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Fate of Nitrogen-15 in a Perennial Ryegrass Seed Field and Herbaceous Riparian Area

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ABSTRACT

Intensive management of grass seed fields in the poorly drained soils of the Willamette Valley, Oregon, has prompted concern in the capacity of these landscapes and their associated minimally managed riparian areas to process and retain fertilizer N. Our goal was to determine the extent of N losses and effectiveness of a riparian area and an adjacent perennial ryegrass seed field to retain N. The fate of fertilizer $^{15}NH_4$ and $^{15}NO_3$ was determined with a ^{15}N tracer experiment. During the second year of the study, ^{15}N recovery in the plant and soil (0–30 cm) from the cropping system was 51% for $^{15}NH_4$ and 43% for $^{15}NO_3$ whereas recovery in the riparian area was only 20% of $^{15}NH_4$ and 31% of $^{15}NO_3$. Greater cropping system retention of ^{15}N resulted from both greater uptake by the crop and greater retention of ^{15}N in the soil. Low recovery of ^{15}N in the riparian area was possibly the result of two significant spring flood events saturating the surface soil of the riparian area but not the cropping system. The prolonged seasonal saturated conditions significantly reduced riparian plant biomass production and N uptake and increased the potential of N loss through overland flow and denitrification. Results indicate that the cropping system had larger available N pools and a larger potential to retain fertilizer N than the riparian zone. However, both areas were prone to substantial loss of applied N.

Intensive forage and grass seed production requires high levels of fertilizer N applications. The production of forage and grass seed are often in areas characterized by seasonally cool wet climates such as the Pacific Northwest. Grass seed production is one of the largest agricultural industries in the Willamette Valley, Oregon, making up 55% of the land-use (Nelson, 2003). Grass seed cropping systems in western Oregon typically receive fertilizer applications in the range of 140 to 235 kg N ha^{-1} yr^{-1} (Griffith et al., 1997b) that are, on average, 30% greater than extension service recommendations (Horneck and Hart, 1988). Intensive fertilizer N management of these systems, high seasonal rainfall, and an increasing awareness of the economic and environmental consequences of N loss have led to a growing interest in understanding how these systems, and their associated riparian areas, process and retain N.

Denitrification, leaching, and overland flow of seasonal precipitation have been shown to be significant pathways of N loss from these systems (Horwath et al., 1998; Wigington et al., 2003). In addition, differences in management (e.g., tillage, monoculture, rotation length, fertilizer applications) and landscape position of grass seed fields and riparian areas will influence the cycling of N into and through the soil and plant pools and determine if these systems will prevent or contribute to NO_3^- contamination of surface and ground water.

Competition for N between plants and soil microbes is the primary process controlling the potential for NO_3^- loss through water movement. In unfertilized grassland systems, competition for inorganic N between plants and the soil microbial biomass can be significant because the demands for N are met by (Dell and Rice, 2005) and can exceed supply by mineralization (Woodmansee et al., 1981; Williams et al., 2001). Plant and microbial consumption of NO_3^- was greater in a native prairie compared with a fertilized cultivated soil (DeLuca and Keeney, 1995). Microbial biomass has been shown to control plant N uptake in a N-limited tall grass prairie (Williams et al., 2001), grass swards (Hatch et al., 2000), and perennial ryegrass plots (Hodge et al., 2000). The absence of cultivation can increase the size and activity of the soil microbial biomass (Smith and Paul, 1990; Horwath et al., 1998) and available C (DeLuca and Keeney, 1994). In addition, higher soil C/N ratios often found in unfertilized systems could potentially increase the amount of N immobilized by microbes and incorporated into soil organic matter pools and limit the N available for plant uptake (Hodge et al., 2000). The landscape position of riparian areas exposes them to high, fluctuating water tables further increasing competition for N by reductive soil N processes such as denitrification (Lowrance et al., 1984; Peterjohn and Correll, 1984; Jacobs and Gilliam, 1985; Haycock and Pinay, 1993; Horwath et al., 1998) and dissimilatory NO_3^- reduction to NH_4^+ (Davis, 2003). Competition for inorganic N between plants and the soil microbial biomass may be less intense in agricultural systems. High inputs and decreased competition for N in highly managed systems creates the potential for larger N pools susceptible to leakage (Scholefield et al., 1993).

Differential processing of NH_4^+ versus NO_3^- in the plant–soil systems of grass seed fields and associated herbaceous riparian areas is not well understood. It is often assumed that the fate of soil NO_3^- is limited to immobilization by plants, denitrification, and water movement through mass flow and diffusion (Davidson et al., 1990). Schimel et al. (1989) and Hatch et al. (2000) found greater plant uptake of NO_3^- in grassland soils due to microbial immobilization of NH_4^+. Microbial preference for NH_4^+ has been documented as well as the inhibition of microbial immobilization of NO_3^- in the presence of NH_4^+ (Rice and Tiedje, 1989). However, studies have also shown significant microbial uptake of NO_3^- in soil even in the presence of NH_4^+ and this has been attributed to microsite depletion of NH_4^+ (Jackson et al., 1989;

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Davidson et al., 1990; Dell and Rice, 2005). Although many crops are equally capable of taking up NH_4^+ and NO_3^- (Haynes and Goh, 1978), perennial ryegrass (*Lolium perenne*) has shown greater uptake and utilization of NH_4^+ than of NO_3^- at low temperatures (<10°C) (Clarkson and Warner, 1979; Clarkson et al., 1986; Griffith et al., 1997a).

Intensive management of grass seed cropping systems could lead to a greater supply of N than demand and result in leaching of NO_3^- to surface and ground water. Conversely, higher demand than supply for N in a minimally managed riparian zone may allow these systems the capacity to process excess N entering from upland intensively managed systems. Understanding the dynamics of plant and microbial utilization of NH_4^+ and NO_3^- and the potential for incorporation into organic matter is essential for managing these systems to achieve optimal crop performance with minimal environmental degradation. To better understand how inorganic N is processed, $^{15}NH_4^+$ and $^{15}NO_3^-$ were added to an herbaceous riparian area and grass seed cropping system and the partitioning of ^{15}N into the plant, microbial, and soil pools were measured 2 and 14 mo after the label was applied. Our hypothesis was that the demand and therefore retention of ^{15}N would be greater in the riparian area than the cropping system. In addition, we hypothesized that NH_4^+ would be retained to a greater degree in both systems.

