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TITLE: Modifications of Soil Nitrogen Pools in Response to Alfalfa Root Systems and Shoot Mulch

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ABSTRACT

Decomposing alfalfa (Medicago sativa L.) shoots and roots generate large amounts of NO[sub3]-N available to the next crop but also susceptible to deep leaching. This study was aimed at determining the specific contributions of above- and belowground alfalfa biomass to soil N pools. Dynamics of soil and plant N pools were studied in a Kalamazoo loam soil (fine-loamy, mixed, mesic Typic Hapludalfs) over a 2-yr period under bare fallow (BF), bare fallow to which alfalfa shoot mulch was applied (BFSM), living alfalfa plants with shoots removed after harvest (A), and living alfalfa with shoot mulch remaining on the soil surface after harvest (ASM). Organic N pools were monitored in alfalfa plant parts, soil-incorporated debris, and soil organic matter to depths of 150 cm. Inorganic N pools were monitored by suction lysimeters, soil extraction, and evaluation of soil denitrification rates. Living alfalfa stands kept soil inorganic N at very low levels, whether shoot mulch was applied or not. Soluble inorganic N concentrations decreased earlier in the fall in the upper horizons of bare fallow soils receiving alfalfa shoot mulch, suggesting enhanced leaching from bare soil under alfalfa mulch. Alfalfa crown and roots contained an average of 115 kg N ha[sup-1] after 2 yr of treatment. In conclusion, alfalfa shoot mulch contributed little to sustained increases in soil N pools, while crowns and roots contributed larger quantities to the soil N pool.

Abbreviations: A, living alfalfa plants (shoots removed after harvest); ASM, living alfalfa plants with shoots left as mulch after harvest; BF, bare fallow; BFSM, bare fallow with shoot mulch; PVC, polyvinyl chloride; SOM, soil organic matter; TDR, time-domain reflectometry.

THE FATE of soil inorganic N generated by decaying plant tissues and mineralized soil organic matter (SOM) is of crucial interest to sustainable agriculture. Reduced-input farming requires the optimal usage of green manure N by the following cash crop (Eltun, 1995). Green-manure N has to be contained in the top of the soil profile in order to (i) maximize N availability to crops, (ii) minimize NO[sub3]-N leaching losses to ground-water, and (iii) minimize N[sub2]O gaseous losses involved in the greenhouse effect (Peterson and Russelle, 1991).

Alfalfa (Medicago sativa L.) N[sub2] fixation contributes more than 1 billion kg N yr[sup-1] to N inputs in the U.S. Corn Belt (Peterson and Russelle, 1991). Net production of soil N (i.e., soil N gain + plant uptake) has been reported to be greater for alfalfa than red clover (Trifolium pratense L.), pea (Pisum sativum L.), and hairy vetch (Vicia villosa Roth) (Lyon and Bizzell, 1933). The improvement of the soil N status has been attributed to the high amounts of N[sub2] fixed by perennial alfalfa (Fox and Piekielek, 1988).

Alfalfa has been considered beneficial as well as detrimental to the reduction of nitrate leaching losses. Lamb et al. (1995) reported the great potential for alfalfa to absorb nitrates--especially during the spring and fall, when nitrate leaching is the greatest. The deep root system of alfalfa can recycle nitrates leached to depths inaccessible to other crops (Blumenthal and Russelle, 1996). On the other hand, mineralization bursts can lead to high levels of nitrate leached to the ground water if not matched by crop consumption (Campbell et al., 1994; Philipps and Stopes, 1995).

Establishing best management practices for cash crops following alfalfa plowdown requires the determination of the specific contributions by above- and belowground alfalfa plant parts to soil N pools. Alfalfa roots and shoots potentially differ in their N contents, decomposition rates, denitrification rates, and specific migration of mineralization products through the soil. Efficient storage of belowground N under alfalfa stands is influenced by the distribution over time of the different N pools (root N, SOM N, inorganic N) within the different soil horizons. Incorporation of fresh plant residues modifies SOM decomposition rates, or basal mineralization. Broadbent and Nakashima (1974) reported increased rates of SOM mineralization (called the priming effect), when plant residues were incorporated to soil. On the other hand, Watkins and Barraclough (1996) argued that the soil microbial activity can be diverted from SOM to plant tissue mineralization, thereby decreasing SOM mineralization rates. Soil incorporation of alfalfa shoots results in a direct increase in net SOM mineralization, due to the low C/N ratio of alfalfa tissues (Li and Mahler, 1995; Smith and Sharpley, 1990). Application of inorganic or easily mineralizable N increases alfalfa yields (Mathers et al., 1975; Schmitt et al., 1994). Most studies report that increased levels of soil inorganic N diminish symbiotic fixation (Cherney and Duxbury, 1994; Phillips and DeJong, 1984). Others report that alfalfa presents the remarkable ability to continue symbiotic fixation at high soil inorganic N levels (Lory et al., 1992; Lamb et al., 1995).

