Biogeochemistry **15**: 47—63, 1991

The effect of harvesting intensity on the fate of applied ^{15}N-ammonium to the organic horizons of a coniferous forest in N. Wales

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Received 11 June 1991; accepted 1 October 1991

Key words: ^{15}N-ammonium sulphate, forest soil, nitrification, microbial biomass, clear-felling, Sitka spruce

Abstract. ^{15}N-ammonium sulphate equivalent to 0.5 kg N/ha was added as a tracer to lysimeters containing the organic horizons of an acid forest soil. The effect of logging debris (brash), vegetation and second rotation *Picea sitchensis* seedlings on the amount of the ^{15}N found in various soil, vegetation and leachate pools was followed over a period of 60 days. Transformation of ^{15}N-ammonium to nitrate occurred within 24 hours. Although total nitrate leachate losses were high, tracer-derived nitrate represented only 0.4%—4.2% of the applied ^{15}N-ammonium. The atom % excess of the KCl-extractable organic-N pool was initially lower than for the inorganic species but due to the large pool size, consistently represented 3—6% of the applied ^{15}N-ammonium. The similarity of the atom % excess of the ammonium and nitrate pools indicated an autotrophic nitrification pathway.

A significant proportion of the ^{15}N-ammonium passed through the microbial biomass which contained between 16 and 48% of the ^{15}N-ammonium 2 days after addition of the ^{15}N-ammonium. This nitrogen was in a readily available form or short-term pool for the first two weeks (with no change in the overall biomass pool), after which the nitrogen appeared to become transformed into more stable compounds representing a long-term pool. Total recovery of the ^{15}N was between 68% and 99% for the different treatments. The presence of brash reduced microbial immobilisation of the ^{15}N-ammonium and total retention in the organic matter. This is suggested to be a consequence of greater nitrification and denitrification rate in organic horizons beneath a brash covering due to different microclimatic conditions.

Introduction

Clearfelling of forest ecosystems frequently results in elevated nitrate losses in streamwater (Likens et al. 1970; Wiklander 1981) due to a combination of increased mineralisation and nitrification rates in the forest floor and removal of the plant biomass. The magnitude of these

losses is dependent on the intensity of harvesting and the nitrogen status of the site prior to felling (Vitousek 1984).

Input-output budgets showed that the magnitude of nitrate losses in a Sitka spruce forest in N. Wales was dependent on the presence or absence of logging debris (brash) and the extent of revegetation (Stevens & Hornung 1990; Emmett et al. 1991a, b). A covering of brash increased nitrate leaching by 100% over 1.5 years, whereas reestablishment of ground vegetation in the absence of brash significantly reduced nitrate losses. The differences between treatments may also reflect microbial immobilisation which was found to be a major mechanism for nitrogen retention in the year following clearcutting of a loblolly pine plantation in the southern US (Vitousek & Matson 1984, 1985b).

The internal cycling of nitrogen under these different treatments can be investigated by the use of ^{15}N-ammonium as a tracer. Vitousek & Andariese (1986) demonstrated that following intensive forest site preparation there was reduced immobilisation of nitrogen by the soil microbial biomass and an increase in gross nitrogen mineralisation rates. The tracer can also be used to determine the partitioning of incoming nitrogen between the various soil, vegetation and leachate pools.

In the study described here, ^{15}N-ammonium was added to lysimeters that had been subjected to different brash and vegetation treatments for 1.5 years. The incorporation of the applied ^{15}N-ammonium was followed into KCl extractable microbial and non-microbial soil-N pools, vegetation biomass and lysimeter leachates. Changes in ^{15}N enrichment of these various pools were recorded during the initial 60 days following application of the ^{15}N-ammonium.

Materials and methods

Site

Beddgelert forest was established between 1931 and 1936 on unploughed, unimproved grassland in the uplands of N. Wales (UK) (National Grid Reference SH55550). The plantation is predominantly *Picea sitchensis* (Bong.) Carr. with 10% *Picea abies* (L.) Karst. The soils are predominantly ferric stagnopodzols (Avery 1980) with thin organic horizons consisting of 2 cm Sitka spruce forest floor (L and F horizon) and 4 cm of humified peat (O horizon). More details on general soil characteristics are described by Stevens & Hornung (1988). Conventional and whole-tree harvesting was carried out in the experimental plots in 1983—1984 as described in Stevens & Hornung (1988).

