaerobic incubation (AN-N)

A modification of the technique of Keeney and Bremner (1966) was used to determine N release by anaerobic incubation. Dry soil (10 g) was weighed into conical flasks and made into a slurry by the addition of 25 ml distilled water. The neck was sealed and the soil slurries were incubated at 40°C for 7 days. At the end of the incubation, ammonium was extracted from the soils by the addition of 25 ml of 2 M KCl. Samples were then shaken and filtered prior to analysis of NH_4^+ by continuous flow analysis. Where samples had been labelled with ^{15}N, the filtrates were steam distilled, and the isotopic enrichment of the NH_4^+-N was determined by mass spectrometry. Nitrogen availability (AN-N) was calculated as the difference between the ammonium pool extracted after anaerobic incubation and that present in the samples extracted for mineral nitrogen.

N released by drying and reincubation (DRY-N)

A modification of a technique suggested by Marumoto et al (1982) was used to determine the amount of N released by drying. Samples of moist soil were weighed into conical flasks and dried at 70°C for 24 hours. They were then rewetted to a tension of -8kPa and reinoculated with c. 1% fresh soil. The flasks were incubated at 25°C for seven days. At the end of the incubation, NH_4^+ and NO_3^- were extracted from the soils with 1 M KCl. Extracts used for the determination of ^{15}N enrichment were distilled for NO_3^- and NH_4^+ together. The N availability (DRY-N) was calculated as the difference between the mineral N pool extracted after drying and reincubation and that present in the fresh soils (MIN-N).

Chemically extracted N (CHEM-N)

A mild chemical extraction techique, based on a method proposed by Gianello and Bremner (1986) and modified by McTaggart and Smith (1993), was employed. Samples of moist soil were weighed into conical flasks, and 2 M KCl was added in a ratio of 7:1 (KCl:soil). The soil slurry was then gently refluxed for 4 hours. Once the extracts had cooled, a volume of distilled water equivalent to the initial volume of KCl was added and the solution was filtered through Whatman No 42 filter paper. Concentrations of NH_4^+ were determined by continuous flow analysis. Extracts for determination of ^{15}N enrichment were

steam distilled for NH_4^+ only. N availability (CHEM-N) was calculated as the difference between the ammonium pool extracted after the extraction and that present initially in the samples extracted for mineral N.

Uptake in pot experiment with ryegrass (RYE-N)

This experiment was carried out with labelled soils in 1991 only. Duplicate pots, 10 cm deep and 12 cm in diameter, were filled with 1 kg of soil from each replicate of labelled soil and 0.25 g ryegrass seed was sown by rough broadcasting. The above ground plant material was harvested after 80 days growth under greenhouse conditions, but without supplementary lighting. The plant material was dried at 100°C overnight and the dry matter yield was determined. The plant samples were then progressively milled to a fine 'flour'. The samples were analysed in a Carlo-Erba 1400 automatic N analyser linked to a VG Isogas MM622 mass spectrometer, which allowed the N concentration and the $^{15}N/^{14}N$ ratio of a sample to be determined in a single analysis. Total N uptake and the amount of ^{15}N uptake were calculated for each soil replicate.

RESULTS AND DISCUSSION

Nitrogen recovered by the extraction techniques

At sampling in 1992, mineral-N concentrations in the soils were very low, and always less than 15 mg kg^{-1} (Table 3). The NH_4^+ concentration was frequently below the detection limit (equivalent to 0.2 mg kg^{-1}) of the continuous flow system. Nitrate concentrations were also relatively low (3.8-13.1 mg N kg^{-1}), but in many soils significant differences ($P<0.05$) occurred between replicates, underlining the high spatial variability of mineral N in the soil (Stockdale et al, 1993). The mineral N concentrations of soils used in the labelling experiment increased during their incubation with $^{15}NH_4^+$. At the end of the 7-day incubation, the mean concentration of mineral-N was 2.3 mg kg^{-1} (Table 4), with the highest ^{15}N enrichment occurring in this pool (Table 5).

TABLE 3. Mean values (with standard errors) of N released by a number of extraction techniques (mg kg^{-1} dry soil).
Tr. = Trace amount, not able to be measured accurately, with a concentration of < 1.0 mg N kg^{-1}.

	MIN-N					
	NO_3^-	NH_4^+	BIO-N	AN-N	DRY-N	CHEM-N
Bush (Cowloan)	10.6	Tr.	49.5	35.1	60.5	37.0
	(0.64)		(2.86)	(0.57)	(5.26)	(0.43)
Tranent	4.0	1.5	25.2	21.3	37.0	30.2
	(0.09)	(0.19)	(0.75)	(0.62)	(0.17)	(0.88)
Quixwood	4.6	Tr.	51.1	42.0	68.1	37.8
	(0.13)		(1.23)	(3.30)	(2.23)	(0.19)
Treaton	6.6	2.0	36.4	47.8	55.0	35.7
	(0.37)	(0.08)	(0.51)	(6.70)	(1.63)	(0.19)
WM East Bank	9.8	Tr.	58.6	65.3	91.7	36.4
	(0.85)		(2.82)	(1.13)	(2.43)	(0.97)
Bush (Farmers holding)	7.3	Tr.	34.6	27.2	54.9	28.8
	(0.14)		(0.58)	(0.92)	(2.43)	(0.44)
Bush (No. 3 field)	13.1	Tr.	48.3	39.0	70.8	28.9
	(0.60)		(3.78)	(2.49)	(3.05)	(0.46)
Glencorse	7.4	Tr.	46.6	32.5	58.1	23.1
	(0.36)		(3.70)	(4.63)	(6.21)	(0.72)
Lintlaw	5.7	Tr.	35.7	26.8	60.6	28.2
	(0.62)		(0.25)	(0.23)	(9.74)	(1.46)
Greenlaw	7.1	Tr.	53.3	39.2	61.8	27.8
	(0.49)		(5.99)	(1.10)	(2.44)	(0.34)
Greenknowe	6.0	Tr.	54.3	43.9	64.5	21.6
	(0.26)		(2.86)	(3.28)	(1.63)	(0.41)
Panlathy	6.1	Tr.	28.1	24.3	34.3	19.7
	(0.30)		(0.61)	(2.53)	(0.92)	(0.36)
Upper Cairnie	3.8	2.0	24.6	21.1	34.7	23.4
	(0.28)	(0.06)	(0.44)	(1.26)	(0.63)	(0.42)
Manorhill	4.6	Tr.	34.2	33.6	68.5	24.9
	(0.16)		(1.74)	(3.95)	(3.02)	(1.06)
Balmano A	7.6	2.5	30.4	22.0	35.9	20.7
	(0.62)	(0.39)	(0.91)	(1.07)	(1.84)	(0.24)
Jamesfield	10.2	Tr.	22.5	15.7	25.4	17.8
	(0.13)		(0.05)	(0.47)	(1.31)	(0.48)
Kettle	9.6	1.9	17.7	25.0	27.6	20.5
	(0.13)	(0.18)	(1.65)	(7.69)	(4.50)	(0.74)

TABLE 4. Mean values (with standard errors) of N released by a number of extraction techniques (mg kg^{-1} dry soil) corrected for mineral N present initially as appropriate. Biomass N was calculated using the k_n adjustment (Voroney and Paul, 1984).
N.d. = Not determined.

	Soils			r values for	
	Quixwood	Kettle	Jamesfield	Total C	Total N
NO_3^--N	22.7 (1.16)	15.0 (0.12)	13.6 (0.47)	N.d.	N.d.
NH_4^+-N	7.3 (0.61)	6.0 (0.32)	4.4 (0.32)	N.d.	N.d.
MIN-N	29.9 (1.69)	20.9 (0.38)	18.1 (0.71)	0.86	0.74
BIO-N	40.8 (0.49)	20.4 (0.49)	20.3 (0.50)	0.97	0.94
AN-N	45.2 (2.28)	23.0 (0.67)	18.5 (0.62)	0.98	0.88
DRY-N	59.9 (2.84)	33.5 (2.85)	22.7 (1.43)	0.98	0.81
CHEM-N	28.3 (1.41)	18.4 (0.54)	18.7 (0.43)	0.92	0.90
RYE-N	126.7 (3.91)	102.7 (6.77)	91.0 (4.38)	0.89	0.72

TABLE 5. The mean atom% ^{15}N in the pools of N extracted by various methods. S.E. of mean in brackets beneath.

	Quixwood	Kettle	Jamesfield
MIN-N	4.857	0.861	2.025
	(0.408)	(0.182)	(0.483)
BIO-N	0.913	0.536	0.758
	(0.018)	(0.087)	(0.002)
AN-N	0.690	0.444	0.568
	(0.036)	(0.024)	(0.012)
DRY-N	1.461	0.645	1.049
	(0.061)	(0.147)	(0.012)
CHEM-N	0.432	0.388	0.393
	(0.026)	(0.006)	(0.002)
RYE-N	0.907	0.484	0.613
	(0.035)	(0.065)	(0.019)

The mean concentration of mineral N in the KCl extracts (in the 17 soils used in the 1992 experiment) was 7.9 mg kg^{-1}. This was consistently lower than the mean concentration of N extracted as BIO-N (38 mg kg^{-1}), AN-N (33 mg kg^{-1}), DRY-N (54 mg kg^{-1}) or CHEM-N (27 mg kg^{-1}). The extracted N was much less variable than mineral-N within soils from the same site, but highly significant differences were found between soils ($\underline{P}<0.01$) for all measurements (Table 3). Most of the N extracted was as NH_4^+. The restoration of a nitrifying population in the soils after fumigation or drying had probably not occurred during the incubation period as has been observed in previous studies (Powlson and Jenkinson, 1976; Marumoto et al, 1982). Drying-reincubation (DRY-N) resulted in a consistently greater release of N than fumigation (BIO-N), as was shown by Marumoto et al (1982), while extraction with hot KCl (CHEM-N) generally resulted in the smallest release of N. In the labelling experiment the mean ^{15}N enrichment of the DRY-N (1.05 atom %) was higher than that of BIO-N (0.75), AN-N (0.57), CHEM-N (0.40) and RYE-N (0.67).

The C flush measured during the fumigation incubation method was very variable within sites, however this was used to correct k_n values in order to calculate the biomass-N pool size (Table 6). The biomass N estimates were within the range quoted by previous authors: 20-50 mg N kg^{-1} dry soil (Ritz and Robinson, 1988), 28-218 mg N kg^{-1} dry soil (Myrold, 1987), 59-144 mg N kg^{-1} dry soil (Bonde et al, 1988) and 35-110 mg N kg^{-1} dry soil (Drury et al, 1991). The occurrence of ^{15}N in the biomass (Table 6) was an indication that immobilization had occurred, with significantly lower amounts of biomass ^{15}N in the Kettle soil (0.07 mg N kg^{-1}) than in those at Quixwood (0.20 mg N kg^{-1}) or Jamesfield (0.17 mg N kg^{-1}). However, the amounts of ^{15}N immobilized were very small and the values may even be overestimates since relatively low values of k_n were used. The small amounts of ^{15}N immobilized by the biomass may have resulted from a lack of carbon substrate in the soil (Kelley and Stevenson, 1985) and/or spatially separated pre-existing and added NH_4^+ pools (Drury et al, 1991), where immobilization may be restricted due to the slow rates of diffusion of NH_4^+ in soils. The model of Drury et al (1991) may well be more appropriate to describe the soil internal N cycle than the non-diffusion limited model of Jansson (1958).

TABLE 6. Assesssment of the ^{15}N in the soil microbial biomass.

	Quixwood	Kettle	Jamesfield
N in extract (mg kg^{-1})	70.7	41.3	38.4
Atom% excess	0.546	0.169	0.391
^{15}N in extract (mg kg^{-1})	0.386	0.070	0.150
Initial $^{15}NH_4^+$ (mg kg^{-1})	0.314	0.046	0.093
N mineralized (mg kg^{-1})	63.4	35.3	34.0
^{15}N mineralized (mg kg^{-1})	0.072	0.024	0.057
k_n	0.360	0.359	0.346
Biomass N (mg kg^{-1})	176.1	98.3	98.3
Biomass ^{15}N (mg kg^{-1})	0.200	0.066	0.165

Relationships between extraction techniques and soil characteristics

Where two extraction techniques are extracting N from only one pool it is to be expected that the actual amounts extracted should be very well correlated. In circumstances where two techniques extract N from the same pool and different pools simultaneously the amounts extracted may still be well correlated, but the gradient of the relationship need not necessarily be unity. In these circumstances, if the pool is also labelled, then it would be anticipated that the ^{15}N enrichment of the different extracts would also be well correlated with a slope close to unity (Myrold, 1987). Some strong linear relationships were observed, when the amounts of N extracted were correlated with each other and with some of the measured soil characteristics (Table 7). All extraction techniques showed highly significant correlations with one another ($r = 0.54$-0.82, $P < 0.01$), although the correlation of CHEM-N with the other techniques was not so strong ($r = 0.54$-0.62). The release of N by anaerobic incubation and fumigation incubation were strongly correlated ($r = 0.91$; $P < 0.001$) with a gradient close to unity, as found by Myrold (1987).

As expected the concentrations of organic-C, total-N and soil texture varied considerably between sites (Table 2), with all extraction techniques showing significant correlations with the concentration of organic-C and TOTAL-N (Table 7). Despite the very highly significant linear correlation ($r = 0.88$, $P < 0.001$) between the concentration of soil organic-C and total soil-N, there were significant differences in the soil C:N ratio between sites. Most of the soils had C:N ratios between 12 and 16, irrespective of organic matter content, but 5 sites with soils of widely varying organic matter contents had higher C:N ratios above 16. The soil C:N ratio showed a significant negative correlations with three of the extraction techniques: BIO-N ($r = -0.52$; $P < 0.001$), AN-N ($r = -0.50$; $P < 0.001$) and DRY-N ($r = -0.53$; $P < 0.001$).

The chemical extraction technique (CHEM-N) showed markedly different correlations with the concentration of soil organic-C, total soil N and soil C:N ratio from the other extraction techniques. However, CHEM-N showed a near perfect linear relationship ($r = 0.95$; $P < 0.001$) with total soil N. Therefore the correlations obtained for CHEM-N with all the other factors are very similar to those for total soil N.

TABLE 7. Correlation coefficients between the amounts of mineral nitrogen (mg kg^{-1}) extracted by various methods and some soil characteristics.
49 degrees of freedom. $P < 0.05$ when $r > 0.276$.

	% Clay	Total C	Total N	Soil C:N	BIO-N	AN-N	DRY-N
O.M.	0.286						
N	0.333	0.876					
C:N	-0.173	0.216	-0.260				
BIO-N	0.421	0.371	0.602	-0.513			
AN-N	0.051	0.383	0.647	-0.495	0.746		
DRY-N	0.172	0.366	0.610	-0.528	0.824	0.797	
CHEM-N	0.215	0.860	0.946	-0.192	0.540	0.616	0.617

Although BIO-N, AN-N and DRY-N were highly correlated with total soil N (with r values between 0.60-0.65), the relationships were much less strong than that with CHEM-N ($r=0.95$). The similarity of the correlation coefficients obtained for BIO-N, AN-N and DRY-N implies that they have been extracted from the same N pool, presumably the microbial biomass. This hypothesis is supported by the work of Myrold (1987) and Marumoto et al (1982), who found that AN-N and DRY-N both included N extracted from the microbial biomass (BIO-N). However, in this experiment it is difficult to confirm this result with reference to the correlations between the ^{15}N enrichments of different extracts (Table 8), since a variable proportion of ^{15}N from the highly labelled mineral N pool is carried over in the extract. Some of the correlations are significant ($r>0.67$), however, none of the relationships have gradients close to unity. Although the correlation coefficients are difficult to interpret, it can be seen that the ^{15}N enrichment of CHEM-N does not correlate well with any of the other pools extracted, again suggesting that this technique extracts N from a different pool or pools.

