SHORT TERM ALFALFA STANDS IN CROPPING SYSTEMS: BENEFITS RELATED TO NITROGEN

A Thesis
Submitted to the Faculty of
Graduate Studies
by

David J. Kelner

In Partial Fulfillment of the
Requirements for the Degree of

Master of Science

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University of Manitoba
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SHORT TERM ALFALFA STANDS IN CROPPING SYSTEMS:
BENEFITS RELATED TO NITROGEN

BY

DAVID J. KELNER

A Thesis submitted to the Faculty of Graduate Studies of the University of Manitoba in partial fulfillment of the requirements for the degree of

MASTER OF SCIENCE

ABSTRACT

Kelner, David J. M.Sc., The University of Manitoba, May, 1994. **Short Term Alfalfa Stands in Cropping Systems: Benefits Related to Nitrogen.** Major Professor: Dr. J.K. Vessey, Department of Plant Science.

The inclusion of alfalfa (*Medicago sativa* L.) in a cropping sequence has long been recognized as a means of providing nitrogen for the subsequent crops in a rotation. Relatively little information is available, however, concerning the nitrogen dynamics of short term alfalfa stands. Field experiments were conducted in 1991 and 1992 to examine growth and N_2 fixation characteristics of a non-dormant, "annual" alfalfa (cv. Nitro) intended as a one year hay source and fall plow-down green manure crop. Additional studies were performed to develop a basic nitrogen budget for one to three years of alfalfa in a crop rotation. Nitro was compared with two dormant alfalfas in 1991, and to three dormant cultivars as well as another non-dormant alfalfa in 1992. Nitro yielded similarly to the other cultivars at the summer hay harvests, but produced significantly more herbage for soil incorporation in the fall relative to the dormant alfalfas (25% average increase in 1991, 39% increase in 1992). The quantities of N_2 fixed were also similar among treatments at the hay harvests, but were greater in Nitro in the fall relative to the dormant cultivars. Nitro and the dormant alfalfas added an average 151.9 and 53.4 kg ha^{-1} of fixed nitrogen to the soil in the fall of 1992, respectively. Nitro was not significantly different from CUF 101, the other non-dormant cultivar tested, in any of the measured parameters. Studies with established alfalfa demonstrated that the total seasonal amount of N_2 fixed increased with each additional year of alfalfa, ranging from 173 kg N ha^{-1} for the seedling year to a high of 466 kg N ha^{-1} for the third year stand. The estimated net soil N balance was 84, 148 and 137 kg N ha^{-1} for first, second and third year alfalfa, respectively. Total soil N measurements indicated that as much as 2510 kg N ha^{-1} could be added to the soil after three years of alfalfa.

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Mom, Dad, and Uncle Joe, for all their moral and financial support.

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Suzanne, for being there when I needed her.

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1. Introduction

Alfalfa (*Medicago sativa* L.) has long been recognized as a source of nitrogen for the subsequent crops in a rotation. The addition of large amounts of symbiotically fixed nitrogen to the soil resource has the potential to significantly reduce the nitrogen fertilizer requirements of the crops following alfalfa in a sequence.

Despite the potential for reducing N inputs by rotating crops with alfalfa, little is grown for strictly rotational reasons. Farms with substantial livestock components have large, steady requirements for high quality forage. A certain amount of the land resource must be designated to forage at any one time. A recent survey of Manitoba and Eastern Saskatchewan farmers showed that the average length of pure alfalfa stands was 6 years (Katepa-Mupondwa et al., 1993). As alfalfa can be difficult to remove and establish, stands are usually maintained as long as they remain productive and satisfy the forage requirements. The rotational benefits of alfalfa are often overlooked when considering stand removal. Katepa-Mupondwa et al. (1993) found that only 12% of the surveyed producers worked up their alfalfa stands for rotational reasons. Working up stands more frequently would mean that more land would be "treated" with alfalfa more often, while still maintaining the same level of forage production. The number of acres receiving the rotational benefits of alfalfa would increase with shorter stand lengths. The need therefore exists to identify a stand length such that the benefits of alfalfa are optimized before rotating the crop to a different part of the farm's land base.

When considering the nitrogen related benefits of alfalfa, determinations of optimum stand lengths should not be based solely on the amounts of residual N available for subsequent crops. The amounts of nitrogen derived from atmospheric or soil sources, and their partitioning into harvested forage or incorporated green manure, are all factors that contribute to the nitrogen dynamics of alfalfa-based cropping systems. Very little research exists that examines nitrogen fixation and nitrogen partitioning in alfalfa stands of increasing age (Heichel et al., 1984; Wivstad et al., 1987). A better understanding of the N dynamics of rotations including alfalfa could help to identify optimum stand lengths as far as N related benefits are concerned.

Although established stands have the ability to supply substantial amounts of N to subsequent crops, they can be impractical for producers who do not feed forage to livestock and rely mainly on grain production. A cropping system that utilizes a single year of alfalfa would give producers the option of using the forage as hay or as an N input for subsequent crops, or a combination of the two, without limiting the choice of crops in the following year (Sheaffer et al., 1989). Non-dormant alfalfa cultivars are well suited to annual cropping systems in northern areas. They are not adapted for winter survival, as partitioning of carbohydrates to the roots does not increase substantially with the onset of fall. Rather, this energy is used for prolonged top-growth and nitrogen fixation relative to the more traditional, dormant cultivars. Herbage growth generally continues in the non-dormant alfalfas until there is a heavy frost, unlike the dormant cultivars where growth reduction begins in late summer or early fall. The poor winter survival generally observed with the non-dormant cultivars facilitates the introduction of other crops in the subsequent year (Pfarr, 1988).

Nitro alfalfa was developed in Minnesota as a special purpose alfalfa for use as a one year hay source and fall plow-down green manure crop. Nitro is the first alfalfa cultivar selected for specialized N accumulation attributes (Barnes et al., 1988). The majority of the research involving Nitro has examined its role as a hay source or as a plow down crop by estimating the amount of total nitrogen incorporated into the soil (Griffin and Hesterman, 1991; Hesterman et al., 1986; Hesterman et al. 1992; Sheaffer et al., 1988; Sparrow et al., 1993). Little research actually looks at the contribution of symbiotically fixed nitrogen to subsequent crops (Hesterman et al., 1987; Sheaffer et al., 1989). No research exists that examines the applicability of Nitro as a single year rotational crop in Manitoba.

An alfalfa-based cropping systems project was initiated in 1990 at Portage la Prairie and Winnipeg, Manitoba by Dr. M. H. Entz of the Department of Plant Science, University of Manitoba. The central concept of the project was to determine the feasibility of including short term alfalfa stands in crop rotations. This thesis examines some of the nitrogen related aspects of that project.

The main objectives of this research were to evaluate the applicability of Nitro as a one year hay and fall nitrogen source in Manitoba, and to develop a basic nitrogen budget that examines the nitrogen dynamics of varying periods (one to three years) of alfalfa in a cropping system.

2. Literature Review

2.1. THE BENEFITS OF ALFALFA IN CROP ROTATIONS

"Some crops are to be planted not so much for the immediate yield as with a view to the following year, since when cut down and left they improve the soil" (Varro, *Rerum rusticarum* 1.23.3). These words, spoken by a first century B.C. agronomist, describe one of the major advantages of including legumes in a cropping sequence, and consequently form the basis for the concept of crop rotation.

The practice of growing alfalfa (*Medicago sativa* L.) in a rotation has long been identified as a means of improving the yield and quality of other crops in the sequence. Research conducted in the early 1930's showed that the protein concentration of the first crop of wheat following alfalfa was substantially higher (16.56%) than after forage grasses (13.58%). Likewise, protein levels were also higher in the second wheat crop after forage breaking, with protein contents of 15.21 and 12.59% for wheat preceded by alfalfa and the forage grass average, respectively (Ellis, 1943). Ellis advocated "using legumes in appropriate mixtures with grasses, in order to maintain yield and quality in subsequent grain crops". Similarly, research conducted by Hedlin et al. (1957) in the late 1940's demonstrated wheat yields of 2313, 1722, and 2112 kg ha^{-1} when wheat was the first crop after alfalfa, forage grass, or summerfallow, respectively. The second crop after breaking produced respective yields of 1601, 1305, and 1238 kg ha^{-1} after alfalfa, grass or summerfallow. Hedlin et al. concluded that "the use of legumes, either as green manures or for hay, results in higher yields and protein contents of wheat than where these crops are not grown".

Yield increases in crops following alfalfa were soon taken for granted, and it was generally accepted that the main source of this increased yield was a greater amount of fixed nitrogen in the soil as a result of symbiotic nitrogen fixation (Gardner and Robertson, 1954). It became apparent, however, that other factors played a role in increasing yield, as crops grown in rotation generally yielded 5 to 10% higher than crops grown continuously, when fertility management was optimal (Hesterman et al., 1987). These non-N related factors are called rotation effects, and are thought to include benefits such as reduced weed, insect, and disease infestations, and improved soil physical properties such as better soil structure, higher organic matter levels, and improved water infiltration (Baldock et al., 1981; Hesterman et al., 1986). Rotation effects have been known to account for as much as 25% of the yield increase in rotated crops (Baldock et al., 1981).

Although rotation effects can play a substantial role in increasing the yield of crops following alfalfa, the addition of symbiotically fixed nitrogen to the soil is responsible for the majority of the observed yield increase where additional N fertilizer has not been applied. Legume N contributions in rotations have been measured using the fertilizer N replacement value, which is the amount of inorganic N that would be required to produce an equivalent yield under otherwise comparable test conditions. This measure provides a rough indication only of the legume N contribution, as it assumes that fertilizer N and N from crop residues are equally available, and it does not account for the influence of rotation effects (Hesterman et al., 1987).

Hesterman et al. (1992) measured a fertilizer N equivalence of 55 kg ha^{-1} when alfalfa was interseeded in the spring with fall sown winter wheat, and worked down the following spring. Bruulsema and Christie (1987) measured fertilizer N replacement values of 90 to 125 kg ha^{-1} for seeding year alfalfa that was cut once for hay and incorporated in the fall. When the alfalfa was cut twice in the seeding year and the herbage spread across the soil, the subsequent wheat yield was similar to that achieved with applications of 150 kg ha^{-1} fertilizer N (Badaruddin and Meyer, 1990). The fertilizer N equivalence of three years of alfalfa followed by first year corn was 136 kg ha^{-1}, while an equivalent of 36 and 16 kg N ha^{-1} was supplied to second and third year corn, respectively (Fox and Piekielek, 1988). The amount of N supplied by alfalfa to following crops tends to be quite varied, and is influenced by a range of factors, but it seems clear that alfalfa has the ability to supply the majority, and in some cases all, of the subsequent crop's nitrogen requirements.

2.2. NITRO: A SPECIAL PURPOSE ALFALFA

2.2.1. Rationale

Despite the potential for reducing N inputs by rotating crops with alfalfa, little is grown for strictly rotational reasons. In Minnesota, alfalfa is normally produced for two or more years as a hay source on farms that have a large livestock component. Producers often commit to longer periods of alfalfa due to difficulties in establishing and removing the crop. These rotations can be impractical on farms that feed little forage and rely mainly on grain production. A cropping system that utilizes a single year of alfalfa would give producers the option of using the forage as hay or as an N input for subsequent crops, or a combination of the two, without limiting the choice of crops in the following year (Sheaffer et al., 1989). As well, older alfalfa stands tend to deplete soil moisture reserves in dry areas (Campbell et al., 1990), making annual rotations more appropriate in these regions.

Non-dormant alfalfa cultivars are well suited to annual cropping systems in northern areas. They are not adapted for winter survival, as partitioning of carbohydrates to the roots does not increase substantially with the onset of fall. Rather, this energy is used for prolonged top-growth and nitrogen fixation relative to the more traditional, dormant cultivars. Herbage growth generally continues in the non-dormant alfalfas until there is a heavy frost, unlike the dormant cultivars where growth reduction begins in late summer or early fall. The poor winter survival generally observed with the non-dormant cultivars facilitates the introduction of other crops in the subsequent year (Pfarr, 1988).

2.2.2. Development

Renewed interest in low input sustainable agricultural practices and forage legume-based crop rotations has resulted in the development of a new non-dormant alfalfa for use in the upper midwestern United States as a one year crop. Nitro alfalfa was developed cooperatively by the USDA-ARS and the Minnesota Agricultural Experiment Station as a special purpose alfalfa for use as a one year hay source and fall plow-down green manure crop. The producer would have the option of managing the alfalfa to meet the needs of on-farm forage requirements, the potential for selling hay, or the need to provide N for subsequent crops, without limiting cropping choices the following year. Selection criteria included fall growth characteristics, root mass and root N concentration, as well as disease and insect resistance. Nitro is the first alfalfa cultivar selected for specialized N accumulation attributes (Barnes et al., 1988).

2.2.3. Summer Herbage Harvested for Hay

If an alfalfa variety is to be successful as a dual purpose hay and fall plow-down green manure crop, summer hay yields should be as good or better than that achieved with other available cultivars. Sheaffer et al. (1989) measured identical total summer forage yields (8500 kg ha^{-1}) for Nitro and representative non-dormant and dormant varieties, when cut three times at bud by early September, averaged over four Minnesota locations. Griffin and Hesterman (1991) observed significantly higher total forage yields in moderately dormant Saranac (7100 kg ha^{-1}) compared with non-dormant Nitro (6300 kg ha^{-1}) at one Michigan location, when the yields of the two summer harvests and late fall cut were combined. The amounts of N removed in the forage were not significantly different, however, with 217 and 195 kg N ha^{-1} harvested with the forage of Saranac and Nitro, respectively.

In a comparison of harvest management systems, Sheaffer et al. (1988) found that Nitro and Saranac had similar summer forage yields when cut twice at bud, or when three summer harvests were performed at bud. However, when the two summer cuts were delayed until first flower, Saranac yielded significantly more forage than Nitro. When averaged over three Minnesota locations under this harvest management system, Saranac and Nitro yielded 7530 and 6280 kg ha^{-1}, respectively. Research conducted by Hesterman et al. (1986) indicated that MN ROOT N (Syn 2 generation of the commercial cultivar Nitro) and Saranac were not significantly different in terms of total and individual summer harvest yields, under two different management systems (one or two summer cuts at one-tenth bloom), at four Minnesota locations.

It seems that the summer hay yields of Nitro are, for the most part, similar to other alfalfa cultivars in the seeding year, although this may be influenced by harvest management.

2.2.4. Fall Herbage and Root Production

2.2.4.1. Fall Herbage Production. As Nitro is a non-dormant cultivar, it should exhibit superior fall herbage production relative to other dormant varieties. Sheaffer et al. (1989) observed significantly greater fall forage yields in Nitro compared to representative non-dormant and dormant cultivars, when cut three times at bud in the summer. When averaged over four Minnesota locations, Nitro, the non-dormants and the dormant alfalfas produced an average of 2010, 1760, and 1000 kg ha^{-1} of forage in the fall, respectively. Sheaffer et al. (1988) measured significantly greater fall herbage yields in Nitro relative to Saranac under two of three harvest management systems. When two summer cuts were taken at bud and the fall growth was sampled at first flower, Nitro and Saranac produced 1760 and 1500 kg ha^{-1} of forage, respectively. Under a system that utilized three summer cuts (at bud) with the fall regrowth harvested at bud, 1260 and 500 kg ha^{-1} of fall forage was produced by Nitro and Saranac, respectively. Nitro and Saranac were similar in a system that employed two summer cuts at first flower, with the regrowth sampled at first flower in the fall. Likewise, under a harvest management system where only one summer cut was taken, with the regrowth sampled in the late fall, MN ROOT N and Saranac were not significantly different at four Minnesota locations, producing an average 2100 and 2000 kg ha^{-1} of fall forage, respectively (Hesterman et al., 1986). Although non-dormant

Nitro seems to produce more forage in the fall relative to other dormant and moderately dormant cultivars, harvest management appears to play a role.

Groya and Sheaffer (1985) noted lower October forage yields in two non-dormant alfalfas (1000 kg ha^{-1}) compared with two dormant cultivars (1600 kg ha^{-1}). The non-dormants, however, were cut three times in the summer while the dormant cultivars were only cut twice. Kroontje and Kehr (1956) measured average fall forage yields of 1460 and 1530 kg ha^{-1} for non-dormant and dormant cultivars when cut once in the summer, and 570 and 300 kg ha^{-1}, when two summer cuts were performed, respectively.

2.2.4.2. **Root Production**. One of the primary criteria in developing Nitro was selection for large crowns and roots with high nitrogen concentrations, such that plow down N yield could be improved. Root yields and N concentrations of 1760, 1760 and 1500 kg ha^{-1} and 2.7, 2.6 and 2.4% were measured for Nitro and representative non-dormant and dormant cultivars respectively (Sheaffer et al. 1989). Nitro and the other non-dormants were significantly greater than the dormant cultivars in root yield and N concentration, but were not significantly different from each other. MN ROOT N and Saranac produced root yields and N concentrations of 3800 and 3200 kg ha^{-1}, and 2.6 and 2.2% in a system where two summer cuts were taken, and 4000 and 3900 kg ha^{-1}, and 2.8 and 2.6% when only one summer harvest was employed, respectively (Hesterman et al., 1986).

Sheaffer et al. (1988) observed that root yields were not significantly different between Nitro and Saranac, even when a number of different harvest management systems were tested. When averaged over three systems that varied in the number and timing of summer hay cuts taken, Nitro and Saranac both produced root yields of 2510 kg ha^{-1}.

2.2.5. Nitrogen Incorporated into the Soil in the Fall

2.2.5.1. Crops Managed for Summer Hay and Fall Nitrogen. The amount of nitrogen worked down into the soil with the alfalfa residues in the fall determines, in part, the value of a fall plow down green manure crop. Griffin and Hesterman (1991) measured plow down nitrogen values of 166 and 230 kg N ha^{-1} for Nitro and Saranac, respectively. The alfalfa was, however, worked down the following spring after a substantial amount of spring regrowth had occurred, which was included in the plow down estimates. The non-dormant nature of Nitro would set it at a disadvantage relative to moderately dormant Saranac when spring regrowth is considered. MN ROOT N had significantly greater fall plow down yields relative to Saranac, producing 100 and 71 kg N ha^{-1} in a two hay cut system, and 167 and 147 kg N ha^{-1} in a single summer cut system (Hesterman et al., 1986). Sheaffer et al. (1988) also observed consistently greater plow down N yields in Nitro relative to Saranac. For the two cut (at bud), three cut (at bud), and two cut (at first flower) harvest systems, plow down yields were 119 and 100, 101 and 68, and 105 and 97 kg N ha^{-1} for Nitro and Saranac, respectively. In a comparison of Nitro and twelve other non-dormant alfalfas, Nitro produced 68 kg N ha^{-1} for plow down compared with an average 61 kg N ha^{-1} generated by the other non-dormants. Of the twelve other non-dormants tested, Nitro was significantly greater than seven varieties in terms of plow down nitrogen (Sheaffer et al., 1989).

Nitro seems to produce more N for soil incorporation, although not always significantly more, than other dormant and many non-dormant alfalfas when managed as a dual purpose hay and fall nitrogen crop. Other non-dormant varieties have been tested as combination hay and green manure crops. Groya and Sheaffer (1985) found that fall N production was significantly less in two non-dormant alfalfas compared with two dormant cultivars, producing 85 and 127 kg N ha^{-1} for plow down. The non-dormants were managed more intensively (greater number of summer hay cuts) which could have affected fall N production. Smith (1956) measured fall N yields for non-dormant (Kansas) and dormant (Grimm) cultivars, under different harvest strategies. For systems that utilized one cut in September, one cut in October, and two cuts (September and October), non-dormant and dormant alfalfas produced an average 60.4 and 55.1, 42.7 and 46.5, and 28.2 and 44.9 kg N ha^{-1} for fall soil incorporation, respectively. In this case, the non-dormant alfalfa seemed to be disadvantaged when two late fall cuts were taken. Kroontje and Kehr (1956), however, noted similar performance between non-dormant and dormant cultivars despite the intensity of harvest management. For systems that utilized one, two and three summer hay cuts, plow down N yields for the average of four non-dormant and two dormant cultivars were 88 and 85, 88 and 90, and 67 and 58 kg N ha^{-1}, respectively. The wide variety of harvest management systems used in these studies makes it difficult to determine if differences truly exist between non-dormant and dormant alfalfas in terms of fall plow down nitrogen yields.

2.2.5.2. Crops Managed for Fall Nitrogen Only. An alternate way of managing these "annual" alfalfas is to allow them to grow throughout the year, without any forage removal for hay, and incorporate them into the soil in the fall. Their main purpose in this case would be to supply nitrogen to subsequent crops. Such a strategy would be simplified over systems requiring intensive harvest management, and may be more appropriate on farms that have relatively small forage requirements.

Sheaffer et al. (1988) measured average fall plow down N values of 144.3 and 131.6 kg ha^{-1}, for Nitro and Saranac, respectively, when the alfalfa was allowed to accumulate dry matter in situ until fall. Nitro produced significantly more plow down N at one of the three sites in this case. In an experiment comparing different green manure crops in Alaska, Nitro accumulated an average 126.5 kg ha^{-1} of nitrogen in the herbage, ranking lowest of the tested crops (Sparrow et al., 1993). Root nitrogen was not included in the plow down estimate of this study. Hesterman et al. (1992) examined a system where alfalfa was frost seeded (in early spring) into winter wheat that had been sown the previous fall. Nitro produced an average of 61 kg N ha^{-1} in the fall herbage and roots for plow down, compared with 73 and 63 kg N ha^{-1} for dormant Big Ten and moderately dormant Saranac alfalfas, respectively. Contrasts comparing non-dormant and dormant alfalfa varieties showed no significant differences in the amount of plow down N present in the herbage or roots at any of the three Michigan experimental sites.

Experiments with other non-dormant (CUF 101 and Ardiente) and dormant (Saranac and Agate) alfalfas demonstrated plow down averages of 184 and 207 kg N ha^{-1} for the non-dormant and dormant cultivars, respectively, when the forage was allowed to accumulate over the growing season (Groya and Sheaffer, 1985). Two non-dormant alfalfas performed similarly to a dormant line in terms of fall nitrogen production, with plow down yields of 65.5, 59.9, and 64.7 kg N ha^{-1} for Arizona Common, Kansas and Grimm alfalfas, respectively. Fribourg and Johnson (1955) also noted similar performance between Grimm and Southern common alfalfa when allowed to accumulate in situ, with plow down yields of 66.4 and 62.5 kg N ha^{-1}, respectively. Stickler and Johnson (1959), however, noted slightly higher fall nitrogen yields in two non-dormant alfalfas compared with a dormant cultivar, with measured nitrogen yields of 90, 84, and 62 kg ha^{-1} for Indian, African and Ranger alfalfas, respectively.

These studies indicate that it is, at best, unclear whether Nitro, or other non-dormant cultivars, hold some advantage over dormant varieties in terms of fall nitrogen plow down yields when the herbage is allowed to accumulate during the growing season.

2.2.6. Nitrogen Fixation

A green manure legume crop benefits the soil nitrogen status by symbiotically fixing atmospheric nitrogen, which is added to the soil as the plant residues decompose after the crop is worked down. An alfalfa that has high plow down N yields, but derives the majority of it's nitrogen from the existing soil N pool such that the nutrient is merely recycled, is of little value as a fall nitrogen source.

Relatively little research has been directed at examining the actual amounts of symbiotically fixed nitrogen returned to the soil. Sheaffer et al. (1989) determined the amounts of biologically fixed nitrogen present in alfalfa roots and herbage at fall plow down, when the forage was cut three times at bud during the summer. Nitro and representative non-dormant and dormant varieties produced 105, 94 and 63 kg ha^{-1} of biologically fixed nitrogen, respectively, for fall soil incorporation. Nitro produced significantly greater amounts of fixed N than the dormant, but not the non-dormant, cultivars in this case. Hesterman et al. (1987) measured amounts of fixed N_2 incorporated into the soil for MN ROOT N and Saranac under two different harvest schedules. For one and three cut harvest systems, an average of 103 and 75, and 75 and 50 kg ha^{-1} of biologically fixed N_2 was worked into the soil in the fall for MN ROOT N and Saranac, respectively. Sparrow et al. (1993) estimated the amount of fixed N_2 in the herbage of

Nitro, when the forage was allowed to accumulate over the season, in a subarctic environment. Nitro fixed an average of 45 kg N ha^{-1} over the growing season (measured in herbage only), which ranked lowest of the other green manure species tested.

Heichel et al. (1984) measured N_2 fixation of two dormant alfalfa cultivars over a four year period. In the seeding year, the proportion of N derived from symbiosis increased from the first summer harvest (late July) to the second (late August), and decreased substantially by the third harvest (late October). Patterns of N_2 fixation were thought to be related to factors such as mineral soil N availability and seasonal precipitation. The average %N derived from symbiosis and the quantity of N fixed on a land area basis was 59.2% and 169 kg N ha^{-1}, respectively, in the seeding year. Heichel et al. (1981) examined nitrogen fixation of dormant and moderately dormant experimental populations of alfalfa that had undergone one cycle of recurrent selection for characteristics associated with whole plant nitrogenase activity. The forage was not harvested for hay during the growing season, but was sampled (whole plant) at four times during the year. The moderately dormant and dormant populations fixed 170 and 125 kg N ha^{-1} over the year, and derived an average 44.6 and 42.3% of their N from symbiosis, respectively. The moderately dormant population continued to grow later in the fall due to the earlier onset of fall dormancy in the dormant alfalfa, which resulted in extended N_2 fixation and, consequently, greater amounts of N_2 fixed in the moderately dormant population.

2.2.7. Winter Survival

In order to facilitate the introduction of subsequent crops, the winter survival of an "annual" alfalfa should be sufficiently poor, such that the use of excessive tillage or herbicides is not required to remove the stand. Barnes et al. (1988) report that Nitro will not reliably survive most upper Midwest U.S. winters, but it may survive during winters with sufficient snow cover and/or mild temperatures. At latitudes with milder winters, non-dormant alfalfas may overwinter and act as true perennials. In Oklahoma, Nitro was among the more productive cultivars in the second and third years of production trials (Sheaffer et al., 1989).

Sheaffer et al. (1992) measured the survival of a number of alfalfa cultivars subjected to four harvest schedules at five locations in Minnesota. Dormant Rambler, moderately dormant Saranac, and non-dormant Nitro had average stand survivals of 84% (range 56% to 95%), 63% (range 27% to 95%) and 14% (range 3% to 25%), respectively.

Although winter survival is substantially poorer in Nitro than the more dormant cultivars, it seems that it is not consistently low, and volunteer alfalfa control could be a concern in the following year.

2.2.8. Yield and Nitrogen Uptake of the Subsequent Crop

Nitro was developed as a special purpose rotational alfalfa that could accumulate large amounts of nitrogen during the seeding year, nitrogen that would be available to the subsequent crops in the rotation. N fertilizer requirements would theoretically be lower following Nitro compared with non-legume crops, or other alfalfa cultivars for that matter.