The objectives of our study were (i) to identify and quantify the storage pools for the N released from decaying alfalfa shoot tissues in soils under living alfalfa stands, (ii) to quantify and compare pathways of N losses in alfalfa systems in comparison with bare soil fallows, and (iii) to identify periods of maximum soluble inorganic N release attributable to soil organic matter, alfalfa shoot mulch, and alfalfa root systems.

MATERIAL AND METHODS

EXPERIMENTAL DESIGN AND TREATMENTS

A field experiment was conducted at the NSF-supported Long-Term Ecological Research (LTER) site at the Kellogg Biological Station in southwestern Michigan. Four treatments were considered: bare fallow (BF), bare fallow to which alfalfa shoots were applied after each harvest (BFSM), living alfalfa plants with shoots removed after harvest (A), and living alfalfa with shoot mulch remaining on the soil surface after harvest (ASM). Each treatment was replicated four times in a randomized complete block design. Experimental plots, 6 by 10 m each, were installed in a Kalamazoo loam soil (fine-

loamy, mixed, mesic Typic Hapludalfs) in late August 1994. The preceding crop was corn (Zea mays L.), fertilized at 123 kg N ha[sup-1]. Moldboard plowing was performed to a depth of 23 cm following corn harvest. All plots were tilled and trafficked equally. Alfalfa was planted in half of the plots at a rate of 22 kg ha[sup-1] on 30 Aug. 1994. The bare soil plots were also drilled without seeds. The bare soil plots were kept free of weeds by applications of glyphosate [N-(phosphonomethyl)-glycine] (7 L ha[sup-1]) at approximately 6-wk intervals between April and August of each year. Plots received no N fertilizer. According to soil testing, K was applied at a rate of 232 kg ha[sup-1], together with 2.2 kg ha[sup-1] of B, on 13 June 1996. Lime was applied at a rate of 2.5 Mg ha[sup-1] on 14 June 1996.

The alfalfa plots were harvested on 9 June 1995, 24 July 1995, 31 Aug. 1995, 31 May 1996, 3 July 1996, and 21 Aug. 1996. At harvest, alfalfa plants were cut 5 cm above the soil with a 90-cm-wide sickle-bar power mower. After cutting, the alfalfa shoots were raked from the A and ASM plots, and redistributed equally across both the ASM and BFSM plots. At the first harvest, the alfalfa was applied as uniformly as possible across all plots. The layer of alfalfa mulch proved to physically impede alfalfa regrowth. Consequently, subsequent applications were conducted by uniform manual applications of the alfalfa shoots between the rows. Alfalfa shoots were applied using similarly compacted containers (0.076 m[sup3]), some of which were sampled for density and moisture content. Similar quantities of alfalfa shoots were applied to BFSM and ASM treatments. At the end of the second growing season, cumulative shoot mulch applications approached 16.4 Mg ha[sup-1] of biomass, corresponding to total applications of 590 kg N ha[sup-1]. All sampled areas were located within 1 m from the edge of the plots, to avoid border effects. The surface area of each plot was allocated to (i) nondestructive sampling with in situ instruments (2 by 4 m), (ii) yield assessment (4 by 4 m), and (iii) destructive sampling (2 by 4 m). Plots were separated by surface plastic barriers installed to depths of 10 cm and protruding 5 cm above the soil surface, to prevent runoff and/or run-on between plots.

MEASUREMENTS

Alfalfa yields at each harvest were estimated by sampling on a nondisturbed 4- by 4-m area reserved for this purpose. Subsamples for moisture content were taken immediately, weighed, dried at 60°C, and reweighed. Subsamples were finely ground (<0.5 mm) for total C and N analyses by dry combustion method (Kirsten, 1983) using a CNS analyzer NA1500 series 2 (Carlo Erba Strumentazione, Milan, Italy).