TABLE 8 Correlation coefficients between the atom% ^{15}N in the pools of N extracted by various methods. 7 degrees of freedom. $\underline{P} < 0.05$ when $r > 0.666$.

	BIO-N	AN-N	DRY-N	CHEM-N	MIN-N
AN-N	0.906				
DRY-N	0.969	0.938			
CHEM-N	0.059	0.059	-0.010		
MIN-N	0.845	0.942	0.876	0.168	
RYE-N	0.927	0.886	0.969	0.066	0.881

BIO-N, AN-N and DRY-N appear to have extracted N mainly from the same N pool. However, the relationship between each of these parameters with the soil's clay content is very different. It is unclear how this could arise unless the techniques are extracting slightly different, though well correlated, pools of N from the soil. It is well established independently that the capacity of a soil to preserve a microbial biomass population is related to texture (Van Veen et al, 1984; Hassink, 1992) and so the significant correlation between BIO-N and clay content ($r = 0.42$; $\underline{P} < 0.01$) is not surprising. Transmission electron micrographs of soil have revealed clusters of microorganisms surrounded by clay particles (Foster and Martin, 1981), producing relatively closed microbial communities possibly protected from predators or harsh and changeable environments.

In the experiment with ryegrass, more N was taken up by the plant (an average of 107 mg N kg^{-1} in the three soils) than was released by any of the extraction techniques (amounts varying between 4-60 mg N kg^{-1})(Table 4). The isotopic enrichment of the ^{15}N taken up in the ryegrass correlated highly significantly with BIO-N, AN-N and DRY-N. However, the interpretation of this data is difficult and no conclusions were drawn about the source of the N taken up by the ryegrass.

CONCLUSIONS

The mild chemical extraction technique (CHEM-N) appeared to extract N from a different pool or pools to that of the other techniques employed. It is possible that the extract is mainly composed of mineral N released from the stabilized and active non-biomass pools of organic nitrogen, since it fails to predict changes in N availability that result from recent additions of organic manures (Fox and Piekielek, 1984; McTaggart and Smith, 1993). This was not a hypothesis supported by Kelley and Stevenson (1985), who suggested that mild extraction procedures are effective in removing newly immobilized N. The success of this technique as an index for predicting soil-N uptake for malting barley (McTaggart and Smith, 1993) may lie in the relatively stable management of long-term index 0 sites (East of Scotland College of Agriculture (ESCA), 1985). Where sites receive the same residue input annually, the differences in the amount of N released by mineralization are attributable to the differences in the basal rate of mineralization from the stabilised and active non-biomass pools of N in the soil. This extraction technique seems to provide an index of this basal rate of mineralization.

The turnover of the soil biomass may provide an important source of N for crop growth. However, there is some evidence that in organic farming systems that the biomass gradually increases throughout the growing season (Ritz, pers. comm.) and this implies that N is supplied to the crops through the action of the biomass in releasing other soil-N fractions, rather than by depletion of the biomass-N pool itself. Under conditions of stress, however, the microbial biomass seems to release a large proportion its nutrients, as cells die and surviving microorganisms mineralize their contents. Wet-dry cycles are a dominant feature of the growing season in many areas and the nutrients released from the biomass after such a cycle seem to be an important component of the N mineralized. The size of the biomass-N pool, as provided by the biological extraction techniques, may therefore be a less useful estimator of potentially available N than the turnover of the biomass pool. Microbial biomass constitutes a variable but significant part of the potentially mineralizable-N pool of the soil. The biomass may act as a sink or source for nutrients depending on the environmental conditions in the soil.

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PAPER IV

NITROGEN SUPPLY FOR ORGANIC CEREAL PRODUCTION IN SCOTLAND

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Abstract

The problems in supplying sufficient N to an organic cereal crop to meet the early N uptake demand of a cereal crop were highlighted. The use of manures containing a greater proportion of N in mineral forms did not seem to give any yield advantage to the crop. However, crop N uptake was closely correlated with the mineral N recovered in the profile after the manure had been applied. This mineral N was recovered more efficiently where poultry manure rather than where farmyard manure had been applied and this was linked to the higher microbiological activity after poultry manure applications.
Leaching was reduced where a cover was maintained on the soil overwinter. However, the use of rye-grass as a cover crop, restricted the yield and N accumulation of a following cereal crop. Leaving the stubble present overwinter was the most suitable overwinter strategy. However, this may cause problems with Take all transmission between crops.

INTRODUCTION

The supply of nitrogen is one of the principal factors limiting organic crop production. Nitrogen is applied to the soil pre-dominantly as animal manures and crop residues, which contain much of their N in organic forms. The release of N in forms available for plant uptake (NH_4^+ and NO_3^-) from these complex materials is effected by the soil micro-organisms. The decomposition of organic materials in soil is controlled by the soil organisms, the environmental conditions under which they function and by the type and variety of the chemical compounds of which the material is composed (Daji, 1934).

There have been a great number of studies into the amounts and timing of of nitrogen released from a wide range of manures and crop residues under laboratory (Chae and Tabatabai, 1986; Chescheir et al, 1986); greenhouse (Rees et al, 1993) and field conditions (Pomares-Garcia and Pratt, 1978; Beauchamp, 1986). Prediction of the release of N has been attempted using empirical (Pratt et al, 1973), statistical-empirical (Barbarika et al, 1985) and computer simulation (Bhat et al, 1980) approaches. The main factors controlling N release from manures have been known at a qualitative level since the 1930's (Jensen, 1931; Daji, 1934). However, quantitative prediction of N release has remained difficult and innaccurate (de Willingen, 1991).

Coupled to the release of nitrogen from manures is the ability of crops to take up and utilise this N efficiently. The demand of crops for N is not constant throughout the season, and one of the main challenges for organic crop production is not just to provide a sufficient quantity of nitrogen for crop growth, but to match the timing of N supply with crop demand. Where this can be achieved, maximum yields will be obtained with minimum losses of N to the environment (Powlson, 1988). The efficiency of the use of mineralised N by crops has been linked to the sowing date and length of the crop season (Sluijsmans and Kolenbrander, 1977). In an organic system, a potato crop used N released from heap-composted FYM more efficiently than spring barley, despite the higher total N uptake of the potato crop (Stockdale et al, 1991) and this was linked to the later sowing and longer period of N demand of the potato crop. Cereals on the other hand require N early in the season and N uptake usually declines after early July.

Scotland can be regarded as having an adverse climate for crop production (Speirs, 1990). The growing season is restricted by cold winter temperatures and summer temperatures are also lower on average than in southern Britain. Cereal growth is generally at a disadvantage, since cereals have a high nutrient demand early in spring when temperatures are still low and mineralisation rates are restricted (Redman et al, 1989), and late harvests also restrict the choice of cover crops that can be used after cereals. Scotland is also significantly wetter than much of southern England. Rainfall in Fife is on average 30% greater than that measured at Bedford. However, when evapotranspiration is considered, excess winter rain may be more than double that in southern England (Speirs, 1990), leading to an increased likelihood of leaching, where NO_3^- remains in the profile after harvest.

Field trials carried out over two seasons in southern Scotland studied the release of N from a range of manures, including heap composted farmyard manure, poultry manure and grass-clover leys, and the efficiency with which the N was used by a following cereal crop. The risk of N losses overwinter by leaching was also assessed and monitored directly after the ploughing-out of grass-clover leys.

SITES, MATERIALS AND METHODS

Trial design and management

Trial 1 Rates and form of manures for spring barley, Jamefield, 1991.

A large trial was carried out to investigate the effect of different manure types and rates on the yield and N uptake pattern of spring barley. The efficiency of use of the inorganic N pool added in the manure was also estimated using ^{15}N. Topsoil properties and previous cropping for the trial are shown in Table 1.

Table 1 Topsoil properties (0 - 30 cm) and previous cropping for the cereal trials. Textures are given using the coding: sand (S), clay (C), silt (Z) and loam (L).

	Trial 1	Trial 2	Trial 3
Year	1991	1992	1993
Crop	Spring barley	Oats	Spring barley
Farm	Jamesfield	Jamesfield	Boghall
Town Grid Ref. No.	Abernethy 204182	Abernethy 198178	Penicuik 248653
Soil series	Carpow/ Carey	Carey	Macmerry/ Duncrahill
Textural group	SCL	S(C)L	SL
Organic matter %	3.1	3.2	8.6
Total N mg kg^{-1}	1500	1600	3400
pH	6.8	6.6	6.3
Previous crop	Potatoes	Spring barley	Grass/clover Spring barley

The experimental treatments (no manure and 3 rates of heap-composted farmyard manure and poultry manure; Table 2) were replicated four times and laid out in a Youden Square design (Cochran and Cox, 1957) with four blocks of seven plots of 18 x 6 m. Manure was applied by hand to the plot surface on the 9th April and this was incorporated by Rotaspike. The trial was drilled with spring barley on the 18th April 1992. Microplots were included in each manured plot, in which the inorganic N pool of the manure was labelled with ^{15}N. A solution of $(^{15}NH_4)_2SO_4$ was applied to the plot surface with a fine mist sprayer, just after manure application, to give ^{15}N enrichments in the inorganic N pool of approximately 13 atom% for the poultry manure and 8 atom% for the farmyard manure.

Table 2 Rates of poultry manure and heap-composted farmyard manure applied to spring barley on April 9th 1992 at Jamesfield Farm.

Treatment	Manure rate t ha^{-1}	Total N kg ha^{-1}	NH_4^+-N kg ha^{-1}
Control	0.0	0.0	0.0
Low FYM	6.5	40.2	1.7
Medium FYM	27.8	172.2	7.2
High FYM	56.5	350.2	14.7
Low PM	2.8	27.8	3.8
Medium PM	13.0	129.6	17.9
High PM	20.4	203.7	28.2

Soil samples were taken in the microplots after drilling (23rd April) to assess the actual ^{15}N enrichment in the mineral N in the soil as seen by the crop. Anaerobic incubations (Keeney, 1982) and an extraction with hot KCl (McTaggart and Smith, 1993) were also carried out on these soils, to determine whether these methods were suitable N availability indices for manured soils. Soil and plant samples in the plots and microplots were taken in conjunction at intervals throughout the season and a plot combine was also used to assess the grain yield at the final harvest.

Trial 2 Cover crops trial, Jamesfield, 1991-1992.

The efficiency of four management strategies in using residual mineral N in the profile was monitored overwinter. Topsoil properties and previous cropping for the trial are shown in Table 1. The treatments were: autumn ploughed and fallow; autumn ploughed with rye grass sown; autumn ploughed with a rye grass - red clover mix sown and stubble left overwinter. Four replicate plots (20 x 6 m) of each treatment were laid out in a Latin Square design in October 1991. Microplots (2.5 x 2 m) were laid out in each treatment. $(^{15}NH_4)_2SO_4$ (99.2 atom %) was applied to the microplots with a fine mist sprayer, at a rate

of 1.66 kg N ha^{-1} to label the residual profile mineral N. Soil samples were taken after application of the ^{15}N, to allow initial ^{15}N enrichments of the profile residual N to be calculated.

The cover crops were ploughed out on 11th March 1992, after a sample had been taken to determine the yield and ^{15}N enrichment of the cover crop in each treatment. The trial was then drilled with oats in early April. The nitrogen uptake pattern of the crop and the efficiency of use of the N contained in the cover crops was monitored by sequential sampling of plant and soil in the plots and microplots throughout the season. A plot combine was also used to assess the grain yield at the final harvest.

Trial 3 Ploughed-out leys, Bush, 1992.

Three old ley trial plots were ploughed out in late February: a 5 year grass-clover ley; a one year grass-clover ley; a 5 year rye-grass ley and these were compared in the trial with a plot under continued arable cultivation. Topsoil properties and previous cropping for the trial are shown in Table 1. Spring barley was drilled on 25th March and soil and plant samples in the plots and microplots were taken in conjunction at intervals throughout the season, to monitor the yield and N uptake of the spring barley crop. After harvest, soil sampling continued and porous cup samplers were installed in the plots on 20th October, with samplers also installed in the continued grass-clover guard area to provide a control. Three porous cups were installed in each plot at a depth of 40 cm. Water samples were withdrawn from the cups weekly following the application of 0.2 bar pressure for 48 hours, to allow leachate concentrations over the winter to be assessed (Grossman and Udluft, 1991). Drainage data from hydrologically isolated plots on the Bush estate (Vinten et al, 1992) were used to calculate leaching losses.

Laboratory analysis

Plant samples, cut to within a few mm of ground level, were oven dried at 100°C and then progressively milled to produce a very fine flour. This was necessary to achieve adequate homogeneity in the very small samples taken for ^{15}N analysis (Robinson and Smith, 1991). Samples were analysed for total N content and the

^{15}N enrichment in a single determination on a Carlo-Erba 1400 automatic N analyser linked to a VG Isogas MM622 mass spectrometer.

Soil samples were taken in the 1 m^2 area from which the plants had been removed for estimation of dry matter. Three cores from this area were taken to a depth of 30 - 35 cm and sealed in plastic bags to prevent moisture loss. Samples were stored at 5°C overnight, where analysis was to occur the next day. Otherwise soil samples were stored frozen (-15°C) until analysis could be carried out. After sieving, available ammonium and nitrate were extracted from the soil using 1 M KCl (Bremner and Keeney, 1966). The extractant was filtered and NH_4^+-N and NO_3^--N determined by continous flow analysis (Crooke and Simpson, 1971; Best, 1976). Extracts were prepared for ^{15}N analysis, where appropriate, by steam distillation (Hauck, 1982)

RESULTS AND DISCUSSION

Yields of 5 t ha^{-1} are possible with organic oats (SAC, 1989), and demand remains high for the grain so that it is a profitable cereal for Scottish organic farmers. Even in the absence of spring manuring yields of oats in the trial in 1992 were close to this expected yield (Table 3). Yields of spring barley grown organically usually lie in the range 3.5 - 5 t ha^{-1} (SAC, unpublished data). Yields in the trial of 1991 were low (Table 3), due in part to the droughty May and June, but yields of spring barley following leys in 1992 lay within this expected range. The N concentration of the grain did not exceed the 1.7 % ceiling for malting, in either of the trials.

Early mineral N for cereals

Poultry manure contains significantly more N in available forms dominantly as NH_4^+ and easily hydrolysed uric acid compounds than the heap composted farmyard manure (Hadas et al, 1983). Very highly significant differences in NO_3^- concentration were seen between treatments after drilling (Trial 1; Figure 1), which were strongly correlated with the amount of N added as NH_4^+ ($r = 0.8$). These differences persisted until early July when NO_3^- concentrations in the soil fell to a very low level (Figure 1), mirroring crop N uptake.

Nitrogen uptake by the spring barley was not significantly affected by the form of manure used, but significant differences ($p < 0.05$) were seen between rates of manure application (Table 4). The low rates of manure application did not increase the N uptake of the barley above the control.