Lysimeters

The experimental lysimeters were established within a clearfelled plot and consisted of zero-tension plastic containers (50 cm × 50 cm × 25 cm) with a base sloping to one corner to improve drainage. The organic horizons were collected from 50 cm × 50 cm quadrats located beneath undisturbed stands of Sitka spruce. Individual horizons were collected and bulked by horizon and carefully introduced into the lysimeters, reconstructing the depth and profile of the original quadrats. The possibility of disturbance stimulating nitrogen mineralisation and/or nitrification was reduced as much as possible by not mixing material from different horizons, avoiding excessive destruction of soil aggregates, and allowing the material to stabilise for 1 month prior to introduction of treatments. Evidence to suggest that no stimulation of soil nitrogen transformations occurred after the initial stabilisation period and more information on the establishment of the lysimeters can be found in Emmett et al. (1991a).

The total dry weight of organic matter introduced into each lysimeter was 1.18 kg (S.E. ± 0.02 kg). Nitrogen concentrations were 1.4%, 2.0% and 2.1% for the litter (L), fermented (F), and humic (O + Ah) horizons respectively. Following the initial stabilisation, treatments were introduced to the lysimeters. These included:

i) the addition of 0.85 kg of fresh logging debris (brash), (the brash used was branches and twigs with a diameter < 2 cm with attached needles),
ii) introduction of 5 small plants of *Agrostis tenuis* (L.) (grass),
iii) the introduction of three 15 month old Sitka spruce seedlings (trees).

Combinations of these treatments were established in a limited factorial design: control (no added treatments), trees, brash, grass, trees + brash, trees + grass. Other treatments established in the original experiment (Emmett et al. 1991a) were not included in the ^{15}N experiment and are not discussed here. All treatments consisted of three replicates.

Leachates were collected fortnightly from the lysimeters from July 1986 to September 1987 and analysed for inorganic and dissolved organic nitrogen and major cations (Emmett et al. 1991a). The ^{15}N tracer was added in October 1987 in a one litre solution as a spray such that each lysimeter received 12.5 mgN equivalent to 0.5 kgN ha^{-1}. In the brash lysimeters the solution was sprayed onto the surface of the organic horizons after careful removal of woody debris. This was replaced immediately afterwards. The sampling strategy was designed to follow the initial movement of the tracer ^{15}N into soil and leachate pools with final destructive sampling of the lysimeters after 60 days.

Leachates were collected 1 and 2 days after addition of the ^{15}N-ammonium. Some leachate was lost during the first two days due to unexpected storms that caused leachate collection bottles to overflow. Sample-splitter tipping buckets were used to obtain a representative sample of leachate over the 14 and 60 day period after ^{15}N application and to record leachate volumes. Studies indicated that no nitrogen transformations occurred in the collection bottles during this period.

On the 2nd and 14th day after ^{15}N application, 3 soil cores were collected from each of the lysimeters using a 6.5 cm-corer. The samples were kept in cold storage during transportation to the laboratory. The 3 cores were bulked and large root systems were removed, although many fine roots remained in the samples. From the remaining soil, 50 g was removed and either fumigated with ethanol free chloroform for 24 h and then extracted with 250 ml 1M KCl or directly extracted with KCl. Extracts were analysed for ammonium-N + organic-N and the difference between fumigated and unfumigated extracted nitrogen used to estimate microbial biomass-N (Brookes et al. 1985). Unfumigated KCl extracts were also analysed for nitrate-N using steam distillation and both fumigated and unfumigated samples were prepared for ^{15}N analysis. The remaining material was oven dried at 70 °C and ground for total-N analysis with prior reduction of nitrate (Pruden et al. 1985).

Sixty days after application of the ^{15}N, the lysimeters were removed from the field and destructively sampled. Above-ground vegetation was harvested. Grass was divided into live and dead components. Sitka seedlings were separated into needles, new woody growth and old woody growth. Below-ground vegetation was mainly collected with the above-ground parts as the organic material fell away easily from the roofs. The remaining roots were collected by direct visual examination of subsamples of the bulked and well-mixed lysimeter organic material using a wet sieving technique and a 2-mm sieve. The vegetation was dried at 70 °C and ground to be analysed for total N with prior reduction of nitrate (Pruden et al. 1985). The organic material in the lysimeters was weighed and samples were taken for KCl extractions pre- and post-chloroform fumigation and dry mass determinations.