Each field plot was equipped with 100 kPa high-flow, suction lysimeters (Model 1900, Soil Moisture, Santa Barbara, CA) for soil solution extraction. Three suction lysimeters per plot were installed at depths of 15, 35, and 60 cm, to intercept the soil solutions in the Ap, Bt[sub1], and Bt[sub2] horizons, respectively, as determined by preliminary soil sampling along the central area of each plot. The depth of the Bt[sub2]-C interface averaged 70 cm. Ceramic-cup suction lysimeters, installed at a 45° angle into the soil, were sampled by applying a negative pressure of 0.07 MPa for periods of 2 d. Following sampling, vials were immediately placed in a cooler in the field, and were frozen a few hours later in the laboratory. Soil solution samples were analyzed for NH[sub4]-N (USEPA, 1979) and NO[sub3]-N (USEPA, 1983) by spectrophotometry using a QuickChem automated flow injection analyzer (Lachat Instruments, Milwaukee, WI). Volumetric soil water contents were assessed using time-domain reflectometry (TDR) technology. Stainless steel TDR probes were inserted horizontally at 15, 35, and 60 cm in a small profile dug in every plot before planting, and shielded cables were brought to the soil surface. Instrument installation profiles were described and used for horizon delineation. The profiles were recompacted horizon by horizon and the cable was buried before planting. Parallel TDR probes, 0.5 cm in diameter, were assembled at the Michigan State University Soil Biophysics Laboratory. Effective length of the probe into the soil profile was 28.5 cm. TDR meter readings were collected from the cables at the soil surface using a Tektronix cable tester Model 1502C (Tektronix, Beaverton, OR). The Topp's equation was used for TDR curve analysis (Topp et al., 1980).

Disturbed soil samples were extracted on 14 May and 12 Oct. 1996 with a Giddings hydraulic probe (Giddings Co., Ft. Collins, CO) equipped with an 8.9-cm-diameter probe. Spring samples were taken to depths of 150 cm. Dry soils often limited the depths of fall samplings to the middle regions of the Bt[sub2] horizon, as the hydraulic probe could not be inserted through the deeper dry clay layers of the Bt[sub2] horizon. Two cores were extracted from each plot and divided longitudinally into two equal halves. This lengthwise division was conducted by placing the undisturbed core in a longitudinally split PVC pipe of identical diameter to the soil core and cutting the two halves of the soil sample with a sharp stainless steel knife. Each half-core was then divided into Ap, Bt[sub1], Bt[sub2], C[sub1], and C[sub2] horizons. One half was used for inorganic N and total C and N analyses. Roots were washed from the other half. Gravimetric soil water contents were determined on subsamples of soils dried in a forced-air oven at 105°C for 24 h. Field-moist 20-g subsamples were extracted for NO[sub3]-N and NH[sub4]-N by shaking for 1 h in 50 mL of 1 M KCl. Solutions were filtered through KCl-rinsed Whatman no. 1 paper and analyzed for NH[sub4]-N and NO[sub3]-N by spectrophotometry using a QuickChem automated flow injection analyzer. The remaining part of the soil samples was air-dried and finely ground (<0.5 mm) for total C and N analyses, by the dry combustion method using a CNS analyzer.

Alfalfa roots were extracted from the soil matrix by hydropneumatic elutriation (Smucker et al., 1982) and stored at 4°C in 20% (v/v) methanol solution. The diameter of the Giddings probe (8.9 cm) was considered insufficient for proper root estimations in the upper 15 cm of the soil profile, as it did not accurately represent the surface ratio of row to interrow. Consequently, two 200-cm[sup2] samples per plot were taken in October 1996 to depths of 15 cm, centered on an alfalfa row and extending into one half of an interrow on each side. Alfalfa plants were clipped at the crowns and the soil surface was cleared of debris. Samples were washed by hydropneumatic elutriation, then divided into roots and soil-incorporated organic debris. Each fraction was analyzed separately for total C and N by the dry combustion method.

The Ap horizon was sampled on 15 June 1995, 25 Nov. 1995, and 12 Oct. 1996 to a depth of 23 cm to monitor total C, N, and the C/N ratio. Ten subsamples per plot were collected, mixed into one composite sample, air-dried, and finely ground (<0.5 mm).

Alfalfa shoot mulch biomass remaining on the soil surfaces of the BFSM and ASM plots at the end of the second year were sampled in December 1996. Two 30-cm-diameter PVC rings were randomly positioned on each plot, and all mulch material was carefully hand-picked inside the ring. The two samples per plot were combined, oven-dried at 60°C, finely ground (<0.5 mm), and analyzed for total C and N by the oxygen-enriched dry combustion method, using a CNS analyzer.

Soil denitrification rates were estimated in June 1996 by acetylene inhibition in static cores (Tiedje et al., 1989). Five 15-cm-long cores were collected per plot in the first three replicated blocks. Cores were immediately capped with rubber stoppers, kept in the shade in the field, and injected with acetylene directly after collection of the entire batch of samples.

STATISTICAL ANALYSES

Data sets with a maximum of six sampling dates were analyzed separately by date, using the general linear model procedure of the SAS system (SAS Inst., 1989). Data were analyzed considering four individual treatments, with means and Fisher's LSD (0.05) reported. Denitrification data, often reported as log-normally distributed (Tiedje et al., 1989), were tested for normality. Data were not improved upon by logarithmic transformation, so analyses proceeded on the original scale of measurement.