Griffin and Hesterman (1991) observed significant increases in the growth of a potato crop when Nitro and Saranac alfalfa, rather than potatos, were the preceding crops. Nitro and Saranac were, however, similar in their effect on the subsequent potato crop. With no additional applied nitrogen, potato vine dry matter yields, N concentrations and N yields of 3770, 3360, and 2010 kg ha^{-1}, 3.03, 3.38 and 2.70%, and 116, 113 and 55 kg N ha^{-1} were measured when Nitro, Saranac and potatos, respectively, were the preceding crops. Hesterman et al. (1986) also observed similar yield responses in subsequent corn that was preceded by Nitro or Saranac. When the alfalfa was subject to harvest schedules where three hay cuts were taken with the crowns and roots incorporated, or only one summer harvest was performed with the herbage regrowth, crowns and roots incorporated, subsequent corn yields were 6500 and 6300 kg ha^{-1}, and 7100 and 7400 kg ha^{-1} for MN ROOT N and Saranac, respectively. When corn was grown two years in a row, the yield of the second crop was 3700 kg ha^{-1}, when no N fertilizer was applied. In a later paper, Hesterman et al. (1987) reported the whole plant uptake of legume-supplied nitrogen in subsequent corn. For the three and one cut harvest management systems, average legume-N uptake in corn was 62 and 68, and 47 and 45 kg N ha^{-1} for MN ROOT N and Saranac, respectively.

Other researchers have noted similar nitrogen responses of crops following non-dormant and dormant alfalfas. The nitrogen uptake of sudangrass was an average 149 kg N ha^{-1} when preceded by non-dormant alfalfa varieties, and 157 kg N ha^{-1} when preceded by dormant cultivars. The alfalfa in this case was cut once during the summer, with the herbage regrowth, crowns and roots incorporated into the soil in the fall. Kroontje and Kehr (1956) measured the N uptake in barley preceded by a number of non-dormant and dormant alfalfas subject to three different harvest schedules. For systems that had one, two or no (forage allowed to accumulate) summer hay cuts, with the fall regrowth, crowns and roots incorporated, average barley N uptakes of 57 and 43.8, 51.5 and 48.2, and 46.3 and 41.6 kg N ha^{-1} were measured for four non-dormant and two dormant alfalfa varieties, respectively, when alfalfa was the preceding crop.

Significant yield and nitrogen responses in subsequent crops are often observed when the preceding alfalfa is compared with a non-leguminous crop. Differences among cultivars of alfalfa, however, are much more rare. Small differences in nitrogen fixation or N accumulation in the plant tissues, although significant, are often not translated into substantial differences in the yield or N uptake of the subsequent crop.

2.3. Nitrogen Dynamics of Established Alfalfa Stands

Producers that have a substantial livestock component to their farming enterprise have large, steady requirements for high quality forage. A certain amount of the land resource must be planted to forage at any one time. A recent survey of Manitoba and Eastern Saskatchewan farmers showed that the average length of pure alfalfa stands was 6 years (Katepa-Mupondwa et al., 1993). As alfalfa can be difficult to remove and establish, stands are usually maintained as long as they remain productive and satisfy the forage requirements. The rotational benefits of alfalfa are often overlooked when considering stand removal. For example, Katepa-Mupondwa et al. (1993) found that only 12% of the surveyed producers worked up their alfalfa stands to receive the rotational benefits it is has to offer. Working up stands more frequently would mean that more land would be "treated" with alfalfa more often, while still maintaining the same level of forage production. The number of acres receiving the rotational benefits of alfalfa would increase with shorter stand lengths. The need therefore exists to identify a stand length such that the benefits of alfalfa are optimized before rotating the crop to a different part of the farm's land base.

One of the most significant rotational benefits that alfalfa has to offer is it's ability to supply nitrogen to subsequent crops, potentially reducing N fertilizer requirements the following year. The amount of residual N available for subsequent crops is not the only factor that should be considered when selecting an optimum stand length. The amounts of nitrogen derived from atmospheric or soil sources, and their partitioning into harvested forage or incorporated green manure, are all factors that contribute to the nitrogen dynamics of alfalfa-based cropping systems. Very little research exists that examines nitrogen fixation and nitrogen partitioning in alfalfa stands of increasing age. A better understanding of the N dynamics of rotations including alfalfa could help to identify optimum stand lengths as far as N related benefits are concerned.

2.3.1. The Nitrogen Cycle

In order to study the nitrogen dynamics of alfalfa in crop rotations, a basic understanding of the nitrogen cycle and how it applies to cropping systems is required. Cabon et al. (1991) divided the various processes involved in the N cycle of the soil-water-plant-atmosphere system into three main categories.

1. Processes governing the input of the system:
    a. biological fixation
    b. contribution from the atmosphere
    c. artificial additions
2. Processes governing the output of the system:
    a. denitrification
    b. volatilization of gaseous nitrogen
    c. leaching of nitrate
    d. plant uptake
3. Processes governing internal transformations:

a. mineralization

b. nitrification

c. immobilization of N from ammonium, immobilization of N from nitrate

d. adsorption-desorption of ammonium

The interaction of these individual processes with one another largely determines the amounts of residual nitrogen available for use by crops following alfalfa in a cropping sequence, and therefore influences alfalfa's role as a rotational crop.

2.3.1.1. System Inputs

One of the major inputs of N into cropping systems is biological nitrogen fixation. The average annual input of biologically derived nitrogen in legume based agricultural ecosystems is estimated at 140 kg N ha^{-1}, while the input of biological nitrogen in non-legume and grassland agricultural systems is estimated at 35 and 15 kg N ha^{-1} yr^{-1}, respectively (Burns and Hardy, 1975 *in* Haynes, 1986). The majority of the nitrogen fixed in temperate agricultural zones is performed by the symbiotic fixers, organisms that fix N_2 in association with higher plants. Of this group, the bacterial symbionts of the genus *Rhizobium*, in association with plants of the *Leguminosae* family, are the most significant in agricultural systems. Atmospheric N_2 is converted to NH_3 in a process catalyzed by the nitrogenase enzyme complex. The equation for the reaction is:

$$N_2 + 6e^- + 6H^+ \longrightarrow 2NH_3$$

The plant exchanges biologically fixed nitrogen, which is used in growth and development, for carbohydrates required by the bacteria, thereby establishing the symbiotic relationship. Biological N_2 fixation is also performed by a number of non-symbiotic organisms that can fix molecular N_2 apart from a specific host. Estimates of annual N input through non-symbiotic N_2 fixation are generally less than 17 kg N ha^{-1} (Russelle, 1992), but can be substantial in moist tropical environments.

The addition of artificially derived nitrogen forms (fertilizers) constitutes another major N input into the plant-soil system. In the arable land of North America, the average intensity of N fertilizer use is estimated at 48.6 kg N ha^{-1} yr^{-1} (Hauck, 1981 *in* Haynes, 1986). Synthesized fertilizer is a major nitrogen input on the Prairies, particularly in cropping systems that do not utilize legumes.

Nitrogen may also be added to the plant-soil system by atmospheric deposition. Various forms of N can be added by dry or wet deposition, ranging from insignificant amounts to 60 kg N ha^{-1} yr^{-1} (Goulding, 1990 *in* Powlson, 1993). Atmospheric depositions would be considered a minor input on the Prairies relative to additions by biological N_2 fixation and artificial fertilizer additions.

2.3.1.2. System Outputs

Nitrogen is removed or lost from the plant-soil system by a number of different processes. Rapid losses of nitrogen from the soil can occur under suitable conditions through bacterial denitrification. Denitrification is the reduction of nitrate to molecular nitrogen or nitrogen oxides by microbial activity. N losses through denitrification vary greatly, depending on nitrate levels, available organic carbon, temperature, pH, and the moisture status of the soil.

Ammonia volatilization represents another route of nitrogen loss in the plant-soil system. Losses can be quite large when NH_3-forming fertilizers are applied to the surface of calcareous soils, particularly under conditions that favour soil drying. N losses due to ammonia volatilization are generally less than 10 kg N ha^{-1} in most agricultural systems (Goulding et al., 1993 *in* Powlson, 1993).

Nitrogen (mainly in the nitrate form) may also be lost by leaching. Leaching losses occur when soil nitrate levels are high, and when the downward movement of water is sufficient to move nitrate below the rooting depth of crops. The type of cropping system will also influence N losses due to leaching. Crop rotations that promote the conversion of N to nitrate forms, and also contribute to large amounts of downward water movement, will increase N leaching. For example, 9.8 kg N ha^{-1} was lost from a permanent grass pasture, while 25.0 kg N ha^{-1} was leached in a fallow/spring wheat cropping system (Catt et al., 1992 *in* Powlson, 1993).

N removal with harvested plant materials represents another significant method of nitrogen loss from the plant-soil system. Approximately 45 kg N ha^{-1} is removed with the grain of a typical Prairie wheat crop, while as much as 400 kg N ha^{-1} can be removed annually with the forage of an average hay crop.

2.3.1.3. Internal System Transformations

Within the plant-soil system, an internal nitrogen cycle exists that operates almost independently of the overall cycle. The main feature of the internal N cycle is the biological turnover of nitrogen through the processes of mineralization and immobilization. Mineralization causes the conversion of organic N to inorganic forms, while immobilization is the reverse procedure. The end result of these two co-occurring processes is a continuous interchange of inorganic and organic N forms.

Mineralization is divided into two main steps: ammonification and nitrification. Ammonification refers to the conversion of organic N to NH_3 by heterotrophic microorganisms. This is followed by nitrification, which results in the conversion of NH_4^+ to NO_2^- (mediated by *Nitrosomonas* bacteria), and the subsequent conversion of NO_2^- to NO_3^- (mediated by *Nitrobacter* bacteria) (Sprent, 1990).

N immobilization is also divided into a two step process. In the first step, mineral N forms are incorporated into amino acids and ultimately to organic substances composing microbial cell constituents in a process called assimilation. Humification is the ultimate conversion of the N in microorganisms to more permanent forms, such as humic and fulvic acids, which are the building blocks of soil humus.

Another factor affecting the internal N cycle, that does not involve the activity of soil microorganisms, is the fixation of ammonium ions in certain clay minerals. Ammonium can be held by 2:1 clay minerals (vermiculites and montmorillonites) in a nonexchangeable, fixed form. This nitrogen would be effectively removed from the workings of the internal N cycle. Estimates of fixed ammonium range from 7-14% of the total N in certain Prairie topsoils (Moore, 1965 *in* Haynes, 1986).

2.3.2. Nitrogen Fixation of Established Alfalfa

The amount of existing soil nitrogen removed from the soil with the harvested herbage, and the amount of biologically fixed nitrogen returned to the soil with incorporated herbage and roots will affect N contributions by alfalfa to subsequent crops (Sheaffer et al., 1989). High levels of N_2 fixation throughout the life of the stand will ensure that minimal amounts of existing soil N are removed with the forage, thereby increasing the overall contribution of biological nitrogen to the soil.

Wivstad et al. (1987) determined the proportion of nitrogen derived from the atmosphere (%Ndfa) at three hay harvests (two summer cuts, one late fall) for two and three year old alfalfa stands. For the first, second, and third harvests, %Ndfa levels of 61, 77, and 67%, and 72, 87, and 84% were reported for second and third year alfalfa, respectively. This resulted in quantities of fixed N_2 in the forage of 64, 117, and 61 kg ha^{-1}, and 123, 135, and 61 kg N ha^{-1} for the two and three year old stands at the first, second and third harvests, respectively. The average %Ndfa and total amount of N_2 fixed in the herbage over the season for second and third year alfalfa was 70 and 80%, and 242 and 319 kg N ha^{-1}, respectively.

Heichel et al. (1984) looked at N_2 fixation and N partitioning over the life of a four year alfalfa stand. The %N from symbiosis generally increased as the season progressed, except in year two where abnormally high summer rainfall significantly decreased N_2 fixation after the first harvest. Seasonal N_2 fixation averages for first, second, third and fourth year alfalfa were 59, 36, 61 and 78%, respectively. Quantities of N_2 fixed were closely related to the %N from symbiosis with 169, 127, 168, and 198 kg N ha^{-1} fixed for one to four year old stands, respectively.

Both experiments indicated increased N_2 fixation with increasing stand age. Heichel et al. (1984) suggested that interannual increases in the proportion of N_2 fixed during the life of the stand could be related to a decreasing availability of readily mineralizable N in the soil.

2.3.3. Nitrate Extraction

An often overlooked benefit of alfalfa is it's ability to extract and utilize nitrate that has leached past the rooting zone of most annual crops. Alfalfa roots have the ability to absorb water and nutrients to 11 m (Peterson and Russelle, 1991), giving it access to large amounts of leached nitrogen that can be removed with the herbage. The utilization of this nitrate by alfalfa may decrease levels of symbiotic N_2 fixation and, subsequently, the addition of biologically fixed N to the soil system. Because this leached nitrogen cannot be recovered by other crops, however, it may be thought of as additional nitrogen, not unlike that added to the soil system by biological means through the nitrogen fixation process.

Schuman and Elliott (1978) observed the removal of large amounts of nitrate-nitrogen from a 4.6 m soil profile with each additional year of alfalfa growth in an abandoned feedlot. Nitrate levels were 113, 171, 1560, and 1703 kg N ha^{-1} less than the control treatment after one, two, three and four years of alfalfa, respectively. Mathers et al. (1975) noted similar results when alfalfa was cropped to plots that had previously received varying amounts of manure. Nitrate nitrogen was removed to a depth of 1.8 and 3.6 m in the first and second years of alfalfa growth, respectively. Nitrate removal and water extraction patterns were very similar, indicating that nitrate was removed to the same depth that water was utilized. A concern of using alfalfa, however, is the potential for large amounts of mineralization and releaching of nitrogen once the alfalfa is killed, especially in areas of high rainfall or under irrigation (Robbins and Carter, 1980; Peterson and Russelle, 1991).

2.3.4. Total Soil Nitrogen

The addition of biologically fixed nitrogen by alfalfa to the soil system should be reflected in the soil N status as an increase in total soil N. Lyon and Bizzell (1934) measured total N increases as large as 566 kg N ha^{-1} in the top 20 cm of soil when alfalfa was grown continuously for 10 years.

The effect of alfalfa, however, seems to depend on the initial nitrogen level of the soil. An annual increase of 8 kg N ha^{-1} was observed on a soil with 0.120% total N when alfalfa was grown 6 times in 10 years, compared with an increase of 68 kg N ha^{-1} on a soil with an initial total N content of 0.084% (Lyon and Bizzell, 1934). No increase was observed on a soil with an initial N content of 0.200% when alfalfa was grown for 4 years out of seven (Dubetz and Hill, 1964), while a decrease of 0.039% was observed on plots that had been in alfalfa for eight out of 12 previous years, when the initial soil N content was 0.245% (Ferguson and Gorby, 1971).

Increases in total soil N seem to be influenced by stand length. Holford (1990) measured total N increases of 103 kg N ha^{-1} yr^{-1} in the top 15 cm of soil when alfalfa was grown for a 4 year period. Holford observed a highly significant regression of total soil nitrogen on years of alfalfa. When 4 years of sorghum followed the alfalfa, the total nitrogen decreased, but was still significantly higher than at the outset of the experiment, and greater than the total N level in the continuous wheat control treatment. In an earlier study, Holford (1981) reported increases in total soil N of 0.008%, 0.011%, and 0.027%, corresponding to 156, 332, and 527 kg N ha^{-1} (based on a bulk density of 1.3 Mg m^{-3}) in the top 15 cm of a Black earth soil for 1.5, 2.5 and 3.5 years of alfalfa, respectively. On a Red Brown earth soil, there was a decrease of 0.009% (176 kg N ha^{-1}) in total soil N after the first 1.5 years of alfalfa, followed by subsequent increases of 0.01% and 0.016% (195 and 312 kg N ha^{-1}) after 2.5 and 3.5 years of alfalfa. The general trend was an increase in total soil N with each additional year of alfalfa.

2.3.5. Yield and N Uptake of Crops Following Established Alfalfa

The response of crops following alfalfa in crop rotations constitute an important part of the N dynamics of cropping systems, and plays a significant role in determining optimum stand lengths.

Some researchers noted little effect of stand age on the subsequent crop. Sheaffer et al. (1991) looked at the effects of one to three years of alfalfa, red clover, and birdsfoot trefoil on the subsequent corn N uptake. As differences were not detected among legume species, the data was pooled. For treatments that were subject to harvest management, total N uptake in corn was 100, 74, and 116 kg N ha^{-1}, when preceded by one, two and three years of legumes, respectively. Likewise, Hoyt and Hennig (1971) noted that there was no difference in the yield of a single subsequent wheat crop when preceded by two to six years of alfalfa.

Bolton et al. (1976) measured corn yields after corn, one year of alfalfa, and two years of alfalfa. The average unfertilized corn yield following two years of alfalfa was 5170 kg ha^{-1}, compared with 3560 kg ha^{-1} after one year, and 1670 kg ha^{-1} in a continuous corn rotation. Holford (1980) observed little difference in the yield of the first crop following different periods of alfalfa, but responses were apparent in the second and subsequent crops. He determined that the effects of alfalfa were significant in five wheat crops following a 2.5 year stand. A period of 3.5 years of alfalfa was determined to be the optimum stand length, as it eliminated the need for N fertilizer in an average four subsequent wheat crops. It seems that effects on a number of subsequent crops should be considered when evaluating the residual benefits of alfalfa in crop rotations.

2.3.6. Alfalfa Nitrogen Budgets

Nitrogen budgets or balances, defined as "the application of mass conservation principles so that N is conserved in the various transformations and biological processes of a system" have been useful tools in studying and understanding the various flows of N in agricultural productions systems (Legg and Meisinger, 1982). A great deal of research has been done at the farm, regional, and global levels in developing N budgets and models for agricultural systems (Groot et al., 1991; Rosswall and Paustian, 1984; Stinner et al., 1984; Wetselaar et al., 1973) although relatively little research has been conducted in the field on alfalfa specifically.

Paustian et al. (1990) developed annual nitrogen budgets for four different crops: barley (unfertilized), barley (120 kg N ha^{-1}), grass ley (200 kg N ha^{-1}), and alfalfa (unfertilized). N_2 fixation was the major N input for the alfalfa, providing 380 kg N ha^{-1} to the system. Atmospheric deposition was also identified as an input, contributing 5 kg N ha^{-1}. Measured N outputs included forage removal (246 kg N ha^{-1}), gaseous losses (20 kg N ha^{-1}) and leaching losses (1 kg N ha^{-1}), resulting in a change in total nitrogen of +120 kg N ha^{-1}. Of the studied treatments, only the alfalfa resulted in a positive change in the total N status. Much of this surplus nitrogen was accounted for in the plant standing crop (90 kg N ha^{-1}), while the remainder (30 kg N ha^{-1}) was assumed to have been incorporated into the soil organic matter.

Peterson and Russelle (1991) developed a nitrogen budget for alfalfa production in the Corn Belt states. They estimated the average annual amounts of forage produced and the amounts of atmospheric and soil N in the herbage, for eight states between 1984 and 1986. Average annual forage production was 8240 kg ha^{-1} during this period, with an average annual herbage N yield of 196 kg N ha^{-1}. They determined that 148 kg N ha^{-1} in the harvested forage was derived from biological N_2 fixation, and 48 kg N ha^{-1} was supplied by soil sources. The average annual input of fixed N_2 directly to the soil was estimated at 104 kg N ha^{-1}, while the total amount of nitrogen produced through symbiotic fixation was 252 kg N ha^{-1}. They estimated that 49% (73 kg N ha^{-1}) of the original 148 kg ha^{-1} of fixed N_2 in the harvested forage ended up in the soil system after feeding to livestock. Using these figures, they estimated that fertilizer use in the Corn Belt could be reduced by 198 to 365 kg N ha^{-1} for two crops of corn following alfalfa.

2.4. Summary

It is clear that alfalfa has much to offer as a component in modern cropping systems. Of the benefits alfalfa has to offer, it's ability to add substantial amounts of symbiotically fixed nitrogen to the soil resource is among the most impressive. Whether alfalfa plays the role of a special purpose, annual alfalfa like Nitro or a more forage oriented, established stand, the potential exists to improve the yield and quality of subsequent crops in the sequence, and reduce the nitrogen fertilizer requirements of those crops.

3. Nitrogen Fixation and Growth of a Non-dormant, Special Purpose Alfalfa (cv. Nitro)

3.1. ABSTRACT

Nitro is a non-dormant, special purpose alfalfa (*Medicago sativa* L.) developed in Minnesota for use as a one year hay source and fall plow-down green manure crop. Field experiments were established at two Manitoba sites to examine growth and N_2 fixation characteristics of Nitro. Nitro was compared with two dormant alfalfas in 1991, and to three dormant cultivars and another non-dormant alfalfa in 1992. N_2 fixation was measured using the ^{15}N isotope dilution technique and the difference method. Comparison of N_2 fixation estimates using barley and ineffective alfalfa as reference crops are discussed. Nitro yielded similarly to the other cultivars at the summer hay harvests, but produced significantly more herbage for soil incorporation in the fall relative to the dormant alfalfas (25% average increase in 1991, 39% increase in 1992). The quantities of N_2 fixed were also similar among treatments at the hay harvests, but were greater in Nitro in the fall relative to the dormant cultivars. Nitro and the dormant alfalfas added an average 151.9 and 53.4 kg ha^{-1} of fixed nitrogen to the soil in the fall of 1992, respectively. Nitro was not significantly different from CUF 101, the other non-dormant cultivar tested, in any of the measured parameters. Nitro seems to have an advantage over the traditionally used dormant alfalfas as a one year hay and fall nitrogen source, although further testing is required to determine it's superiority over other non-dormant alfalfas in Manitoba.

3.2. INTRODUCTION

Alfalfa (*Medicago sativa* L.) has long been recognized as a source of nitrogen for the subsequent crops in a rotation (Ellis, 1943; Hedlin et al., 1957). Nitrogen fertilizer replacement values of 125 to 135 kg N ha^{-1} for alfalfa followed by corn are not unusual (Baldock and Musgrave, 1980; Bruulsema and Christie, 1987). The amount of alfalfa N typically incorporated with three to five year old alfalfa stands in the midwestern United States has been estimated at 140 to 196 kg N ha^{-1} (Hesterman et al., 1987).

Although these established stands have the ability to supply substantial amounts of N to subsequent crops, they can be impractical for producers who do not feed forage to livestock and rely mainly on grain production. A cropping system that utilizes a single year of alfalfa would give producers the option of using the forage as hay or as an N input for subsequent crops, or a combination of the two, without limiting the choice of crops in the following year (Sheaffer et al., 1989). As well, older alfalfa stands tend to deplete soil moisture reserves in dry areas (Campbell et al., 1990), making annual rotations more appropriate in these regions.

Non-dormant alfalfa cultivars are well suited to annual cropping systems in northern areas. They are not adapted for winter survival, as partitioning of carbohydrates to the roots does not increase substantially with the onset of fall. Rather, this energy is used for prolonged top-growth and nitrogen fixation relative to the more traditional, dormant cultivars. Herbage growth generally continues in the non-dormant alfalfas until there is a heavy frost, unlike the dormant cultivars where growth reduction begins in late summer or early fall. The poor winter survival generally observed with the non-dormant cultivars facilitates the introduction of other crops in the subsequent year (Pfarr, 1988).

The concept of managing non-dormant alfalfas as annual crops is not new. Fribourg and Johnson (1955) reported no difference in dry matter yields or nitrogen contents between Grimm alfalfa (dormant) and a strain of Southern common alfalfa (non-dormant), when managed as one year green manure crops without hay removal. A similar study found the nitrogen yields comparable between four non-dormant and two dormant alfalfas, ranging from 56 to 101 kg N ha^{-1}, although the subsequent barley yield was significantly higher in three of the four non-dormant cultivars when compared with a non-fertilized check plot (Kroontje and Kehr, 1956). Stickler and Johnson (1959), however, discovered significant increases in the seeding year dry matter and nitrogen production of two non-dormant alfalfas relative to a dormant cultivar. Indian, African and Ranger alfalfa yielded 3549, 3188 and 2377 kg ha^{-1} of dry matter and produced 90, 84 and 62 kg ha^{-1} of nitrogen for soil incorporation, respectively. They attributed the differences to greater growth rates and lack of hardening in the Southern strains with the onset of fall.

Legumes receive nitrogen through two sources, existing soil N and biological N_2 fixation. If an annual alfalfa is to be successful as a fall nitrogen source, there must be a net increase in the soil N balance at the end of the season, an increase supplied by N_2 fixation. Published estimates of annual N_2

fixation by alfalfa range from 70 kg N ha^{-1} for seedling stands to 400 kg N ha^{-1} for established stands (Peterson and Russelle, 1991). Values representative of seasonal N_2 fixation in seeding year stands of various dormant and moderately dormant alfalfas in Minnesota range from 125 to 177 kg N ha^{-1} (Heichel et al., 1981; Heichel et al., 1984). The amount of biologically fixed N_2 returned to the soil and the subsequent increase in soil N will depend on the harvest management of the seedling alfalfa (Sheaffer et al., 1989).

Renewed interest in low input sustainable agricultural practices and forage legume-based crop rotations has resulted in the development of a new non-dormant alfalfa for use in the upper midwestern United States as a one year crop (Sheaffer et al., 1989). Nitro alfalfa was developed cooperatively by the USDA-ARS and the Minnesota Agricultural Experiment Station as a special purpose alfalfa for use as a one year hay source and fall plow-down green manure crop. Selection criteria included fall growth characteristics, root mass and root N concentration, as well as disease and insect resistance. Nitro is the first alfalfa cultivar selected for specialized N accumulation attributes (Barnes et al., 1988).

Given the potential for increasing interest in low input sustainable cropping systems, a study was initiated to evaluate the applicability of Nitro as a one year hay and fall nitrogen source under the climatic conditions of south-central Manitoba. Specific objectives were to evaluate growth and N_2 fixation characteristics of Nitro relative to the more traditionally used dormant cultivars, and examine the response of a subsequently grown wheat crop.

3.3. MATERIALS AND METHODS

Studies were carried out at Portage la Prairie and Glenlea, Manitoba in 1991 and 1992. Experiments were conducted on a Dugas clay at Portage la Prairie and a Scanterbury heavy clay at Glenlea. Temperature and precipitation data for the two sites is presented in Table 1. Climatic information was derived from Environment Canada data for the Glenlea site, and from information supplied by the University of Manitoba and Environment Canada at Portage la Prairie.

The 1991 treatments included Nitro (non-dormant), Excalibur (moderately dormant) and Algonquin (dormant), while the 1992 treatments were expanded to include CUF 101 (non-dormant) and Saranac (moderately dormant). A 1 to 9 fall dormancy scale (Barnes et al., 1991) has been used to rate the tested cultivars for fall dormancy, where 1 is the most fall dormant and 9 is the least fall dormant: CUF 101 (9), Nitro (8), Excalibur (4), Saranac (4), and Algonquin (2) (Certified Alfalfa Seed Council, 1992). Non-N_2 fixing controls included Argyle barley (*Hordeum vulgare* L.) in 1991 and 1992 and Ineffective Saranac alfalfa (a near isogenic line derived from Saranac incapable of forming effective nodules) in 1992 (Barnes et al., 1990). The experiment was arranged as a randomized complete block design with six replicates in 1991 and four replicates in 1992. Individual plots were 2 m by 6 m in 1991 and 3 m by 6 m in 1992.

Both years of the Glenlea experiments were established on land that had been cropped to barley the previous year. Canola was sown the year previous to the 1991 experiment at Portage la Prairie, while the 1992 experiment was established on land that had been cropped to wheat the previous year.