MATERIALS AND METHODS

Site Description

The site was located on a tributary of Lake Creek in Linn County, Oregon, USA (44° 32′ N, 123° 03′ W). The intermittent stream was bordered by an uncultivated mixed perennial herbaceous species riparian area that was 30 to 48 m wide. A perennial ryegrass (*Lolium perenne* L.) seed field (cropping system) was adjacent and upslope to the riparian area. The Dayton soil series (fine, smectitic, mesic Vertic Albaqualfs) extended from the streambed into the riparian area approximately 25 m. The Holcomb soil series (fine, smectitic, mesic Typic Argialbolls) extended from the Dayton soil through the remainder of the riparian area and out into the cropping system.

Precipitation was 1860 mm in 1996 and 1134 mm in 1997, with 88 and 76% falling between November and June, respectively (Oregon Climatic Service, 2005). Precipitation in 1996 was the highest on record for the last 110 yr. During the wet winter period (November to March), the water table was close to the soil surface and soils were frequently saturated (Wigington et al., 2003). In the spring of 1996, two precipitation events occurred that produced average water table heights in the riparian zone that were above the soil surface. These events occurred approximately 7 (25 Mar. 1996; 7 cm above soil surface) and 35 (22 Apr. 1996; 5 cm above soil surface) days after the ^{15}N label was applied and did not raise the cropping system water table above the surface soil (25 Mar. 1996; 50 cm below soil surface; 22 Apr. 1996; 7 cm below soil surface). The riparian area also experienced a significant drying period between these flood events where the water table dropped to an average of 20 cm below the soil surface (8 Apr. 1996). By the end of June the surface soils had dried out and the water table dropped below 1.5 m until consistent precipitation began again in the following fall.

Table 1. Summary of differences in soil characteristics and management of the riparian area and cropping system soils from 0 to 15 cm.

Soil characteristic	Riparian area	Cropping system
Vegetation	grass/herbaceous	perennial ryegrass
Soil type†	Holcomb	Holcomb
Tillage management	uncultivated > 20 yr	cultivated fall 1994
Fertilizer N, kg ha^{-1}	0.0	177
Water content, %‡	54.8	38.6
Bulk density$_{-1}$	1.0	1.3
Soil C g kg^{-1}	35.5	29.7
Soil N g kg^{-1}	2.6	2.2
% organic matter (LOI)	15.8	6.27
% sand	8.0	7.6
% silt	67.2	69.1
% clay	24.8	23.3

† Fine, smectitic, mesic Typic Argialbolls.
‡ Average gravimetric soil water content.

Table 1 outlines differences between soil properties and management of the riparian area and cropping system. The riparian area was vegetated predominantly with grasses (native and introduced), forbs, sedges, and rushes (McAllister et al., 2000) and had not been cultivated for over 20 yr. On 15 Sept. 1994, the previous perennial ryegrass (*Lolium perenne* L.) crop was tilled into the soil to a depth of approximately 20 cm and the cropping system was replanted with perennial ryegrass using conventional tillage practices. The same crop was kept for the following three growing seasons. The cropping system received 162 kg N ha^{-1} as split applications in the spring of 1996 (1 Mar. 1996 and 20 Apr. 1996) and 191 kg N ha^{-1} as split applications in the spring of 1997 (14 Mar. 1997 and 14 Apr. 1997). The fertilizer was applied as urea-ammonium sulfate solutions.

Nitrogen-15 Labeling

Sixteen, 3 by 3 m experimental plots were established in the Holcomb soil, eight in the riparian area and eight in the cropping system. Riparian and cropping system plots were located on opposite sides of Lake Creek to prevent cross contamination of label. Plots within each area were positioned along a line that ran perpendicular to the stream to prevent label from contaminating other plots through overland or ground water flow. Four of the eight plots in each area received 0.82 kg ^{15}N ha^{-1} as $^{15}NH_4NO_3$ (99.9 atom %) and the remaining four plots received 0.82 kg ^{15}N ha^{-1} as $NH_4{}^{15}NO_3$ (99.9 atom %). These amounts of ^{15}N were expected to enrich soil ^{15}N to 0.5 atom % ^{15}N above background from the soil depth of 0 to 30 cm. On 18 Mar. 1996, 1-L solutions containing the ^{15}N were uniformly applied to the soil surface of all plots using a pump sprayer. One additional liter of water was applied to each plot as a rinse. Plant and soil samples were collected from a 2 by 2 m interior plot, creating a 0.5-m border around each of the 16 primary plots.

Plant and Soil Analyses

Plant and soil samples were collected from plots in May of 1996 and 1997 to determine the fate of applied ^{15}N. The N and ^{15}N content of soil and plant N was measured on a Europa 20/20 isotope ratio-mass spectrometer (IRMS) (Europa Scientific, Crewe, England). Aboveground plant samples were collected from three randomly selected 10-cm^2 subplots within each replicate plot. Plant samples were dried at 70°C for 24 h, ground to pass a 50-μm sieve, and analyzed for total N and ^{15}N content. Aboveground plant biomass samples were also periodically collected from January to July of 1996 and were dried and weighed for biomass production and analyzed for total N content.

Two replicate soil cores were collected from all plots at 0- to 10-, 10- to 20-, and 20- to 30-cm depths. In 1996, cropping system soil samples were collected both 'on the row' of ryegrass and 'between the rows'. Surface soil cores (0–10 cm) collected 'on the row' of the ryegrass crop were 10 cm in diameter. Roots were collected from one half of the core by washing the sample on a fine sieve (0.5 mm) and removing non-root debris by hand. Root material was then dried, ground, and analyzed for total N and ^{15}N content. Soil total N and ^{15}N content were quantified on soil collected from the other half of the core after coarse and fine roots and non-root debris were removed by hand and the soil was dried, ground, and thoroughly mixed. Cropping system soil cores collected from the 'on the row' subsoil (10–20 and 20–30 cm) and from all soil depths 'between the rows' were 5 cm in diameter. These cores had debris removed before soil was analyzed for total N and ^{15}N content. In the cropping system, analytes quantified from soil samples collected from 'between the rows' and 'on the row' were combined for a total after analysis as [(0.75 × 'between') + (0.25 × 'on row')] to account for the incomplete coverage of the ryegrass crop in 1996. The ratio of 'between' and 'on row' was based on measurements of root biomass distribution from row center. In the riparian system, surface soil cores (0–10 cm) were also 10 cm in diameter. Roots collected from one half of this core were dried, homogenized, ground, and analyzed for total N and ^{15}N content. Soil total N and ^{15}N content were quantified on soil collected from the other half of the core after roots and debris were removed and the soil was homogenized, dried, and ground. Soil cores collected from the subsoil (10–20 and 20–30 cm) were 5 cm in diameter. These cores had debris removed and soil was analyzed for total N and ^{15}N content. In 1997, the sampling was repeated, except that in the cropping system, 'on the row' and 'between the rows' were not differentiated due to complete crop coverage.