Suction lysimeter data, including more than 20 measurement dates, were analyzed using univariate repeated measures techniques with a spatiotemporal correlation structure based on the mixed procedure of the SAS system (Gregoire et al., 1995; Schabenberger and Gregoire, 1995; Littell et al., 1996). This technique permits inference on main effects, simple effects, and interactions between treatments and treatments × time. The analysis accounts for the fact that repeated observations on the same experimental unit are not independent. Since measurements were collected at three depths at every given point in time, multiple correlations over time and depths needed to be incorporated. Traditional methods of analysis of variance, such as square decompositions and means comparisons, are inappropriate in this case. The unequal spacing of measurements in time, along with the pattern of missing observations for certain depths, required a continuous spatiotemporal correlation process. Thorough investigation of various candidate correlation models resulted in the choice of an exponential correlation model. The treatment structure was entered as a 2 × 2 factorial with a plant factor P (levels: alfalfa and bare soil) and a mulch factor M (levels: applied and not applied) to gain further insight in the interaction structure. Analyses were conducted separately for 1995 and 1996. Soluble inorganic N contents were statistically modeled for each treatment in 1995 and 1996 (Fig. 1 and 2), using polynomial functions in time.

RESULTS AND DISCUSSION

HERBAGE BIOMASS AND N CONTENTS

Alfalfa herbage yields were significantly decreased in 1995 by application of harvested shoots, while a significant yield increase by shoot application was observed in 1996 (Table 1). Initial yield reduction by alfalfa shoot mulch appears to have resulted from the physical impedance to shoot growth from the randomly scattered mulch. For 1996, the significant yield increase is consistent with other studies reporting that application of inorganic or easily mineralizable N increases alfalfa yields (Mathers et al., 1975; Schmitt et al., 1994). Nitrogen concentration of the shoots was not significantly altered by shoot application at harvest (Table 1). Groya and Sheaffer (1985) also reported that alfalfa shoot application did not significantly alter total alfalfa N yields in the seedling year. However, total N contents of alfalfa herbage per harvest mimicked biomass yields, where 169 kg N ha[sup-1] were harvested from the ASM treatment for the first harvest of the second growing season. The amount of N from alfalfa shoot mulch that was directly recycled into the growing alfalfa plant is difficult to assess, as the rate of N[sub2] fixation can be influenced by soil nitrate levels (Heichel et al., 1984; Cherney and Duxbury, 1994; Kelner et al., 1997). During the second growing season, plant N contents totaled 100 kg N ha[sup-1] more in the ASM than in the A treatments (329 vs. 229 kg N ha[sup-1]), indicating that alfalfa plants absorbed at least this amount from the mineralization of surface applied alfalfa shoots. Alfalfa plant N in the ASM treatment increased 152 kg N ha[sup-1] from the first to the second year, but increased only 25 kg N ha[sup-1] for the A treatment.

ROOT, CROWN, SOIL-INCORPORATED DEBRIS, AND MULCH N CONTENTS

Dry biomass of alfalfa roots was not significantly altered by shoot application after 2 yr of treatment. Root biomass, manually sampled to 15 cm, averaged 3.55 Mg ha[sup-1] on 12 Oct. 1996 (Table 2). Root biomass, coresampled between depths of 26 to 55 cm, averaged 0.39 Mg ha[sup-1] on 11 Oct. 1996 (data not shown). Reported alfalfa root biomass varies greatly among studies. Groya and Sheaffer (1985) reported dry matter root biomass of 2.5 Mg ha[sup-1], with samples collected to depths of 15 to 20 cm, using less quantitative methods of root recovery. Lory et al. (1992) extracted, by hand picking and sieving, 3.22 Mg and 0.83 Mg of alfalfa root biomass per hectare to depths of 0 to 35 cm and 35 to 70 cm, respectively. Blumenthal and Russelle (1996), who sampled directly over 17-mo-old alfalfa plants, reported up to 7.85 Mg ha[sup-1] of dry biomass alfalfa roots in the top 35 cm of the soil profile and 1.12 Mg ha[sup-1] from 37 to 70 cm. In our study, N contents of the roots were not significantly modified by alfalfa shoot mulch application (Table 2).

Alfalfa crown biomass and N content were not significantly modified by application of alfalfa shoot mulch (Table 2). Total amounts of N present in alfalfa crowns and roots (0-15 cm) in October 1996 averaged 86 and 119 kg N ha[sup-1] for A and ASM, respectively. These estimations are raised to 97 and 133 kg N ha[sup-1], respectively, when root contents of the Bt[sub1] are considered (data not shown). These results are consistent with values reported by Hesterman et al. (1986), ranging from 85 to 106 kg N ha[sup-1] in alfalfa crowns and roots in the fall of the seeding year. Soil-incorporated debris presented highly significant ($P < 0.01$) increases in N concentration with shoots applied to the soil surface (Table 2). However, total biomass of soil-incorporated debris were not significantly modified by treatment. This suggests that little surface-applied alfalfa mulch was incorporated into the Ap horizon during the 2 yr of treatment.