Triple superphosphate fertilizer (0-46-0) was applied at 60 kg P_2O_5 ha^{-1} to the 1991 sites, and 40 kg P_2O_5 ha^{-1} in 1992. The fertilizer was broadcast and cultivated to an approximate 10 cm depth in all cases except Portage in 1991, where the fertilizer was banded (no prior cultivation) with a press drill (15 cm row spacing) to about 5 cm. All plots were harrowed three times in opposing directions prior to seeding. The Portage site in 1991 was sprayed with glyphosate (1.23 kg active ingredient [a.i.] ha^{-1}) two days prior to seeding.

Alfalfa seed was inoculated with commercial *Rhizobium meliloti* L. peat based inoculum (Type A, LiphoTech Ltd.) before seeding, which was carried out at a rate of 800 readily germinable seeds per m^2. Alfalfa plots were broadcast seeded by mixing the seed with sand and spreading by hand, except at Portage in 1991 where a press drill (15 cm row spacing) with a cone attachment was used. All plots were packed immediately after seeding. Alfalfa seed was placed less than 0.5 cm deep when broadcast seeding was used, while drilled seed was sown about 2.5 cm deep. Barley was seeded at a rate of 122 kg ha^{-1} in 1991, and 126 kg ha^{-1} in 1992, with a press drill (15 cm row spacing) to a 2.5 cm depth. Alfalfa and barley were sown in the third week of May at Glenlea, and the last week of May at Portage, in both years.

Nitrogen fixation was measured in both years using the ^{15}N isotope dilution technique (McAuliffe et al., 1958) as well as the difference method in 1992. Argyle barley and Ineffective Saranac were the ^{15}N reference crops in 1992, while only the barley was used in 1991. Ineffective Saranac was the non-fixing control crop for the difference method. As it was difficult to determine which method was providing the best estimate of N_2 fixation, the average of the difference method and the two isotope dilution calculations (barley and Ineffective Saranac as reference crops) was used to estimate N_2 fixation in 1992. Boddey et al. (1990) suggested that the most practical approach in estimating N_2 fixation by the isotope dilution technique was to average values determined from a range of reference crops, as the true value of soil ^{15}N enrichment was likely represented in this range.

The proportion of nitrogen derived from the atmosphere (%Ndfa) was calculated for the ^{15}N isotope dilution technique using the following formula:

$$100 - \left(\frac{\%^{15}\text{N Excess in the Fixing Crop}}{\%^{15}\text{N Excess in the Reference Crop}} \times 100 \right)$$

The percentage ^{15}N Excess was determined by subtracting the natural abundance of ^{15}N (0.3663%) from the $\%^{15}N$ abundance of the analyzed samples. The quantity of nitrogen fixed (QNF) was determined by multiplying the total amount of nitrogen in the sampled crop (kg N ha^{-1}) by %Ndfa/100.

The quantity of nitrogen fixed by the difference method was determined by subtracting the total amount of nitrogen present in the reference crop from the total amount present in the N_2 fixing crop. %Ndfa for the difference method is subsequently calculated by taking the amount of nitrogen fixed as a proportion of the total amount of nitrogen present in the fixing crop.

For the isotope dilution technique, ^{15}N labelled $(NH_4)_2SO_4$ fertilizer (15% atom enrichment) was applied at a rate of 10 kg N ha^{-1} in 1 L of water with a pressurized garden sprayer to 1 m^2 subplots at Glenlea in 1991. At Portage in 1991, the same rate of enriched fertilizer was mixed with sand and broadcast by hand across the 1 m^2 subplots, followed by an application of 4 to 5 L of water with a pressurized backpack sprayer. In 1992, ^{15}N labelled $(NH_4)_2SO_4$ (10% atom enrichment) was applied at 15 kg N ha^{-1} in 0.75 L of water with a pressurized garden sprayer to 1 m^2 subplots at both sites. Non-labelled $(NH_4)_2SO_4$ was hand broadcast on the remainder of the plots at the same rates used for ^{15}N application in all cases. The plots were lightly raked in two opposing directions immediately after fertilizer addition. $(NH_4)_2SO_4$ application always took place after seeding but prior to crop emergence. Both sites in both years were irrigated as necessary until the crop emerged.

A combination of herbicides and hand-weeding were employed for weed control. Sethoxydim (0.199 kg a.i. ha^{-1}) was used at Glenlea in both years and Portage la Prairie in 1992. Imazethapyr (0.035 kg a.i. ha^{-1}) was applied at Portage la Prairie in 1991 and Glenlea in 1992. 2,4-DB (1.41 kg a.i. ha^{-1}) was employed at Portage la Prairie in 1992. Grasshoppers were present at Glenlea in 1991, and were controlled with a split application of carbaryl (1.42 kg a.i. ha^{-1}) and malathion (0.805 kg a.i. ha^{-1}).

Above ground biomass accumulation was measured on an approximate bi-weekly basis using 0.25 m^2 samples clipped to the soil surface. Alfalfa treatments were cut twice for hay, except at Glenlea in 1991 (one hay cut only), with the subsequent regrowth incorporated into the soil with the crowns and roots. The first cut was taken in the last week of July or the first week of August, while the second hay harvest was taken in the first week of September. Non-dormant cultivars generally bloomed earlier than the dormant types, and were usually more advanced than the dormant cultivars at each hay cut. ^{15}N subplots were removed by hand immediately prior to hay harvest and fall soil incorporation. The approximate inner 0.64 m^2 of each 1 m^2 subplot was removed to ensure sampling of plants uniformly exposed to ^{15}N labelled fertilizer. In 1991, a forage plot harvester was used to remove a 1.5 m strip in the center of each alfalfa plot. Sub-samples were taken to determine forage moisture contents and calculate plot dry weights. Final plot yield in 1991 was determined by adding the dry weight of the harvested plot strip, the ^{15}N subplot, and the weights of any previously sampled biomass accumulation areas. In 1992, two 1 m^2 areas per plot were randomly designated at the beginning of the year and used for all yield determinations. The forage harvester was used to remove any remaining alfalfa after sampling had taken place. Plots in both years were clipped to a 7.5 to 10 cm stubble height.

The amount of herbage present for soil incorporation was measured in mid to late October. In 1991, two 0.25 m^2 samples were taken, while the 1 m^2 designated yield subplots were used in 1992. Alfalfa was clipped to the soil surface. In 1992, root contribution was measured by taking twelve soil cores (10 cm width, 15 cm depth) per plot, four from the ^{15}N subplot and the remainder from each of the two yield subplots. Roots were washed, clipped to a 10 cm length, dried and weighed. Although attempts were made to isolate individual plants when taking the soil cores, there were often as many as nine or ten plants per core. Final plot root yield was determined by multiplying the average individual root weight by the average number of plants per core and then by the average number of individually appearing plants per plot (determined by plant counts prior to root sampling using two 0.25 m^2 quadrats per plot).

The 1991 plots at Glenlea were roto-tilled three times (10 cm depth) in a lengthwise direction immediately after final sampling. At Portage la Prairie, the plots were roto-tilled the following spring before any substantial regrowth had occurred (less than 10 cm). They were worked twice in a lengthwise fashion to a depth of 10 cm on May 19, 1992. Plant counts (two 0.25 m^2 quadrats per plot) were performed prior to tillage to determine alfalfa survival. Plots were not roto-tilled in the fall of 1992, and plant survival counts (two 0.25 m^2 quadrats per plot) were performed on April 20, 1993 at Glenlea and April 26, 1993 at Portage la Prairie.

The ^{15}N subplots of the barley control plots were harvested at the soft dough stage in the last week of July or the first week of August, making sure to harvest the inner 0.64 m^2 of the subplots, such that sampled plants had equal access to ^{15}N labelled fertilizer. The whole shoot was harvested, except for 10 cm of stubble that was the cutting height. The remaining barley was harvested with a plot combine at maturity. All plant material was oven dried at 70°C for a minimum of 72 hours prior to weighing.

After drying, all plant material was ground in a Wiley mill to pass a 2.0 mm screen. Material to be analyzed for ^{15}N:^{14}N ratios was ground once more in a Udy cyclone mill to pass a 0.5 mm sieve (passed three times in 1992). Material from the biomass accumulation samplings was analyzed for N concentration using the kjeldahl technique in 1991, and a Leco N Determinator (Model FP 428, Mississauga, Ontario) in 1992. N concentration of the material harvested for hay or incorporated into the soil was determined by the micro-kjeldahl procedure (Bremner and Mulvaney, 1982) in 1991 and by a nitrogen determinator (see below) in 1992. After determination of total nitrogen content by the micro-kjeldahl procedure in 1991, the titrated solution was evaporated to a small volume (less than 5 mL) and subsequently analyzed for ^{15}N:^{14}N ratios, performed at the laboratory of Dr. A. Blackmer, Iowa State University, on a Finnigan Mat 250 mass spectrometer. ^{15}N:^{14}N ratios were determined in 1992 on an ANCA-MS nitrogen determinator/mass spectrometer (Europa Scientific, Crewe, U.K.), equipped with a single inlet and triple collectors, in the laboratory of Dr. C. van Kessel, University of Saskatchewan.

In 1992, Katepwa wheat (*Triticum aestivum* L.) was sown into the previous year's plots. In addition to roto-tilling (see previous), the plots were harrowed once prior to seeding. Small strips of volunteer alfalfa were sprayed with a pressurized garden sprayer using a 10.7 g L^{-1} solution of glyphosate before seeding. Wheat was seeded at Glenlea on May 16, 1992 and Portage la Prairie on May 23, 1992 at rates of 133 and 137 kg ha^{-1}, respectively. Triple superphosphate was applied with the seed at a rate of 49 kg P_2O_5 ha^{-1} at Glenlea and 58 kg P_2O_5 ha^{-1} at Portage la Prairie. In both cases, a press drill (15 cm row spacing) was used for seeding (2.5 cm depth). Clopyralid (0.148 kg a.i. ha^{-1}) was employed for weed control at both sites.

Wheat was harvested at the soft dough stage on August 13, 1992 and August 16, 1992 for Glenlea and Portage la Prairie, respectively. Five rows (1 m length) were cut to a stubble height of 10 cm. The shoot material was dried (70°C, minimum 72 hours) and weighed. The remainder of the plot was harvested with a plot combine at maturity on September 16, 1992 at Glenlea and September 23, 1992 at Portage la Prairie. The grain was cleaned, weighed, and tested for moisture content with a grain moisture tester. Plot grain yield was determined on a dry weight basis, with the previous removal of the total shoot subplot (0.75 m^2) taken into consideration. Shoot material was ground in a Wiley mill to pass a 2.0 mm screen, while the grain was ground in a Udy cyclone mill to pass a 1.0 mm screen. N concentration was determined on a Leco N Determinator (Model FT 428, Mississauga, Ontario).

Plant survival data from Portage la Prairie in 1992 and Glenlea in 1993 showed non-homogenous variance and were arcsin transformed before an analysis of variance (ANOVA) was performed. ANOVA was conducted on all data at the 0.05 and 0.01 levels. Mean separations were performed by the LSD test at the 0.05 level, but only if the ANOVA proved significant. Single degree of freedom orthogonal contrasts were used to compare non-dormant cultivars with moderately dormant and dormant cultivars in 1992. Excalibur, Saranac, and Algonquin were grouped together as dormant cultivars as they were similar in fall growth and had a very different fall dormancy response than either of the non-dormant cultivars. Contrasts were also performed to compare Nitro and CUF 101.

3.4. RESULTS AND DISCUSSION

3.4.1. Seasonal Growth Patterns

Above-ground biomass accumulation and fluctuations in N concentrations are shown for Portage la Prairie in Figures 1 through 4 (trends at Glenlea were similar; see Appendix Figures A1 through A4). In general, the non-dormant Nitro (and CUF 101 in 1992) grew faster immediately after seeding, and had faster regrowth after each of the hay cuts (Figures 1 and 2), with the exception of Glenlea in 1991. This initial growth advantage by the non-dormant alfalfas was not usually maintained at the time of hay harvest, as forage yields for all cultivars tended to be similar (see below). The non-dormants did, however, maintain their growth after the second hay cut into the late fall, while the dormant alfalfas slowed their growth in response to cooler temperatures and shorter daylengths. The rapid early growth and regrowth advantage of the non-dormant cultivars could be important in early weed competition and, consequently, initial stand establishment.

N concentrations were lower in the non-dormant cultivars for the majority of the sampling dates, except in the fall (Figures 3 and 4). This might be related to the stage of the non-dormant alfalfas at sampling relative to the dormant alfalfas.

The non-dormants were generally in a more advanced stage at each of the sampling dates due to faster growth rates, and plant N was likely being diluted by the accumulation of structural and non-structural carbohydrates. The higher N concentrations in the non-dormant alfalfas after the second cut was likely related to the continued growth of these cultivars into the fall, while the other varieties became dormant shortly after the second harvest. Single degree of freedom orthogonal contrasts between Nitro and CUF 101 (not shown) were not significant at any of the sampling dates for above ground biomass accumulation or N concentration.

3.4.2. Herbage Harvested for Hay

Alfalfa cultivars were similar in terms of the yield, N concentration, and the nitrogen yield of the herbage harvested for hay in 1991 at both sites (Table 2). However, Nitro had a significantly lower N concentration (12% less than the dormant average) for the second cut at Portage la Prairie. A second hay cut was not taken at Glenlea in 1991 due to dry conditions and poor regrowth after the first cut.

Alfalfa cultivars also yielded similarly in 1992 at Glenlea, although differences were observed at Portage la Prairie (Table 3). Saranac yielded significantly more hay than the non-dormants and Algonquin at the first hay cut, while Nitro and CUF 101 were generally better than the dormant cultivars (12% greater than the dormant average) at the second cut (contrast significant). N concentrations were not different at the first cut at both sites, but were higher by 5 to 21% in the dormant cultivars at the second hay cut (contrasts significant). Nitrogen yields were similar among cultivars at the first hay cut at Glenlea and the second cut at Portage la Prairie in 1992, although Saranac had a significantly greater N yield for the Portage la Prairie first cut relative to the other cultivars. As well, the dormant alfalfas yielded significantly more nitrogen (average 20% more) in the herbage at the Glenlea second hay cut when compared with the average of Nitro and CUF 101.

Others have reported first and second cut yields for seeding year stands in the range of 2400 to 3900 kg ha^{-1}, with N concentrations and N yields ranging from 24.6 to 39.0 mg g^{-1} and 82 to 115 kg N ha^{-1}, respectively, for dormant and non-dormant alfalfa cultivars (Hesterman et al., 1986; Groya and Sheaffer, 1985). The majority of observations in this experiment are within these reported ranges. In general, alfalfa cultivars were very similar at the first hay cut, while the non-dormants yielded slightly higher (Portage la Prairie, 1992 only), with lower N concentrations, than the dormant alfalfas at the second hay cut. This could be attributed to the advanced stage of the non-dormant cultivars at the time of the second harvest. Non-dormant alfalfas ranged from first flower to 20% bloom, while the dormant cultivars ranged from mid-bud to 5% bloom at the time of cutting. Later cut alfalfa tends to yield slightly higher with a lower N concentration than earlier cut alfalfa. Sheaffer (1983) observed consistent increases in yield and progressive decreases in N concentration in alfalfa that was cut at the mid-vegetative, early flower and late flower stages.

3.4.3. Herbage and Roots Incorporated into the Soil in the Fall

Nitro yielded significantly more herbage for soil incorporation in the fall compared with Algonquin, but not Excalibur, at Glenlea, and proved superior to both dormant cultivars at Portage la Prairie in 1991 (Table 4). Nitro contributed an additional 737 (36% increase) and 802 kg ha^{-1} (39% increase) of herbage to the soil at Portage la Prairie relative to Excalibur and Algonquin, respectively. N concentrations and N yields were similar among cultivars at Glenlea, but were significantly higher for Nitro at Portage la Prairie in 1991. Nitro had 7.0 mg g^{-1} more N (21% increase) in the herbage tissue, and provided 34.5 kg ha^{-1} (50% increase) more N for soil incorporation when compared with an average of the dormant cultivars. Differences were not apparent at Glenlea in 1991 due to poor regrowth caused by dry conditions after the first hay cut, and a failure to resume growing when moisture conditions improved later in the season.

Similar trends were observed in 1992 at both sites (Table 5), with significantly higher herbage yields, N concentrations, and N yields in the incorporated herbage for Nitro and CUF 101 compared with the dormant cultivars. The non-dormant alfalfas added an additional 1031 kg ha^{-1} of herbage to the soil, an increase of 39% over the dormant cultivar average for both sites. The average N concentration was 12.3 mg g^{-1} greater (33% increase), and there was 57.8 kg ha^{-1} more N (58% increase) present for soil incorporation, in the herbage of the non-dormant alfalfas relative to the average of the dormant cultivars at both sites.

The greater herbage production by Nitro and CUF 101 in the fall is likely related to their non-dormant characteristic that allows prolonged growth late in the season. Sheaffer et al. (1988) measured significantly greater fall herbage yields in Nitro relative to Saranac under two of three harvest management systems. When two summer cuts were taken at bud and the fall growth was sampled at first flower, Nitro and Saranac produced 1760 and 1500 kg ha^{-1} of fall forage, respectively. Under a system that utilized three summer cuts (at bud) with the fall regrowth harvested at bud, 1260 and 500 kg ha^{-1} of fall forage was produced by Nitro and Saranac, respectively. Nitro and Saranac were similar in a system that employed two summer cuts (first flower) with the regrowth sampled at first flower in the fall, suggesting that the relative increases in fall herbage production observed by non-dormant cultivars can be influenced by harvest management.

There were no significant differences among cultivars in terms of the upper root yield (top 10 cm of root sampled) at either site in 1992 (Table 5), but Nitro and CUF 101 did have significantly higher N concentrations in the roots, and generally yielded more N for soil incorporation than the dormant cultivars (contrasts significant). N concentrations and N yields were higher in the non-dormant alfalfas by 7.6 mg g^{-1} and 55.9 kg ha^{-1}, corresponding to increases of 30% and 43%, respectively, when compared with the dormant cultivar average over both sites.

One of the main criteria in developing Nitro was selection for large root mass and high root N concentration (Barnes et al., 1988). Sheaffer et al. (1989) found that Nitro had a significantly greater root yield (1757 kg ha^{-1}) and a higher N concentration (27 mg g^{-1}) relative to representative dormant cultivars with root yields and N concentrations of 1506 kg ha^{-1} and 24 mg g^{-1}, respectively. MN ROOT N (Syn 2 generation of the commercial cultivar Nitro) and Saranac produced root yields and N concentrations of 3800 and 3200 kg ha^{-1}, and 26 and 22 mg g^{-1} in a system where two summer cuts were taken, while yields and N concentrations of 4000 and 3900 kg ha^{-1}, and 28 and 26 mg g^{-1} were measured when only one summer harvest was employed, respectively (Hesterman et al., 1986). Griffin and Hesterman (1991), however, noted lower N yields in the roots of Nitro (92 kg N ha^{-1}) relative to Saranac alfalfa (109 kg N ha^{-1}) when managed similarly (three hay cuts, fall stubble and roots incorporated). This observation might still be related to harvest management as the second hay cut was taken in mid-August, thereby allowing sufficient time for herbage and root growth in Saranac before the onset of dormancy and the final cut in October. The root growth advantage of the non-dormant Nitro could have therefore been masked by the time of cutting.

3.4.4. Total Herbage Production

The dry matter and nitrogen yields of the total herbage produced (hay cuts and incorporated material) over the season are shown for the two sites in Table 6. Nitro was similar to the dormant cultivars in terms of herbage dry matter and nitrogen yields in 1991 at Glenlea, but was significantly greater than Algonquin at Portage la Prairie. Total herbage production at Glenlea in 1991 was notably lower than Portage la Prairie due to the poor growth (resulting in fewer hay cuts) caused by dry conditions in the summer (Table 1).

Significant differences were not present among cultivars at Glenlea in 1992, similar to the 1991 data. At Portage la Prairie, however, the non-dormant alfalfas generally had significantly greater herbage dry matter and N yields than the dormant cultivars (contrasts significant), although moderately dormant Saranac was not different from either Nitro or CUF 101. The non-dormants produced 13% more total herbage, and yielded 17% more total nitrogen, than the average of the dormant cultivars.

It should be noted that CUF 101 was never different from Nitro (contrasts non-significant, not shown) in terms of the herbage removed for hay or the roots and herbage incorporated into the soil. Sheaffer et al. (1989) did not find significant differences between CUF 101 and Nitro in terms of the forage removed for hay or the crown and root yields and N concentrations, but did observe that Nitro produced significantly more forage in the fall (11% more) and had significantly greater N yields in the incorporated crowns and roots (13% more) relative to CUF 101.

3.4.5. N_2 Fixation

The percentage of nitrogen derived from the atmosphere (%Ndfa) was similar among alfalfa cultivars at both sites in 1991, ranging from 22.2 to 83.4% (Table 7). The quantity of N_2 fixed (QNF) was also similar among cultivars for the hay cuts at both sites, although significantly more fixed nitrogen was returned to the soil in the fall with the herbage of Nitro relative to both dormant alfalfas at Portage la Prairie, and Algonquin at Glenlea. Nitro added 51 and 21% more fixed N_2 to the soil via the incorporated herbage compared with the dormant varieties at

Portage la Prairie and Glenlea, respectively, in 1991. As %Ndfa was similar among cultivars, the greater quantity of N_2 fixed in Nitro was likely a function of it's non-dormant ability to grow late into the fall, resulting in greater amounts of incorporated herbage, and consequently more fixed N_2 being contributed to the soil. The notably smaller differences in the amounts of N_2 fixed between Nitro and the dormant alfalfas at Glenlea in the fall of 1991 was likely related to the poor growth exhibited by all cultivars after the first hay cut until the season's end.

The total amounts of N_2 fixed in the herbage over the 1991 season are shown in Table 10. The quantities of N_2 fixed at Glenlea were substantially lower than Portage la Prairie, due to lower %Ndfa values and reduced herbage yields. Nitro fixed significantly more nitrogen than Algonquin, but not Excalibur, at Glenlea in 1991. Significant differences in total seasonal N_2 fixation were not observed at Portage la Prairie.

As in 1991, %Ndfa in 1992 was similar among cultivars for the hay cuts at both sites (range 32.4 to 75.7%, Table 8). However, unlike the 1991 data, significant differences in %Ndfa were detected among treatments for the incorporated herbage (Table 9). The contrasts proved highly significant at both sites, with the non-dormant and dormant cultivars averaging 71.0 and 48.2% of their N from symbiotic N_2 fixation, respectively.

QNF followed a pattern similar to %Ndfa in 1992, with similar amounts of N_2 being fixed among cultivars at the hay cuts (except the Glenlea second cut, where the non-dormants had a slightly lower QNF) and significantly more N_2 being fixed by the non-dormants later in the fall (Tables 8 and 9). Nitro fixed more N_2 than CUF 101 at the Portage la Prairie second hay cut (contrast significant, not shown) but was, in general, very similar to CUF 101 in %Ndfa and QNF, which is consistent with the herbage and root yield data. An average of 69.4 and 19.4 kg ha^{-1} of fixed N_2 was incorporated with the herbage of the non-dormant and dormant cultivars, respectively, in the fall of 1992. Part of the increase in QNF for the non-dormant cultivars in 1992 was caused by greater %Ndfa (unlike 1991), although the extended growth of the non-dormant cultivars in the fall and the correspondingly greater amount of incorporated herbage played a substantial role in increasing QNF.

The upper roots of the non-dormants also had significantly higher levels (contrasts significant) of %Ndfa (26% greater) and QNF (60% greater) relative to the dormant cultivars (Table 9). The amount of fixed N_2 added to the soil by Nitro and CUF 101 through upper root contribution was similar to that added with the herbage at Portage la Prairie, and almost double the amount worked in with the herbage at Glenlea. The QNF in the roots as presented in this experiment are no doubt underestimates of the true values because only the top 10 cm of the root system was measured, thereby stressing the importance of considering roots when calculating N contributions to the soil by N_2 fixation.

Total amounts of N_2 fixed in the herbage and upper roots in 1992 are shown in Table 10. The non-dormant cultivars fixed significantly more nitrogen over the season than the dormant varieties (contrasts significant). Respective amounts of N_2 fixed for the non-dormant and dormant alfalfas were 290.0 and 201.0 kg N ha^{-1} at Portage la Prairie, and 230.9 and 134.9 kg N ha^{-1} at Glenlea, in 1992. The relatively larger amounts of N_2 fixed in the fall herbage and upper roots of the non-dormants was great enough to observe significant differences in the total seasonal amounts between the non-dormant and dormant cultivars.

Heichel et al. (1981) reported values ranging from 19 to 67% for %Ndfa, and 8 to 87 kg N ha^{-1} for QNF, for two dormant cultivars that had been sampled four times in the seeding year. Values in the present experiment tend to fall in the middle to upper end of this range, with lower values observed earlier in the season. Soil tests in the spring of 1992 indicated levels of available nitrate (101 and 141 kg N ha^{-1}, at Glenlea and Portage la Prairie, respectively) that could have inhibited early N_2 fixation.

Hesterman et al. (1987) showed that an average of 75 and 50 kg ha^{-1} of fixed N_2 was incorporated into the soil with the herbage, crowns and roots of MN ROOT N and Saranac, respectively, when cut three times in the seeding year. Nitro and Saranac contributed an average of 151.9 and 61.8 kg ha^{-1} of fixed N_2, respectively, in the herbage and roots incorporated into the soil in the fall of 1992 of the present study.

If a crop is to be successful as a fall nitrogen source, there should be a net positive effect on the soil nitrogen balance, such that fertilizer requirements can be reduced in the subsequent year. An average of 84.0 and 95.1 kg ha^{-1} of soil N (calculated by subtracting fixed N_2 from total N) was removed with the first and second hay harvests while an average of 154.1 and 53.4 kg ha^{-1} of fixed N_2 was incorporated into the soil with the herbage and roots (upper 10 cm) of the non-dormant and dormant cultivars, respectively, in 1992. The net N balance was therefore 70.1 and -41.7 kg N ha^{-1}, for the non-dormant and dormant alfalfas, respectively. Under the harvest management employed in this experiment, the non-dormant cultivars had a positive effect on soil N, while the dormant alfalfas exerted a negative effect on the end of season soil N balance. Altering the harvest management so that only one hay cut is taken, or cutting and leaving the herbage on the soil surface may help to improve the soil N balance (Sheaffer et al., 1989). Changing the time of cutting, however, may serve to mask the fall growth advantage of the non-dormant cultivars if maximum yield in the dormant cultivars is reached before the onset of fall dormancy. Another factor to consider is the initial soil N status; if alfalfa is established on low N soils the %Ndfa might be increased which could also work to improve the net soil N balance. As well, measurement of the entire root system (instead of the top 10 cm) would have undoubtedly increased the N contribution to the soil, resulting in a more positive net N balance.

3.4.6. Methods of Calculating N_2 Fixation

The three methods of calculating N_2 fixation, isotope dilution with Ineffective Saranac as the reference crop (IDS), isotope dilution with barley as the reference crop (IDB), and the difference method using Ineffective Saranac as the reference (DS), generally gave very different results. Respective %Ndfa values for IDS, IDB and DS were 57.9, 38.6, and 54.0% for the first cut, 46.7, 77.5 and 59.3% for the second cut, 39.0, 77.9, and 65.3% for incorporated herbage, and 47.1, 69.3, and 58.1% for incorporated roots when averaged over all treatments and both sites in 1992.