Analytical methods

Ammonium and nitrate determinations in lysimeter leachates and KCl extracts

The methodology used during steam distillation was that of O'Deen &

Porter (1980) using an all glass still. In this method, 150 ml of lysimeter leachate or KCl extract was placed in a 500 ml distillation flask, 0.2 g MgO added, and the flask immediately connected to the distillation apparatus. The first 90 ml of distillate were collected in 0.025 M H_2SO_4 (0.75 ml/0.5 mgN) and made up to 100 ml with deionised water. A 10 ml sample was taken for determination for ammonium-N using a Bemas continuous flow autoanalyser and the alkaline phenate-nitroprusside method. Devarda's alloy (0.4 g) was then added to the distillation flask and a further 90 ml of distillate collected and made up to 100 ml. Again, 10 ml was removed for determination of ammonium-N. A spike of natural abundance ammonium sulphate was added to the remaining distillate prior to evaporation on a hot plate at 90 °C such that the sample contained at least 0.5 mgN for the mass spectrometer. When samples were reduced to approximately 1—2 ml, the flasks were removed from the hotplate and the sample washed into glass vials with deionised water. Final evaporation was carried out under an ammonia-scrubbed forced air system. Frequent blanks were carried through the entire process and the distillation apparatus was checked for quantitative recovery of ammonium and nitrate at the beginning of each session. To prevent cross contamination, 10 ml of ethanol was distilled between samples. ^{15}N analysis was carried out on a VG SIRA9 mass spectrometer.

KCL-extracted organic nitrogen determinations

Digestion of 20 ml of extract was carried out in a Taylor flask on a hot plate using a digest mix of H_2O_2/H_2SO_4 containing a selenium catalyst (Allen et al. 1974). The digest was transferred to a distillation flask and attached to the distillation apparatus. Sufficient sodium hydroxide-sodium thiosulphate solution was added to the flask to ensure the digest was strongly alkaline. Approximately 40 ml of distillate was collected in 0.75 ml of 0.025 M H_2SO_4 and made up to 50 ml. 10 ml were removed for determination of ammonium-N. Spikes of natural abundance ammonium suplhate were added and prepared for the mass spectrometer as described above. Organic-N concentrations were calculated as the total nitrogen recovered after digestion minus the ammonium concentration in the original extract.

Total nitrogen determinations in plant and organic horizons

The dried and ground material was digested after reducing oxidised nitrogen compounds using the method of Pruden et al. (1985). Aliquots of 0.4 g of material were weighed into Kjeldahl flaks and 300 mg Zn powder

placed on top of each sample. Then, 4.5 ml of chromic reagent was added, swirled to rewet all the sample and the slurry left overnight. The following day, 2 g K_2SO_4-HgO followed by 9 ml of concentrated H_2SO_4 was added to the flasks which were subsequently heated on a rack until the digests had been clear for 30 minutes. The digest was made up to 100 ml with deionised water and 25 ml of this digest was distilled following the addition of 12 ml of sodium hydroxide-sodium thiosulphate solution. 25 ml of distillate were collected in boric acid indicator and titrated with M/140 HC1 for ammonium-N (Allen et al. 1974). The volume required to obtain a distillate containing 1 mgN was placed in the distillation flask and the distillate collected in 2.25 ml 0.025M H_2SO_4, evaporated and prepared for the mass spectrometer.

Statistical analysis and analytical precision

The coefficient of variation (standard deviation/mean) for the ^{15}N analysis was 0.1% and for the total N analyses < 1%. Differences between treatments were tested by one-way ANOVA with $P < 0.05$. Individual treatments were compared using Fisher's L.S.D. statistic. Arcsin transformation of the data was carried out on percentage data prior to statistical analysis. Atom % excess is defined as the % ^{15}N above natural abundance (0.3663 atom % ^{15}N).

