

NITROGEN DYNAMICS AFTER ALFALFA AS INFLUENCED BY  
TERMINATION TECHNIQUE

BY

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A Thesis  
Submitted to the Faculty of Graduate Studies  
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for the Degree of

DOCTOR OF PHILOSOPHY

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**Ramona Maria Mohr**

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## ABSTRACT

Herbicide application may provide an alternative to intensive tillage for the termination of established alfalfa (*Medicago sativa*) stands but may alter N dynamics in the plant-soil system. A series of field and controlled environment studies was conducted to quantify and identify mechanisms influencing the plant-available N supply following alfalfa termination. From 1992 to 1993, four field experiments were initiated to determine the effect of a factorial combination of three methods (herbicide, tillage, herbicide plus tillage) and two times (early-summer after the first cut of alfalfa or late-summer after the second cut of alfalfa) of termination on soil  $\text{NO}_3^-$  status and N accumulations by subsequent spring wheat (*Triticum aestivum*). At three of four sites, tillage or herbicide+tillage treatments produced a larger short-term plant-available N supply than herbicide application alone; however, grain yields in herbicide treatments were similar to or higher than tilled treatments. Regardless of termination method, delaying termination reduced the available N supply. Controlled environment studies showed that reductions in the available N supply under herbicide-terminated alfalfa are primarily due to reduced mineralization of surface residues and partly due to increased volatile N losses from surface residues. In controlled environment studies, cumulative N uptake by barley (*Hordeum vulgare*) established in residue-incorporated treatments was nearly double that in residue-unincorporated treatments 125 d after alfalfa termination. Volatile N losses were <1% of the N in soil-incorporated legume top-growth, but were 8-12% of surface-applied legume N by 95 d after termination. In a subsequent  $^{15}\text{N}$  study, in herbicide treatments in which alfalfa top-growth was retained on the soil surface, 1%

of the  $^{15}\text{N}$  present was recovered in barley top-growth, 8% in soil and 91% in residues; however, in tillage treatments in which alfalfa top-growth was incorporated, 10% of the  $^{15}\text{N}$  present was recovered in barley top-growth, 52% in soil and 38% in residues. Termination technique did not affect the distribution of  $^{15}\text{N}$  from labelled alfalfa roots. In summary, termination of alfalfa by herbicide application may improve synchrony between N release and N needs of a subsequent wheat crop. Although surface residues in herbicide treatments may be subject to small N losses via volatilization, reductions in the short-term plant-available N supply may decrease the potential for leaching and denitrification losses thereby increasing the N use efficiency of subsequent crops. Despite a smaller short-term available N supply, herbicide termination allowed sufficient mineralization to attain grain yields similar to, or greater than, termination by tillage.

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## 1. INTRODUCTION

Concerns regarding agricultural and environmental sustainability have generated interest in the development of crop management systems that reduce inputs of non-renewable resources but maintain soil and environmental quality. Incorporating perennial legumes like alfalfa into today's cropping systems may be one approach. Legumes not only contribute substantial amounts of symbiotically-fixed N to the plant-soil system (Heichel et al. 1984; Kelner and Vessey 1995), but also may improve soil physical condition (Mazurak et al. 1955; Toogood and Lynch 1959; MacRae and Mehuys 1985; Blackwell et al. 1990), reduce the population of certain weeds (Harvey and McNevin 1990; Entz et al. 1995), and remove deep-leached nitrates from the soil profile (Schertz and Miller 1972; Mathers et al. 1975).

Although increases in total forage area are restricted by the demand for forage, by reducing the duration of forage stands, the proportion of arable land deriving benefits from legumes could be increased without increasing total acreage. Under current management systems in western Canada, alfalfa stands are maintained in crop rotations for extended periods that often exceed both the economic optimum and the period required to obtain N benefits. One factor that contributes to these long-term stands is that established alfalfa is difficult to terminate. Therefore, management practices that effectively terminate alfalfa but maximize the benefits derived from alfalfa must be developed to improve the feasibility of short-term alfalfa stands.

Alfalfa usually is terminated by intensive tillage that may leave the exposed soil prone to erosion and moisture loss. An alternative is to apply herbicides and leave

residue standing on the soil surface. Herbicide application not only provides effective control of alfalfa but reduces the potential for soil and water losses and, through increased snow trapping by standing residue, may increase soil moisture reserves.

Although herbicide application has many advantages over intensive tillage, the impact of alternative termination management techniques on N dynamics within the plant-soil system is not clear. Many studies have quantified N release from legume crops, but few studies have directly compared the plant-availability of N under a range of termination management systems. Further information is required in order to develop effective termination techniques that maximize N use efficiency by subsequent crops and minimize N losses from the plant-soil system.

A series of field and laboratory studies was conducted to quantify N release following the termination of established alfalfa stands and to identify the mechanisms influencing N release. The specific objectives of these studies were:

- 1) to determine, under field conditions, the effects of method (herbicide application, tillage, tillage+herbicide application) and time (after first and second cut of alfalfa) of alfalfa termination on the plant-available N supply.
- 2) to determine the effect of method of alfalfa termination (chemical, mechanical) and residue placement (surface, incorporated) on volatile N losses and on the plant-available N supply under controlled conditions; and to determine the effects of residue particle size on N mineralization.
- 3) to determine, using  $^{15}\text{N}$  methodologies, the distribution of symbiotically fixed  $^{15}\text{N}_2$  in the plant/soil system following growth of an alfalfa crop; and the effect of

method of alfalfa termination on the fate and plant-availability of symbiotically fixed  $^{15}\text{N}_2$ .

## 2. LITERATURE REVIEW

### 2.1. Nitrogen contributions and non-nutritional benefits derived from legumes

Dinitrogen fixation by legume-*Rhizobium* symbiosis has traditionally been a primary source of N for agricultural production. However, reliance on these legume N sources has declined substantially in favour of manufactured N fertilizers (Power and Doran 1984). In recent years, interest in legume-based cropping systems has been renewed as concerns regarding agricultural and environmental sustainability have encouraged the development of cropping systems that reduce inputs of non-renewable resources but preserve or enhance soil and environmental quality. Legumes not only provide an inexpensive source of N in terms of non-renewable inputs but may promote improved soil and environmental quality.

Legumes contribute N to the plant-soil system both during growth and upon senescence. During alfalfa growth, N release may occur by several mechanisms including direct exudation of N-containing compounds (primarily ammonia, glutamate, serine, alanine and aspartate) from the root system, and decomposition of above-ground and below-ground residues (Ta et al. 1986; Dubach and Russelle 1994; Tomm et al. 1995).

Although estimates vary, N release from alfalfa by direct exudation appears to be minimal. Ta et al. (1986) reported exudation losses from the nodulated root system of hydroponically grown alfalfa equivalent to 3% of daily fixed N while Brophy and Heichel (1989) reported N release equivalent to 4.5% of symbiotically fixed plant N from alfalfa grown in sand culture. Even smaller amounts of N were directly excreted from soil-grown alfalfa; the soil surrounding alfalfa roots and nodules contained the equivalent of



$\leq 1\%$  of the symbiotically fixed N present in the alfalfa shoot at time of sampling, the equivalent of approximately  $1 \text{ kg symbiotically fixed N ha}^{-1}$  (Lory et al. 1992). Decomposition of above- and below-ground residues may account for considerably larger N losses than direct exudation (Dubach and Russelle 1994; Russelle et al. 1994). Root and nodule decomposition has been estimated to result in plant N losses ranging from less than  $15 \text{ kg of symbiotically fixed N ha}^{-1}$  ( $\sim 13 \text{ kg ha}^{-1}$  from decomposing roots and  $< 2 \text{ kg ha}^{-1}$  from decaying nodules in the top 60 cm of the soil) under a first year alfalfa stand (Dubach and Russelle 1994) to  $24 \text{ kg of fixed N ha}^{-1} \text{ yr}^{-1}$  under an established alfalfa stand (Andrén et al. 1990). Harvest and litterfall losses may also be significant, accounting for a total of  $28 \text{ kg N ha}^{-1} \text{ yr}^{-1}$  from alfalfa in an alfalfa/bromegrass mixture (Tomm et al. 1995).

Although significant amounts of N may be released from growing alfalfa, this N may be subsequently reabsorbed by the legume (Tomm et al. 1995) or absorbed by intercropped nonlegumes (Ta and Faris 1987; Burity et al. 1989). Only a portion of the N released during legume growth may accumulate in the soil. In an early study, Lyon and Bizzell (1934) estimated soil N accumulations of  $56 \text{ kg N ha}^{-1} \text{ yr}^{-1}$  under a pure alfalfa stand; a more recent study estimated accumulations of  $24 \text{ kg fixed N ha}^{-1} \text{ yr}^{-1}$  (Andrén et al. 1990).

Considerably larger quantities of legume-derived N may be returned to the soil upon alfalfa termination. The N contributions provided by alfalfa are well-documented (Lyon and Bizzell 1933; Bowren et al. 1969; Hoyt and Hennig 1971; Hoyt and Leitch 1983; Bruulsema and Christie 1987; Badaruddin and Meyer 1990; Westcott et al. 1995;

Hossain et al. 1996b) although estimates vary, likely because of differences in stand composition, management practices, environmental conditions and the manner in which N contributions are reported.

Alfalfa contributes substantial amounts of N to the plant-soil system via symbiotic  $N_2$  fixation. In field studies conducted in southern Manitoba,  $N_2$  fixation by alfalfa ranged from 98 to 247 kg N ha<sup>-1</sup> during the seeding year (Kelner and Vessey 1995). In Minnesota, perennial alfalfa stands fixed an estimated 160 to 177 kg N ha<sup>-1</sup> during the seeding year and up to 224 kg N ha<sup>-1</sup> during the fourth year of the alfalfa stand (Heichel et al. 1984). Similar levels of  $N_2$  fixation, ranging from 148 to 290 kg N ha<sup>-1</sup> yr<sup>-1</sup>, have been reported elsewhere (LaRue and Patterson 1981).

The amount of N actually returned to the soil in the form of legume residues will depend on various factors including management (harvested for forage versus plowdown), crop cultivar (dormant versus nondormant) and the timing and number of cuttings taken (Groya and Sheaffer 1985; Hesterman et al. 1986; Westcott et al. 1995). On Black Chernozemic soils near Melfort, SK, a second year alfalfa stand managed as a green manure crop (ie. terminated in late June or early July) returned an estimated 60 to 95 kg N ha<sup>-1</sup> to the soil in the form of alfalfa residue (shoots+roots excavated to 20 cm); considerably less N, approximately 20 kg N ha<sup>-1</sup>, was returned in residues of alfalfa harvested for hay or silage before termination (Bowren et al. 1969). Substantially larger amounts of N were returned in fall-incorporated alfalfa residues (shoots+roots excavated to 30 cm) in studies conducted in Minnesota (Hesterman et al. 1986). Alfalfa managed in a one-cut system (one summer harvest) returned 147 and 167 kg N ha<sup>-1</sup> in the herbage

regrowth, crown and root tissue of dormant and nondormant alfalfa, respectively; however, in a three-cut system (two summer and one fall harvest) only 71 and 100 kg N ha<sup>-1</sup> was returned in the crown and root tissue of dormant and nondormant alfalfa, respectively. Differences between cultivars were also evident in field studies conducted in Manitoba. 'Nitro' alfalfa, a nondormant cultivar selected specifically for its ability to accumulate N through a larger root mass and higher root N concentrations (Barnes et al. 1988), returned 121 kg symbiotically fixed N ha<sup>-1</sup> to the soil in the fall; dormant cultivars returned an average of only 40 kg fixed N ha<sup>-1</sup> (Kelner and Vessey 1995). Earlier studies have reported residue N contributions from alfalfa ranging from 39 to 135 kg N ha<sup>-1</sup> (Fribourg and Johnson 1955; Kroontje and Kehr 1956; Smith 1956; Stickler and Johnson 1959).

Of the N returned to the soil as legume residues, not all is immediately available for uptake by subsequent crops. In field studies conducted in western Canada, wheat accumulated 12% to 17% of the N present in residues of Tangier flatpea (*Lathyrus tingitanus*) or lentil (*Lens culinaris*) incorporated into the soil approximately 8 months prior to seeding of the wheat crop (Janzen et al. 1990). Only 1% to 2% of N applied as legume residue was recovered by a second wheat crop. Comparable recoveries of legume N have been reported elsewhere. In field studies conducted in Australia, 11% to 28% of N present in soil-applied strand medic (*Medicago littoralis*) was recovered by the initial wheat crop established after residue addition (Ladd et al. 1981; Ladd et al. 1983; Ladd and Amato 1986). Nitrogen uptake by a subsequent crop declined to 5% of legume N (Ladd et al. 1983). Consecutive barley crops established for 3 years after application of

alfalfa residues accumulated 18% of applied legume N (Ta and Faris 1990). In a previous study, only 6 to 25% of N present in decomposing plant material was recovered by barley (Müller and Sundman 1988).

The relatively low recoveries of legume-derived N observed in these  $^{15}\text{N}$  studies suggest that legumes are of greater importance for long-term than for short-term soil fertility (Janzen et al. 1990). In fact, in field studies, the majority of N applied in the form of legume residues (56 to 71%) was found to be retained in the soil as stabilized residues, inorganic N and N in roots of a subsequent wheat crop (Ladd et al. 1983). Low recoveries of legume-derived N by subsequent crops may also be attributable, in part, to the pool substitution effect (Hart et al. 1986). For example, labelled N derived from alfalfa may stand proxy for unlabelled N that would otherwise have been immobilized. Consequently, the amount of labelled N available for uptake by subsequent crops is reduced.

Although the proportion of legume-derived N recovered by subsequent crops is relatively small, legumes significantly influence soil N status and the N-supplying power of the soil (Campbell et al. 1991a,b; Campbell et al. 1993). Cropping systems which include legumes frequently contain larger amounts of total soil N, organic matter, light fraction organic matter and active soil N than non-legume rotations and also have a higher potential rate of N mineralization (Campbell et al. 1991a,b; Janzen et al. 1992; Bremer et al. 1994; Wani et al. 1994; Hossain et al. 1996a). In addition, applications of legume residues often increase soil inorganic N concentrations, N accumulations by subsequent crops, and yields of subsequent crops (Fribourg and Bartholomew 1956; Boawn et al.

1963; Hoyt and Hennig 1971; Hoyt and Leitch 1983; Bruulsema and Christie 1987; Hossain et al. 1996b).

In studies conducted in northern Alberta, the yield of spring wheat crops established in the first, second, fourth and fifth years after alfalfa termination were 71, 82, 75 and 68% greater, respectively, than that of wheat following fallow (Hoyt and Hennig 1971). The authors attributed these yield increases primarily to increased N availability: immediately after alfalfa termination, the mineralizable N content of surface soils was 42.4 mg N kg<sup>-1</sup> in alfalfa plots compared to 29.8 mg N kg<sup>-1</sup> in a fallow-wheat control. In a second study conducted in the Peace River region of Alberta, N contributions by alfalfa increased the yield of subsequent barley crops over that of an unfertilized fallow control at 3 of 5 sites (Hoyt and Leitch 1983). At those sites, the authors estimated that residual soil N provided by alfalfa ranged from approximately 50 to 150 kg N ha<sup>-1</sup>. Substantial increases in N accumulations by barley were also evident. These beneficial effects of alfalfa on succeeding crops may be long-lasting. During the initial 8 years of a 15 year period following alfalfa termination, yields of wheat following alfalfa were 66 to 114% higher than wheat established after a fallow-wheat rotation; wheat yields after alfalfa were also significantly higher than those in the fallow-wheat control during the tenth and thirteenth years after alfalfa termination (Hoyt 1990).

The positive responses attributed to legumes may be a function not only of N fertility, but also of non-nutritional benefits (Baldock et al. 1981; Wright 1990). Perennial forages in crop rotations may reduce the severity and/or incidence of plant disease (Campbell et al. 1990; Osunlaja 1990) and the population of a variety of annual grass and

broadleaf weeds (Harvey and McNevin 1990; Entz et al. 1995; Chung and Miller 1995). Improvements in soil physical condition, including aggregate stabilization and improved drainage, have also been attributed to legumes (Mazurak et al. 1955; Toogood and Lynch 1959; Spratt 1966; MacRae and Mehuys 1985; Blackwell et al. 1990). An additional benefit of legumes with respect to environmental quality is that deep-rooted legumes like alfalfa may recover N leached below the root zone of other crops and thus potentially reduce groundwater contamination (Schertz and Miller 1972; Mathers et al. 1975).

## 2.2. Current and future roles of alfalfa in crop management systems

Although the nutritional and non-nutritional benefits provided by legumes are well-known, the role of legumes in agricultural production has been reduced as legume N sources have been replaced by manufactured N fertilizer. However, concerns regarding agricultural sustainability, environmental degradation and rising input costs have prompted the development of cropping systems which reduce non-renewable inputs, pesticide use and soil erosion, and maintain, or improve, soil quality (Stinner and House 1989; Morrison and Kraft 1994). Perennial legumes such as alfalfa may be an important component of these developing cropping systems both as an inexpensive N source (in terms of non-renewable inputs) and an aid in pest control and the maintenance or enhancement of soil quality.

As of 1994, legumes occupied an estimated 9.6% of the seeded cropland (25 million hectares) in western Canada (Biederbeck et al. 1996). Forage legumes comprised approximately 52% of the area seeded to legumes but accounted for an estimated 65%

of the N fixed. Over 70% of the net N gain resulting from legume crops was attributed to forage legumes, the equivalent of 45 million kg N.

Alfalfa is one of the most common forage legumes, particularly in moist and irrigated areas of western Canada. Alfalfa is a highly productive, perennial legume with a deep, penetrating taproot system and is usually grown alone or with grass species for use as hay or pasture. Under current management systems, pure alfalfa and alfalfa/grass stands are often kept in rotation for extended periods ranging from 3 to 5 years in moist areas like south-central Manitoba to as long as 6 to 9 years in the drier areas of southern Saskatchewan (Entz et al. 1995). Based on a recent survey of producers in Manitoba and Saskatchewan, most stands are terminated due to reduced yield although other factors including pocket gophers (*Thomomys talpoides* and *Geomys bursarius*), weeds, and the establishment of higher value crops were sometimes involved (Entz et al. 1995). Only 11.6% of producers surveyed cited "rotational considerations" as the reason for termination of alfalfa which suggests that few forage crops are being managed to maximize rotational benefits (e.g. increases in the yield of subsequent crops, reductions in weed populations).

Although increases in total forage area are restricted by the demand for forage, the proportion of arable land deriving benefits from legumes could be increased without increasing total forage acreage by reducing stand duration. Currently, the average stand duration in many areas of Manitoba and Saskatchewan exceeds the economic optimum of 4 to 5 years reported by Jeffrey et al. (1993). In fact, in a recent survey, 57% of producers reported alfalfa stand durations of 6 years or more (Entz et al. 1995). These

long-term stands exceed the duration required to gain the N benefits associated with alfalfa (Heichel et al. 1984; Kelner 1994). Lyon and Bizzell (1933) demonstrated large increases in the yield of wheat following alfalfa whether the preceding alfalfa stand has been in production for one or three years. Similarly, Hoyt and Hennig (1971) found that alfalfa stands in production from 2 to 6 years had similar effects on a subsequent barley crop.

However, one of the challenges of incorporating short-term alfalfa stands into current cropping systems is that alfalfa is often difficult to terminate. Therefore, management practices which effectively terminate alfalfa while maximizing the benefits derived from alfalfa must be developed in order to improve the feasibility of short-term alfalfa stands.

At present, tillage is the most common method of terminating alfalfa. A recent survey of producers in Manitoba and Saskatchewan reported that 76.6% of producers terminated established alfalfa stands by tillage (Entz et al. 1995). Less than 25% used alternate management practices such as herbicide application (1.3%) or a combination of herbicide application and tillage (22.1%).

Despite its widespread use, tillage is not without disadvantages. Tillage is expensive, with costs ranging from approximately \$50 to 75 per ha (\$20 to 30 per acre), and may provide inconsistent control of alfalfa (Manitoba Agriculture 1993). Moreover, tillage reduces aggregation and buries surface residue, thereby leaving soils prone to erosion. In addition, tillage may promote soil moisture losses thereby further reducing soil moisture reserves already depleted during the growth of alfalfa. Tillage may also



encourage the germination of dormant weed seeds and promote the spread of perennial weeds such as quackgrass. An added difficulty often associated with tillage of alfalfa is that coarse residues, such as alfalfa crowns, retained on the soil surface obstruct equipment and result in an uneven soil surface and a loose, dry seedbed.

Although specific circumstances may warrant tillage (e.g., to level fields damaged by pocket gophers) some alternatives are now emerging. One option is to apply a herbicide to the alfalfa either during the growing season prior to establishment of a subsequent crop or in the spring immediately before establishment of a subsequent crop, and to leave residue standing on the soil surface. A variety of registered herbicides and tank-mixes (Roundup, Roundup+Banvel, Roundup+2,4-D) can provide reliable, cost-effective control of alfalfa and control of common weeds such as quackgrass and thistles (Manitoba Agriculture 1995). (Roundup was also registered recently as a pre-harvest treatment for application 4 to 5 days prior to alfalfa harvest.) By eliminating or reducing tillage, many of the disadvantages associated with tillage may be overcome. For example, by maintaining standing residue on the soil surface, soil moisture reserves may be increased through greater snow trapping (Bullied, Dept. of Plant Science, University of Manitoba, unpublished data). As well, a no-till management system often produces a moister, firmer seedbed which favours seed-soil contact and germination (Manitoba Agriculture 1993).

Given its many apparent benefits, herbicide application may be a practical alternative to intensive tillage for the termination of established alfalfa stands. However, since one of the primary benefits derived from legumes is N fertility, the impact of

alternative termination management practices on N dynamics within the plant-soil system must be considered. Many differences may exist between tilled and herbicide-treated untilled systems: placement of alfalfa residues, method by which alfalfa is terminated (ie. chemical versus mechanical), degree of soil disturbance, soil moisture levels, soil aeration, and soil temperature. These could influence N mineralization and N availability to subsequent crops.

### 2.3. Impacts of termination management on N benefits derived from legumes

In order to maximize the fertility benefits derived from legumes, N release from legume residues must be synchronized with N uptake by the subsequent crop. In field studies, soil-incorporation of hairy vetch (*Vicia villosa*) and red clover (*Trifolium pratense*) residues resulted in a pattern of N release which not only matched the N demands of a subsequent corn (*Zea mays*) crop, but also provided sufficient amounts of N to meet its N requirements (Stute and Posner 1995). In addition, post-harvest soil  $\text{NO}_3^-$  accumulations in the legume-based rotation were similar to, or less, than those resulting from fertilization. This suggests that the legume N source did not increase the potential for N losses by leaching or denitrification.

However, if N mineralization is not sufficient to meet the N needs of a subsequent crop, crop yield and quality may be adversely affected. In field experiments conducted in Idaho, alfalfa terminated by fall-applied herbicide did not release sufficient N to meet the requirements of subsequent winter or spring wheat crops, resulting in lower wheat yields and protein contents (Westermann and Crothers 1993). Similarly, N release from

hairy vetch terminated by spring-applied herbicide was poorly synchronized with the N uptake needs of a subsequent corn crop (Huntington et al. 1985). Again, the yield of the subsequent crop was reduced.

Poor synchrony may also result in the accumulation of excess inorganic N which may subsequently be lost from the plant-soil system via leaching or denitrification (Robbins and Carter 1980; Firestone 1982). In irrigated field experiments, 85 to 96 kg  $\text{NO}_3^-$ -N  $\text{ha}^{-1} \text{yr}^{-1}$  was leached from the root zone of an unfertilized bean (*Phaseolus vulgaris*) crop established after alfalfa (Robbins and Carter 1980). During the next growing season, an additional 17 to 29 kg  $\text{NO}_3^-$ -N  $\text{ha}^{-1} \text{yr}^{-1}$  was lost from the root zone of unfertilized wheat and bean crops. Recent studies suggest that potential may also exist for leaching losses of legume-derived N under dryland conditions in semi-arid areas (Campbell et al. 1984; Campbell et al. 1994). Campbell et al. (1994) noted that although deep-rooted legumes like alfalfa may remove water and  $\text{NO}_3^-$  from lower soil depths, legumes also increase the N supplying power of the soil. Particularly in cases where a legume crop is followed by fallow, concurrent N mineralization and water storage may increase the potential for  $\text{NO}_3^-$  leaching.

Few studies have compared the effects of method of legume termination on subsequent crops. In field studies with wheat, termination of legume green manure by herbicide application without incorporation resulted in a 22% lower grain yield than termination by soil-incorporation of legume green manure (Biederbeck and Slinkard 1988). The observed yield reduction was attributed to an insufficient nutrient supply resulting from delayed decomposition of the herbicide-treated residue. Contrasting results

were reported in field studies of corn following alfalfa (Triplett et al. 1979; Levin et al. 1987). In those studies, conventional and no-till systems did not differ in the fertilizer N response of corn established after alfalfa; however, Triplett et al. (1979) noted that grain yield may not have been sensitive enough to detect differences in N mineralization.

Termination technique could conceivably affect N release from legumes by several mechanisms. Perhaps foremost among these is an effect on residue placement. Soil incorporation of legume residues often results in greater N release than surface application and may increase N availability to subsequent crops (McCalla and Russel 1948; McKay et al. 1952; Wilson and Hargrove 1986; Varco et al. 1989), presumably due to more extensive contact between residues and microbial populations (Cogle et al. 1987). Possibly, more favourable moisture conditions in residue incorporated treatments also contribute to enhanced mineralization. Under controlled environment conditions, soil-incorporation of vetch residues resulted in mineralization of 51% of residue N after 35 d; only 36% of residue N was mineralized where vetch residues were surface applied (Aulakh et al. 1991a). Similar trends were evident under field conditions. Incorporation of sweetclover by mold-board plow resulted in the accumulation of 16 kg N ha<sup>-1</sup> more NO<sub>3</sub><sup>-</sup> than tillage with a subsurface plow which left residues on the soil surface (McKay et al. 1952). In an earlier study with sweetclover, subsurface tillage also reduced soil NO<sub>3</sub><sup>-</sup> accumulations, from approximately 110 kg N ha<sup>-1</sup> in plowed treatments to 103 kg N ha<sup>-1</sup> in subsurface-tilled treatments (McCalla and Russel 1948). Similarly, Varco et al. (1989) found that the proportion of N in <sup>15</sup>N-labelled hairy vetch recovered by a subsequent corn crop was 20% in no-till treatments compared to 32% in conventional-till

treatments.

These differences in N mineralization of surface and soil-incorporated residues tend to be most pronounced in early stages of decomposition. Crimson clover (*Trifolium incarnatum*) residue placed in mesh bags and buried in the plow layer of tilled soil for four wk retained 40% of initial residue N while residue retained on the soil surface retained 63% of its N after the same period. After 16 wk of incubation, differences between treatments had declined: 31% of initial residue N remained in incorporated residues compared to 36% in surface-applied residues (Wilson and Hargrove 1986). Similar results were obtained in field studies with <sup>15</sup>N-labelled hairy vetch; differences in N release between soil-incorporated and surface-applied residues were greatest within 30 d of residue application and declined thereafter (Varco et al. 1993).

In addition to effects of residue placement, N release may be influenced by the alfalfa shoot:root ratio at termination, the N content of alfalfa residues, and the rates of mineralization of individual plant parts. The shoot:root ratio, which can change substantially due to harvest management or growth stage, and residue N concentrations may influence the relative N contributions of shoot and root residues (Bowren et al. 1969; Heichel et al. 1984). Estimates of the amount of N supplied by roots vary, in part, because of difficulties in sampling and measuring the root mass. In studies conducted in Saskatchewan, roots accounted for an estimated 20 to 32% of the total residue N (in tops+roots to a soil depth of 20 cm) provided by an alfalfa crop terminated in midsummer as a green manure crop (Bowren et al. 1969); whereas roots would account for the majority of residue N if alfalfa were harvested and only the stubble incorporated. In

studies conducted in Manitoba, roots often accounted for >50% of total residue N (in stubble+roots to a soil depth of 15 cm) present in mid to late October in a two cut system (alfalfa hay cut in midsummer and early fall) (Kelner and Vessey 1995). Subsequent N release from these residues is influenced not only by the N content of root and shoot components but also their relative decomposability. In studies with medic (*Medicago littoralis* and *Medicago truncatula*), leaf tissue decomposed more rapidly and to a greater degree than stem and root tissue (Amato et al. 1984). After 4 wk, leaf tissue retained only 64% of its initial organic  $^{15}\text{N}$  whereas stem and root tissue retained 87% and 81%, respectively; after 2 years of decomposition, 40%, 56% and 50% of the  $^{15}\text{N}$  present in leaf, stem and root tissue remained. Differences in the decomposability of plant residues are primarily a function of residue composition which traditionally has been characterized by the carbon to nitrogen (C/N) ratio (Millar et al. 1936). Other factors, including the lignin and carbohydrate content, total N content and the soluble and intermediately-available C content of residues, have also been associated with differences in residue decomposability (Iritani and Arnold 1960; Nyhan 1975; Herman et al. 1977; Reinertsen et al. 1984; Janzen and Kucey 1988). Seasonal changes in the chemical composition of plant parts may also contribute to differential decomposability. For example, in alfalfa roots, the sugar, N and soluble protein concentrations increase substantially in the fall whereas in spring, the N and soluble protein concentrations decline and starch concentrations increase (Li et al. 1996).

The presence of herbicide residues in a desiccated legume green manure could also inhibit or delay the decomposition of legume residues (Biederbeck and Slinkard 1988).

However, in field studies with lentil, canola, rye, barley, wheat and flax residues, up to three applications of herbicide (glyphosate or paraquat or 2,4-D) did not alter the rate of residue degradation (Blackshaw and Lindwall 1995).

Although increased N release from incorporated residues has generally been attributed to enhanced mineralization, recent studies suggest that reduced volatile N losses from incorporated residues may also contribute to the larger available N supply observed. During the decomposition of plant residues, substantial N losses may occur presumably from volatilization of  $\text{NH}_4^+$  produced during the ammonification of soluble organic N and subsequent mineralization of legume N (Janzen and McGinn 1991). Perennial ryegrass (*Lolium perenne*) incubated for 70 d lost the equivalent of 20 to 47% of herbage N via volatilization (Whitehead et al. 1988). Lentil green manure, lentil straw and wheat straw incubated without soil for 28 d lost the equivalent of 42, 3.4 and 0.5% of residue N (Bremer and van Kessel 1992). Janzen and McGinn (1991) also reported volatile N losses equivalent to 14% of residue N from lentil green manure placed on the soil surface or suspended above the soil surface. However, incorporating the lentil residue into the soil essentially eliminated volatile N losses. Volatile N losses from other organic amendments have also been substantially reduced following soil incorporation (Adamsen and Sabey 1987).

Another factor which may contribute to differential N mineralization from tilled versus herbicide-treated alfalfa is residue particle size. Intensive tillage physically breaks down alfalfa residues whereas termination by herbicide application without tillage leaves alfalfa residues physically intact and presumably with a smaller surface area available for

microbial decomposition. Several laboratory studies suggest that, within relatively small size fractions (<50 mm diameter), reductions in residue particle size may enhance residue decomposition. For example, reductions in the particle size of corn residues from 19 mm to <0.25 mm significantly increased CO<sub>2</sub> evolution, particularly within the first few days of the decomposition period (Sims and Frederick 1970). Similarly, in studies with legume residues, fine grinding (<1 mm) increased net <sup>15</sup>N mineralization in stem, root and pod material of strand medic although it did not affect mineralization of leaf material (Amato et al. 1984). However, particle size (which ranged from <1 mm to 50 mm lengths of stem and 2.5 cm<sup>2</sup> root mats) had little effect on the residual <sup>15</sup>N content of *Medicago* sp. decomposing under field conditions (Amato et al. 1984). In contrast, Stickler and Frederick (1959) observed greater net NO<sub>3</sub><sup>-</sup> accumulations from coarse (<1.25 cm) than from finely ground alfalfa top-growth, but little effect of particle size on net NO<sub>3</sub><sup>-</sup> accumulations from alfalfa root.

Another factor which may contribute to differences in the supply of available N under different termination treatments is the magnitude of denitrification losses which occur following termination. Although field studies with white clover (*Trifolium repens*) have demonstrated significant effects of termination method on denitrification losses (shallow tillage > herbicide > plough = rotary hoe), losses estimated 15 d after termination were relatively small, ranging from 2 kg N ha<sup>-1</sup> in herbicide treatments to 6 kg N ha<sup>-1</sup> in shallow tillage treatments (King and Ball 1992). Under controlled conditions, surface application of residues resulted in lower initial denitrification losses than incorporation; however, cumulative N losses were the same after 35 d (Aulakh et al.



1991a). Denitrification following soil-incorporation of residues has been attributed to the increased supply of labile organic C provided by residues (Aulakh et al. 1991b; McKenney et al. 1993) whereas denitrification in soil under a herbicide-terminated grass sward was attributed to increased soil moisture and  $\text{NO}_3^-$  contents (Tenuta and Beauchamp 1996).

Differences in soil temperature and moisture in herbicide-terminated and tillage-terminated systems may also contribute to differences in N release. Surface residue cover often results in lower soil temperatures and higher soil moisture contents than bare soil (Mitchell and Teel 1977; Gauer et al. 1982; Wall and Stobbe 1984; Sarrantonio and Scott 1988). The influence of soil moisture, soil temperature and interaction between these and other factors (soil organic C content, N availability) on N mineralization has been the subject of numerous studies (e.g., Stanford et al. 1973; Stanford and Epstein 1974; Cassman and Munns 1980; Addiscott 1983; MacDonald et al. 1995). In general, the rate of N mineralization increases with increasing temperature over the range of temperatures normally encountered under field conditions (Stanford et al. 1973; MacDonald et al. 1995). Increases in soil moisture content also increase N mineralization rate up to a maximum water content above which mineral N accumulations decline presumably due to denitrification (Stanford and Epstein 1974).

The many factors which may influence N release under different termination management systems make the prediction of N mineralization under these systems very difficult. Although the N contributions made by alfalfa to the plant-soil system have been well-documented, few studies have directly compared the available N supply under

various termination management systems. Further information is required regarding the impact of termination technique on N dynamics in order to develop management strategies which effectively terminate alfalfa while maximizing N use efficiency by subsequent crops and minimizing N losses. Strategies which allow more effective management of legumes may improve the feasibility of using legumes in today's cropping systems as an alternative N source.

### 3. Plant-Available N Supply under Field Conditions as Affected by Method of Alfalfa Termination

#### ABSTRACT

Herbicide application may provide an alternative to intensive tillage for the termination of alfalfa stands but could alter the pattern of N release and N availability to subsequent crops. Four field experiments were initiated on alfalfa (*Medicago sativa*) stands in southern Manitoba in 1992 and 1993. A factorial combination of three methods (herbicide, tillage, herbicide+tillage) and two times (after first alfalfa cut, after second alfalfa cut) of termination was arranged in a randomized complete block design. One additional treatment of a spring-applied herbicide was also included. Spring wheat (*Triticum aestivum* cv. Katepwa) was established following alfalfa termination, and soil  $\text{NO}_3^-$  status, plant N accumulations and wheat yield were monitored for one to two years. In three of four experiments, termination by tillage or herbicide+tillage produced a larger short-term plant-available N supply than termination by herbicide application alone. Regardless of method, early-summer termination generally produced a greater short-term plant-available N supply than late-summer termination. However, despite a smaller short-term plant-available N supply in herbicide treatments, wheat yields in herbicide treatments were similar to or greater than those in tillage treatments. Differences in the N status of termination treatments diminished with time; by the fall of the second growing season after termination, differences in the cumulative available N supply were no longer evident. Under conditions like those in southern Manitoba, herbicide application may improve the synchrony between N release from alfalfa residues and N needs of a subsequent spring

wheat crop and could improve the N use efficiency of subsequent crops.

## INTRODUCTION

Concerns regarding agricultural sustainability, soil and environmental quality and energy conservation have renewed interest in the use of legumes in cropping systems. Legumes not only provide substantial amounts of plant-available N to subsequent crops (Hoyt and Leitch 1983; Bruulsema and Christie 1987) but may also reduce weed populations (Harvey and McNevin 1990; Entz et al. 1995), reduce the incidence and/or severity of plant disease (Campbell et al. 1990; Osunlaja 1990) and improve soil physical condition (Mazurak et al. 1955; Toogood and Lynch 1959; Campbell et al. 1990).

One strategy for increasing the proportion of arable land deriving benefits from legumes without increasing the total acreage of legume crops is to reduce the duration of alfalfa stands. Currently, in Manitoba and Saskatchewan, the average duration of pure alfalfa (*Medicago sativa*) or alfalfa/grass stands is 6.5 years (Entz et al. 1995). This exceeds the economic optimum of 4 to 5 years reported by Jeffrey et al. (1993) and the duration required to attain maximum N benefits (Hoyt and Hennig 1971; Heichel et al. 1984). However, successful adoption of shorter-term alfalfa stands requires the development of management practices which effectively terminate alfalfa but maximize the benefits provided to subsequent crops.