A number of researchers have used barley as the ^{15}N reference crop (see review by Chalk, 1985). Although good success in estimating N_2 fixation with barley as the reference crop has been reported for annual crops like soybeans and fababeans (Rennie, 1982; Rennie et al., 1976), barley may not be ideal as the control for crops having later maturity and different N uptake patterns (Witty, 1983). One of the primary assumptions of the isotope dilution technique is that the roots of the legume and reference crops absorb the same proportion of soil and applied fertilizer N during the growing period (McAuliffe et al., 1958). In this study, the barley was harvested at the same time as the first alfalfa cut, and the ^{15}N enrichment of that sampling was used in N_2 fixation calculations for the second cut and incorporated herbage and roots late in the fall. Changes in soil ^{15}N enrichment likely occurred during the period after barley harvest, hence the estimates of N_2 fixation after the first hay cut may be inaccurate, likely giving overestimates of %Ndfa due to ^{15}N dilution after the first cut. Despite this, qualitative comparisons can still be made among cultivars in this study using barley as the ^{15}N reference crop.

Comparisons between the isotope dilution technique and the difference method have been made in a number of studies, generally with inconsistent results (Chalk, 1985). Henson and Heichel (1984) evaluated N_2 fixation in Saranac alfalfa using Ineffective Saranac as the reference crop, for the isotope dilution and difference techniques. They found that the two methods produced similar results, although the difference method was more variable and tended to underestimate N_2 fixation slightly. Factors which could cause error in the estimation of N_2 fixation by violating the assumption that fixing and control crops receive N from fertilizer and soil in the same proportions include: differences in ability to assimilate N from different soil depths, in association with differences in the distribution of added N with soil depth, and differences in N assimilation rate in conjunction with changes in isotopic composition of added plus soil N with time (Ledgard et al., 1985). These factors might be used in an attempt to explain the different results between the difference and isotope dilution techniques in this experiment. It is not unreasonable to assume that the Ineffective Saranac reference had a different rooting pattern than the fixing cultivars as it yielded 23.8% less root biomass (individual plant basis) in the top 10 cm of soil when compared with an average of the other alfalfas. If the fixing cultivars had access to leached nitrogen that was unavailable to the Ineffective Saranac reference because of a more shallow root system, the difference method would have overestimated N_2 fixation. As well if the soil enrichment was non-uniform with depth, such that more ^{15}N was available to the fixing alfalfas than the reference crop, N_2 fixation might have been underestimated. Such a scenario could have arisen if some of the labelled fertilizer leached out of the rooting zone of the Ineffective Saranac reference, but was still accessible to the fixing cultivars, or if there was a dilution of ^{15}N labelled fertilizer in the surface soil layers, but not at depth, caused by microbial immobilization of the fertilizer and (or) mineralization of non-labelled soil organic matter.

3.4.7. Plant Survival

Non-dormant cultivars had significantly lower survival rates compared with the dormant alfalfas (Table 11). At Portage la Prairie in 1992 and 1993, plant survival was very poor in the non-dormants (less than 8.0%) compared with the dormant survival rate (52.9 to 70.1%). Survival levels at Glenlea in 1993 were substantially higher in all cultivars with the best survival in dormant Algonquin (87.1%) and the worst in non-dormant CUF 101 (46.4%). Significantly more Nitro plants survived the winter (62.2%) compared with CUF 101 at Glenlea in 1993. There was slightly more snowfall at Glenlea (128.4 cm) compared with Portage la Prairie (109.8 cm) over the 1992-1993 winter, possibly giving better plant insulation that may have resulted in the higher survival rates observed at the Glenlea site in the spring of 1993. The substantial reduction in winter survival observed in the non-dormant alfalfas should facilitate subsequent crop establishment relative to a preceding dormant cultivar. It is likely, however, that volunteer alfalfa control will be a concern, even with the higher levels of winter kill observed in the non-dormant alfalfas.

Barnes et al. (1988) report that Nitro will not reliably survive most upper Midwest U.S. winters, but it may survive during winters with sufficient snow cover and/or mild temperatures. Sheaffer et al. (1992) measured the survival of a number of alfalfa cultivars subjected to different harvest schedules at five locations in Minnesota. Dormant Rambler, moderately dormant Saranac, and non-dormant Nitro had average stand survivals of 84% (range 56% to 95%), 63% (range 27% to 95%) and 14% (range 3% to 25%), respectively, which is similar to the results observed at Portage la Prairie.

3.4.8. N Uptake in the Subsequent Wheat Crop

A nitrogen response to alfalfa was not observed at Glenlea in 1992, although N uptake was significantly higher in wheat following alfalfa (compared with wheat following barley) at Portage la Prairie (Table 12). Total shoot N uptake in wheat was not different among alfalfa cultivars, but they all supplied more N than the previous barley crop. Grain N uptake at Portage la Prairie was highest in the wheat following Nitro, which was significantly better than wheat following either Excalibur or barley, but not Algonquin. Average total shoot and grain N uptake in wheat following alfalfa was 117.0 and 69.2 kg N ha^{-1}, respectively, compared with 74.9 and 58.6 kg N ha^{-1} for barley at Portage la Prairie.

Increases in yield and N uptake have been observed when a non-leguminous crop follows a single year of alfalfa (Badaruddin and Meyer, 1990; Bruulsema and Christie, 1987; Hesterman et al., 1986). Results in this experiment are similar to those of Griffin and Hesterman (1991), who observed significant increases in potato vine dry matter yield, N concentration, and N yield when Saranac and Nitro alfalfa, rather than potatoes, were the preceding crops. There were no differences, however, between Saranac and Nitro in terms of the subsequent potato vine growth. Hesterman et al. (1986) observed similar yield responses when corn was preceded by MN ROOT N or Saranac. Average subsequent corn yields were 6800 and 6850 kg ha^{-1} for MN ROOT N and Saranac, respectively, compared with 3700 kg ha^{-1} for corn preceded by corn (no N fertilizer applied).

3.5. SUMMARY AND CONCLUSIONS

Nitro was, in general, very similar to the dormant cultivars in terms of the yield and N_2 fixed in the herbage removed for hay. Small discrepancies among cultivars might be explained by different stages of maturity at cutting time. Nitro was, however, superior as a fall green manure crop, as it returned significantly more biologically fixed N_2 to the soil in the fall relative to the dormant cultivars. Nitro's ability to fix more N_2 in the fall comes, in part, from a greater specific fixation capacity (increased proportion of N from N_2 fixation, observed in 1992 only), but also from it's non-dormant ability to maintain growth late into the fall. Nitro was not significantly different from the other non-dormant cultivar tested, CUF 101, in terms of growth or N_2 fixation.

The survival of the non-dormant cultivars was significantly lower than the dormant alfalfas, and was likely low enough to facilitate the introduction of a subsequent crop at two of the three site years tested, relative to a preceding dormant cultivar. A nitrogen response was only detected at one of the two sites, and even then differences were mainly between the alfalfas and the previous barley, and not between alfalfa cultivars.

The higher levels of fixed N_2 returned to the soil in the fall by the non-dormant cultivars resulted in a positive net soil N balance that was not observed with the dormant alfalfas. Harvest management can have significant effects on the amounts of N returned to the soil, but may also have a profound influence on the growth advantage normally demonstrated by non-dormant alfalfas. Further work is necessary to fine tune the harvest management for Manitoba conditions, such that hay and plow down yields are optimized, and the yields of subsequent crops are better characterized. More work is necessary to determine if Nitro truly is superior to other non-dormant cultivars, as the observed differences between Nitro and the other alfalfas may simply be a function of it's non-dormant characteristic. Under the conditions and parameters examined in this study, however, Nitro does seem to be superior in fall growth and N_2 fixation compared with the dormant cultivars, and this, combined with it's similarity in performance as a hay source, gives it some advantage as a one year hay and green manure crop relative to the more traditionally used dormant alfalfa cultivars.

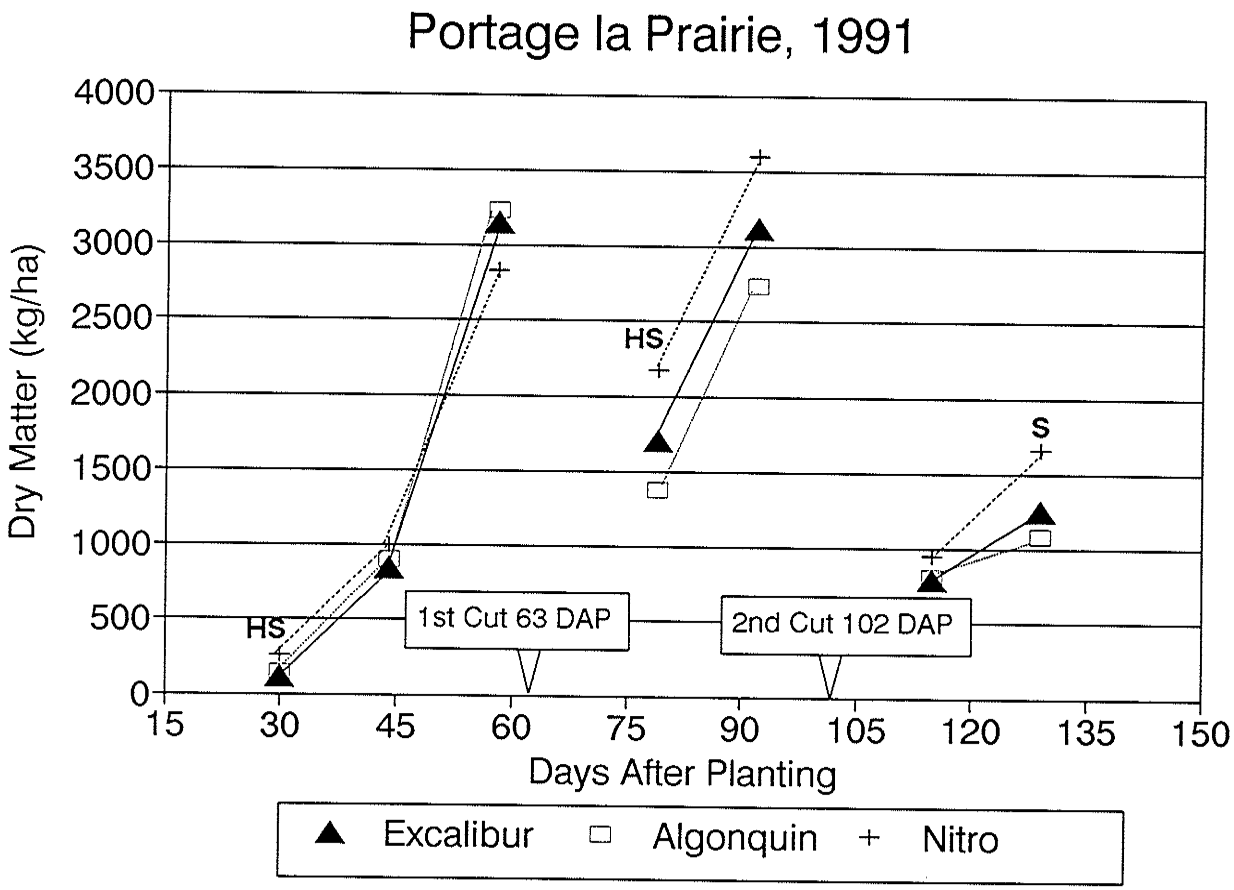


Figure 1. Seasonal above-ground biomass accumulation at Portage la Prairie in 1991. S, HS - Analysis of variance significant at the 0.05 and 0.01 levels, respectively.

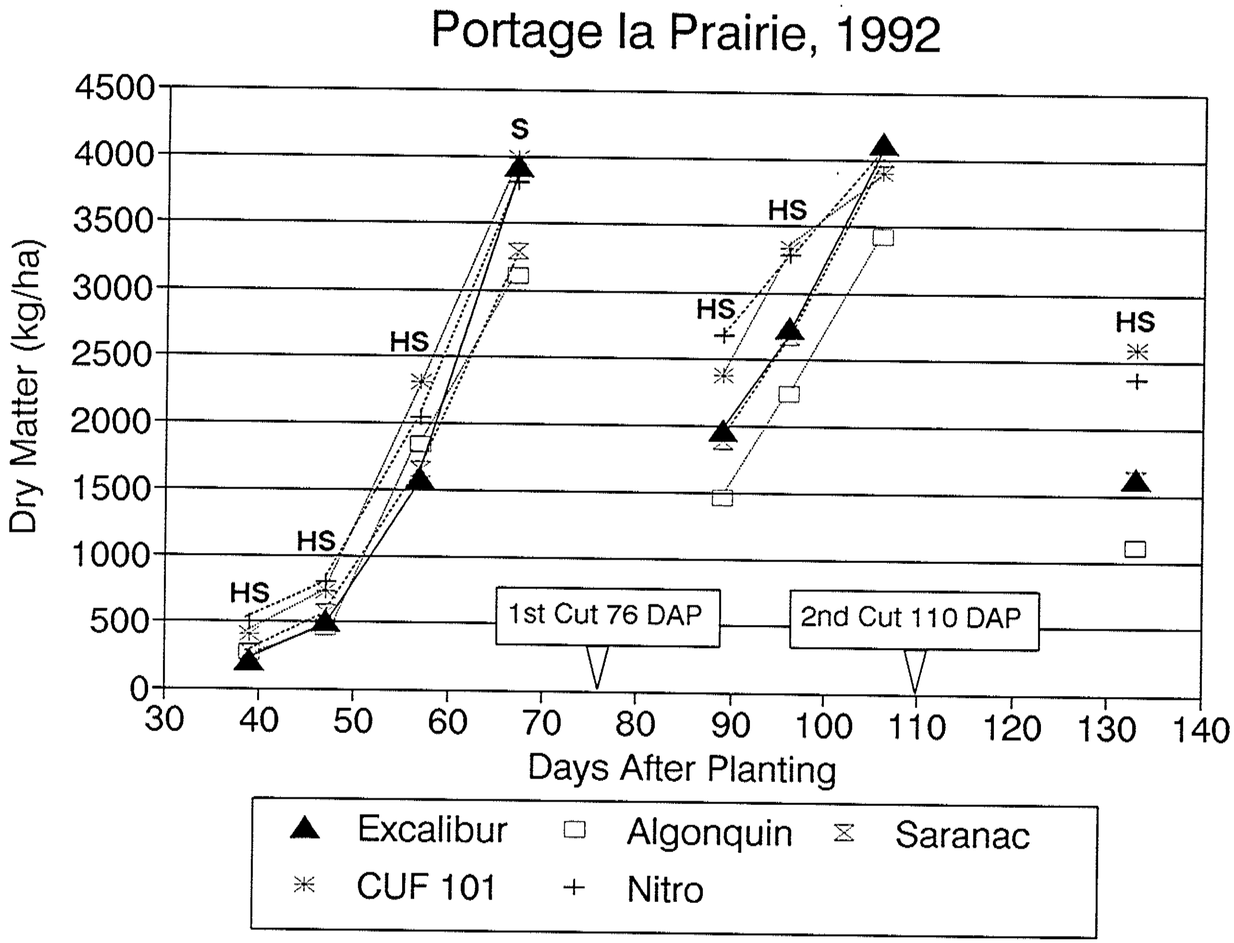


Figure 2. Seasonal above-ground biomass accumulation at Portage la Prairie in 1992. S, HS - Single degree of freedom orthogonal contrast (non-dormant vs dormant cultivars) significant at the 0.05 and 0.01 levels, respectively.

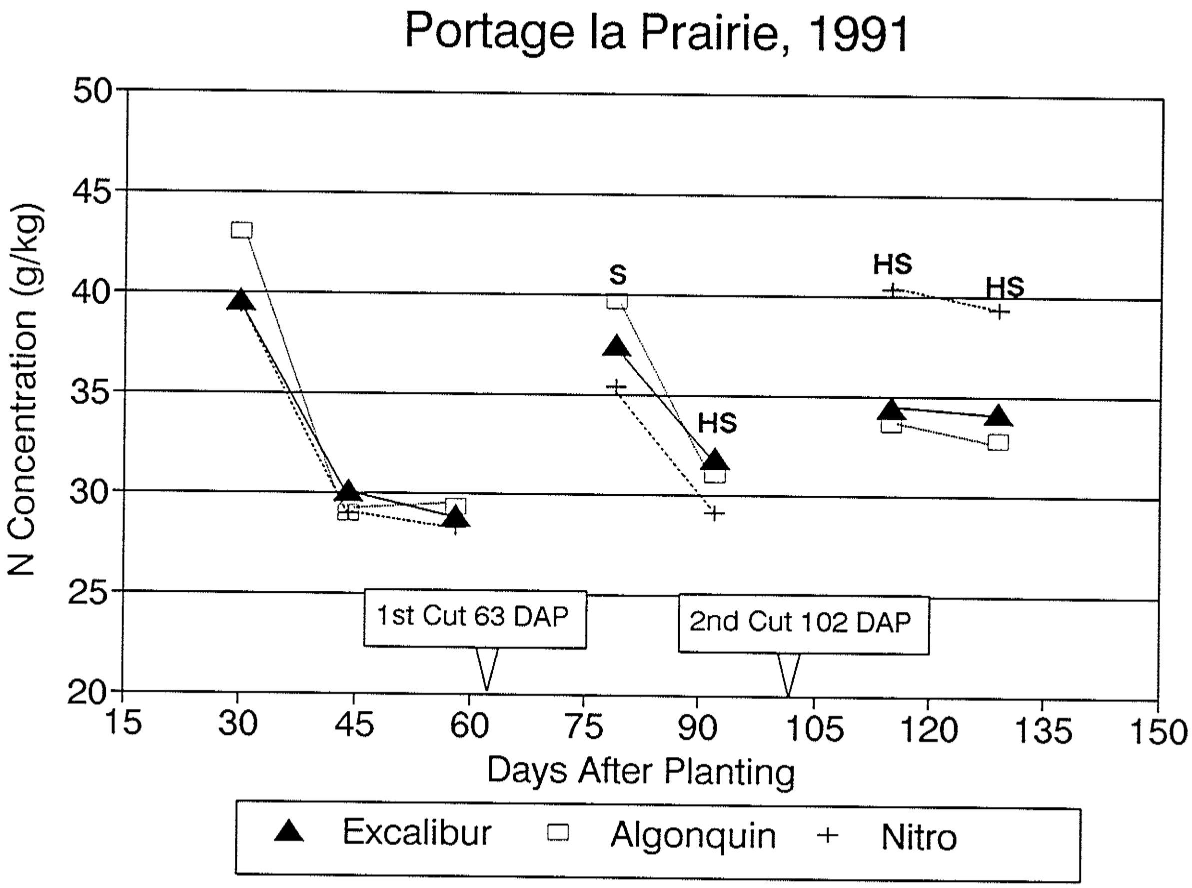


Figure 3. Seasonal changes in herbage N concentration at Portage la Prairie in 1991. S, HS - Analysis of variance significant at the 0.05 and 0.01 levels, respectively.

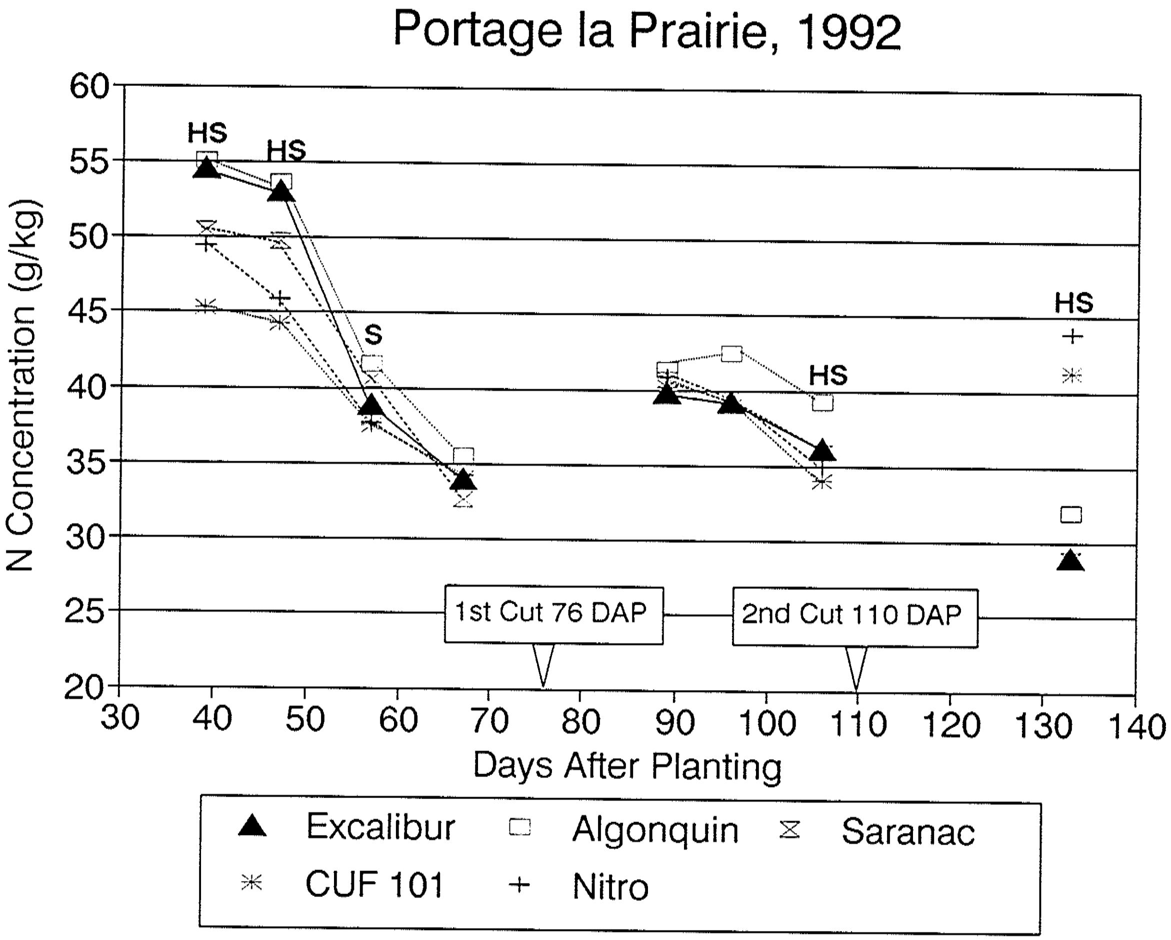


Figure 4. Seasonal changes in herbage N concentration at Portage la Prairie in 1992. S, HS - Single degree of freedom orthogonal contrast (non-dormant vs dormant cultivars) significant at the 0.05 and 0.01 levels, respectively.

Table 1. Precipitation and mean temperature (long term averages, 1991, and 1992) for the growing season months at Portage la Prairie and Glenlea, Manitoba.

Temperature and Precipitation	Month	Portage 1961 to 1990	Portage 1991	Portage 1992	Glenlea 1967 to 1990	Glenlea 1991	Glenlea 1992
Mean Temp. (°C)	May	11.6	14.1	12.8	11.9	14.3	13.2
	June	17.1	18.8	15.4	16.6	19.1	15.1
	July	19.8	19.5	16.3	19.4	19.2	15.2
	August	18.4	20.3	16.3	18.1	8.2	16.3
	September	12.5	12.2	10.8	12.3	23.2	11.4
	October	6.1	2.2	5.5	4.9	1.7	4.6
Precip. (mm)	May	56.8	59.0	12.6	56.8	82.0	26.4
	June	75.0	75.0	44.0	94.9	120.1	98.4
	July	76.9	95.0	109.0	70.6	110.8	95.8
	August	78.8	10.0	49.0	60.5	41.2	68.0
	September	50.1	68.0	50.0	52.9	92.8	70.4
	October	32.7	43.1	6.2	37.9	71.9	3.8

[z] Climatic information for Portage la Prairie derived from University of Manitoba and Environment Canada data.
[y] Climatic information for Glenlea derived from Environment Canada data.

Table 2. Herbage yield, N concentration (conc.), and N yield for the first and second hay cuts at Portage la Prairie, and the first hay cut at Glenlea, in 1991.

	Portage la Prairie, 1991						Glenlea, 1991		
	Cut 1			Cut 2			Cut 1		
Cultivar	Yield kg ha^{-1}	N Conc. g kg^{-1}	N Yield kg N ha^{-1}	Yield kg ha^{-1}	N Conc. g kg^{-1}	N Yield kg N ha^{-1}	Yield kg ha^{-1}	N Conc. g kg^{-1}	N Yield kg N ha^{-1}
Nitro	3584*a*	30.2*a*	98.1*ab*	4098*a*	26.7*b*	108.7*a*	4053*a*	24.8*a*	100.3*a*
Excalibur	3731*a*	31.2*a*	116.4*a*	3810*a*	30.0*a*	113.8*a*	4414*a*	26.0*a*	115.2*a*
Algonquin	3292*a*	30.0*a*	98.1*b*	3627*a*	30.7*a*	110.4*a*	4339*a*	24.4*a*	105.7*a*
CV (%)^{z}	16.4	6.5	16.6	17.1	10.4	15.5	10.6	14.6	18.0

^{z}Coefficient of variation (%).

a-b Means within a column followed by the same letter are not significantly different at $P < 0.05$ by LSD.

Table 3. Herbage yield, N concentration (conc.), and N yield for the first and second hay cuts at Portage la Prairie and Glenlea in 1992.

Portage la Prairie, 1992

	Cut 1			Cut 2		
Cultivar	Yield kg ha^{-1}	N Conc. g kg^{-1}	N Yield kg N ha^{-1}	Yield kg ha^{-1}	N Conc. g kg^{-1}	N Yield kg N ha^{-1}
Nitro	3489*b*	32.2*a*	112.2*b*	2776*a*	37.3*bc*	103.5*a*
CUF 101	3363*b*	32.1*a*	107.9*b*	2688*ab*	33.7*c*	90.6*a*
Excalibur	3752*ab*	30.8*a*	110.6*b*	2501*b*	39.1*ab*	97.8*a*
Algonquin	3418*b*	34.7*a*	118.0*b*	2121*c*	42.8*a*	90.9*a*
Saranac	4121*a*	33.9*a*	139.4*a*	2612*ab*	39.4*ab*	102.8*a*
CV (%)^{z}	7.5	6.1	7.4	5.3	6.8	9.4
Contrast^{y}	*	NS	**	**	**	NS

Glenlea, 1992

	Cut 1			Cut 2		
Cultivar	Yield kg ha^{-1}	N Conc. g kg^{-1}	N Yield kg N ha^{-1}	Yield kg ha^{-1}	N Conc. g kg^{-1}	N Yield kg N ha^{-1}
Nitro	3107*a*	34.3*a*	106.4*a*	1939*a*	38.6*b*	74.5*b*
CUF 101	3318*a*	32.2*a*	106.6*a*	1975*a*	37.5*b*	72.7*b*
Excalibur	3207*a*	31.2*a*	100.5*a*	2054*a*	44.8*a*	92.1*a*
Algonquin	3617*a*	33.9*a*	120.8*a*	1906*a*	46.9*a*	89.2*a*
Saranac	3261*a*	35.9*a*	117.3*a*	2188*a*	44.5*a*	95.9*a*
CV (%)^{z}	9.8	16.5	19.3	11.1	5.1	9.3
Contrast^{y}	NS	NS	NS	NS	**	**

^{z}Coefficient of variation (%).