Table 3 Grain yields (t ha^{-1}) @ 85 % dry matter and grain N concentrations (%) for the three cereal trials 1991-1992. Values given are means of four replicate plots

Trial 1 1991 Spring barley	Yield (t ha^{-1} @ 85% DM)	N concentration %
Control	2.4	1.3
Low rate poultry manure	2.2	1.3
Medium rate poultry manure	3.1	1.4
High rate poultry manure	3.1	1.4
Low rate farmyard manure	2.0	1.2
Medium rate farmyard manure	2.8	1.4
High rate farmyard manure	3.1	1.4
Trial 2 1992 Oats		
Bare fallow	4.6	1.5
Stubble	4.7	1.6
Rye-grass	3.9	1.3
Rye/clover	3.9	1.3
Trial 3 1992 Spring barley		
5 yr grass/clover	5.4	1.3
1 yr grass/clover	4.2	1.3
5 yr grass	5.0	1.4
Continuous arable	2.7	1.3

The plots, which had received medium and high rates of manure application, showed increased N uptake, although the difference between the highest measured N uptake and the control was usually the only significant difference. There was a high degree of variability between replicate plots and this may have masked further treatment differences. Recovery of the N from the whole manure was estimated by the difference method and ranged from 5 to 21 % at harvest. Recovery of N from the poultry manure was significantly higher on average (16.1 %) than that from the farmyard manure (7.5 %), and only a little higher than the percentage of manure N present as mineral N, 14 % for the poultry manure and 4 % for the farmyard manure. Recoveries were significantly lower for the high rate of application ($p < 0.05$).

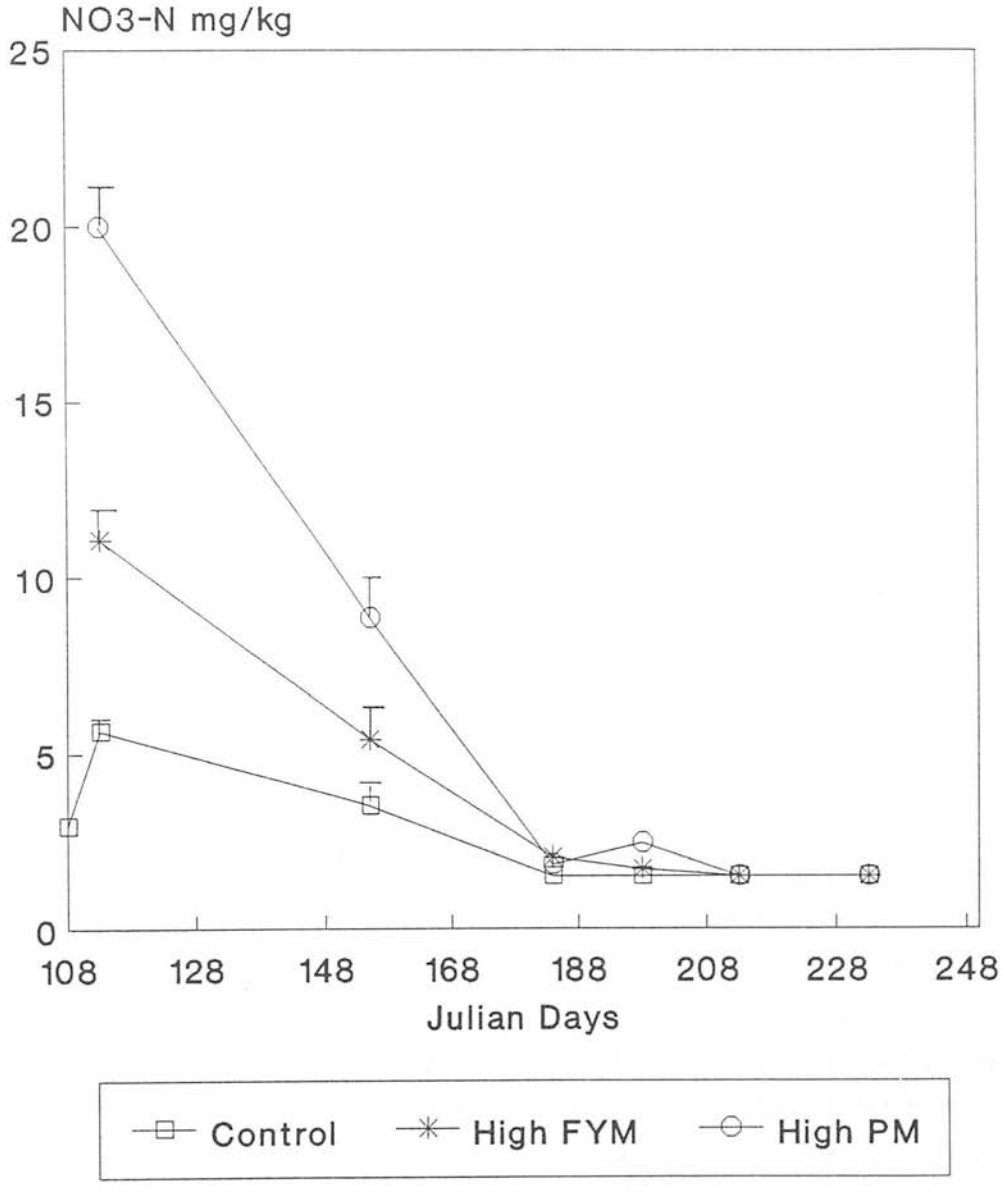

Figure 1 Nitrate concentration in the topsoil (0 -30 cm), where three rates of poultry and farmyard manure had been applied to the soil on April 9th (Day 99). The trial was drilled with spring barley on 18th April (Day 108). All the other treatments lay between the lines shown. The high poultry manure treatment received approximately 11.3 mg N kg^{-1} as NH_4^+; the high farmyard manure treatment approximately 5.9 mg N kg^{-1} as NH_4^+; and the control plots no added N.

Table 4 N uptake of spring barley in Trial 1, kg ha^{-1}, after application of 3 rates of poultry and farmyard manure. Values are means of four replicate plot for each treatment, with the least significant difference between the means for each harvest given.

Treatment	Cut 1	Cut 2	Cut 3	Cut 4
Control	13.48	25.17	24.37	32.33
Poultry manure				
Low	12.90	27.61	29.00	37.26
Medium	33.00	41.89	51.33	59.45
High	29.21	46.17	49.41	52.17
Farmyard manure				
Low	13.24	27.20	30.48	34.36
Medium	27.68	42.39	45.14	53.71
High	29.10	41.91	44.04	49.94
L.S.D.	18.676	18.439	19.468	22.104

Recovery of the ^{15}N labelled mineral N pool of the manure was significantly different between manure types ($p < 0.01$). Recovery reached a maximum in most plots at the third harvest, when on average 103% and 83% of the mineral N added in the poultry and farmyard manure respectively was recovered in the crop. There were no significant differences in recovery between the rates of poultry manure application, but much greater recoveries (approximately 100 %) were measured at the low rate of farmyard manure application than at the higher rates (65.9 % at the third harvest). The very small pool of mineral N added in the low rate of farmyard manure application may have been used more efficiently, or difficulties in determining such a small pool may have lead to overestimates of recovery.

The recovery of the mineral N from poultry manure was larger than that measured for conventional fertiliser, 40 - 50 % (Smith et al, 1984; Powlson et al, 1986; Mary et al, 1988), while that from farmyard manure was similar at harvest. The uptake of ^{15}N from the mineral N pool of the manures (Figure 2) was similar to the pattern seen when ^{15}N labelled fertilisers are used, where

^{15}N uptake is most rapid during stem elongation and then slows, with losses occurring in some seasons between anthesis and harvest (McTaggart, 1992; Recous et al, 1988).

Increased nematode populations were seen in the presence of added poultry manure and the population structure was altered in favour of bacterial-feeding nematodes compared to the farmyard manure treated and control plots (Griffiths et al, 1993). These changes probably indicated that the microbial population was increased by the addition of poultry manure, but not where farmyard manure was added, and this was probably related to the amount of soluble C supplied by the manures. Since the mineral N pool of the manure was closely spatially associated with the available C, this pool was probably the main source of N immobilised by the micro-organisms (Bernal and Kirchmann, 1992). More of the mineral N added in the poultry manure is therefore expected to have been immobilised than that added in farmyard manure. The increased efficiency of uptake of inorganic N from the poultry manure as compared to the farmyard manure, suggests that such immobilisation is beneficial, as immobilised N was probably released for plant uptake as the soil microbes died or were harvested by predators. (Robinson et al, 1989)

Uptake of the N from the mineral N added in the manure was a variable fraction of the total N taken up by the plant (Figure 2) ranging from 6 to 42 % of total plant uptake at harvest. The proportion of plant uptake derived from the mineral N pool of the manures was significantly higher in the plots treated with poultry manure than those receiving farmyard manure ($p < 0.01$), and there was some evidence at harvest, though not earlier, that the proportion of plant uptake derived from the mineral N pool increased with increasing addition of mineral ^{15}N. ^{15}N in the plant declined in most plots between the third and final harvests, probably due to senesence of leaves and some volatilisation losses from the plants (Schorring et al, 1989).

A stepwise regression procedure, including all possible factors, identified NO_3^- measured after drilling as the only significant factor, which explained differences in N uptake by the barley crop ($r = 0.57$). This was probably a better predictor than applied NH_4^+, since it allowed for losses and fixation of ammonium during and after application.

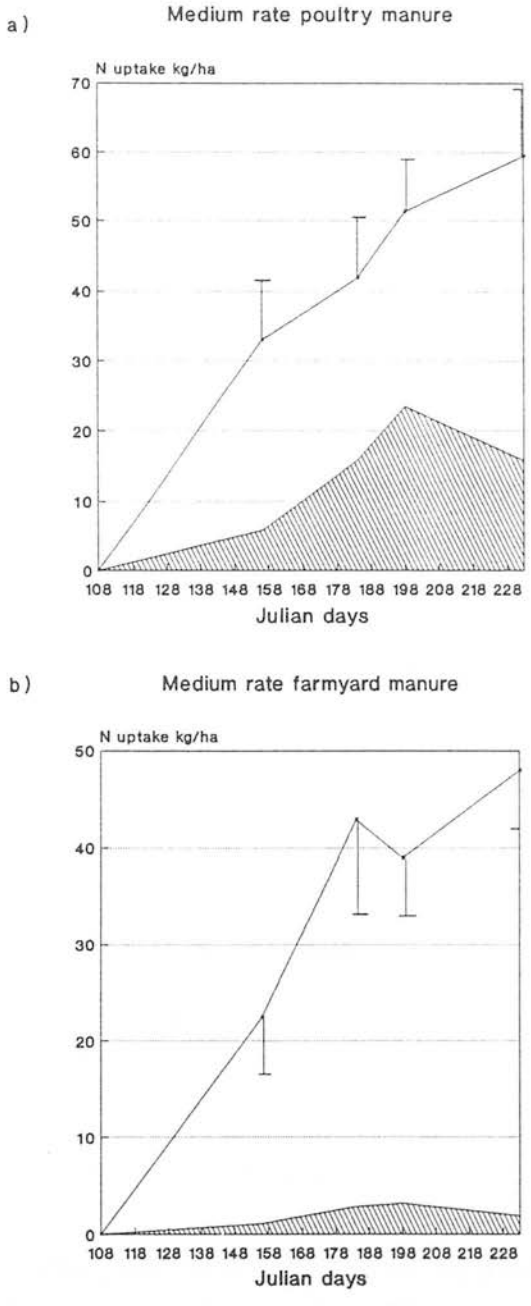

Figure 2 N uptake of a spring barley crop (Trial 1) treated with:
a) medium rate of poultry manure
b) medium rate of farmyard manure.
In each case the total N uptake is represented by the line through the mean of four replicate plots, standard errors of each mean are indicated by the bar. The N uptake from the inorganic N pool of the manure is represented by the shaded area. This was calculated using the static isotope dilution equation using the ^{15}N enrichments of the manure inorganic N pool and the ^{15}N enrichments of the harvested plant material.

The significance of the regression describing the relationship between N uptake and NO_3^- measured after drilling was reduced by the presence of two outliers (Figure 3). The increased soil heterogeneity after manure application causes problems for the representative sampling of soil for mineral nitrogen analysis shortly after incorporation and therefore may give misleading results.

It has been proposed that ammonium N added in the manure is the most important factor controlling the N uptake of the crop (Beauchamp, 1986). However, where large amounts of soluble C are added in the manure, the incorporation of the added mineral N into the soil biomass seem to have rendered it more available for plant uptake over the season than mineral N supplied in conventional fertiliser. This may be due to the prevention of losses of N before the crop roots are fully developed or that N released as a result of microbial death and/or predation is more available for crop uptake.

The incorporation of leys (Trial 3) seemed to provide a large supply of N available for uptake by the spring barley crop, and this trial gave the largest yields of spring barley (Table 3). Dry matter yields following the incorporation of any of the previous leys were very significantly higher ($p < 0.01$) than those from the continued arable plots. As expected, the barley crop following the oldest ley, where most organic matter is expected to have accumulated (Whitehead et al, 1990), showed the highest increase in yield. N uptake following the incorporation of the grass ley continued for longer through the season, than that on the plots following grass-clover leys, although this is most likely to be due to the variability in soil organic matter and moisture content across the site.

Overwinter management and leaching of NO_3^-

The ploughing of the stubble after harvest stimulates the mineralisation of crop residues and soil organic matter (Colbourn, 1985). Increases in NO_3^- (Trial 2) were reduced where a cover crop had been sown after ploughing or the plots had been left in stubble fallow (Figure 4). Leaching losses of residual NO_3^- are therefore thought to have been reduced in the presence of any active cover overwinter (Atallah and Lopez-Real, 1991).

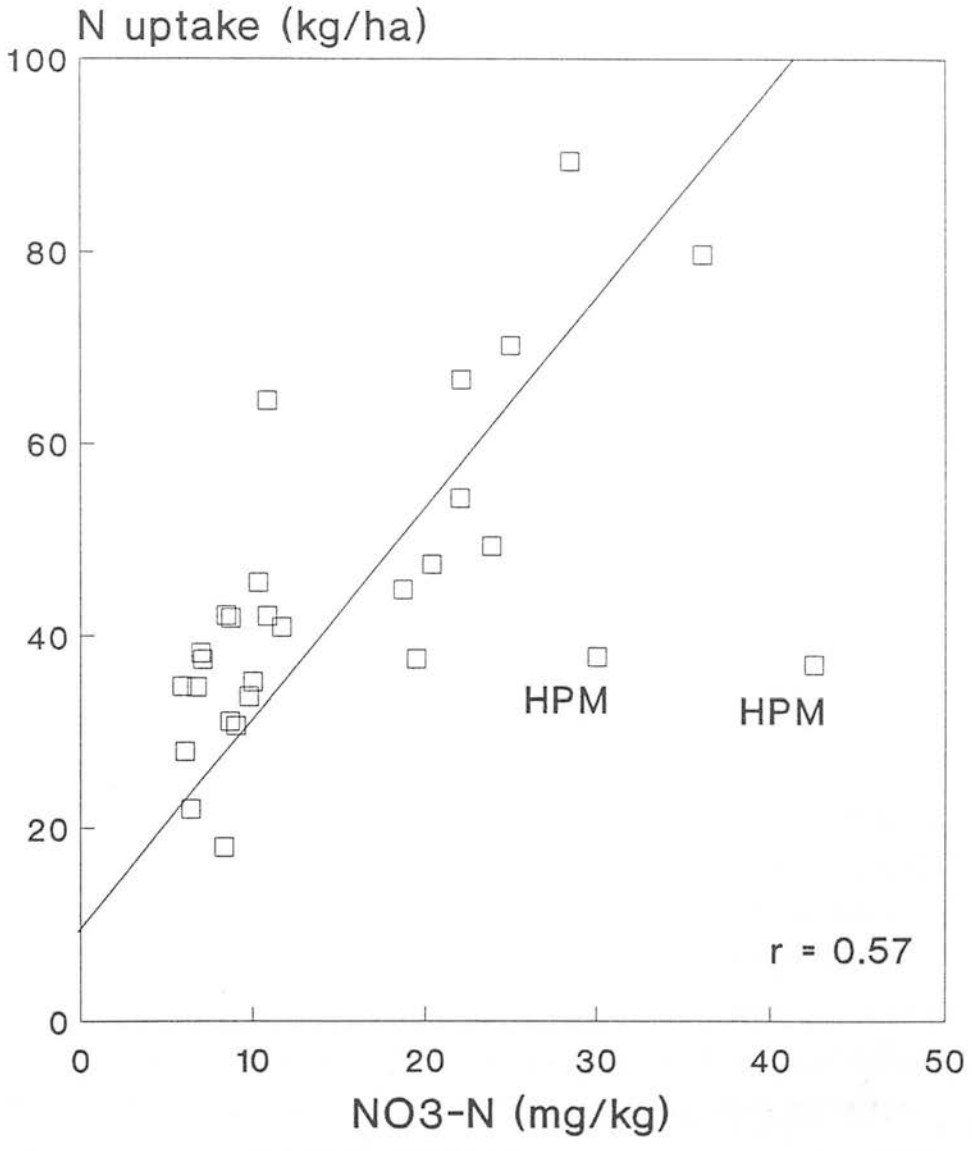

Figure 3 N uptake of the spring barley (Trial 1) at harvest plotted against mineral N present in the soil approximately 2 weeks after manure application and drilling. Outliers may have been caused by non-representative sampling for mineral N, or for the high rate poultry manure plots by continued losses following sampling.