Intensive tillage, the usual method of terminating alfalfa, leaves the soil prone to erosion and moisture loss. An alternative is to apply herbicides either the summer before, or the spring immediately before, establishment of a subsequent crop and leave residue

standing on the soil surface. Herbicide application provides reliable, cost-effective control of alfalfa and concurrent control of a broad spectrum of weeds (Bullied, Dept. of Plant Science, University of Manitoba, unpublished data). In addition, standing surface residues protect the soil from erosion and moisture loss and may increase snow trapping, thereby increasing soil moisture reserves (Bullied, Dept. of Plant Science, University of Manitoba, unpublished data).

Although use of herbicide in place of tillage has many apparent benefits, it could affect the amount and timing of N release from alfalfa residue and the availability of this N to subsequent crops. If the rate of N mineralization is too slow to meet the N uptake needs of a subsequent crop, crop yield and/or quality may be adversely affected (Huntington et al. 1985; Westermann and Crothers 1993). Conversely, accumulations of excess amounts of inorganic N may be prone to leaching or denitrification (Robbins and Carter 1980; Firestone 1982). Even under dryland conditions in semi-arid areas, potential may exist for leaching losses of legume-derived N (Campbell et al. 1984; Campbell et al. 1994).

Few studies have directly compared N release from alfalfa terminated by chemical versus mechanical means. Under controlled conditions, incorporation of alfalfa residues resulted in more rapid N release from alfalfa residues and a larger short-term plant-available N supply than termination by herbicide application in which alfalfa residues were retained on the soil surface (Chapter 5). In field experiments, termination of a green manure crop by herbicide application resulted in a 22% lower grain yield in a subsequent wheat (*Triticum aestivum*) crop than termination by tillage (Biederbeck and Slinkard

1988). The observed yield reduction was attributed to delayed decomposition of herbicide-treated residues which may have reduced the available nutrient supply. In contrast, in field studies with corn (*Zea mays*), the fertilizer N response of corn established after alfalfa was the same under conventional till and no-till management (Triplett et al. 1979; Levin et al. 1987).

The objective of this study was to determine the effect of timing and method of termination of alfalfa on the pattern of N release under field conditions. This was accomplished by measuring soil  $\text{NO}_3^-$  accumulations, soil mineralizable N content and N accumulations by subsequent spring wheat crops for up to two years after alfalfa termination.

## MATERIALS AND METHODS

Four field experiments were conducted in southern Manitoba from 1992 to 1994 (Table 3.1). Field experiments were initiated on established alfalfa stands at the Glenlea Research Station (Glenlea-92) and at the Carman Research Station at 23-6-5W (Carman-92) in 1992 and were monitored during the 1993 and 1994 growing seasons. Two additional experiments were initiated on established alfalfa stands at "the Point" on the University of Manitoba campus at Winnipeg (Winnipeg-93) and at the Carman Research Station at 23-6-5W (Carman-93) in 1993 and were monitored from time of establishment through the 1994 growing season. At all sites, a hay-type cultivar was used. Alfalfa stands at the Carman-92, Winnipeg-93, Carman-93 and Glenlea-92 sites were 4, 4, 5 and 6 years old, respectively. (Climatic data for Glenlea-92 and Carman are reported in

Tables A.1 and A.2 of Appendix A).

Termination treatments consisted of a factorial combination of three termination methods (herbicide application, tillage, herbicide application followed by tillage) applied at two times during the growing season (early summer-after the first alfalfa harvest, late summer-after the second alfalfa harvest). One additional treatment was included in which herbicide was applied in the spring immediately prior to establishment of a subsequent spring wheat crop. All termination treatments were included in the experiments initiated in 1992. In experiments initiated in 1993, selected treatments were omitted. At the Carman-93 site, only five termination treatments were applied: early, late and spring herbicide application; and early and late tillage. At Winnipeg-93, only two termination treatments were applied: late termination by herbicide or by tillage. In all experiments, treatments were arranged in a randomized complete block design with four replicates.

#### Field Techniques

At all sites, the first cut of alfalfa was harvested in mid-June to early July and the second cut in late July to mid-August (Table A.3 in Appendix A). For both harvests, alfalfa was cut, baled and removed from the plot area.

In plots designated for early termination, treatments were applied approximately 3-5 wk after the first cut of alfalfa. Alfalfa in plots designated for late termination was allowed to regrow after the first cut and treatments were applied approximately 3-5 wk after the second cut of alfalfa. (Based on random square meter samples of alfalfa top-growth harvested inside the plot area, the amount of alfalfa top-growth present within

several days of termination averaged about 2000 kg dry matter ha<sup>-1</sup> in experiments established in 1993. Alfalfa dry matter yields were not determined prior to termination in experiments established in 1992.)

Herbicide+tillage treatments consisted of an initial application of 2.5 L ha<sup>-1</sup> Roundup followed by several tillage operations. Herbicide treatments consisted of 1-2 applications of 2.5 or 5 L ha<sup>-1</sup> Roundup (in 1992) or a tank-mix of 1.85 L ha<sup>-1</sup> Roundup, 1.25 L ha<sup>-1</sup> Banvel and 1 L ha<sup>-1</sup> 2,4-D (in 1993). Tillage treatments consisted of repeated tillage by a combination of disc, cultivator and harrow from time of termination throughout the remainder of the growing season except at Winnipeg-93 where tillage treatments were rototilled three times during the year of termination. (Table A.3 in Appendix A gives a detailed description of termination management.)

Plot dimensions were 5.5 m x 16 m at Winnipeg-93 and 12 m x 12 m at all other sites. Neutron access tubes were installed in the centre of each plot.

At all sites, spring wheat (cv. Katepwa) was established during the first growing season following alfalfa termination. In herbicide treatments, wheat was seeded directly into untilled soil using a zero till offset disc drill. All tillage and herbicide+tillage treatments were tilled in the spring prior to seeding except at Winnipeg-93 where wheat was directly seeded without spring tillage. After harvest, wheat straw was baled and removed from all treatments, but no tillage was done. At sites initiated in 1992, a second spring wheat crop (cv. Katepwa) was established in 1994. Wheat was seeded directly into untilled soil in all treatments.

Triple super phosphate (0-46-0) was applied in the seedrow at time of seeding at



a rate of about 13 kg P ha<sup>-1</sup>. Where required based on soil test recommendations, S and K were applied in the form of spring-broadcast potassium sulfate. No N-containing fertilizers were applied.

#### Sampling Procedures and Analytical Techniques

Soil samples to a depth of 60 or 120 cm (in increments of 0 to 15, 15 to 30, 30 to 60, 60 to 90, 90 to 120 cm) were collected periodically throughout the study. In experiments initiated in 1992, soil samples (to 120 cm) were collected in May 1993, September to early October 1993, May 1994 and August 1994; soil samples (to 60 cm) were collected in June to July of 1993. In experiments initiated in 1993, soil samples (to 120 cm) were collected in September to early October 1993, May 1994 and August 1994; soil samples (to 60 cm) were collected in June of 1994. At all sites, spring samples were taken shortly before or immediately after wheat was seeded; fall samples were taken after the wheat was harvested. Midsummer soil sampling coincided with the third plant tissue sampling, approximately 6 wk after wheat emergence. Soil samples consisted of a composite of three to six soil cores per plot.

Soil samples were air dried immediately after collection except in a few cases in which soil samples were frozen briefly before air drying. Air dry soil samples were ground (<2 mm) using a rotating sieve. Plant residues which did not pass through the sieve were discarded. During selected sampling periods, soil samples were weighed prior to grinding to determine bulk density. [In some cases, this procedure resulted in uncharacteristically low bulk densities for the Glenlea-92 and Winnipeg-93 sites (e.g. ~0.9

Mg m<sup>-3</sup> at Glenlea-92 in 1993; ~1.1 Mg m<sup>-3</sup> for the 0-15 cm soil depth at Winnipeg-93 in 1994) which may have resulted in underestimation of the soil NO<sub>3</sub><sup>-</sup>-N content.]

Soil inorganic N was extracted with 2 M KCl and the concentration of NO<sub>3</sub><sup>-</sup> and NH<sub>4</sub><sup>+</sup> in the extract was determined by a colorimetric procedure (Keeney and Nelson 1982). (Soil NO<sub>3</sub><sup>-</sup>-N concentrations are reported in Tables A.4 to A.7 in Appendix A.)

Mineralizable C and N was determined for surface soil samples (0 to 15 cm) collected during the first spring after alfalfa termination using a procedure similar to that described by Bremer et al. (1994). A 50 g soil sample was moistened to 80% of field capacity and placed in a sealed 1 L glass jar containing a CO<sub>2</sub> trap (10 mL of 2 M NaOH). Samples were incubated at 25°C for 6 wk. Jars were aerated and the NaOH traps replaced at 1, 2, 4 and 6 wk. To quantify the carbonate content in each sample, the entire NaOH sample was placed in a 1 L airtight jar, acidified with an excess of HCl, and the CO<sub>2</sub> evolved was measured on a gas chromatograph. The CO<sub>2</sub> concentration in the headspace of each jar was calculated using a standard curve run with each set of samples. At the end of the incubation period, soil samples were air dried and analyzed for inorganic N as described previously (Keeney and Nelson 1982) to estimate mineralizable N.

Soil physical and chemical characteristics for each field site (Table 3.1) were determined on a composite sample of soil samples collected in the spring of the first year following alfalfa termination.

Plant tissue samples were taken at about 2 wk intervals throughout the growing season of the first wheat crop established after alfalfa termination. (The plant tissue

sampling schedule is reported in Table A.8 in Appendix A.) The second wheat crop established after alfalfa termination was sampled at the soft dough stage and at crop maturity. At all sites, plant tissue samples consisted of wheat top-growth from a 1 m length of 6 adjacent rows (approx. 0.9 m<sup>2</sup>); wheat was cut approximately 2 cm above the soil surface and immediately air dried. (Dry matter yields are reported in Tables A.9 to A.12 in Appendix A.)

To determine grain yields at crop maturity, wheat was harvested from a 10 to 19 m<sup>2</sup> area within each plot using a small plot combine. To determine straw yields, wheat was hand-harvested from 1 m lengths of 6 adjacent rows as described previously. In 1993, straw yields were calculated as the difference between the total dry matter yield of hand-harvested samples and the grain yield of combine-harvested samples. In 1994, hand-harvested samples were threshed and the straw yield measured directly. For several plots at Glenlea-92 which could not be harvested by combine due to uneven crop growth, 1994 grain yields were based on the grain yield of hand-harvested samples.

Air-dried plant samples were oven dried, weighed to determine plant dry matter yield and ground to pass a 2 mm sieve using a Wiley mill. A subsample of coarsely ground plant material was taken, finely ground using a Cyclone sample mill (Udy Corporation, Fort Collins, CO) and analyzed for total N by an automated combustion technique (Carlo Erba<sup>TM</sup>, Milan, Italy). (Plant tissue N concentrations are reported in Tables A.13 to A.16 in Appendix A.)

As part of a concurrent study, soil moisture to a depth of up to 180 cm was monitored periodically throughout the duration of the experiment using a Troxler Series

4300 depth moisture gauge (Troxler Electronic Laboratories Inc., North Carolina) (Bullied, Dept. of Plant Science, University of Manitoba, unpublished data).

Data were analyzed by analysis of variance for a factorial experiment arranged in a randomized complete block design using the Proc GLM procedure (SAS Institute Inc. 1985). To determine the effect of spring-applied herbicide treatments, data were re-analyzed by two-way analysis of variance for a randomized complete block design and by single degree of freedom contrasts using the Proc GLM procedure.

## RESULTS AND DISCUSSION

### Soil $\text{NO}_3^-$ Accumulations

Large amounts of  $\text{NO}_3^-$  accumulated in the soil after the termination of alfalfa (Figs. 3.1 and 3.2). At all sites except Winnipeg-93, both timing and method of alfalfa termination significantly influenced soil  $\text{NO}_3^-$  content.

Tillage, alone or in combination with herbicide application, increased soil  $\text{NO}_3^-$  accumulations in the first spring after alfalfa termination. At Carman-92, soil  $\text{NO}_3^-$  content averaged  $128 \text{ kg ha}^{-1}$  in tilled treatments compared to  $100 \text{ kg ha}^{-1}$  in herbicide treatments (Fig. 3.1). Similarly, at Glenlea-92, termination involving tillage produced greater amounts of spring soil  $\text{NO}_3^-$  ( $84 \text{ kg ha}^{-1}$ ) than termination by herbicide application alone ( $61 \text{ kg ha}^{-1}$ ). The largest differences among termination methods were evident at Carman-93. Spring soil  $\text{NO}_3^-$  accumulations were  $62$  and  $84 \text{ kg ha}^{-1}$  in late and early herbicide treatments, respectively, compared to  $125$  and  $196 \text{ kg ha}^{-1}$  in late and early tillage treatments. These differences among termination methods were already evident

at Carman-93 the previous fall (Fig. 3.2). Similar results have been reported for green manure crops: accumulations of soil inorganic N were lower under winter green manure crops terminated by herbicide application in a no-till system than by tillage in a conventional-till system (Sarrantonio and Scott 1988).

Tillage of a previously untilled system like a perennial alfalfa stand may result in incorporation of surface residues, redistribution of organic matter within the soil, increases in soil aeration, and soil drying which, in turn, may influence microbial activity and thus processes such as N mineralization. Greater accumulations of inorganic N under conventional till management may be due to factors which improve conditions for N mineralization: higher soil temperatures (Mitchell and Teel 1977) and more suitable soil moisture levels (Sarrantonio and Scott 1988). In part, our observation may simply reflect greater N release from incorporated than from surface residues (McKay et al. 1952; Wilson and Hargrove 1986; Varco et al. 1993), presumably the result of more extensive exposure of legume residues to soil microbial populations (Cogle et al. 1987).

Regardless of termination method, delaying termination until after the second cut of alfalfa substantially reduced soil  $\text{NO}_3^-$  accumulations measured in the spring immediately following alfalfa termination. The same trend was evident for early and late herbicide treatments at Carman-93, although differences were not statistically significant. Presumably, lower soil  $\text{NO}_3^-$  accumulations in the delayed treatments result from the shortened period of decomposition. Differences between early and late termination by the same method ranged from 13 kg  $\text{NO}_3^-$ -N ha<sup>-1</sup> (herbicide treatments at Glenlea-92) to 71 kg  $\text{NO}_3^-$ -N ha<sup>-1</sup> (herbicide treatments at Carman-92, tillage treatments at Carman-93).

The effect on spring soil  $\text{NO}_3^-$  accumulations of delaying herbicide application until the spring immediately prior to establishment of a subsequent wheat crop was measured only at Carman-93. At that site, delaying herbicide application until spring reduced the soil  $\text{NO}_3^-$  content compared with termination early in the previous season, but produced soil  $\text{NO}_3^-$  levels similar to termination late in the previous season (Fig. 3.2). These results suggest either that minimal N mineralization occurred in the treatment applied late the previous growing season or that a portion of the N mineralized in the fall treatment was lost from the system.

The differences in soil  $\text{NO}_3^-$  accumulations observed among termination treatments were still evident at midsummer, but N uptake by the growing wheat crop had substantially depleted soil  $\text{NO}_3^-$  reserves at all sites (Figs. 3.1 and 3.2).

Observed differences between termination treatments in their soil  $\text{NO}_3^-$  accumulations diminished with time. In both experiments conducted at Carman, effects of termination treatment on soil  $\text{NO}_3^-$  accumulations were no longer evident in the fall after harvest of the initial wheat crop. In contrast, at Glenlea-92, effects of both time and method of alfalfa termination were evident until the second spring after alfalfa termination. In the fall after harvest of the initial wheat crop at Glenlea-92, soil  $\text{NO}_3^-$  levels remained significantly lower in late than early termination treatments regardless of termination method; this effect was still evident in the spring of 1994, the second spring after alfalfa termination. Effects of termination method were also evident at Glenlea-92 in the spring of 1994; soil  $\text{NO}_3^-$  accumulations were greater in tillage than herbicide only treatments even though no differences among termination methods had been measured the

previous fall. Following harvest of the second wheat crop at Glenlea-92, effects of termination treatment on soil  $\text{NO}_3^-$  accumulations were no longer evident (Fig. 3.1).

At Winnipeg-93, soil  $\text{NO}_3^-$  accumulations were comparable to those observed at other sites and followed similar trends over time but showed no effect of termination method, perhaps because later termination (second week in September) and cool conditions delayed mineralization in both treatments (Table A.1 in Appendix A).

Method and time of termination influenced not only the amount of  $\text{NO}_3^-$  accumulated in the soil, but also the distribution of  $\text{NO}_3^-$  within the soil profile. In the first spring after alfalfa termination, tillage increased the  $\text{NO}_3^-$  concentration in surface soils (0 to 30 cm) but had variable effects on subsurface soil  $\text{NO}_3^-$  concentrations (Figs. 3.3 and 3.4). At Carman-92, no effects of termination method on soil  $\text{NO}_3^-$  concentrations were evident below a soil depth of 30 cm; however, in the coarser-textured soil at the Carman-93 site, tillage increased  $\text{NO}_3^-$  concentrations to a soil depth of 120 cm suggesting that downward movement of  $\text{NO}_3^-$  may have occurred. In contrast, in subsurface soils at Glenlea-92 (30 to 90 cm), early-season herbicide application resulted in higher soil  $\text{NO}_3^-$  concentrations than tillage. One possible explanation is that larger fall soil moisture reserves (Bullied, Dept. of Plant Science, University of Manitoba, unpublished data), combined with intact root channels in the untilled herbicide system, allowed downward movement of water through the fine-textured soil and concurrent leaching of  $\text{NO}_3^-$  (Blackwell et al., 1990). Regardless of method, delaying termination until after the second cut of alfalfa often reduced soil  $\text{NO}_3^-$  concentrations; however, effects were not consistent for all soil depths at all sites. Further delaying herbicide application until the

following spring did not influence soil  $\text{NO}_3^-$  concentrations in the top 30 cm of the soil profile, but reduced subsurface soil  $\text{NO}_3^-$  concentrations (30 to 90 cm) compared to herbicide application during the previous growing season (Fig. 3.4).

Observed differences in the vertical distribution of soil  $\text{NO}_3^-$  under different termination management systems suggest that both method and time of alfalfa termination may affect the potential for  $\text{NO}_3^-$  leaching. In order for substantial leaching losses to occur, two conditions must exist: the presence of large amounts of  $\text{NO}_3^-$ , and substantial water movement (Legg and Meisinger 1982). Termination techniques like herbicide application which reduce the quantity of  $\text{NO}_3^-$  in the soil profile (Figs. 3.1 and 3.2) also reduce the amount of  $\text{NO}_3^-$  which could potentially be lost through leaching. However, herbicide termination may also increase soil moisture reserves compared to tillage. Although these increases in soil moisture reserves could potentially contribute to downward movement of  $\text{NO}_3^-$  in some cases, the potential for  $\text{NO}_3^-$  leaching will depend on various other factors including soil texture, precipitation, evapotranspiration, N transformations within the soil and drainage (Follett 1989).

#### Mineralizable N

The quantity of mineralizable N (determined in a laboratory incubation procedure) in the surface soils at Glenlea-92, Carman-92 and Winnipeg-93 was not influenced by time or method of alfalfa termination (Table 3.2). However, at Carman-93, early termination decreased the mineralizable N content in tillage treatments but had no effect on the mineralizable N content in herbicide treatments.



Although the laboratory-determined mineralizable N revealed minimal differences among termination treatments, net N mineralization in the field during the growing season (measured as the change in the cumulative available N supply between spring and fall) varied considerably among treatments (Table 3.3). At Glenlea-92, 1993 growing season net mineralized N was higher in herbicide ( $57 \text{ kg ha}^{-1}$ ) than in herbicide+tillage treatments ( $28 \text{ kg ha}^{-1}$ ) and was intermediate in tillage treatments ( $44 \text{ kg ha}^{-1}$ ). However, at Carman-92, 1993 growing season net mineralized N did not differ among termination methods but was substantially higher in late-terminated ( $113 \text{ kg ha}^{-1}$ ) than early-terminated ( $65 \text{ kg ha}^{-1}$ ) treatments regardless of termination method. No treatment differences were observed at Glenlea-92 or Carman-92 during the subsequent growing season. At Carman-93, 1994 growing season mineralized N was substantially higher in herbicide ( $111 \text{ kg ha}^{-1}$ ) than tilled treatments ( $19 \text{ kg ha}^{-1}$ ), although termination time did not influence the amount of N mineralized during the growing season. Therefore, treatments in which N mineralization was initially delayed (based on low spring soil  $\text{NO}_3^-$  accumulations), such as the herbicide and late-terminated treatments, often had higher rates of N mineralization during the first growing season following alfalfa termination. It should be noted, however, that although termination method clearly influences N mineralization, the quantity of N mineralized and hence the ability of each termination management system to meet the N needs of a subsequent crop will be strongly influenced by the prevailing environmental conditions.

Differences between field and laboratory determinations suggest that most treatment effects are related to the rate of residue decomposition, not to differences in the

mineralization rate of soil N; laboratory incubations (which measure only mineralizable N in the soil) demonstrated minimal differences among treatments whereas large differences among treatments were evident in field soils containing alfalfa residues. This is in agreement with results of a subsequent  $^{15}\text{N}$  study which showed no difference in the soil mineralizable N content of herbicide and tillage treatments following alfalfa termination (Chapter 6).

#### Available N supply

Substantial amounts of plant available N (calculated as the sum of soil  $\text{NO}_3^-$  accumulations to 60 cm and kg N  $\text{ha}^{-1}$  removed in the grain and straw of subsequent wheat crops) were released following alfalfa termination (Fig. 3.5 and 3.6). [This calculation may have slightly overestimated the available N supply since the soil  $\text{NO}_3^-$  content at time of termination was not determined at all sites and therefore was not subtracted from the available N supply. Presumably, soil  $\text{NO}_3^-$  levels under these long-term alfalfa stands which had not received N fertilizers would be fairly low at all sites. Sites established in 1993 generally contained  $<20$  kg  $\text{NO}_3^-$ -N  $\text{ha}^{-1}$  at time of termination.] By the fall of the first growing season after alfalfa termination, plant-available N release averaged 107 kg N  $\text{ha}^{-1}$  at Winnipeg-93, 119 kg N  $\text{ha}^{-1}$  at Glenlea-92, 186 kg N  $\text{ha}^{-1}$  at Carman-93 and 211 kg N  $\text{ha}^{-1}$  at Carman-92 (Figs. 3.5 and 3.6). At the end of the second growing season after alfalfa termination, the cumulative plant-available N was 244 kg N  $\text{ha}^{-1}$  at Glenlea-92 and 256 kg N  $\text{ha}^{-1}$  at Carman-92. These values are comparable to those reported in the literature. Alfalfa residues have been shown to contribute from 20 to 167

kg N ha<sup>-1</sup> to the soil depending on various factors including alfalfa management (harvested for forage versus plowdown; number and timing of cuttings) and cultivar (Bowren et al. 1969; Groya and Sheaffer 1985; Hesterman et al. 1986; Westcott et al. 1995). These values may, in fact, underestimate N contributions because N in the root system is often not completely accounted for. The fertilizer-N replacement value of alfalfa to a subsequent corn crop ranged from 135 to 180 kg N ha<sup>-1</sup> in studies conducted in the United States (Baldock and Musgrave 1980; Hesterman et al. 1987). Even larger alfalfa N contributions have been reported in long-term studies. Boawn et al. (1963) found that five consecutive corn crops established in an area previously seeded to alfalfa accumulated a total of 383 kg ha<sup>-1</sup> more N than corn established in a previously uncropped area.

At Glenlea-92, the available N supply in fall 1993 was higher in tillage only (131 kg N ha<sup>-1</sup>) than in herbicide+tillage treatments (107 kg N ha<sup>-1</sup>) and intermediate in herbicide only treatments (118 kg ha<sup>-1</sup>) (Fig. 3.5). Regardless of termination method, delaying termination until after the 2nd cut of alfalfa reduced the available N supply by an average of 24 kg N ha<sup>-1</sup>. Significant effects of termination method and time were still evident in spring 1994 but, by fall 1994 the available N supply at Glenlea-92 was the same in all treatments regardless of method or time of termination.

In contrast, at the Carman-92 site, effects of method and time of termination diminished rapidly; by the fall of 1993, differences among termination treatments were no longer evident. Substantial differences in the pattern of N release were also evident between Carman-92 and Glenlea-92. Whereas the available N supply at Glenlea-92

increased slowly and steadily between spring 1993 and fall 1994, the available N supply at Carman-92 increased rapidly until fall 1993 then increased at a markedly slower rate. Presumably, environmental factors at Carman-92 and Glenlea-92 contributed to these differences. In 1993, Carman-92 experienced substantially higher July and August temperatures than Glenlea-92 which may have contributed to the rapid increase in available N supply observed at Carman-92 during the last half of the 1993 growing season (Table A.1 in Appendix A). In addition, Carman-92 received considerable rainfall which appeared to result in soil moisture conditions conducive to N mineralization (Table A.2 in Appendix A). In contrast, at Glenlea-92, extremely wet conditions (the result of high rainfall combined with a poorly-drained heavy clay soil) may have enhanced denitrification and thus reduced the available N supply.

At Carman-93, termination treatment had a significant effect on the short-term available N supply, but these effects diminished over time (Fig. 3.6). By fall 1994, differences among termination methods were no longer evident, but effects of termination time remained. Delaying tillage until after the second cut of alfalfa reduced the available N supply by  $59 \text{ kg N ha}^{-1}$ . In herbicide treatments, delaying termination application until after the second cut of alfalfa did not reduce the available N supply; however, further delaying herbicide application until immediately before establishment of a subsequent spring wheat crop reduced the available N supply by 65 and  $56 \text{ kg N ha}^{-1}$  compared to termination early and late in the preceding year.

At Winnipeg-93, termination method had no effect on the available N supply (Fig. 3.6).

### Nitrogen Uptake by Wheat

For all treatments, total N accumulation by wheat increased rapidly until approximately 30 d before crop maturity then increased at a markedly slower rate or declined (Fig. 3.7). Total N accumulations by wheat at crop maturity were not consistently related to spring soil  $\text{NO}_3^-$  content. A relationship between total N accumulations at crop maturity and spring soil  $\text{NO}_3^-$  content was evident only at the Carman-92 site in 1994 ( $R^2=0.51$ ,  $P=0.0004$ ,  $n=25$ ).

At all sites except Winnipeg-93, the N concentration in wheat top-growth sampled prior to filling (2 to 3%) was sufficient according to plant tissue guidelines developed for Manitoba conditions (Manitoba Provincial Soil Testing Laboratory 1982, unpublished) (Tables A.13 to A.16 in Appendix A). At Winnipeg-93, N concentrations in wheat tissue (1.5 to 2%) were marginal; however, no visual symptoms of N deficiency were evident.

Nitrogen uptake patterns at Glenlea-92 and Carman-92 followed similar trends in 1993. Significant method\*time interactions indicated differential responses to termination time among termination treatments during a number of sampling periods (Table 3.4). At Glenlea-92 and Carman-92, patterns of N uptake in tillage treatments were similar regardless of time of termination (Fig. 3.7). In contrast, in herbicide treatments, late termination delayed N uptake by wheat compared to early termination. However, N uptake in late herbicide-termination treatments appeared to increase late in the growing season after N uptake in early herbicide-terminated plots had begun to level off or decline. Late termination also appeared to delay somewhat N uptake in herbicide+tillage treatments although differences between early and late termination were not as marked

as those observed between early and late termination in the herbicide only treatments. By crop maturity, no differences among treatments were evident at Carman-92. However, at Glenlea-92, N uptake was found to be lower in herbicide+tillage treatments than in herbicide or tillage treatments.

Nitrogen uptake patterns at Carman-93 in 1994 did not follow the general patterns observed at Glenlea-92 and Carman-92 in 1993. Overall, termination treatments had minimal effects on N uptake (Table 3.4). Early-season N accumulations were slightly lower in tillage than in herbicide treatments, and also lower in late than early termination treatments. At crop maturity, N accumulations in wheat were less in late than early-terminated treatments but were similar regardless of termination method; further delaying herbicide application until the spring immediately prior to alfalfa termination further reduced N accumulations. The reason time of herbicide termination had a stronger impact on N uptake patterns at Glenlea-92 and Carman-92 in 1993 than at Carman-93 in 1994 is not clear.

At Winnipeg-93, early-season N accumulations by wheat were greater in tillage than herbicide treatments, but termination method had no effect on N accumulations throughout the remainder of the growing season or at crop maturity (Fig. A.1 and Table 3.4).

#### Grain Yield and N Concentration

Grain yield of the initial wheat crop established after alfalfa termination varied considerably among termination treatments at sites established in 1992 (Table 3.5). At

Glenlea-92, grain yields in herbicide and tilled treatments did not differ ( $1844 \text{ kg ha}^{-1}$ ) despite significantly lower spring soil  $\text{NO}_3^-$  accumulations in herbicide treatments; lower yields were observed in herbicide+tillage treatments ( $1709 \text{ kg ha}^{-1}$ ). At Carman-92, grain yields were highest in herbicide treatments ( $2853 \text{ kg ha}^{-1}$ ), intermediate in herbicide+tillage treatments ( $2703 \text{ kg ha}^{-1}$ ) and lowest in tillage treatments ( $2508 \text{ kg ha}^{-1}$ ) although spring soil  $\text{NO}_3^-$  accumulations averaged  $28 \text{ kg ha}^{-1}$  higher in tilled than in herbicide treatments. The reason for higher yields in herbicide than tillage treatments is not clear. Wheat accumulated similar amounts of N throughout the growing season regardless of termination method which suggests that N nutrition likely had minimal effects on yield. Results of a concurrent study assessing the impact of termination technique on soil moisture suggested that larger soil moisture reserves in herbicide treatments during the spring of 1994 may have contributed to higher yields (Bullied, Dept. of Plant Science, University of Manitoba, unpublished data).

Delaying termination until after the 2nd alfalfa cut did not affect grain yield at either Carman-92 or Glenlea-92 despite substantially lower soil  $\text{NO}_3^-$  accumulations in late than early termination treatments. Further delaying herbicide application until the next spring immediately prior to wheat establishment substantially reduced the grain yields at Glenlea-92 and Carman-92. Termination treatments had no effect on the yield of subsequent wheat crops established at either Glenlea-92 or Carman-92 during the second growing season (1994) after alfalfa termination (Table 3.5).

Although termination method strongly influenced grain yield at Glenlea-92 and Carman-92, method had no effect on the grain yield of the initial wheat crop grown at

sites established in 1993 (Table 3.5). However, at Carman-93, delaying termination until after the 2nd cut of alfalfa decreased grain yield regardless of termination method; yields were further reduced by delaying herbicide application until the following spring.

Termination treatment had minimal effects on the harvest index of wheat crops established after alfalfa (Table A.17 in Appendix A). At Glenlea-92 in 1993 and 1994, delaying termination increased the harvest index of wheat indicating that a larger proportion of dry matter was converted into grain yield where termination and hence N release had been delayed. No other effects of termination treatment on harvest index were evident.

Reported effects of legume termination techniques on the grain yield of subsequent crops are inconsistent. In field studies conducted in Saskatchewan, the grain yield of a wheat crop following a legume green manure was 22% less where the green manure was terminated by herbicide application than where it had been terminated by tillage (Biederbeck and Slinkard 1988). In field studies conducted in Idaho, alfalfa termination by fall-applied herbicide did not release sufficient N to meet the needs of subsequent winter or spring wheat crops resulting in lower wheat yield and protein contents (Westermann and Crothers 1993). In contrast, corn yields following a winter green manure crop were higher in a no-till than a conventional-till system; the authors attributed this to the higher soil moisture content of the no-till system (Sarrantonio and Scott 1988). However, in other field studies, the fertilizer N response of corn was found to be the same under conventional and no-till management (Triplett et al. 1979; Levin et al. 1987).

In the present study, termination treatment affected the N concentration of grain



at one site only (Table 3.5). At Carman-92 in 1993, the grain N concentration was higher in treatments involving tillage than in herbicide only treatments and, regardless of termination method, the N concentration in grain was significantly higher in early termination treatments. At Glenlea-92 in 1993 and 1994 and at Carman-92 in 1994, grain N concentrations in herbicide treatments tended to be slightly lower than in herbicide+tillage and/or tillage treatments, but these effects were not statistically significant. At Glenlea-92 in 1994, delaying herbicide application until the spring immediately before wheat establishment resulted in a significantly higher grain N concentration than herbicide termination in the previous growing season. This effect was not evident at the remaining sites.

## SUMMARY AND CONCLUSIONS

Method and timing of termination strongly affected the pattern of N release from alfalfa residues. Compared to intensive tillage which has traditionally been used to terminate established alfalfa, herbicide application without tillage delayed N release from alfalfa residues and reduced the short-term plant-available N supply. Results of controlled environment studies suggest that observed differences are primarily a function of residue placement rather than termination method *per se* (Chapter 5): N mineralization of soil incorporated residues is enhanced with greater exposure of residues to soil microbial populations (Cogle et al. 1987). In addition, incorporation may substantially reduce volatile N losses from alfalfa residues (Janzen and McGinn 1991; Chapter 4).

Under Manitoba conditions, delayed N release from the residues of herbicide-

terminated alfalfa appeared to improve the synchrony between N release from alfalfa residues and N needs of a subsequent spring wheat crop. Although herbicide treatments had lower spring soil  $\text{NO}_3^-$  levels than tilled treatments, mineralization during the growing season appeared to provide sufficient N to attain yields similar to or greater than wheat established after tillage-terminated alfalfa. In the one case in which termination by herbicide application resulted in higher yields than tillage, larger soil moisture reserves in the untilled system may have contributed to higher grain yields (Bullied, Dept. of Plant Science, University of Manitoba, unpublished data). Because smaller amounts of inorganic N accumulate in herbicide than tilled treatments prior to establishment of a subsequent crop, leaching and denitrification losses may be reduced.

Although delaying alfalfa termination until after the second cut of alfalfa reduced soil  $\text{NO}_3^-$  accumulations at three of four sites, delaying termination until after the second alfalfa cut reduced grain yields only at one site. Thus, delaying termination until after the second cut of alfalfa might occasionally reduce the grain yield of a subsequent crop compared to early termination; however, if the value of a second hay harvest exceeds the value of grain yield lost (or the value of N fertilizer required to compensate for reduced N release in late-terminated treatments), delaying termination may be an economically viable option. Further delaying herbicide application until the spring immediately prior to wheat establishment delayed N release from alfalfa residues and significantly reduced grain yields at 3 of 5 sites.

At all sites, the effects of method and time of alfalfa termination had the greatest impact on N dynamics and grain production during the initial year following alfalfa

termination. By the second growing season following alfalfa termination, differences among termination treatments had largely disappeared although significant amounts of plant-available N were present.

In summary, under conditions like those in southern Manitoba, herbicide application may be a viable, or possibly even a superior, alternative to tillage for the termination of established alfalfa stands. Herbicide application allowed sufficient mineralization to meet the N needs of a subsequent wheat crop but may reduce the potential for N losses by reducing the size of the soil inorganic N pool. Despite the benefits associated with herbicide termination, extrapolation of these findings to other situations demands caution. Because of the many differences between termination management systems including differences in residue placement, soil temperature and degree of soil disturbance, predicting the timing, amount and fate of N released following alfalfa termination is complex. Effects of termination method on N dynamics are further complicated by the confounding effect of soil moisture. Herbicide-termination often results in larger soil moisture reserves than tillage which, depending on prevailing conditions, may enhance N mineralization or produce conditions conducive to denitrification and leaching (Thomas et al. 1973). In addition, N dynamics may be influenced by factors unique to each field situation like environmental conditions and the quantity and N concentration of alfalfa residues, both of which strongly influence the availability of N. Nonetheless, results of this study demonstrate that, under conditions conducive to N mineralization, herbicide application has potential to improve synchrony between N release from alfalfa residues and N needs of a subsequent spring wheat crop

which may improve N use efficiency and reduce the potential for N losses.

Table 3.1. Physical and chemical characteristics of soils at field sites in Manitoba established in 1992 and 1993.

Characteristic <sup>†</sup>	Depth (cm)	Site			
		Glenlea-92	Carman-92	Carman-93	Winnipeg-93
Soil name		Osborne	Denham	Denham	Riverdale
Soil classification		Rego Black Chernozem	Orthic Black Chernozem	Orthic Black Chernozem	Cumulic Regosol
Texture					
% Sand	0-15	9	50	78	13
% Silt	0-15	26	21	9	45
% Clay	0-15	66	30	13	42
pH	0-15	7.1	6.6	6.2	7.4
EC (dS m <sup>-1</sup> )	0-15	0.32	0.36	0.50	0.30
Total N (g kg <sup>-1</sup> )	0-15	3.90	3.36	1.72	2.63
	15-30	2.56	2.25	1.25	2.45
	30-60	1.44	1.22	0.74	1.77
	60-90	1.03	0.91	0.56	1.38
	90-120	0.87	0.75	0.48	0.99
Extr. P (mg kg <sup>-1</sup> )	0-15	10.6	10.1	6.8	29.4
Extr. K (mg kg <sup>-1</sup> )	0-15	493	390	162	442

<sup>†</sup> Soil texture was determined by pipette method, total N by an automated combustion technique (Carlo Erba<sup>™</sup>, Milan, Italy), sodium bicarbonate extractable P by a colorimetric method (Olsen and Sommers 1982) and ammonium acetate extractable K by atomic absorption (Knudsen et al. 1982). Soil pH and electrical conductivity were determined in water (1:2 ratio of soil:water).