^{y}Single degree of freedom orthogonal contrast; nondormant cultivars vs. dormant cultivars. NS, non-significant. *,** Significant at $P < 0.05$ and 0.01, respectively.

a-c Means within a column followed by the same letter are not significantly different at $P < 0.05$ by LSD.

Table 4. Yield, N concentration (conc.), and N yield of the herbage incorporated into the soil at Portage la Prairie and Glenlea in 1991.

	Portage la Prairie, 1991 Incorporated Herbage			Glenlea, 1991 Incorporated Herbage		
Cultivar	Yield kg ha^{-1}	N Conc. g kg^{-1}	N Yield kg N ha^{-1}	Yield kg ha^{-1}	N Conc. g kg^{-1}	N Yield kg N ha^{-1}
Nitro	2058*a*	33.6*a*	69.5*a*	1765*a*	23.6*a*	42.2*a*
Excalibur	1321*b*	27.5*b*	36.9*b*	1511*b*	23.4*a*	36.0*a*
Algonquin	1256*b*	25.8*b*	33.1*b*	1591*ab*	19.8*a*	31.6*a*
CV (%)[z]	11.8	16.4	45.8	15.6	22.8	31.5

[z] Coefficient of variation (%).

a-b Means within a column followed by the same letter are not significantly different at $P < 0.05$ by LSD.

Table 5. Yield, N concentration (conc.), and N yield of the herbage and upper roots[z] incorporated into the soil in the fall at Portage la Prairie and Glenlea in 1992.

Portage la Prairie, 1992

	Incorporated Herbage			Upper Roots		
Cultivar	Yield kg ha^{-1}	N Conc. g kg^{-1}	N Yield kg N ha^{-1}	Yield kg ha^{-1}	N Conc. g kg^{-1}	N Yield kg N ha^{-1}
Nitro	2582*a*	37.1*a*	95.5*a*	3424*a*	25.2*a*	89.6*ab*
CUF 101	2699*a*	37.4*a*	101.2*a*	3863*a*	25.0*a*	96.8*a*
Excalibur	1593*b*	21.9*c*	34.5*b*	3335*a*	17.5*b*	57.8*bc*
Algonquin	1278*b*	21.4*c*	26.3*b*	2826*a*	18.4*b*	52.9*c*
Saranac	1475*b*	26.1*b*	38.3*b*	3084*a*	18.5*b*	57.4*bc*
CV (%)[y]	15.7	8.5	15.2	23.8	15.7	32.0
Contrast[x]	**	**	**	NS	**	**

Glenlea, 1992

	Incorporated Herbage			Upper Roots		
Cultivar	Yield kg ha^{-1}	N Conc. g kg^{-1}	N Yield kg N ha^{-1}	Yield kg ha^{-1}	N Conc. g kg^{-1}	N Yield kg N ha^{-1}
Nitro	2545*a*	38.1*a*	96.4*a*	6418*a*	26.4*a*	177.3*a*
CUF 101	2629*a*	36.8*a*	97.0*a*	5753*a*	26.4*a*	151.3*ab*
Excalibur	1853*b*	28.6*b*	52.8*b*	4945*a*	18.3*b*	92.5*bc*
Algonquin	1504*b*	24.7*c*	37.3*c*	4055*a*	18.8*b*	76.5*c*
Saranac	1793*b*	28.0*b*	49.8*bc*	5665*a*	17.7*b*	100.0*bc*
CV (%)[y]	13.1	6.0	13.9	26.0	17.1	34.1
Contrast[x]	**	**	**	NS	**	**

[z] Sampled roots were trimmed to a length of 10 cm before analysis.

[y] Coefficient of variation (%).

[x] Single degree of freedom orthogonal contrast; nondormant cultivars vs. dormant cultivars. NS, non-significant. *,** Significant at $P < 0.05$ and 0.01, respectively.

a-c Means within a column followed by the same letter are not significantly different at $P < 0.05$ by LSD.

Table 6. Total herbage (hay and incorporated) dry matter and nitrogen yields for Portage la Prairie and Glenlea in 1991 and 1992.

	Portage la Prairie, 1991 Total Herbage		Glenlea, 1991 Total Herbage	
Cultivar	Yield kg ha^{-1}	N Yield kg N ha^{-1}	Yield kg ha^{-1}	N Yield kg N ha^{-1}
Nitro	9739*a*	286.1*a*	5819*a*	142.4*a*
Excalibur	8862*ab*	267.1*ab*	5930*a*	151.2*a*
Algonquin	8175*b*	241.6*b*	5925*a*	137.3*a*
CV (%)^z	16.7	16.6	8.9	12.3

	Portage la Prairie, 1992 Total Herbage		Glenlea, 1992 Total Herbage	
Cultivar	Yield kg ha^{-1}	N Yield kg N ha^{-1}	Yield kg ha^{-1}	N Yield kg N ha^{-1}
Nitro	8847*a*	311.2*a*	7590*a*	277.3*a*
CUF 101	8751*a*	299.7*ab*	7922*a*	272.4*a*
Excalibur	7846*b*	246.0*c*	7380*a*	257.7*a*
Algonquin	6817*c*	235.2*c*	7027*a*	247.3*a*
Saranac	8208*ab*	280.5*b*	7174*a*	262.2*a*
CV (%)^z	5.1	5.4	8.5	12.8
Contrast^y	**	**	NS	NS

^{z}Coefficient of variation (%).

^{y}Single degree of freedom orthogonal contrast; nondormant cultivars vs. dormant cultivars. NS, non-significant. *,** Significant at $P <$ 0.05 and 0.01, respectively.

a-c Means within a column followed by the same letter are not significantly different at $P < 0.05$ by LSD.

Table 7. Percent N derived from N_2 fixation (%Ndfa) and the quantity of N_2 fixed (QNF - kg N ha^{-1}), using barley as the reference crop, for the hay harvests and the incorporated herbage at Portage la Prairie and Glenlea in 1991.

Portage la Prairie, 1991

	Cut 1		Cut 2		Incorporated Herbage	
Cultivar	%Ndfa	QNF	%Ndfa	QNF	%Ndfa	QNF
Nitro	49.9*a*	52.8*a*	79.6*a*	87.3*a*	83.4*a*	58.0*a*
Excalibur	53.1*a*	61.6*a*	81.9*a*	93.3*a*	80.3*a*	29.9*b*
Algonquin	50.4*a*	47.0*a*	81.8*a*	89.9*a*	81.5*a*	27.1*b*
CV (%)[z]	22.8	23.3	10.5	15.7	5.9	13.1

Glenlea, 1991

	Cut 1		Incorporated Herbage	
Cultivar	%Ndfa	QNF	%Ndfa	QNF
Nitro	26.8*a*	27.1*a*	78.6*a*	33.3*a*
Excalibur	24.7*a*	27.3*a*	79.0*a*	28.1*ab*
Algonquin	22.2*a*	20.2*a*	76.0*a*	24.2*b*
CV (%)[z]	29.8	32.7	3.8	17.8

[z] Coefficient of variation (%).

a-b Means within a column followed by the same letter are not significantly different at $P < 0.05$ by LSD.

Table 8. Percent N derived from N_2 fixation (%Ndfa) and the quantity of N_2 fixed (QNF - kg N ha^{-1}) in the herbage as determined by the average of the isotope dilution technique (barley and Ineffective Saranac reference crops) and the difference method (Ineffective Saranac reference crop). N_2 fixation data is presented for the hay harvests at Portage la Prairie and Glenlea in 1992.

	Portage la Prairie, 1992				Glenlea, 1992			
	Cut 1		Cut 2		Cut 1		Cut 2	
Cultivar	%Ndfa	QNF	%Ndfa	QNF	%Ndfa	QNF	%Ndfa	QNF
Nitro	64.7*a*	72.3*a*	75.7*a*	78.6*a*	37.2*a*	39.0*a*	46.3*a*	34.4*a*
CUF 101	65.1*a*	70.5*a*	73.0*a*	65.8*a*	40.2*a*	43.0*a*	46.8*a*	34.9*a*
Excalibur	60.5*a*	66.5*a*	74.9*a*	73.4*a*	32.4*a*	33.4*a*	50.3*a*	47.0*a*
Algonquin	62.7*a*	73.8*a*	71.9*a*	65.7*a*	36.2*a*	44.5*a*	50.7*a*	45.6*a*
Saranac	64.7*a*	90.3*a*	72.7*a*	75.0*a*	35.0*a*	41.6*a*	50.2*a*	48.4*a*
CV (%)[z]	7.4	12.8	2.9	10.4	15.8	30.0	10.6	17.0
Contrast[y]	NS	NS	NS	NS	NS	NS	NS	*

[z] Coefficient of variation (%).

[y] Single degree of freedom orthogonal contrast; nondormant cultivars vs. dormant cultivars.

NS, non-significant. *,** Significant at $P < 0.05$ and 0.01, respectively.

a-c Means within a column followed by the same letter are not significantly different at $P < 0.05$ by LSD.

Table 9. Percent N derived from N_2 fixation (%Ndfa) and the quantity of N_2 fixed (QNF - kg N ha^{-1}) in the biomass as determined by the average of the isotope dilution technique (barley and Ineffective Saranac reference crops) and the difference method (Ineffective Saranac reference crop). N_2 fixation data is presented for incorporated herbage and upper roots[z] at Portage la Prairie and Glenlea in 1992.

	Portage la Prairie, 1992				Glenlea, 1992			
	Incorporated Herbage		Upper Roots		Incorporated Herbage		Upper Roots	
Cultivar	%Ndfa	QNF	%Ndfa	QNF	%Ndfa	QNF	%Ndfa	QNF
Nitro	78.2*a*	74.9*a*	71.2*a*	65.7*ab*	63.3*a*	60.9*a*	52.7*a*	102.2*a*
CUF 101	79.2*a*	80.0*a*	73.2*a*	72.2*a*	63.1*a*	61.7*a*	64.7*a*	98.5*a*
Excalibur	59.3*b*	21.3*bc*	64.1*a*	36.6*bc*	45.7*b*	24.5*b*	36.9*a*	32.2*b*
Algonquin	51.5*c*	14.3*c*	54.7*a*	28.1*c*	30.8*c*	11.8*c*	37.1*a*	27.9*b*
Saranac	62.4*b*	24.6*b*	57.5*a*	36.0*bc*	39.6*b*	19.9*bc*	41.8*a*	43.1*b*
CV (%)[y]	6.6	15.0	15.1	44.6	8.8	20.0	28.9	48.9
Contrast[x]	**	**	*	**	**	**	*	**

[z] Sampled roots were trimmed to a length of 10 cm before analysis.

[y] Coefficient of variation (%).

[x] Single degree of freedom orthogonal contrast; nondormant cultivars vs. dormant cultivars.

NS, non-significant. *,** Significant at $P < 0.05$ and 0.01, respectively.

a-c Means within a column followed by the same letter are not significantly different at $P < 0.05$ by LSD.

Table 10. Total N fixed in 1991 (hay and incorporated herbage) and 1992 (hay, incorporated herbage, and upper roots[z]) at Portage la Prairie and Glenlea.

Cultivar	Portage la Prairie, 1991 Total N Fixed (herbage) kg N ha^{-1}	Glenlea, 1991 Total N Fixed (herbage) kg N ha^{-1}
Nitro	195.1*a*	60.4*a*
Excalibur	184.7*a*	55.4*ab*
Algonquin	164.0*a*	44.4*b*
CV (%)[y]	14.0	18.0

Cultivar	Portage la Prairie, 1992 Total N Fixed (herbage and upper roots[z]) kg N ha^{-1}	Glenlea, 1992 Total N Fixed (herbage and upper roots[z]) kg N ha^{-1}
Nitro	291.5*a*	236.5*a*
CUF 101	288.5*a*	225.3*ab*
Excalibur	196.1*bc*	117.4*a*
Algonquin	181.9*c*	129.8*c*
Saranac	225.9*b*	157.4*bc*
CV (%)[y]	11.3	24.7
Contrast[x]	**	**

[z] Sampled roots were trimmed to a length of 10 cm before analysis.

[y] Coefficient of variation (%).

[x] Single degree of freedom orthogonal contrast; nondormant cultivars vs. dormant cultivars. NS, non-significant. *,** Significant at $P < 0.05$ and 0.01, respectively.

a-c Means within a column followed by the same letter are not significantly different at $P < 0.05$ by LSD.

Table 11. Plant survival in the spring of 1992 (Portage la Prairie) and 1993 (Portage la Prairie and Glenlea).

Cultivar	Fall Dormancy[z]	Portage, 1992 Plant Survival (%)	Portage, 1993 Plant Survival (%)	Glenlea, 1993 Plant Survival (%)
Algonquin	2	52.9a	70.1*a*	98.5*a*
Excalibur	4	57.2*a*	64.7*a*	87.1*b*
Saranac	4	_	60.1*a*	88.3*b*
Nitro	8	2.4*b*	7.4*b*	62.2*c*
CUF 101	9	_	5.6*b*	46.4*d*
CV (%)[y]		26.1	23.8	8.0
Contrast[x]		_	**	**

[z] Fall dormancy ratings by the Certified Alfalfa Seed Council (1992).
[y] Coefficient of variation (%).
[x] Single degree of freedom orthogonal contrast; nondormant cultivars vs dormant cultivars. *,** Significant at $P < 0.05$ and 0.01, respectively.
a-d Means within a column followed by the same letter are not significantly different at $P < 0.05$ by LSD.

Table 12. Grain yield, N concentration (conc.), grain N uptake, and total shoot N uptake (soft dough stage) in wheat preceded by alfalfa and barley at Portage la Prairie in 1992.

Portage la Prairie, 1992				
Cultivar	Grain Yield kg ha^{-1}	Grain N Conc. g kg^{-1}	Grain N Uptake kg N ha^{-1}	Total Shoot N Uptake kg N ha^{-1}
Nitro	3365*a*	22.2*a*	74.1*a*	111.7*a*
Excalibur	2762*a*	23.3*a*	63.6*bc*	115.5*a*
Algonquin	3150*a*	22.2*a*	69.9*ab*	123.8*a*
Barley	2815*a*	20.8*b*	58.6*c*	74.9*b*
CV (%)^{z}	13.3	4.4	12.3	18.7
Contrast^{y}	NS	**	*	**

^{z}Coefficient of variation (%).

^{y}Single degree of freedom orthogonal contrast; alfalfa cultivars vs barley.

*, ** Significant at $P < 0.05$ and 0.01, respectively.

a-c Means within a column followed by the same letter are not significantly different at $P < 0.05$ by LSD.

4. The Nitrogen Dynamics of Varying Periods of Alfalfa in a Cropping System

4.1. ABSTRACT

Alfalfa (*Medicago sativa* L.) has long been recognized as a source of nitrogen for the subsequent crops in a rotation. The relationship between symbiotically fixed and soil nitrogen sources, and the way they are partitioned into harvested hay or incorporated biomass, is not well understood. Field experiments were initiated to develop a basic nitrogen budget for one to three years of alfalfa in a crop rotation. The role of extracted deep leached nitrate was assessed and changes in total soil N were monitored. The effects on a subsequent wheat crop were also measured. N_2 fixation was measured using the ^{15}N isotope dilution technique. The total seasonal amount of N_2 fixed generally increased with each additional year of alfalfa, ranging from 174 kg N ha^{-1} for first year alfalfa to a high of 466 kg N ha^{-1} for third year alfalfa. The net soil N balance showed that an average of 84, 148 and 137 kg N ha^{-1} was added to the soil system for one, two and three year old alfalfa stands. Total soil N measurements indicated that as much as 2510 kg N ha^{-1} could be added to the soil after three years of alfalfa.

4.2. INTRODUCTION

The inclusion of alfalfa (*Medicago sativa* L.) in a cropping sequence has long been perceived as a means of improving the yield and quality of the subsequent crops in a rotation (Ellis, 1943; Hedlin et al., 1957). A number of factors are involved in increasing subsequent crop yields, some of which include reduced disease, weed and insect levels, as well as higher soil organic matter contents, better soil aggregation and improved water infiltration (Campbell et al., 1990; Meek et al., 1990; Raimbault and Vyn, 1991; Angers, 1992; Caporali and Onnis, 1992). A significant benefit of including alfalfa in a crop rotation, however, is it's contribution of nitrogen to subsequent crops. Nitrogen fertilizer replacement values of 125 to 135 kg N ha^{-1} for alfalfa followed by corn have been reported (Baldock and Musgrave, 1980; Bruulsema and Christie, 1987). The amount of alfalfa N typically incorporated with three to five year old alfalfa stands in the midwestern United States has been estimated at 140 to 196 kg N ha^{-1} (Hesterman et al., 1987).

Alfalfa's role as a nitrogen source is made possible by it's ability to symbiotically fix large amounts of atmospheric nitrogen. Published estimates of annual N_2 fixation by alfalfa range from 70 to 400 kg N ha^{-1} (Peterson and Russelle, 1991). Symbiotically fixed N_2 is made available to subsequent crops when the alfalfa is killed and the tissues decompose, resulting in mineralization of organic nitrogen to plant available mineral forms. In addition to N released from alfalfa residue, a certain amount of N is excreted directly to the soil during the life of the stand, estimated at 3% of the daily N fixed (Ta et al., 1986). The amount of biologically fixed N_2 returned to the soil will depend on the proportion of N derived from symbiosis and the amount of plant biomass added to the soil, which is in turn affected by the harvest management of the alfalfa (Bowren et al., 1969; Sheaffer et al., 1989; Sheaffer et al., 1991).

Alfalfa's contribution of nitrogen is usually reflected in the total N status of the soil. Average annual increases of 100 to 566 kg N ha^{-1} have been reported (Lyon and Bizzell, 1934; Holford, 1990). The effect of alfalfa, however, seems to depend on the initial nitrogen level of the soil. A soil with 0.120% total N observed an annual increase of 8 kg N ha^{-1} when alfalfa was grown 6 times in 10 years, compared with an increase of 68 kg N ha^{-1} on a soil with an initial total N content of 0.084% (Lyon and Bizzell, 1934). No increase was observed on a soil with an initial N content of 0.200% when alfalfa was grown for 4 years out of seven (Dubetz and Hill, 1964), while a decrease of 0.039% was observed on plots that had been in alfalfa for eight out of 12 previous years, when the initial soil N content was 0.245% (Ferguson and Gorby, 1971).

An often overlooked benefit of including alfalfa in a cropping sequence is it's ability to extract and utilize nitrate that has leached past the rooting zone of most annual crops. Alfalfa roots have the ability to absorb water and nutrients from a depth of 11 m (Peterson and Russelle, 1991), giving it access to large amounts of leached nitrogen that can be removed with the herbage. When alfalfa was cropped to an abandoned feedlot, nitrate-nitrogen was 113 kg ha^{-1} less than the control treatment in the year of establishment, and 1703 kg ha^{-1} less after the fifth year of alfalfa growth (Schuman and Elliot, 1978). Mathers et al. (1975) noted similar results when alfalfa was cropped to plots that had previously received varying amounts of manure. Nitrate nitrogen was removed to a depth of 1.8 and 3.6 m in the first and second years of alfalfa growth, respectively. Nitrate removal and water extraction patterns were very similar, indicating that nitrate was removed to the same depth that water was utilized. A concern of using alfalfa, however, is the potential for large amounts of mineralization and releaching of nitrogen once the alfalfa is killed, especially in areas of high rainfall or under irrigation (Robbins and Carter, 1980; Peterson and Russelle, 1991).

The dynamics of nitrogen flow in a cropping system containing alfalfa are not well understood. How much nitrogen is derived from symbiosis or from soil sources such as leached nitrate, and how this N is partitioned into hay removal or green manure sources for different years of alfalfa are questions lacking complete answers. Nitrogen budgets or balances, defined as "the application of mass conservation principles so that N is conserved in the various transformations and biological processes of a system" have been useful tools in studying and understanding the various flows of N in agricultural productions systems (Legg and Meisinger, 1982). A great deal of research has been done at the farm, regional, and global levels in developing N budgets and models for agricultural systems (Groot et al., 1991; Rosswall and Paustian, 1984; Stinner et al., 1984; Wetselaar et al., 1973) although relatively little research has been conducted in the field on alfalfa specifically. The objective of this study is, therefore, to develop a basic nitrogen budget for varying periods (one to three years) of alfalfa in a crop rotation and to define an optimum stand length in terms of the N related benefits. The major N sources of N_2 fixation and soil N (including leached nitrate) are considered, with particular attention paid to their partitioning into harvested hay or incorporated green manure. Changes in total soil N and the effects on a subsequent wheat crop are also measured.

4.3. MATERIALS AND METHODS

Investigations to establish N budgets were initiated in 1991 on a previously established alfalfa-based cropping systems study located at Winnipeg and Portage la Prairie, Manitoba. Climatic information for the studied years is presented in Tables 1 and 2 (derived from data supplied by the University of Manitoba and Environment Canada). The cropping systems study was established in 1990 on a Black Lake heavy clay soil at Winnipeg and a Dugas clay at Portage la Prairie.

Treatments for study included one to three years of O.A.C. Minto alfalfa (*Medicago sativa* L.) followed by one year of Katepwa wheat (*Triticum aestivum* L.) that received 0 or 80 kg ha^{-1} of nitrogen fertilizer. A summerfallow treatment provided a baseline for total soil N measurements, while an annual wheat-pea-barley rotation was used as the control for crop N uptake and nitrate extraction comparisons. The experiment had four replicates and was laid out as a split plot design with cropping sequence as the main plot and nitrogen fertility of the wheat crop as the subplot. Individual plot size was 5.5 m by 16 m at Winnipeg and 9 m by 12 m at Portage la Prairie. As budget measurements were not initiated until 1991, certain data was not available for the first year of alfalfa. In an attempt to estimate nitrogen fixation and crop growth for the first year stand, alfalfa was re-established in the spring of 1991 at Winnipeg.

Alfalfa was sown (no companion crop) on June 15 and May 23 of 1990 at rates of 10 and 8 kg ha^{-1} for Winnipeg and Portage la Prairie, respectively. Alfalfa established in 1991 was sown on May 18, at a rate of 12 kg ha^{-1}. The seed was previously treated with a clay based *Rhizobium meliloti* L. inoculum, but received an additional application of peat based inoculum (Nitragin Type A, LiphaTech Ltd.) at the time of seeding to ensure adequate *Rhizobium* populations. Alfalfa seed was placed approximately 0.5 cm deep in rows 15 cm apart.

The Winnipeg alfalfa stand was fertilized with 85 kg P_2O_5 ha^{-1} (11-52-0) in October of 1990, and 55 kg P_2O_5 ha^{-1} (12-55-0) in May of 1992. Alfalfa at Portage la Prairie received 45 kg P_20_5 ha^{-1} in May of 1991 (12-51-0) and 1992 (12-55-0). Imazethapyr (0.035 kg active ingredient [a.i.] ha^{-1}) was used at both sites for weed control in 1990, as well as sethoxydim (0.500 kg a.i. ha^{-1}) at Portage la Prairie. Handweeding was employed for weed control in alfalfa sown in 1991.

Seedling alfalfa in 1990 and 1991 was harvested for hay at the end of July (10% bloom) and incorporated into the soil in mid to late August when there was about 30 cm of regrowth. Two hay harvests were taken from established alfalfa at 10% bloom, the first in the second or third week of June and the second in the last week of July or the first week of August. The two and three year old alfalfa stands were worked down near the end of August when there was approximately 30 cm of regrowth. Alfalfa plots that were not worked down usually received an additional hay cut. Second (1990, Portage la Prairie only) and third (1991, 1992) harvests were taken in early to mid October.

Katepwa wheat was sown in mid May of 1991 and 1992. Wheat was sown at a rate of 135 kg ha^{-1} to a depth of 2.5 cm in rows 15 cm apart. Phosphate fertilizer was applied with the seed at a rate of 30 kg P_2O_5 ha^{-1} (12-51-0). Subplots fertilized with nitrogen received 80 kg N ha^{-1} as 34-0-0, broadcast shortly after seeding (except at Winnipeg in 1991, where it was broadcast and incorporated before seeding). A combination of fenoxaprop-p-ethyl (0.092 kg a.i. ha^{-1}), MCPA ester (0.420 kg a.i. ha^{-1}) and thifensulfuron methyl (0.015 kg a.i. ha^{-1}) was used for weed control in 1991. A mixture of dicamba (0.122 kg a.i. ha^{-1}), 2,4-D amine (0.327 kg a.i. ha^{-1}) and mecoprop (0.089 kg a.i. ha^{-1}) was used in 1992, as well as tralkoxydim (0.247 kg a.i. ha^{-1}) at Portage la Prairie. The wheat was harvested in the third week of August in 1991 and the first week of September in 1992.

Nitrogen fixation was measured using the ^{15}N isotope dilution technique (McAuliffe et al., 1958). The proportion of nitrogen derived from the atmosphere (%Ndfa) was calculated for the ^{15}N isotope dilution technique using the following formula:

$$100 - \left(\frac{\%^{15}\text{N Excess in the Fixing Crop}}{\%^{15}\text{N Excess in the Reference Crop}} \times 100 \right)$$

The percentage ^{15}N Excess was determined by subtracting the natural abundance of ^{15}N (0.3663%) from the %^{15}N abundance of the analyzed samples.

The quantity of nitrogen fixed was determined by multiplying the total amount of nitrogen in the sampled crop (kg N ha^{-1}) by %Ndfa/100.

^{15}N labelled $(NH_4)_2SO_4$ (60-85% enrichment) was applied in 1 L of water with a pressurized garden sprayer to 0.60 m by 1.5 m alfalfa subplots at a rate of 2.8 kg N ha^{-1} at both sites in 1991. In 1992, 15.9% enriched $(NH_4)_2SO_4$ fertilizer was applied to 0.75 m by 1.4 m alfalfa subplots at a rate of 9.5 kg N ha^{-1}, respectively, in 0.75 L of water with a pressurized garden sprayer. Labelled fertilizer was applied to the soil in between the rows of the alfalfa, such that application was made directly to the soil and the herbage was avoided. Four subplot interrows were treated in 1991 while five were treated in 1992, with a corresponding harvest of three and four rows of alfalfa, respectively, such that each harvested row had labelled fertilizer applied on either side of it. Plant residue was removed from the interrows prior to application to allow the fertilizer to contact bare soil. The fertilizer was incorporated by lightly raking the soil immediately following application. Wheat served as the ^{15}N reference crop in both years, using the same rates and methods of labelled fertilizer addition that were used for alfalfa, in each respective year. ^{15}N application was made to established alfalfa and wheat in mid May and the end of May, respectively. There was approximately 25 cm of new growth on the alfalfa at the time of application, while the wheat ranged from the one to three leaf stage. Seedling alfalfa established in 1991 received ^{15}N labelled fertilizer on June 7, when the majority of the plants were still in the cotyledon stage. The alfalfa subplots were removed immediately prior to hay harvest or soil incorporation, while the wheat was harvested at the soft dough stage (whole shoot, 10 cm cutting height) in the last week of July in 1991, or the second week of August in 1992.

Nitrogen uptake in the wheat following alfalfa was assessed in whole shoot samples taken at the soft dough stage, and also in grain harvested at maturity. Wheat following barley from the annual wheat-peas-barley rotation was sampled for comparison. A 0.75 m^2 area (5 rows, 1 m length) was harvested at soft dough, while a grain subsample was retained for N analysis from plots harvested at maturity.