Rye-grass and the rye-grass/clover mixture yielded approximately 1 t ha^{-1} dry matter between October and March, with little clover establishment seen overwinter, while little weed growth was seen on the bare fallow plots (Trial 2). However, the stubble fallow treatment produced the largest dry matter yield overwinter (4 t ha^{-1}) and showed the largest N uptake approximately 75 kg ha^{-1}, compared to only 7 kg ha^{-1} in the bare fallow treatment. The plots with a cover crop or stubble fallow overwinter recovered 24 % of the residual profile mineral N on average, with no significant differences between these treatments. However, the ploughed fallow plot recovered significantly less of the residual profile mineral N (2 %) than any of the plots with an overwinter cover ($p < 0.05$). There was little weed growth after ploughing and the soil surface remained bare throughout the winter

Estimated leaching losses following the ploughing out of leys have varied widely (Cameron and Wild, 1984; Ryden et al, 1984) and are related to the previous seasons management and the age of the ley (Whitehead et al, 1990). Although the ploughed-out 5 year grass-clover ley resulted in the highest losses of N by leaching (Trial 3; Table 5) they were not significantly greater than those from the continued ungrazed grass-clover sward. Since porous cups were installed after the onset of drainage these estimates (Table 5) must be considered as underestimates, though data from soil cores and porous cups suggest that the majority of nitrate loss was accounted for. Organic farming systems rule out the use of herbicide in the crop and therefore strong regrowth was seen beneath the barley crop following the ploughed-out leys. Since the plots were not ploughed in the autumn, regrowth following ploughed-out leys provided a substantial sink for nitrate overwinter and therefore potential leaching from these plots was reduced. This may account for the lower losses of N after the ploughed-out grass compared to the plot under continued arable cultivation, where there was little soil cover overwinter. Leachate nitrate concentrations in all the plots were below the EEC maximum acceptable concentration of 50 mg NO_3 per litre (Tunney, 1992) throughout the sampling period.

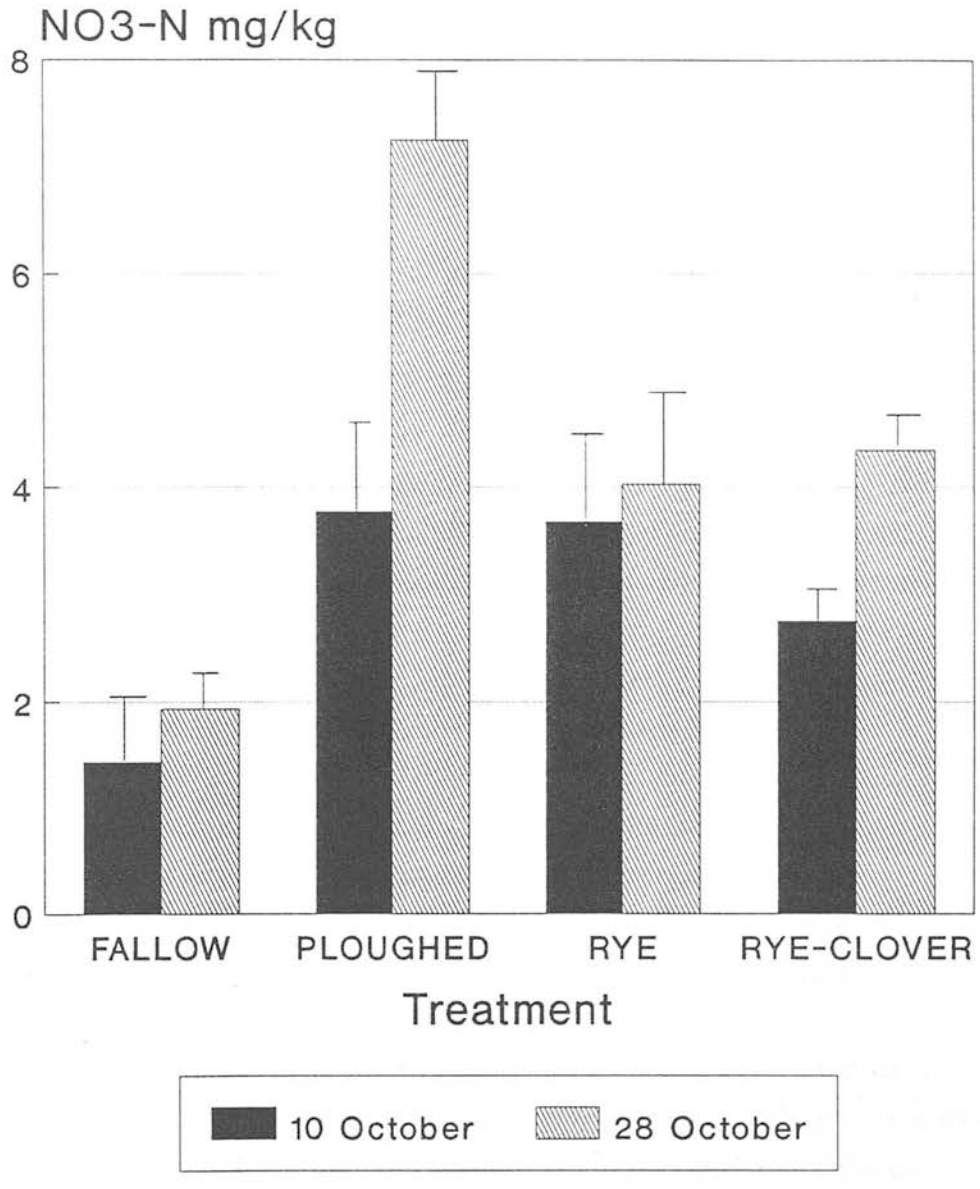

Figure 4 NO_3^- concentration in the soil (0 - 30 cm) in Trial 2 two weeks after ploughing (10th October) and four weeks after ploughing (28th October), when the cover crops had emerged.

Table 5 Mean estimated leaching losses for the period 16th October 1992 to 22nd February 1993 under an ungrazed grass-clover ley compared to three plots cropped with spring barley in 1992, which had followed a 5 year grass-clover ley, a 5 year rye-grass ley and continued arable cultivation. Leaching losses were calculated using leachate concentrations, as measured by porous cup samplers at 40 cm depth, and drainage data from hydrologically isolated plots on the Bush Estate. Standard errors of the means (SE) are also given.

Treatment	Mean leaching loss (kg N ha^{-1})	SE
Continued grass-clover ley	3.1	0.40
5 year grass-clover ley	12.8	4.45
5 year rye-grass ley	6.1	1.40
Continued arable cultivation	10.1	3.12

The use of rye-grass or a rye-clover mix seemed to restrict the N uptake of the following oat crop (Figure 5), with significant differences seen between plots which had remained fallow (bare or stubble) and plots which had had a rye or rye/clover cover. Nitrate concentrations in the plots, which had grown a cover crop overwinter, were significantly lower than the concentrations in the stubble plots for 6 weeks after the cover crops were incorporated ($p < 0.05$). This was thought to be because the C:N ratio of the rye-grass was sufficiently high to immobilise significant quantities of mineral N, as decomposition occurred. The nitrate concentration in these plots would therefore have been significantly lower as the oat crop began to emerge and take up N. Jensen (1991) and Drysdale (1992) also observed that incorporation of an rye-grass cover crop reduced the yield and N accumulation of a following spring barley crop.

Stubble fallowing seems to be the most appropriate, and cheapest, overwinter management technique before a spring cereal crop. However, such practices can lead to the transmission of disease between cereal crops, especially Take all (Scopes and Stables, 1989). Forage rape may be a more suitable crop than rye-grass as an overwinter cover established after the cereal harvest, though trials would have to be carried out to determine its value. In situations where leaching of NO_3^- is not perceived to be important, a winter bare fallow might be the better option.

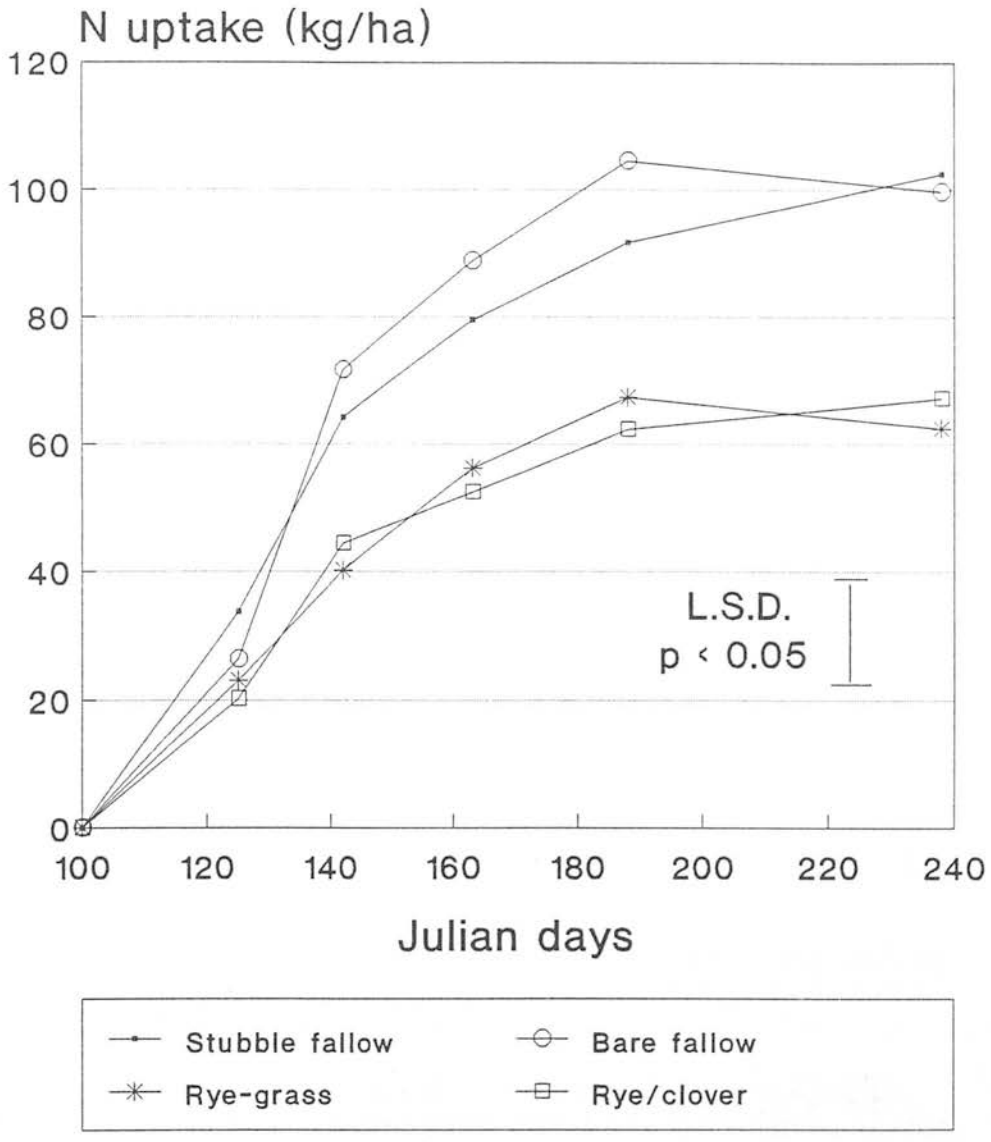

Figure 5 Mean N uptake (kg ha^{-1}) by an oat crop (Trial 2) following various overwinter treatments: bare fallow; stubble fallow; rye-grass and rye/clover cover crop. Each point is the mean of four replicate plots, with the S.E. of the mean indicated by a bar.

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PAPER V

RELEASE OF NITROGEN FROM PLANT AND ANIMAL RESIDUES AND CONSEQUENT PLANT UPTAKE EFFICIENCY

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ABSTRACT

Plant N uptake and mineralisation were studied in a pot experiment using two soils of contrasting textures with a range of manures: poultry manure, cattle slurry, fresh swine manure, composted cattle manure, sewage sludge, straw, cabbage residues, pea residues and grass-clover turf. The manures were labelled with ^{15}N before use, so that crop uptake efficiency could be measured directly and by comparison with control treatments. A range of manure and soil properties were determined before the incubations to examine if any combination of these properties would be useful to predict the amount and timing of N supply. Three replicate pots with and without ryegrass were used. Soil mineral nitrogen and rye grass yield, N uptake and ^{15}N enrichment were measured throughout the 16 week incubation. Volatilisation losses of NH_3 and losses of N as N_2O were also monitored.

The total amount of mineral N produced in pots in the absence of rye grass ranged from 46 to 165 mg N kg^{-1}. N uptake by the rye grass ranged from 17 to 89 mg N kg^{-1}. Volatilisation losses were very small: 0.5 - 12 μg N kg^{-1}; and losses of N as N_2O amounted to approximately 1 % of the mineral N released. Stepwise regression procedures did not produce an acceptable model of the N release based on the results of this experiment. However, it did identify the most important of the measured factors for predicting N release from manures: initial NO_3^-concentrations; C:N ratio and N concentration of the manure. None of the chemical indices tested were found to be valuable for the prediction of N release. Our understanding of the biological processes occurring in soil needs to be much improved, before we are able to model the N release from manures and the consequent uptake efficiency of this N for crop growth adequately.

INTRODUCTION

In organic agriculture, nitrogen is supplied to crops through the addition of plant and animal residues to the soil. Such materials are chemically and physically complex and contain a wide range of macro- and micro-nutrients in addition to N. Although some of the N added in these manures is found in the simple inorganic ions nitrate and ammonium, which are immediately available to plants, most is contained in complex organic forms. Micro-organisms present in the soil break down this material into simpler compounds, part of which they consume for building their body tissues, and the remainder is released as byproducts (Daji, 1934). The additions of crop residues and manures has been shown to increase biomass (Jensen, 1931; Ocio et al, 1991), as well as supplying N for plant growth.