Table 3.2. Mineralizable N concentration in surface soil (0-15 cm) collected from field sites in the spring following alfalfa termination (based on a laboratory incubation procedure). [The soil used in the laboratory incubation procedure had been air dried and ground (<2 mm) using a rotating sieve. Plant residues which did not pass through the sieve were discarded.]

Time	Method	1993		1994	
		Glenlea-92	Carman-92	Carman-93	Winnipeg-93
-----mg N kg <sup>-1</sup> soil <sup>†</sup> -----					
Early	herbicide	65	44	35	n.d.
	herb.+till	80	48	n.d.	n.d.
	tillage	69	48	30	n.d.
Late	herbicide	72	46	36	22
	herb.+till	72	53	n.d.	n.d.
	tillage	72	50	39	22
Spring	herbicide	n.d.	n.d.	42	n.d.
Significance (P) <sup>‡</sup>					
	Rep	0.52	0.12	0.09	0.82
	Time	0.83	0.23	0.003	n.d.
	Method	0.46	0.19	0.91	0.98
	M*T	0.47	0.86	0.006	n.d.
C.V. (%)		16.9	13.1	13.6	22.2

<sup>†</sup> At all sites, mineralizable C and N were significantly correlated with correlation coefficient (r) and significance as follows: Glenlea-92  $r=0.74^{**}$ , Carman-92  $r=0.75^{**}$ , Winnipeg-93  $r=0.82^{**}$  and Carman-93  $r=0.65^{**}$  where  $^{**}$  indicates significance at  $P=0.01$ . Termination treatment had no effect on mineralizable C which averaged 750 mg C kg<sup>-1</sup> soil at Glenlea-92, 580 at Carman-92, 380 at Carman-93 and 340 at Winnipeg-93. The term n.d. indicates that the value was not determined.

<sup>‡</sup> At Glenlea-92 and Carman-92, degrees of freedom (df) for effects of rep, time, method and M\*T were 3, 1, 2 and 2 respectively. At Carman-93, df for effects of rep, time, method and M\*T were 3, 1, 1, and 1, respectively. At Winnipeg-93, df for effects of rep and method were 3 and 1, respectively.

Table 3.3. Plant-available N mineralized during the first or second growing season [calculated as the difference in the cumulative available N supply (soil  $\text{NO}_3^-$ -N to 60 cm + wheat N removed) between spring and fall of a growing season] following alfalfa termination as influenced by method and time of alfalfa termination.

Time	Method	1st growing season after alfalfa termination				2nd growing season after alfalfa termination	
		Glenlea-92	Carman-92	Carman-93	Winnipeg-93	Glenlea-92	Carman-92
-----kg N ha <sup>-1</sup> -----							
Early	herbicide	62	99	108	n.d.	74	39
	herb.+till	23	49	n.d.	n.d.	110	35
	tillage	40	48	7	n.d.	83	26
Late	herbicide	51	143	114	27	103	35
	herb.+till	33	110	n.d.	n.d.	94	35
	tillage	47	84	30	32	80	23
Spring	herbicide	n.d.	n.d.	78 <sup>†</sup>	n.d.	n.d.	n.d.
Significance (P) <sup>§</sup>							
	Time	0.75	0.03	0.59	n.d.	0.73	0.81
	Method	0.01 <sup>‡</sup>	0.10	0.004	0.55	0.38	0.36
	M*T	0.39	0.87	0.75	n.d.	0.87	0.83
C.V. (%)		38	48	33	32	23	62

<sup>†</sup> Single degree of freedom contrasts indicated no difference between spring and early-summer herbicide application (P=0.10) or between spring and late-summer herbicide termination (P=0.11).

<sup>‡</sup> The least significant difference (LSD) for testing among methods is 18. The term n.d. indicates that N mineralization was not determined.

<sup>§</sup> At Glenlea-92 and Carman-92, degrees of freedom (df) for effects of time, method and M\*T were 1, 2 and 2 respectively. At Carman-93, df for effects of time, method and M\*T were 1, 1, and 1, respectively. At Winnipeg-93, df for the effects of method was 1.

Table 3.4 Summary of the statistical significance (P value) of method and time of alfalfa termination on N accumulations by the first wheat crop established after alfalfa termination.

ANOVA	df	Sampling time						Crop maturity
		1	2	3	4	5	6	
Glenlea-92								
Rep	3	0.03	0.07	0.04	0.17	0.39	0.50	0.86
Method (M)	2	0.36	0.50	0.02	0.06	0.21	0.40	0.03
Time (T)	1	0.23	0.97	0.0004	0.0002	0.0001	0.003	0.09
M*T	2	0.17	0.50	0.005	0.03	0.19	0.004	0.82
C.V. (%) <sup>†</sup>		25.8	25.5	12.9	11.9	12.3	14.4	15.3
Carman-92								
Rep	3	0.0001	0.05	0.02	0.13	0.10	0.07	0.97
Method (M)	2	0.08	0.72	0.25	0.70	0.87	0.27	0.42
Time (T)	1	0.004	0.0001	0.0001	0.01	0.002	0.05	0.82
M*T	2	0.007	0.001	0.60	0.26	0.02	0.07	0.86
C.V. (%)		7.45	7.71	9.46	15.52	13.13	14.14	24.04
Carman-93								
Rep	3	0.93	0.13	0.45	0.57	0.87	n.d.	0.56
Method (M)	1	0.008	0.32	0.49	0.79	0.72	n.d.	0.16
Time (T)	1	0.03	0.006	0.34	0.08	0.23	n.d.	0.005
M*T	1	0.27	0.55	0.98	0.77	0.06	n.d.	0.99
C.V. (%)		15.58	12.96	12.38	15.75	20.39	n.d.	8.82
Winnipeg-93								
Rep	3	0.32	0.61	0.57	0.45	n.d.	n.d.	0.06
Method (M)	1	0.04	0.67	0.61	0.36	n.d.	n.d.	0.49
C.V. (%)		21.48	25.95	20.42	8.02	n.d.	n.d.	5.34

<sup>†</sup> The reported C.V. is based on all treatments sampled including, where applicable, spring herbicide treatments. 'n.d.' indicates that N uptake was not determined for a given sampling time, or that N uptake was measured but missing values prevented statistical analysis (Winnipeg-93, 5th sampling period).



Table 3.5. Grain yield and grain N concentration of spring wheat crops established during the first and second growing seasons following alfalfa termination.

Time	Method	1st growing season after alfalfa termination								2nd season after alfalfa termination			
		Glenlea-92		Carman-92		Carman-93		Winnipeg-93		Glenlea-92		Carman-92	
		Yield (kg ha <sup>-1</sup> )	g N kg <sup>-1</sup>	Yield (kg ha <sup>-1</sup> )	g N kg <sup>-1</sup>	Yield (kg ha <sup>-1</sup> )	g N kg <sup>-1</sup>	Yield (kg ha <sup>-1</sup> )	g N kg <sup>-1</sup>	Yield (kg ha <sup>-1</sup> )	g N kg <sup>-1</sup>	Yield (kg ha <sup>-1</sup> )	g N kg <sup>-1</sup>
Early	herbicide	1828	34.1	2926	32.9	2810	33.7	n.d.	n.d.	2419	30.4	2443	28.7
	herb.+till	1751	35.1	2624	34.1	n.d. <sup>†</sup>	n.d.	n.d.	n.d.	2648	30.8	2414	30.6
	tillage	1860	34.4	2481	34.2	2690	34.5	n.d.	n.d.	2852	31.6	2349	29.5
Late	herbicide	1853	34.7	2779	31.4	2402	33.6	2868	24.8	2452	30.5	2390	28.1
	herb.+till	1667	35.1	2782	33.1	n.d.	n.d.	n.d.	n.d.	2881	31.3	2425	28.9
	tillage	1835	33.8	2534	33.3	2213	33.3	2898	25.3	2707	31.8	2432	29.9
Spring	herbicide	1678	34.2	2625	32.1	1546	33.7	n.d.	n.d.	2685	32.8	2307	28.8
LSD <sub>(0.05)</sub> (method)		121	0.8	148	0.8	229	1.0	n.d.	n.d.	418	1.1	160	1.3
Significance (P) <sup>‡</sup>													
Rep		0.69	0.18	0.11	0.009	0.17	0.17	0.06	0.53	0.003	0.002	0.08	0.01
Time		0.56	0.97	0.71	0.001	0.002	0.18	n.d.	n.d.	0.81	0.55	0.82	0.43
Method		0.05	0.06	0.0007	0.0009	0.16	0.65	0.77	0.48	0.18	0.09	0.91	0.11
M*T		0.64	0.32	0.12	0.61	0.74	0.22	n.d.	n.d.	0.64	0.88	0.67	0.46
Contrasts													
Spring vs late herbicide		0.03	0.38	0.16	0.16	0.0001	0.97	n.d.	n.d.	0.39	0.04	0.46	0.38
Spring vs early and late herbicide		0.02	0.68	0.02	0.92	0.0001	0.99	n.d.	n.d.	0.29	0.02	0.26	0.52
C.V. (%)		6.0	2.3	5.6	2.2	8.5	3.6	4.6	3.3	14.0	4.6	6.5	3.8

<sup>†</sup> n.d. indicates that the value was not determined.

<sup>‡</sup> At Glenlea-92 and Carman-92, degrees of freedom (df) for effects of rep, time, method and M\*T were 3, 1, 2 and 2 respectively. At Carman-93, df for effects of rep, time, method and M\*T were 3, 1, 1, and 1, respectively. At Winnipeg-93, df for effects of rep and method were 3 and 1, respectively.

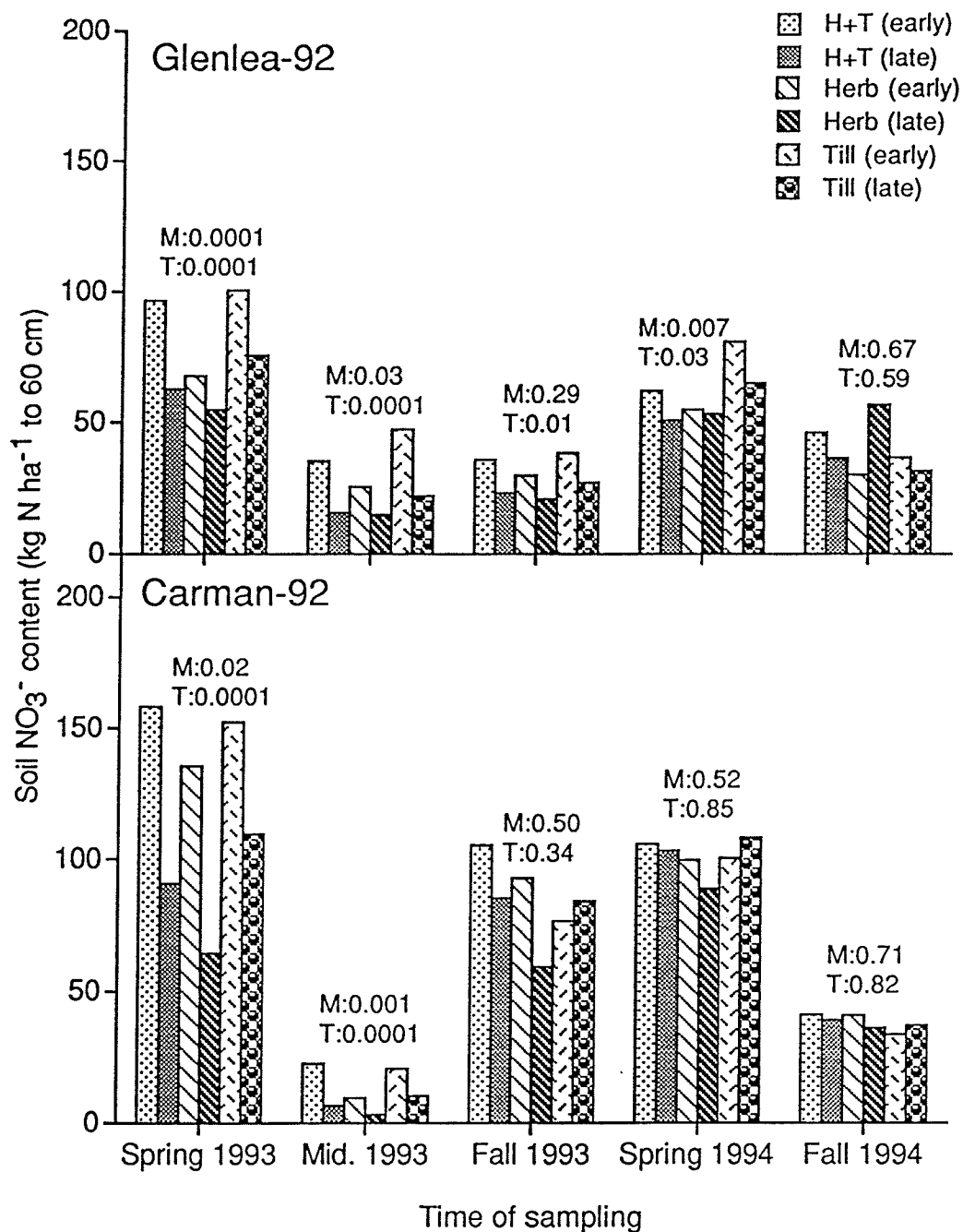


Fig. 3.1. Soil  $\text{NO}_3^-$ -N accumulations at Glenlea-92 and Carman-92 during two growing seasons (1993 and 1994) following the termination of perennial alfalfa stands as influenced by method and time of termination. The statistical significance (P value) of method (M) and time (T) is shown for each sampling time. Method\*time interactions were not significant at  $P=0.05$ .

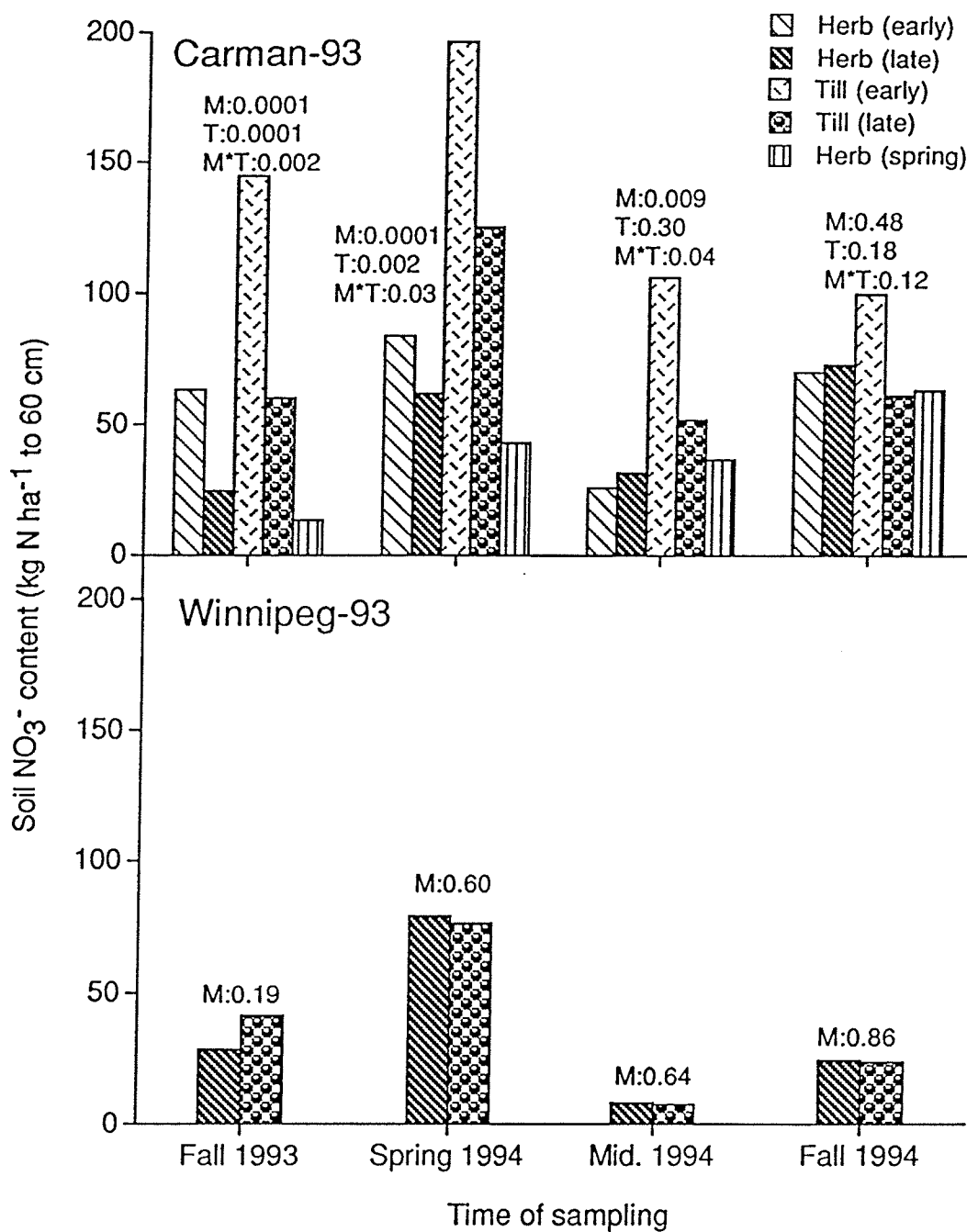


Fig. 3.2. Soil  $\text{NO}_3^-$ -N accumulations at Carman-93 and Winnipeg-93 during one growing season (1994) following the termination of perennial alfalfa stands as influenced by method and time of termination. The statistical significance (P value) of method (M), time (T) and method\*time interactions (M\*T) is shown for each sampling time.

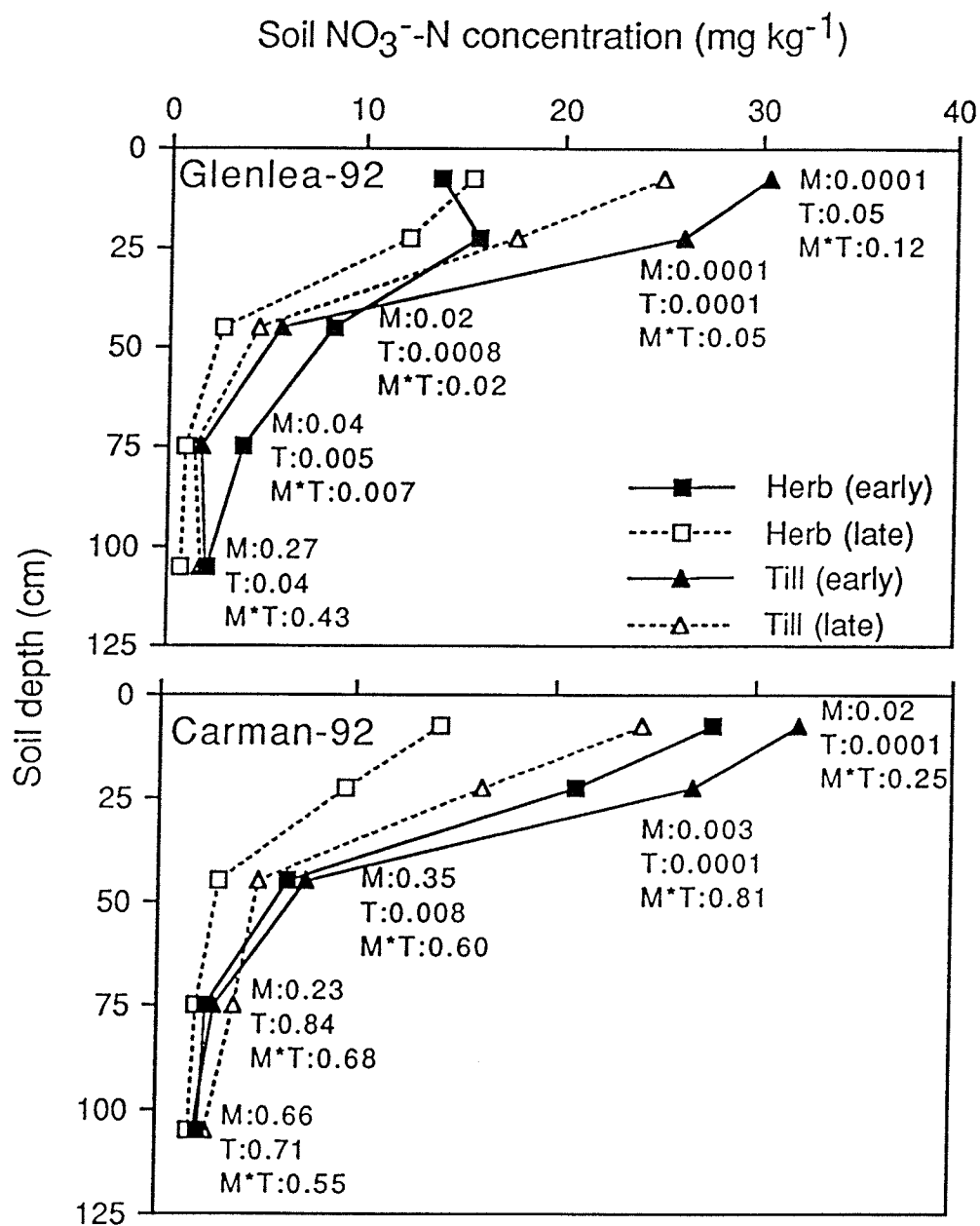


Fig. 3.3. Soil  $\text{NO}_3^-$ -N concentrations at Glenlea-92 and Carman-92 during the first spring after termination of perennial alfalfa as influenced by method and time of termination. The statistical significance (P value) of method (M), time (T) and method\*time interactions (M\*T) is shown for each depth.

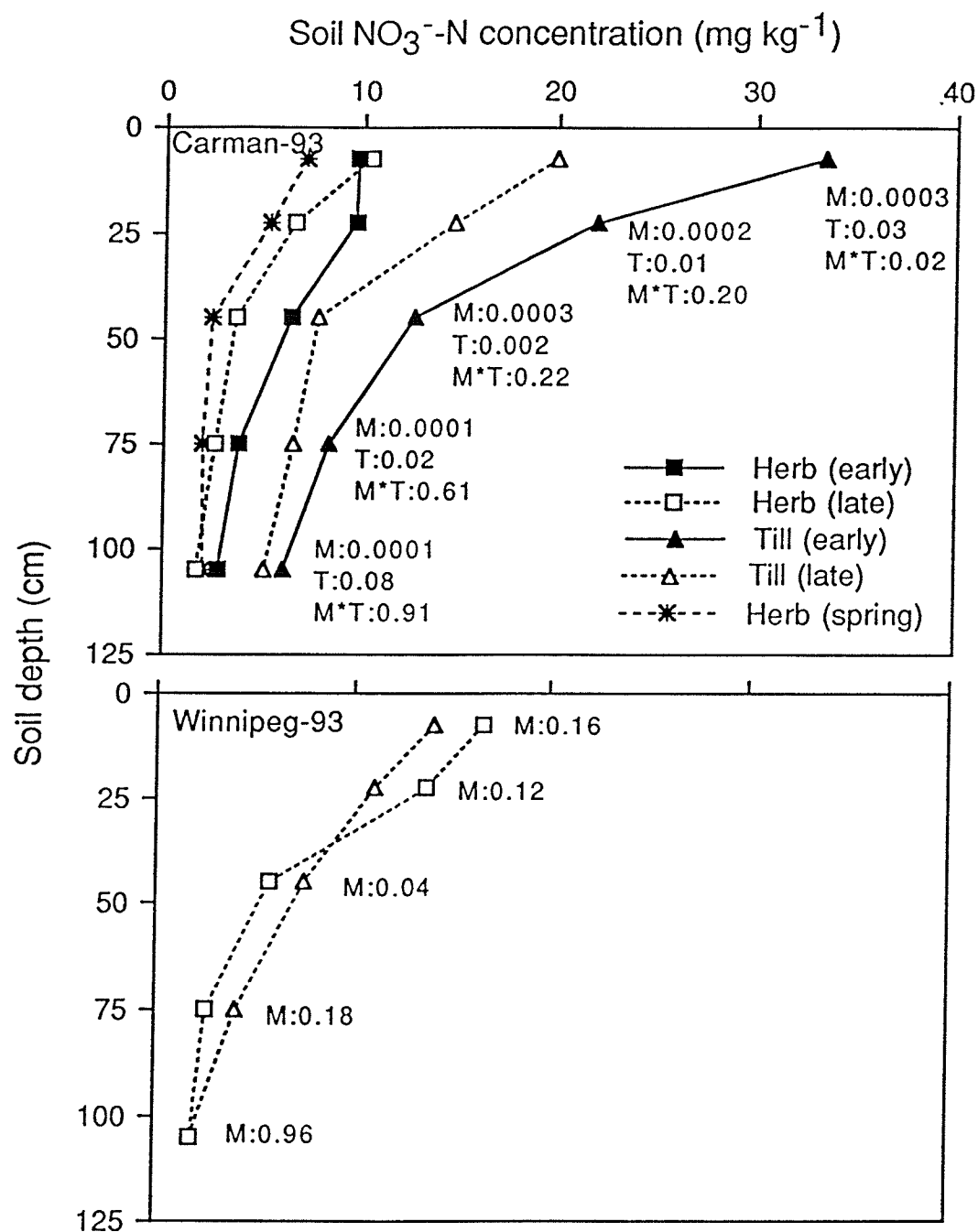


Fig. 3.4. Soil  $\text{NO}_3^-$ -N concentrations at Carman-93 and Winnipeg-93 in the first spring following termination of perennial alfalfa as influenced by method and time of termination. The statistical significance (P value) of method (M), time (T) and method\*time interactions (M\*T) is shown for each depth.

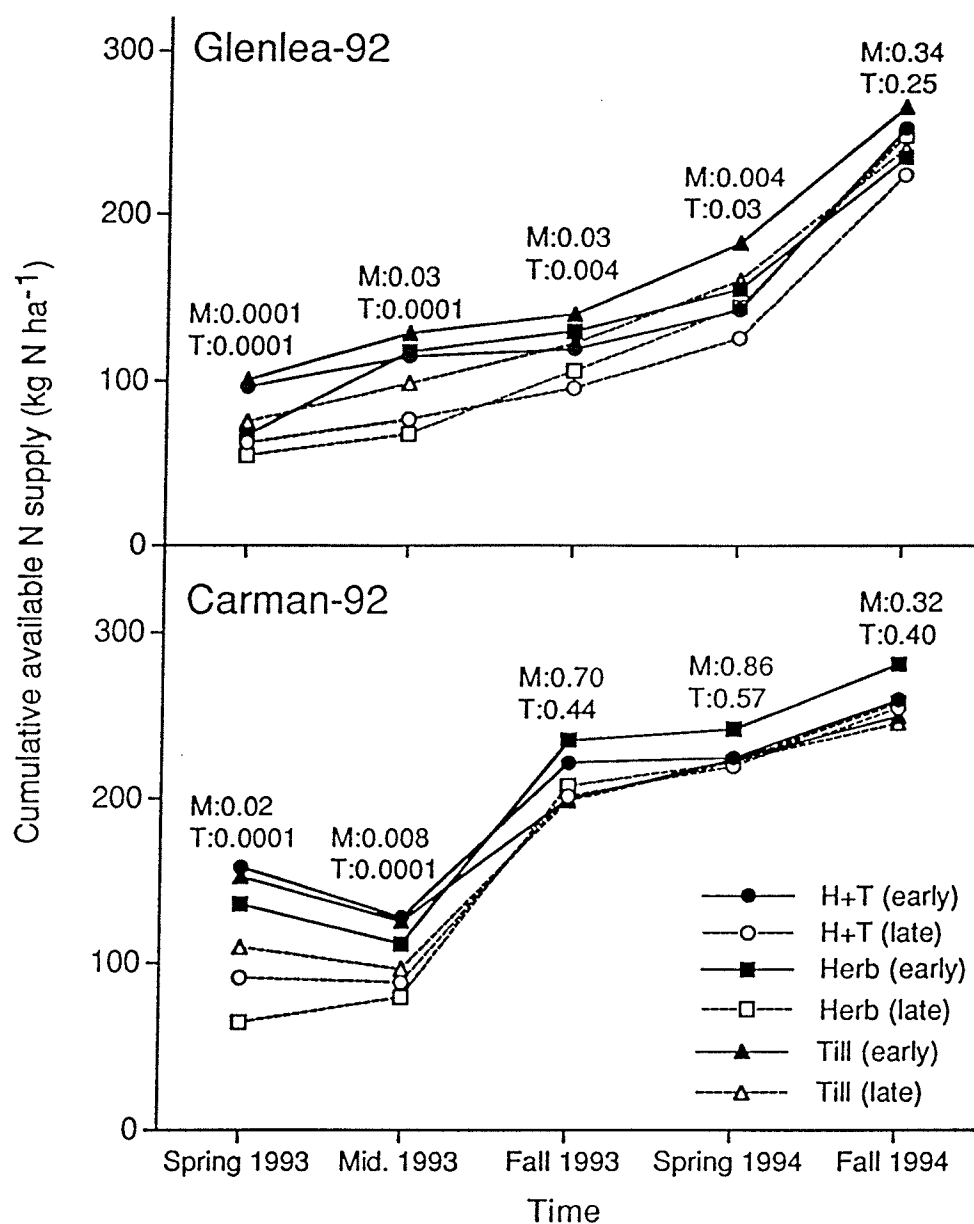


Fig. 3.5. Cumulative available N supply (calculated as the sum of N present as soil  $\text{NO}_3^-$ -N to 60 cm and cumulative N removed in the grain and straw of wheat) at Glenlea-92 and Carman-92 during two growing seasons following the termination of perennial alfalfa stands as influenced by method and time of termination. The statistical significance (P value) of method (M) and time (T) is shown for each sampling time. Method\*time interactions were not significant at  $P=0.05$ .

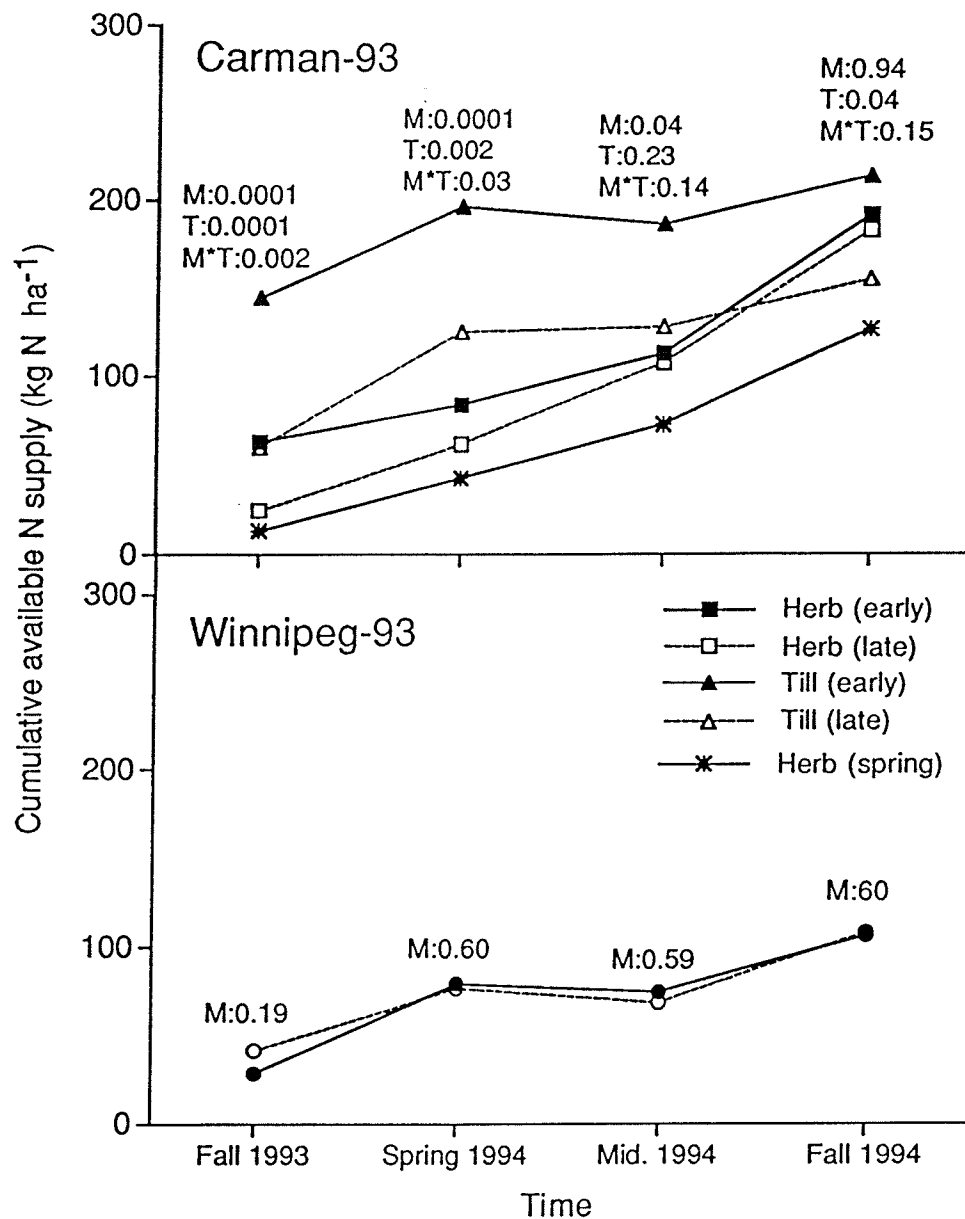


Fig. 3.6. Cumulative available N supply (calculated as the sum of N present as soil NO<sub>3</sub><sup>-</sup>-N to 60 cm and N removed in the grain and straw of wheat) at Carman-93 and Winnipeg-93 during one growing season following the termination of perennial alfalfa stands as influenced by method and time of termination. The statistical significance (P value) of method (M), time (T) and method\*time (M\*T) interactions is shown for each sampling time.

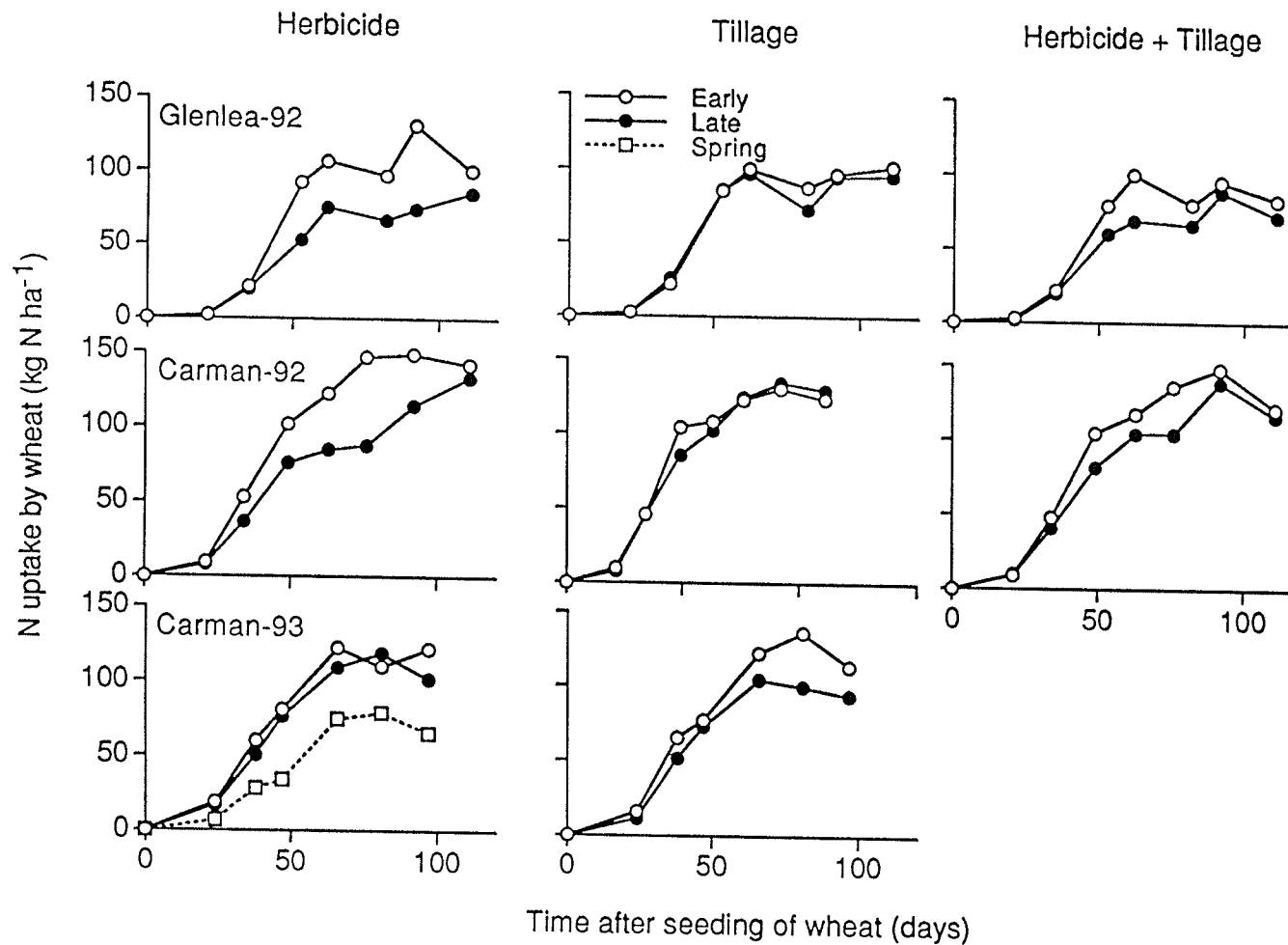


Fig. 3.7. Nitrogen accumulations by wheat at Glenlea-92, Carman-92 and Carman-93 during the initial growing season following alfalfa termination by various methods at several times.