Differences in means, of untransformed data, between the sites (cropping system and riparian area) and ^{15}N treatments were determined by analysis of variance using a split-plot design, with site as the main plot and ^{15}N treatments as subplots using SPSS statistical software (SPSS Inc., 2002). The model for this design was:

$$y_{ijkl} = \mu + S_i + B_j + \mathrm{SB}_{ij} + \delta_{ij} + T_k + \mathrm{ST}_{ik} + \mathrm{SBT}_{ijk} + \varepsilon_{(ijk)}$$

Where S is site (cropping system and riparian area), B is block, and T is ^{15}N treatments. The restrictions on randomization were represented by: δ_{ij} the restriction error. The MS for SB was used to test S main effect, and the MS for SBT used to test the T main effects and ST interaction. Means separations were done using Fisher's Protected Least Significant Difference test (Sokal and Rohlf, 1981). All differences reported are significant at $p \leq 0.05$, unless otherwise stated.

Long-Term Incubations

The mineralization of organic matter is a key process that regulates the cycling and availability of N in soil. Long-term aerobic incubations (Horwath, 1993) were used in conjunction with ^{15}N to understand how fertilizer N was stabilized into the organic matter and labile N pools. Incubations were performed on soil collected in the spring of 1996, from the riparian and cropping system plots, at the 0- to 10-, 10- to 20-, and 20- to 30-cm depths. Before the incubations began, the soil was sieved (5 mm), mixed, and moistened to bring the water content to 55% of field capacity. Replicates within each plot were composited, and a total of 250 g of moist weight soil were weighed into mason jars fitted with lids that had 1-mm holes to allow aerobic conditions to persist inside the jars. Soil samples from the cropping system plots were collected both 'between the rows' and 'on the row', and were incubated separately and then combined for a total after analysis. Soils were incubated in the dark at 25°C for 150 d and maintained at 55% of field capacity. A 25-g subsample was collected from each jar at 0, 10, 20, 30, 45, 60, 80, 110, and 150 d and extracted for NH_4^+ and NO_3^- with 2 *M* KCl.

Soil subsamples were extracted with 100 mL of 2 *M* KCl after shaking for 1 h at 300 rpm. After shaking, the slurry was allowed to settle for 30 min and was then filtered through a Whatman Qualitative no. 1 filter (Whatman Inc., Florham Park NJ) that had been washed three times with 1% (v/v) HCl and three times with 2 *M* KCl. Concentrations of NH_4^+ and NO_3^- were measured colorimetrically on soil extracts using a flow injection autoanalyzer (Lachat, Milwaukee, WI). The ^{15}N content of NH_4^+ and NO_3^- was determined by a modified diffusion method (Brooks et al., 1989) and analysis on a Europa 20/20 isotope ratio-mass spectrometer (IRMS) (Europa Scientific, Crewe, England). The amount of extract (10–60 mL) used in the diffusions was calculated to contain 100 μg of N based on the measured concentration of NH_4^+ and NO_3^-. If the extract contained <20 μg of N, it was spiked to bring the amount of N to 100 μg. To determine the ^{15}N content of soil NH_4^+, the extracts were treated with 0.2 g of MgO and gently stirred with acid-washed glass beads for 6 d. Volatilized NH_3 was collected on a 7-mm diam. GF/D glass fiber filter disk that was acidified with 2.5 *M* $KHSO_4$ and suspended over the solution by a stainless steel wire. The disks were dried and put into Sn capsules for ^{15}N analysis. The extracts were then treated with 0.4 g of Devarda's Alloy for 6 d for the collection of reduced NO_3^-–N on a second acidified disk.

Microbial biomass N (MBN) and ^{15}N were determined at Day 0 of the long-term incubation using the chloroform fumigation incubation (CFI) method (Horwath, 1993; Horwath and Paul, 1994). The CFI microbial biomass N and ^{15}N were estimated by incubating fumigated (ethanol free chloroform) and unfumigated soil subsamples (25 g) for 10 d at 25°C. After 10 d the soils were extracted with 2 *M* KCl and analyzed for inorganic N and ^{15}N with colorimetric and diffusion/IRMS methods as described previously. Microbial biomass N and MB^{15}N were calculated as the difference between the mass of fumigated and non-fumigated N and ^{15}N divided by a K_n of 0.48.

Mineralizable N pool sizes (N_{min}), rate constants (k), and mean residence times (MRT) of the labile organic N pool were determined for both mineralized soil N and applied ^{15}N. Rate constants and MRT for the cumulative data were calculated using a first-order, one-pool model (Paul and Clark, 1996):

$$N_t = N_o e^{-kt} \quad [1]$$

where N_t is the concentration of substrate remaining at time increment t, k is the rate constant (d^{-1}), and N_o is the concentration of N_{min} at Day 0 (μg N g soil^{-1}). Mean residence times was calculated as:

$$\mathrm{MRT} = 1/k. \quad [2]$$

A double model (Molina et al., 1980) was used to determine if two organic N pools, active and resistant, could be distinguished. A two-component, first-order exponential equation was used for the cumulative data:

$$N_t = N_1 e^{-k1t} + N_2 e^{-k2t} \quad [3]$$

where N_1 is the active fraction, N_2 is the resistant fraction and k_1 and k_2 are the corresponding rate constants of these fractions. Significance of difference between the one and two pool models was detected using an F-test.

Inherent statistical problems with using cumulative data (Eq. [1] and [3]) to estimate N pool parameters has led to model fitting of incremental data (Hess and Schmidt, 1995; Kiovunen and Horwath, 2004). A first-order rate equation (Whitmore, 1996) was used on the incremental data:

$$\Delta N = N_o e^{-kt}(1 - e^{-k\Delta t}) \quad [4]$$

where the amount of N mineralized is estimated for the time interval directly before t. N_o is the amount of N mineralized at time zero (N_{min}) and k_1 is the rate constant of the active fraction. All models were fitted to the data using the nonlinear curve-fitting programs in Sigma Plot 8.0 (Systat Software, Inc., 2002). All differences were significant at $p \leq 0.05$, unless otherwise stated.

RESULTS AND DISCUSSION

Plant Nitrogen Dynamics

The percentage of crop shoot N derived from added ^{15}N was three times higher than for roots (Fig. 1). Labeled ^{15}N was applied at the beginning of the linear phase of shoot N accumulation for perennial ryegrass (Griffith et al., 2003). Periods of rapid root growth have been shown to precede and follow periods of active shoot growth (Griffith et al., 1997a), which explains the lower percentage of N derived from ^{15}N in root biomass than in shoots during the measured period (Fig. 1). There was no difference between the amounts of N that were derived from $^{15}NH_4^+$ or $^{15}NO_3^-$ in crop shoot or root material in 1996. We hypothesized that there would be preferential uptake of NH_4^+ by the crop due to physiological preferences of ryegrass for NH_4^+ at cool temperatures (<10°C) and the dominance of NH_4^+ in soil under cool wet spring conditions in the Willamette Valley (Clarkson and Warner, 1979; Clarkson et al., 1986; Griffith et al., 1997b). Higher mobility of NO_3^- into the root area may have counterbalanced the NH_4^+ preference, so that similar amounts of N were derived from $^{15}NH_4^+$ and $^{15}NO_3^-$ by the crop.