The timing of N supply from manures to crops is as important as the total amount of N supplied and where supply and demand can be closely matched, crop yields can be maximised, while losses of N to the environment are minimised (Powlson, 1988). The amount and timing of N release and consequent plant uptake are dependent on a number of interacting factors, including soil characteristics, manure characteristics, application rates and the population size, structure and activity of soil micro-organisms (Waksman, 1924; Barbarika et al, 1985; Douglas and Magdoff, 1991). Recoveries by plants are usually higher in greenhouse experiments (Rees et al, 1993), but such recoveries have been shown to be well correlated with those under field conditions (Beauchamp, 1986).

The main losses of N from manures are thought to occur within the first two weeks after application (Chescheir et al, 1986). Addition of manures may create conditions favourable to denitrification (Epstein et al, 1978; Hsieh et al, 1981). Ammonia volatilisation will also occur where the ammonium content of the added material is high and the soil is at or near neutral pH. However, rapid and efficient incorporation of manures can minimise losses by volatilisation (Pratt et al, 1976; Beauchamp, 1986).

The following experiment was carried out to allow a more complete study of N transformations in the soil, after additions of manures and crop residues, than had been possible in field experiments (Stockdale et al, 1993). A range of characteristics of the soils and manures was also measured, and three N

availability indices used. The objectives were to determine whether manures have a characteristic N release pattern and whether this can be predicted from the measured manure and soil properties.

MATERIALS AND METHODS

Two soils of contrasting textures (Quixwood, clay loam, Etrick soil series and Kettle, sandy loam, Eckford soil series) were sampled in autumn 1992 (0 - 30 cm). The soils were sieved to pass a 6 mm mesh and stored in large plastic bags at 5 °C until needed.

The animal manures and sewage sludge were collected in September 1992. These manures were aerobically incubated with a mixture of ^{15}N salts [NH_4NO_3, KNO_3, and $(NH_4)_2SO_4$] with an isotope enrichment of approximately 15 atom % for 3 weeks before use. Turf was cut from a grass-clover sward, and peas and winter cabbage grown from seed in peat-sand mixtures. These were placed in the greenhouse and watered with ^{15}N labelled nutrient solution once a week for 12 weeks. Straw was collected from a labelled microplot in the field after harvest, dried and chopped into 1 - 2 cm lengths. The properties of the soils and manures are presented in Table 1.

Samples of the manures, except straw, were taken 4 days before the experiment was to begin and their N content (wet mass basis) was estimated by micro-Kjedahl digestion. The straw was applied at 20 g kg^{-1} soil, since the N content was assumed to be approximately 0.5 %, and the sewage sludge was applied at 50 g sludge wet mass kg^{-1} of soil, since the N content was too low to be determined on a wet mass basis. The application rates of the other materials were calculated using the measured N contents, so that all the pots received approximately 100 mg N kg^{-1} soil. The manures were mixed with the 2 kg portions of the soils by hand, with the cabbage residues being applied only to the Quixwood soil. Samples of the manures were also taken at application. These were dried at 100 °C and progressively milled to a fine flour before analysis for N content and ^{15}N enrichment using a Carlo-Erba 1400 automatic N analyser linked to a VG Isogas MM622 mass spectrometer. Actual application rates are shown in Table 1.

Table 1 Properties of the soils (Quixwood and Kettle) and manures (pea residues, PEA; grass-clover turf, TURF; cabbage residues, CAB; straw, STR; composted farmyard manure, FYM; fresh swine manure, SwM; sewage sludge, SS; cattle slurry, SL; and poultry manure, PM.) used in the pot experiment, and manure application rates

	%DM	As % of D.M. %C	%N	C:N	Excess atom%	Applied N mg kg^{-1}
Quixwood	N.A.	2.9	0.19	15	N.A.	N.A.
Kettle	N.A.	1.6	0.12	13	N.A.	N.A.
PEA	11	29.2	4.13[1]	7	1.056	85.5
TURF	28	15.4	0.71[1]	22	0.607	97.8
CAB	16	15.9	3.15[1]	5	2.871	172.6
STR	100	43.1	1.30[1]	33	0.777	260.0
FYM	13	31.7	2.95[2]	11	2.316	100.3
SwM	13	30.5	2.36[2]	13	2.315	100.1
SS	1	37.1	3.80[1]	10	12.688	18.4
SL	13	37.2	4.46[2]	8	1.046[3]	128.9
PM	49	37.1	3.46[2]	11	3.129[3]	77.5

NOTES

N.A. not applicable

1 % N determined on dried sample by 1400 Carlo-Erba automatic N analyser.

2 % N determined on wet sample by micro-Kjeldahl digestion, as high losses of N result on drying (Giddens and Rao, 1975).

3 Values of ^{15}N enrichment may be underestimates for these manures, since N is lost on drying, which is probably preferentially labelled.

Three replicate pots were filled with 2 kg of the soil-manure mixtures and sown with rye-grass (1 g pot^{-1}). Three further replicate samples of the soil-manure mixtures were placed in plastic buckets (21 cm diameter, 18 cm high) with air-tight lids, to be incubated alongside the sown pots, but remaining unsown throughout the experiment. The Quixwood soil-manure mixtures (500 g) were also placed in Kilner Jars and incubated in the laboratory, so that the N_2O production over the first five days could be studied in greater detail.

The sown and unsown pots were transferred to the greenhouse and incubations carried out for 16 weeks. The temperature ranged from 15 - 20 °C, and 12 hours of artificial light were provided in each 24 hour cycle. The sown pots were watered from the surface every second day and allowed to drain freely. The moisture content in the unsown pots was maintained close to field capacity throughout the incubations. Soil samples were taken from the unsown pots at the beginning of the experiment and after 1, 2, 4, 6, 8, 10, 12, 14 and 16 weeks. Soil samples were also taken from the sown pots after 16 weeks. The soils were extracted with 1 M KCl and the NO_3^--N and NH_4^+-N content determined by continuous flow analysis. Soils sampled at the beginning of the experiment were used to determine N released by anaerobic incubation (Keeney, 1982), N released by an extraction with hot 2 M KCl (McTaggart and Smith, 1993) and C and N recovered after a Walkley-Black wet oxidation (Douglas and Magdoff, 1991). Soils sampled after 16 weeks were used to determine N released by anaerobic incubation and analyses of pH, P, K and Mg levels were also carried out (MAFF, 1986). Mineral N extracted from the soil samples after the anaerobic incubations and also from the soils sampled from the unsown pots after 16 weeks incubation, were analysed for ^{15}N enrichment, after steam distillation (Hauck, 1982).

Plant samples (cut with a scalpel to approx. 0.25 cm above the soil surface) were taken after 2, 4, 8, 12 and 16 weeks. These samples were dried at 100 °C and progressively milled to a fine flour before analysis for N content and ^{15}N enrichment using a Carlo-Erba 1400 automatic N analyser linked to a VG Isogas MM622 mass spectrometer.

Vials containing 50 ml 0.05 M H_2SO_4 were placed in sealed buckets containing the sown pots, and sealed into the buckets containing the unsown replicates for two consecutive periods of 5 days at the beginning of the experiment. The acid was then analysed for absorbed NH_3 using the autoanalyser system slightly

modified to prevent precipation of salicylates in the system, by pre-neutralising the samples with a small amount of 0.4 M NaOH.

N_2O production from the soils was also monitored. Sown pots were placed in plastic buckets (21 cm diameter, 18 cm high) and the lids sealed. The buckets had been modified to give a sampling port in the lids, and after one hour a gas sample was withdrawn for gas chromatographic analysis (Smith and Arah, 1991). In the unsown replicates, the lids were removed for at least an hour to allow the gas concentrations in the buckets to return to atmospheric, the lids were then replaced and samples were taken after an hour. Gas samples from the three treatment replicates were bulked in one syringe. Gas samples were taken immediately after mixing and after 1, 2, 4, 10 and 16 weeks. N_2O production from the Quixwood soil-manure mixtures was measured twice daily for the first five days.

RESULTS AND DISCUSSION

Growth and N uptake of rye-grass

The growth of the rye-grass in the sown pots was initially very rapid, with no significant differences in rye-grass yield between treatments seen in the first four weeks. For the remainder of the experiment, cumulative rye-grass dry matter yield (D.M.) increased approximately linearly, but at a slower rate than over the initial two week period. The N content of the rye-grass harvested varied between 2 % and 6 % N, with lower N concentrations occurring in the later harvests.

Very highly significant differences were observed in N uptake by the rye-grass between treatments, soils and harvests ($p < 0.001$; Table 2). Differences in N uptake between treatments and soils were very much smaller in the first 4 weeks, since much of the N was supplied by the seed, which contained approximately 14 mg N kg^{-1} soil. Cumulative N uptake by the rye-grass increased approximately linearly with time in the period 4 to 16 weeks.

Table 2. Mean nitrogen uptake (mg N kg^{-1} soil) by rye-grass in the pot experiment, calculated from DM yield and N concentration data, standard error of the mean in brackets beneath. Treatments were two soil types (Quixwood and Kettle) with 9 residues or manures: pea residues (PEA); grass-clover turf (TURF); cabbage residues (CAB) on Quixwood soil only; straw (STR); farmyard manure (FYM); swine manure (SwM), sewage sludge (SS); cattle slurry (SL) and poultry manure (PM). The standard error of the difference of the means resulting form an analysis of variance including treatment, soil and cut is also indicated.

Soil	Treatment	Time of harvest (weeks) 4	8	12	16
Quixwood	CONTROL	16.6	10.8	7.3	9.5
		(0.84)	(0.55)	(1.24)	(2.00)
	PEA	18.7	21.9	18.5	14.1
		(1.81)	(5.02)	(4.62)	(3.64)
	TURF	12.6	8.5	10.2	16.3
		(0.96)	(1.01)	(1.63)	(2.37)
	STR	11.8	16.5	22.0	25.5
		(1.25)	(4.02)	(4.24)	(1.26)
	FYM	15.8	15.8	9.7	13.0
		(2.23)	(1.32)	(1.39)	(1.65)
	SwM	11.1	13.6	14.9	14.3
		(0.44)	(3.32)	(1.15)	(3.70)
	SS	16.2	4.2	14.1	10.7
		(2.58)	(0.02)	(3.20)	(4.38)
	SL	20.7	19.6	9.0	9.9
		(0.26)	(1.78)	(0.63)	(2.88)
	PM	21.0	24.5	9.6	12.8
		(3.42)	(3.74)	(0.42)	(0.89)
	CAB	17.4	25.1	15.9	27.5
		(2.52)	(7.06)	(2.21)	(1.46)
Kettle	CONTROL	14.3	3.1	5.1	5.3
		(4.25)	(1.15)	(0.90)	(0.18)
	PEA	10.7	13.5	15.3	20.2
		(2.56)	(4.68)	(4.39)	(3.70)
	TURF	11.4	7.6	4.9	10.6
		(5.40)	(1.03)	(1.52)	(1.82)
	STR	17.4	17.2	10.5	9.0
		(2.98)	(2.52)	(3.61)	(1.84)
	FYM	15.7	14.0	6.3	9.0
		(1.67)	(2.63)	(0.89)	(1.63)
	SwM	18.0	8.1	7.9	10.6
		(2.03)	(2.27)	(1.25)	(4.67)
	SS	14.8	10.0	7.7	5.0
		(1.05)	(0.98)	(1.59)	(0.96)
	SL	17.2	7.7	6.4	3.5
		(2.21)	(2.08)	(1.79)	(0.74)
	PM	19.0	13.4	10.8	10.1
		(2.28)	(2.29)	(1.28)	(0.54)

Standard error of difference of the means = 3.555

Rye-grass grown on the Quixwood soil showed greater cumulative N uptake in the period 4 to 16 weeks, than that on the Kettle soil in all treatments. The effect of the treatments on rye-grass N uptake was not significantly different between the soils (the soil*treatment interaction was not significant). Even where small correlation coefficients were obtained for the linear regression (Kettle soil, control; Table 3), this was caused by variability between replicates not by a significant deviation of the relationship from linearity. Three main groups of treatments were separated (Table 3), by studentised range analysis of total cumulative N uptake of the rye-grass, and these divisions were also reflected in the fitted rate constants, which ranged from 0.16 to 0.79 mg N taken up kg^{-1} soil day^{-1} (Table 3).

Table 3 Correlation coefficients (r^2) and rate constants for straight lines fitted to the relationship between cumulative N uptake by rye-grass and time for the period 4 to 16 weeks of the incubation. Treatments are grouped according to cumulative N uptake over 16 weeks, by studentised range test, following analysis of variance, which showed highly significant differences between treatments. Where treatments are followed by the same letter, then the cumulative N uptake of rye-grass was not significantly different between treatments.

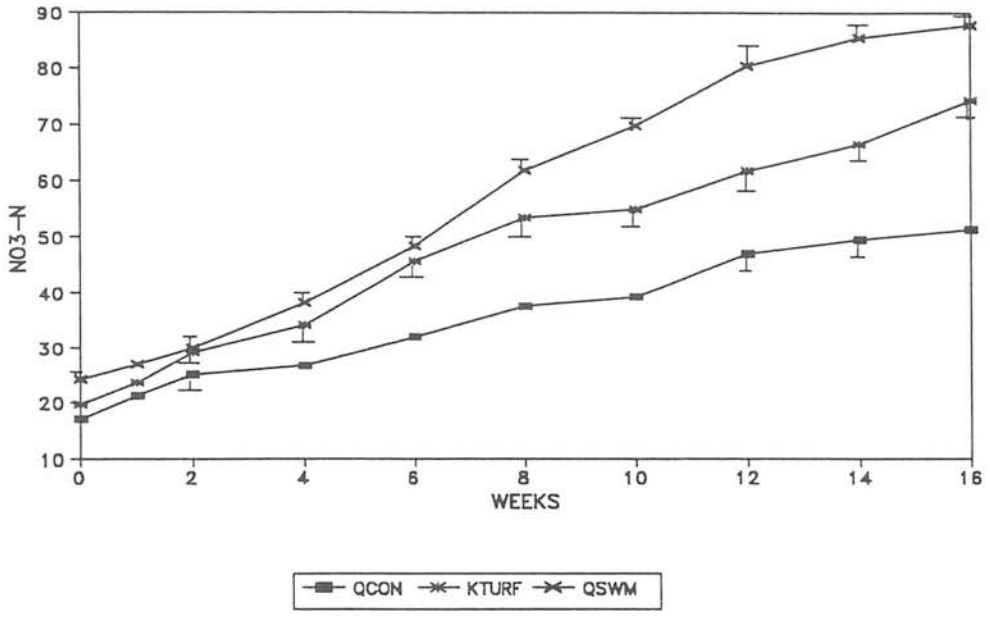
Soil and treatment		fitted rate constant mg N taken up kg^{-1} soil day^{-1}	r^2 %
Quixwood			
Control	a	0.32	91.7
Turf	ab	0.41	95.4
Sewage Sludge	ab	0.36	88.4
Slurry	b	0.44	91.1
Swine manure	b	0.54	93.9
Farmyard manure	b	0.45	92.2
Poultry manure	c	0.79	90.9
Straw	c	0.77	84.2
Pea	c	0.65	75.4
Cabbage	c	0.79	82.5
Kettle			
Control	a	0.16	31.3
Turf	ab	0.29	51.4
Sewage Sludge	ab	0.27	93.3
Slurry	b	0.28	69.9
Swine manure	b	0.33	79.0
Farmyard manure	b	0.39	77.2
Poultry manure	c	0.41	94.3
Straw	c	0.43	62.7
Pea	c	0.58	71.7

Release of N from manures

Soil samples taken from the unsown pots at the beginning of the incubations showed very significantly different ($p < 0.001$) levels of nitrate (NO_3^-) and ammonium (NH_4^+) to the control soils. The addition of turf, farmyard manure, swine manure and sewage sludge did not increase the available mineral N pool in the soil significantly over the controls. However, the other manures supplied significant quantities of mineral N: poultry manure 30 mg N kg^{-1} soil; slurry 40 mg N kg^{-1} soil; pea residues 70 mg N kg^{-1} soil; cabbage residues 90 mg N kg^{-1} soil and straw 160 mg N kg^{-1} soil. The straw contained most of the mineral N in the form of NH_4^+, whereas the other manures and residues dominantly supplied mineral N as NO_3^-. The barley straw used in the experiment had a very high content of mineral N (61.5 % of total N) and a much higher N content (1.3 % N) than had been expected. The soluble N content of straw is usually found in the range 20 - 44 % of total N (Rice, 1979; Christensen, 1986; Ocio et al, 1991). However, as the N content of straw increases, the proportion of the N in straw stored in soluble forms increases rapidly (Reinersten et al, 1984).