Decomposition and mineralization of alfalfa shoot mulches were greatly enhanced by the presence of a living alfalfa stand. Total dry matter biomass of alfalfa mulch remaining on the soil surface in December 1996 showed a highly significant ($P < 0.01$) treatment effect, and averaged 3747 kg ha[sup-1] for ASM and 10067 kg ha[sup-1] for BFSM treatments (data not shown). Neither N concentrations (avg. 22.4 g N kg[sup-1]) nor C/N ratios (avg. 17.9) of the remaining mulch were significantly altered by treatment (data not shown). Total N contents in remaining mulch were 84 and 225 kg N ha[sup-1] in ASM and BFSM, respectively. The alfalfa shoot mulch contained from 14% (ASM) to 38% (BFSM) of the total shoot N applied over the 2-yr period.

SOLUBLE SOIL INORGANIC N

Inorganic N concentrations (i.e., the sum of NO[sub3]-N and NH[sub4]-N concentrations) of suction lysimeter extractions were combined with TDR volumetric soil water contents and soil bulk density for each horizon depth to report soluble inorganic N contents in kilograms per hectare (the same units as for 1 M KCl soil extractions).

Few changes in soluble inorganic N were observed on the alfalfa plots containing living plant root systems, while the bare soil exhibited high soil N levels (Fig. 1 and 2). Alfalfa stands kept inorganic N contents in the soil solution at very low levels throughout the year. Soluble soil N contents became significantly lower under alfalfa stands than under bare fallow

soils at all depths, whether shoot mulch was applied or not (Table 3). Alfalfa roots absorbed nearly all nitrates from the soil solution at any sampling depth, regardless of shoot mulch, resulting in no significant shoot mulch effect on alfalfa plots (Table 4). These results confirm the high prevention potential of alfalfa for nitrate leaching previously reported by several authors (Lamb et al., 1995; Blumenthal and Russelle, 1996; Kelner et al., 1997).

Peaks of maximum soluble N were reached under BFSM treatment in early October, November, and December 1995 in the Ap, Bt[sub1], and Bt[sub2] horizons, respectively. Similar dynamics were observed under BF treatment; however, peaks of soluble inorganic N were delayed compared with BFSM treatment (Fig. 1). These findings suggest that substantial inorganic N leaching occurred under bare fallow (BF and BFSM) and that alfalfa shoot mulch application promoted leaching earlier in the fall. Hence, soluble inorganic N contained in the Ap horizon of bare fallow soils during the spring of 1996 was significantly lowered by previous alfalfa shoot mulch application (Fig. 2; Table 4). In fall 1996, soluble inorganic N contents in bare fallow soils peaked earlier in upper than in deeper horizons, which confirms the leaching dynamics observed in 1995. In contrast to 1995, however, no temporal differences in leaching patterns were observed between BF and BFSM plots. This probably was due to the excessively dry growing season (Rasse and Smucker, 1999).

The time sequence of soluble inorganic N contents in the different horizons was used to estimate soluble inorganic N gains and losses for BF and BFSM treatments. It was assumed that there was no significant upward movement of soil nitrates in these profiles. Each plot was uniquely characterized by the depths of its horizons, and soluble inorganic N gains and losses were summed over the top 70 cm of the soil profile. Soluble inorganic N gain in a horizon was accounted for only if it could not be explained by a leaching loss from an upper horizon. Soluble inorganic N loss from each horizon was accounted for only if it could not be retrieved in lower horizons. Differences in soluble inorganic N contents for each horizon between two dates represent the minimum value for N gain or N loss, as both mineralization and leaching potentially took place between the two measurement dates. For example, on 28 June 1996 soluble inorganic N contents were 39 (Ap), 23 (Bt[sub1]), and 22 kg N ha[sup-1] (Bt[sub2]), while on 11 July 1996 they were 36 (Ap), 32 (Bt[sub1]), and 16 kg N ha[sup-1] (Bt[sub2]) in the BF treatment (Fig. 2). Minimum N loss from the 70 cm profile between these two dates was therefore 6 kg N ha[sup-1], corresponding to the decrease in the Bt[sub2] horizon. On the other hand, minimum N gain between these two dates was also 6 kg N ha[sup-1], corresponding to an apparent gain of 9 kg N ha[sup-1] in the Bt[sub1] horizon, compensated by a leaching loss of 3 kg N ha[sup-1] from the Ap horizon. Using this methodology, inorganic N gains and losses were computed for every pair of consecutive dates and summed for the period extending from 26 June 1995 to 13 Dec. 1996. Minimum amounts of soil soluble N produced by mineralization under bare fallow were 255 kg N ha[sup-1] without shoot mulch application and 365 kg N ha[sup-1] when shoot mulch was applied. Estimated minimum leaching losses during the same period of time were 202 kg N ha[sup-1] for the BF treatment and 292 kg N ha[sup-1] for the BFSM treatment.