Results

Leachates

The loss of ammonium from the lysimeters in the first leachate following application (Table 1) was higher than previous recorded from the lysimeters presumably as a result of the extra input from the ^{15}N tracer. In addition, the rapid passage of water through the system as a consequence of the high rainfall immediately following the tracer application probably increased the leaching of ammonium. This is reflected in the large losses of tracer-derived ammonium-N in the first leachates. The amounts of tracer lost as leachate-N during Day 1 and 2 should be considered as minimum values because of the loss of leachate from the overflowing bottles.

There were no significant differences between treatments for the loss of tracer-derived ammonium on Day 1. On Day 2 however, there were significant differences between treatments ($F = 4.96$, $P < 0.05$) with ammonium present only in the brash treatment leachates. Soil-derived

Table 1. Leachate losses of soil and tracer derived ammonium-N and nitrate-N (mgN) from the lysimeters following application of the ^{15}N-ammonium tracer and the percentage of the tracer leached.

Treatment	Days	Ammonium-N			Nitrate-N		
		Soil	Tracer	% Tracer	Soil	Tracer	% Tracer
Control	1	0.6	2.3	18.6	8.9	0.4	3.3
	2	0.3	0.0	0.0	3.7	0.0	0.0
	14	0.4	0.0	0.0	2.2	0.0	0.1
	60	0.0	ND	ND	5.7	0.0	0.0
Brash	1	3.6	2.6	20.7	23.4	0.4	3.2
	2	6.1	0.1	1.1	8.5	0.2	0.0
	14	3.0	0.0	0.1	7.6	0.1	0.7
	60	1.5	0.0	0.0	15.7	0.0	0.3
Grass	1	0.7	1.3	10.1	1.0	0.1	0.6
	2	0.5	0.0	0.0	0.6	0.0	0.0
	14	0.0	ND	ND	1.7	0.0	0.0
	60	0.0	ND	ND	1.9	ND	ND
Trees	1	0.3	1.8	14.7	0.7	0.1	0.9
	2	0.3	0.0	0.0	1.4	0.1	0.0
	14	0.4	ND	ND	0.5	0.0	0.0
	60	0.0	ND	ND	3.2	ND	ND
Trees + brash	1	1.0	1.8	14.3	8.5	0.3	2.4
	2	0.8	0.0	0.2	3.4	0.1	0.0
	14	0.2	ND	ND	3.0	0.0	0.1
	60	0.0	ND	ND	9.1	ND	ND
Trees + grass	1	0.4	1.5	12.3	0.7	0.1	0.4
	2	0.3	0.0	0.0	0.4	0.0	0.0
	14	0.4	ND	ND	0.9	0.0	0.0
	60	0.0	ND	ND	0.4	ND	ND

ND — not determined
Values represent the total nitrogen leached between sampling days. See text for details.

ammonium was also higher in these treatments. On day 14 and 60 ammonium concentrations in the leachates were below detection limits.

Within 24 hours, there were detectable amounts of tracer-derived nitrate in the leachates from all treatments. Both total leachate losses of nitrate ($F = 8.53$, $P < 0.01$) and tracer-derived nitrate ($F = 3.98$, $P < 0.05$) were significantly different between treatments. Tracer and soil-derived nitrate were greater from the control, brash and tree + brash treatment relative to the remaining vegetated treatments.

The loss of soil-derived nitrate was consistently higher from the brash-

treatments (brash and tree + brash) leachates compared to the other treatments. Tracer-derived nitrate generally fell below detection limits between Day 2 and 60, with the exception of the brash treatment which lost 1% of the tracer as nitrate during this period.

Cumulative inorganic-N leachate losses of labelled nitrogen ranged from 12.7% from the tree + grass treatment to 26.1% from the brash treatment.

KCl extracts

The amount of tracer in the form of ammonium two days after application was 1—2% with the exception of the brash treatment, which had 8.1% in the form of ammonium ($F = 11.99$, $P < 0.001$) (Table 2). There was significantly less in the form of nitrate with greater tracer-derived nitrate from the brash treatment (2.3%) and control treatment (1.5%) relative to the vegetated treatments ($F = 27.93$, $P < 0.001$). A significant proportion of the added tracer was recovered as KCl-extractable organic-N

Table 2. The percentage of ^{15}N in the KC1 extractable ammonium, nitrate and organic-N pools, 2, 14 and 60 days after application of the tracer to the lysimeters.