Soil samples were taken in 30 cm increments to a depth of 240 cm. Sampling took place in early September of 1990, the first week of May in 1992 (representative of the 1991 growing season) and in mid October of 1992 (representative of the 1992 growing season). Summerfallow treatments sampled in 1990 provided a baseline for total soil N, while the annual wheat-peas-barley (nitrogen fertilized) rotation was used as the control for the nitrate extraction comparison.

Alfalfa roots were not harvested in this study, and root yields were therefore determined by deriving shoot:root ratios from the literature. The data of Pettersson et al. (1986) was chosen due to the similar harvest management employed in that study (two summer hay harvests with the subsequent regrowth measured in the fall for two and three year old alfalfa stands). Ratios were derived by combining the first and second harvest yields with the amount of herbage remaining at the end of the year, and relating this figure to the end of season root yield (0.5 m depth). The ratio was averaged for two and three year old alfalfa stands, giving a shoot:root ratio of 1.59.

Root yields for the nitrogen budget were estimated by applying this ratio to the total herbage yield in each year (hay cuts plus incorporated herbage). Root N yields were estimated by using the end of season root N concentration averaged for second and third year stands (25.7 g kg^{-1}) measured in the experiment by Pettersson et al. (1986). The proportion of N derived from N_2 fixation (%Ndfa) for roots was estimated by averaging the %Ndfa measured from the herbage sampled at the hay cut(s), and the material incorporated into the soil.

All herbage and soil samples were dried in an oven at 70 and 50°C, respectively, for a minimum of 72 hours, with the exception of soil samples from control plots for nitrate comparison which were air dried. Grain samples were tested for moisture and converted to a dry weight basis prior to N uptake calculations. Plant samples for ^{15}N:^{14}N analysis were ground once in a Wiley mill to pass a 2 mm screen, followed by one more pass (three passes in 1992) through a Udy cyclone mill with a 0.5 mm screen. Grain samples were ground once in a Udy cyclone mill to pass a 1.0 mm screen. Soil samples were broken up in a Christi hammer mill equipped with a 2.0 mm screen.

All soil samples were analyzed for N content on a Leco N Determinator (Model FP 428, Leco Corp., Mississauga, Ontario), except for those taken in 1990, which were tested for total N by the kjeldahl procedure. Grain samples and whole shoot wheat samples were analyzed for N concentration by kjeldahl in 1990 and

1991, and by the Leco N determinator in 1992. Prior to analysis, the samples were redried at 60°C for a minimum of 12 hours. N concentration of the material analyzed for $^{15}N:^{14}N$ ratios was determined using the micro-kjeldahl procedure (Bremner and Mulvaney, 1982) in 1991 and by a nitrogen determinator (see below) in 1992. After determination of total nitrogen content by the micro-kjeldahl procedure in 1991, the titrated solution was evaporated to a small volume (less than 5 mL) and subsequently analyzed for $^{15}N:^{14}N$ ratios, performed at the laboratory of Dr. A. Blackmer, Iowa State University, on a Finnigan Mat 250 mass spectrometer. $^{15}N:^{14}N$ ratios were determined in 1992 on an ANCA-MS nitrogen determinator/mass spectrometer (Europa Scientific, Crewe, U.K.), equipped with a single inlet and triple collectors, in the laboratory of Dr. C. van Kessel, University of Saskatchewan.

Soil nitrate of the second and third year alfalfa plots was measured on an ion chromatograph, as described by Dick and Tabatabai (1979). Soil samples were redried at 50°C for 48 hours prior to analysis, and a 10 g sample was extracted in 20 mL of 10 m*M* KCl by shaking for 30 minutes. The suspension was filtered (Whatman #100 filter paper) and analyzed on a Dionex Model 4000 ion chromatograph (Mississauga, Ont.). Soil nitrate of the first year alfalfa plots and the control plots (annual wheat-peas-barley rotation) from 1990 and 1991 was analyzed by the methods described in McKeague (1978). The soil was extracted using 50 mL of extractant ($0.5M$ Na_2CO_3) and 1 g of soil, with shaking for 30 minutes. The suspension was filtered using Whatman #40 filter paper, and the extract analyzed by hydrazine sulfate reduction on a Technicon autosampler (Model AA I). Control samples from 1992 were extracted using a 25 g size sample in 50 mL of extractant (10 m*M* KCl) with 30 minutes of shaking. After filtering the extract (Whatman #40 paper), the samples were analyzed by cadmium reduction on a Model AA II Technicon autosampler.

Nitrate nitrogen was converted from ppm to kg N ha^{-1} for each 30 cm increment below 0.9 m. Bulk density values required for the conversion were derived from measurements performed by the Manitoba Land Resource Unit Center for Land and Biological Resources Research (W. Michalyna, pers. comm.). Bulk density values for 0.9 to 1.2 m and 1.2 to 1.5 m were used; the latter value assumed for the lower depth increments. For a Neuhorst soil (similar to the Dugas clay at Portage la Prairie) bulk densities of 1.37 and 1.45 Mg m^{-3} were used for the 0.9 to 1.2 m and 1.2 to 1.5 m depths, respectively. The bulk density was 1.37 Mg m^{-3} between 0.9 m and 1.5 m for the Black Lake soil. The amount of nitrate extracted was calculated by summing the total amount of nitrate in the 0.9 to 2.4 m zone for each year of alfalfa and subtracting that from the nitrate level of the corresponding annual control plot. This is meant to represent the removal of nitrate that could no longer be accessed by the roots of annual crops. The amount of nitrate extracted in different years was determined by subtracting individual stand years from one another (ie. nitrate extracted in the third year of alfalfa = year three nitrate uptake - year two nitrate uptake).

4.4. RESULTS AND DISCUSSION

4.4.1. N Partitioning

The partitioning of biologically fixed and soil nitrogen into herbage removed for hay, or incorporated herbage and roots, for the varying stand lengths is illustrated for the two sites in Figures 1 and 2 (first year stand data available for Winnipeg site only). The total amount of N (fixed plus soil) in the herbage (removed with hay plus incorporated) was similar between years of established alfalfa, with 407 and 404 kg N ha^{-1} present in the herbage of alfalfa at Winnipeg, and 391 and 392 kg N ha^{-1} in the herbage at Portage la Prairie, for second and third year alfalfa, respectively. The total N herbage yield of first year alfalfa at Winnipeg was 167 kg N ha^{-1} (Figure 1), a substantially smaller amount when compared with the older stands. Wivstad et al. (1987) and Heichel et al. (1984) noted substantial increases in the herbage and nitrogen yields of established alfalfa relative to the seeding year. Heichel et al. (1984) observed that herbage yields decreased by 12% in the third year of production relative to the second, while Wivstad et al. (1987) noted a 12% increase in total herbage yield and a 15% increase in total nitrogen yield from the second to the third year of alfalfa growth.

In general, more nitrogen (fixed plus soil) was removed with the herbage at the first cut relative to the second, which corresponds to normal alfalfa yield patterns in Manitoba, as first cut hay generally yields better than hay taken at the second cut. The amount of N incorporated into the soil with the herbage was less than that removed for individual hay cuts, due to the relatively immature state of the incorporated alfalfa (30 cm regrowth, prior to bud).

Regarding biological N_2 fixation, there was a trend for increasing levels of activity as the season progressed, measured as the percentage of nitrogen derived from the atmosphere (%Ndfa) in the herbage. %Ndfa for the first and second hay cuts and the herbage incorporated into the soil was 47.3, 82.2, and 89.3%, respectively, when averaged for the second and third years of alfalfa over both sites. Wivstad et al. (1987) reported %Ndfa levels of 67, 82 and 76% for measurements taken at the first and second harvests and at the season's end, respectively, when averaged for the second and third years of alfalfa. Increased mineral N availability caused by the turnover of organic N from dead, overwintered plant material could cause a reduction in N_2 fixation in the spring. As mineral N availability decreases, the alfalfa might respond by increasing N_2 fixation later in the season.

The mean level of N_2 fixed (averaged for hay and incorporated materials) for the two and three year old alfalfa stands was 70.2 and 75.6%, respectively, when averaged over both sites. The average %Ndfa in the herbage of first year alfalfa at Winnipeg was 63.7%. In general, the percent N derived from symbiosis increases with increasing stand age, particularly after the establishment year (Peterson and Russelle, 1991). Increases in %Ndfa from 59 to 78% from the first to the fourth year of alfalfa (Heichel et al., 1984) and 70 to 80% from the second to third year of alfalfa growth (Wivstad et al., 1987) have been observed.

The contribution of nitrate extracted from depth to the N budgets is presented for the Winnipeg site only, as there was little nitrate accumulated at depth at Portage la Prairie and therefore no discernable extraction (Figure 1). Nitrate extraction increased with each additional year of alfalfa growth (by 56% over the three year period), possibly due to greater rooting at depth and improved access to deep leached nitrate. Nitrate and water extraction patterns were similar over the three year period (Appendix Figures D1 to D6), suggesting that nitrate was removed from the same depth that water was utilized (Mathers et al., 1975). Despite the increase in extracted nitrate over time, there was still an increasing trend in N_2 fixation with each year of alfalfa growth. The amount of symbiotically fixed nitrogen in the herbage increased by 31 kg N ha^{-1} from the second to the third year, while the total amount of N in the herbage remained the same between years. A decrease in N_2 fixation, or an increase in the total amount of herbage N produced, would have been expected with increased extraction of mineral N. A corresponding decrease in the amount of soil N used above 0.9 m, or possibly a shift in the shoot:root N ratios, between years of alfalfa might explain such an observation, although neither can be verified here.

The estimated amount of nitrogen (fixed plus soil) returned to the soil with the crowns and roots was substantial, ranging from 207 to 259 kg N ha^{-1} for established alfalfa (Figures 1 and 2). The crown and root total nitrogen addition of first year alfalfa was notably smaller (107 kg N ha^{-1}) than that added by the two or three year old alfalfa stands. Most of the previous research on crown and root N contributions of alfalfa has been conducted on seeding year stands. N additions of

37 to 99 kg N ha^{-1} have been reported (Bruulsema and Christie, 1987; Griffin and Hesterman, 1991; Hesterman et al., 1986; Kroontje and Kehr, 1956). All of these studies examined the top 30 cm of soil or less. The values reported in the present experiment are exceed this range, possibly due to the differences in rooting depth (50 cm depth was used for estimation of root biomass). Little research has been done on older alfalfa stands, but it seems reasonable that crown and root N contributions of established alfalfa would be substantially greater than that added by seedling stands. The estimated N addition by the crowns and roots for second and third year alfalfa was an average 2.3 times the N added to the soil with the incorporated herbage, stressing the importance of below ground biomass in alfalfa soil N contributions.

4.4.2. Soil N Balance

The estimated amount of biologically fixed nitrogen added to the soil system with the incorporated herbage and roots was substantial. For first, second and third year alfalfa, an average 131, 272, and 241 kg ha^{-1} of fixed N_2 was added to the soil. However, the overall effect of the cropping system must consider the removal of existing soil N, in addition to that added by biological N_2 fixation. Basic nitrogen budgets are presented for different years of alfalfa in Tables 3 and 4. The major crop N inputs of biological N_2 fixation and soil N sources are estimated, and this N is accounted for in the major crop outputs of harvested hay, roots, and incorporated herbage. Using these figures, it is possible to calculate a soil N balance, a measurement of the effect of the cropping system on the soil N status.

Although large amounts of fixed nitrogen were added to the soil by alfalfa, significant quantities were also removed with the harvested forage. For the one, two and three year old stands, an average of 47, 125 and 105 kg ha^{-1} of existing soil N was removed with the herbage. When a net N balance is calculated, however, there was still a substantial positive effect exerted on the soil N status. The net N balance was 84 kg N ha^{-1} for the seedling stand and 148 and 137 kg N ha^{-1} for second and third year alfalfa, respectively, when averaged over both sites.

Two major factors influence the amount of N that alfalfa adds to the soil system: harvest management and the proportion of plant N derived from biological N_2 fixation (Sheaffer et al., 1989). A harvest system that returns more herbage to the soil (reduced number of hay cuts, or cutting and leaving the residue on the field) will increase the amount of N added to the soil. As well, the more N that is derived from N_2 fixation (as opposed to the soil), the greater will be the soil N contribution. Low fixation levels (less than 50%), caused by high mineral N availability or adverse environmental conditions, will result in a mining of soil N by alfalfa, and consequently a negative net N balance.

4.4.3. N Uptake in a Subsequent Wheat Crop

The ability of alfalfa to supply N to subsequent crops was examined by monitoring the N uptake of wheat following alfalfa and comparing this with the N uptake of wheat in an annual cropping system. Table 5 shows the N uptake of wheat in 1991, when preceded by one year of alfalfa or one year of barley. The barley - wheat rotation was split into fertilized (80 kg N ha^{-1}) or unfertilized subplots, while only the wheat phase of the alfalfa - wheat rotation received a fertilization sub treatment. At the Winnipeg site, N uptake in unfertilized wheat (whole shoot and grain) following alfalfa was intermediate to N uptake in unfertilized wheat preceded by barley and the average of the fertilized treatments, but was not significantly different from any treatment. Although the N yield was not increased significantly by alfalfa relative to the barley control treatment, it was also not significantly reduced relative to the treatments receiving fertilizer. At the Portage la Prairie site N uptake was greatest in unfertilized wheat preceded by alfalfa, and was significantly greater than unfertilized wheat following barley.

Table 6 shows the nitrogen uptake of wheat in 1992, when preceded by two years of alfalfa or by a peas - barley sequence. The results at Winnipeg were similar to those of 1991, with unfertilized wheat preceded by alfalfa taking up amounts of nitrogen intermediate to that extracted by the fertilized treatments and the unfertilized annual sequence. It was not, however, significantly different from either extreme. The Portage la Prairie site also produced similar results to those observed in 1991, with the two years of alfalfa supplying amounts of N similar to those treatments receiving N fertilizer, and providing significantly more N than the unfertilized annual cropping sequence.

The N uptake of unfertilized wheat preceded by alfalfa was 45% greater at Portage la Prairie and 25% greater at Winnipeg relative to unfertilized wheat in the annual sequences, when averaged over both years for whole shoot and grain. When the N uptake of unfertilized wheat following alfalfa is similarly compared with an average of the fertilized treatments (wheat preceded by alfalfa or annual crops), there was a 9% increase at Portage la Prairie and a 17% decrease at Winnipeg.

The literature generally reports yields and N uptakes of crops following alfalfa greater than unfertilized controls, but similar to control plots receiving N fertilizer (Kroontje and Kehr, 1956; Hesterman et al., 1986; Bruulsema and Christie, 1987; Badaruddin and Meyer; 1990), which follows the general trend observed in this study. Results between years at either site were similar, suggesting that both one and two years of alfalfa had a similar effect on the subsequent wheat crop. Other researchers have reported little effect of stand age on the yield of the subsequent crop (Hoyt and Hennig, 1971; Sheaffer et al., 1991). The present study, however, only examined the response of the first crop after alfalfa. The residual effects of alfalfa on several subsequent crops has shown that older (3.5 year) stands can be better than younger (1.5 year) stands when considering long term N supply (Holford, 1980).

4.4.4. Total Soil N

The effect of alfalfa stand length on the total N status of the soil is presented in Table 7. The change in total soil nitrogen, as determined by comparison with a non-crop treatment sampled at the beginning of the study, is given for the top 30 cm of soil. In general, alfalfa had an extremely large positive effect on the total N status of the soil. The first year of alfalfa at Winnipeg was the only treatment to exert a negative effect on soil total N. The size of the effect seemed to increase with stand age, with a 2% decrease, 15% increase and a 23% increase in total soil

N after one, two, and three years of alfalfa, respectively. The trend was similar at Portage la Prairie, with increases in soil N of 13%, 19%, and 20% for the first, second, and third year stands, respectively.

The observed increases in total soil N under alfalfa were impossibly large, with additions as high as 2510 kg N ha^{-1}. An increase of 2000 kg N ha^{-1} would require the incorporation of 80,000 kg ha^{-1} of biomass, assuming 2.5% N in the tissue. The maximum estimated amount of alfalfa biomass added to the soil in this study was 13,682 kg ha^{-1}. Changes in soil N of this magnitude have not been observed elsewhere. It is important to note, however, that the analysis of variance was not significant at either site. One year of alfalfa, which depleted the total soil N by 125 kg ha^{-1}, was not significantly different from three years of alfalfa, which added 2510 kg N ha^{-1} to the soil at Winnipeg.

Lyon and Bizzell (1934) measured increases in soil N (top 15 cm of soil) of 450 kg ha^{-1} after 10 years of alfalfa, while Holford (1990) measured a similar increase (400 kg N ha^{-1}) after only 4 years. Holford (1981) reported increases in total soil N of 0.008%, 0.011%, and 0.027%, corresponding to 156, 332, and 527 kg N ha^{-1} (based on a bulk density of 1.3 Mg m^{-3}) in the top 15 cm of a Black earth soil for 1.5, 2.5 and 3.5 years of alfalfa, respectively. On a Red Brown earth soil, there was a decrease of 0.009% (176 kg N ha^{-1}) in total soil N after the first 1.5 years of alfalfa, followed by subsequent increases of 0.01% and 0.016% (195 and 312 kg N ha^{-1}) after 2.5 and 3.5 years of alfalfa. The general trend was an increase in total soil N with each additional year of alfalfa, similar to the results observed in this experiment. The initial decrease in soil N after the first 1.5 years of alfalfa on the Red Brown earth also resembles the results observed after the first year of alfalfa at the Winnipeg site.

4.4.5. Potential Sources of Error in the N Budget

The budget makes direct comparisons between different stand lengths of alfalfa based on measurements taken in different years. The environmental conditions in a given year will have a profound influence on the growth, nitrogen fixation, N partitioning and, ultimately, the N balance of an alfalfa stand in that year. Determining if measured effects are a function of alfalfa stand age or environmental parameters is difficult, particularly if conditions vary greatly from year to year. Temperature and precipitation were quite similar, however, over the studied years of this experiment and were generally close to, or above, the thirty year average for each site (Tables 1 and 2).

The estimated soil N balance showed that alfalfa had a substantial positive effect on the N status of the soil. This agrees with the observation that N uptake in the subsequent wheat crop was similar to treatments receiving 80 kg N ha^{-1} of fertilizer, but does not explain the large apparent increases in measured total soil N (as high as 2510 kg ha^{-1}). Higher estimated levels of N_2 fixation might account for part of the large observed increases in total soil N. If fixation levels of 100% were observed, the positive effect on the soil N balance could have been as large as 367 kg N ha^{-1}. The use of wheat as the ^{15}N control crop in estimating N_2 fixation might be questioned, due to differences in maturity and rooting depth. ^{15}N levels were determined in the alfalfa at three times during the season, corresponding to the harvest dates, while it was only measured once in the wheat (at the soft dough stage). A dilution of available ^{15}N likely occurs over time, either through plant use or soil immobilization. The second alfalfa hay cut and the soft dough stage of the wheat took place at about the same time, hence N_2 fixation estimates should be reasonably accurate for the second hay harvest. N_2 fixation at the first hay cut, however, could have been underestimated while the estimates at the time of soil incorporation may have been overestimated. All three dates are used in N balance calculations, and despite discrepancies at individual dates, the averaged N_2 fixation levels should have been reasonably well estimated. N_2 fixation in the roots was based on an average of the levels measured in the herbage over the season, and should have also been fairly well estimated. Another factor that might be considered is the greater depth of rooting in the alfalfa relative to the wheat control crop. It was shown that alfalfa had the ability to extract nitrate from depths that the annual crops could not reach. This would tend to dilute the level of ^{15}N taken up by the alfalfa, inflating the estimate of N_2 fixation. Taking this into consideration, the estimates of N_2 fixed presented in this paper would be, if anything, overestimates of the true values.

As discussed earlier, the estimates of total N added to the soil by alfalfa were excessively large. Part of this may lie in the sensitivity of detecting changes in soil N. Standard deviations were generally as large or larger than the means themselves (Table 7) and any effect caused by treatments was likely obscured by the variation associated with sampling and analyzing soil for total nitrogen.

Root biomass was estimated in this study using a shoot:root ratio derived from the literature. If the chosen value was higher than the actual shoot:root ratio, an underestimation of the root biomass would have resulted, consequently reducing the estimated contribution of fixed N_2 to the soil by the roots. However, even a doubling of root biomass would not have accounted for the apparent changes in total soil N under alfalfa.

At the very least, it is reasonable to say that the increases in total soil N under alfalfa were large and were not accounted for in the soil N balance estimates. One possible explanation that might account for part of this discrepancy is the occurrence of root tissue turnover during the growing season (Russelle, 1992). The root production estimates in this study considered live roots only; if significant root death and regrowth were occurring, the impact on the soil N balance estimates could have been significant. Direct excretion of nitrogen by alfalfa roots to the soil could have a similar effect, although the size of the effect would likely be small, as direct excretion has been estimated at only 3% of the daily N fixed (Ta et al., 1986).

4.5. SUMMARY AND CONCLUSIONS

Second and third year alfalfa were similar to each other but greater than first year alfalfa in terms of forage N yields, incorporated N with the biomass, amounts of N_2 fixed, and effects on the net soil N balance. Although the effect of alfalfa on the estimated N balance was substantial, it was not reflected in the total soil N measurements, which illustrated large, progressive increases in total soil N with each additional year of alfalfa. Large apparent differences in total soil nitrogen among treatments were not significantly different, however, suggesting that the variation associated with testing for total soil N was sufficiently high to obscure true treatment differences. N uptake in the subsequent wheat crop was similar between one and two years of alfalfa, although the effects of residual N on the second and subsequent wheat crops were not determined.

Based on the N yields of hay, incorporated biomass, N_2 fixation, and the apparent changes in total soil N, it would seem that at least two years of alfalfa is necessary to optimize N related benefits. Although third year alfalfa fixed slightly more nitrogen and seemed to add more nitrogen to the soil than the second year stand, a discernible advantage of three years of alfalfa was not obvious in this study.

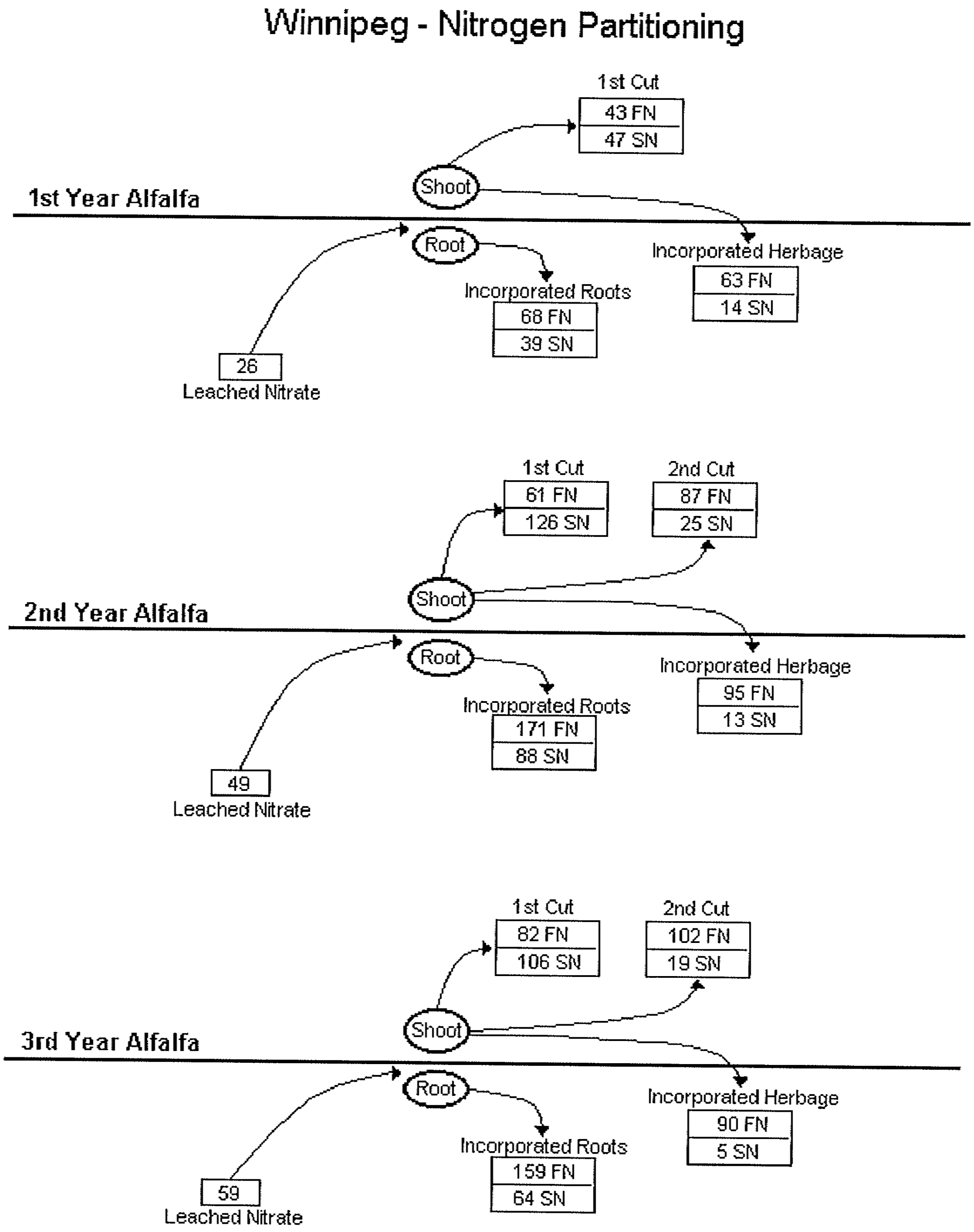
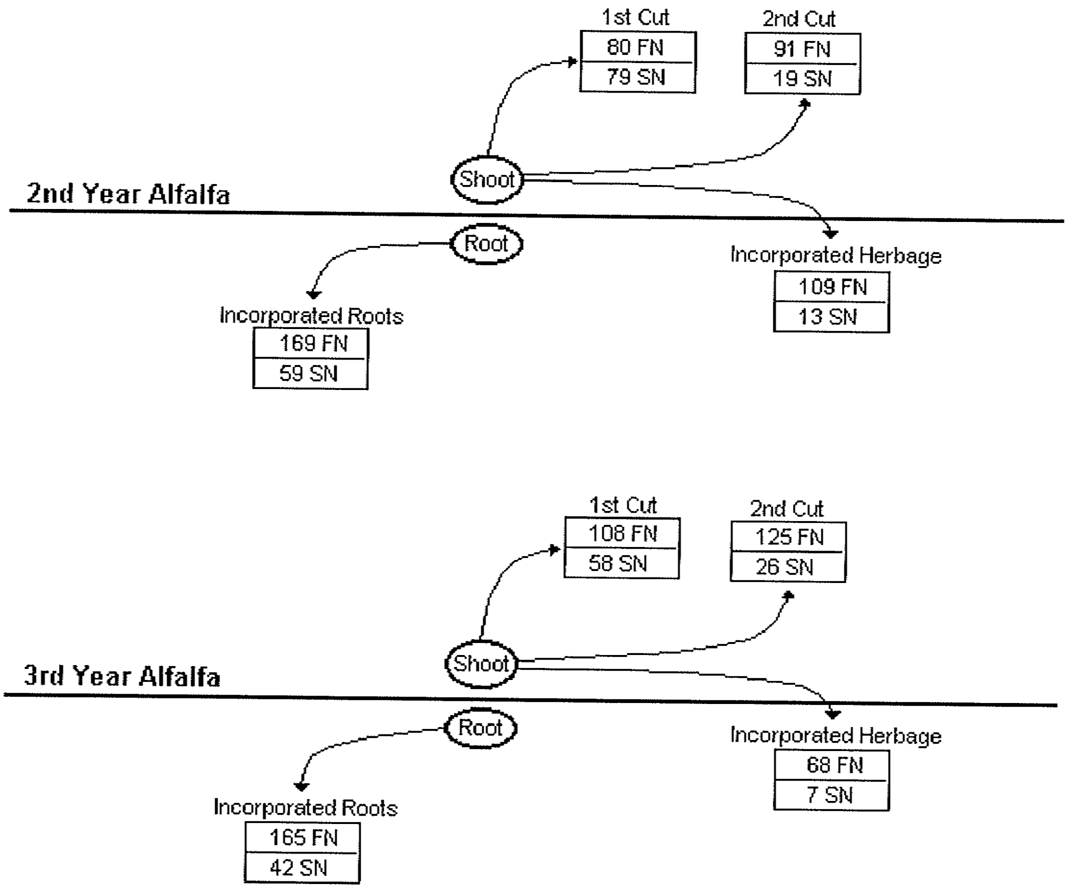


Figure 1. Partitioning of nitrogen (kg N ha^{-1}) into hay or incorporated roots and herbage for one, two and three years of alfalfa at Winnipeg. FN - nitrogen derived from symbiotic N_2 fixation. SN - nitrogen derived from soil sources. Leached nitrate - nitrate extracted from the 0.9 to 2.4 m soil depth zone.