EXTRACTABLE SOIL INORGANIC N

Soil inorganic N contents, as extracted by 1 M KCl, were always higher than soluble inorganic N from suction lysimeters. When substantial amounts of inorganic N were present in the soil solution (i.e. in the BF and BFSM plots), soluble and extractable inorganic N contents followed an exponential relationship (r[sup2] = 0.86). In spring 1996, extractable inorganic N contents in BF and BFSM soils were five times higher than in A and ASM soils (Table 5). Highest inorganic N contents were found in the Bt[sub1] horizon of BF plots and the C[sub1] horizon of BFSM plots. These findings confirm suction lysimeter data showing higher nitrate leaching from fall to spring when shoots were applied to bare soils. Extractable inorganic N contents were increased in BF and BFSM treatments from May to October 1996, while A and ASM showed little differences over time (Table 5). The Ap horizons of the BFSM plots accumulated an average of 163 kg NO[sub3]-N + NH[sub4]-N ha[sup-1], increasing from 15 to 178 kg N ha[sup-1]. Total inorganic N accumulations over summer and fall 1996 in the Ap and Bt horizons were 205 kg N ha[sup-1] in BFSM plots and 87 kg N ha[sup-1] in BF plots. During the same period of time, suction lysimeter data indicate that minimum losses of soluble inorganic N out of the Ap and Bt horizons were 67 kg N ha[sup-1] for BF and 60 kg N ha[sup-1] for BFSM. Consequently, total inorganic N produced from May to October 1996 was 154 and 265 kg N ha[sup-1] for BF and BFSM, respectively.

TOTAL SOIL C AND N

Total C and N levels in the Ap horizon were analyzed in spring and fall of 1995 and 1996 (data not shown). No significant differences between treatments were observed for any date. Data analyses were complicated by a very strong gradient of SOM contents along the 96-m transect of the 16 plots. The block effect for total C was pronounced ($P < 0.001$), with C contents 35% greater in Block 2 than in Block 4. A decreasing trend in total C content was observed in the Ap horizon of the BF treatment, from 10.1 g C kg[sup-1] in June 1995 to 9.0 g C kg[sup-1] in October 1996. Nevertheless, this variation proved to be nonsignificant. Angers (1992) reported that a minimum of 3 yr were necessary to observe significant differences in SOM contents between alfalfa and bare soil plots in Canada.

Deep samples, taken to depths of 150 cm on 14 May and 12 Oct. 1996, revealed increased total C contents in the Bt[sub1] and Bt[sub2] horizons for A and ASM treatments and decreased C contents for BF and BFSM treatments (Table 6). Total C and N and C/N ratios were not significantly affected by treatments in May 1996. By fall 1996, C content in the Bt[sub1] horizon of ASM plots was significantly higher than in BFSM plots (Table 6). Soil C/N ratio in the Bt[sub1] horizon of BFSM plots was significantly lower than in any other plots. Factorial analysis for October 1996 showed total C contents and C/N ratios of the Bt[sub1] horizon to be significantly higher under living alfalfa stands than under bare fallow soils, whether shoots were applied or not (data not shown). These results suggest that alfalfa root systems have a substantial effect on SOM levels below the Ap horizon.

Total amounts of soil organic C in the Ap, Bt[sub1], and Bt[sub2] horizons were computed on the basis of horizon thickness, bulk density and C content. In 1996, bare fallow soils contained 57 632 kg C ha[sup-1] on 14 May and 55 885 kg C ha[sup-1] on 12 October. The mineralization rate of the SOM, inferred from this decrease in total C from May to October 1996 was 3.0%. Mineralization rates of the SOM were also computed from the amount of N mineralized in the bare fallow plots. SOM mineralization was the only source of inorganic N in the soil profile of the BF plots, excluding atmospheric deposition. Total soil N mineralized in the Ap, Bt[sub1], and Bt[sub2] horizons (top 70 cm) of BF plots from 14 May to 12 Oct. 1996 was assessed at 154 kg N ha[sup-1], as mentioned at the end of the previous section. Average C/N ratio of SOM in BF plots was 9.9, as adjusted to respective amounts of SOM in Ap, Bt[sub1], and Bt[sub2] horizons.

Consequently, the total amount of soil organic C corresponding to the mineralized N was approximately 1532 kg C ha[sup-1]. This value, compared with total soil C contents, corresponds to a SOM mineralization rate of 2.6%. Consequently, differences in soil organic C contents and assessment of mineralized N by suction lysimeters and soil extractions gave comparable results with respect to mineralization rates of the SOM. These results are similar to evaluations made by Barber (1979), who found that 2.4% of the SOM was lost annually to mineralization during a 6-yr study of a bare Raub silt loam soil in Indiana.