There was no difference in the percentage of shoot and root N derived from added ^{15}N in the riparian vegetation (Fig. 1). Riparian shoot biomass contained a larger ($p \leq 0.06$) percentage of N derived from $^{15}NO_3^-$ than $^{15}NH_4^+$. The lower percentage of N derived from $^{15}NH_4^+$ in the riparian shoot biomass was not completely unexpected. Schimel et al. (1989) found that unfertilized grassland plants utilized primarily NO_3^- since the microbial biomass of these soils competed better for soil NH_4^+. However, we did not find greater immobilization of NH_4^+ into the microbial biomass or soil organic matter of the riparian area compared with cropping system NH_4^+. Higher mobility of NO_3^- and quicker transport to the root zone compared with NH_4^+ may have led to greater plant uptake of $^{15}NO_3^-$.

Maximum ^{15}N recovery per plot (41%, pooled ^{15}N treatments), in 1996, occurred in the crop shoot biomass (Table 2). Shoot biomass production and total N accumulation were greater in the crop than in the mixed riparian vegetation in 1996 (Fig. 2). Paul and Juma (1981) also found greatest recovery of ^{15}N in the crop biomass (81.2%, spring wheat) 12 wk after $^{15}NH_4OH$ was applied to soil cores. Lower plot recovery of ^{15}N (10%, pooled mean) in the riparian shoot biomass was associated with low overall production of aboveground biomass and N accumulation (Fig. 2), which was most likely due to spring flood events in the riparian area.

Recoveries of ^{15}N in the plant biomass of the following year (1997) ranged from 3.5 to 11% (Table 2) compared with 6.2 to 42% in 1996, indicating that little of the inorganic ^{15}N applied in 1996 was available for plant uptake in 1997. Decreases in percentage recovery of ^{15}N in plant biomass from 1996 to 1997 were significant especially for crop shoot and riparian root biomass (Table 3). These findings are in contrast to Dell et al.

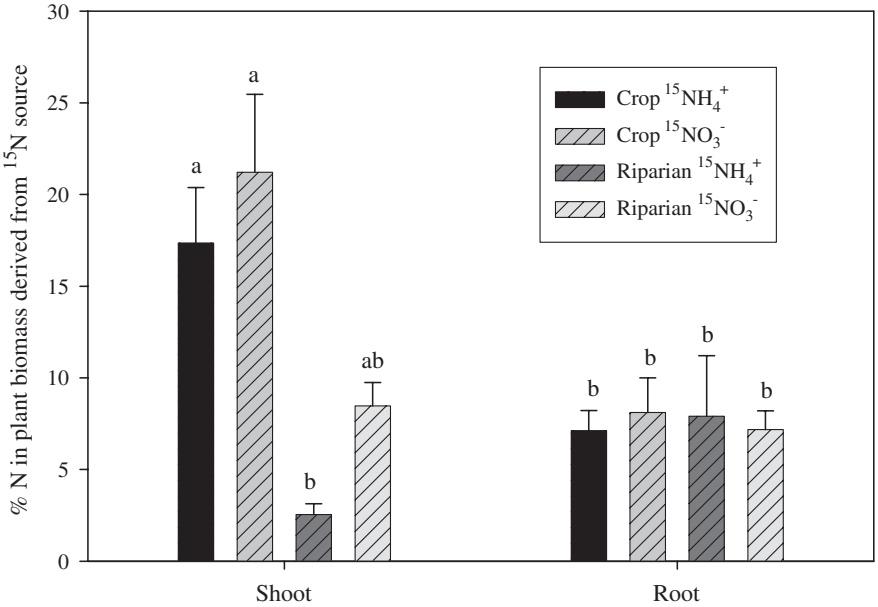

Fig. 1. Percentage of N in shoot and root biomass derived from ^{15}N in the riparian area and cropping system in samples collected in May 1996. Letters denote significant differences between sites (cropping system and riparian area) and ^{15}N treatments ($^{15}NH_4^+$ and $^{15}NO_3^-$) ($p \leq 0.05$).

Table 2. Mean and standard errors of the percent recovery of ^{15}N in soil, shoot and root biomass, and whole plot for samples collected in May 1996 and 1997, 2 and 14 mo after the label was applied. Letters denote significant differences among sites (cropping system and riparian area) and ^{15}N treatments ($^{15}NH_4^+$ and $^{15}NO_3^-$) ($p \leq 0.05$).

			Total soil N % recovery			Plant biomass % recovery		Total plot % recovery
Year	Site	^{15}N Tmt	0–10 cm	10–20 cm	20–30 cm	Shoot	Root	Total
1996	Cropping system	$^{15}NH_4^+$	27(6.9) a	0.7(0.1) a	0.7(0.2) a	39(6.1) a	7.3(1.0) a	75(11) a
		$^{15}NO_3^-$	11(2.2) b	0.8(0.2) a	0.9(0.4) a	42(9.9) a	7.6(1.0) a	62(13) a
	Riparian area	$^{15}NH_4^+$	4.3(1.6) b	0.5(0.4) a	0.5(0.1) a	6.2(4.9) b	14(6.0) a	26(3.0) b
		$^{15}NO_3^-$	5.5(0.6) b	1.3(0.7) a	2.3(0.8) a	13(2.7) b	20(3.7) a	42(5.4) b
Site × treatment			*	ns†	ns	ns	ns	ns
1997	Cropping system	$^{15}NH_4^+$	32(3.5) a	1.2(0.1) a	0.4(0.5) a	6.1(0.2) ab	11(1.8) a	51(2.9) a
		$^{15}NO_3^-$	27(3.7) a	0.8(0.2) a	0.5(0.4) a	7.6(0.3) a	7.2(0.8) b	43(4.7) ab
	Riparian area	$^{15}NH_4^+$	11(1.7) b	0.5(0.1) a	0.5(0.2) a	4.3(0.3) b	3.5(0.3) c	20(1.3) c
		$^{15}NO_3^-$	16(4.2) b	0.5(0.1) a	0.8(0.3) a	7.2(0.8) a	6.6(0.9) b	31(4.5) b
Site × treatment			ns	ns	ns	ns	**	ns

* Significant at the 0.05 probability level.
** Significant at the 0.01 probability level.
† ns = $p > 0.05$.