Portage la Prairie - Nitrogen Partitioning

Figure 2. Partitioning of nitrogen (kg N ha^{-1}) into hay or incorporated roots and herbage for two and three years of alfalfa at Portage la Prairie. FN - nitrogen derived from symbiotic N_2 fixation. SN - nitrogen derived from soil sources.

Table 1. Precipitation and mean temperature (long term averages, 1990, 1991, and 1992)^{z} for the growing season months at Winnipeg, Manitoba.

Temperature and Precipitation	Month	Winnipeg 1961 to 1990	Winnipeg 1990	Winnipeg 1991	Winnipeg 1992
Mean Temp. (°C)	May	11.6	11.9	15.5	14.1
	June	16.9	18.5	20.0	16.3
	July	19.8	20.7	20.8	17.5
	August	18.3	20.9	21.7	17.7
	September	12.4	14.5	12.9	11.9
	October	5.7	5.6	2.6	5.4
Precip. (mm)	May	59.8	26.4	54.4	14.2
	June	83.8	167.4	126.5	117.9
	July	72.0	38.1	112.3	112.8
	August	75.3	15.2	13.0	83.8
	September	51.3	34.5	146.3	88.9
	October	29.5	10.9	54.6	3.3

^{z}Climatic information derived from University of Manitoba and Environment Canada data.

Table 2. Precipitation and mean temperature (long term averages, 1990, 1991, and 1992)^{z} for the growing season months at Portage la Prairie, Manitoba.

Temperature and Precipitation	Month	Portage 1961 to 1990	Portage 1990	Portage 1991	Portage 1992
Mean Temp. (°C)	May	11.6	10.9	14.1	12.8
	June	17.1	18.0	18.8	15.4
	July	19.8	19.2	19.5	16.3
	August	18.4	19.6	20.3	16.3
	September	12.5	13.6	12.2	10.8
	October	6.1	5.2	2.2	5.5
Precip. (mm)	May	56.8	34.0	59.0	12.6
	June	75.0	145.0	75.0	44.0
	July	76.9	57.0	95.0	109.0
	August	78.8	42.6	10.0	49.0
	September	50.1	21.6	68.0	50.0
	October	32.7	8.5	43.1	6.2

^{z}Climatic information derived from University of Manitoba and Environment Canada data.

Table 3. Nitrogen balance (kg N ha^{-1}) of an alfalfa crop for one, two, and three years of growth at Winnipeg.

		1st Year Alfalfa	2nd Year Alfalfa	3rd Year Alfalfa
N Input	Biological N_2 fixation	174	414	433
	Soil N (including leached nitrate)	100 (26[z])	252 (49)	194 (59)
N Output	Harvested hay	90	299	309
	Incorporated herbage and roots	184	367	318
Soil N Balance	Biologically fixed N_2 added to the soil via incorporated herbage and roots	131	266	249
	Soil N removed with harvested hay	47	151	125
	Net soil N balance	84	115	124

[z] Represents contribution from extraction of deep-leached nitrate, which is included in the soil N estimate.

Table 4. Nitrogen balance (kg N ha^{-1}) of an alfalfa crop for two and three years of growth at Portage la Prairie.

		2nd Year Alfalfa	3rd Year Alfalfa
N Input	Biological N_2 fixation	449	466
	Soil N	170	133
N Output	Harvested hay	269	317
	Incorporated herbage and roots	350	282
Soil N Balance	Biologically fixed N_2 added to the soil via incorporated herbage and roots	278	233
	Soil N removed with harvested hay	98	84
	Net soil N balance	180	149

Table 5. N Uptake in the wheat whole shoot (soft dough stage) and the grain (crop maturity) at Winnipeg and Portage la Prairie in 1991.

		Winnipeg - 1991		Portage la Prairie - 1991	
Crop Sequence	Fertility kg N ha^{-1}	Shoot N Uptake kg N ha^{-1}	Grain N Uptake kg N ha^{-1}	Shoot N Uptake kg N ha^{-1}	Grain N Uptake kg N ha^{-1}
Alfalfa - Wheat[z]	0	96*ab*	64.7*ab*	135.8*a*	72.5*a*
Alfalfa - Wheat	80	129.2*a*	72.6*a*	126.3*ab*	69.1*a*
Barley - Wheat	0	70.5*b*	52.1*b*	71.3*c*	49.2*b*
Barley - Wheat	80	140.4*a*	72.5*a*	102.1*b*	65.8*a*
CV (%)[y]		27.1	15.4	22.7	12.6
Crop Sequence		NS[x]	NS	*	**
Fertility		*	*	NS	NS
Crop*Fertility		NS	NS	NS	*

[z] Wheat sampled in 1991 was preceded by 1 year of alfalfa. The alfalfa - wheat sequence received N fertilizer in the wheat phase only. The annual (barley - wheat) sequence received (or did not receive) N fertilizer in both years of the rotation.

[y] Coefficient of variation (%).

[x] Analysis of variance. NS, non-significant. *,** Significant at $P < 0.05$ and 0.01, respectively.

a-c Means within a column followed by the same letter are not significantly different at $P < 0.05$ by LSD.

Table 6. N Uptake in the wheat whole shoot (soft dough stage) and the grain (crop maturity) at Winnipeg and Portage la Prairie in 1992.

		Winnipeg - 1992		Portage la Prairie - 1992	
Crop Sequence	Fertility kg N ha^{-1}	Shoot N Uptake kg N ha^{-1}	Grain N Uptake kg N ha^{-1}	Shoot N Uptake kg N ha^{-1}	Grain N Uptake kg N ha^{-1}
Alf-Alf-Wheat[z]	0	163.2*ab*	77.7*ab*	137.5*a*	77.2*a*
Alf-Alf-Wheat	80	221.3*a*	90.7*ab*	153.9*a*	71.4*a*
Pea-Bar-Wheat	0	96.1*b*	67.7*b*	68.6*b*	37.2*c*
Pea-Bar-Wheat	80	154.4*ab*	95.6*a*	146.7*a*	47.3*b*
CV (%)[y]		10.8	27.6	21.2	12.4
Crop Sequence		NS[x]	NS	NS	**
Fertility		NS	NS	*	NS
Crop*Fertility		NS	NS	NS	NS

[z] Wheat sampled in 1992 was preceded by 2 years of alfalfa. The alfalfa-wheat sequence received N fertilizer in the wheat phase only. The annual (pea-barley-wheat) sequence received (or did not receive) N fertilizer in all three years of the rotation.

[y] Coefficient of variation (%).

[x] Analysis of variance. NS, non-significant. *,** Significant at $P < 0.05$ and 0.01, respectively.

a-c Means within a column followed by the same letter are not significantly different at $P < 0.05$ by LSD.

Table 7. Changes in total soil nitrogen as affected by alfalfa stand length at Winnipeg and Portage la Prairie from 1990 to 1992.

Site	Alfalfa Stand Length	Change in Total Soil N (Percent)[z]	Change in Total Soil N (kg ha^{-1})[y]	Standard Deviation (kg ha^{-1})
Winnipeg	1 Year	-0.0033	-125	1920
	2 Years	0.0378	1470	2230
	3 Years	0.0645	2510	830
		NS[x]	NS	
Portage la Prairie	1 Year	0.0380	1480	1520
	2 Years	0.0550	2150	900
	3 Years	0.0587	2290	2360
		NS	NS	

[z] Change in total soil N determined by comparison with a non-cropped treatment sampled in the fall of 1990, based on a 0.3 m depth.

[y] Quantification of soil N based on 1 ha, 0.3 m deep, with a bulk density of 1.3 Mg m^{-3} (3900 Mg soil).

[x] Analysis of variance not significant at $P < 0.05$.

5. Summary and Conclusions

This study examined the feasibility of using short term alfalfa stands from the standpoint of optimizing nitrogen related benefits in cropping systems. The objectives were to evaluate the applicability of a special purpose, non-dormant alfalfa as a one year hay and fall nitrogen source in Manitoba, and to develop a basic nitrogen budget that examines the nitrogen dynamics of varying periods of alfalfa in a crop rotation.

Nitro was, in general, very similar to the dormant cultivars in terms of the yield and N_2 fixed in the herbage removed for hay. Small discrepancies among cultivars might be explained by different stages of maturity at cutting time. Nitro was, however, superior as a fall green manure crop, as it returned significantly more biologically fixed N_2 to the soil in the fall relative to the dormant cultivars. Nitro's ability to fix more N_2 in the fall comes, in part, from a greater fixation capacity (increased proportion of N from N_2 fixation, observed in 1992 only), but also from it's non-dormant ability to maintain growth late into the fall. Nitro was not significantly different from the other non-dormant cultivar tested, CUF 101, in terms of growth or N_2 fixation.

The survival of the non-dormant cultivars was significantly lower than the dormant alfalfas, and was likely low enough to facilitate the introduction of a subsequent crop at two of the three site years tested, relative to a preceding dormant cultivar. A nitrogen response was only detected at one of the two sites, and even then differences were mainly between the alfalfas and the previous barley, and not between alfalfa cultivars.

The non-dormant alfalfas exerted a positive effect on the net soil N balance, while an estimated negative effect was observed with the dormant cultivars. Harvest management can have significant effects on the amounts of N returned to the soil, but may also have a profound influence on the growth advantage normally demonstrated by non-dormant alfalfas. Further work is necessary to fine tune the harvest management for Manitoba conditions, such that hay and plow down yields are optimized, and the yields of subsequent crops are better characterized. More work is required to determine if Nitro truly is superior to other non-dormant cultivars as a single year hay and fall nitrogen source, as the observed differences between Nitro and the other alfalfas may simply be a function of Nitro's non-dormant characteristic.

Studies with varying periods of alfalfa in rotation indicated that the two and three year old stands were similar to each other but greater than first year alfalfa in terms of forage N yields, incorporated N with the biomass, amounts of N_2 fixed, and the net soil N balance. The substantial positive effects of alfalfa on the soil N balance, however, did not account for the large, progressive increases in total soil N with each additional year of alfalfa. Large apparent differences in total soil nitrogen among treatments were not significantly different, however, suggesting that the variation associated with testing for total soil N was sufficiently high to obscure true treatment differences. N uptake in the subsequent wheat crop was similar between one and two years of alfalfa, although the effects of residual N on the second and subsequent wheat crops were not determined.

Based on the N yields of hay, incorporated biomass, N_2 fixation, and the apparent changes in total soil N, it would seem that at least two years of alfalfa is necessary to optimize N related benefits. Although third year alfalfa fixed slightly more nitrogen and seemed to add more N to the soil, an advantage over two year old stands of alfalfa was not obvious. The single year, non-dormant Nitro seemed to be superior in fall growth and N_2 fixation compared with the dormant alfalfas tested, and this, combined with it's similarity in performance as a hay source, gave it some advantage as a one year hay and green manure crop relative to the more traditionally used dormant alfalfa cultivars.

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7. Appendices

APPENDIX A

Seasonal Biomass and Nitrogen Accumulation of Nitro

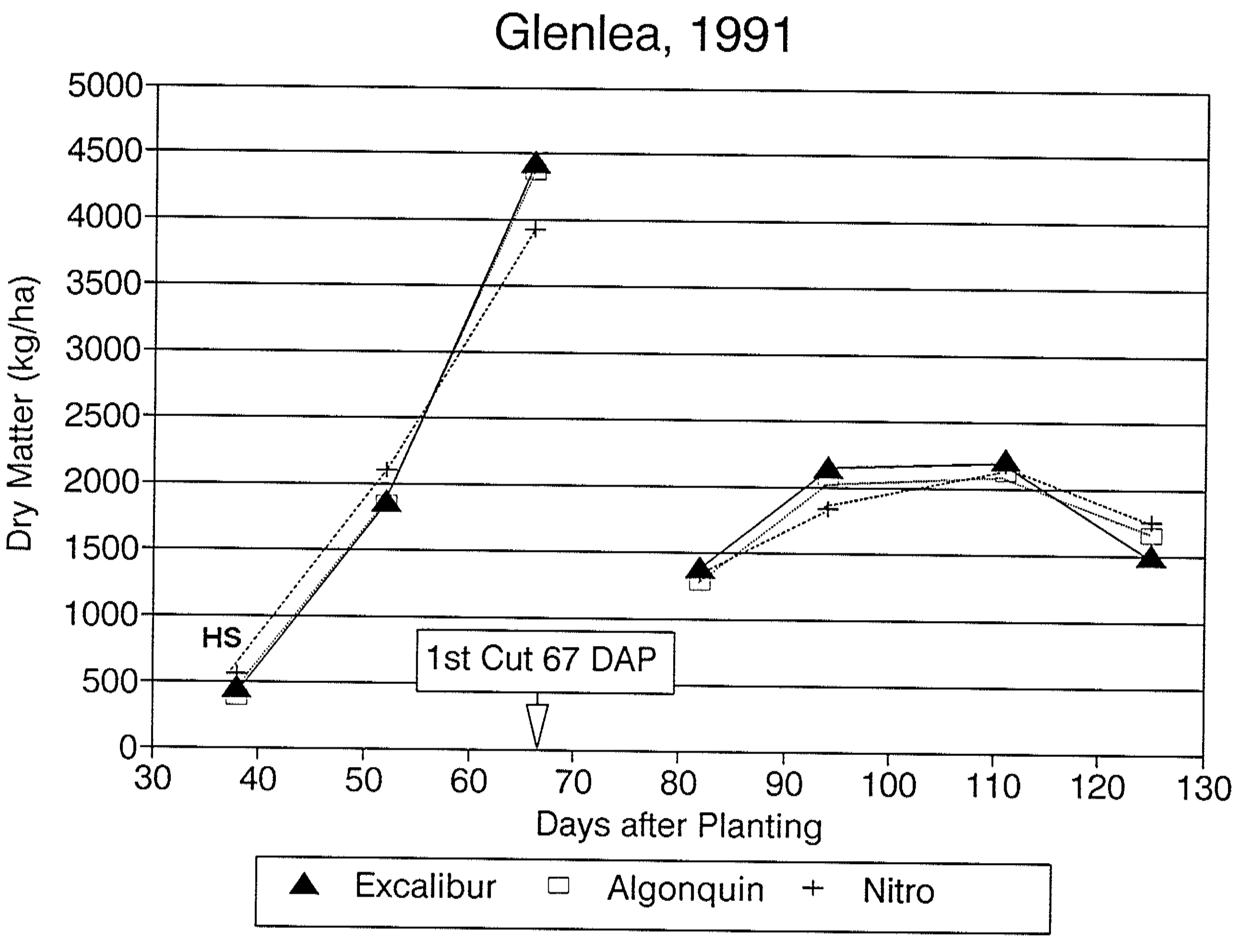

Figure A1. Seasonal above-ground biomass accumulation at Glenlea in 1991. S, HS - Analysis of variance significant at the 0.05 and 0.01 levels, respectively.

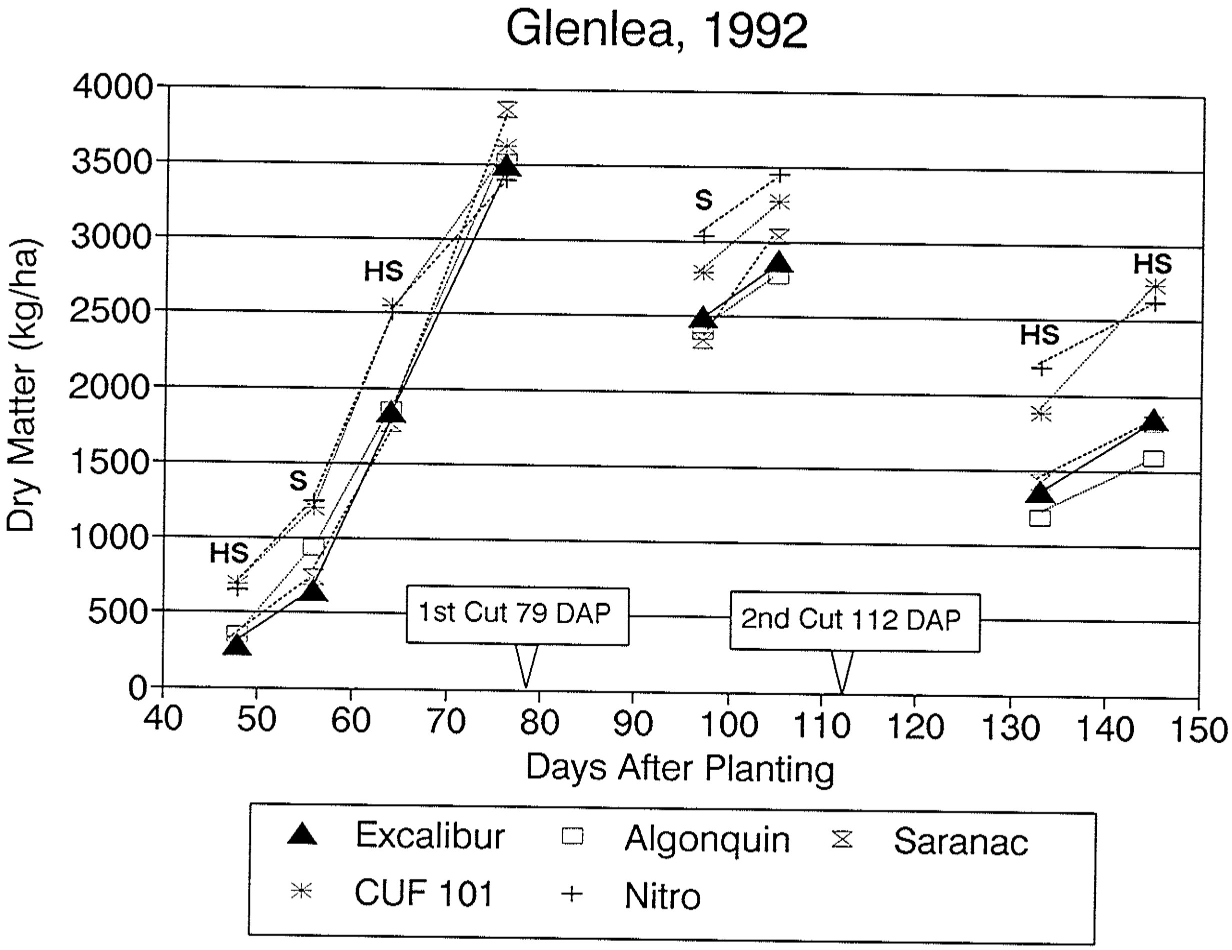

Figure A2. Seasonal above-ground biomass accumulation at Glenlea in 1992. S, HS - Single degree of freedom orthogonal contrast (non-dormant vs dormant cultivars) significant at the 0.05 and 0.01 levels, respectively.

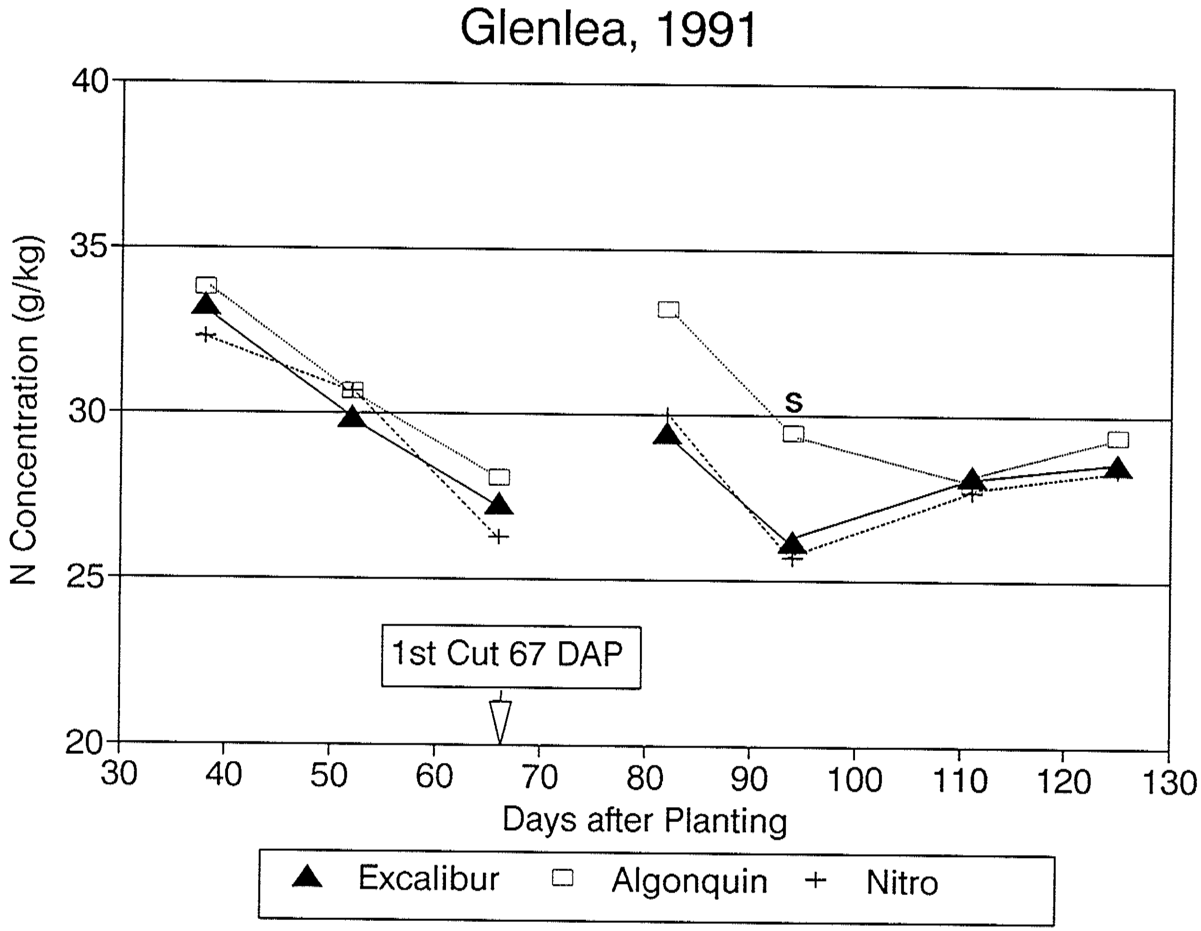

Figure A3. Seasonal changes in herbage N concentration at Glenlea in 1991. S, HS - Analysis of variance significant at the 0.05 and 0.01 levels, respectively.

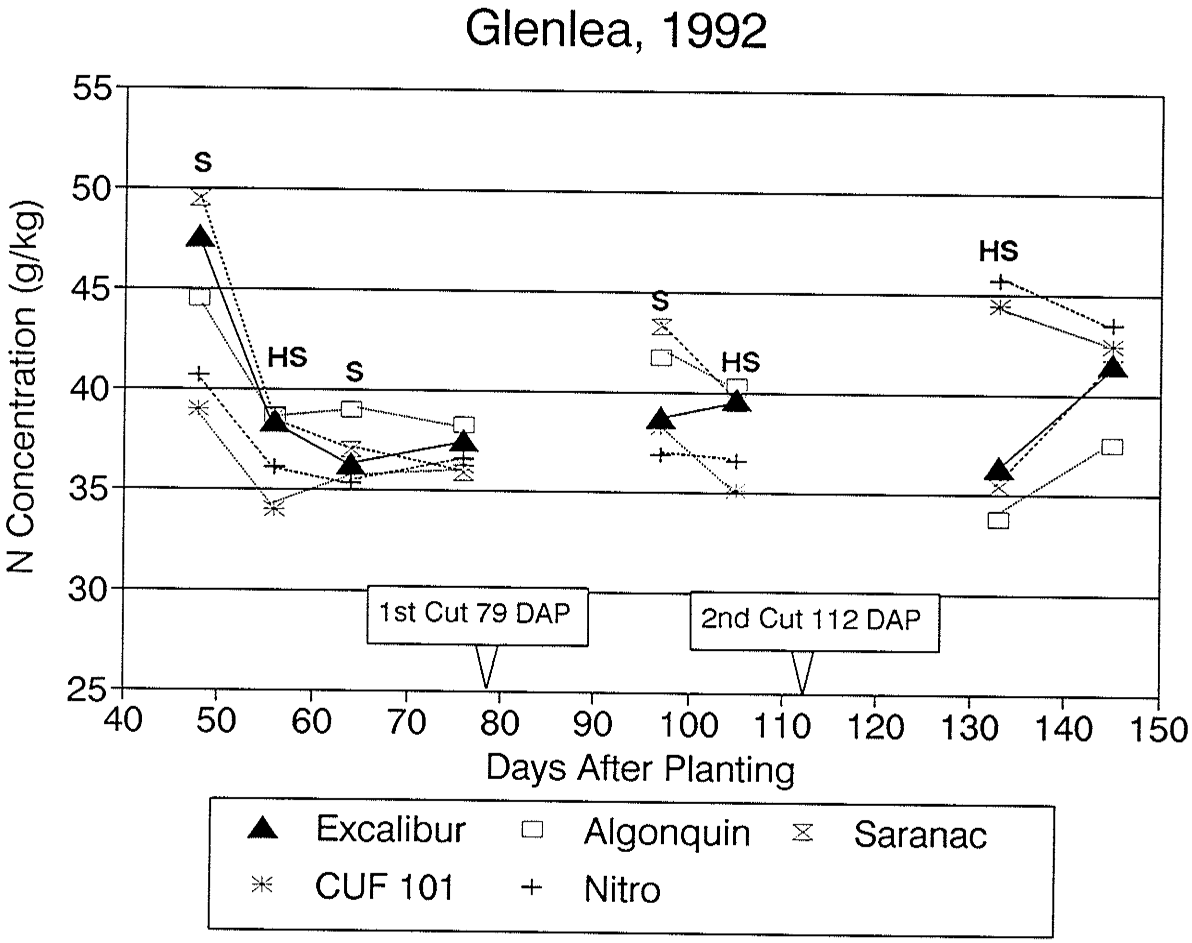

Figure A4. Seasonal changes in herbage N concentration at Glenlea in 1992. S, HS - Single degree of freedom orthogonal contrast (non-dormant vs dormant cultivars) significant at the 0.05 and 0.01 levels, respectively.