#### 4. Nitrogen Dynamics as Influenced by Method of Alfalfa Termination: Volatile N Losses

##### ABSTRACT

Use of herbicide to terminate alfalfa (*Medicago sativa*), while reducing soil erosion, could conceivably enhance volatile N losses from alfalfa residues. A controlled environment study was conducted to determine the effect of termination technique on the quantity of  $\text{NH}_3$  volatilized from residues of alfalfa. A factorial combination of two termination methods (chemical, mechanical) and two methods of residue placement (incorporated, surface-applied) was applied to alfalfa. Treatments were incubated in a flow-through chamber in the greenhouse for 95 d during which  $\text{NH}_3$  and  $\text{CO}_2$  evolution was measured periodically. After 95 d, the equivalent of 8% to 12% of the N in surface-applied legume residues was lost as  $\text{NH}_3$ . These losses amounted to 3% to 4% of total legume N in surface-applied residue treatments. Termination method did not have a significant effect on the amount of alfalfa N lost via volatilization. Incorporation of alfalfa residue essentially eliminated volatile N losses. Residue incorporation increased cumulative  $\text{CO}_2$  evolution regardless of termination method. Results of this study suggest that termination techniques in which alfalfa residue is retained on the soil surface may result in significant volatile N losses which could diminish the fertilizer value derived from alfalfa residues. The benefits of herbicide termination over tillage termination (e.g. reductions in soil erosion and moisture loss) must be weighed against the diminished fertilizer value derived from residues of alfalfa terminated by herbicide application.

## INTRODUCTION

Established alfalfa (*Medicago sativa* L.) stands are typically terminated by intensive tillage. Recently, however, herbicide application has been suggested as an alternative. Herbicide application not only effectively controls alfalfa but also eliminates tillage, thereby reducing the potential for soil erosion and soil moisture loss. In addition, snow trapping by standing residue retained on the soil surface can increase soil moisture reserves (Bullied, Dept. of Plant Science, University of Manitoba, unpublished data).

Although herbicide application offers numerous advantages over tillage, the alfalfa residues maintained on the soil surface may be subject to volatile N losses during senescence and decomposition. Ammonia volatilization losses during senescence have been documented in various crops including corn (*Zea mays*), spring wheat (*Triticum aestivum*) and ryegrass (*Lolium perenne*) (Farquhar et al. 1979; Parton et al. 1988; Whitehead et al. 1988). Also, senescing alfalfa leaves have been identified as a potential source of small amounts of  $\text{NH}_3$  evolved from an alfalfa field (Dabney and Bouldin 1985). In the latter study, increased  $\text{NH}_3$  evolution also appeared to be associated with frost damage of alfalfa plants and the presence of alfalfa residue on the soil surface following hay harvest.

Losses of  $\text{NH}_3$  from decomposing alfalfa may be higher than those observed during senescence. Volatilization from decomposing perennial ryegrass (*Lolium perenne*) incubated for a 70 d period was equivalent to 20 to 47% of herbage N (Whitehead et al. 1988). Bremer and van Kessel (1992) reported volatile N losses of 42, 3.4 and 0.5% of residue N from lentil (*Lens culinaris*) green manure, lentil straw and wheat (*Triticum*

*aestivum*) straw incubated for 28 d in the absence of soil. In both studies, the greater the N concentration in decomposing plant residue, the greater the proportion of N lost by volatilization. In greenhouse studies, lentil green manure placed on the soil surface or suspended above the soil surface lost up to 14% of residue N present within 14 d of application, but incorporation of lentil residue into the soil virtually eliminated  $\text{NH}_3$  loss (Janzen and McGinn 1991). Volatile N losses from other organic amendments have also been sharply reduced by incorporation (Adamsen and Sabey 1987).

Ammonia volatilization may diminish the fertilizer value of surface-applied legume residues. Short term fertility may be particularly affected since labile fractions of legume N are most susceptible to volatile N losses. Various studies have demonstrated that available N supply is greater from incorporated than from surface-applied legume residues (McCalla and Russel 1948; McKay et al. 1952; Wilson and Hargrove 1986; Sarrantonio and Scott 1988). In field studies, incorporation of  $^{15}\text{N}$ -labelled hairy vetch (*Vicia villosa*) resulted in recovery of 32% of residue N by a subsequent corn crop; only 20% of the N present in surface-applied vetch residue was recovered by a subsequent corn crop (Varco et al. 1989). Biederbeck and Slinkard (1988) reported a 22% lower grain yield for wheat following legume green manure terminated by herbicide and retained on the soil surface than for wheat following incorporated legume green manure. Differences between placement methods are generally thought to be the result of a higher rate of N mineralization from incorporated residue. However, greater volatile N losses from surface-applied residues may also contribute to the differences observed (Janzen and McGinn 1991; Bremer and van Kessel 1992).

Significant N contributions made by alfalfa to the available N supply are well-documented (Lyon and Bizzell 1933; Hoyt and Hennig 1971; Bruulsema and Christie 1987) but the effect of termination method on the fertilizer value derived from established alfalfa stands has not been determined. Efficient utilization of alfalfa-derived N requires a better understanding of the magnitude of volatile N losses from alfalfa following termination.

The objective of this study was to determine the effect of termination method and residue placement on the amount of  $\text{NH}_3$  volatilized from alfalfa residue. Our approach involved quantification of  $\text{NH}_3$  evolution from residues of alfalfa terminated by herbicide or simulated tillage under greenhouse conditions.

## MATERIALS AND METHODS

A series of experiments was initiated to study the effect of method of alfalfa termination on N dynamics. Four termination treatments, consisting of a factorial of two termination methods (chemical, mechanical) and two methods of residue placement (surface-applied, incorporated), were established. Three replications of each of the four treatments were incubated in flow-through chambers under greenhouse conditions and  $\text{NH}_3$  evolution was measured periodically throughout the 95 d incubation period. Additional replicates were used in a complementary experiment to determine the effect of termination method on N mineralization and N uptake by subsequent crops (Chapter 5).

### Establishment and Greenhouse Techniques

The A horizon of a Cavendish sandy loam soil (pH in  $\text{CaCl}_2=6.9$ ; total N=0.74 g  $\text{kg}^{-1}$ ; organic C=7.6 g  $\text{kg}^{-1}$ ) was air dried and passed through a 2 mm sieve. (This soil was collected near Purple Springs, AB and stockpiled at the Lethbridge Research Centre prior to its use.) Air dry soil was moistened to 50% of field capacity and nutrient solutions were thoroughly mixed into the soil to provide 7 mg N, 41 mg P, 51 mg K, 10 mg S, 1 mg Cu, 2 mg Mn, 4 mg Zn, 0.4 mg Mo, 1 mg B and 4 mg Fe per kilogram soil. The equivalent of 5.43 kg (oven dry basis) soil was added to each pot resulting in a headspace volume of approximately 0.7 L in each pot.

Twenty presoaked 'Nitro' alfalfa seeds were placed on the soil surface of each pot. At time of seeding, 1-2 drops of a slurry of Rhizobium-peat (commercial inoculant Nitragin, Liphatech, Madison, WI) in water were applied to each seed with a glass pipette. The soil surface was covered with moistened filter paper and each pot watered to field capacity (16.5% w/w). After alfalfa germinated, the filter paper was removed and alfalfa was thinned to six plants per pot.

Alfalfa was grown in a greenhouse from November 1993 through March 1994. During the growing period, alfalfa was harvested twice at the bloom stage by cutting top-growth 8 cm above the soil surface (Fig. 4.1). Following the second harvest, alfalfa was allowed to regrow to a height of approximately 12 cm before termination treatments were applied.

### Termination of Alfalfa

Three replications of four termination treatments were established. Of the twelve pots selected for termination, six were designated for termination by tillage and six for termination by herbicide application.

For tillage treatments, alfalfa in three pots was undercut approximately 2 to 2.5 cm below-ground and alfalfa residue retained on the undisturbed soil surface. In the remaining three pots, the entire alfalfa plant including roots was removed from the soil, cut by hand into 4-5 cm lengths and the residue fragments were thoroughly mixed throughout the soil.

For herbicide treatments, glyphosate at a rate of 4000 g a.i. ha<sup>-1</sup> (the equivalent of 0.04 mg a.i. cm<sup>-2</sup>) was applied to alfalfa plants placed in a spray chamber. To prevent herbicide movement into the soil, a layer of vermiculite was applied to the soil surface of each pot prior to herbicide application and removed immediately after. In three herbicide-treated pots, standing residue was retained on the undisturbed soil surface. In the other three herbicide-treated pots, alfalfa residue was completely incorporated into the soil 2 d after herbicide application. The incorporation method used was the same as that described for tillage treatments. Herbicide was applied 2 d prior to application of tillage treatments in an attempt to minimize differences in alfalfa dry matter accumulation between termination treatments. Soil bulk density determinations were not conducted following alfalfa termination, however, based on visual observation, differences between tilled and untilled treatments appeared to be minimal.

### Sampling and Analytical Techniques

Alfalfa plants in six additional pots (three herbicide-treated and three untreated) were destructively sampled at the time of tillage to provide an estimate of alfalfa N present. Plant tissue was separated into top-growth and root tissue. Soil particles were visible on root surfaces, therefore, a subsample of root tissue was taken and adhering soil removed; the remainder of the root sample was analyzed with soil adhering. Plant tissue was oven dried, dry matter yield determined, and total N and C concentration determined by an automated combustion technique (Carlo Erba<sup>TM</sup>, Milan, Italy). Root mass was calculated as follows:

Let:  $M_{R+S}$  = mass of roots+soil

$M_R$  = mass of roots

$M_S$  = mass of soil

$C_{R+S}$ ,  $C_R$ ,  $C_S$  = % C in root+soil, root and bulk soil, respectively

Then:

$$M_R = (M_{R+S}C_{R+S} - M_{R+S}C_S)/(C_R - C_S)$$

(This calculation was based on the assumption that the C concentration of the root without adhering soil and the C concentration of the bulk soil were the same as the C concentration of these fractions in the root+soil sample.)

The twelve pots each containing terminated alfalfa were watered to field capacity and sealed with a clear plexiglass cover and rubber seal. Pots were arranged randomly along a greenhouse bench and covered with a black plastic sheet to provide shade. Humidified CO<sub>2</sub>-free air was continuously passed through the headspace of each chamber

at a rate of about 0.5 displacements  $\text{min}^{-1}$ . A flow rate of 0.3 displacements  $\text{min}^{-1}$  was shown to maximize  $\text{NH}_3$  evolution from lentil green manure (Janzen and McGinn 1991). Prior to entering soil chambers, air was passed through a series of three flasks, two with 4 M NaOH and one with  $\text{CO}_2$ -free water, to remove  $\text{CO}_2$  and humidify the air, respectively. Airflow into each chamber was regulated using a system similar to that described by Weaver (1974).

Air exiting each chamber was passed through one acid trap containing 200 mL 0.1 M HCl to collect  $\text{NH}_3$  and two alkaline traps each containing 200 mL 2 M NaOH to collect  $\text{CO}_2$  (Fig. 4.2). The  $\text{NH}_3$  and  $\text{CO}_2$  concentration of air entering the sealed chambers was determined at three points along the inflow manifold using the same system of one acid and two alkaline traps. Acid and alkaline traps were sampled for 95 d following alfalfa termination at increasing intervals ranging from 1 to 21 days. The  $\text{CO}_2$  content of alkaline traps was determined by titration as described by Tiessen et al. (1981). The  $\text{NH}_3$  content of acid traps was determined by steam distillation in the presence of an excess of NaOH followed by titration with  $\text{H}_2\text{SO}_4$ . Net  $\text{NH}_3$  and  $\text{CO}_2$  accumulation from each chamber was calculated by subtracting the average  $\text{NH}_3$  and  $\text{CO}_2$  content of the incoming air.

Cumulative and daily  $\text{NH}_3$  and  $\text{CO}_2$  accumulations and cumulative  $\text{NH}_3$  evolution as a proportion of alfalfa N were analyzed by analysis of variance for a completely randomized design using the Proc GLM procedure (SAS Institute Inc. 1985).



## RESULTS

### Plant Dry Matter and N Yield

Alfalfa dry matter was higher in tillage treatments than in herbicide treatments (Table 4.1). The N yield of alfalfa top-growth was also greater in tillage than in herbicide treatments; however, the N yield of root tissue did not differ between treatments since lower dry matter accumulations in herbicide treatments were offset by significantly higher N concentrations. Attempts had been made to minimize differences among treatments by delaying tillage for 2 d following herbicide application. However, the high rate of glyphosate applied quickly halted plant growth apparently resulting in higher dry matter and N accumulations in tillage than in herbicide treatments.

### CO<sub>2</sub> Evolution

For all treatments, the rate of CO<sub>2</sub> evolution peaked within several days of alfalfa termination and then declined gradually (Fig. 4.3). This general pattern of CO<sub>2</sub> evolution is similar to that observed for decomposing lentil green manure (Janzen and McGinn 1991) and may be attributed to a three stage decomposition process comprised of an immediate increase in microbial populations followed by rapid mineralization of labile C and subsequent slower mineralization of less labile fractions.

Evolution of CO<sub>2</sub> for herbicide-incorporated residue treatments increased rapidly and peaked after 2 d of incubation. The CO<sub>2</sub> evolution in tillage-incorporated residue and herbicide-surface residue treatments followed a similar pattern. In contrast, tillage-surface residue treatments did not produce a distinct, large peak in CO<sub>2</sub> evolution rate as did the

other treatments. Lower levels of CO<sub>2</sub> evolution during early stages of residue decomposition suggest that smaller amounts of soluble and intermediately-available C were available for decomposition in tillage-surface treatments than in other treatments. Reinertsen et al. (1984) found that the size of soluble and intermediately-available C pools strongly influenced early stages of wheat straw decomposition. Differences in CO<sub>2</sub> evolution among treatments diminished over time; by the final sampling period, neither residue placement nor termination method had a significant effect on the rate of CO<sub>2</sub> evolution. (A summary of the statistical significance of effects of residue placement and termination method on daily and cumulative CO<sub>2</sub> evolution at individual sampling times is presented in Table B.1 of Appendix B.)

Regardless of termination method, the incorporation of alfalfa residue increased cumulative CO<sub>2</sub> evolution (Table 4.2), presumably as a result of greater exposure of alfalfa residue to microbial populations (Cogle et al. 1987). Herbicide application also increased cumulative CO<sub>2</sub> evolution throughout the first half of the incubation period (Table B.1 in Appendix B) but this effect was no longer evident at the end of the incubation period (Table 4.2). Greater cumulative CO<sub>2</sub> evolution in herbicide treatments may be the result of increased availability of labile C compounds to microbial populations, possibly due to root exudation of C-containing compounds following herbicide application. Herbicide application has been shown to increase exudation of C-containing compounds from root tissue (Lai and Semeniuk 1970; Lee and Lockwood 1977).

### NH<sub>3</sub> Evolution

Incorporation of alfalfa top-growth virtually eliminated NH<sub>3</sub> volatilization losses from alfalfa residue regardless of termination method (Fig. 4.4). By 95 d after alfalfa termination, cumulative NH<sub>3</sub> loss from incorporated treatments averaged 0.8 mg N which was equivalent to 0.5% of total N in alfalfa top-growth.

Large losses of NH<sub>3</sub> occurred from surface alfalfa residue. Volatilization rate increased to a maximum 9 d after alfalfa termination and declined rapidly thereafter (Fig. 4.4). By 74 d after alfalfa termination, the rate of volatilization from surface-applied alfalfa residue had declined to near that of incorporated residue. (A summary of the statistical significance of effects of residue placement and termination method on daily and cumulative NH<sub>3</sub> evolution at individual sampling times is presented in Table B.2 of Appendix B.) The majority of N volatilization occurred soon after alfalfa termination; within 18 d of alfalfa termination, the equivalent of 10% and 6% of the total amount of N in surface-applied residues had been volatilized in herbicide and tillage treatments, respectively. This pattern of NH<sub>3</sub> evolution is typical of that observed for decomposing crop residue (Whitehead et al. 1988; Janzen and McGinn 1991). Initial high rates of N loss have been attributed to volatilization following ammonification of soluble organic N while subsequent lower rates of NH<sub>3</sub> evolution likely result from volatilization of NH<sub>4</sub><sup>+</sup> produced during mineralization of legume N (Janzen and McGinn 1991).

Termination method did not have a statistically significant effect on the proportion of surface-applied N lost via volatilization (Table 4.2). By the end of the 95 d incubation period, NH<sub>3</sub> loss averaged 10% of surface-applied legume N in tillage and herbicide

treatments (Table 4.2). These losses accounted for 3.3 to 4.3% of total alfalfa N present.

## DISCUSSION

Retaining alfalfa residue on the soil surface may result in N loss via volatilization. During a 95 d incubation period, volatile N losses equal to 8.0% to 11.9% of surface-applied alfalfa N were measured in surface-applied residue treatments. The majority of volatile N loss occurred within several days of alfalfa termination. However, volatile N losses were essentially eliminated by incorporation of alfalfa residue.

The method of herbicide termination proposed as an alternative to tillage would involve maintenance of standing residue on the soil surface for an extended period of time. Although surface residue would reduce the potential for soil moisture loss and soil erosion, these surface-applied alfalfa residues may be subject to volatile N loss. Because  $\text{NH}_3$  volatilization is a diffusive process and therefore a function of the concentration gradient between the N source and the atmosphere, factors affecting the concentration gradient (e.g. the amount of air movement around residues, the quantity of residue N prone to volatile N loss) may influence the quantity of N lost via volatilization. Because a portion of this N lost via volatilization may subsequently be adsorbed by surrounding soil or plant tissue, these volatile N losses do not necessarily represent a net loss from the plant-soil system.

The relative N contributions from alfalfa shoot and root material to subsequent crops may vary substantially with the amount of alfalfa top-growth and root material present at time of termination. Compared to green manure crops, in which much of the

plant's N is present in top-growth, alfalfa roots may contain a greater proportion of the plant's N than shoot material. In this experiment, although volatile N losses accounted for up to 12% of surface-applied alfalfa N, observed losses accounted for less than 4.5% of total alfalfa N.

Alfalfa top-growth, particularly new regrowth, contains relatively high concentrations of N, much of which may be present in intermediate, labile forms susceptible to loss. Under field conditions, wetting and drying cycles and air movement may result in greater N losses from surface-applied alfalfa residues than were observed under the controlled conditions of this experiment. Volatile N loss from surface residues represents a reduction in the fertilizer value of alfalfa residue and therefore a reduction in the efficiency with which crops following alfalfa are able to utilize alfalfa-derived N.

Although volatile N losses from decomposing alfalfa residues may be significant, they may not be high enough to preclude the use of herbicides to terminate established alfalfa stands. For example, if 2000 kg ha<sup>-1</sup> above-ground alfalfa residue containing 3.5% N were present at termination, NH<sub>3</sub> volatilization of the magnitude observed in this experiment would result in losses of less than 10 kg N ha<sup>-1</sup>. Volatile N losses, although small, represent a loss from the most N labile fraction and therefore may have a greater impact on short-term than long-term soil fertility. Whether the losses are sufficient to discourage the adoption of herbicide termination of alfalfa stands depends on the relative value ascribed to its benefits.

Table 4.1. Composition of alfalfa residue present in herbicide treatments 2 d after glyphosate application and in tillage treatments at time of tillage.

		Termination method <sup>†</sup>		P>F
		Herbicide	Tillage	
Top-growth	%C	43.9	44.2	0.55
	%N	3.3	3.7	0.02
	N yield (mg)	130	181	0.003
	Mass (g)	3.93	4.95	0.008
Root	%C	44.6	43.6	0.002
	%N	3.7	2.7	0.006
	N yield (mg)	231	266	0.17
	Mass (g)	6.18	9.96	0.0008

<sup>†</sup> The C:N ratio of alfalfa top-growth was 13.3 and 11.9 in herbicide and tillage treatments, respectively. The C:N ratio of alfalfa roots was 12.1 and 16.1 in herbicide and tillage treatments, respectively.

Table 4.2. Effect of termination method (herbicide application, tillage) and residue placement (soil-incorporated, surface-applied) on cumulative  $\text{NH}_3$  and  $\text{CO}_2$  evolution from decomposing alfalfa residue.

Treatment		$\text{CO}_2$ evolution g C $\text{pot}^{-1}$	$\text{NH}_3$ evolution		
Placement	Method		mg N $\text{pot}^{-1}$	% of surface-applied legume N	% of total legume N
Incorporated	herbicide	6.81	0.8	0.63	0.23
	tillage	6.06	0.8	0.45	0.18
Surface	herbicide	5.34	15.5	11.93	4.29
	tillage	4.32	14.5	8.02	3.25
Significance (P) <sup>†</sup>					
Placement		0.01	0.0001	0.0001	0.0001
Method		0.12	0.80	0.13	0.26
M*P		0.80	0.80	0.16	0.30

<sup>†</sup> The df for effects of placement, method and M\*P are 1, 1, and 1, respectively.

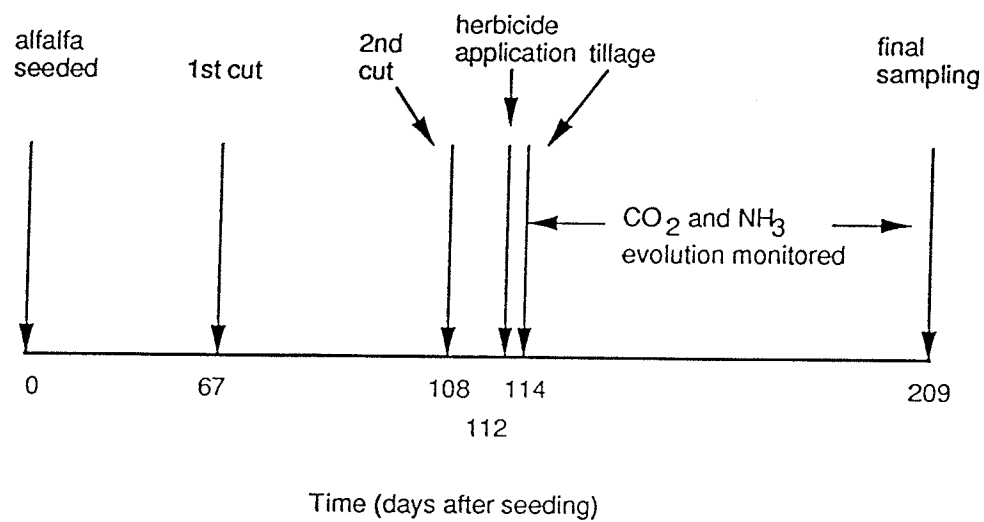


Fig. 4.1. Schedule of establishment of alfalfa, alfalfa termination, and measurement of CO<sub>2</sub> and NH<sub>3</sub> evolution.



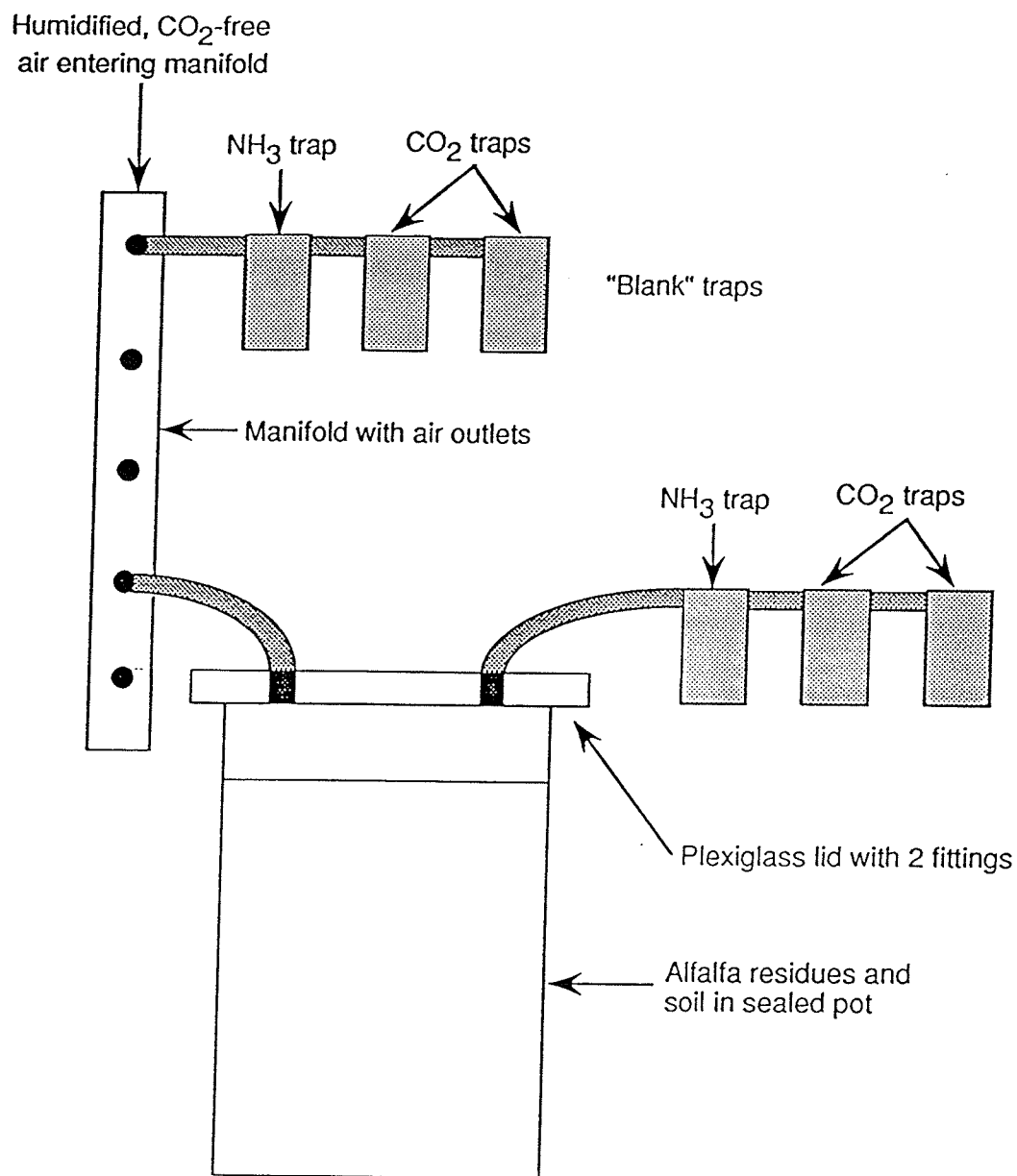


Fig. 4.2. Schematic diagram of the system used to measure  $\text{NH}_3$  and  $\text{CO}_2$  evolution following alfalfa termination.

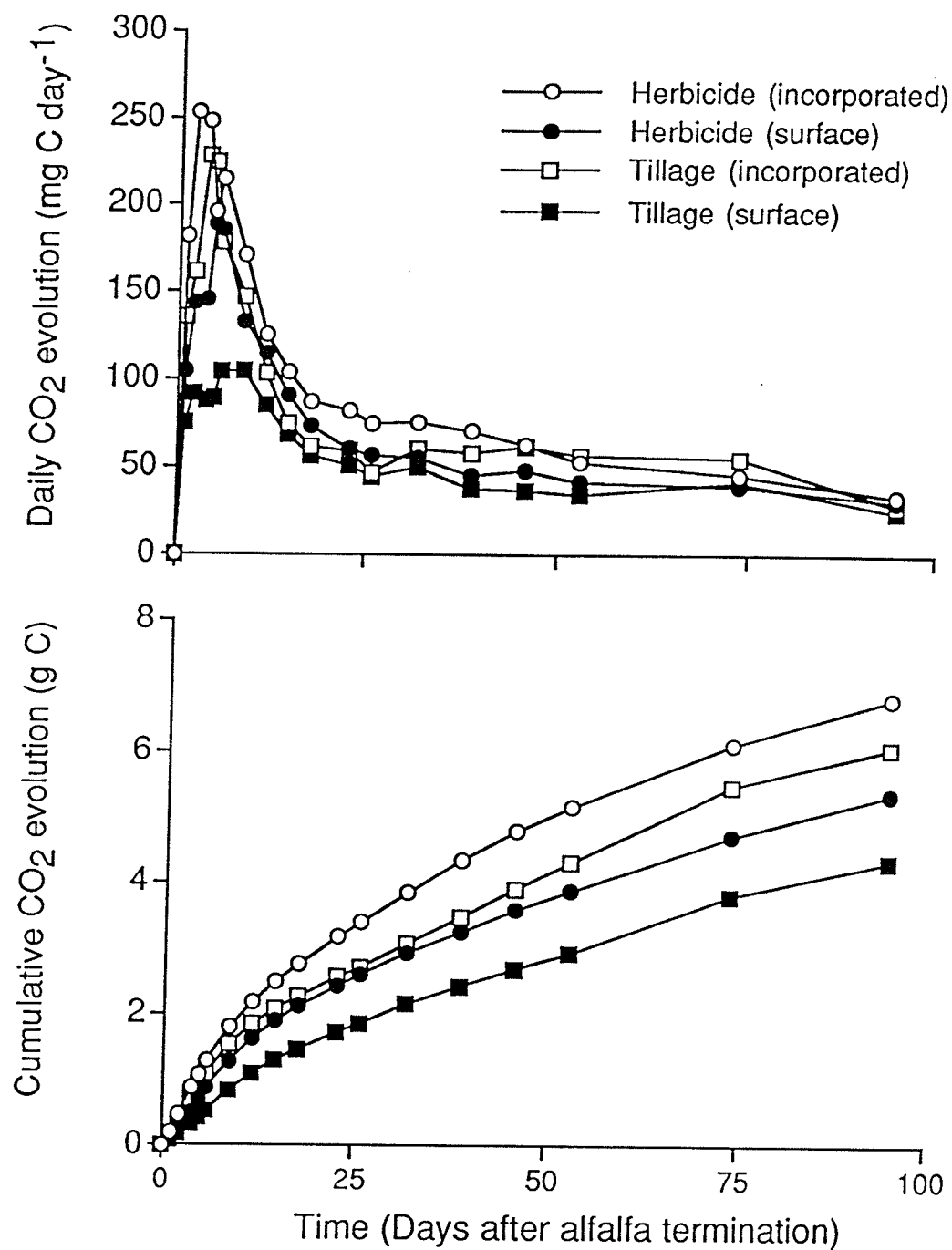


Fig. 4.3. Effect of method of alfalfa termination on daily and cumulative CO<sub>2</sub> evolution.

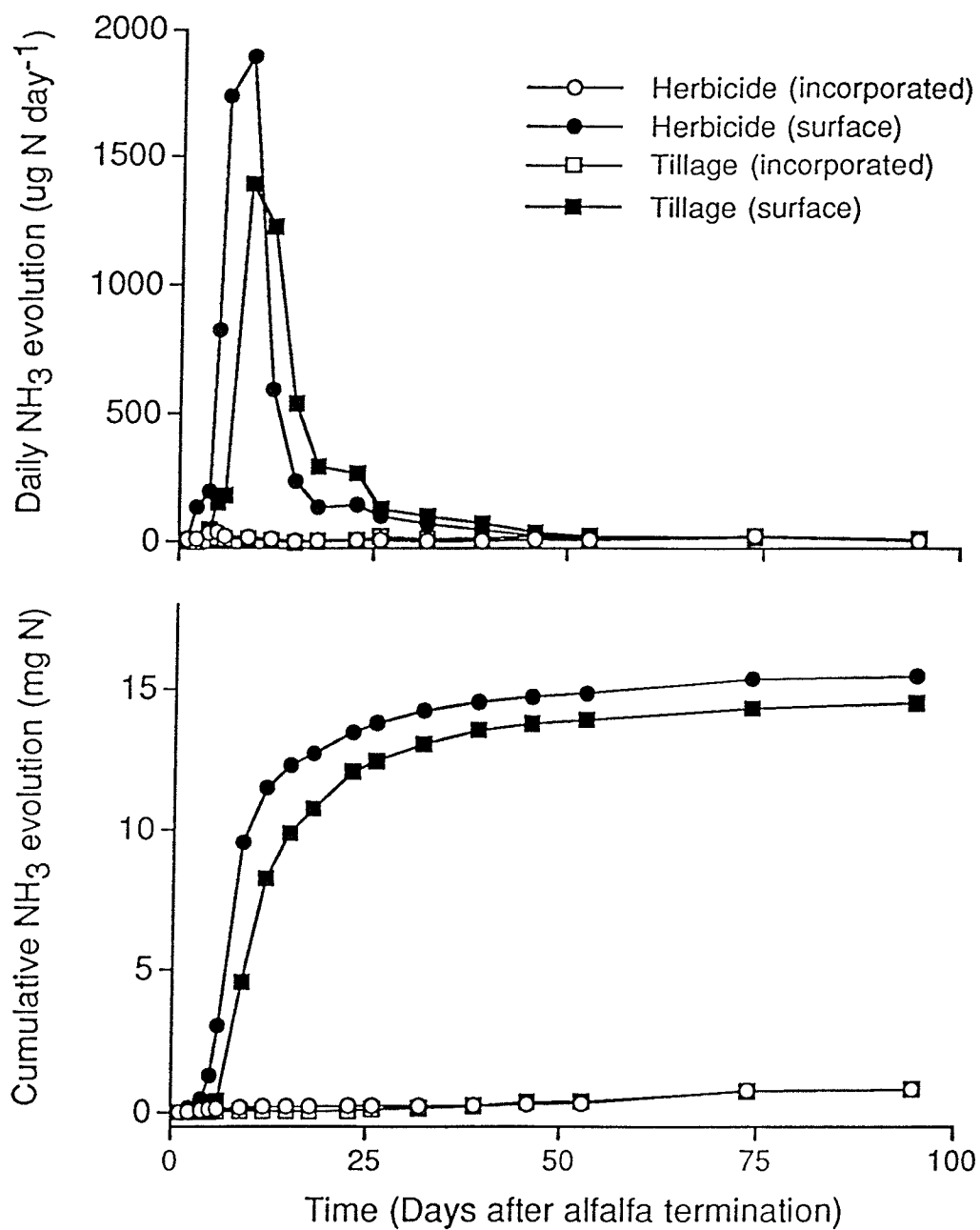


Fig. 4.4. Effect of method of alfalfa termination on daily and cumulative  $\text{NH}_3$  evolution.

## 5. Nitrogen Dynamics as Influenced by Method of Alfalfa Termination: Plant-available N Release

### ABSTRACT

Herbicide application has been proposed as an alternative to tillage for termination of established alfalfa (*Medicago sativa*) stands but may alter the pattern and amount of N released from alfalfa residues. A controlled environment study was conducted to investigate the effect of termination technique on the available N supply following alfalfa termination. Four treatments consisting of a factorial combination of two termination methods (chemical, mechanical) and two methods of residue placement (surface, incorporated) were established. Nitrogen uptake by four consecutive crops of barley (*Hordeum vulgare*) was measured during a 125 d period after termination. Termination method, particularly residue placement, strongly affected N release from alfalfa residues. Nitrogen accumulation by the initial barley crop accounted for >60% of cumulative N uptake in incorporated treatments compared to 39% and 24% for herbicide and tillage treatments in which alfalfa residue was surface applied. Herbicide application also slightly increased N uptake by the initial barley crop. Nitrogen uptake by subsequent barley crops was not affected by termination method; however, cumulative N uptake remained substantially greater for incorporated treatments throughout the 125 d experiment. Effects of residue particle size on N release from alfalfa residues were small. These results suggest that herbicide termination in which residue is retained on the soil surface may reduce the short-term plant-available N supply. Provided mineralization is sufficient to meet the N needs of subsequent crops, maintaining a smaller reservoir of soil

inorganic N may be beneficial in reducing the potential for leaching or denitrification losses.