(2005) who found that the recovery and distribution ^{15}N in the plant biomass of a tallgrass prairie did not change much from the first to the second growing season after application of $^{15}NH_4^+$. However, large decreases in plant ^{15}N from the second to the fifth growing season were matched by increases in soil organic N. In the present study, the decrease in crop biomass percentage of recovery (−22%; root plus shoot; pooled ^{15}N treatment) was significantly ($p = 0.04$) greater than the decrease (−16%) in riparian area plant biomass.

Soil Nitrogen Dynamics

Total microbial biomass N (MBN) was significantly higher in the riparian soil than the cropping system in the surface soil (0–10 cm) and decreased significantly with depth at both soils (Table 4). Agronomic practices such as cultivation and pesticide use have been shown to decrease microbial biomass (Smith and Paul, 1990; Horwath et al., 1998). Microbial biomass C has been shown to be higher in this riparian soil (Horwath et al., 1998; Davis, 2003) and a tallgrass prairie (Groffman et al., 1993) as compared with a complementary cultivated soil. At the lower soil depths sampled, MBN was significantly higher in the cropping system soil than in the riparian soil. Conventional tillage with incorporation of residues into the plow layer likely led to higher MBN with depth in the cropping system. Cropping system and riparian soil total MBN (0–30 cm) was approximately 18 and 29% less than reported for a tall grass prairie (Garcia and Rice, 1994).

By mid-May, 2 mo after ^{15}N-label was applied, <6% of ^{15}N was recovered in the soil microbial biomass (Table 5). This was significantly ($p \leq 0.05$) less than that found in plants (20–50%). These findings differ from those of Jackson et al. (1989), who found that soil microbes were the main consumers of both NH_4^+ and NO_3^-, in an unfertilized California grassland, even during periods of rapid plant growth. Hatch et al. (2000) also found that microbial uptake of N was the major consumer of ^{15}N in unfertilized grass swards. Both studies examined the movement of ^{15}N for short periods (1–4 d). Longer-term studies (2–4 mo) have found more ^{15}N in the plant biomass than microbial biomass (P.D. Brooks and E.A. Paul, unpublished data as cited in Jackson et al. 1989) and soil (Paul and Juma, 1981) of grass swards and spring wheat, respectively. In our soils, it is possible that the microbial biomass initially took up more ^{15}N than the plant biomass, but with time, turnover of the microbial biomass released ^{15}N for plant uptake in the following months. Dell and Rice (2005) found that a 6-d incubation was sufficient for microbial biomass to turn over applied ^{15}N.

The microbial biomass in the surface soil of the cropping system recovered more ^{15}N than in the riparian area (Table 5). This trend was somewhat unexpected. Our original hypothesis was that the riparian area microbial biomass would be the better competitor for ^{15}N due to lower native inorganic N concentrations (Ledgard et al., 1998; Griffith et al., 2003). Spring flooding and drying events in the riparian area may have increased competition for inorganic N by denitrifiers and reduced the activity of the more general microbial community and their capacity to immobilize N.

The cropping system microbial biomass contained more $^{15}NH_4^+$ than $^{15}NO_3^-$ in the 0- to 10-cm depth (Table 5). Preferential utilization of NH_4^+ over NO_3^- by microbes has been well established (Winsor and Pollard, 1956; Jansson, 1958). The presence of NH_4^+ has been shown to inhibit uptake of NO_3^- (Rice and Tiedje, 1989). In addition, preferential uptake of NH_4^+ occurs within the soil matrix because the spatial distribution of microbes brings them into closer proximity to the immobile NH_4^+ ions compared with plant roots (Schimel et al., 1989). Only about 1% of the ^{15}N label was found in the riparian microbial biomass, with similar amounts of $^{15}NH_4^+$ and $^{15}NO_3^-$ being recovered (Table 5). Dell and Rice (2005) found similar amounts of ^{15}N incorporated into MB and SOM pools despite the form of N applied. They suggest that microsite depletion of NH_4^+ may have led to microbial uptake of $^{15}NO_3^-$.

Soil N content in the top 10 cm was 2.3 ± 0.12 kg N plot^{-1} in the riparian area and 2.2 ± 0.05 kg N plot^{-1} in the cropping system and did not differ significantly between sites. In the 10- to 30-cm soil layer, however, total N in the cropping system soil increased to 3.1 ± 0.05 kg