DENITRIFICATION

Denitrification rates, measured on 16 June 1996, were always higher when alfalfa shoots were applied to either bare or alfalfa plots (Table 7) and were consistent with a previous report of higher denitrification rates following fresh residue applications (Avalakki et al., 1995). Average denitrification rates doubled from the BF treatment (76 g N ha d[sup-1]) to the A treatment (159 g N ha d[sup-1]). Alfalfa shoot application increased denitrification rates of bare soil and alfalfa plots on 16 June 1996 by nearly 250 g N ha d[sup-1]. Van Kessel et al. (1993) reported an average denitrification rate of 331 g N ha[sup-1] d[sup-1] in a pea field during the month of June in Saskatchewan. Since denitrification is a highly variable process, both temporally and spatially (Tiedje et al., 1989), it is difficult to convert these results into kilograms of N lost over the season. The data do, however, represent a reasonable range of denitrification values.

CONCLUSIONS

Our study demonstrates that nitrate leaching is not an environmental risk under living alfalfa stands, not even when large quantities of organic N are applied as alfalfa shoots to the soil surface. In bare fallow systems, much of the mineralized N from shoot mulch and SOM was leached from upper regions of the soil profile during the cold season. These results imply that alfalfa stands should be kept alive into the spring for as long as possible before planting successive crops. Living alfalfa stands promote nitrate removal from the soil profile by actively absorbing root systems and prevent early N release into the vadose zone when above- and belowground portions of alfalfa plants decompose following spray killing. Alfalfa crowns and roots retained an average of 115 kg N ha[sup-1] by the end of the second growing season. This conservative estimation, which excludes root activities deeper than the Bt[sub1] horizon, does not take into account recently deposited SOM from alfalfa root turnover. We therefore conclude that spray killing of perennial alfalfa plants provides substantial quantities of slow-release soil N from alfalfa root systems alone.

ADDED MATERIAL

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Table 1. Alfalfa herbage yield and total N content for plots with alfalfa shoots removed at harvest (Treatment A) and with alfalfa shoot mulch remaining on the soil surface after harvest (Treatment ASM).

	Yield		N conc.		Total N	
Harvest date	A	ASM	A	ASM	A	ASM
	kg ha[sup-1]		g kg[sup-1]		kg ha[sup-1]	
9 June 1995	1393	1045	35.7	35.2	50	37
24 July 1995	2960	2517	32.3	30.2	95	77
31 Aug 1995	1439	1567	41.0	39.8	59	63
Total 1995	5792(FN**)	5129			204(FN**)	177
31 May 1996	3102	4163(FN*)	37.8	40.7	118	169(FN*)
3 July 1996	1782	1946	34.5	35.9	62	70
21 Aug 1996	1512	2792	32.2	32.9	49	90
Total 1996	6396	8901(FN*)			229	329(FN*)

FOOTNOTE

*, ** Within dates, significant treatment difference at P [less or equal] 0.05 and P [less or equal] 0.01, respectively.

Table 2. Biomass and N content of alfalfa crowns, roots, and soil-incorporated organic debris for plots with alfalfa shoots removed at harvest (Treatment A) and with alfalfa shoot mulch remaining on the soil surface after harvest (Treatment ASM), sampled to a depth of 15 cm on 12 Oct. 1996.

Biomass pool	Treatment	Biomass yield	N conc.	Total N
		kg ha[sup-1]	g kg[sup-1]	kg ha[sup-1]
Crowns	A	889	23.3	20.7
	ASM	1274	25.3	32.2
Roots	A	3329	19.7	65.6
	ASM	3776	23.0	86.8
Soil debris	A	1070	17.6	18.8
	ASM	1115	22.6(FN**)	25.1

FOOTNOTE

** Within biomass pools, significant treatment difference at P [less or equal] 0.01.

Table 3. Simple effects tests for the factor Plant: comparison of bare fallow (BF) with alfalfa stand (A) and bare fallow

with alfalfa shot mulch (BFSM) to alfalfa with alfalfa shoot mulch (ASM), from repeated measures analyses of soluble mineral N contents for 1995 and 1996.

		Significance of Plant effect	
Treatment comparison	Depth cm	1995	1996
BF vs. A	15	(FN***)	(FN***)
BFSM vs. ASM	15	NS(FN+)	(FN***)
BF vs. A	35	(FN**)	(FN***)
BFSM vs. ASM	35	NS	(FN***)
BF vs. A	60	NS	(FN**)
BFSM vs. ASM	60	NS	(FN***)

FOOTNOTES

, * Significant at P [less or equal] 0.01, and P [less or equal] 0.001, respectively.
+ Nonsignificant at P [less or equal] 0.05.