The patterns of nitrogen release from manures observed in the unsown pots correspond to those described by Chae and Tabatabai (1986): (i) immobilisation of N during an initial period followed by mineralisation of N later in the experiment (straw; Figure 1d); (ii) rate of release decreasing with time (cabbage residues, sewage sludge; Figure 1b); (iii) a steady linear release with time over the whole incubation period (control soils, turf, FYM, SwM; Figure 1a); (iv) a rapid release of nitrate during the first few days followed by a slower linear release (pea, slurry, PM; Figure 1c). In general, the pattern of treatment differences established at 4 weeks of incubation (control ~ turf ~ FYM ~ SwM ~ SS < PM ~ SL ~ straw < pea ~ cabbage) persisted throughout the incubation. However, the NO_3^--N concentration of the straw treatments declined for the first 6 weeks, followed by a slow linear accumulation of NO_3^--N (Figure 1d). After 16 weeks of incubation their NO_3^--N concentration was not significantly different to that in the controls. As the incubation proceeded significant differences in the NO_3^--N concentrations between the soils became apparent, and at the end of the incubation the treatments on the Quixwood soil contained significantly more NO_3^--N than the treatments on the Kettle soil ($p < 0.01$).

a)



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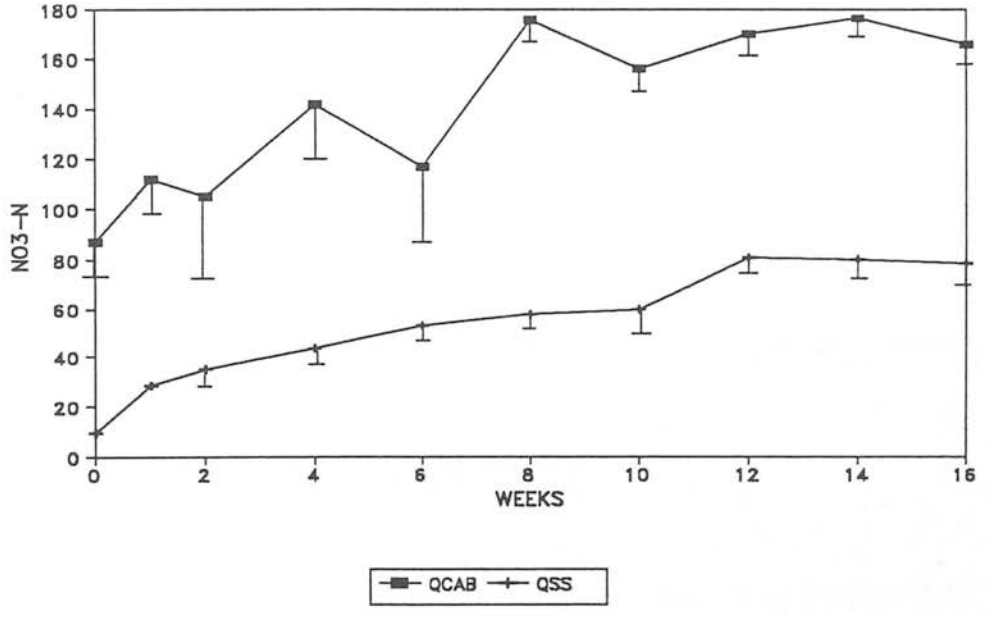
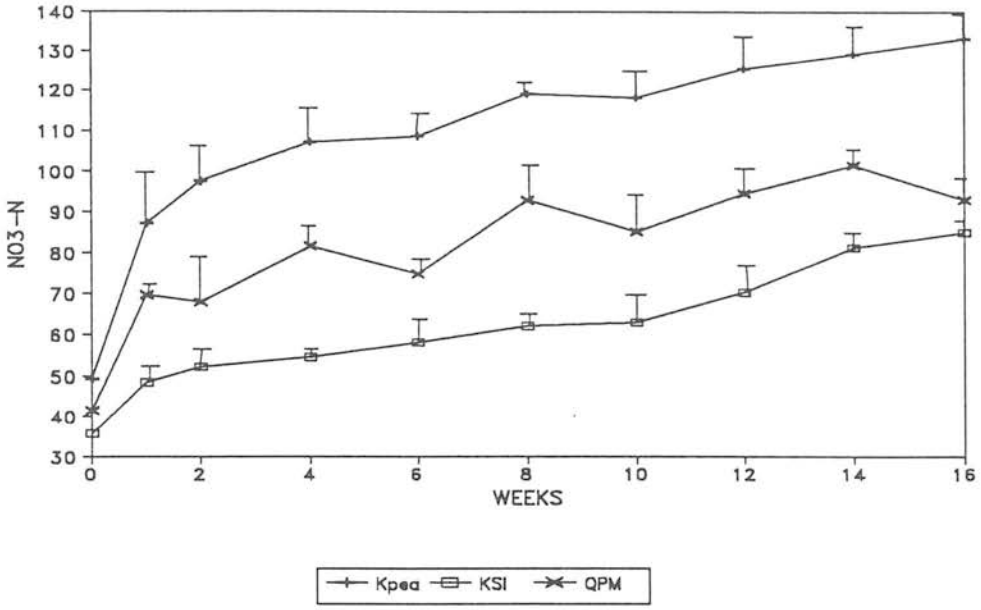


Figure 1

a) Mean NO_3^--N (mg kg^{-1}) accumulating in the unsown pots for the Quixwood control soil (QCON), turf added to the Kettle soil (KTURF) and the swine manure added to the Quixwood soil (QSwM), showing approximately linear NO_3^--N accumulation. Standard error of the mean indicated at each point.

b) Mean NO_3^--N (mg kg^{-1}) accumulating in the unsown pots for the cabbage residues added to the Quixwood soil (QCAB) and the sewage sludge added to the Quixwood soil (QSS), showing curvilinear accumulation of NO_3^--N with time. Standard error of the mean indicated at each point.

c)



d)

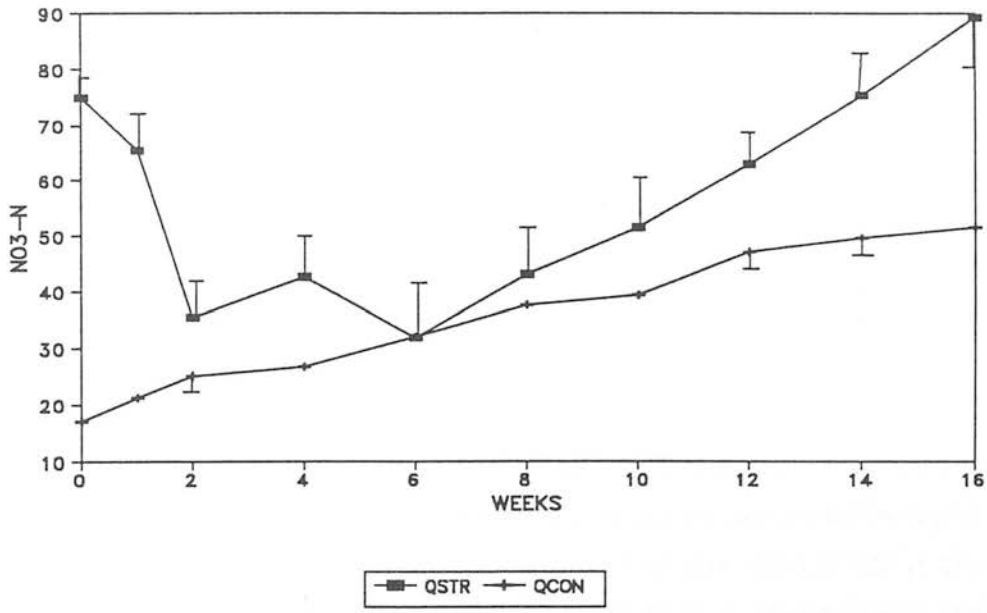


Figure 1

c) Mean NO_3^--N (mg kg^{-1}) accumulating in the unsown pots for the pea residues on the Kettle control soil (KPEA), slurry treatment on the Kettle soil (KSL) and poultry manure treatment on the Quixwood soil (QPM), showing rapid NO_3^--N accumulation initially, followed by a slower linear accumulation. Standard error of the mean indicated at each point.

d) Mean NO_3^--N (mg kg^{-1}) accumulating in the unsown pots for the straw treatment on the Quixwood soil (QSTR) compared to the Quixwood control soil (QCON), showing the decline in NO_3^--N concentration followed by linear accumulation of NO_3^--N with time. Standard error of the mean indicated at each point.

The decline in NO_3^--N during the first six weeks of the incubation in the straw-amended soils was not fully accounted for by increased gaseous losses of N. The addition of straw to soils is usually accompanied by a rapid increase in the biomass (Ocio et al, 1991), with a preferential increase in the fungal over microbial population (Jensen, 1931). Assimilation of NO_3^- by the biomass to meet their N demand may have accounted for the decline in NO_3^- during the first six weeks of the incubation. The turnover of this biomass, once the soluble C of the straw had been exhausted, might also have explained the accumulation of mineral N in the soil after 6 weeks (Jensen, 1931). Due to the very high initial mineral N content of the straw, the mineral N content of the straw-amended soil did not decline below that of the control treatments.

A steady linear accumulation of NO_3^- occurred throughout the incubation of the control soils and the soils treated with turf, farmyard manure and swine manure. None of these treatments contained significant amounts of N as NH_4^+ during the incubations, the rate of NO_3^- accumulation was therefore approximately equivalent to the net rate of mineralisation, since denitrification losses were low. The soils and manures were used moist and so no curvilinearity was introduced into the relationships by the Birch effect (Addiscott, 1983) and a constant rate of net mineralisation may indicate that the substrate was similar to the pre-existing soil organic matter. Chae and Tabatabai (1986) also observed zero order kinetics for NO_3^- accumulation from swine and cattle manure, after a lag period in which no mineralisation occurred.

The rapid release of nitrate during the first few days of the incubations with pea residues, cattle slurry and poultry manure can be partly explained by rapid nitrification of the added NH_4^+. The occurrence of this rapid phase in the decay of poultry manure has been noted by many workers (Castellanos and Pratt, 1981; Hadas et al, 1983; Bitzer and Sims, 1988). However, in each case this phase lasted for various lengths of time (24 hours to 3 weeks) and different reaction kinetics were fitted. The initial rapid release of N from poultry manure is probably due to the hydrolysis of uric acid type compounds in the manure and their subsequent nitrification (Hadas et al, 1983). The slower linear phase of mineralisation, followed once the initial pool of NH_4^+ and easily hydrolysable compounds had been exhausted. The rate of mineralisation of the remaining organic matter became the rate limiting process, and rates of nitrate accumulation slowed.

The N released from the cabbage residues and the sewage sludge tended to a curvilinear relationship rather than a straight line. The decrease in the rate of NO_3^- accumulation with time may have reflected the declining availability of easily mineralisable substrate, and the achievement of almost complete mineralisation of the added material. Rapid and almost complete release of N was observed from cauliflower residues in the field by Rahn et al (1992). First order relationships were also observed for the accumulation of NO_3^--N after the addition of sewage sludge to soils by Hsieh et al (1981), Parker and Sommers (1983) and Lindemann and Cardenas (1984).

Losses

An initial increase in N_2O production, monitored in Kilner jars on the Quixwood soil, was observed during the first two days for all treatments, so that losses of N were significantly higher than those from the control soil ($p < 0.05$). Cumulative losses were still very low, approximately 0.2 mg N kg^{-1} soil in all the treatments, except straw and sewage sludge, which lost 0.54 mg N kg^{-1} and 1.4 mg N kg^{-1} as N_2O respectively. During the 16 week incubations in the greenhouse, losses of N as N_2O were not significantly different between soils or sown and unsown replicates. Measured in this way, only the straw treatment lost significantly more N as N_2O (3.5 mg N kg^{-1} soil over 16 week incubation) than the control pots (1 mg N kg^{-1} soil over 16 weeks).

The addition of the sewage sludge, which had a low percentage solids and contained some active carbon, probably stimulated the formation of anaerobic sites in which denitrification could occur (Epstein et al, 1978; Hsieh et al, 1981). However, the soil only contained a small amount of NO_3^--N and this would have been depleted rapidly. However, the straw-amended soils were dry and well aerated, and it seems likely that a large proportion of the N_2O was released during nitrification (Freney et al, 1979). Anaerobic micro-sites conducive to denitrification may have also been formed, where active carbon leached from the straw was rapidly utilised by micro-organisms.

Straw-amended soils also had the greatest cumulative losses of N by volatilisation over the first 10 days (11 μg N kg^{-1} soil), with most of the N trapped in the acid in the first five days. The other treatments lost very small amounts of N by volatilisation in the range 0.5 μg N kg^{-1} soil to 3 μg N kg^{-1} soil, over the first ten days. The addition of straw is not usually associated with

increased gaseous losses of N, however, the high mineral N content of the straw-amended soils almost certainly explain the increased losses in this experiment.

Recovery of added N

The ^{15}N enrichment of the rye-grass samples were used to quantify the total N uptake from either labelled (manure) or unlabelled (soil) sources. Uptake of labelled N was approximately linear with time (Figure 2a and b), although for some treatments a curve might have been fitted (Figure 2c and d). Over the first four weeks, assuming much of the unlabelled N was supplied by the seed, manure N seems to have been the main pool of N used by the growing rye-grass. After this time, total N uptake, soil N uptake and manure N uptake can all be described approximately by straight line relationships.

The differences in percentage recovery of applied N resulting from the treatments were greater than those related to the method of estimation of the recovery (Table 4). In general, the recovery of N determined by the difference method gave slightly lower values than the percentage recovery determined using the static isotope dilution equation, indicating that some pool substitution of ^{14}N with ^{15}N had probably occurred during the incubations. The recovery of N in the rye-grass was also generally lower than the recovery in the mineral N extracted from the unsown pots. Correcting the N recovered, using the measured losses of N as N_2O to correct for denitrification and/or volatilisation, made a negligible difference to the estimates. The N not recovered in this way was assumed to have remained in the soils. Anaerobic incubations carried out on the soils after the rye-grass was harvested indicated a slight increase in 'active' soil N, where the soils had been treated with straw, swine manure and poultry manure, but there were no significant differences between treatments. Between 5 and 30% of the manure N applied was recovered by the anaerobic incubation after harvest, with a mean recovery of 13.6 %.