Treatment	Days	Ammonium-N μg/g	% Tracer	Nitrate-N μg/g	% Tracer	Org-N μg/g	% Tracer
Control	2	27.1	1.6	19.9	1.5	195.0	3.1
	14	28.6	0.7	13.4	0.2	225.3	3.3
	60	54.4	1.1	34.0	0.6	232.1	3.1
Brash	2	59.0	8.1	20.2	2.3	233.4	3.1
	14	46.0	0.8	22.5	0.3	247.2	3.0
	60	159.4	2.1	45.6	3.5	188.5	2.1
Grass	2	28.6	1.5	10.6	0.3	269.5	4.7
	14	30.5	0.7	10.5	0.1	246.9	4.3
	60	195.0	3.4	14.4	0.2	236.2	2.6
Trees	2	22.4	1.3	12.3	0.3	233.0	3.9
	14	28.3	0.8	10.2	0.1	243.5	4.6
	60	47.0	0.8	29.1	0.4	179.0	2.6
Trees +	2	38.3	2.7	19.9	0.7	262.5	4.5
brash	14	39.5	1.0	15.8	0.2	243.7	4.3
	60	153.6	2.4	32.5	0.5	88.2	
Trees +	2	16.3	0.9	9.4	0.2	204.6	3.8
grass	14	32.7	1.0	10.8	0.2	311.5	6.0
	60	124.5	1.8	14.5	0.2	186.9	1.9

(3.1—4.7%). In the control and brash treatments the atom % excess (% ^{15}N above natural abundance) of this fraction (0.14—0.24) was significantly lower than that of the inorganic fractions (0.25—1.41), but the total pool size much larger. Atom % excess of the ammonium and nitrate pools were similar, for example in the control treatment on Day 2, atom % excess of ammonium, nitrate and organic-N was 0.70, 0.92 and 0.19, respectively. Atom % excess of the inorganic-N pools was generally lower in the lysimeters containing vegetation relative to the control and brash treatment.

After 14 days, atom % excess had fallen in all treatments for nitrate-N and ammonium-N (< 0.35), but remained the same for organic-N. The amount of tracer N in KCl extractable form was below 1% for all treatments. There was a further fall in the ^{15}N enrichment of the KCl extractable ammonium-N pool after a further 6 weeks. The enrichment of the nitrate and organic-N generally remained unchanged. The amount of the tracer nitrogen in the combined ammonium and nitrate pools was below 6% for all treatments and greatest in the brash treatment (5.6%). KCl extractable organic-N represented 1.9—3.1% of the labelled-N with atom % excess similar to the inorganic-N pools at the time of the 60 day sampling.

Microbial biomass-N

Assuming a K_N value of 0.54 (Brookes et al. 1985), the microbial population in the control treatment contained 37%, 43% and 48% of the added nitrogen in the control, trees and trees + grass treatments respectively, two days after application of the tracer (Table 3). The percentage of tracer in the microbial population in the brash treatments, was lower with 16% in both the brash and trees + brash treatment. The values, however, are highly variable between the three replicates for all treatments and there were no significant differences between individual treatments. If the treatments were regrouped according to the presence or absence of brash, grass or trees, brash significantly reduced incorporation of the tracer nitrogen into microbial biomass for all three sampling times, ($F = 9.87$, $F = 7.41$, $F = 11.4$, $P < 0.05$ for 1, 14 and 60 day sampling time respectively). Grass and trees had no significant effect.

After 14 days, the amount of tracer-N found as microbial-N had fallen to $\approx$ 50% of the previous values with no significant change in the overall size of the microbial-N pools. After 60 days, there was a reduction in the percentage of tracer held in the microbial pool relative to the measurements after 14 days in the vegetated treatments but no change in the non-vegetated treatments.

Table 3. The percentage of the ^{15}N-ammonium tracer incorporated into the soil microbial biomass pool assuming a K_N factor of 0.54.