## INTRODUCTION

Established alfalfa stands are typically terminated by intensive tillage which leaves the exposed soil prone to erosion and moisture loss. One alternative is to apply herbicides and leave residue standing on the soil surface. Surface residue not only protects against soil erosion and moisture loss, but also traps snow which contributes to soil moisture reserves.

Although the use of herbicides to terminate alfalfa has apparent benefits, it could conceivably alter the amount and pattern of N release from alfalfa residue and its use by subsequent crops. Contributions made by alfalfa to the soil N supply have long been recognized (Lyon and Bizzell 1933; Boawn et al. 1963; Hoyt and Hennig 1971; Campbell et al. 1993). However, poor synchrony between N release from legume residues and crops N demands may reduce the fertility benefits of the legume (Huntington et al. 1985; Westermann and Crothers 1993). Moreover, it may result in the accumulation of plant-available N which can be lost by leaching or denitrification losses (Campbell et al. 1994; Robbins and Carter 1980; Firestone 1982).

Tillage has been shown to increase the available N supply from green manure crops as compared to herbicide application (Sarrantonio and Scott 1988). However, in field studies with corn, the fertilizer N response following alfalfa was similar whether the previous alfalfa crop had been terminated by herbicide application or tillage (Triplett et

al. 1979; Levin et al. 1987). Triplett et al. (1979) noted, however, that grain yield may not have been sensitive enough to detect small differences in the amount or rate of N mineralization from alfalfa residues in their study.

Termination method could conceivably affect N release from legumes by several mechanisms. Perhaps foremost among these is the effect on residue placement. Various studies have demonstrated greater amounts of available N from incorporated than from surface-applied legume residues (McCalla and Russel 1948; McKay et al. 1952; Wilson and Hargrove 1986). These differences were generally thought to be the result of enhanced mineralization of incorporated residues although reduced volatile N losses from incorporated residue may also contribute to these differences (Janzen and McGinn 1991; Chapter 4).

Differences in residue particle size and surface area among tillage treatments could also affect N mineralization from alfalfa stands. In laboratory studies, reducing residue particle size by grinding increased net  $^{15}\text{N}$  mineralization of stem, root and pod material of *Medicago littoralis*, but did not affect mineralization of leaf material; however, particle size had little effect on residual  $^{15}\text{N}$  content of *Medicago* sp. decomposing under field conditions (Amato et al. 1984). In contrast, Stickler and Frederick (1959) observed greater net  $\text{NO}_3^-$  accumulations from coarse than from finely ground alfalfa top-growth, but little effect of particle size on net  $\text{NO}_3^-$  accumulations from alfalfa root.

The objectives of this study were to determine the effect of method of alfalfa termination on the timing and amount of N release from alfalfa residues and to identify the mechanisms influencing N release. A controlled environment study was conducted

to measure N uptake by barley during a 125 d period following alfalfa termination. A supplementary incubation experiment was conducted to determine the possible effects of reduced residue particle size in tilled treatments.

## MATERIALS AND METHODS

### Experiment 1: Effect of Termination Method on N Release from Alfalfa

Four treatments, consisting of a factorial combination of two termination methods (chemical, mechanical) and two methods of residue placement (surface, incorporated) were established. Three replications of each of the four treatments were seeded to four consecutive barley crops and barley N uptake was monitored for 125 d following alfalfa termination. An additional six replications of each treatment were incubated, unplanted, alongside the barley crops. Soil  $\text{NO}_3^-$ -N concentration in uncropped pots was determined by destructively sampling three replications at each of 25 d and 125 d after alfalfa termination.

*Alfalfa growth and termination.* Procedures for alfalfa establishment and termination have been described in Chapter 4. In brief, 'Nitro' alfalfa (6 plants  $\text{pot}^{-1}$  each containing 5.43 kg sandy loam soil) was grown in a greenhouse for 112 d. During this period, alfalfa was harvested twice at the bloom stage by cutting top-growth 8 cm above the soil surface. Termination of alfalfa by the treatments described above was initiated 4 d after the second harvest when alfalfa regrowth had reached a height of approximately 12 cm. Alfalfa plants in six additional pots (three untreated and three herbicide-treated) were

destructively sampled at the time of termination to provide an estimate of dry matter yield and N and C concentrations of alfalfa top-growth and root tissue (Table 5.1).

*Barley establishment and growth.* Four consecutive barley crops were grown in pots in which alfalfa had been harvested and the appropriate termination methods had been applied. In all cases, 15 barley seeds (*Hordeum vulgare* cv. 'Galt') were planted in each pot. Pots were watered to field capacity (16.5% w/w) and placed in a growth chamber (16 h day, 22°C day temperature; 8 h night, 15°C night temperature). Shortly after emergence, barley was thinned to eight plants pot<sup>-1</sup>. For the first barley crop, nutrient solutions were surface-applied at the time of seeding to provide 30 mg P kg<sup>-1</sup>, 62 mg K kg<sup>-1</sup> and 10 mg S kg<sup>-1</sup> soil.

In general, barley crops were harvested 25 d after planting by cutting the top-growth at the soil surface; however, the third barley crop was harvested only 10 d after planting due to poor plant growth and crop senescence subsequently attributed to severe N deficiency. Barley top-growth was oven dried, dry matter yield determined and total N concentration determined by an automated combustion technique (Carlo Erba<sup>TM</sup>, Milan, Italy).

The first barley crop was seeded immediately after alfalfa termination and the second barley crop immediately after harvest of the first. Establishment of the third and fourth crops was delayed for 5 d and 35 d after harvest of the preceding crops.

*Incubation of uncropped pots.* Six replicates of each treatment were left unplanted and



incubated in the growth cabinet alongside barley plants. Pots were weighed periodically to determine the soil moisture content and distilled water was added as required throughout the incubation period to maintain soil moisture near field capacity (16.5% w/w). Three replicates were destructively sampled 25 d after alfalfa termination, the remaining three replicates were destructively sampled 125 d after alfalfa termination. Soil within each pot was mixed thoroughly and a subsample removed and air dried. Soil inorganic N was extracted with 2 M KCl. The concentration of  $\text{NO}_3^-$  and  $\text{NH}_4^+$  in the extract was determined by a colorimetric procedure using an autoanalyzer (Keeney and Nelson 1982).

*Statistical analysis.* N uptake was analyzed by two way analysis of variance for a completely randomized design using the Proc GLM procedure (SAS Institute Inc. 1985).

#### Experiment 2: Effect of Particle Size on N Release from Alfalfa Leaf, Stem and Root Tissue

An incubation experiment was conducted to determine the effect of residue particle size on N mineralization from soil-incorporated alfalfa leaf, stem and root tissue. Eight treatments, consisting of leaf (ground or whole), stem (ground, 1.5 cm and 5 cm lengths) and root (ground, 1.5 cm and 5 cm lengths) tissue, were established in addition to a control (soil only). Three replications of each treatment were incubated at 25°C for 12 wk and accumulations of soil  $\text{NO}_3^-$  and  $\text{NH}_4^+$  were measured periodically.

Alfalfa root and shoot material was collected in late fall from a field plot at the Lethbridge Research Centre. Alfalfa plants were separated by hand into leaf, stem and

root tissue. Stems of uniform diameter (1-2 mm) and roots of uniform diameter (2-4 mm) were cut into 5 cm lengths. Leaf, stem and root tissue was then oven dried.

Approximately one-half of the oven-dried leaves were ground using a Wiley mill and the remaining leaves were left whole. Stem and root tissue was subsampled and either left in 5 cm lengths, cut by hand into ~1.5 cm lengths or ground using a Wiley mill. Total N and C content of ground leaf, stem and root tissue was determined by an automated combustion technique (Carlo Erba<sup>TM</sup>, Milan, Italy) (Table 5.2).

The A horizon of a Cavendish sandy loam soil (pH in  $\text{CaCl}_2=6.9$ ; total N=0.74 g  $\text{kg}^{-1}$ ; organic C=7.6 g  $\text{kg}^{-1}$ ) was air dried and passed through a 2 mm sieve. The equivalent of 1 kg soil (oven dry basis) was added to each pot and moistened to 50% of field capacity. For each treatment (except the control), oven dry plant tissue at a rate of 3 g  $\text{pot}^{-1}$  was thoroughly mixed into the soil. [This application of 3 g residue  $\text{kg}^{-1}$  soil would be equivalent to approximately 4000 kg residue  $\text{ha}^{-1}$  (to 10 cm) assuming a bulk density of approximately 1.3  $\text{Mg m}^{-3}$ . In the field study (Chapter 3), it was estimated that alfalfa top-growth present at time of termination averaged about 2000 kg dry matter  $\text{ha}^{-1}$ ; root residues present at time of termination were not quantified.] Soil was then moistened to field capacity (16.5% w/w) and covered with a plastic sheet to prevent moisture loss. Small holes were made in the plastic to provide aeration.

Pots were incubated in the dark at 25°C for a 12 wk period. Every 1 to 2 d, pots were aerated, weighed to determine the soil moisture content, and distilled water added as required to maintain the soil moisture content near field capacity (generally between 85% and 100% of field capacity). At 2, 5, 9 and 12 weeks the soil in each pot was

mixed thoroughly and a 20 g subsample removed and air dried. Pots were then returned to 25°C for the remainder of the incubation period. Air dried soil was shaken with 2 M KCl for 1 h and the concentration of  $\text{NO}_3^-$  and  $\text{NH}_4^+$  in the extract determined by a colorimetric procedure using an autoanalyzer (Keeney and Nelson 1982).

This experiment was analyzed by one way analysis of variance for a completely randomized design using the Proc GLM procedure (SAS Institute Inc. 1985). The effects of plant part and residue particle size were further analyzed by single degree of freedom contrasts.

## RESULTS

### Effect of Termination Method on N Release from Alfalfa

Incorporation of alfalfa residue increased N uptake for the first barley crop established after alfalfa termination, regardless of termination method (Fig 5.1). In incorporated treatments, the majority (>60%) of N uptake occurred within 25 d of alfalfa termination; surface application of alfalfa residues reduced the proportion of N accumulated by the first barley crop to 39% and 24% of total N uptake in herbicide and tillage treatments, respectively. No differences among treatments were observed in subsequent barley crops (Table C.1 in Appendix C). Herbicide application also increased N uptake by the first barley crop established after alfalfa termination (Fig. 5.1), but N uptake by subsequent crops was not affected by termination method (Table C.1 in Appendix C).

Although the effect of incorporation on N uptake diminished with each barley crop

grown, cumulative N uptake in incorporated treatments remained higher than in unincorporated treatments throughout the 125 d experiment (Fig. 5.1). In incorporated treatments, total N uptake after 125 d averaged 104 mg N pot<sup>-1</sup>, almost double that of surface residue treatments, which averaged 57 mg N pot<sup>-1</sup>. Although termination method did not affect cumulative N uptake beyond 25 d after alfalfa termination, cumulative N uptake as a proportion of alfalfa N present was also higher ( $P < 0.05$ ) for herbicide (22.9%) than for tillage (17.5%) treatments at the conclusion of the 125 d experiment.

The N content of barley top-growth harvested 25 d after alfalfa termination was closely correlated ( $r = 0.969$ ,  $P = 0.0001$ ) with the accumulation of NO<sub>3</sub><sup>-</sup>-N in uncropped soil incubated for the same period. Incorporation of alfalfa top-growth increased the NO<sub>3</sub><sup>-</sup>-N concentration in uncropped soil regardless of termination method (Table 5.3); although herbicide application had increased N uptake by the first barley crop, termination method had no effect on the NO<sub>3</sub><sup>-</sup> concentration in uncropped soil incubated for 25 d.

Analysis of uncropped soil incubated for 125 d showed very low NO<sub>3</sub><sup>-</sup> concentrations presumably because of N loss by denitrification (Table C.2 in Appendix C). But, in a similar study (Chapter 4), in uncropped soil containing terminated alfalfa which was incubated for 95 d, soil NO<sub>3</sub><sup>-</sup>-N accumulations were 51.3 and 37.8 mg NO<sub>3</sub><sup>-</sup>-N kg<sup>-1</sup> for incorporated and unincorporated treatments, respectively ( $P = 0.03$ ). Again, termination method had no effect on soil NO<sub>3</sub><sup>-</sup> concentration.

#### Effect of Particle Size on N Release from Leaf, Stem and Root Tissue

Nitrogen in leaf tissue was rapidly mineralized, resulting in net N accumulations

equivalent to 50% of leaf N within 2 wk; only small amounts of additional N were mineralized after 2 wk (Fig. 5.2; Table 5.4). Nitrogen mineralization from stem and root tissue progressed at a comparatively slower rate; after 2 wk, 11% of stem N and 2% of root N had been mineralized. However, unlike leaf tissue, significant amounts of N continued to be mineralized from root and stem tissue throughout the 12 wk incubation.

Plant part affected not only the pattern of N release, but also the quantity and proportion of residue N mineralized (Table 5.4). Net mineralization from 3 g leaf tissue totalled 81 mg N pot<sup>-1</sup> after 12 wk; net mineralization from equal masses of root or stem tissue averaged 22 mg N pot<sup>-1</sup>. The higher N concentration in leaf tissue contributed to this difference as did more extensive mineralization from leaf tissue. After 12 wk, 51% of leaf N had been mineralized but only 34% of stem N and 27% of root N. (Net N mineralized and the % of alfalfa N mineralized after 5 and 8 wk of incubation is reported in Table C.3 in Appendix C.) The pattern of N release from root and stem tissue suggests that the proportion of N mineralized from stem and root tissue may have been higher if the incubation period had been longer. Based on the pattern of N release from the various plant parts, it appears that mineralization of leaf N has a stronger influence on short-term fertility whereas mineralization of stem and root tissue has a greater long-term effect.

Overall, residue particle size had little influence on N mineralization from alfalfa residue (Table 5.4). Particle size did not have a significant effect on the amount of N mineralized from any of the plant parts, although grinding tended ( $P=0.07$ ) to decrease the amount of N released from stem tissue after 2 wk. In contrast, grinding tended

( $P=0.06$ ) to increase the amount of N released from leaf tissue after 12 wk. Grinding had no effect on the proportion of N mineralized from leaf or root tissue, but decreased the proportion of N mineralized from stem tissue after 2 wk. Although this trend was apparent throughout the 12 wk incubation, the effect was not statistically significant.

## DISCUSSION

### Mechanisms

More rapid release of greater amounts of N from incorporated alfalfa residue presumably results from greater exposure of alfalfa residue, particularly decomposable leaf tissue, to microbial populations. In a previous study by Cogle et al. (1987), greater initial decomposition of soil-incorporated than of surface-applied wheat straw was similarly attributed to greater positional availability of residues to microbial populations. Reduced volatile N losses from incorporated residues may also contribute slightly to the increased plant-available N supply observed. In a previous study (Chapter 4), the equivalent of 3 to 4% of total legume N was lost from surface-applied alfalfa residues by 95 d after alfalfa termination.

Greater initial N uptake by barley in herbicide treatments suggests increased availability of labile N from alfalfa residues. This may be due, in part, to more rapid senescence and subsequent decomposition of herbicide-treated alfalfa. In a previous study (Chapter 4), greater cumulative  $\text{CO}_2$  evolution in herbicide treatments was speculated to be the result of increased root exudation following herbicide application.

The physical breakdown of residues by incorporation was not an important factor

influencing N mineralization. In the current study, residue particle size had minimal effects on N release. Amato et al. (1984) also found that particle size had little effect on N mineralization from *Medicago* sp. decomposing under field conditions. In contrast, several laboratory studies have demonstrated that reductions in residue particle size may enhance residue decomposition (Sims and Frederick 1970; Amato et al. 1984).

Plant part appeared to have a stronger influence on N release patterns than residue particle size; N release from leaf tissue was more rapid and greater than from stem or root tissue. Amato et al. (1984) similarly observed greatest mineralization from leaf material. In field experiments, leaf, stem and root material of medic contained an average of 64%, 87% and 81% of initial organic  $^{15}\text{N}$  4 wk after soil incorporation (Amato et al. 1984). After 2 years of decomposition, 40%, 56% and 50% of  $^{15}\text{N}$  present in leaf, stem and root tissue remained. These authors reported similar results under laboratory conditions. The observed differences in residue decomposability are presumably a function of residue composition including factors such as the C/N ratio, lignin and carbohydrate content, total N content and soluble and intermediately-available C contents of residues (Iritani and Arnold 1960; Nyhan 1975; Herman et al. 1977; Reinertsen et al. 1984; Janzen and Kucey 1988).

### Implications

These findings indicate that incorporation accelerates the release of N from alfalfa residues and significantly increases the short-term plant-available N supply.

The proposed method of alfalfa termination would involve applying herbicide either the summer prior to or the spring immediately prior to establishment of an annual crop and leaving alfalfa residue standing on the soil surface. Provided N mineralization under this system were sufficient to meet the N requirements of a subsequent crop, reduced mineralization resulting from surface application of residues may have some advantage. Although surface residue may be subject to small N losses via volatilization (Chapter 4), the potential for substantial N losses via leaching or denitrification may be reduced if accumulations of excess amounts of inorganic N are avoided. Lower N losses may also contribute to more efficient use of alfalfa-derived N by subsequent crops. However, the mineralization of smaller amounts of available N or delayed release of N in unincorporated treatments may be detrimental if N requirements of a subsequent crop are not met and lower grain yield or grain quality results. In field studies, Huntington et al. (1985) reported poor synchrony between N release from a spring-terminated winter annual cover crop and N uptake by a subsequent corn crop. Similarly, Westermann and Crothers (1993) found that N mineralization from fall-terminated alfalfa was insufficient to meet the N requirements of a subsequent soft winter wheat crop. These authors emphasized that crops should be selected such that sufficient N is mineralized from alfalfa residue prior to crop needs and suggested that, in the case of their study, synchrony between N release and N uptake might be improved by selecting crops with later-season N uptake (eg. sugarbeet, potato, corn).

Results of this study demonstrate that termination technique, particularly residue placement, affects both the pattern and amount of N release from alfalfa residues and



could conceivably affect the efficiency with which alfalfa-derived N is used by subsequent crops. Under field conditions, however, additional factors such as the duration between alfalfa termination and establishment of a subsequent crop, N requirements of a subsequent crop, the amount of alfalfa N present, and environmental conditions, may act in combination with termination technique to influence the short-term plant-available N supply.

Table 5.1. Composition of alfalfa residue present in herbicide treatments 2 d after glyphosate application and in tillage treatments at time of tillage.

		Termination method		P>F
		Herbicide	Tillage	
Top-growth	%C	43.9	44.2	0.55
	%N	3.3	3.7	0.02
	N yield (mg)	130	181	0.003
	Dry mass (g)	3.93	4.95	0.008
Root	%C	44.6	43.6	0.002
	%N	3.7	2.7	0.006
	N yield (mg)	231	266	0.17
	Dry mass (g)	6.18	9.96	0.0008

Table 5.2. Composition of leaf, stem and root tissue from field-grown alfalfa. Residues were subsequently applied to moistened soil and incubated at 25°C for a 12 wk period during which N mineralization was monitored.

Plant part	mg C g <sup>-1</sup>	mg N g <sup>-1</sup>	C:N
Leaf	438	52.8	8.3
Stem	448	22.7	19.7
Root	446	25.1	17.8

Table 5.3. Effect of termination method (herbicide application, tillage) and residue placement (soil-incorporated, surface-applied) on soil  $\text{NO}_3^-$  accumulations 25 d after alfalfa termination.

Treatment		Soil $\text{NO}_3^-$ -N concentration
Method	Placement	
		--mg $\text{NO}_3^-$ -N $\text{kg}^{-1}$ --
Herbicide	incorporated	18.2
	surface	5.7
Tillage	incorporated	17.3
	surface	5.0
Significance (P) <sup>†</sup>		
Method		0.57
Placement		0.0001
M*P		0.91

<sup>†</sup> The df for effects of method, placement and M\*P are 1, 1, and 1, respectively.

Table 5.4. Effect of residue particle size on net N mineralization of alfalfa leaf, stem and root tissue.

Treatment		Net mineralization <sup>†</sup>		% Recovery <sup>‡</sup>	
Residue	Size	Wk 2	Wk 12	Wk 2	Wk 12
-mg inorganic N pot <sup>-1</sup> -					
Leaf	ground	80.2	84.7	50.6	53.5
	whole	77.9	77.1	49.2	48.7
Stem	ground	5.4	20.8	8.0	30.5
	1.5 cm	10.3	26.9	15.1	39.5
	5 cm	7.7	22.5	11.3	33.0
Root	ground	1.6	22.2	2.1	29.4
	1.5 cm	1.3	17.9	1.8	23.7
	5 cm	2.5	21.4	3.3	28.4
Significance (P) <sup>§</sup>					
Treatment		0.0001	0.0001	0.0001	0.0001
Contrasts					
Leaf 'ground vs whole'		0.29	0.06	0.41	0.30
Stem 'ground vs unground'		0.07	0.23	0.003	0.15
Root 'ground vs unground'		0.87	0.44	0.78	0.39

<sup>†</sup> Net mineralization=[Soil NO<sub>3</sub><sup>-</sup>+NH<sub>4</sub><sup>+</sup>]<sub>sample at Week X</sub>-[Soil NO<sub>3</sub><sup>-</sup>+NH<sub>4</sub><sup>+</sup>]<sub>soil control at Week X</sub>  
in mg N pot<sup>-1</sup>

<sup>‡</sup> % recovery=[Net mineralization (mg N pot<sup>-1</sup>)/Alfalfa N added (mg N pot<sup>-1</sup>)]\*100

<sup>§</sup> The df for the effect of treatment was 7. All contrasts were single df contrasts.

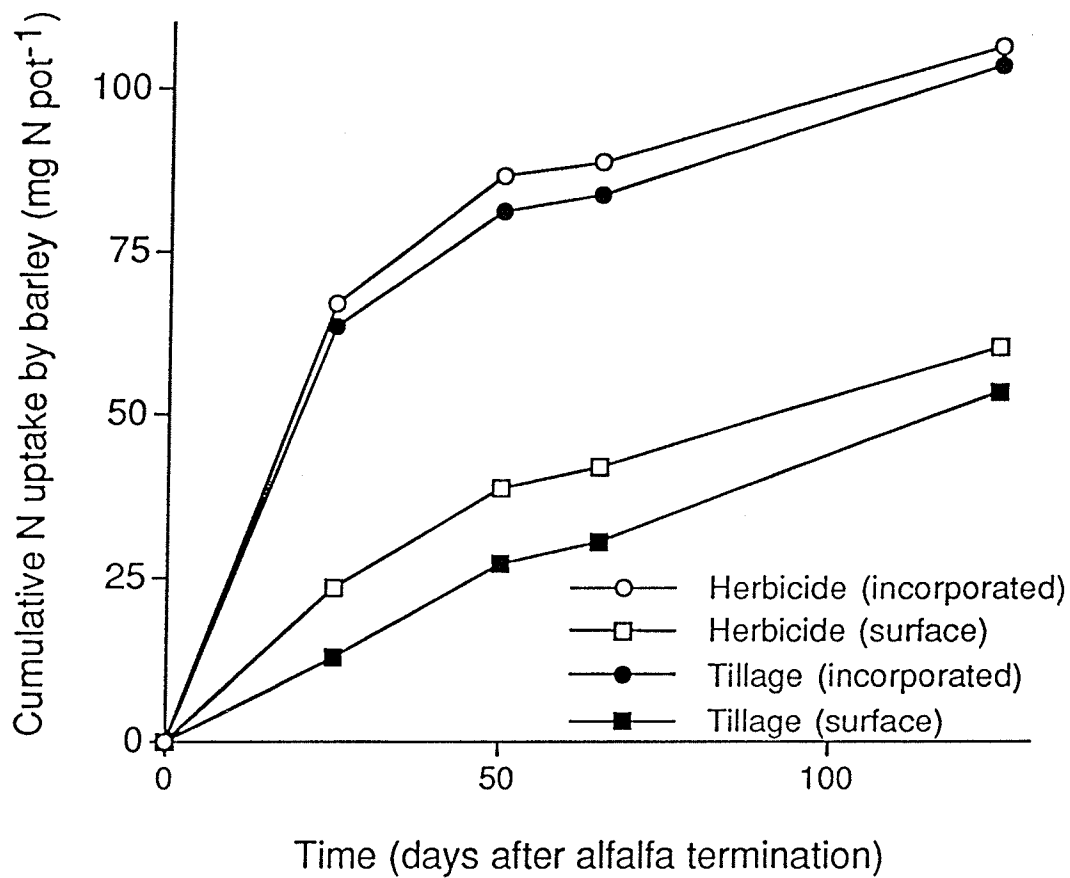


Fig. 5.1. Effect of method of alfalfa termination on cumulative N uptake by barley. For cumulative N uptake after 25 d, the statistical significance (P-value) of method (M), placement (P) and M\*P was 0.02, 0.0001 and 0.20, respectively. For cumulative N uptake after 125 d, the statistical significance (P-value) of method (M), placement (P) and M\*P was 0.60, 0.0006 and 0.70, respectively (Table C.1 in Appendix C).

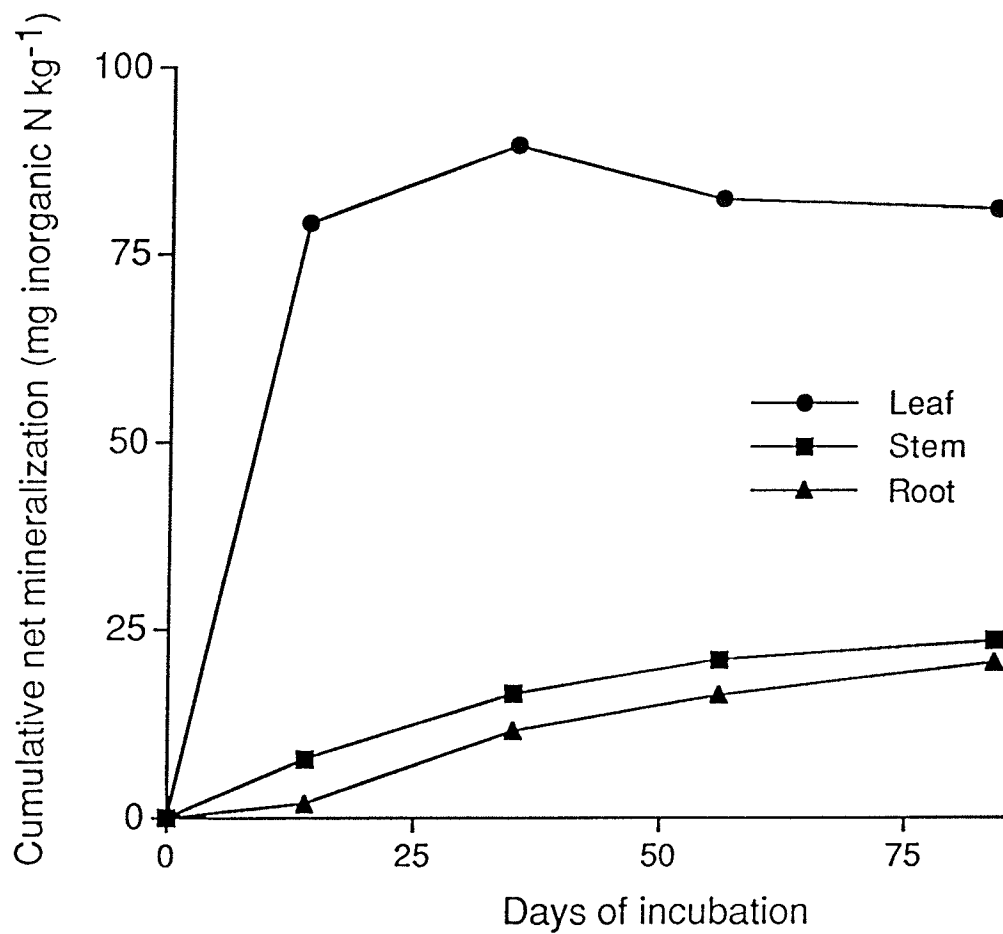


Fig. 5.2. Cumulative net N mineralization of alfalfa leaf, stem and root tissue (calculated as mean of all particle sizes).

## 6. The Fate of Symbiotically Fixed $^{15}\text{N}_2$ as Influenced by Method of Alfalfa Termination

### ABSTRACT

Alfalfa (*Medicago sativa*) may contribute appreciable amounts of fixed N to subsequent crops, but this may depend on method of crop termination. In a controlled environment study, alfalfa was labelled by continuous, prolonged exposure to a  $^{15}\text{N}_2$ -containing soil atmosphere to allow direct measurement of the fate of fixed  $\text{N}_2$  from growing and terminated alfalfa. The distribution of  $^{15}\text{N}$  in plant and soil components was measured 74 d after alfalfa establishment at the time of alfalfa termination and again 33 d later following the growth of a 4 wk crop of barley (*Hordeum vulgare*). At time of termination, 88% of the  $^{15}\text{N}$  present was in alfalfa and 12% in soil (5% in root fragments, 5% in microbial biomass and 2% in root and microbial products). The distribution of  $^{15}\text{N}$  after alfalfa termination was primarily a function of residue placement rather than termination method. In herbicide treatments in which alfalfa top-growth was retained on the soil surface, 1% of the  $^{15}\text{N}$  present was recovered in barley top-growth, 8% in soil and 91% in residues; in tillage treatments in which alfalfa top-growth was incorporated, 10% of the  $^{15}\text{N}$  present was recovered in barley top-growth, 52% in soil and 38% in residues. Regardless of termination method, 10% of the  $^{15}\text{N}$  present in alfalfa roots was recovered in barley top-growth, 30% in soil and 60% in residues. Although tillage and herbicide application result in a similar degree of N release from alfalfa root tissue, termination of alfalfa by tillage greatly accelerates mineralization of alfalfa top-growth resulting in a larger short-term supply of plant-available N than herbicide application.

## INTRODUCTION

Legumes release N into the soil during growth and senescence. This N release occurs by exudation from living roots and nodules, decomposition of roots, nodules and shoot residues, and leaching from living plants (Lory et al. 1992; Dubach and Russelle 1994; Tomm et al. 1995). Under growing alfalfa (*Medicago sativa* L.) stands, soil N accumulations of 21 to 56 kg N ha<sup>-1</sup> yr<sup>-1</sup> have been estimated (Lyon and Bizzell 1934; Andrén et al. 1990). In fact, these values may somewhat overestimate the net accumulation of N under alfalfa stands since they include not only symbiotically fixed N but also soil N absorbed by alfalfa. In intercropped alfalfa/non-legume systems N transfers equivalent to 5 to 20 kg N ha<sup>-1</sup> yr<sup>-1</sup> have been reported (Ta and Faris 1987; Burity et al. 1989).

Substantial amounts of N may also be released from the residues of terminated legumes. Most legume-derived N is retained in the soil in organic forms which are only slowly made available to subsequent crops. Field studies have demonstrated recoveries of 11 to 28% of N present in soil-applied legume residue by the initial wheat (*Triticum aestivum*) crop established after residue additions (Ladd et al. 1981; Ladd et al. 1983; Janzen et al. 1990); N recoveries by a subsequent crop declined to <5% of applied legume N (Ladd et al. 1983; Janzen et al. 1990).

One factor which may influence the distribution of legume-derived N in the plant/soil system and its availability to subsequent crops is method of legume termination. Currently, herbicide application is being suggested as an alternative to tillage for the termination of established alfalfa stands, but, few studies have directly compared the fate



of legume N under different termination methods.

Much of the information regarding N release from legume residues is based on studies involving the fate of N from  $^{15}\text{N}$ -labelled residues applied to the soil. Although this technique allows direct measurement of N release from residues, it does not allow quantification of N release from root systems under undisturbed conditions. Also, N contributions made by legumes to subsequent crops may be underestimated if N lost from legumes during growth is not included (Heichel 1987).

Direct quantification of the fate of legume-derived N requires that plants be labelled *in situ*. In a recent study, alfalfa in an undisturbed plant-soil system was pulse labelled by repeatedly applying  $^{15}\text{N}$ -labelled urea directly onto the foliage of non-fixing alfalfa plants (Jordan et al. 1993). Suppression treatments were then applied and differences in the distribution of alfalfa-derived N were directly measured. Although this technique effectively labelled alfalfa tissue, the means by which  $^{15}\text{N}$  was assimilated into the plant may not reflect that of a  $\text{N}_2$ -fixing plant under natural conditions. Also, only the N absorbed at the time of urea application was labelled. Consequently, the  $^{15}\text{N}$  present in the alfalfa may not simulate N assimilated by conventional means.

An alternative technique for labelling plants *in situ* is to expose the legume root system to  $^{15}\text{N}$ -labelled  $\text{N}_2$ . Because  $^{15}\text{N}$  is incorporated into the plant by biological  $\text{N}_2$  fixation, assimilated  $^{15}\text{N}$  is a true tracer of symbiotically fixed N (Warembourg 1993). This technique has been used previously to measure N transfers from legumes to intercropped non-legumes (Ta et al. 1989; McNeill and Wood 1990) and to measure the loss of symbiotically-fixed N into the rhizosphere of alfalfa (Russelle et al. 1994).

Exposure times ranging from several hours (Ta et al. 1989) to several days (Ruschel et al. 1979; McNeill and Wood 1990; Russelle et al. 1994) have been reported but continuous, prolonged exposure is uncommon (Bremer et al. 1995). In general, previous studies have used a relatively short labelling period; hence, only that N assimilated during the brief exposure period is labelled. Continuous, prolonged exposure of plants to  $^{15}\text{N}$ -labelled  $\text{N}_2$  would allow labelling of a greater proportion of symbiotically fixed N which may be more representative of N fixed throughout alfalfa growth.

The objectives of this study were: 1) to determine the distribution of symbiotically fixed  $^{15}\text{N}$  in the plant/soil system following growth of an alfalfa crop and 2) to determine the effect of method of alfalfa termination on the fate and plant-availability of symbiotically fixed  $^{15}\text{N}$ . Our approach involved labelling alfalfa by continuous, prolonged exposure to a soil atmosphere containing  $^{15}\text{N}$ -labelled  $\text{N}_2$ . Before and after termination of alfalfa, the distribution in the plant-soil system of symbiotically-fixed  $^{15}\text{N}$ -labelled  $\text{N}_2$  was measured. Potential mineralization of  $^{15}\text{N}$ -labelled alfalfa leaf, stem, crown, fine root and taproot tissues was also measured.

## MATERIALS AND METHODS

### Alfalfa establishment, growth and labelling

Alfalfa was established in cylinders (10 cm diameter x 32 cm ABS pipe attached with silicone to a base of ABS sheeting or plexiglass) each containing the equivalent of 3188 g (oven dry basis) of a Cavendish sandy loam soil (pH in  $\text{CaCl}_2=6.9$ ; organic C=7.6 g  $\text{kg}^{-1}$ ). (This soil had been collected from near Purple Springs, AB and stored in a

stockpile at the Lethbridge Research Centre prior to its use.) Air dry soil was moistened to 50% of field capacity and nutrient solutions thoroughly mixed into the soil to provide the following (in mg kg<sup>-1</sup> soil): 6.6 N, 40 P, 50.5 K, 10 S, 1 Cu, 2 Mn, 4 Zn, 4 Fe, 1 B and 0.40 Mo.

Three 'Nitro' alfalfa seeds were placed just below the soil surface in each cylinder. One to two drops of a slurry of *Rhizobium*-peat (commercial inoculant Nitragin, Liphatech, Madison, WI) in water were applied to each seed by pipette and the soil surface was covered with moist paper towels. Several cylinders were left unplanted as controls. Cylinders were watered to field capacity (16.5% w/w) and placed in a growth chamber (16 h day, 22°C; 8 h night, 15°C). Alfalfa was thinned to 1 plant per cylinder 1 wk after emergence.

One to two days prior to labelling the soil atmosphere, selected cylinders were capped with ABS sheeting or painted plexiglass. Each lid had four holes equipped with brass or plastic fittings to allow for watering, aeration and emergence of the alfalfa plant (Fig. 6.1). One brass fitting was fitted with a subaseal to allow the addition of water by syringe. A plastic tube was placed around the alfalfa plant, and the area surrounding the alfalfa stem was sealed with a layer of medical grade elastomer (Dow Corning Corporation, Midland, Michigan) and a layer of lanolin-wax. (Additional layers of sealant were applied during the study to maintain an airtight seal around the stem.) Two brass fittings in each lid were connected to Nalgene tubing (1.25 cm diameter) to allow air exchange.

Each cylinder was connected by Nalgene tubes to a central airtight chamber. The

chamber, constructed from ABS pipe (10 cm diameter) and sheeting, contained an electric fan to continuously circulate air throughout the system. The central chamber also contained a soda lime trap to collect CO<sub>2</sub> and a Cryovac gas bag (Grace Chemicals, Mississauga, ON) to allow volume changes within the system (Fig. 6.1). O-ring seals were installed in all joints to produce an airtight system. Two identical <sup>15</sup>N labelling systems (System 1 which consisted of 6 cylinders attached to one central chamber, System 2 which consisted of 5 cylinders attached to one central chamber) were established and operated as described by Bremer et al. (1995).