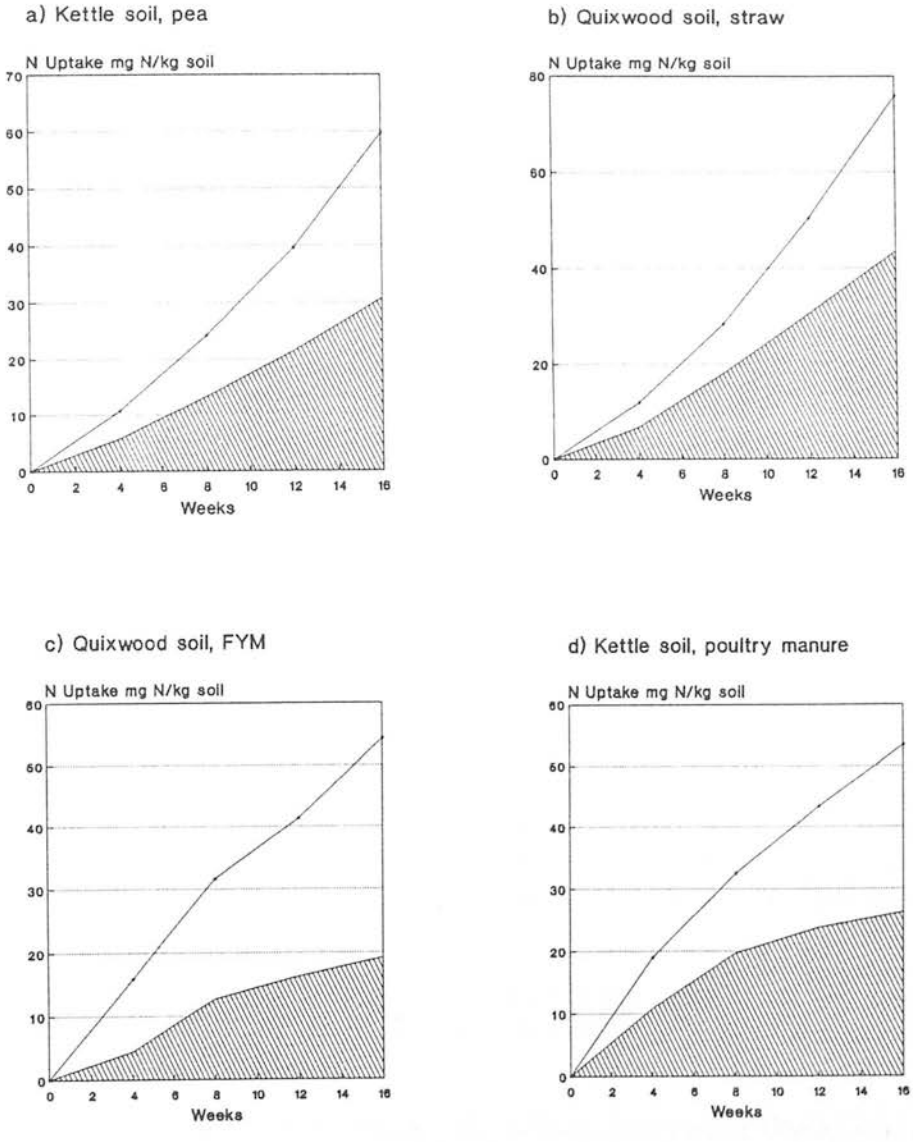


Figure 2 N uptake by the rye grass, partionned into that from labelled (manure) and that from unlabelled (soil) sources. The total N uptake is represented by the area under the line. The uptake of N from manure is shown as the shaded area, the uptake of N from soil is unshaded.

Table 4 Percentage of N applied in manure (pea residues, grass-clover turf, cabbage residues, barley straw, farmyard manure (FYM), swine manure (SwM), sewage sludge, slurry and poultry manure (PM)) recovered in above-ground rye-grass uptake and mineral N accumulated in unsown pots after 16 weeks incubation. Values were calculated from treatment means by the difference method (DIFF)and by static isotope dilution equations (^{15}N).

Treatment	Quixwood DIFF	^{15}N	Kettle DIFF	^{15}N
FROM PLANT UPTAKE				
Pea	33.9	38.8	41.5	37.1
Turf	9.9	18.8	10.0	16.9
Cabbage	24.2	27.1	-	-
Straw	14.0	16.6	10.1	13.8
FYM	10.9	19.1	17.1	20.0
SwM	15.2	20.6	16.8	18.9
Sewage sludge	36.9[1]	60.9[1]	54.3[1]	75.4[1]
Slurry	11.6	ND	6.8	ND
PM	24.2	11.6	32.9	33.8
FROM MINERAL N				
Pea	>100	92.3	>100	97.2
Turf	42.6	47.2	38.7	50.6
Cabbage	66.2	63.9	-	-
Straw	14.5	20.7	9.4	19.2
FYM	34.0	43.3	15.4	36.8
SwM	36.2	40.4	26.0	38.7
Sewage sludge	>100	>100	6.1	84.3
Slurry	46.7	ND[2]	30.4	ND[2]
PM	53.8	71.3	46.7	76.3

Notes

1 No ^{15}N enrichment data was available for rye-grass from the sewage sludge treatments, after 16 weeks. Percentage recovery is therefore calculated for the period 0 - 12 weeks.

2 ^{15}N enrichment determined for the whole slurry after drying was lower than the ^{15}N enrichment determined for the rye-grass harvests and that measured in the mineral N recovered in the unsown pots. Percentage recovery could therefore not be calculated.

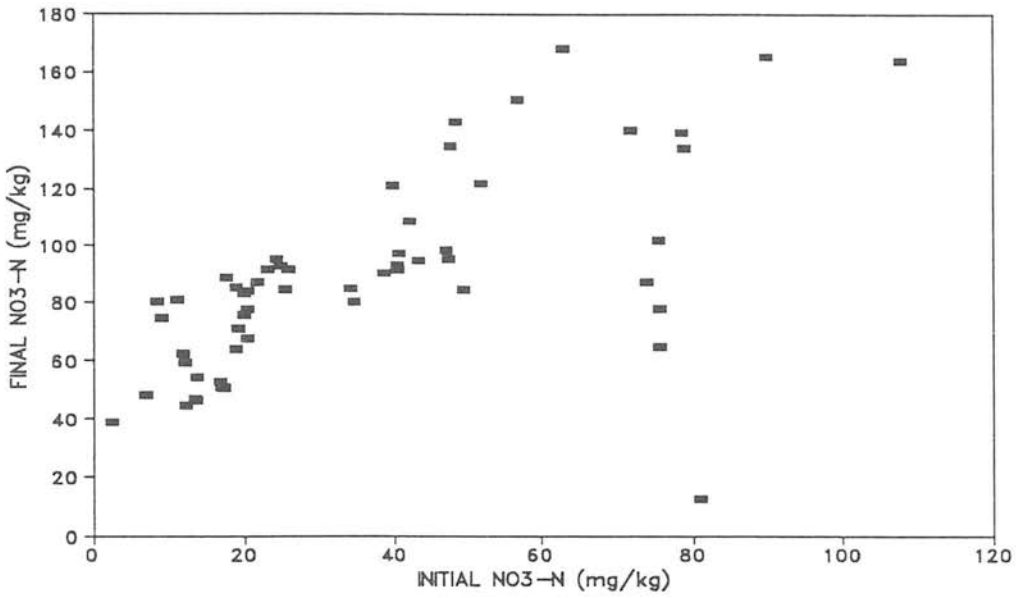
Soil N uptake, measured as the difference between total plant uptake and the N uptake derived from labelled sources, was generally lower than the N uptake of the rye-grass on the control soils. On the Quixwood soil, the N uptake of the rye-grass on the control soil was 31.2 mg kg^{-1}, while soil N uptake on the treated soils ranged from 16.2 - 26.2 mg kg^{-1}. On the Kettle soil, the N uptake of the rye-grass was 14.8 mg kg^{-1}, while soil N uptake on the treated soils ranged from 5.02 - 15.95 mg kg^{-1}. Where pea residues had been mixed with the Kettle soil a positive added N interaction (Jenkinson et al, 1985) was seen, ie. soil N uptake was greater than N uptake in the control soil. The negative added N interaction increased as the C:N ratio of the manure increased. Such an effect is consistent with previous results (Powlson et al, 1985; Rees et al, 1993), and may be due to increased immobilisation in the presence of residues with high carbon contents.

Prediction

N uptake by the rye-grass and nitrate accumulation in the unsown pots were highly significantly correlated (r^2 = 51 %). However, the gradient of the relationship was less than unity. Rye-grass was either not able to recover all of the available N or the N transformations in the soil were altered in the presence of growing plants so that less N was released (Wang and Bakken, 1989). However, the harvest of above-ground plant material only may have led to significant underestimates of the N taken up by the plant (Saffigna, 1988).

Stepwise regression procedures, including a range of soil and manure factors (%N, %C, C:N, application rate) and the indices tested in the experiment, identified initial nitrate levels in the soil-manure mixtures as the most important factor for predicting either cumulative N uptake by the rye-grass (r^2 = 70 %) or total NO_3^- accumulation in the unsown pots (r^2 = 62 %; Figure 3a). The C:N ratio of the manure (-ve coefficient), N concentration of the manure (+ve coefficient) and C concentration of the soil (+ve coefficient) were also introduced at lower levels. However, the correlation coefficient, r^2, never exceeded 73 %.

a)



b)

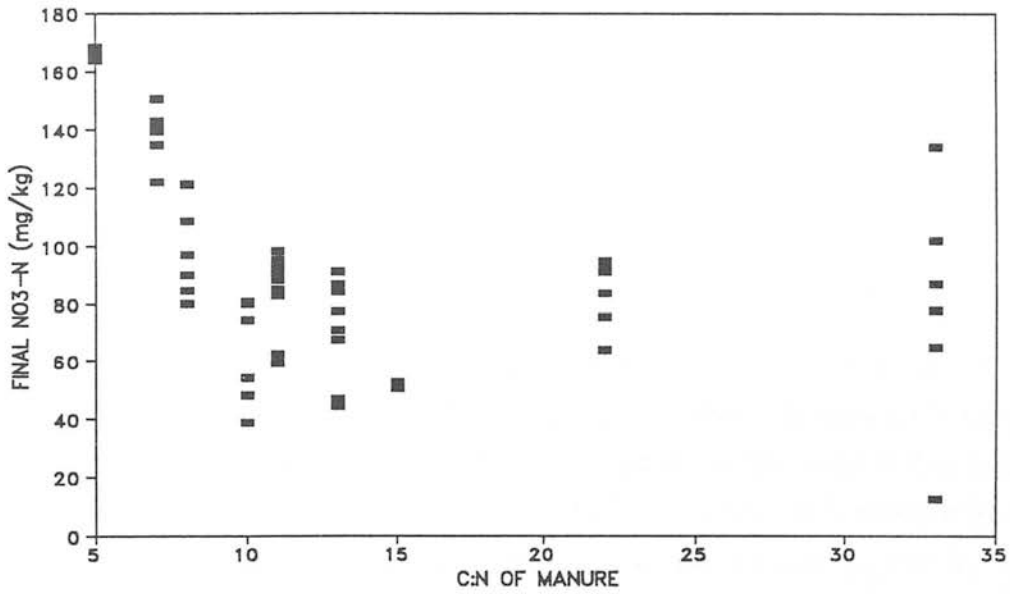


Figure 3 Final NO_3^--N content of the unsown pots plotted against
a) Initial NO_3^--N content (r^2 = 0.389***)

b) C:N ratio of manure (r^2 = 0.106***), however relationship is better described by a curve or two straight lines

The relationship between the C:N ratio of the manure and NO_3^- accumulation could be described by two straight lines (Figure 3b), with the breakpoint for net mineralisation occurring at a C:N ratio of about 15. The relationship between N content of the manures and NO_3^- accumulation was not so clear, and no breakpoint could be identified. The C content of soil was only increased significantly ($p < 0.01$) in the straw-amended soils. None of the chemical indices tested were able to predict N availability from the manures accurately, although the N released during anaerobic incubation ranked the N supply from the manures correctly.

The statistical-empirical approach to the modelling of the release of N from manures, using multiple regressions and curve fitting, did not produce an acceptable model of the N release based on the results of this experiment. Other workers (King, 1984; Barbarika et al, 1985) have found regression equations which describe data sets very well, but which fail to describe the data generated by other groups using different soils or manures. Such an approach identified the most important of the measured factors controlling N release: initial NO_3^- concentrations; manure C:N ratio and N concentration of the manure. Much work has been carried out in the search for a chemical index, which is able to predict N availability from manures (Parker and Sommers, 1983; Chescheir et al, 1986; Serna and Pomares, 1991). However, there has been little success. Many indices, including anaerobic incubation, are able to rank manures for N availability. However, perhaps not surprisingly no index has been found, which is able to match the complex processes leading to N supply from manures. Our understanding of the biological processes occurring in soil needs to be much improved, before we are able to model the N release from manures and the consequent uptake efficiency of this N for crop growth adequately.

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APPENDIX I

THE USE OF BLACK POLYETHYLENE AS A PRE-PLANTING MULCH IN VEGETABLES: ITS EFFECT ON WEEDS, CROP AND SOIL

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ABSTRACT

There is interest in alternative methods of weed control as the availability of herbicides for vegetable crops decline and there is increased interest in alternative reduced pesticide input systems. A series of trials on organic farms is described in which polyethylene is tried as a pre-planting mulch to increase weed emergence and improve the stale seedbed approach to weed control. The use of black polyethylene, laid for 2-8 weeks and lifted prior to planting, prevented weed establishment, and greatly reduced weed growth in the following planted vegetable crop. In comparison weed growth was vigorous in the following crop after use of stale seedbed and clear polyethylene mulch laid for 2-8 weeks prior to planting, with weed removal prior to planting. Crop yield was also improved by the use of the pre-planting black polyethylene mulch. There is evidence that moisture may be less limiting where the black mulch had been used, and nitrate leaching and depletion by weed growth is reduced. However, there is no clear explanation for the reduction in weed growth following use of the black mulch. The technique could be of interest to small-scale growers, but the cost requires repeat use of materials, which will require further machinery and materials development for large-scale use.

INTRODUCTION

As the costs of developing novel herbicide products increase, or re-registration is not sought for older products, it is probable that the range of herbicides available to the vegetable crop grower will diminish. There is also some interest in the development of reduced pesticide input systems of production which use little or no herbicide.

This paper describes a series of trials in 1990-93 on land already converted to organic standards, as part of a research programme on the use of mulches in reduced input systems for vegetables. The series examined whether the traditional stale seed-bed approach to weed control could be improved by encouraging early weed growth with the use of clear polyethylene ground cover, so depleting the seedbank, or masking early weed growth with black polyethylene mulch. The polyethylene was then removed prior to crop planting after any weeds present were removed, to look at the effect on consequent weed and crop growth. Soil conditions under the black polyethylene covers were measured in one of the trials.

METHODS AND MATERIALS

The experiments in 1990 and 1991 were undertaken at the Edinburgh School of Agriculture Organic Farming Centre field site at Jamesfield Farm, Fife in a silty sandy loam over clay loam. The 1993 experiments were at the Sustainable Farming Systems field site at Woodside Farm, Morayshire, on a light sandy loam. Both farms are arable with a long history of vegetable crop production.