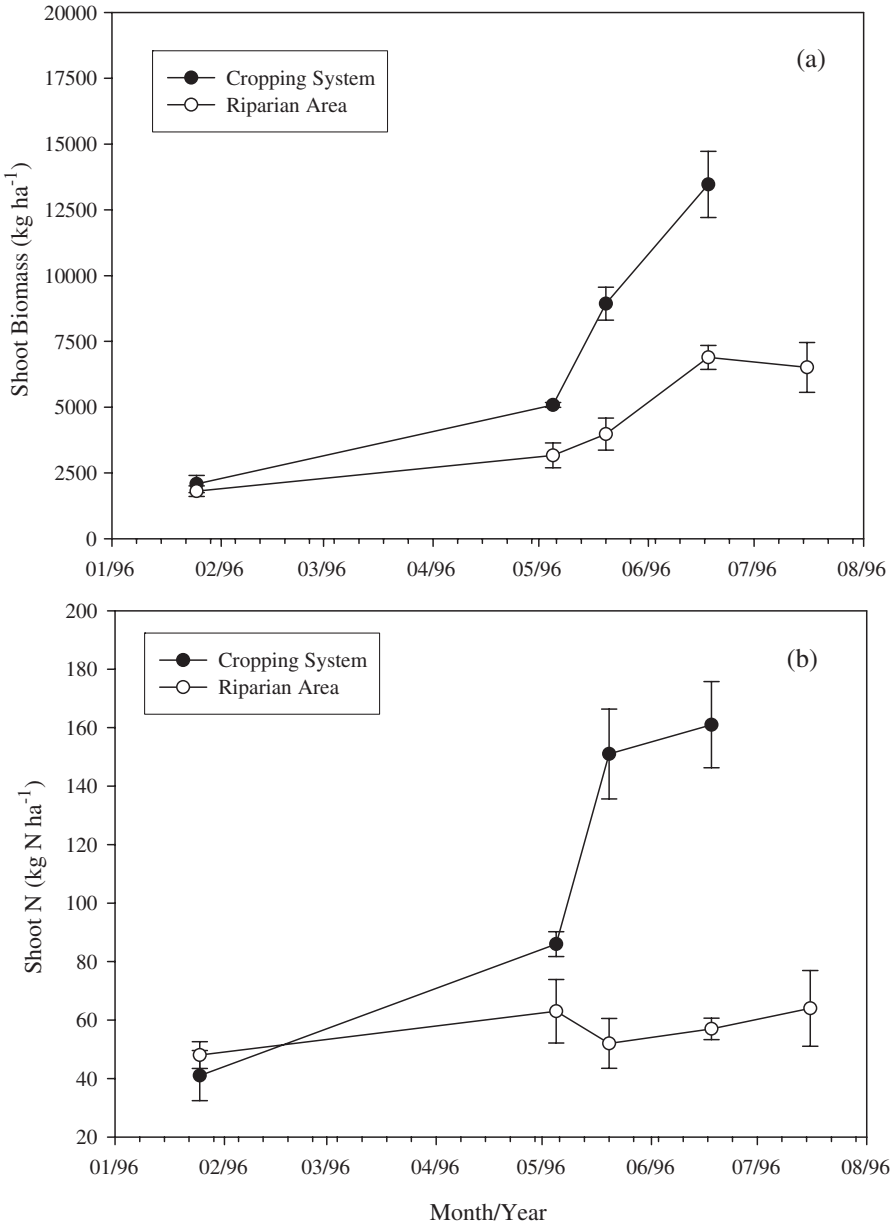

Fig. 2. Mean and standard error of (a) above ground plant biomass accumulation and (b) shoot N accumulation in plant samples collected from the cropping system and riparian area in 1996.

N plot^{-1} while the riparian soil decreased with depth to 1.8 ± 0.08 kg N plot^{-1}. Incorporation of plant residues into the cropping system soil likely led to the higher subsurface total N, although some fertilizer N applied at the surface may have leached into the 10- to 30-cm layer. The riparian soil lacked both fertilizer additions and tillage incorporation of plant residues. These results are similar to those of Needelman et al. (1999), who found higher soil organic matter in the surface soil and less in the subsoil of a no-till compared with conventionally tilled soil, leading to no overall greater SOM sequestration with no-till. All totaled, the top 30 cm of the cropping system soil profile contained significantly more total N (5.4 kg N per plot) than the riparian area (4.2 kg N per plot) most likely from fertilizer applications (Horwath et al., 1998).

A small percentage (<3.6%) of ^{15}N was recovered in the lower soil profiles (10–30 cm) of both the cropping system and riparian soil (Table 2). Paul and Juma (1981) conducted a similar experiment where $^{15}NH_4OH$ (56 kg N ha^{-1}) was added to a gray-black chernozemic soil planted with spring wheat. This study concluded that leaching was not a significant pathway for loss of ^{15}N because only 0.5% of the label was recovered in their B soil horizon.

Less ^{15}N was recovered from the 0- to 10-cm depth of the riparian area than from the cropping system (Table 2). Low recovery of ^{15}N in the riparian soil may have been the result of two significant spring flood events occurring in the riparian area but not in the cropping system. These events occurred approximately 7 and 35 d after the ^{15}N label was applied and significantly increased the potential of ^{15}N loss through overland flow. The ri-

Table 3. Differences between 1996 and 1997 mean percentage of recoveries for the cropping system and riparian area. Positive numbers indicate a net increase in percentage of recovery from 1996 to 1997 and negative numbers indicate a net decrease in recovery from 1996 to 1997.

		Total soil N % recovery			Plant biomass % recovery		Total plot % recovery
Site	^{15}N Tmt	0–10 cm	10–20 cm	20–30 cm	Shoot	Root	Total
Cropping System	$^{15}NH_4^+$	+5 ns†	+0.5**	−0.3 ns	−24 **‡	+3.7 ns	−16 ns
	$^{15}NO_3^-$	+16**	0.0 ns	−0.4 ns	−25*‡	−0.4 ns	−10 ns
Riparian Area	$^{15}NH_4^+$	+6.7**	0.0 ns	0.0 ns	−1.9*	−11**	−6 ns
	$^{15}NO_3^-$	+11*	−0.8 ns	−1.5 ns	−5.8 ns	−13*	−11 ns

* Significant at the 0.05 probability level.
** Significant at the 0.01 probability level.
† ns $p > 0.05$.
‡ Difference in shoot recovery takes into account seed removal from 1996 plant biomass.

parian area also experienced a significant drying period between these flood events when the water table dropped to an average of 20 cm below the soil surface. Nitrification would be promoted during this drying period and denitrification of subsequently formed $^{15}NO_3^-$ and applied $^{15}NO_3^-$ would have occurred during and/or after the second flood event making ^{15}N less available for incorporation into soil organic matter (Devevre and Horwath, 2001). The riparian zone has been shown to have higher denitrification potential that the cropping system (Davis, 2003).

More $^{15}NH_4^+$ (27%) than $^{15}NO_3^-$ (11%) was recovered from the cropping system surface soil in 1996 (Table 2) suggesting greater stabilization of $^{15}NH_4^+$ into organic matter and/or greater binding of $^{15}NH_4^+$ to soil particles. There was no difference between the amount of $^{15}NH_4^+$ and $^{15}NO_3^-$ retained in the riparian soil in 1996 or 1997. Nitrification of $^{15}NH_4^+$ during the dry period may have led to a more equitable loss of $^{15}NH_4^+$ and $^{15}NO_3^-$ by denitrification during the second flood event. In addition, greater uptake of $^{15}NO_3^-$ by the riparian vegetation may have been balanced by a greater loss of soil bound $^{15}NH_4^+$ through overland flow, both contributing to equal retention of $^{15}NH_4^+$ and $^{15}NO_3^-$ in the riparian soil.

In the surface soils (0–10 cm), 5 to 16% more ^{15}N was recovered as soil N in 1997 than 1996 from the riparian area and cropping system (Table 3). The increase in ^{15}N soil recovery in 1997 coincided with a decrease in plant biomass recovery indicating that the 1996 plant biomass ^{15}N was turned over and was subsequently incorporated into soil organic matter. This is similar to Dell et al. (2005) where ^{15}N lost in tallgrass prairie plant biomass from the second to fifth growing season after $^{15}NH_4^+$ was applied was accounted for in soil organic N pools. The differences in total plot (plant plus soil) recovery of ^{15}N from 1996 to 1997 were 6 to 16% less but were not significantly different.

Native soil N mineralized during aerobic incubations is shown in Fig. 3. A cumulative, first-order, one-pool model (Eq. [1]) provided a good fit to the native soil N mineralization data (r^2 = 0.81 to 0.98) (Table 6). In the riparian soil, both the size of the labile N pool and the mean residence time (MRT) decreased with depth, again showing the stratification effect from surface applications of plant residues. In the cropping system soil, labile soil N decreased with depth, but the rate constants (k) and MRT of the labile N pool did not change with depth (Table 6). The continuity of MRT in the cropping system most likely resulted from incorporation and homogenization into the plow layer of both fertilizer N and the perennial ryegrass crop during cultivation the previous year.