Prior to the addition of <sup>15</sup>N<sub>2</sub>, positive pressure was applied to the cylinders and central chambers to ensure all were airtight. Cylinders were then purged with a combination of He and O<sub>2</sub> to reduce the concentration of N<sub>2</sub> in the atmosphere of the labelling system to approximately 50% (Based on periodic analyses of the atmosphere by gas chromatography, the concentration of N<sub>2</sub> in the labelling system increased to >80% during the first few weeks of labelling, presumably due to small leaks in the system and the addition of <sup>15</sup>N<sub>2</sub>.)

For a 7 wk period beginning 25 d after seeding, the soil atmosphere of 11 sealed cylinders (9 alfalfa, 2 unplanted) was continuously exposed to air containing <sup>15</sup>N<sub>2</sub> (Fig. 6.2). Twice weekly, gas samples were collected from the central chamber and analyzed for <sup>15</sup>N by mass spectrometry (VG Optima, VG Isogas Limited, Middlewich, England) using the mass 29/28 and 30/28 ratios to determine <sup>15</sup>N abundance. Purified, highly enriched <sup>15</sup>N-labelled N<sub>2</sub> (>99 atom % <sup>15</sup>N; Isotech Inc., Miamisburg, OH) was usually added twice each week. The <sup>15</sup>N enrichment of the soil atmosphere was usually

maintained between 4 and 6 atom %, although approximately 2 wk after the start of labelling,  $^{15}\text{N}$  enrichment in both labelling systems dropped briefly below this range due to air leaks (Fig. 6.3). When leaks were identified, System 1 was dismantled, repaired and reassembled; in System 2, seals around plant stems were repaired or removed and replaced. After repairs were made,  $^{15}\text{N}_2$  was added to both systems and the  $^{15}\text{N}$  concentration returned to >5 atom %.

Additional gas samples were collected from the central chamber every 1-2 d and analyzed for  $\text{O}_2$  and  $\text{N}_2$  by gas chromatography. Because the concentration of  $\text{O}_2$  tended to fluctuate, pure  $\text{O}_2$  was added once or twice daily to maintain the  $\text{O}_2$  concentration near that of atmosphere (Fig. 6.3). Average  $\text{O}_2$  concentrations ranged from 16 to 23%.

Alfalfa was watered every 1-2 d by injection of water through a subseal in the lid of each cylinder. The amount of water required was estimated from the size of labelled alfalfa plants and water usage by unlabelled alfalfa grown in capped cylinders placed in the growth cabinet alongside the plants being labelled.

Alfalfa was harvested 42 d and 64 d after seeding by cutting the top-growth approximately 9.5 cm above the soil surface. Termination treatments were applied 10 d after the second alfalfa harvest (Fig. 6.2).

#### Measurement of the distribution of $^{15}\text{N}_2$ fixed by alfalfa

Ten days after the second alfalfa harvest (at the time of termination), three labelled and three unlabelled alfalfa plants were sampled. Top-growth was removed at the soil surface and separated into leaf, stem and crown tissue. (Crown tissue was defined as

"old" growth including lower stems remaining from previous harvests.)

Fine roots and taproots were manually removed from the soil with forceps. Soil loosely adhering to roots was removed by sieving (2 mm sieve) and air dried. Root samples were then rinsed several times with distilled, deionized water and sonified for two 60 s periods to remove any remaining soil. The resulting water/soil mixture was acidified with several drops of concentrated  $\text{H}_2\text{SO}_4$  to prevent  $\text{NH}_3$  loss and dried at  $70^\circ\text{C}$ . This procedure generated three fractions: 1. loosely-adhering rhizosphere soil, 2. closely associated rhizosphere soil and 3. bulk soil.

One subsample of bulk soil was air dried and analyzed for total N,  $^{15}\text{N}$ , inorganic N, and light fraction N and  $^{15}\text{N}$ . A second subsample of the bulk soil was stored moist at  $0.5^\circ\text{C}$  and analyzed for microbial biomass N and  $^{15}\text{N}$  and mineralizable C, N and  $^{15}\text{N}$ . Despite attempts at careful removal, small amounts of root tissue remained in the bulk soil. To determine the  $^{15}\text{N}$  and N contributions made by this root tissue to the bulk soil, root fragments were carefully removed from a 10 g subsample of the bulk soil. This 'root-free' soil was analyzed for N and  $^{15}\text{N}$  content.

The total N and  $^{15}\text{N}$  concentration in alfalfa tissue, rhizosphere soil, root-free soil, bulk soil and light fraction organic matter was determined using an automated combustion technique (Carlo Erba<sup>TM</sup>, Milan, Italy) and a continuous flow mass spectrometer (VG Optima, VG Isogas Limited, Middlewich, England) standardized against a reference gas (0.36623 atom %). Soil inorganic N was extracted with 2 M KCl and the concentration of  $\text{NO}_3^-$  and  $\text{NH}_4^+$  in the extract was determined by a colorimetric procedure using an autoanalyzer (Keeney and Nelson 1982).

Microbial biomass N was determined on moist samples of the bulk soil by a chloroform fumigation extraction procedure (Brookes et al. 1985). After extraction, a 40 mL aliquot of each sample was acidified, evaporated and digested with sulfuric acid and hydrogen peroxide (Wolf 1982). Nitrogen in the digest was quantified by steam distillation, collection in boric acid and titration with  $\text{H}_2\text{SO}_4$ . Following titration, samples were acidified, dried at 70°C and a 20 mg sample of the boric acid crystals was analyzed on a continuous flow mass spectrometer. Reported  $^{15}\text{N}$  values for microbial biomass were corrected for the N content of blank samples which were assumed to have an  $^{15}\text{N}$  abundance of 0.3663 atom %.

Light fraction organic matter was determined using a procedure similar to that described by Janzen et al. (1992). In brief, soil was vigorously mixed with NaI (specific density of 1.7), and the suspension allowed to settle for approximately 2 d. The "light fraction" floating on the solution surface was removed by vacuum and washed with a dilute  $\text{CaCl}_2$  solution and then water. The procedure was then repeated. The organic material collected was dried at 70°C, finely ground and analyzed for N and  $^{15}\text{N}$ .

Mineralizable C and N was determined in a 12 wk incubation using a procedure similar to that described by Bremer et al. (1994). Five 50 g subsamples were taken from each of the three bulk soil samples, moistened to 80% of field capacity and individually incubated in sealed 1 L jars at 25°C. Jars were aerated periodically and the  $\text{CO}_2$  trap (10 mL of 2 M NaOH) in each jar was replaced at 1, 2, 4, 8 and 12 wk. To quantify carbonate content, the NaOH samples were placed in an airtight 1 L jar, acidified with HCl and the  $\text{CO}_2$  evolved was measured by gas chromatography. After 1, 2, 4, 8 and 12

wk, 1 of the 5 subsamples being incubated was air dried and analyzed for inorganic N as described previously. The inorganic  $^{15}\text{N}$  content was determined by a diffusion method (Brooks et al. 1989).

#### Measurement of the effect of termination method on the subsequent fate of $^{15}\text{N}$

Termination treatments were applied to the remaining alfalfa plants. Four treatments were established: 1) labelled roots from alfalfa terminated by herbicide application; 2) labelled roots from alfalfa terminated by tillage; 3) labelled top-growth from alfalfa terminated by herbicide application and 4) labelled top-growth from alfalfa terminated by tillage.

Prior to termination, six labelled plants were selected and grouped according to size in a randomized complete block design with three replications. Six unlabelled plants were similarly arranged. Within each replicate, one cylinder was designated for herbicide application and the other for tillage. In total, herbicide was applied to 3 labelled and 3 unlabelled plants; the remaining three labelled and three unlabelled plants were designated for tillage.

In herbicide treatments, glyphosate at a rate of 6.72 mL per 100 mL water was applied to the alfalfa top-growth with a hand-held spray applicator. The soil surface was covered with vermiculite to avoid contamination with herbicide.

In tillage treatments, alfalfa top-growth was cut about 2 cm below-ground, air dried for several days and weighed. A small subsample of top-growth was oven dried and dry matter yield and total N and  $^{15}\text{N}$  concentrations were determined.



To produce "labelled root" and "labelled top-growth" treatments, the top-growth (including crown) of all labelled and unlabelled plants was removed. Labelled top-growth was transferred to the corresponding unlabelled cylinders; unlabelled top-growth was oven dried, ground and analyzed. Thus, "labelled root" treatments contained only  $^{15}\text{N}$ -labelled root material and no top-growth; "labelled top-growth" treatments contained unlabelled root material and  $^{15}\text{N}$ -labelled top-growth.

To simulate tillage, alfalfa roots or roots+top-growth were cut into 4-5 cm lengths and mixed thoroughly throughout the soil. In treatments to which herbicide had been applied, labelled roots were left undisturbed in the soil and, in labelled top-growth treatments, alfalfa top-growth was left intact and placed on the soil surface.

Five days after herbicide application, 10 'Galt' barley seeds were planted in each cylinder. Barley was thinned to 5 plants cylinder<sup>-1</sup> a few days after emergence.

All cylinders were destructively sampled 4 wk after barley was seeded. Barley top-growth was removed at the soil surface and oven dried. Where present, surface alfalfa residue was removed and oven dried. Barley roots and alfalfa residue were removed from the soil by manual picking with forceps. Plant material collected from the soil was separated into fine roots (barley and alfalfa), intermediate roots, taproot and alfalfa stems. The soil adhering to the roots and incorporated plant tissue was removed as described previously for rhizosphere soil.

The bulk soil was analyzed for N,  $^{15}\text{N}$ , inorganic N, mineralizable C and N and light fraction organic matter. Chemical analyses were conducted as described previously except mineralizable C and N was determined after 8 instead of 12 wk.

For calculations of  $^{15}\text{N}$  in alfalfa prior to termination, the natural abundance of alfalfa in unlabelled control treatments was used and is indicated in the tables; for any other calculations of  $^{15}\text{N}$  in plant tissue, 0.37 atom % was used. For calculations of  $^{15}\text{N}$  in soil, a natural abundance of 0.3685 atom % (the atom % of the bulk soil in pots in which unlabelled alfalfa plants had been grown) was used unless otherwise indicated.

The % of nitrogen derived from legume (%NDFL) was calculated for all plant and soil samples as follows:

$$\% \text{NDFL} = (A_s - A_b) / (A_l - \text{NA})$$

where:

$A_s$  =  $^{15}\text{N}$  abundance (atom %) in amended sample

$A_b$  =  $^{15}\text{N}$  abundance in background sample

$A_l$  =  $^{15}\text{N}$  abundance in appropriate legume N source (ex. roots, top-growth)

NA = natural abundance of legume N source

Data from shoot and root treatments were analyzed by analysis of variance using the Proc GLM procedure (SAS Institute Inc. 1985). If the effect of replicate was significant, data were analyzed as a randomized complete block design; if the effect of replicate was not significant, data were re-analyzed as a completely randomized design. The %NDFL in barley and % recovery of alfalfa-derived N by barley was tested for homogeneity of variance using the modified Bartlett test statistic developed by Box (Neter et al. 1990). In cases where the Bartlett test revealed heterogeneity of variance, data were log transformed prior to analysis.

Relative rates of N and C mineralization from  $^{15}\text{N}$ -labelled alfalfa leaf, stem, crown, fine root and taproot tissue

An incubation experiment was conducted to measure N and C mineralization from  $^{15}\text{N}$ -labelled alfalfa. Five treatments, consisting of leaf, stem, crown, fine root and taproot tissue, and a 'soil only' control were established and incubated at 25°C. The evolution of  $\text{CO}_2$  and concentrations of soil inorganic N were measured periodically.

Alfalfa top-growth and roots harvested in the  $^{15}\text{N}$  labelling experiment were oven dried and ground. Tissue from each of the three labelled alfalfa plants was ground and stored separately; treatments were assigned to each replicate on the basis of the alfalfa plant from which tissue had been harvested.

A Cavendish sandy loam soil was air dried and passed through a 2 mm sieve. (This soil had been collected near Purple Springs, AB and stored in a stockpile at the Lethbridge Research Centre prior to its use.) Fifty grams of soil (oven dry basis) were placed in a specimen cup and oven dry plant tissue was mixed into the soil at a rate of 0.2 g cup<sup>-1</sup> (except for stem tissue which was applied at a rate of 0.15 g cup<sup>-1</sup> because of small sample size). Soils were moistened to 80% of field capacity and placed in a sealed 1 L glass jar with a vial containing 10 mL of 2 M NaOH to collect  $\text{CO}_2$ . Jars were aerated periodically during the incubation. At 1, 2, 4, 8 and 12 wk, NaOH was replaced. At 4 and 12 wk, three replications of each treatment were destructively sampled and soil inorganic N determined. Inorganic N,  $^{15}\text{N}$  and  $\text{CO}_2$  were determined as described previously.

Data were analyzed by one way analysis of variance for a completely randomized design and by least significant difference procedure using the Proc GLM procedure (SAS

Institute Inc. 1985).

## RESULTS

### Distribution in the plant-soil system of $^{15}\text{N}_2$ fixed by alfalfa

Nitrogen concentration in the labelled alfalfa ranged from 1.1% in crown tissue to 5% in leaf tissue (Table 6.1). At time of termination, the majority (62%) of alfalfa N occurred in roots. This estimate may have been conservative because of incomplete recovery of fine roots. Stem and leaf material comprised a smaller proportion (26%) of total alfalfa N since alfalfa top-growth had been harvested shortly before sampling. Despite its high dry matter yield, the crown comprised only 12% of total alfalfa N.

All plant parts were effectively labelled during exposure of the alfalfa to a soil atmosphere containing  $^{15}\text{N}_2$ .  $^{15}\text{N}$  concentrations were fairly uniform throughout the plant ranging from 3.68 atom % in fine roots to 4.62 atom % in leaves (Table 6.1). The slightly higher  $^{15}\text{N}$  concentrations in leaf and stem tissues likely resulted because virtually all of the leaf and stem material harvested was produced during exposure to the  $^{15}\text{N}$ -labelled atmosphere whereas crown and root tissue had partially developed before exposure to  $^{15}\text{N}$ . In addition, the  $^{15}\text{N}$  concentration measured for roots may have been diluted with N in adhering soil particles, though the C concentration in the plant material (43 to 47%) suggests that soil contamination was minimal.

Small amounts of alfalfa-derived N were also recovered in the soil. Of the alfalfa-derived N in the soil following the growth of alfalfa, 83% was recovered in the bulk soil (Table 6.2). Based on analysis of root-free and bulk soil samples, an estimated 36% of

alfalfa-derived N in the bulk soil (the equivalent of 7.5 mg alfalfa-derived N cylinder<sup>-1</sup>) occurred as alfalfa roots not recovered during manual picking. An additional 11.8 mg alfalfa-derived N occurred in forms other than root material.

A significant proportion of the legume-derived N recovered in the bulk soil occurred in labile soil fractions. The light fraction of the bulk soil contained 9 mg of alfalfa-derived N, and accounted for 41% of the alfalfa-derived N in the soil at sampling. Much of the light fraction organic matter may have been unrecovered roots; the quantity of legume-derived N in the bulk soil not attributable to light fraction (10 mg) was nearly equivalent to that of legume-derived N in root-free soil (12 mg).

Microbial biomass N comprised only 2% of the total N in soil but 42% of the alfalfa-derived N present in the soil. The %NDFL in microbial biomass was 20.4%, the equivalent of 9 mg alfalfa-derived N cylinder<sup>-1</sup>. These values, however, are only rough approximations because of uncertainty arising from estimates of  $k_N$  and the assumptions made in correcting for the <sup>15</sup>N content of blanks.

Mineralizable N in the bulk soil averaged 60 mg N cylinder<sup>-1</sup>. The proportion of mineralized N derived from alfalfa ranged from 21% after 1 wk to 12% after 12 wk (Table D.1 in Appendix D). After a 1 wk incubation, the equivalent of 7% (1.6 mg cylinder<sup>-1</sup>) of the alfalfa-derived N present in the soil at time of sampling had been mineralized; after 12 wk, the equivalent of 32% (7.2 mg cylinder<sup>-1</sup>) had been mineralized (Fig. 6.4).

Of alfalfa-derived N deposited in the soil, significant amounts were present in the rhizosphere soil. An average of 18 g rhizosphere soil was recovered from each alfalfa

plant although the mass of rhizosphere soil varied depending largely on soil moisture content at time of sampling. The average N concentration in rhizosphere soil was twofold that in the bulk soil and totalled 21 mg N cylinder<sup>-1</sup> (Table 6.2). Considerably more of the N in the rhizosphere soil was derived from alfalfa than in bulk soil; the % NDFL in rhizosphere soil ranged from 13 to 26% for a total of 4 mg alfalfa-derived N cylinder<sup>-1</sup>. Alfalfa N present in the rhizosphere soil as root fragments or root exudates accounted for 17% of the total alfalfa-derived N present in soil.

Of the total <sup>15</sup>N present in the plant-soil system, only 12% occurred in the soil. Of this, most was present in the bulk soil equally distributed between the light and heavy fractions; only 2% of total <sup>15</sup>N occurred in rhizosphere soil. The majority of below-ground (82%) and total (88%) <sup>15</sup>N was present in the roots and shoots of alfalfa (Table 6.3). Of the <sup>15</sup>N in the alfalfa plant remaining after the second harvest of top-growth and a brief regrowth period, 60% was in the roots (28% in fine roots and 32% in the taproot) and 40% in the top-growth.

#### Effect of termination method on the distribution of <sup>15</sup>N<sub>2</sub> fixed by alfalfa

Method of alfalfa termination did not affect dry matter yield, plant tissue N concentration or N uptake by the barley crop established immediately after termination of alfalfa (Table 6.4).

Termination method had a strong influence on the plant-availability of alfalfa-derived N, however. Approximately 12.8% of barley N was derived from alfalfa top-growth incorporated into the soil in simulated tillage treatments whereas only 1.3% of

barley N was derived from herbicide-treated alfalfa top-growth retained on the soil surface. As well, we observed more than an eightfold greater recovery by barley of N from alfalfa top-growth in tillage (10.4%) than in herbicide (1.2%) treatments.

In contrast, in  $^{15}\text{N}$ -labelled root treatments, the proportion of barley N derived from alfalfa roots was significantly greater in herbicide-treated (42.4%) than in tilled treatments (25.3%) (Table 6.4). However, the % of alfalfa N recovered by barley did not differ significantly between treatments.

The average recovery of applied  $^{15}\text{N}$  in labelled shoot treatments ranged from 91% in herbicide treatments to 101% in tillage treatments. Recoveries for treatments involving labelled alfalfa roots could not be calculated directly because root weights could not be determined. Therefore, the distribution of  $^{15}\text{N}$  presented in Table 6.5 has been reported on the basis of total  $^{15}\text{N}$  excess present at the time of barley harvest. This calculation assumes no losses of N via volatilization.

Regardless of termination method, a relatively small proportion (1 to 10%) of the  $^{15}\text{N}$  present occurred in the top-growth of the barley crop established after alfalfa termination (Table 6.5). In tilled top-growth treatments, most of the  $^{15}\text{N}$  was found in the soil; in the remaining treatments, most of the  $^{15}\text{N}$  occurred in residue comprised of alfalfa residues and barley roots. (Detailed analyses of barley, residue and soil components are reported in Tables D.2 to D.4 in Appendix D.)

Termination of alfalfa top-growth by tillage increased the proportion of  $^{15}\text{N}$  present in the soil and decreased the proportion retained in crop residue (Table 6.5). Almost 90% of the  $^{15}\text{N}$  in herbicide treatments was retained in surface residues compared to 38% in

incorporated residues of tilled treatments. In herbicide treatments, only 8% of  $^{15}\text{N}$  was recovered from the soil whereas 52% of  $^{15}\text{N}$  occurred in the soil of tilled treatments. Tillage also increased the proportion of  $^{15}\text{N}$  taken up by barley; barley top-growth contained 10% of  $^{15}\text{N}$  present in tillage treatments but only 1% of  $^{15}\text{N}$  in herbicide treatments.

In contrast, termination treatment had no measurable effect on the distribution of  $^{15}\text{N}$  from labelled alfalfa roots (Table 6.5). The soil and recovered residues contained an average of 30% and 60% of  $^{15}\text{N}$ , respectively. For both methods of alfalfa termination 10% of  $^{15}\text{N}$  was recovered in the top-growth of the subsequent barley crop.

Although termination method altered the distribution of  $^{15}\text{N}$  among barley top-growth, residue and soil components, detailed analysis revealed minimal effects of termination treatments on labile N fractions. Termination method had no effect on mineralizable N in shoot or root treatments (Table D.5 in Appendix D). Mineralizable N after 8 wk averaged 18.8 and 13.3 mg N kg<sup>-1</sup> soil for alfalfa shoot and root treatments. Regardless of treatment, mineralizable N accounted for a significant proportion of the  $^{15}\text{N}$  present in the soil after barley was harvested. Mineralizable N accounted for 16 to 34% of the  $^{15}\text{N}$  present in the soil of shoot treatments and 9 to 19% of the  $^{15}\text{N}$  present in the soil of root treatments.

Termination treatment also had no effect on the amount of N present as light fraction organic matter. Light fraction organic matter averaged 145 mg N cylinder<sup>-1</sup> in top-growth treatments and 159 mg N cylinder<sup>-1</sup> in root treatments (Table D.6 in Appendix D). Incorporation of alfalfa top-growth increased the %NDFL ( $P=0.01$ ) and the mg N



derived from alfalfa ( $P=0.03$ ) in the light fraction. In tillage treatments, 0.53% (0.76 mg N cylinder<sup>-1</sup>) of the light fraction N was derived from alfalfa top-growth compared to 0.22% (0.29 mg N cylinder<sup>-1</sup>) in herbicide treatments. Termination method did not affect the proportion or mass of light fraction derived from alfalfa roots; approximately 2% of light fraction N was derived from alfalfa roots, the equivalent of 3 mg N cylinder<sup>-1</sup>.

#### Relative rates of N and C mineralization from alfalfa leaf, stem, crown, fine root and taproot

Leaf tissue was rapidly mineralized resulting in release of 37% of applied N after 4 wk and 46% of applied N after 12 wk (Table 6.6). Stem and root tissue mineralized more slowly with 26 to 31% of applied N released after 12 wk. Mineralization of crown tissue was substantially delayed. After 4 wk, net mineralization totalled only 1.5% of applied N; however, after 12 wk, the quantity of N released had increased to 21% of applied N. These data agree with results of a previous study in which the equivalent of 51, 34 and 27% of applied legume N was mineralized from leaf, stem and root tissue after a 12 wk incubation period (Chapter 5).

The addition of leaf tissue resulted in accumulations of 5.24 mg N container<sup>-1</sup>. Significantly lower N accumulations averaging 1.41 mg N container<sup>-1</sup> were observed with the addition of root tissue. Lowest net N mineralization from alfalfa tissue resulted from soil-applied crown material; after 12 wk net mineralization totalled 0.30 mg N container<sup>-1</sup>. Low N concentrations and an initial period of net immobilization of N present likely contributed to the low levels of net mineralization observed for crown tissue. The %NDFL ranged from 36% in crown treatments to 74% in leaf treatments; the % NDFL

in stem and root treatments was intermediate ranging from 53 to 58%.

## DISCUSSION

### Effectiveness of labelling with $^{15}\text{N}$ -labelled $\text{N}_2$

Exposure of alfalfa to a soil atmosphere containing  $^{15}\text{N}$ -labelled  $\text{N}_2$  effectively labelled the alfalfa tissue and allowed direct measurement of N release from alfalfa residues. Several limitations of this technique were apparent in our study, however. Regulation of moisture content and  $\text{O}_2$  concentrations in the soil was difficult with the labelling system used. The system used could accommodate only a limited number of plants and appeared to stress plants somewhat after prolonged, continuous exposure; these factors both contributed to the variability observed among labelled alfalfa plants. In addition, the pool substitution effect (Hart et al. 1986), which may vary in magnitude in herbicide and tillage treatments, may have resulted in underestimation of the uptake of legume-derived N by the subsequent barley crop.

### N release during alfalfa growth

Results of this study indicate that a relatively small proportion of symbiotically fixed N is released into the soil during alfalfa growth. Several days after harvest, the alfalfa contained 88% of  $^{15}\text{N}$  present in the plant/soil system; the remaining 12% of the  $^{15}\text{N}$  present was recovered in the soil, in part as unrecovered root fragments. A similar distribution of symbiotically fixed N was observed for  $^{15}\text{N}$ -labelled beans (*Phaseolus* spp.) and soybeans (*Glycine max*); approximately 12 to 18% of symbiotically fixed N was

recovered in the soil and the remaining 72 to 78% was retained in the plant (Ruschel et al. 1979). In contrast, Poth et al. (1986) estimated that a greater proportion of  $^{15}\text{N}_2$  fixed by pigeon pea (*Cajanus cajan*) was released into the soil than retained in the plant.

Of N released into the soil, a significant proportion was deposited into labile N pools. In our experiment, of the  $^{15}\text{N}$  recovered in the soil, 41% occurred as light fraction organic matter, 42% as microbial biomass and 32% as mineralizable N. Presumably, the light fraction organic matter derived from alfalfa was comprised for the most part of sloughed roots or root fragments. This being the case, and given that 12% of the total amount of  $^{15}\text{N}$  present occurred in the soil, an estimated 5% (41% of the 12% in soil) of the total amount of  $^{15}\text{N}$  present several days after harvest occurred as sloughed roots or root fragments, 5% (42% of the 12% in soil) as microbial biomass and the remaining 2% (32% of the 12% in soil) as root and microbial products.

Assuming soil N accumulations of the magnitude observed in this experiment (23.1 mg symbiotically fixed N plant<sup>-1</sup> present in the soil including unrecovered root fragments) and a stand density of 50 alfalfa plants m<sup>-2</sup>, an estimated 12 kg fixed N ha<sup>-1</sup> would have been released from living alfalfa during the 74 d duration of alfalfa growth. Given that an estimated 227 mg N plant<sup>-1</sup> was removed as top-growth in the two harvests taken during the experiment (based on an average total harvest of 8.3 g top-growth per  $^{15}\text{N}$ -labelled plant and the average N concentration of alfalfa top-growth removed during the first harvest of 2.74%), the amount of N released into the soil by growing alfalfa would be equivalent to 10% of the total amount of N removed in alfalfa top-growth during two harvests. Similar results have been reported elsewhere. Dubach and Russelle

(1994) estimated that less than 15 kg symbiotically fixed N ha<sup>-1</sup> could be released from decomposing roots and nodules during the first year of a pure alfalfa stand. Andrén et al. (1990) estimated N release from the below-ground biomass of an established alfalfa stand of 32 kg N ha<sup>-1</sup> yr<sup>-1</sup> (24 kg fixed N ha<sup>-1</sup> yr<sup>-1</sup> assuming 76% of alfalfa N resulted from symbiotic N<sub>2</sub> fixation) which is equivalent to 13% of the N removed as alfalfa top-growth during harvest (246 kg N ha<sup>-1</sup> yr<sup>-1</sup>).

Previous studies have attempted to identify the source of soil N accumulations under growing alfalfa. Direct excretion results in relatively small N losses and may be less important for N release than other processes (Russelle et al. 1994). Decomposition of below-ground residue has been identified by Dubach and Russelle (1994) as a significant source of N release from growing alfalfa; however, neither decomposition nor direct excretion account for the amounts of N reportedly transferred from legumes to intercropped non-legumes. These authors suggested above-ground residues as a potential N source. In an earlier study, Andrén et al. (1990) reported that under an established alfalfa stand, stubble and litter contributed 103 kg N ha<sup>-1</sup> yr<sup>-1</sup> to the soil organic pool whereas the below-ground biomass contributed 32 kg N ha<sup>-1</sup> yr<sup>-1</sup>. For an alfalfa/bromegrass intercrop, litterfall losses combined with harvest losses contributed an estimated 38 kg N ha<sup>-1</sup> yr<sup>-1</sup> to the soil (Tomm et al. 1995).

In our study, symbiotically fixed N released into the soil presumably resulted from a combination of direct excretion and decomposition of roots and nodules. Because the soil surface was covered throughout the labelling period, above-ground residues could not provide N to the soil.

Under field conditions, various factors including environmental conditions, stand composition and health, and management factors (number of harvests per growing season, stand duration) may affect the amount of N released from a growing alfalfa stand and also the relative contributions of below and above-ground N sources.

#### N distribution as influenced by termination method

Results of our study indicate that regardless of termination method, a relatively small proportion (1-10%) of N symbiotically fixed by alfalfa becomes plant-available shortly after termination. In general, the values obtained were comparable to values reported in the literature which range from 6 to 28% (Ladd et al. 1981; Ladd et al. 1983; Müller and Sundman 1988).

Approximately 5 wk after alfalfa termination, in all termination treatments, a significant proportion of the symbiotically fixed N present was retained in residues or deposited into the soil. Thirty-eight to 65% of the symbiotically fixed N present remained in below-ground or incorporated residues whereas 91% remained in the residues of surface-applied alfalfa top-growth. Eight to 52% of the symbiotically fixed N present occurred in the soil. These data support the results of previous studies which have demonstrated that the main benefit of legume crops is to long-term soil fertility and productivity (Ladd et al. 1981; Janzen et al. 1990).

Based on results of the incubation study conducted, it appears that mineralization of readily decomposable leaf tissue is important for the short-term soil fertility benefits derived from alfalfa residues whereas mineralization of more slowly decomposable crown,

stem and root tissue may be of comparatively less importance in the short term. More rapid, extensive decomposition of leaf tissue has also been observed for *Medicago* spp. incubated under field conditions (Amato et al. 1984). Leaf, stem and root tissue contained 64%, 87% and 81% of initial organic  $^{15}\text{N}$  after 4 wk and 40%, 56% and 50% of initial organic  $^{15}\text{N}$  after 2 yr. Observed differences in the decomposability of residues have been attributed to various factors including the C/N ratio, concentration of water-soluble constituents, and lignin and carbohydrate content of the plant residue (Nyhan 1975; Herman et al. 1977).

Residue placement appeared to have a greater influence on the fate of symbiotically fixed N than termination method *per se*. Termination method had no measurable effect on the distribution of symbiotically fixed N from alfalfa roots which suggests that neither differences in residue particle size nor differences resulting from chemical versus mechanical disruption of the plant affected N release. A previous study demonstrated minimal effects of residue particle size on N mineralization from alfalfa residues (Chapter 5); however, herbicide application was found to increase short-term plant-available N. In that study, greater short-term N availability was attributed to greater root exudation and/or more rapid senescence and subsequent decomposition of alfalfa residue following herbicide application.

Tillage enhanced the release of symbiotically fixed N from alfalfa top-growth. Residue incorporation substantially increased the proportion of  $^{15}\text{N}$  present in the soil and decreased the proportion of  $^{15}\text{N}$  retained in residues. This increased the availability of symbiotically fixed N to barley grown for a 4 wk period after alfalfa termination.

Presumably, incorporation resulted in greater exposure of alfalfa residue, particularly readily decomposable leaf tissue, to soil microbial populations which enhanced mineralization (Cogle et al. 1987). Results of this study support a previous study which demonstrated substantially greater release of short-term plant-available N from incorporated than surface-applied alfalfa residue (Chapter 5). Residue incorporation has also been shown to increase short-term N release under field conditions. Varco et al. (1993) found that after 15 d, 47% of the N in incorporated vetch residue was recovered as soil inorganic N compared to 12% of the N in surface-applied vetch residue. After 30 d, 89% and 60% of the applied N had been lost from legume residue in conventional till and no-till treatments, respectively.

## CONCLUSIONS

During the growth of alfalfa, a relatively small proportion of symbiotically fixed N (12%) is released into the soil, much of it into labile N pools; most of the symbiotically fixed N (88%) is retained in the alfalfa plant.

Termination may substantially increase the proportion of symbiotically fixed N released from alfalfa; however, the fate and short-term plant availability of this symbiotically fixed N may be strongly influenced by the termination method employed, particularly as it affects residue placement. Based on the relative amounts of alfalfa leaf, stem, crown and root tissue present at time of sampling and the relative mineralization rates of alfalfa plant parts measured in our study, alfalfa shoots and roots would be expected to contribute almost equally to the mineralizable N pool in the short-term. In

our study, an estimated 28% of mineralizable N would be derived from leaves, 8% from stems, 8% from crowns, 27% from fine roots and 29% from taproots. Therefore, termination methods like tillage which enhance the mineralization of alfalfa top-growth increase the short-term plant-available N supply whereas surface application of herbicide-terminated alfalfa delays N mineralization from alfalfa top-growth resulting in a smaller supply of plant-available N in the short-term.



Table 6.1. Composition of alfalfa (mean of three plants) labelled by continuous, prolonged exposure of the root system to  $^{15}\text{N}$  labelled  $\text{N}_2$ . Alfalfa was destructively sampled 10 d after the second cut of alfalfa top-growth was taken.

Component	Mass (mg)	% C	%N	% of plant N	$^{15}\text{N}$ atom % (excess) <sup>†</sup>
leaf	635 (195) <sup>‡</sup>	45.5 (0.6)	5.0 (0.4)	18 (2.0)	4.26 (0.09)
stem	480 (125)	43.3 (0.1)	2.8 (0.1)	8 (0.6)	4.04 (0.07)
crown	1840 (418)	46.7 (0.8)	1.1 (0.1)	12 (0.3)	3.38 (0.23)
fine root	1990 (813)	43.2 (0.2)	2.7 (0.1)	32 (10.8)	3.32 (0.22)
taproot	2300 (589)	43.7 (0.01)	2.2 (0.1)	30 (11.4)	3.85 (0.09)

<sup>†</sup> Natural abundance values based on the mean of 3 unlabelled alfalfa plants grown and sampled concurrently (leaf=0.3662, stem=0.3658, crown=0.3664, fine root=0.3668, taproot=0.3664 atom %).

<sup>‡</sup> Standard deviation given in parenthesis following the mean.

Table 6.2. Distribution of symbiotically fixed  $^{15}\text{N}$  in the bulk soil and rhizosphere soil following growth of alfalfa. Alfalfa was destructively sampled 10 d after the second cut of alfalfa top-growth was taken. (Values represent the mean of three cylinders.)

Component	mg N kg <sup>-1</sup>	mg N <sup>†</sup>	%NDFL	mg N <sub>(alf)</sub> <sup>‡</sup>	% of soil N <sub>(alf)</sub>
Bulk soil <sup>§</sup>	670	2140	0.90 (0.23)	19.3 (4.98)	83 (2)
light fraction	54	171	5.31 (1.08)	9.1 (1.90)	41 (15)
microbial biomass	14	44.2	20.4 (5.5)	9.0 (2.00)	42 (22)
mineralizable N <sup>¶</sup>	19	59.7	12.0 (1.6)	7.2 (1.52)	32 (4)
root-free	640	2040	0.58 (0.06)	11.8 (0.90)	53 (14)
Rhizosphere soil					
closely associated	1760	11.0	25.5 (3.4)	2.8 (0.11)	12 (3)
loosely adhering	900	9.8	12.5 (4.5)	1.1 (0.40)	5 (1)

<sup>†</sup> The mg N in components of the bulk soil are based on the mass of bulk soil (3221 g - mass of rhizosphere soil) in each cylinder. For selected measurements, the standard deviation is given in parenthesis following the mean.

<sup>‡</sup> mg N<sub>(alf)</sub> indicates the mg N derived from alfalfa in each cylinder. Natural abundance values are based on the mean of 3 unlabelled pots of alfalfa grown and sampled concurrently (bulk soil=0.3685, closely associated rhizosphere soil=0.3687, loosely adhering rhizosphere soil=0.3684, root-free soil=0.3692).

<sup>§</sup> Light fraction, microbial biomass, mineralizable N and root-free samples are based on a subsample taken from the bulk soil. The bulk soil contained 0.60 mg NO<sub>3</sub><sup>-</sup>-N kg<sup>-1</sup> and 1.51 mg NH<sub>4</sub><sup>+</sup>-N kg<sup>-1</sup>.

<sup>¶</sup> Mineralizable N and C were measured after 12 wk; mineralizable C=168 mg C kg<sup>-1</sup>.

Table 6.3. Distribution among soil and plant components of  $^{15}\text{N}$  symbiotically fixed by alfalfa (as % of the below-ground and total  $^{15}\text{N}$  excess present). Alfalfa was destructively sampled 10 d after the second cut of alfalfa top-growth was taken.

Component	% of below-ground $^{15}\text{N}$ excess <sup>†</sup>	% of total $^{15}\text{N}$ excess
Alfalfa		
top-growth	-	35.6
fine root	38.7	24.8
taproot	43.5	28.0
	82.2	88.4
Soil		
rhizosphere soil	3.0	1.9
light fraction	7.2	4.7
heavy fraction (by difference)	7.7	4.9
	17.9	11.5

<sup>†</sup> Calculations were based on a total of 3 cylinders.