Treatment	Days	N released ugN/g	% Tracer	% Tracer as microbial biomass-N
Control	2	355	20.1	37.2 (1.4)
	14	249	9.4	17.4 (0.5)
	60	282	9.4	17.5 (0.6)
Brash	2	435	8.8	16.3 (0.4)
	14	306	3.2	5.9 (0.1)
	60	310	3.6	6.7 (0.1)
Grass	2	456	NA	NA
	14	503	13.4	24.8 (0.1)
	60	266	7.6	14.2 (0.1)
Trees	2	470	23.1	42.8 (1.4)
	14	413	15.3	28.3 (0.3)
	60	372	9.4	17.5 (0.1)
Trees + brash	2	342	8.4	15.6 (0.1)
	14	391	4.2	7.8 (0.1)
	60	434	6.0	11.1 (0.1)
Trees + grass	2	435	26.1	48.3 (0.1)
	14	412	13.3	24.6 (0.1)
	60	373	9.2	17.0 (0.1)

NA — missing value
Values are the mean of three replicates. Standard errors of arcsin transformed data are in parentheses.

Vegetation uptake

Vegetation retained a significant proportion of the tracer-N, 60 days following the application of the tracer (Table 4). The grass immobilised 17.7% in the grass and 12.9% in the grass + trees mixture treatment (not significantly different at $P < 0.05$). The sitka seedlings immobilised between 1.5% and 7.7% of the tracer. The larger seedlings in the trees + brash treatment immobilised significantly more than the tree seedlings in a tree + grass mixture.

^{15}N recovery

Retention of the labelled nitrogen in the lysimeter organic material after

60 days was 41%—66% (Table 4). This fraction represents microbial and non-microbial organic-N and inorganic-N held in exchange forms on organic matter. Incorporation into the non-microbial or residual organic-N (RO-N) of soil organic-N pool can be calculated from the total recovery in the dried ground material minus the microbial biomass-N and the KCl extractable-N pools. Total recovery of the tracer-N was significantly different between treatments ($F = 10.33$, $P < 0.01$) with the lowest recovery in the brash treatment (68%) and the highest from the grass treatment (99%) (Table 4).

Discussion and conclusions

Although large nitrate losses are known to occur at this site (Stevens & Hornung 1988), leachate losses of added nitrogen were low. Losses of nitrate ranged from 0.4 to 4.2% of the added tracer with the higher values from the brash treatments. Stamms et al. (1990) indicate that as a consequence of simultaneous mineralisation and immobilisation by the microbial biomass an exchange of ^{15}N and ^{14}N-ammonium will result in nitrification of the latter and an underestimate of the importance of oxidation of added ammonium. A rapid uptake of ^{15}N into the microbial

Table 4. ^{15}N recovery in soil, vegetation and leachate pools 60 days after application to the lysimeters.

	% Tracer						
Treatment	RO-N	Microbial-N	KC1-extr N	Leachates	Trees	Grass	Total
Control	34.1	17.5	3.9	22.1	—	—	78.6 (0.3)
Brash	29.9	6.7	4.6	26.0	—	—	67.6 (0.1)
Grass	45.4	14.2	6.2	15.5	—	17.7	99.0 (0.1)
Trees	39.8	17.5	3.6	15.6	3.6	—	76.8 (0.2)
Trees + brash	26.1	11.1	5.4	17.0	7.7	—	69.7 (0.2)
Trees + grass	36.7	17.0	12.1	12.1	1.5	12.9	83.1 (0.1)

Abbreviations: RO-N — residual organic N
Microbial N — microbial biomass-N ($K_N = 0.54$)
KC1-extr N — KC1 extractable N (NH_4-N + NO_3-N + organic-N)
Leachates — cumulative inorganic-N leachate losses

Values are the mean of three replicates. Standard errors of arcsin transformed data are in parentheses.

biomass has been demonstrated in this study and may limit conclusions that can be drawn concerning the role of incoming ammonium as a substrate in the nitrification process.

The rapid conversion into nitrate-N and the similar enrichment of the ammonium and nitrate pools suggest an ammonium substrate for the nitrifiers and not an organic-N substrate indicating an autotrophic pathway. This has been reported in acid forest soils (Stams et al. 1990); however, as de Boer et al. (1990) indicate, all evidence is indirect as no acid-tolerant ammonium-oxidising bacteria has been isolated. In the UK, heterotrophic nitrification had previously been reported to predominate in acid coniferous forest soils (Kilham 1987).