Neither the MRT nor the size of the labile native N pool were different between the cropping system surface soil and the riparian surface soil. However, fertilizer applications and incorporation of residues into the soil profile in the cropping system produced a larger available N pool with slower turnover in the subsurface than the riparian area. Hatch et al. (2000) also found that fertilized grass swards had larger available N pools which turned over more slowly than unfertilized grass swards and concluded that this combination increased the potential of the fertilized swards to lose N to the larger environment.

Mineralization of ^{15}N during aerobic incubations is shown in Fig. 4a and 4b. A cumulative, first-order, one-pool model (Eq. [1]) was also used on the surface soil ^{15}N mineralized during the incubation to provide rate constants for recently incorporated ^{15}N (r^2 = 0.96 to 0.98) (Table 7). Very little original ^{15}N was recovered from the mineralization of N at the lower soil depths (0.0- 0.9%) and therefore rate constants and pool sizes from 10 to 30 cm were not calculated. There was no significant difference between the surface soil rate constants of native soil N and added labeled ^{15}N, showing the rapid equil-

Table 4. Microbial biomass N in samples collected in May 1996. Letters denote significant differences between sites (cropping system and riparian area) at each depth ($p \leq 0.05$).

	Microbial biomass N, μg N g soil^{-1}		
Site	0–10 cm	10–20 cm	20–30 cm
Cropping system	48 (3.3) a	29 (1.7) a	18 (1.2) a
Riparian area	61 (3.9) b	16 (1.2) b	5.9 (0.8) b

Table 5. Mean and standard errors of the percentage of recovery of ^{15}N in soil microbial biomass samples collected in May 1996, 2 mo after the label was applied. Letters denote significant differences among sites (cropping system and riparian area) and ^{15}N treatments ($^{15}NH_4^+$ and $^{15}NO_3^-$) ($p \leq 0.05$).

		Microbial biomass ^{15}N % recovery		
Site	Label	0–10 cm	10–20 cm	20–30 cm
Cropping System	$^{15}NH_4^+$	5.2 (0.6) a	0.2 (0.08) a	0.01 (0.01) a
	$^{15}NO_3^-$	3.6 (0.6) b	0.8 (0.37) a	0.01 (0.01) a
Riparian Area	$^{15}NH_4^+$	1.0 (0.2) c	0.1 (0.06) a	0.00 (0.00) a
	$^{15}NO_3^-$	1.2 (0.1) c	0.1 (0.02) a	0.00 (0.00) a

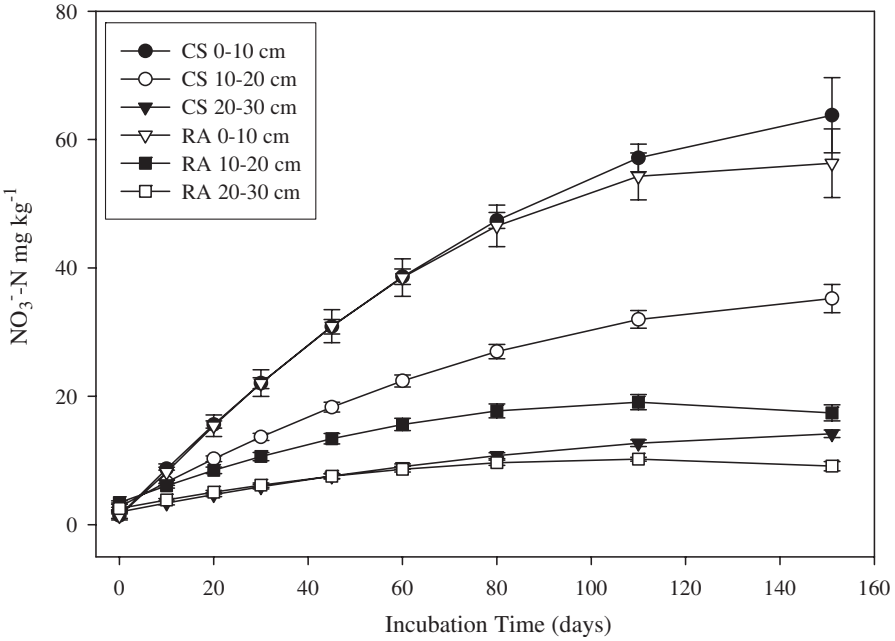

Fig. 3. Nitrate-N mineralized during aerobic incubations of cropping system (CS) and riparian area (RA) soil.

ibration of newly added N with labile N pools. Although the recently added ^{15}N was turned over at a similar rate in the cropping system and riparian area, the size of the recently added $^{15}N_{min}$ pool was significantly larger in the cropping system than the riparian area (Table 7). This indicates that there was more available N in the cropping system soil than the riparian soil, which correlates well with the accumulation of inorganic N (NO_3^-) in the cropping system during summer when plant N uptake is low and soils are dry (Griffith et al., 2003).

The double model (Eq. [3]) did not provide a better fit to the incubation data than the single pool model (data not shown). This was not surprising since studies have shown that the resistant fraction of organic N in grassland soils can take longer than 150 d to turn over (Ajwa et al., 1998; Collins and Allinson, 2002).

Unlike Kiovunen and Horwath (2004), parameter estimates from the first order modeling of our incremental data (Eq. [4]) gave the same trends for N_{min} and k as that of our cumulative data (Table 6). Similar to Kiovunen and Horwath (2004), r^2 values for nonlinear regression of the incremental data were much lower than for the cumulative data. Modeling incremental data has been shown to give more realistic estimates of parameters and their associated error due to the inherent problem of correlated residuals from using an integral model with the accumulated data (Hess and Schmidt, 1995).

CONCLUSIONS

We hypothesized that retention of ^{15}N would be higher in the riparian system than in the adjacent cropping system due to higher competition for N in an unfertilized system. We found however, that the cropping system retained more $^{15}NH_4^+$ and $^{15}NO_3^-$. The ^{15}N label was applied to the site in March and samples were collected in May, during the time when the photoperiod and soil temperature became conducive to plant growth. Greater retention of ^{15}N in the cropping system resulted from both greater uptake of N by the perennial ryegrass crop and greater retention of N in the soil than the riparian area. Low recovery of ^{15}N in the riparian area likely resulted from dynamic flooding and drying events in the spring which reduced riparian plant biomass production and N uptake and increased the potential of ^{15}N label loss through overland flow and denitrification. Additional research is warranted to look at how these systems behave in years with annual precipitation closer to average.