Table 6.4. Effect of alfalfa termination by herbicide application or simulated tillage on dry matter yield, N uptake and  $^{15}\text{N}$  content of a subsequent barley crop. Barley was established immediately after the application of termination treatments to  $^{15}\text{N}$ -labelled alfalfa top-growth and root tissue and grown for 4 wk.

	Barley provided with labelled alfalfa top-growth			Barley provided with labelled alfalfa root		
	Herbicide	Tillage	P>F <sup>†</sup>	Herbicide	Tillage	P>F <sup>†</sup>
dry matter yield (g)	1.6	1.4	0.57	1.0	1.3	0.48
% N	2.0	1.8	0.14	1.7	1.5	0.29
N uptake (mg)	32.0	24.1	0.34	16.6	19.9	0.59
% NDFL	1.3	12.8	0.03	42.4	25.3	0.009
% of alfalfa N recovered by barley <sup>‡</sup>	1.2	10.4	0.03	6.1	3.6	0.37

<sup>†</sup> indicates P>F for termination method. The effect of replicate was significant (P=0.01) only for %NDFL in labelled alfalfa root treatments. %NDFL and % recovery data were tested for homogeneity of variance by the modified Bartlett test statistic developed by Box. The ANOVA was conducted on log transformed data for top-growth data only. The df for effects of rep and termination method were 2 and 1, respectively.

<sup>‡</sup> The % of alfalfa N recovered by barley was calculated as [(mg N in barley top-growth derived from alfalfa)/(mg N added in labelled alfalfa)]\*100.

Table 6.5. Effect of alfalfa termination by herbicide application or simulated tillage on the distribution of  $^{15}\text{N}$  excess following the harvest of a subsequent barley crop.

Component <sup>†</sup>	Labelled alfalfa top-growth			Labelled alfalfa root		
	Herbicide	Tillage	P>F <sup>‡</sup>	Herbicide	Tillage	P>F <sup>‡</sup>
	--% of $^{15}\text{N}$ present--			--% of $^{15}\text{N}$ present--		
Barley top-growth	1.1	10.1	0.03	9.7	10.2	0.92
Soil	7.5	51.7	0.0003	25.0	35.4	0.47
Residue <sup>§</sup>	91.4	38.2	0.0007	65.3	54.4	0.56

<sup>†</sup> A detailed breakdown of the % $^{15}\text{N}$  present in individual components of residue and soil is presented in Table D.4 in Appendix D.

<sup>‡</sup> indicates P>F for effect of termination treatment. The df for effects of rep and termination method were 2 and 1, respectively. The effect of replicate was not significant for any measurement.

<sup>§</sup> This value is based on residue + any soil adhering following washing. Except for herbicide-treated, labelled alfalfa top-growth treatments, residue in all treatments was recovered below-ground. Below-ground residues consisted of fine roots (barley+alfalfa), intermediate roots, taproots and, in the case of incorporated top-growth treatments, incorporated alfalfa stems. Where top-growth was surface-applied, most of the  $^{15}\text{N}$  present occurred in surface residues; only 2% of the  $^{15}\text{N}$  present occurred below-ground. [This was significantly (P=0.002) less than in the corresponding tillage treatment.]

Table 6.6. Mineralization of C and N from <sup>15</sup>N-labelled alfalfa leaf, stem, crown, fine root and taproot. Ground plant tissue was incorporated into moistened soil and incubated at 25°C for a 12 wk period during which inorganic N accumulation and CO<sub>2</sub> evolution were measured.

Residue	Net mineralization of alfalfa <sup>†</sup>			% NDFL		% of alfalfa N mineralized <sup>‡</sup>	
	4 wk	12 wk	4 wk	12 wk	4 wk	12 wk	12 wk
	--mg C container <sup>-1</sup> --§			---mg N container <sup>-1</sup> ---			
leaf	52.2	53.4	4.27	5.24	76.2	73.7	45.9
stem	40.7	50.7	1.30	1.79	57.3	55.0	31.2
crown	35.5	47.6	-0.55	0.30	36.1	35.7	20.8
fine root	31.7	39.3	0.78	1.42	60.6	58.1	25.9
taproot	49.1	59.5	0.35	1.40	54.5	52.7	29.2
LSD (P=0.05)	4.8	8.6 <sup>¶</sup>	0.41	0.73	4.1	3.9	3.6

<sup>†</sup> For all treatments, net C and N mineralization of alfalfa was based on 0.2 mg plant material. Net mineralization was calculated as (net mineralization of treatment containing soil+alfalfa residue)-(net mineralization of soil control). The df for the effect of treatment was 4.

<sup>‡</sup> % of alfalfa N mineralized=[(%NDFL/100)\*(net mineralization)]/alfalfa N added]\*100.

<sup>§</sup> The analytical method used to quantify carbonates provides a conservative estimate of mineralizable C.

<sup>¶</sup> For comparisons involving fine root treatments, the appropriate LSD=9.6.

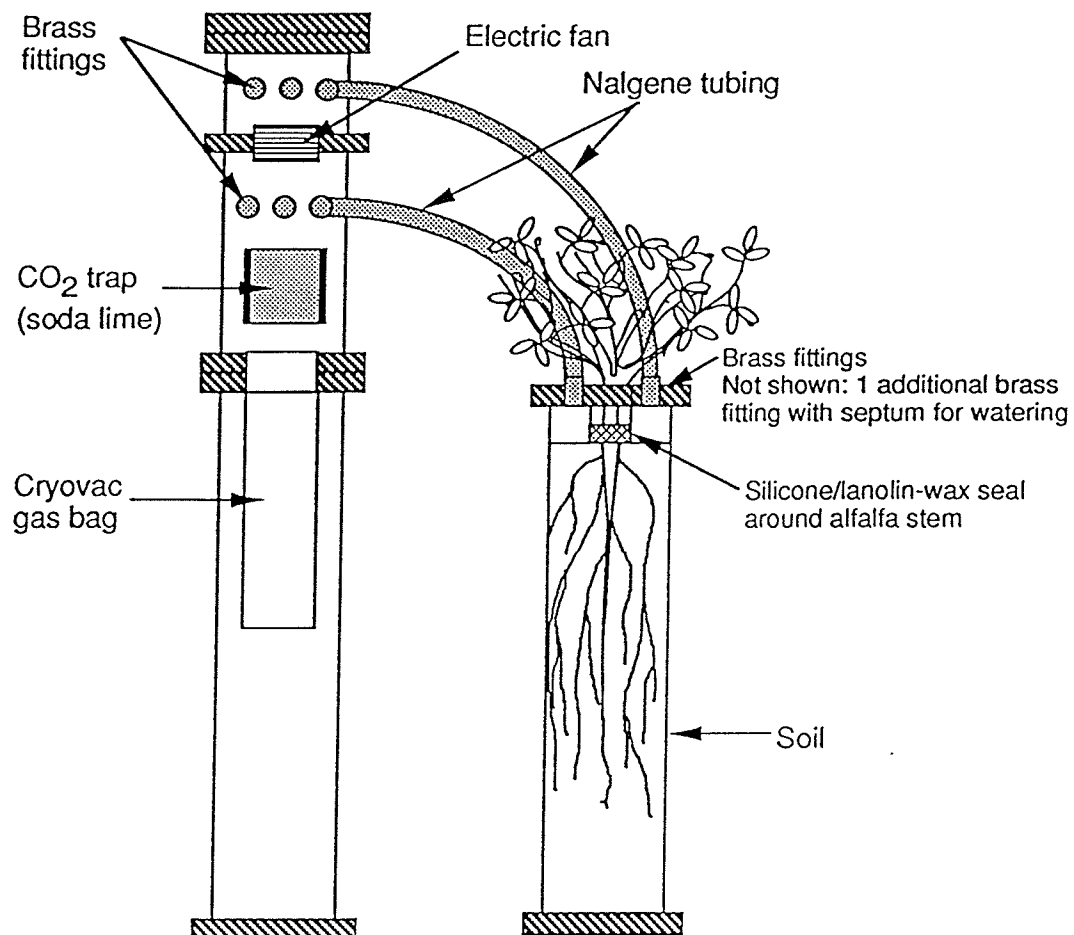


Fig. 6.1. Schematic diagram of the  $^{15}\text{N}$ -labelling system used for continuous exposure of the root system of alfalfa to  $^{15}\text{N}_2$ .

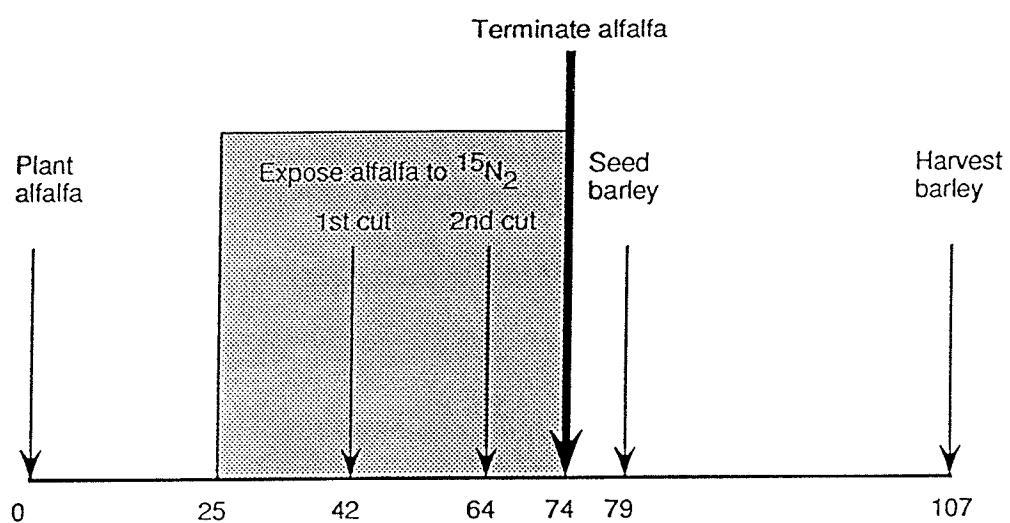


Fig. 6.2. Schedule of the establishment, exposure to  $^{15}\text{N}_2$  and termination of alfalfa and the establishment and harvest of a subsequent barley crop.



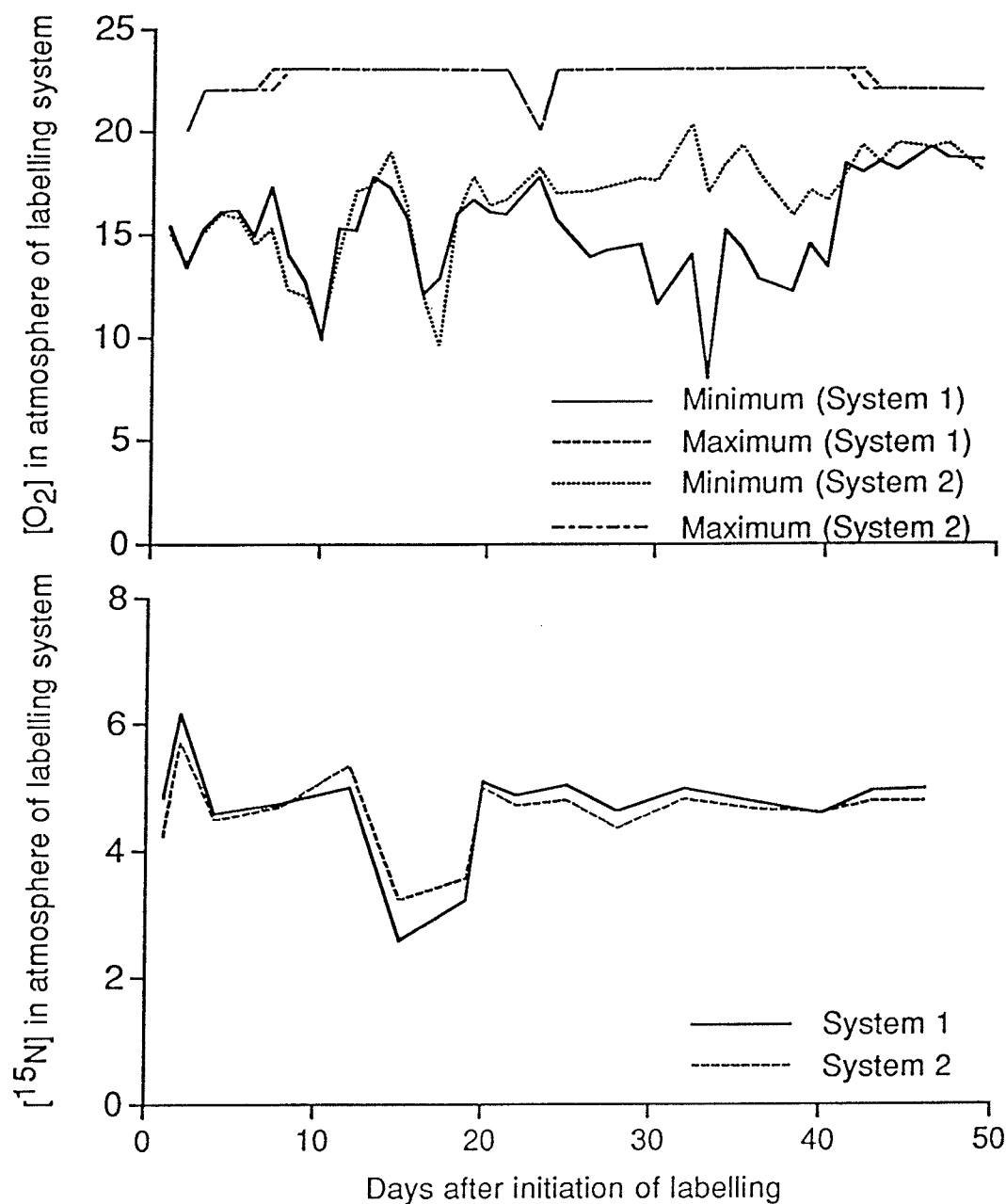


Fig. 6.3.

Concentrations of  $O_2$  and  $^{15}N$  in the atmosphere of systems used to label alfalfa. (Minimum  $O_2$  concentrations were measured prior to  $O_2$  additions; maximum  $O_2$  values are calculated values which approximate the maximum  $O_2$  concentration based on the mL of  $O_2$  added to the system.  $^{15}N$  concentrations were values measured just prior to the addition of highly enriched  $^{15}N_2$ ).

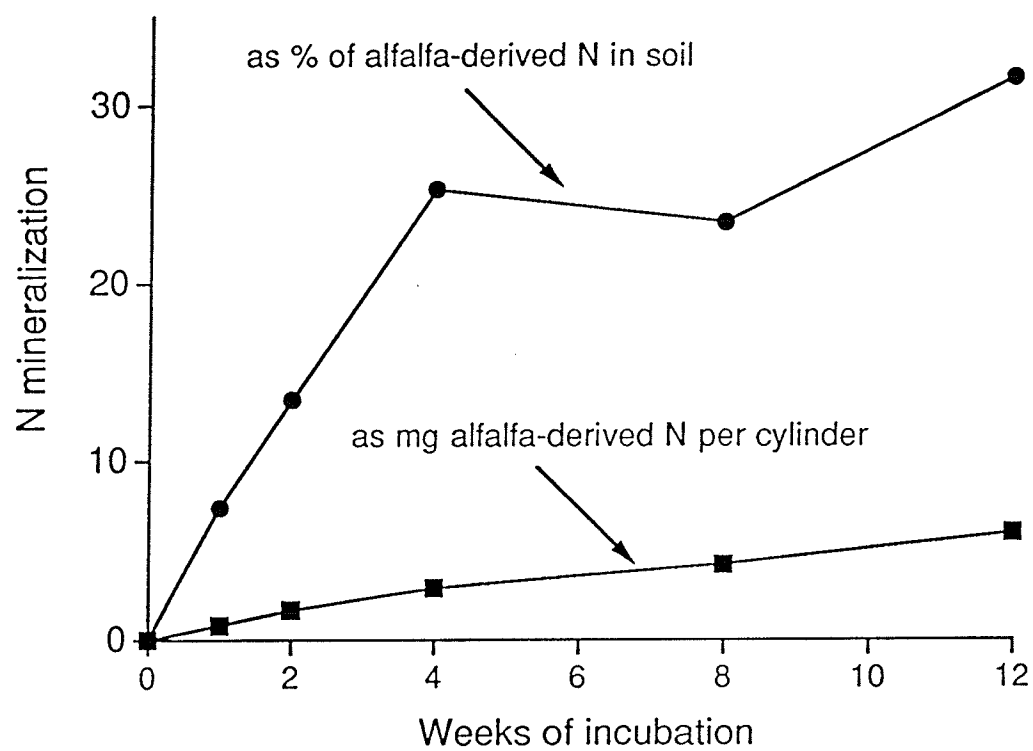


Fig. 6.4. Mineralizable alfalfa-derived N present in soil following growth of alfalfa labelled by exposure of its root system to  $^{15}\text{N}_2$ .

## 7. SUMMARY AND CONCLUSIONS

Concerns regarding agricultural and environmental sustainability have renewed interest in perennial legumes like alfalfa which not only contribute N to the plant-soil system, but also help reduce weed and disease levels and maintain soil quality. One strategy for increasing the proportion of arable land deriving benefits from alfalfa without increasing total forage acreage is to shorten stand duration. However, successful adoption of shorter-term stands demands that management systems be developed which effectively terminate alfalfa while maximizing benefits to subsequent crops.

Intensive tillage remains the most common method of alfalfa termination in western Canada, but it is expensive, often provides inconsistent control of alfalfa, and leaves soil prone to erosion and moisture loss. An alternative which avoids many of these problems is to apply herbicides and leave alfalfa residues standing on the soil surface; however, this practice may alter N dynamics in the plant-soil system.

Since one of the primary benefits of legumes is as a source of N, information regarding the impact of termination technique on N dynamics is required in order to select the most appropriate termination management system. Therefore, a series of field and controlled environment studies was conducted to determine the effect of termination technique on the short-term plant-available N supply and to identify factors influencing N dynamics under different termination management systems.

Four field experiments were initiated on established alfalfa stands in southern Manitoba to determine the effect of method (herbicide, tillage, herbicide+tillage) and time (after first and second cuts of alfalfa) of alfalfa termination on the short-term plant-

available N supply (Chapter 3). Under field conditions, both method and time of termination strongly influenced the plant-available N supply. In 3 of 4 experiments, termination by tillage or herbicide+tillage resulted in larger accumulations of soil  $\text{NO}_3^-$ -N in the spring following alfalfa termination than herbicide application alone. However, greater growing-season N mineralization in herbicide treatments appeared to compensate for these initial differences so that herbicide treatments produced grain yields similar to or greater than tillage treatments. In the one case in which termination by herbicide application resulted in higher yields than tillage, larger soil moisture reserves in the untilled system may have contributed to higher grain yields (Bullied, Dept. of Plant Science, University of Manitoba, unpublished data); differences in N uptake by wheat had not been evident. Previous studies have similarly reported either no yield differences between crops established after legumes in no-till and conventional-till systems (Triplett et al. 1979; Levin et al. 1987), or higher yields in no-till systems which were attributed to increased soil moisture reserves (Sarrantonio and Scott 1988).

Regardless of termination method, delaying termination reduced the soil  $\text{NO}_3^-$  content in the spring following alfalfa termination, presumably because of the shorter period of residue decomposition. Despite the smaller initial available N supply, delaying termination until the second cut of alfalfa did not generally reduce grain yield. However, further delaying herbicide application until the spring immediately before wheat establishment reduced N uptake and yield of the initial wheat crop established after alfalfa termination. Previous studies have also reported poor synchrony between N release and N uptake when legumes were terminated shortly before establishment of a subsequent

crop (Huntington et al. 1985; Westermann and Crothers 1993).

Based on results of the field study, termination management may provide one means of influencing both the amount and timing of N released following the termination of perennial alfalfa stands. By selecting termination management practices and subsequent crops such that N release and N needs by the subsequent crops are well-matched, potential may exist to improve synchrony between N release and N uptake. For example, Westermann and Crothers (1993) suggested that, although N release from alfalfa terminated by fall-applied herbicide was insufficient to meet the N needs of a subsequent winter wheat crop in their study, synchrony might be improved by selecting crops with later-season N uptake (e.g. sugarbeet, potato, corn). In the present study, where N release from alfalfa terminated by a spring-applied herbicide application was not sufficient to meet N needs of a subsequent spring wheat crop, selecting crops with smaller N requirements or supplementing the N supply with fertilizer N may be beneficial. Thus, the influence of different combinations of termination management and subsequent crops, and possibly the use of starter N fertilizers, on N use efficiency, potential N losses and subsequent crop yields may deserve further consideration. Further, although prediction of N release following alfalfa termination is complex, the development of procedures which would allow more accurate prediction of N release would also be beneficial. This information may allow better management of legume N sources and improve the feasibility of utilizing legume N sources into today's cropping systems.

In order to identify factors which influence N transformations and thereby contribute to differences in N availability among management systems, a series of

controlled environment studies was conducted. These studies examined the impact of different termination techniques, specifically effects of differences in residue placement, termination method and residue particle size, on N mineralization and volatilization. In addition, N mineralization from different plant parts was quantified.

An initial greenhouse study was conducted to determine the influence of termination method (chemical, mechanical) and residue placement (incorporated, surface) on volatile N losses from terminated alfalfa (Chapter 4). After a 95 d incubation period, volatile N losses from soil-incorporated residues were negligible. However, where alfalfa residues were surface-applied, volatile N losses amounted to 8% to 12% of surface legume N, the equivalent of 3% to 4% of total legume N (roots+shoots). Termination method did not have a significant effect on volatile N losses. Results of this study suggested that volatile N loss may contribute to a smaller short-term plant-available N supply in herbicide treatments. However, volatile N losses did not account completely for differences in the available N supply of herbicide and tillage treatments, nor were they large enough to preclude the use of herbicides for alfalfa termination.

A growth chamber study was conducted concurrently to determine the effect of termination method (chemical, mechanical) and residue placement (incorporated, surface) on the plant-availability of N following alfalfa termination (Chapter 5). By 25 d after residue incorporation, N accumulations by a subsequent barley crop accounted for >60% of the total amount of N accumulated by four consecutive barley crops during the 125 d study. Nitrogen accumulations by barley in surface-residue treatments accounted for only 24% to 39% of the total amount of N accumulated after 125 d. Large differences in

plant-available N supply were still evident between soil-incorporated ( $104 \text{ mg N pot}^{-1}$ ) and surface-residue ( $57 \text{ mg N pot}^{-1}$ ) treatments at the end of the 125 d experiment. Although herbicide application increased initial N accumulations by barley, no effects of termination method were evident beyond 25 d after alfalfa termination. Perhaps most significant was that residue placement had a substantially larger effect on N availability than did termination method. Thus, the method of residue placement associated with a termination technique, rather than termination method *per se*, appeared to be one of the primary factors influencing the available N supply.

In order to examine more closely the effect of alfalfa termination by herbicide application and tillage on the fate of alfalfa N in the plant-soil system, a growth chamber study was conducted in which alfalfa was continuously labelled with  $^{15}\text{N}_2$  for 7 weeks prior to termination (Chapter 6). Immediately prior to termination, 88% of the  $^{15}\text{N}$  in the plant-soil system was present in alfalfa whereas only 12% was present in the soil (5% in root fragments, 5% in microbial biomass and 2% in root and microbial products). This suggested that a relatively small proportion of symbiotically fixed N was released into the soil during alfalfa growth. Results of this study also confirmed that residue placement strongly influenced the available N supply following alfalfa termination. Moreover, the distribution of symbiotically-fixed  $^{15}\text{N}$  following alfalfa termination demonstrated that the larger short-term available N supply in tilled treatments was due primarily to accelerated mineralization of alfalfa top-growth, not alfalfa roots. In herbicide treatments in which alfalfa top-growth was retained on the soil surface, 1% of the  $^{15}\text{N}$  present was recovered in the top-growth of a subsequent barley crop, 8% in the soil and 91% in plant residues.

However, when alfalfa top-growth was incorporated into the soil, the proportion of  $^{15}\text{N}$  released into the soil and made available for uptake by a subsequent crop increased substantially: in the tilled system, 10% of the  $^{15}\text{N}$  present was recovered in the top-growth of a subsequent barley crop, 52% in the soil and 38% in plant residues. In contrast, termination method had no effect on the fate of symbiotically-fixed  $^{15}\text{N}$  in soils containing roots of terminated alfalfa. Regardless of termination method, 10% of the  $^{15}\text{N}$  present was recovered in the top-growth of a subsequent barley crop, 30% was recovered in the soil and 60% was recovered in plant residues.

The lack of difference between N mineralization from intact roots in herbicide treatments and segmented roots in tilled treatments suggested that neither termination method *per se* nor residue particle size strongly influenced N mineralization. A supplementary 12 wk incubation study which quantified N mineralization from several size fractions of alfalfa leaf (ground, whole), stem (ground, 1.5 cm, 5 cm lengths) and root (ground, 1.5 cm, 5 cm lengths) tissue confirmed that residue particle size had minimal effects on N mineralization (Chapter 5). Thus, reductions in residue particle size resulting from tillage would be expected to have little influence on the short-term plant-available N supply. The effect of tillage therefore appears to be exerted via residue placement rather than physical disruption.

Incubation studies revealed large differences in N mineralization among plant parts. In an initial 12 wk incubation study, 51, 34 and 27% of applied legume N was mineralized from alfalfa leaf, stem and root tissue, respectively (Chapter 5). Similar results were obtained in a second incubation study. After 12 wk, N mineralization from



<sup>15</sup>N-labelled alfalfa leaf, stem, crown and root tissue amounted to 46, 31, 21 and 28% of applied legume N (Chapter 6). Given the highly decomposable nature of leaf material, leaves may be more important than stems, crowns and roots for the short-term N fertility benefit derived from alfalfa. Thus, in tilled systems, not only is a greater proportion of alfalfa residue exposed to soil microbial populations, but the most decomposable fraction is made available to microbial populations. Both factors presumably contribute to the larger short-term plant-available N supply observed in tilled treatments.

In summary, termination of alfalfa by herbicide application reduced the amount of inorganic N accumulated in the soil between time of termination and establishment of a subsequent crop (Chapter 3). These smaller accumulations of soil inorganic N may reduce the potential for N losses by leaching and denitrification; however, surface residues in herbicide treatments may be prone to small volatile NH<sub>3</sub> losses (Chapter 4). Although these volatile N losses may contribute to a small extent to reductions in the short-term plant-available N supply in herbicide treatments, the observed reductions in N availability appeared to be due primarily to reduced mineralization of surface residues (Chapter 6). Neither residue particle size nor termination method (ie. chemical versus mechanical) appeared to strongly influence N availability (Chapter 5; Chapter 6). Even though herbicide termination often reduced the short-term available N supply compared to tillage, greater N mineralization in herbicide treatments during the growing season often compensated for these initial differences. Consequently, despite an initial delay in N release, herbicide-terminated systems contained sufficient plant-available N to achieve yields at least as high as tilled systems.

Termination of established alfalfa stands by herbicide application may improve synchrony between plant-available N release and N needs of a subsequent spring wheat crop, and thereby potentially reduce N losses from the plant-soil system. This may provide significant benefits environmentally by reducing the potential for leaching and denitrification, and their associated hazards: leaching may contribute to  $\text{NO}_3^-$  contamination of water sources whereas  $\text{N}_2\text{O}$  production due to denitrification may contribute to the depletion of atmospheric ozone and to the greenhouse gas effect. From an agronomic perspective, improving synchrony between N release and N utilization by subsequent crops is beneficial in that the efficiency with which N is used by subsequent crops may be improved. Although significant benefits were apparent under the conditions of this study, various factors including environmental conditions, the proportion of alfalfa present in a terminated stand, the duration between termination and establishment of a subsequent crop, and the N needs of a subsequent crop, will ultimately determine the feasibility of herbicide-termination of established alfalfa stands.

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## 9. APPENDICES

## Appendix A



Table A.1. Mean monthly air temperature for selected field sites.

Site	Year	Jan	Feb	Mar	Apr	May	June	July	Aug	Sept	Oct	Nov	Dec
-----mean temperature (°C)-----													
Carman	1992	-	-	-	-	13.0	15.4	15.5	15.8	11.2	5.0	-4.2	-16.7
	1993	-16.1	-13.3	-4.5	4.2	11.5	15.0	17.1	17.5	10.6	3.5	-6.0	-12.0
	1994	-22.8	-18.1	-1.4	4.4	12.9	17.7	18.0	16.8	14.8	-	-	-
Glenlea	1992	-	-	-	2.5	12.8	15.0	15.8	15.5	10.3	4.5	5.0	-17.0
	1993	-18.0	-16.0	-1.5	4.0	12.5	14.8	11.3	13.5	10.3	2.5	-5.0	-12.5
	1994	-24.5	-19.0	-3.0	4.0	12.6	17.9	18.1	16.5	14.5	-	-	-

Source: Environment Canada, Atmospheric Environmental Services

Table A.2. Total monthly precipitation for selected field sites.

Site	Year	Jan	Feb	Mar	Apr	May	June	July	Aug	Sept	Oct	Nov	Dec
-----total monthly precipitation (mm)-----													
Carman	1992	-	-	-	-	35.0	128.9	116.2	64.9	38.4	4.2	21.1	35.6
	1993	17.6	0.2	9.0	17.4	70.0	120.0	152.8	114.0	28.8	30.6	25.0	13.9
	1994	12.9	5.4	13.6	11.2	39.1	53.5	48.0	102.6	54.8	-	-	-
Glenlea	1992	-	-	-	39.7	28.0	106.6	86.8	69.2	70.0	3.8	33.1	44.2
	1993	16.4	0.0	7.6	22.8	41.0	72.8	246.0	160.0	31.8	32.6	15.8	20.2
	1994	11.2	7.2	3.2	10.0	150.7	94.7	96.7	100.8	73.2	-	-	-

Source: Environment Canada, Atmospheric Environmental Services

Table A.3. Management practices used for termination of alfalfa stands at field sites established at Glenlea and Carman in 1992 and at Winnipeg and a second Carman site in 1993.

Termination treatment		1992 <sup>†</sup>				1993 <sup>‡</sup>			
Time	Method	Glenlea-92		Carman-92		Carman-93		Winnipeg-93	
		Date	Operation	Date	Operation	Date	Operation	Date	Operation
<i>First cut of alfalfa:</i>		<i>June 20</i>		<i>June 28</i>		<i>July 9</i>			
Early	herbicide+tillage	July 21	2.5 L ha <sup>-1</sup>	July 22	2.5 L ha <sup>-1</sup>				
		Aug. 28	disc (2x)	Aug. 29	disc (2x)				
		Oct. 1	harrow	Oct. 2	harrow				
	herbicide	July 21	5 L ha <sup>-1</sup>	July 22	5 L ha <sup>-1</sup>	July 26	4.1 L ha <sup>-1</sup>		
		Sept. 8	2.5 L ha <sup>-1</sup>	Sept. 10	2.5 L ha <sup>-1</sup>	Sept 10	4.1 L ha <sup>-1</sup>		
	tillage	July 10	deep till	July 22	deep till	July 30	deep till (2x), disc (2x)		
		July 21	deep till	Aug. 1	deep till	Sept 11	disc (2x)		
		Aug. 11	deep till (2x)	Aug. 15	deep till (2x)	Oct 8	disc, harrow (2x)		
		Aug. 28	disc	Aug. 29	disc				
		Oct. 1	disc, harrow	Oct. 2	disc, harrow				
<i>Second cut of alfalfa:</i>		<i>July 31</i>		<i>Aug 10</i>		<i>Aug 6</i>		<i>Aug 5</i>	
Late	herbicide+tillage	Sept. 8	2.5 L ha <sup>-1</sup>	Sept. 10	2.5 L ha <sup>-1</sup>				
		Oct. 1	disc (2x), harrow	Oct. 2	disc (2x), harrow				
	herbicide	Sept. 8	5 L ha <sup>-1</sup>	Sept. 10	5 L ha <sup>-1</sup>	Sept 10	4.1 L ha <sup>-1</sup>	Aug 26	4.1 L ha <sup>-1</sup>
								Sept 11	4.1 L ha <sup>-1</sup>
	tillage	Aug. 11	deep till	Aug. 15	deep till	Sept 11	deep till (2x), disc (2x)	Sept 9	rototill (2x)
		Aug. 28	disc	Aug. 29	disc	Oct 8	disc (2x), harrow (2x)	Oct 12	rototill (1x)
		Sept. 14	disc	Sept. 14	disc				
		Oct. 1	disc, harrow	Oct. 2	disc, harrow				

<sup>†</sup> In 1992, Roundup was applied at all sites.

<sup>‡</sup> In 1993, herbicide applications consisted of a combination of 1.85 L ha<sup>-1</sup> Roundup, 1.25 L ha<sup>-1</sup> Banvel and 1 L ha<sup>-1</sup> 2,4-D.

Table A.4. Soil NO<sub>3</sub><sup>-</sup>-N concentrations measured periodically following alfalfa termination as influenced by method and time of alfalfa termination at the Glenlea-92 site.

Year	Sampling Time	Treatment		Soil depth (cm)				
		Time	Method	0-15	15-30	30-60	60-90	90-120
-----mg NO <sub>3</sub> <sup>-</sup> -N kg <sup>-1</sup> soil-----								
1993	Spring	Early	herbicide	13.8	15.7	8.5	3.9	2.1
			herb.+till	30.0	21.2	3.7	1.3	1.5
			tillage	30.4	26.0	5.8	1.7	2.0
		Late	herbicide	15.3	12.2	2.7	0.9	0.7
			herb.+till	23.1	12.3	2.6	1.2	0.8
			tillage	24.9	17.6	4.6	1.3	1.7
	Midseason	Early	herbicide	6.7	3.2	4.2	n.d.	n.d.
			herb.+till	9.6	6.5	3.5	n.d.	n.d.
			tillage	7.7	9.9	5.9	n.d.	n.d.
		Late	herbicide	3.5	3.0	1.3	n.d.	n.d.
			herb.+till	5.1	2.5	1.5	n.d.	n.d.
			tillage	5.4	3.0	3.9	n.d.	n.d.
	Fall	Early	herbicide	7.2	5.4	3.8	2.1	1.5
			herb.+till	9.0	5.6	4.8	2.6	1.4
			tillage	8.4	5.9	6.4	4.0	2.2
		Late	herbicide	5.6	3.9	1.9	1.2	1.3
			herb.+till	7.0	3.6	2.3	1.2	0.8
			tillage	7.9	4.0	3.7	2.2	1.4
		Spring	herbicide	5.6	4.5	2.3	0.9	0.8
1994	Spring	Early	herbicide	13.3	10.2	5.8	2.7	1.8
			herb.+till	14.5	12.8	5.7	2.9	2.1
			tillage	19.4	16.5	9.5	4.9	2.2
		Late	herbicide	11.1	8.6	4.6	1.9	1.4
			herb.+till	14.4	9.8	4.5	2.0	1.3
			tillage	16.4	14.1	5.8	3.1	2.1
		Spring	herbicide	12.8	10.3	4.5	1.6	0.9
	Fall	Early	herbicide	11.0	3.4	1.7	1.0	1.6
			herb.+till	15.4	6.3	2.7	1.7	2.1
			tillage	13.2	4.8	2.6	1.8	2.1
		Late	herbicide	17.1	4.9	2.6	1.6	1.5
			herb.+till	13.1	4.6	1.8	1.3	1.1
			tillage	11.3	3.9	1.9	1.1	1.2
		Spring	herbicide	15.5	5.2	2.3	2.0	1.7

n.d. indicates that the value was not determined.

Table A.5. Soil NO<sub>3</sub><sup>-</sup>-N concentrations measured periodically following alfalfa termination as influenced by method and time of alfalfa termination at the Carman-92 site.