Table 4. Simple effects tests for the factor Mulch: application or no application of alfalfa shoot mulch (SM) on bare fallow (BF) and alfalfa plots (A), from repeated measures analyses of soluble mineral N contents for 1995 and 1996.

		Significance of Mulch effect	
Treatment comparison	Depth cm	1995	1996
BF vs. BFSM	15	(FN***)	(FN**)
A vs. ASM	15	NS(FN+)	NS
BF vs. BFSM	35	NS	NS
A vs. ASM	35	NS	NS
BF vs. BFSM	60	NS	NS
A vs. ASM	60	NS	NS

FOOTNOTES

*, *** Significant at P [less or equal] 0.05, and P [less or equal] 0.001, respectively.
+ Nonsignificant at P [less or equal] 0.05.

Table 5. Extractable mineral N content (NO[sub3]-N + NH[sub4]-N) per horizon in a Kalamazoo loam soil under bare fallow (BF), bare fallow to which alfalfa shoot mulch was applied after each harvest (BFSM), and living alfalfa plants with shoots removed after harvest (A), and living alfalfa plants with shoot mulch remaining on the soil surface after harvest (ASM).

		Extractable mineral N						
		14 May 1996				12 Oct. 1996		
Horizon	BF	BFSM	A	ASM	BF	BFSM	A	ASM
				kg ha[sup-1]				
Ap	40.4a(FN+)	15.2b	6.7bc	7.4c	123.0a	178.5a	7.2b	11.7b
Bt[sub1]	54.4a	28.0b	4.6b	6.9b	51.2b	69.5a	3.4c	5.4c
Bt[sub2]	31.8a	30.5a	3.0b	5.3b	39.6a	26.9b	3.3c	4.2c
C[sub1]	39.3b	60.3a	14.6c	14.0c	43.2a	49.1a	--	--
C[sub2]	29.4ab	33.3a	7.6c	12.3bc	--	--	--	--

FOOTNOTE

+ Within horizons and dates, means followed by the same letter are not significantly different according to Fisher's LSD (0.05).

Table 6. Soil carbon and N contents in the Bt[sub1] and Bt[sub2] horizons of soils under bare fallow (BF), bare fallow to which alfalfa shoot mulch was applied after each harvest (BFSM), and living alfalfa plants with shoots removed after harvest (A), living alfalfa with shoot mulch remaining on the soil surface after harvest (ASM).

	Total C		Total N		C/N ratio	
Treatment	Bt[sub1]	Bt[sub2]	Bt[sub1]	Bt[sub2]	Bt[sub1]	Bt[sub2]
		g kg[sup-1]				
BF	2.99ab(FN+)	1.86a	0.340a	0.240a	8.73a	7.60a
BFSM	2.95b	1.71a	0.348a	0.233b	8.47b	7.58a
A	3.21ab	2.35a	0.350a	0.270a	9.18a	8.64a
ASM	3.47a	2.65a	0.375a	0.300a	9.22a	8.74a

FOOTNOTE

+ Within horizons, means followed by the same letter are not significantly different according to Fisher's LSD (0.05).

Table 7. Denitrification rates of bare fallow (BF), bare fallow to which alfalfa shoot mulch was applied after each harvest (BFSM), living alfalfa plants with shoots removed after harvest (A), and living alfalfa with shoot mulch remaining on the soil surface after harvest (ASM), on 16 June 1996. Depth: 0-15 cm.

Treatments	Denitrification rates g N[sub2]O-N ha[sup-1] d[sup-1]
BF	76c(FN+)
BFSM	323ab
A	159bc
ASM	419a

FOOTNOTE

+ Means followed by the same letter are not significantly different according to Fisher's LSD (0.05).

Fig. 1. Soluble inorganic N contained in the top three horizons of soils under bare fallow (BF), bare fallow to which alfalfa shoots were applied after each harvest (BFSM), living alfalfa plants with shoots removed after harvest (A), and living alfalfa with shoots remaining on the soil surface after harvest (ASM) in 1995. Trends are derived from polynomial repeated measures analysis. R[sup2] values are empirical coefficients of determination based on the observed and fitted values for all treatments calculated separately for each depth. See Tables 3 and 4 for significant probability comparisons.

Fig. 2. Soluble inorganic N contained in the top three horizons of soils under bare fallow (BF), bare fallow to which alfalfa shoots were applied after each harvest (BFSM), living alfalfa plants with shoots removed after harvest (A), and living alfalfa with shoots remaining on the soil surface after harvest (ASM) in 1996. Trends are derived from polynomial repeated measures analysis. R[sup2] values are empirical coefficients of determination based on the observed and fitted values for all treatments calculated separately for each depth. See Tables 3 and 4 for significant probability comparisons.

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