There was a persisitent but small leachate loss of tracer-N predominantly in the form of nitrate, on Day 2, 14 and 60. The transformation of tracer and soil derived ammonium-N into nitrate was faster and more sustained in the brash treatments supporting the earlier finding of greater nitrification rates beneath brash piles (Emmett et al. 1991a). Leachate losses are not often directly measured in forest ^{15}N studies or agricultural studies with non-recovery of ^{15}N in the plant and soil system accorded to 'leaching and gaseous losses'. Overrein (1971a, b, 1972) reported that losses after addition of nitrogenous fertiliser were greatest after addition of nitrate fertiliser (100 kgN ha^{-1}) with 82% lost as leachate in the first few months. Losses after application of ammonium-based fertiliser were more gradual and significantly less with 2.8% lost after 40 months.

Immobilisation of the labelled N by the vegetation varied with the species. The grass immobilised 18% and the trees 4% in the single factor treatment although biomass was similar. Emmett et al. (1991b) found improved growth of the trees beneath the brash and suggested this was a consequence of the increase in phosphorus and potassium supply and/or improved microclimate conditions (Emmett et al. 1991b). The nitrogen demand of these trees was greater, reflected by the 7.8% uptake of the labelled N by the trees in the presence of brash compared to only 1.5% when the trees were in competition with the grass. In a similar study, only 3.5% of the fertiliser was recovered in three year old Black spruce trees 2.5 months after application (Weetman 1962 in Knowles 1975). Comparisons with other studies are of limited value due to the longer time post-application, higher rates of application and the different ages of trees in other studies.

Between 41% and 66% of the tracer-N remained in the organic material 60 days after application which is comparable to other studies using significantly higher levels of application. Knowles (1975) cited values ranging between 29—42% of ammoniacal fertiliser held in organic layers, 12—40 months after application. Nômmik (1989) found 54% of

150 kg ha^{-1} ammonium nitrate-N remained in the soil (organic + mineral) after two growing seasons. Leaching and gaseous losses were assumed to account for the 12% of the fertiliser not in the soil and vegetation.

The ammonium may have been retained in the organic horizons by biotic or abiotic processes. Chemical binding of ammonium-N in acid forest humus was thought to be negligible after work on sterile forest humus by Nômmik (1970). Indeed, Vitousek & Matson (1984, 1985b) investigated the importance of microbial immobilisation of nitrogen inputs and the turnover of the microbial-N pool assuming all incorporation would occur through the microbial biomass. However, more recently Schimel & Firestone (1989) reported that 20% of added ammonium-N was immobilised in an acid coniferous humus by abiotic processes. The rapid incorporation of the ^{15}N-ammonium into KCl-extractable organic-N in this study may be a result of such abiotic immobilisation processes. Van Cleve & White (1980) suggested that extractable organic-^{15}N may be microbial nitrogen constituents released by cellular lysis during extraction. This is unlikely as the atom % excess of the microbial biomass was significantly higher than the KCl extractable organic-N in this study. It is more likely that the ammonium has been incorporated into extractable organic-N through abiotic processes although there may be some microbial nitrogen constituents present.

The proportion of the ^{15}N passing through or immobilised in the microbial biomass was significant in this study and had a similar ^{15}N enrichment to the KCl extractable inorganic nitrogen indicating the importance of the microbial biomass-N as a source of mineralisable-N (Paul 1984). These values for microbial biomass-N may be underestimates as described previously, due to the initial extraction of nitrogen held in the biomass in the unfumigated samples. However, if the values presented here are considered as minimum values, the microbial biomass-N accounts for 37—82% after 48 hours decreasing to 19—32% after 60 days, of the soil organic^{15}N. This represents 15—48% and 6—17% of the tracer, 2 and 60 days after application of the tracer respectively, with no significant change in the size of the total microbial biomass-N pool. The initial biological half-life of this microbial-N is less than 12 days perhaps as a consequence of rapid isotope exchange reactions with a labile microbial biomass-N pool described earlier. Initial fast exchange between the soil microbial biomass-N pool and added labelled-N has also been shown in an agricultural soil (Bristow et al. 1987). These authors suggest the fall in the ^{15}N content of the microbial biomass was due to death and decay of the microbial cells and remineralisation of the ^{15}N originally immobilised.