Year	Sampling Time	Treatment		Soil depth (cm)					
		Time	Method	0-15	15-30	30-60	60-90	90-120	
-----mg NO <sub>3</sub> <sup>-</sup> -N kg <sup>-1</sup> soil-----									
1993	Spring	Early	herbicide	27.8	21.0	6.6	2.5	2.1	
			herb.+till	36.0	22.7	6.5	3.8	2.9	
			tillage	32.2	26.8	7.5	2.9	1.9	
		Late	herbicide	14.2	9.5	3.1	2.0	1.7	
			herb.+till	19.1	13.1	5.0	3.7	2.1	
			tillage	24.3	16.3	5.1	3.9	2.6	
		Midseason	Early	herbicide	1.1	1.2	1.1	n.d.	n.d.
				herb.+till	2.6	3.7	1.9	n.d.	n.d.
				tillage	1.7	3.9	2.1	n.d.	n.d.
			Late	herbicide	0.4	0.4	0.4	n.d.	n.d.
				herb.+till	1.1	0.8	0.5	n.d.	n.d.
				tillage	1.0	1.3	1.2	n.d.	n.d.
	Fall	Early	herbicide	23.9	10.8	4.1	3.1	1.8	
			herb.+till	21.8	11.8	5.8	3.4	1.9	
			tillage	18.4	10.1	4.6	3.2	1.5	
		Late	herbicide	16.2	7.0	4.0	1.4	0.9	
			herb.+till	21.9	10.5	3.9	2.4	1.6	
			tillage	21.2	10.8	3.2	2.6	1.4	
		Spring	herbicide	18.7	7.5	2.4	1.2	1.0	
1994	Spring	Early	herbicide	20.6	12.8	6.3	4.1	3.4	
			herb.+till	22.2	14.7	7.4	4.9	3.6	
			tillage	20.6	12.6	6.5	4.9	3.4	
		Late	herbicide	19.3	12.4	4.7	3.3	3.2	
			herb.+till	21.7	14.1	5.8	3.6	3.0	
			tillage	23.0	14.1	6.6	5.2	3.8	
		Spring	herbicide	21.5	13.6	5.6	3.8	3.0	
	Fall	Early	herbicide	11.1	4.8	1.5	0.9	1.2	
			herb.+till	12.4	3.5	1.7	1.5	2.0	
			tillage	8.3	3.7	1.8	1.6	2.1	
		Late	herbicide	10.3	3.5	1.4	0.9	1.0	
			herb.+till	10.2	3.8	1.8	1.0	1.2	
			tillage	11.0	4.3	1.3	0.9	1.0	
		Spring	herbicide	11.5	4.4	1.8	1.4	1.6	

n.d. indicates that the value was not determined.

Table A.6. Soil  $\text{NO}_3^-$ -N concentrations measured periodically following alfalfa termination as influenced by method and time of alfalfa termination at the Carman-93 site.

Year	Sampling Time	Treatment		Soil depth (cm)					
		Time	Method	0-15	15-30	30-60	60-90	90-120	
-----mg NO <sub>3</sub> <sup>-</sup> -N kg <sup>-1</sup> soil-----									
1993	Termination	Late	herbicide	2.5	1.6	0.6	0.3	0.3	
			tillage	1.9	1.4	0.5	0.3	0.1	
	Fall	Early	herbicide	13.5	6.0	2.7	0.9	0.8	
			tillage	33.4	14.1	5.5	2.2	1.5	
		Late	herbicide	6.3	1.8	0.9	0.6	0.5	
			tillage	18.7	3.5	1.6	0.8	0.6	
		Spring	herbicide	2.9	1.2	0.7	0.4	0.4	
	1994	Spring	Early	herbicide	9.7	9.6	6.4	3.8	2.8
				tillage	33.5	22.0	12.7	8.3	6.1
Late			herbicide	10.3	6.5	3.6	2.6	1.7	
			tillage	19.9	14.7	7.8	6.5	5.1	
Spring		herbicide	7.1	5.3	2.4	2.0	2.0		
Midseason		Early	herbicide	4.2	2.7	1.3	n.d.	n.d.	
			tillage	9.2	11.7	3.2	n.d.	n.d.	
		Late	herbicide	5.9	3.2	1.6	n.d.	n.d.	
			tillage	8.3	7.1	2.1	n.d.	n.d.	
		Spring	herbicide	8.1	3.9	1.3	n.d.	n.d.	
Fall		Early	herbicide	18.7	5.3	2.3	1.4	1.7	
			tillage	12.8	13.8	6.0	1.9	1.5	
		Late	herbicide	19.8	6.2	1.7	1.1	0.7	
			tillage	15.7	5.7	1.8	0.9	1.1	
		Spring	herbicide	18.2	5.5	1.4	0.9	0.8	

n.d. indicates that the value was not determined.

Table A.7. Soil  $\text{NO}_3^-$ -N concentrations measured periodically following alfalfa termination as influenced by method of alfalfa termination at the Winnipeg-93 site.

Year	Sampling Time	Treatment		Soil depth (cm)				
		Time	Method	0-15	15-30	30-60	60-90	90-120
-----mg NO <sub>3</sub> <sup>-</sup> -N kg <sup>-1</sup> soil-----								
1993	Termination	Late	herbicide tillage	4.5	2.6	2.4	0.9	0.7
				2.6	2.0	2.2	1.0	0.7
	Fall	Late	herbicide tillage	5.6	4.7	2.3	1.1	0.8
				10.7	5.0	3.3	1.5	1.0
1994	Spring	Late	herbicide tillage	16.6	13.7	5.7	2.6	1.9
				14.1	11.0	7.5	4.1	1.9
	Midseason	Late	herbicide tillage	2.8	0.9	0.4	n.d.	n.d.
				2.6	0.9	0.3	n.d.	n.d.
	Fall	Late	herbicide tillage	5.2	3.4	2.1	0.7	0.5
				6.3	3.5	1.5	0.8	0.8

n.d. indicates that the values were not determined.

Table A.8. Plant tissue sampling schedule and physiological development of wheat at time of sampling.

Sampling time	1993		1994	
	Glenlea-92	Carman-92	Carman-93	Winnipeg-93
Seeding date	May 13	May 6	May 13	May 10
1	June 3 (1) <sup>†</sup>	May 27 (1)	June 6 (n.d.)	June 3 (3)
2	June 17 (3-4)	June 9 (4-5)	June 20 (n.d.)	June 16 (7-8)
3	July 5 (8-9)	June 24 (10)	June 29 (10.2-10.3)	June 29 (10.3)
4	July 14 (10.4)	July 8 (10.5.1-10.5.2)	July 18 (10.5.4-11.1)	July 14 (10.5.4-11.1)
5	August 3 (n.d.)	July 21 (11.1)	August 2 (soft dough)	July 28 (soft dough)
6	August 13 (soft dough)	August 6 (soft dough)	-	-
Mature harvest	September 1	August 25	August 18	August 12

<sup>†</sup> the values given in parenthesis indicate the Feekes developmental stage of wheat at time of sampling; n.d. indicates that Feekes development stage was not determined at time of sampling.



Table A.9. Dry matter accumulations of wheat during the growing season and at crop maturity at the Glenlea-92 site in 1993 and 1994.

Treatment		Sampling Times in 1993							Sampling times in 1994	
Time	Method	1	2	3	4	5	6	Maturity	Soft dough	Maturity
-----kg ha <sup>-1</sup> -----										
Early	Herbicide	40	388	2620	3863	6329	7858	6004	6973	5640
	Herb.+till	53	368	2278	4076	5167	6056	5372	8495	6410
	Tillage	46	375	2357	4074	5506	5864	5952	8285	6972
Late	Herbicide	41	367	1842	3743	4847	5206	4809	6953	5539
	Herb.+till	36	365	1883	3445	5072	5211	4683	7795	6366
	Tillage	45	450	2529	4008	5039	6239	5189	8362	6091
Spring	Herbicide	n.d.	n.d.	1684	3139	4670	5454	4372	7642	5927
Significance (P) <sup>§</sup>										
Time		0.27	0.70	0.007	0.20	0.006	0.002	0.003	0.63	0.37
Method		0.69	0.64	0.05 <sup>†</sup>	0.49	0.21	0.06	0.22	0.05 <sup>†</sup>	0.11
M*T		0.29	0.61	0.009	0.47	0.04	0.002	0.67	0.74	0.59
Contrasts										
Spring vs early herbicide		n.d.	n.d.	0.0001	0.05	0.0002	0.0001	0.0008	0.37	0.65
Spring vs late herbicide		n.d.	n.d.	0.41	0.09	0.62	0.60	0.30	0.36	0.54
Spring vs late and early herb.		n.d.	n.d.	0.004	0.04	0.008	0.02	0.009	0.30	0.54
C.V. (%)		27.1	26.1	12.3	12.7	9.6	11.2	11.0	13.3	14.4

<sup>†</sup> LSD for the effect of method is 281 kg ha<sup>-1</sup>.

<sup>‡</sup> LSD for the effect of method is 1140 kg ha<sup>-1</sup>.

<sup>§</sup> The degrees of freedom (df) for effects of time, method and M\*T are 1, 2, and 2, respectively. All contrasts are single df contrasts.

Table A.10. Dry matter accumulations of wheat during the growing season and at crop maturity at the Carman-92 site in 1993 and 1994.

Treatment		Sampling Times in 1993							Sampling times in 1994	
Time	Method	1	2	3	4	5	6	Maturity	Soft dough	Maturity
-----kg ha <sup>-1</sup> -----										
Early	Herbicide	151	903	3666	6493	9293	9728	9471	9596	6343
	Herb.+till	147	822	3237	5916	8883	9082	8262	9597	6392
	Tillage	151	777	3234	5537	7948	9364	7560	9526	5856
Late	Herbicide	137	711	3106	5446	7320	8831	8300	8662	5996
	Herb.+till	158	747	3169	5996	7896	9044	8410	9458	6591
	Tillage	124	780	3114	5851	8238	8689	8087	8751	6237
Spring	Herbicide	n.d.	n.d.	2313	4388	6600	8213	7923	9006	5990
Significance (P) <sup>§</sup>										
Time		0.04	0.002	0.007	0.26	0.05	0.12	0.55	0.04	0.70
Method		0.04 <sup>†</sup>	0.58	0.10	0.58	0.72	0.80	0.02 <sup>‡</sup>	0.42	0.19
M*T		0.007	0.01	0.05	0.06	0.10	0.56	0.05	0.47	0.31
Contrasts										
Spring vs early herbicide		n.d.	n.d.	0.0001	0.0001	0.0002	0.03	0.02	0.40	0.39
Spring vs late herbicide		n.d.	n.d.	0.0003	0.01	0.19	0.34	0.53	0.62	0.99
Spring vs early and late herb.		n.d.	n.d.	0.0001	0.0002	0.003	0.07	0.08	0.84	0.61
C.V. (%)		7.3	7.1	8.1	9.7	9.9	9.9	10.1	10.6	9.1

<sup>†</sup> LSD for the effect of method is 11 kg ha<sup>-1</sup>; n.d. indicates that the values were not determined.

<sup>‡</sup> LSD for the effect of method is 698 kg ha<sup>-1</sup>.

<sup>§</sup> The degrees of freedom (df) for effects of time, method and M\*T are 1, 2, and 2, respectively. All contrasts are single df contrasts.

Table A.11. Dry matter accumulations of wheat during the growing season and at crop maturity at the Carman-93 site in 1994.

Treatment		Sampling Times					
Time	Method	1	2	3	4	5	Maturity
-----kg N ha <sup>-1</sup> -----							
Early	Herbicide	365	1532	3016	7240	7640	6812
	Tillage	287	1498	2539	6986	9059	6080
Late	Herbicide	324	1203	2635	6244	7486	5262
	Tillage	208	1185	2332	6213	6820	5141
Spring	Herbicide	138	657	1052	3976	4820	3298
Significance (P) <sup>†</sup>							
Time		0.03	0.004	0.05	0.09	0.03	0.01
Method		0.002	0.75	0.01	0.77	0.42	0.30
M*T		0.42	0.93	0.51	0.82	0.04	0.45
Contrasts							
Spring vs early herbicide		0.0001	0.0001	0.0001	0.0004	0.007	0.0001
Spring vs late herbicide		0.0001	0.001	0.0001	0.005	0.01	0.004
Spring vs early and late herb.		0.0001	0.0001	0.0001	0.0004	0.004	0.0001
C.V. (%)		15.9	15.1	10.1	15.4	17.3	14.5

<sup>†</sup> The degrees of freedom (df) for effects of time, method and M\*T are 1, 1, and 1, respectively. All contrasts are single df contrasts.

Table A.12. Dry matter accumulations of wheat during the growing season and at crop maturity at the Winnipeg-93 site in 1994.

Treatment		Sampling Times					
Time	Method	1	2	3	4	5	Maturity
-----kg N ha <sup>-1</sup> -----							
Late	Herbicide	140	965	3605	6166	8293	6145
	Tillage	220	1043	3424	5636	8005	6234
Significance (P) <sup>†</sup>							
Method		0.07	0.64	0.23	0.05	0.52	0.65

<sup>†</sup> The effect of method has 1 df.

Table A.13. Nitrogen concentration in wheat top-growth during the growing season and in grain and straw at crop maturity at the Glenlea-92 site in 1993 and 1994.

Treatment		Sampling Times in 1993								Sampling times in 1994		
Time	Method	1	2	3	4	5	6	Grain	Straw	Soft dough	Grain	Straw
-----g N kg <sup>-1</sup> -----												
Early	Herbicide	50.8	55.1	35.1	27.9	15.4	16.6	34.1	9.4	13.6	30.4	6.9
	Herb.+till	56.8	59.8	35.4	24.9	15.5	15.9	35.1	6.1	14.1	30.8	8.7
	Tillage	56.8	57.1	36.4	24.6	15.9	16.9	34.4	9.6	14.7	31.6	8.7
Late	Herbicide	54.0	55.1	28.9	20.3	13.8	14.2	34.7	6.0	12.3	30.5	7.4
	Herb.+till	54.9	54.7	32.8	20.2	13.3	17.1	35.1	4.2	14.5	31.3	6.9
	Tillage	55.9	57.1	33.8	24.2	14.5	15.2	33.8	10.3	12.8	31.8	7.7
Spring	Herbicide	n.d.	n.d.	31.4	20.4	13.6	16.3	34.2	2.8	14.3	32.8	7.2
Significance (P) <sup>†</sup>												
Time		0.90	0.08	0.04	0.0001	0.006	0.20	0.97	0.35	0.21	0.55	0.17
Method		0.01 <sup>†</sup>	0.15	0.33	0.10	0.46	0.48	0.06	0.08	0.33	0.09	0.27
M*T		0.11	0.07	0.60	0.002	0.82	0.12	0.32	0.57	0.43	0.88	0.21
Contrasts												
Spring vs early herbicide		n.d.	n.d.	0.18	0.0001	0.06	0.81	0.85	0.02	0.58	0.04	0.84
Spring vs late herbicide		n.d.	n.d.	0.35	0.93	0.84	0.10	0.38	0.24	0.12	0.04	0.77
C.V. (%)		4.4	4.0	11.3	7.1	8.5	10.8	2.3	54.4	12.9	4.6	16.0

<sup>†</sup> The LSD for the effect of method is 2.6; n.d. indicates that the values were not determined.

<sup>‡</sup> The degrees of freedom (df) for effects of time, method and M\*T are 1, 2, and 2, respectively. All contrasts are single df contrasts.

Table A.14. Nitrogen concentration in wheat top-growth during the growing season and in grain and straw at crop maturity at the Carman-92 site in 1993 and 1994.

Treatment		Sampling Times in 1993								Sampling times in 1994		
Time	Method	1	2	3	4	5	6	Grain	Straw	Soft dough	Grain	Straw
-----g N kg <sup>-1</sup> -----												
Early	Herbicide	63.1	58.7	27.8	18.8	15.8	15.3	32.9	7.0	13.7	28.7	7.2
	Herb.+till	63.8	58.4	32.2	19.7	15.2	16.3	34.1	5.3	14.9	30.6	7.4
	Tillage	65.3	59.2	32.2	19.6	15.5	14.0	34.2	7.6	12.8	29.5	6.7
Late	Herbicide	60.4	51.6	24.5	15.5	11.9	13.1	31.4	7.8	13.5	28.1	5.9
	Herb.+till	62.2	54.9	25.8	17.4	13.2	15.4	33.1	4.4	13.9	28.9	7.0
	Tillage	63.1	58.6	27.6	17.4	15.1	15.4	33.3	7.9	14.5	29.9	5.8
Spring	Herbicide	n.d.	n.d.	24.4	15.4	13.1	12.7	32.1	2.5	12.9	28.8	6.3
Significance (P) <sup>†</sup>												
Time		0.001	0.001	0.001	0.009	0.001	0.41	0.001	0.96	0.86	0.43	0.04
Method <sup>‡</sup>		0.009	0.007	0.005	0.36	0.10	0.16	0.0009	0.40	0.64	0.11	0.12
M*T		0.001	0.003	0.33	0.85	0.07	0.13	0.61	0.94	0.33	0.46	0.60
Contrasts												
Spring vs early herbicide		n.d.	n.d.	0.02	0.03	0.005	0.04	0.12	0.20	0.50	0.81	0.17
Spring vs fall herbicide		n.d.	n.d.	1.00	0.92	0.37	0.77	0.16	0.14	0.60	0.38	0.53
C.V. (%)		1.6	2.7	6.8	11.2	8.0	11.6	2.2	80.6	12.9	3.8	13.4

<sup>†</sup> The degrees of freedom (df) for effects of time, method and M\*T are 1, 2, and 2, respectively. All contrasts are single df contrasts.

<sup>‡</sup> LSD's for the effect of method are 1.1, 1.6, 2.1 and 0.7 g N kg<sup>-1</sup> for sample times 1, 2 and 3 and grain (1993), respectively; n.d. indicates that the values was not determined.

Table A.15. Nitrogen concentration in wheat top-growth during the growing season and in grain and straw at crop maturity at the Carman-93 site in 1994.

Treatment		Sampling Times						
Time	Method	1	2	3	4	5	Grain	Straw
		-----g N kg <sup>-1</sup> -----						
Early	Herbicide	51.4	39.5	26.8	16.9	14.3	33.7	6.8
	Tillage	56.0	44.1	30.5	17.7	15.1	34.5	6.3
Late	Herbicide	52.6	42.2	28.9	17.5	16.0	33.6	7.9
	Tillage	55.8	44.0	31.4	16.9	14.6	33.3	6.9
Spring	Herbicide	50.0	42.5	32.8	18.8	16.9	33.7	8.4
Significance (P) <sup>†</sup>								
Time		0.55	0.16	0.13	0.90	0.55	0.18	0.08
Method		0.0007	0.005	0.008	0.91	0.73	0.65	0.13
M*T		0.41	0.14	0.52	0.34	0.28	0.22	0.57
Contrasts								
Spring vs early herbicide		0.25	0.02	0.0006	0.06	0.15	0.99	0.06
Spring vs late herbicide		0.05	0.53	0.01	0.18	0.62	0.97	0.39
C.V. (%)		3.1	4.3	6.1	7.1	15.2	3.6	15.8

<sup>†</sup> The degrees of freedom (df) for effects of time, method and M\*T are 1, 1, and 1, respectively. All contrasts are single df contrasts.

Table A.16. Nitrogen concentration in wheat top-growth during the growing season and in grain and straw at crop maturity at the Winnipeg-93 site in 1994.

Treatment		Sampling Times						
Time	Method	1	2	3	4	5	Grain	Straw
		-----g N kg <sup>-1</sup> -----						
Late	Herbicide	50.7	26.8	18.4	11.7	9.1	24.8	3.2
	Tillage	54.0	26.7	17.7	12.2	9.2	25.3	3.2
Significance (P) <sup>†</sup>								
Method		0.09	0.99	0.76	0.58	n.d.	0.48	0.97

<sup>†</sup> The effect of method has 1 df.



Table A.17. Harvest index of spring wheat at crop maturity during the first and second growing seasons following alfalfa termination.

Time	Method	1st growing season following alfalfa termination				2nd season after alfalfa termination	
		Glenlea-92	Carman-92	Carman-93	Winnipeg-93	Glenlea-92	Carman-92
-----% of total dry matter yield at crop maturity comprised of grain-----							
Early	herbicide	30.6	30.9	41.4	n.d.	42.6	38.8
	herb.+till	32.8	32.3	n.d.	n.d.	41.1	37.9
	tillage	31.6	32.8	44.8	n.d.	40.6	40.5
Late	herbicide	39.1	33.8	45.9	46.7	44.7	40.2
	herb.+till	36.2	33.3	n.d.	n.d.	45.1	37.2
	tillage	36.4	31.4	43.2	46.6	44.1	39.2
Spring	herbicide	38.8	33.7	47.7	n.d.	45.2	38.7
LSD <sub>(0.05)</sub> (method)		4.1	3.4	4.2	n.d.	3.2	3.1
Significance (P) <sup>†</sup>							
Rep		0.01	0.06	0.11	0.41	0.001	0.02
Time		0.003	0.91	0.45	n.d.	0.02	0.26
Method		0.90	0.53	0.83	0.94	0.69	0.84
M*T		0.40	0.42	0.13	n.d.	0.80	0.65
Contrasts							
Spring vs late herbicide		0.90	0.96	0.60	n.d.	0.82	0.48
Spring vs early and late herbicide		0.10	0.52	0.18	n.d.	0.41	0.65
C.V. (%)		10.5	10.1	10.5	3.3	6.8	7.5

<sup>†</sup> At Glenlea-92 and Carman-92, degrees of freedom (df) for effects of rep, time, method and M\*T were 3, 1, 2 and 2 respectively. At Carman-93, df for effects of rep, time, method and M\*T were 3, 1, 1, and 1, respectively. At Winnipeg-93, df for effects of rep and method were 3 and 1, respectively. n.d. indicates that the value was not determined.

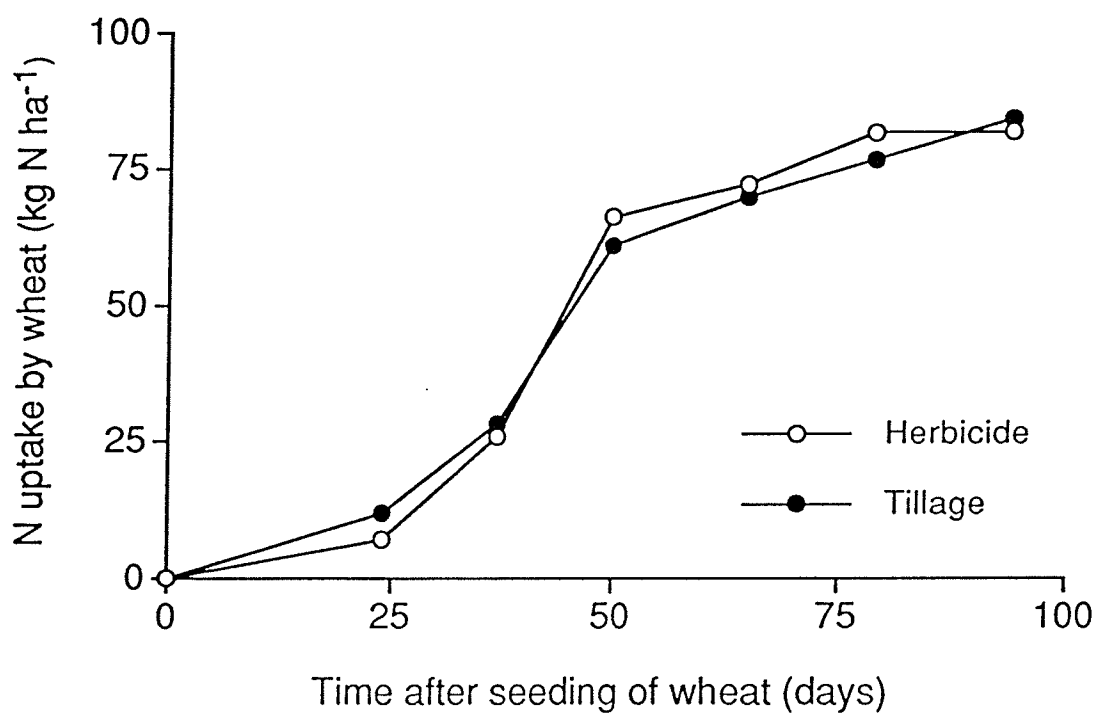


Fig. A.1. Nitrogen accumulations by wheat at Winnipeg-93 during the initial growing season following alfalfa termination by herbicide application or tillage.

## Appendix B

Table B.1. Summary of the statistical significance (P value) of effect of residue placement (surface, incorporated) and termination method (chemical, mechanical) on daily and cumulative CO<sub>2</sub> evolution from terminated alfalfa measured at 17 sampling times during a 95 d period following alfalfa termination

Days of incubation	mg C day <sup>-1</sup>			Cumulative C (mg C)		
	Placement <sup>†</sup>	Method	P*M	Placement	Method	P*M
1	0.0005	0.01	0.50	0.0005	0.01	0.50
2	0.0005	0.002	0.24	0.0004	0.003	0.31
4	0.02	0.37	0.66	0.004	0.08	1.00
5	0.14	0.44	0.18	0.01	0.14	0.65
6	0.0001	0.0001	0.01	0.007	0.08	0.55
9	0.02	0.09	0.87	0.004	0.05	0.56
12	0.09	0.01	0.64	0.004	0.03	0.55
15	0.30	0.02	0.72	0.005	0.02	0.62
18	0.10	0.003	0.43	0.005	0.02	0.68
23	0.02	0.01	0.21	0.005	0.01	0.82
26	0.07	0.003	0.14	0.005	0.01	0.91
32	0.006	0.04	0.27	0.004	0.01	0.98
39	0.001	0.07	0.63	0.003	0.01	0.94
46	0.004	0.25	0.33	0.003	0.01	0.96
53	0.007	0.71	0.26	0.003	0.02	0.87
74	0.05	0.19	0.42	0.004	0.08	0.75
95	0.69	0.48	0.97	0.01	0.12	0.80

<sup>†</sup> The effects of placement, method and P\*M each have 1 df.

Table B.2. Summary of the statistical significance (P value) of effect of residue placement (surface, incorporated) and termination method (chemical, mechanical) on daily and cumulative  $\text{NH}_3$  evolution from terminated alfalfa measured at 17 sampling times during a 95 d period following alfalfa termination

Days of incubation	$\mu\text{g N day}^{-1}$			Cumulative N ( $\mu\text{g N}$ )		
	Placement <sup>†</sup>	Method	P*M	Placement	Method	P*M
1	0.87	0.36	0.76	0.87	0.36	0.76
2	0.07	0.04	0.09	0.10	0.05	0.11
4	0.05	0.09	0.24	0.05	0.06	0.17
5	0.04	0.10	0.12	0.03	0.06	0.11
6	0.05	0.10	0.10	0.05	0.09	0.12
9	0.0002	0.34	0.35	0.001	0.09	0.11
12	0.0003	0.07	0.07	0.0002	0.25	0.29
15	0.0002	0.04	0.03	0.0001	0.41	0.46
18	0.0001	0.01	0.009	0.0001	0.50	0.57
23	0.0008	0.15	0.14	0.0001	0.64	0.72
26	0.0009	0.34	0.74	0.0001	0.67	0.73
32	0.0002	0.14	0.39	0.0001	0.72	0.76
39	0.0001	0.01	0.19	0.0001	0.77	0.78
46	0.13	0.19	0.97	0.0001	0.81	0.77
53	0.002	0.37	0.47	0.0001	0.81	0.78
74	0.98	0.69	0.68	0.0001	0.77	0.79
95	0.12	0.16	0.48	0.0001	0.80	0.80

<sup>†</sup> The effects of placement, method and P\*M each have 1 df.

## Appendix C

Table C.1. Nitrogen accumulations by four consecutive barley crops established following alfalfa termination as influenced by method of alfalfa termination (chemical, mechanical) and residue placement (surface, incorporated)

Treatment		N uptake during individual sampling periods (mg)				Cumulative N uptake (mg)				Soil NO <sub>3</sub> <sup>-</sup> -N
Method	Placement	1	2	3	4	25 d	50 d	65 d	125 d	125 d
		-----mg N pot <sup>-1</sup> -----				-----mg N pot <sup>-1</sup> -----				---mg kg <sup>-1</sup> ---
Herbicide	Incorporated	67.0	19.6	2.1	17.7	66.98	86.62	88.70	104.63	1.16
	Surface	23.6	15.2	3.2	18.4	23.57	38.73	41.97	60.37	1.50
Tillage	Incorporated	63.4	17.7	2.5	19.8	63.44	81.15	83.68	103.51	1.29
	Surface	12.8	14.4	3.4	11.9	12.83	27.23	30.61	53.47	1.20
Significance (P) <sup>†</sup>										
Method		0.02	0.55	0.83	0.18	0.02	0.09	0.35	0.60	0.61
Placement		0.001	0.11	0.45	0.42	0.0001	0.0001	0.0003	0.0006	0.47
M*P		0.20	0.79	0.93	0.62	0.20	0.52	0.45	0.70	0.22

<sup>†</sup> The effects of placement, method and M\*P each have 1 df.

Table C.2. Soil  $\text{NO}_3^-$  accumulations in uncropped pots 125 d after the termination of alfalfa as influenced by termination method and method of residue placement.

Treatment		Soil $\text{NO}_3^-$ -N concentrations
Method	Placement	
		-----mg $\text{NO}_3^-$ -N $\text{kg}^{-1}$ -----
Herbicide	Incorporated	13.07
	Surface	6.33
Tillage	Incorporated	10.54
	Surface	5.80
Significance (P) <sup>†</sup>		
Method		0.34
Placement		0.004
M*P		0.53

<sup>†</sup> The effects of placement, method and M\*P each have 1 df.



Table C.3. Effect of residue particle size on net N mineralization of alfalfa leaf, stem and root tissue after 5 and 8 wk of incubation.

Treatment		Net mineralization <sup>†</sup>		% Recovery <sup>‡</sup>	
Residue	Size	Wk 5	Wk 8	Wk 5	Wk 8
mg inorganic N pot <sup>-1</sup>					
Leaf	ground	88.8	82.7	56.0	52.2
	whole	90.0	81.8	56.8	51.6
Stem	ground	12.9	15.6	19.0	23.0
	1.5 cm	17.5	30.7	25.7	45.1
	5 cm	18.7	16.3	27.5	24.0
Root	ground	14.4	18.6	19.1	24.8
	1.5 cm	8.3	15.9	11.0	21.1
	5 cm	11.7	14.2	15.5	18.8
Significance (P) <sup>§</sup>					
Treatment		0.0001	0.0001	0.0001	0.005
Contrasts					
Leaf 'ground vs whole'		0.79	0.90	0.87	0.95
Stem 'ground vs unground'		0.20	0.20	0.08	0.17
Root 'ground vs unground'		0.28	0.55	0.17	0.56

<sup>†</sup> Net mineralization=[Soil NO<sub>3</sub><sup>-</sup>+NH<sub>4</sub><sup>+</sup>]<sub>sample at Week X</sub>-[Soil NO<sub>3</sub><sup>-</sup>+NH<sub>4</sub><sup>+</sup>]<sub>soil control at Week X</sub> in mg N pot<sup>-1</sup>

<sup>‡</sup> % recovery=[Net mineralization (mg N pot<sup>-1</sup>)/Alfalfa N added (mg N)]\*100

<sup>§</sup> The df for the effect of treatment was 7. All contrasts were single df contrasts.

## Appendix D.

Table D.1. Mineralizable C and N in bulk soil in which alfalfa was grown (Mean of 3 replicates. Roots were removed prior to incubation.)

	Wk 1	Wk 2	Wk 4	Wk 8	Wk 12
N mineralization (mg N kg <sup>-1</sup> soil) <sup>†</sup>	2.5	5.2	9.0	13.1	18.6
%NDFL <sup>‡</sup>	21	19	19	13	12
Cumulative C (mg C kg <sup>-1</sup> soil)	25.8	55.5	107.6	155.6	167.8

<sup>†</sup> Net N mineralization calculated as [total inorganic N (post-incubation)] - [total inorganic N (pre-incubation)].

<sup>‡</sup> Calculated using the <sup>15</sup>N abundance in fine roots of alfalfa.

Table D.2. The C, N and  $^{15}\text{N}$  concentration of barley, residue and soil components following the application of alfalfa top-growth terminated by herbicide application and retained on the soil surface or terminated by tillage and incorporated into the soil.

Component		Herbicide					Tillage				
		Mass (g)	%N	%C	$^{15}\text{N}$ atom %	%NDFL	Mass (g)	%N	%C	$^{15}\text{N}$ atom %	%NDFL
Barley	top-growth	1.61	2.007	40.8	0.4146	1.28	1.36	1.779	40.7	0.8474	12.83
Residue	fine root	8.50	0.643	12.8	0.4182	1.38	5.72	0.881	20.6	0.7988	11.95
	intermediate root	0.40	2.282	29.3	0.3925	0.69	1.18	3.125	42.9	0.4053	0.91
	taproot	1.37	2.819	39.6	0.3723	0.06	2.27	2.220	41.7	0.3739	0.08
	inc. stem	0	n/a	n/a	n/a	n/a	0.67	0.643	46.1	2.6159	66.63
	surface	1.66	1.999	43.3	3.8960	101.3	n/a	n/a	n/a	n/a	n/a
Soil	Bulk	2913.9	0.067	0.634	0.3727	0.12	2898.12	0.067	0.645	0.3893	0.56
	Rhizosoil										
	closely associated	88.9	0.049	0.454	0.3735	0.14	104.54	0.049	0.475	0.5101	4.03
	loosely adhering	119.2	0.072	0.681	0.3725	0.11	119.34	0.069	0.671	0.4185	1.40
	Light fraction	11.79	1.178	21.3	0.3777	0.22	12.87	1.104	19.7	0.3893	0.53
	root-free	n/a	0.076	0.676	0.3731	0.13	n/a	0.071	0.670	0.3849	0.44

n/a indicates that the measurement is not applicable.

Table D.3. The C, N and  $^{15}\text{N}$  concentration of barley, residue and soil components following termination of  $^{15}\text{N}$ -labelled alfalfa roots by herbicide application or tillage.

Component		Herbicide					Tillage				
		Mass (g)	%N	%C	$^{15}\text{N}$ atom %	%NDFL	Mass (g)	%N	%C	$^{15}\text{N}$ atom %	%NDFL
Barley	top-growth	1.006	1.682	40.9	1.8972	42.40	1.308	1.536	40.4	1.2812	25.30
Residue	fine root	3.843	0.917	23.5	2.0876	47.69	3.952	0.791	22.5	1.3871	28.24
	intermediate root	0.379	3.348	44.2	3.7279	93.23	0.327	1.978	28.7	3.4631	85.88
	taproot	1.297	3.007	42.7	3.9059	98.18	0.844	1.910	41.19	3.1285	76.59
Soil	Bulk	3011.74	0.065	0.636	0.4002	0.88	2965.48	0.063	0.624	0.3929	0.68
	Rhizosoil										
	closely associated	55.38	0.070	0.663	0.6331	7.35	63.36	0.065	0.634	0.4896	3.36
	loosely adhering	54.88	0.066	0.661	0.4559	2.43	93.16	0.066	0.669	0.4194	1.41
	Light fraction	12.62	1.256	21.5	0.4550	2.36	12.04	1.239	21.9	0.4184	1.34
	root-free	n/a	0.069	0.642	0.3908	0.62	n/a	0.069	0.655	0.3906	0.61

n/a indicates that the measurement is not applicable.

Table D.4. Distribution of  $^{15}\text{N}$  excess in barley top-growth, residue and soil components following termination of alfalfa by herbicide application or simulated tillage.

Component	Labelled alfalfa top-growth		Labelled alfalfa root	
	Herbicide	Tillage	Herbicide	Tillage
-----% of $^{15}\text{N}$ present-----				
Barley top-growth	1.1	10.1	9.7	10.2
Residue				
surface	89.4	0	0	0
incorporated stem	0	14.1	0	0
fine root	1.8	23.1	20.2	16.8
intermediate root	0.3	1.1	11.8	11.0
taproot	-0.05	-0.14	33.3	26.5
Total	91.4	38.2	65.3	54.4
Soil				
bulk	7.1	39.0	20.9	30.7
loosely adhering	0.3	4.3	1.1	2.0
closely associated	0.2	8.4	2.9	2.7
Total	7.5	51.7	25.0	35.4

Table D.5. Effect of alfalfa termination by herbicide application and simulated tillage on mineralizable N and  $^{15}\text{N}$ . Mineralizable N and  $^{15}\text{N}$  of the bulk soil was determined following harvest of a subsequent barley crop.

Alfalfa residue	Termination method	Mineralizable N <sup>†</sup>	
		--mg N kg <sup>-1</sup> soil--	--% of $^{15}\text{N}$ excess in soil--
Shoot	herbicide	19.9	34.2
	tillage	17.7	16.1
Root	herbicide	13.3	8.7
	tillage	13.2	19.0

<sup>†</sup> Replicate and termination treatment did not have a statistically significant effect on any of the parameters measured. The df for effects of rep and termination method were 2 and 1, respectively.

Table D.6. Light fraction N (LF N) and %NDFL in bulk soil samples in which <sup>15</sup>N-labelled alfalfa top-growth or root tissue was terminated by herbicide application or tillage.

Termination method	Labelled alfalfa top-growth <sup>†</sup>			Labelled alfalfa root		
	LF N	LF N <sub>(alf)</sub> <sup>‡</sup>	%NDFL <sup>§</sup>	LF N	LF N <sub>(alf)</sub>	%NDFL
	--mg N cylinder <sup>-1</sup> --			--mg N cylinder <sup>-1</sup> --		
Herbicide	143	0.29	0.22	163	3.9	2.4
Tillage	147	0.76	0.53	154	2.1	1.3

<sup>†</sup> LF N and LF N<sub>(alf)</sub> refer to total light fraction N and light fraction N derived from alfalfa, respectively.

<sup>‡</sup> Tillage significantly (P=0.03) increased the mg LF N derived from alfalfa top-growth.

<sup>§</sup> Tillage significantly (P=0.01) increased the % NDFL in alfalfa top-growth treatments. Termination treatment did not affect any other measures taken.