Table 6. First-order decay constants (k), mean residence time (MRT) and the amount of labile native soil N (N_{min}) in the cropping system and riparian soil from modeling the cumulative and discrete data. Letters denote significant differences among sites (cropping system and riparian area) and depths ($p \leq 0.05$).

		First order-cumulative				First order-discrete			
	Depth	r^2	k, d^{-1}	MRT, d	N_{min}, μg N g soil^{-1}	r^2	k, d^{-1}	MRT, d	N_{min}, μg N g soil^{-1}
	cm								
Cropping system	0–10	0.97	0.020 a	50 a	60 a	0.60	0.012 a	81 a	90 a
	10–20	0.97	0.018 a	55 a	34 b	0.76	0.010 a	103 a	63 b
	20–30	0.98	0.017 a	60 a	13 c	0.77	0.008 a	119 a	22 c
Riparian area	0–10	0.96	0.023 a	44 a	58 a	0.51	0.016 a	63 a	81 a
	10–20	0.94	0.032 b	30 b	15 c	0.53	0.020 a	50 a	22 c
	20–30	0.81	0.044 c	22 c	7.0 d	0.48	0.023 a	44 a	11 d

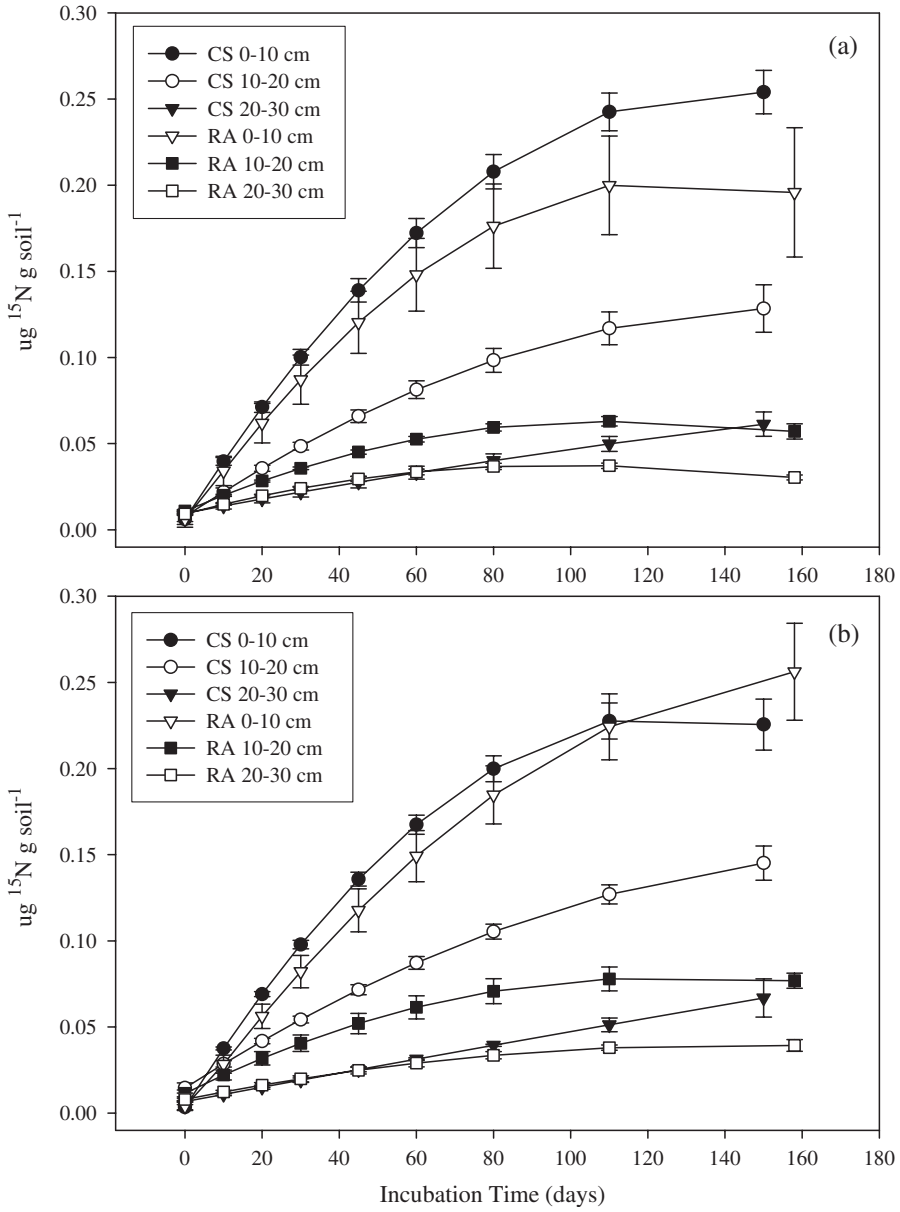

Fig. 4. Mineralization of ^{15}N during aerobic incubations of soil from (a) $^{15}NH_4NO_3$ and (b) $NH_4{}^{15}NO_3$–labeled plots in the cropping system (CS) and riparian area (RA).

This study indicates that even though the cropping system is more efficient at retaining newly applied N than the riparian zone, larger available N pools in the cropping system were potentially prone to leaching. These data are consistent with the conclusions drawn by the Oregon State University extension service that grass seed cropping systems in western Oregon are over fertilized (Horneck and Hart, 1988). Further research should investigate how N retention by the crop and soil changes in these grass seed cropping systems over the 3 to 4 yr that the crop is in production. Nitrogen dynamics in the riparian zone were greatly influenced by landscape position, (e.g., the proximity of this system to surface waters). Depending on the dynamics of the water table, excess N entering the riparian zone from the upland cropping system could be sequestered into soil organic matter or possibly lost from the system through denitrification and overland flow.

Table 7. Cumulative first-order decay constants (k), mean residence time (MRT) and the amount of ^{15}N mineralized during the 150-d incubation ($^{15}N_{min}$) in the cropping system and riparian soil. Letters denote significant differences among sites (cropping system and riparian area) and ^{15}N treatments ($^{15}NH_4^+$ and $^{15}NO_3^-$) ($p \leq 0.05$).

Site	Label	Depth, cm	r^2	k, d^{-1}	MRT, d	$^{15}N_{min}$, µg N g soil^{-1}
Cropping system	$^{15}NH_4^+$	0–10	0.98	0.033 a	30 a	0.015 a
	$^{15}NO_3^-$	0–10	0.98	0.015 b	67 b	0.015 a
Riparian area	$^{15}NH_4^+$	0–10	0.96	0.035 ab	28 ab	0.002 b
	$^{15}NO_3^-$	0–10	0.97	0.018 ab	56 ab	0.003 b

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