In addition to this short-term microbial pool, the labelled-N appeared to be incorporated into stable microbial fractions more resistant to

mineralisation (Ladd et al. 1981, Carter & Rennie 1984). The biological half-life of the microbial-N increased after 14 days, with no significant change in the percentage tracer in the microbial biomass between 14 and 60 days in the control and brash treatments. Another explanation for this increased biological half-life may be; the recycling of labelled-N through microbial attack of ^{15}N labelled humus, the utilisation of senescent labelled microbial biomass or the immobilisation of labelled plant litter (Jenkinson & Parry 1989). These processes would result in the continued enrichment of the microbial biomass pool.

The presence of brash significantly reduced the total ^{15}N immobilised in the microbial population and the overall soil organic matter. The brash will act as an insulating layer thus reducing dessication and temperature fluctuations in the lower soil horizons. An inert covering of polysyrene beads on these lysimeters was found to increase nitrogen leaching similar to those found beneath a brash covering (Emmett et al. 1991b). A change in soil microclimate conditions therefore appears to reduce the retention of incoming atmospheric ammonium in the lower organic horizons by reducing the microbial immobilisation potential. This may be offset by the immobilisation of atmospheric ammonium by the surface brash beneath which the ^{15}N-ammonium was applied in this study.

The reduced microbial immobilisation in the brash lysimeters may be a consequence of the high nitrification rate present beneath the brash. Vitousek & Andariese (1986) found reduced nitrogen immobilisation in high nitrifying felled forest sites and suggested it was a consequence of labelled nitrogen bypassing immobilisation by oxidation to nitrate. Juma & Paul (1983) in an agricultural soil also found nitrification affected the microbial immobilisation rate. The presence of a nitrification inhibitor increased labelled N in the microbial biomass by 100% (as in this study), with no change in the rate of release of the ^{15}N from the microbial biomass pool. This can explain only part of the brash effect on uptake of labelled-N into microbial biomass as there is insufficient nitrate present either as leachate or KCl-extractable nitrate to account for the reduced uptake into the microbial biomass. One explanation may be an under-estimation of nitrate production, resulting from the conversion of nitrate to gaseous forms of nitrogen through the denitrification process. The presence of increased concentrations of nitrate and dissolved organic carbon and increased moisture contents in conventionally harvested plots (i.e. beneath a brash covering) (Stevens & Hornung pers. comm.) may increase denitrification. A relationship between the size of soil nitrate pools and the rate of denitrification has previously been described in forest soils after felling (Vitousek & Matson 1985b).

The reduced immobilisation in the microbial biomass of the brash

treatments is reflected in the total recovery of the applied ^{15}N-ammonium in the lysimeters. The presence of brash significantly reduced microbial immobilisation ($F = 9.28$, $P < 0.01$), with only 68—70% of the tracer was recovered from the lysimeters containing brash. Recovery from the non-brash lysimeters was 77—99%. The ^{15}N unaccounted for may have been lost through denitrification or as a consequence of the overflowing of the collection bottles during the initial 2 days. It is unlikely that the latter can account for 30% of the applied ^{15}N-ammonium. Another possibility is the loss of dissolved organic-N (DON) in the leachates. Earlier results, however, indicated that there was no significant differences in DON losses between treatments (Emmett et al. 1991a). In addition the atom % excess of the KC1 extractable organic-N during was consistently lower than for inorganic species of nitrogen. Assuming DON represented approximately 20% of the nitrogen lost as leachate (Emmett et al. 1991a), losses of applied ^{15}N-ammonium as DON can be calculated to account for a maximum of 5% of the tracer. Thus 25% of the labelled N in the brash treatments and 10% in the non-brash treatments may represent maximum gaseous losses of the ^{15}N-ammonium applied to the lysimeters.

Acknowledgements

The authors would like to thank Dr. J. M. Anderson and Dr. M. Hornung for their advice and P. Splatt for technical assistance. Permission to work at Beddgelert forest was granted by the Forestry Commission. This work was funded by NERC as part of a Ph.D. studentship award.

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