Trial A

Calabrese (cv. Corvet) was planted on 28 June 1990 at 20 cm spacing in 60 cm rows following treatments: (i) Standard stale seedbed managed over 8 wk; (ii) Clear polyethylene (150μm) laid for 2, 4 or 8 wk onto the seedbed prior to lifting and then crop planting after any weeds present had been removed; (iii) Black polyethylene (50 μm) laid for 2, 4 or 8 wk onto the seedbed prior to lifting and then crop planting. Plots were 1.8 m beds x 3 m long, with treatments randomised in four replicate blocks.

Trials B and C

Treatments as for Trial A, excluding the clear polyethylene, with calabrese (cv. Shogun) planted on 18 June 1991, and carrot (cv. Nairobi F1) sown on 9 June 1993 at 60 plants/m.

Trials D and E

Treatments as in Trials B/C, plus a stale seedbed approach with extra hand-weeding in the crop, and crops planted through the black polyethylene retained in place. The crops were (D) calabrese (cv. Shogun) and (E) lettuce (cv. Saladin) planted on 2 June 1993, planted as in Trial A.

Trial F

Treatments as in Trial C, with carrots (cv. Nandor) sown at 60 plants/m on 3 July 1991. This trial was established to monitor soil conditions underneath the black polyethylene. The soil was sampled after seedbed preparation for moisture content and soil mineral nitrogen on 22 April 1991. Thereafter soil samples were taken for the first 2 wk then fortnightly until the mulch was removed. Samples were divided for soil moisture determination (drying at 105°C for 24 h) and extraction with 1M KCl to determine nitrate and ammonium by continuous flow analysis (Best, 1976; Crooke and Simpson, 1971). Soil temperature probes (Grant Instruments) were inserted to 100 mm in all plots.

RESULTS

There were few live weeds present in the plots which had been covered by black polyethylene for 2, 4 or 8 wk, except for *Elymus repens* shoots in Trials A and B. In comparison, covering the ground with clear polyethylene in Trial A allowed considerable weed growth with close to complete ground cover after 8 weeks polyethylene cover, as was the case with the uncovered stale seedbed (Table 1). The remaining weeds in the stale seedbed and clear polyethylene plots were then removed prior to planting the crop. Weed re-growth was much greater following the use of clear polyethylene or stale seedbed methods than if black polyethylene had been used (Table 1).

TABLE 1. Effect of pre-planting treatment on weed ground cover (%) at planting (P), and 7-8 wk (7-8 w) or 4 wk (4w) after planting, calabrese (c), lettuce (l) and carrots (r)

Pre-planting treatment	Trial A(c)		B(c)		C(r)		D(c)		E(l)	
	P	7-8w	P	7-8w	P	7-8w	P	7-8w	P	7-8w
Stale s.-b.	100	42	100	75	NA	NA	28	79	30	91
Stale s.-b.	-	-	-	-	NA	NA	28	6	31	11
Black poly. 2wk	3	38	T	21	NA	NA	1	7	1	9
Black poly. 4wk	0	25	T	23	NA	NA	0	6	1	9
Black poly.8wk	T	28	T	23	NA	NA	0	8	0	7
Clear poly. 2wk	47	85	-	-	NA	NA	-	-	-	-
Clear poly. 4wk	60	63	-	-	NA	NA	-	-	-	-
Clear poly. 8wk	96	60	-	-	NA	NA	-	-	-	-
	21 d.f.		12 d.f.							
SED±	5.5	15.3	-	5.8	NA	NA	3.1	2.9	2.0	2.3

s-b. = seedbed; (nw) = plus hand-weeding; poly. = polyethylene; T = trace
NA = data not available when paper was prepared, as trials were ongoing

TABLE 2. Effect of pre-planting treatment on marketable yield (fresh weight kg/plot) of calabrese (c) and lettuce (l)

Pre-planting treatment	Trial A(c)	B (c)	D (c)	E (l)
Stale s.-b.	0.27	0.24	NA	7.91
Stale s.-b.	-	-	NA	6.92
Black poly. 2wk	0.69	1.14	NA	9.76
Black poly. 4wk	1.07	1.49	NA	11.52
Black poly. 8wk	1.25	1.04	NA	9.12
Clear poly. 2wk	0.10	-	NA	-
Clear poly. 4wk	0.13	-	NA	-
Clear poly. 8wk	0.87	-	NA	-
	21 d.f.	12 d.f.		
SED±	0.430	0.301	NA	1.517

s.-b. = seedbed; (nw) = plus hand-weeding; poly. = polyethylene
NA = data not available when paper was prepared, as trials were ongoing

The use of black polyethylene before planting also significantly improved calabrese crop yield over the other techniques; particularly where the polyethylene had remained in place for 4-6 weeks in Trial A (Table 2). The difference in yield over the stale seedbed approach was also noted in Trial B. (Harvest data are not yet available for Trials C/D/E).

The soil moisture status generally fell throughout the first 4 wk in Trial E, as potential evaporation increased (Figure 1). Thereafter, this reversed as rainfall increased. The plots covered with black polyethylene at the beginning of the trial (8 wks cover) showed very significantly higher ($p < 0.01$) moisture content than the uncovered plots for the first 4 wk. A steady rise in moisture content under the polyethylene was noted throughout the whole period of the cover. The plots covered for 4 or 2 wk showed a rapid rise in moisture content after covering.

There was a rapid rise in nitrate during the first 4 wk in the uncovered plots, but then levels fell rapidly to that seen at the start of the trial (Figure 2). The plots covered for 2, 4 and 8 wk also showed a rise in nitrate, but these continued to rise. All the covered plots had, thereafter, higher levels of nitrate then the uncovered plots. There was no difference in ammonium levels between treatments.

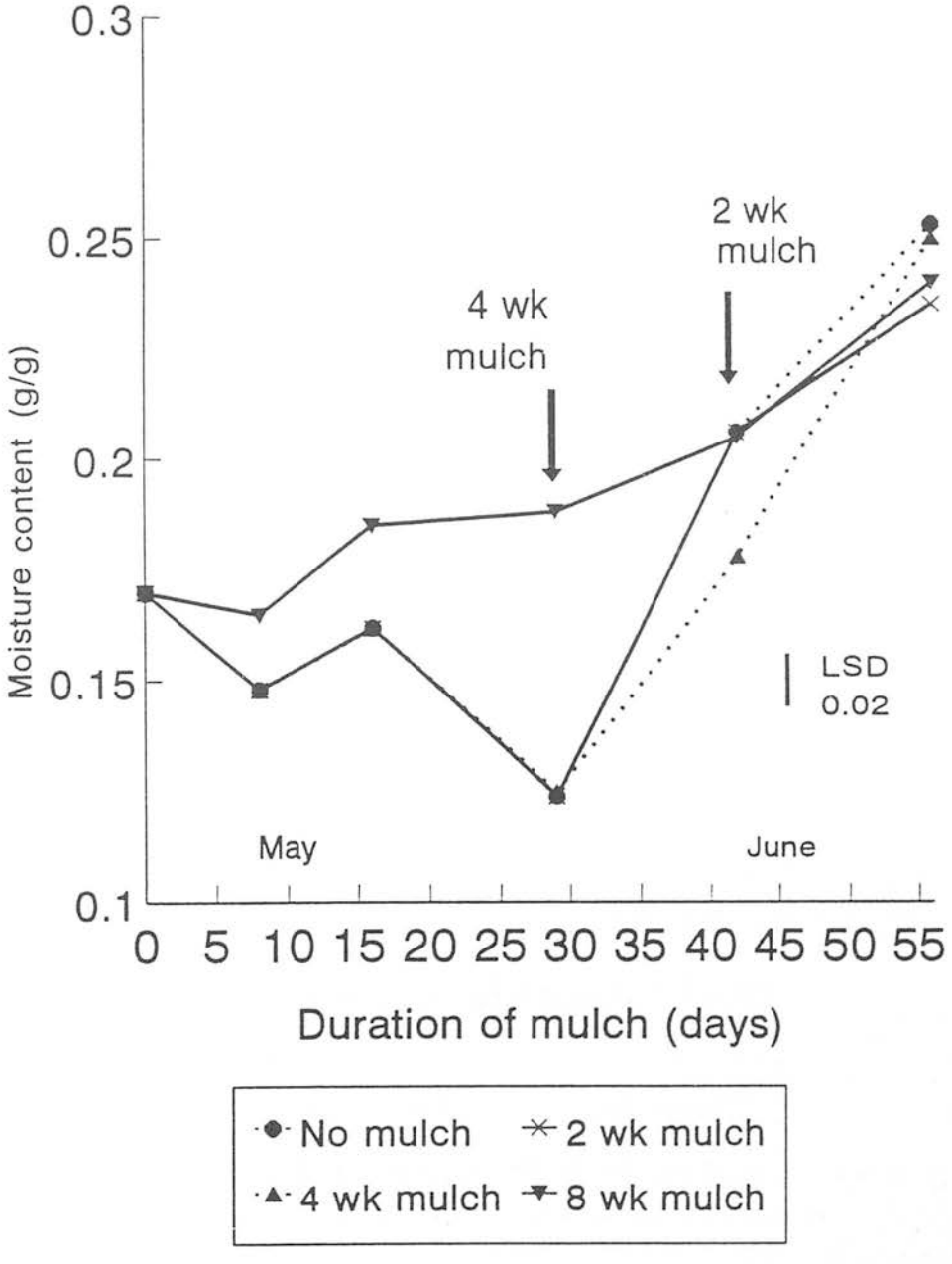

Figure 1 Changes in the gravimetric moisture content of the top 20 cm of soil, either uncovered or covered by a black polyethylene mulch for different periods of time before planting.

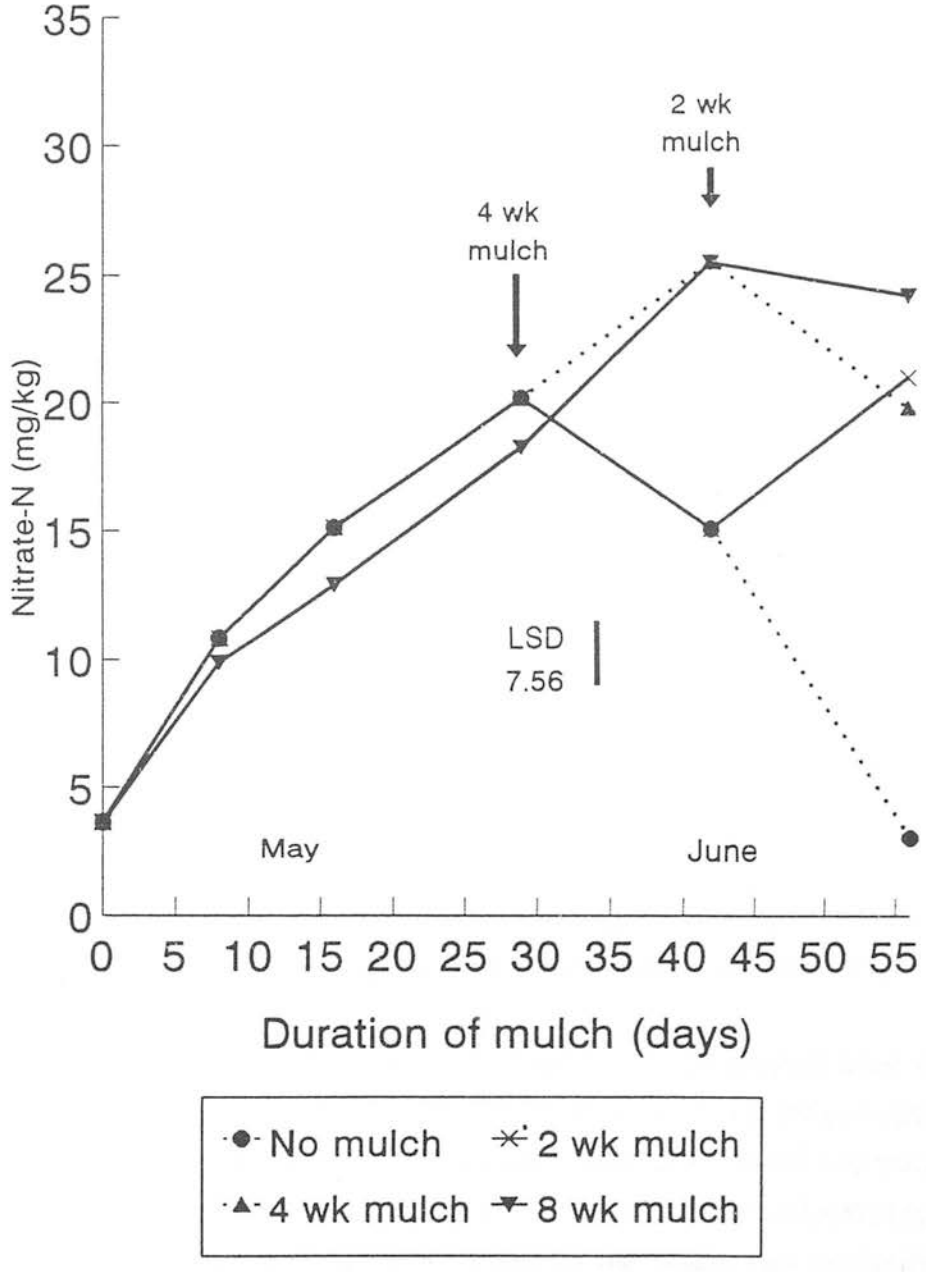

Figure 2 Changes in the nitrate-N concentration of the top 20 cm of soil, either uncovered or covered by a black polyethylene mulch for different periods of time before planting.

Temperature monitoring indicated that temperatures under plastic were higher from the middle of the day than in uncovered plots, but there was little difference in the morning.

DISCUSSION

The consistency of the effect noted in five trials on two differing soil types and sites would indicate that the phenomenon of reduced weed growth following ground cover to prevent light penetration requires further analysis. Of equal interest is the increased crop yield following the use of this technique. In this series, land registered for organic farming was used, where there would normally be limits to available nitrogen and other resources not limiting in other forms of farming. However, the yield responses are marked. Other authors have indicated that the temperature of the ground under black mulches did not consistently increase (Wolfe *et al.* 1992). However, where the soil surface is covered, water loss by evaporation is reduced (Robbins *et al.* 1952). This will leaf to conservation of moisture in the surface horizons of the soil compared to uncovered plots when a dry spell preceded planting. The rapid fall in the nitrate concentration of the uncovered plots after 4 wk coincided with an increase in rainfall and probably resulted from the leaching of nitrate down the profile. This was prevented in covered plots and nitrate will therefore be less limiting in once covered plots where a wet spell preceded planting. Rapid re-emergence of weeds is stale seedbed plots would probably also have reduced available nitrate

However, it is less clear how such conditions may have reduced weed emergence and growth in the following crops following the use of black polyethylene. There was little evidence of high weed germination and death under the polyethylene. Factors associated with weed dormancy, but possible also soil changes such as slight surface compaction from the presence of the mulch may be implicated, but this requires further research.

The practicalities of laying, lifting and re-laying polyethylene mulch limits the usefulness of the technique, except to the small grower, or possibly in experimental situations, until appropriate techniques and machinery have been developed. It is only with such equipment that the economics of use become practical. However, machinery is available for lifting and re-laying especially prepared reinforced plastics which may allow the technique to be used more widely. As the availability

of herbicides and the requisites of customers change, the use of mulches may become of greater interest to the vegetable grower, and the approach described in this paper may have uses for both weed control and improved crop growth.

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