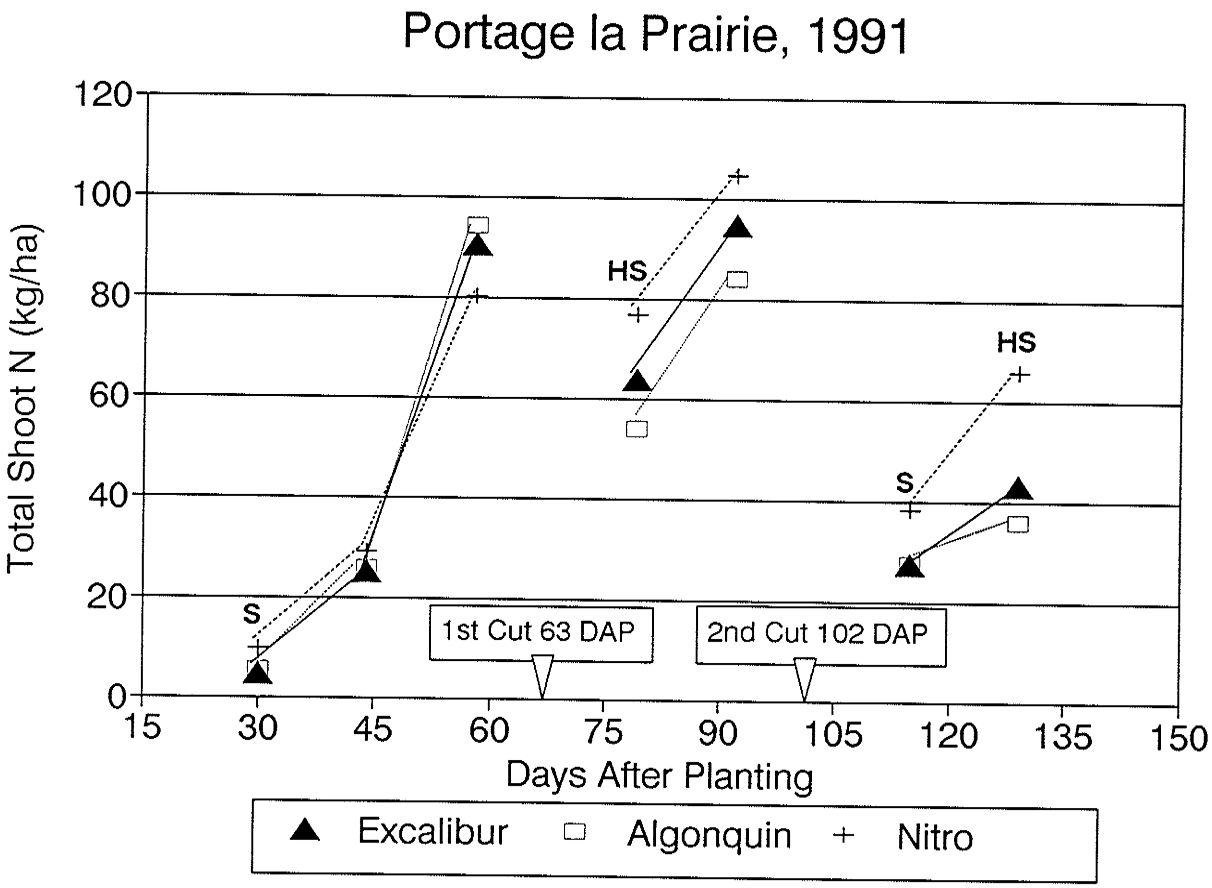

Figure A5. Seasonal changes in total herbage N uptake at Portage la Prairie in 1991. S, HS - Analysis of variance significant at the 0.05 and 0.01 levels, respectively.

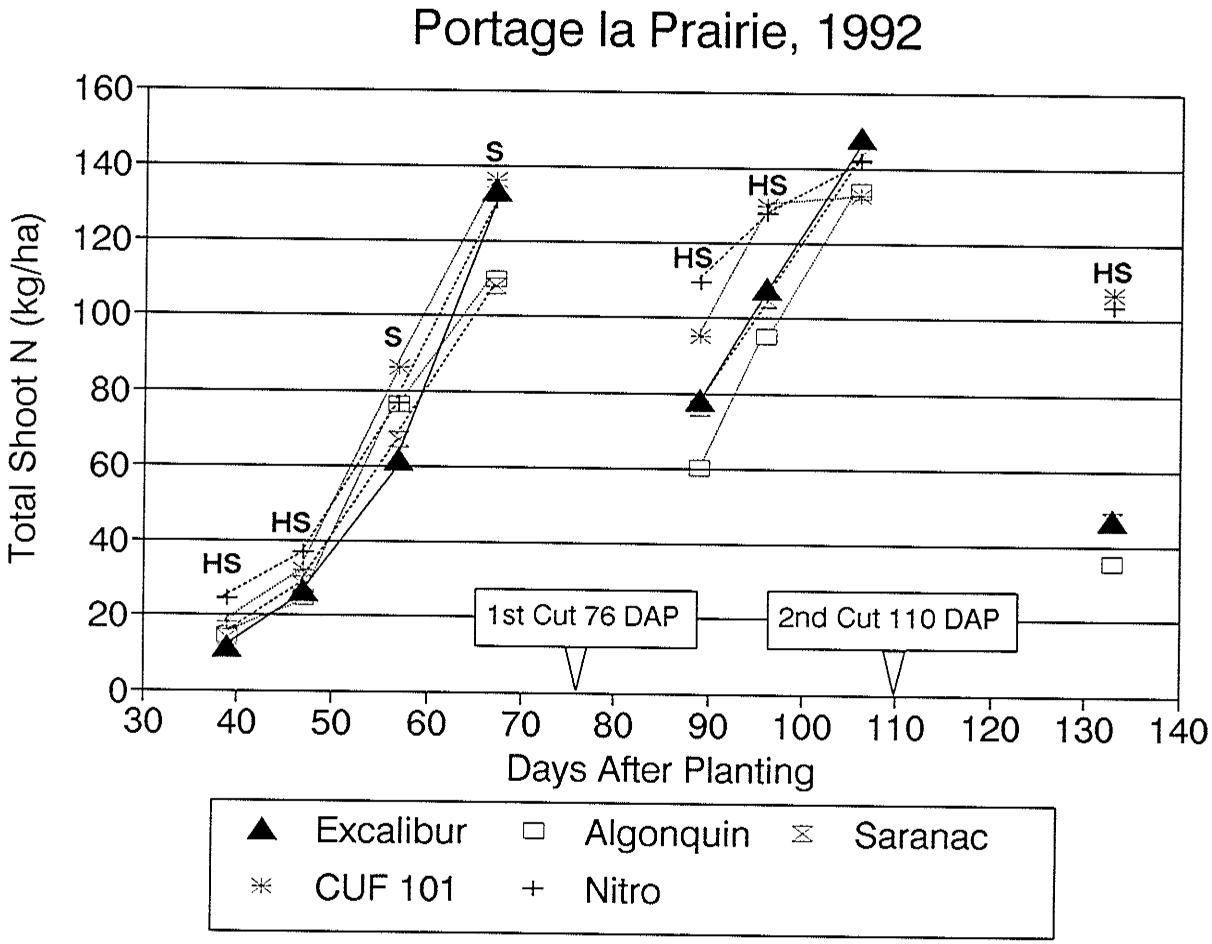

Figure A6. Seasonal changes in herbage N uptake at Portage la Prairie in 1992. S, HS - Single degree of freedom orthogonal contrast (non-dormant vs dormant cultivars) significant at the 0.05 and 0.01 levels, respectively.

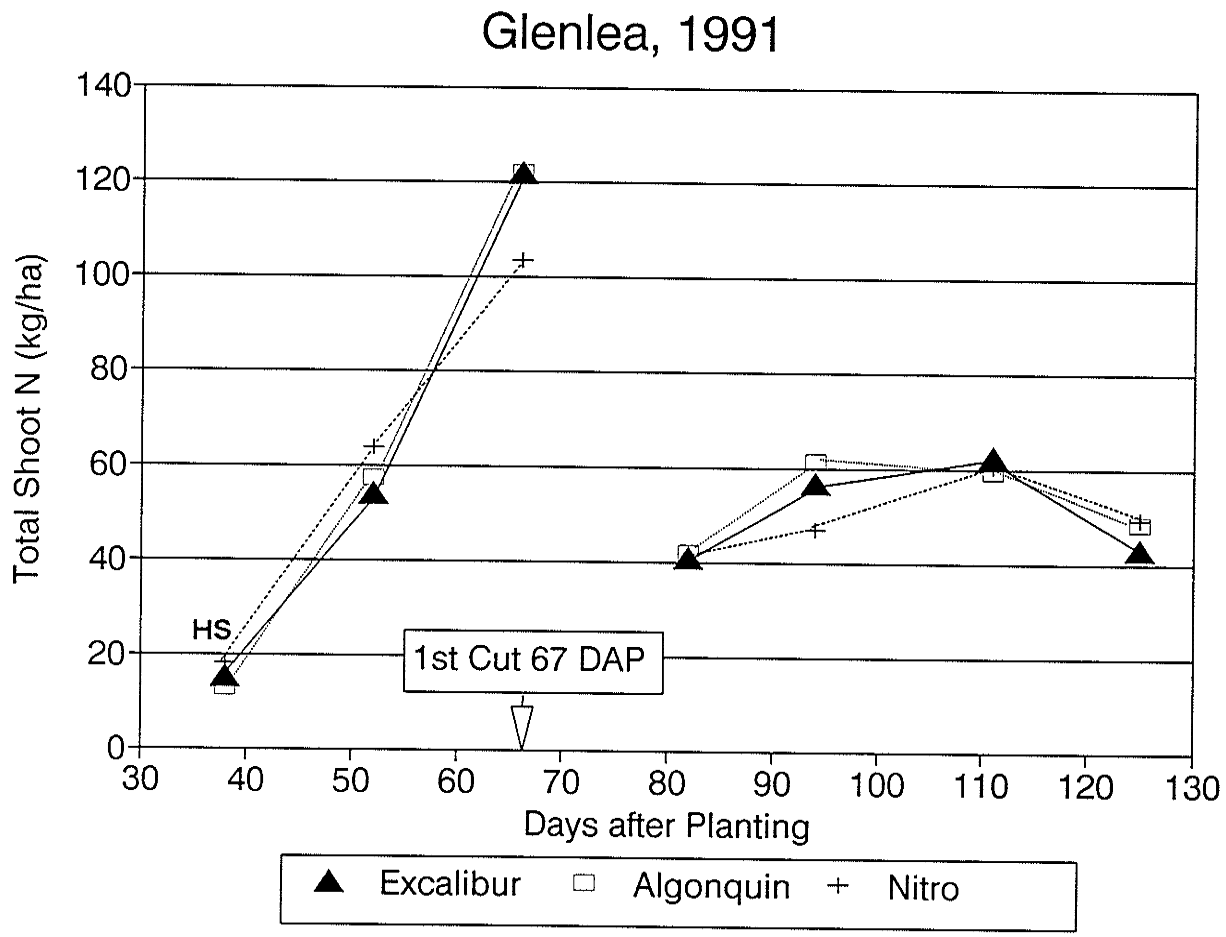

Figure A7. Seasonal changes in total herbage N uptake at Glenlea in 1991. S, HS - Analysis of variance significant at the 0.05 and 0.01 levels, respectively.

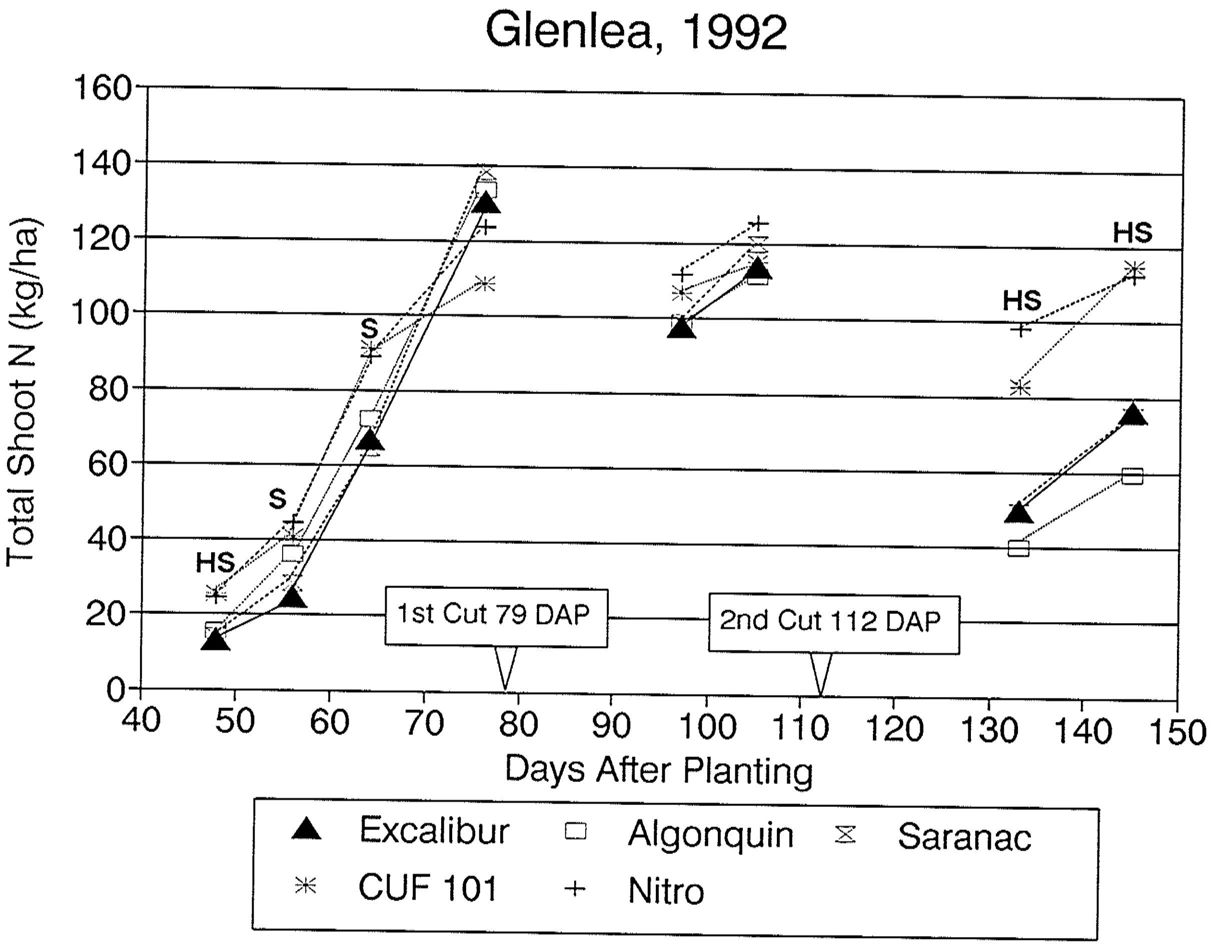

Figure A8. Seasonal changes in herbage N uptake at Glenlea in 1992. S, HS - Single degree of freedom orthogonal contrast (non-dormant vs dormant cultivars) significant at the 0.05 and 0.01 levels, respectively.

APPENDIX B

Nitrogen Fixation of Nitro Determined by Three Different Methods

Table B1. Percent N derived from N_2 fixation (%Ndfa) and the quantity of N_2 fixed (QNF - kg N ha^{-1}) in the herbage as determined by the isotope dilution technique with the barley and Ineffective Saranac (In. Sar.) reference crops, and the difference method (Diff.) using In. Sar. as the reference crop. N_2 fixation data is presented for the first hay harvest at Portage la Prairie and Glenlea in 1992.

Portage la Prairie, 1992 - Cut 1

Cultivar	%Ndfa (Barley)	QNF (Barley)	%Ndfa (In. Sar.)	QNF (In. Sar.)	%Ndfa (Diff.)	QNF (Diff.)
Nitro	74.8*a*	83.6*a*	62.9*a*	70.5*a*	56.2*b*	62.8*b*
CUF 101	76.2*a*	82.7*a*	64.7*a*	70.5*a*	54.3*b*	58.6*b*
Excalibur	70.1*a*	76.8*a*	55.3*a*	58.0*a*	58.4*b*	64.5*b*
Algonquin	71.6*a*	84.1*a*	58.6*a*	68.8*a*	58.1*b*	68.7*b*
Saranac	71.7*a*	100.1*a*	57.7*a*	80.8*a*	64.7*a*	90.0*a*
CV (%)[z]	6.8	11.9	12.1	17.1	5.4	12.5
Contrast[y]	NS	NS	NS	NS	*	**

Glenlea, 1992 - Cut 1

Cultivar	%Ndfa (Barley)	QNF (Barley)	%Ndfa (In. Sar.)	QNF (In. Sar.)	%Ndfa (Diff.)	QNF (Diff.)
Nitro	8.2*a*	8.8*a*	63.9*a*	57.1*a*	47.7*a*	51.2*a*
CUF 101	8.7*a*	9.6*a*	55.8*a*	68.1*a*	48.2*a*	51.4*a*
Excalibur	0.0*a*	0.0*a*	54.6*a*	54.9*a*	42.6*a*	45.3*a*
Algonquin	1.5*a*	1.8*a*	55.5*a*	66.1*a*	51.6*a*	65.6*a*
Saranac	3.1*a*	3.7*a*	49.9*a*	58.9*a*	52.1*a*	62.1*a*
CV (%)[z]	150.0	153.3	14.8	24.5	23.2	38.6
Contrast[y]	*	*	NS	NS	NS	NS

[z] Coefficient of variation (%).

[y] Single degree of freedom orthogonal contrast; nondormant cultivars vs. dormant cultivars. NS, non-significant. *,** Significant at $P < 0.05$ and 0.01, respectively. *a-c* Means within a column followed by the same letter are not significantly different at $P < 0.05$ by LSD.

Table B2. Percent N derived from N_2 fixation (%Ndfa) and the quantity of N_2 fixed (QNF - kg N ha^{-1}) in the herbage as determined by the isotope dilution technique with the barley and Ineffective Saranac (In. Sar.) reference crops, and the difference method (Diff.) using In. Sar. as the reference crop. N_2 fixation data is presented for the second hay harvest at Portage la Prairie and Glenlea in 1992.

Portage la Prairie, 1992 - Cut 2						
Cultivar	%Ndfa (Barley)	QNF (Barley)	%Ndfa (In. Sar.)	QNF (In. Sar.)	%Ndfa (Diff.)	QNF (Diff.)
Nitro	89.8*a*	93.2*a*	59.1*a*	61.4*a*	78.1*a*	81.3*a*
CUF 101	88.8*a*	80.2*a*	54.5*a*	48.9*a*	75.6*a*	68.3*a*
Excalibur	89.6*a*	87.8*a*	58.2*a*	56.9*a*	76.8*a*	75.6*a*
Algonquin	88.2*a*	80.3*a*	52.5*a*	48.3*a*	75.0*a*	68.7*a*
Saranac	88.0*a*	90.6*a*	52.0*a*	53.8*a*	78.2*a*	80.5*a*
CV (%)^z	1.4	9.4	9.2	12.0	2.9	12.2
Contrast^y	NS	NS	NS	NS	NS	NS

Glenlea, 1992 - Cut 2						
Cultivar	%Ndfa (Barley)	QNF (Barley)	%Ndfa (In. Sar.)	QNF (In. Sar.)	%Ndfa (Diff.)	QNF (Diff.)
Nitro	66.4*a*	49.3*a*	37.9*a*	28.1*a*	34.7*b*	25.7*b*
CUF 101	67.3*a*	48.9*a*	39.2*a*	29.4*a*	34.0*b*	26.3*b*
Excalibur	65.6*a*	60.2*a*	37.3*a*	34.9*a*	47.8*a*	45.6*a*
Algonquin	67.0*a*	59.9*a*	40.2*a*	36.4*a*	44.8*a*	40.4*a*
Saranac	63.0*a*	60.5*a*	32.7*a*	36.1*a*	50.3*a*	48.7*a*
CV (%)^z	7.2	12.8	24.3	27.3	13.5	21.3
Contrast^y	NS	*	NS	NS	**	**

^{z}Coefficient of variation (%).

^{y}Single degree of freedom orthogonal contrast; nondormant cultivars vs. dormant cultivars. NS, non-significant. *,** Significant at $P < 0.05$ and 0.01, respectively.

a-c Means within a column followed by the same letter are not significantly different at $P < 0.05$ by LSD.

Table B3. Percent N derived from N_2 fixation (%Ndfa) and the quantity of N_2 fixed (QNF - kg N ha^{-1}) in the herbage as determined by the isotope dilution technique with the barley and Ineffective Saranac (In. Sar.) reference crops, and the difference method (Diff.) using In. Sar. as the reference crop. N_2 fixation data is presented for the incorporated herbage at Portage la Prairie and Glenlea in 1992.

Portage la Prairie, 1992 - Incorporated Herbage

Cultivar	%Ndfa (Barley)	QNF (Barley)	%Ndfa (In. Sar.)	QNF (In. Sar.)	%Ndfa (Diff.)	QNF (Diff.)
Nitro	89.5*a*	85.7*a*	60.0*a*	61.8*a*	85.2*a*	81.6*a*
CUF 101	89.6*a*	90.9*a*	61.7*a*	57.4*a*	86.1*a*	87.3*a*
Excalibur	83.7*bc*	29.0*b*	38.4*bc*	14.3*b*	55.9*b*	20.6*b*
Algonquin	81.3*c*	21.4*b*	30.4*c*	8.7*b*	42.8*b*	12.8*b*
Saranac	84.4*b*	32.5*b*	42.5*b*	17.0*b*	60.4*b*	24.4*b*
CV (%)[z]	2.0	15.5	12.2	19.8	18.8	19.6
Contrast[y]	**	**	**	**	**	**

Glenlea, 1992 - Incorporated Herbage

Cultivar	%Ndfa (Barley)	QNF (Barley)	%Ndfa (In. Sar.)	QNF (In. Sar.)	%Ndfa (Diff.)	QNF (Diff.)
Nitro	76.8*a*	73.9*a*	46.9*a*	44.8*a*	66.1*a*	64.2*a*
CUF 101	76.5*a*	74.6*a*	46.3*a*	45.7*a*	66.4*a*	64.8*a*
Excalibur	69.5*b*	36.8*b*	30.1*b*	16.1*b*	37.5*b*	20.6*b*
Algonquin	62.2*c*	23.4*c*	14.2*c*	5.8*c*	16.1*c*	6.3*b*
Saranac	64.8*bc*	32.3*bc*	19.4*bc*	9.7*bc*	34.6*b*	17.6*b*
CV (%)[z]	5.0	15.1	24.8	26.3	19.4	27.2
Contrast[y]	**	**	**	**	**	**

[z] Coefficient of variation (%).

[y] Single degree of freedom orthogonal contrast; nondormant cultivars vs. dormant cultivars. NS, non-significant. *,** Significant at $P < 0.05$ and 0.01, respectively.

a-c Means within a column followed by the same letter are not significantly different at $P < 0.05$ by LSD.

Table B4. Percent N derived from N_2 fixation (%Ndfa) and the quantity of N_2 fixed (QNF - kg N ha^{-1}) in the herbage as determined by the isotope dilution technique with the barley and Ineffective Saranac (In. Sar.) reference crops, and the difference method (Diff.) using In. Sar. as the reference crop. N_2 fixation data is presented for the incorporated upper roots[z] at Portage la Prairie and Glenlea in 1992.

Portage la Prairie, 1992 - Incorporated Roots

Cultivar	%Ndfa (Barley)	QNF (Barley)	%Ndfa (In. Sar.)	QNF (In. Sar.)	%Ndfa (Diff.)	QNF (Diff.)
Nitro	86.5*ab*	78.2*ab*	63.4*a*	58.5*a*	63.8*a*	60.6*ab*
CUF 101	88.4*a*	86.2*a*	61.4*a*	62.4*a*	69.8*a*	67.8*a*
Excalibur	84.6*abc*	48.7*bc*	57.5*a*	32.3*a*	50.1*a*	28.7*bc*
Algonquin	79.3*c*	41.9*c*	41.6*a*	18.5*a*	43.2*a*	23.8c
Saranac	80.5*bc*	47.1*bc*	48.3*a*	32.5*a*	43.7*a*	28.4*bc*
CV (%)[y]	5.0	35.5	22.9	52.4	32.3	54.1
Contrast[x]	**	**	*	**	*	**

Glenlea, 1992 - Incorporated Roots

Cultivar	%Ndfa (Barley)	QNF (Barley)	%Ndfa (In. Sar.)	QNF (In. Sar.)	%Ndfa (Diff.)	QNF (Diff.)
Nitro	57.2*a*	108.0*a*	37.3*a*	77.7*a*	63.7*a*	120.9*a*
CUF 101	70.9*a*	106.5*a*	61.4*a*	94.0*a*	61.8*a*	95.0*ab*
Excalibur	39.8*a*	34.9*b*	32.2*a*	27.5*b*	38.6*a*	34.1*bc*
Algonquin	46.9*a*	35.9*b*	33.1*a*	24.0*b*	31.2*a*	23.8*c*
Saranac	50.3*a*	52.1*b*	31.3*a*	31.7*b*	43.8*a*	45.6*bc*
CV (%)[y]	29.9	42.4	40.6	50.8	33.3	62.4
Contrast[x]	*	**	*	**	**	**

[z] Sampled roots were trimmed to a length of 10 cm before analysis.

[y] Coefficient of variation (%).

[x] Single degree of freedom orthogonal contrast; nondormant cultivars vs. dormant cultivars. NS, non-significant. *,** Significant at $P < 0.05$ and 0.01, respectively.

a-c Means within a column followed by the same letter are not significantly different at $P < 0.05$ by LSD.

APPENDIX C

Yield and Nitrogen Uptake of Wheat Following Nitro

Table C1. Grain yield, N concentration (conc.), N uptake, and total shoot N uptake (soft dough stage) in wheat preceded by alfalfa and barley at Glenlea in 1992.

	Glenlea, 1992			
Cultivar	Grain Yield kg ha^{-1}	Grain N Conc. g kg^{-1}	Grain N Uptake kg N ha^{-1}	Total Shoot N Uptake kg N ha^{-1}
Nitro	4145*a*	28.1*a*	116.3*a*	129.0a
Excalibur	3903*a*	27.0*a*	105.6*a*	131.9a
Algonquin	3652*a*	28.1*a*	103.3*a*	115.0a
Barley	3974*a*	29.2*a*	116.3*a*	141.1a
CV (%)[z]	14.4	7.5	18.6	23.1
Contrast[y]	NS	NS	NS	NS

[z] Coefficient of variation (%).

[y] Single degree of freedom orthogonal contrast; alfalfa cultivars vs barley.

*,** Significant at $P < 0.05$ and 0.01, respectively.

a-c Means within a column followed by the same letter are not significantly different at $P < 0.05$ by LSD.

APPENDIX D

Nitrate and Water Extraction by Different Years of Alfalfa

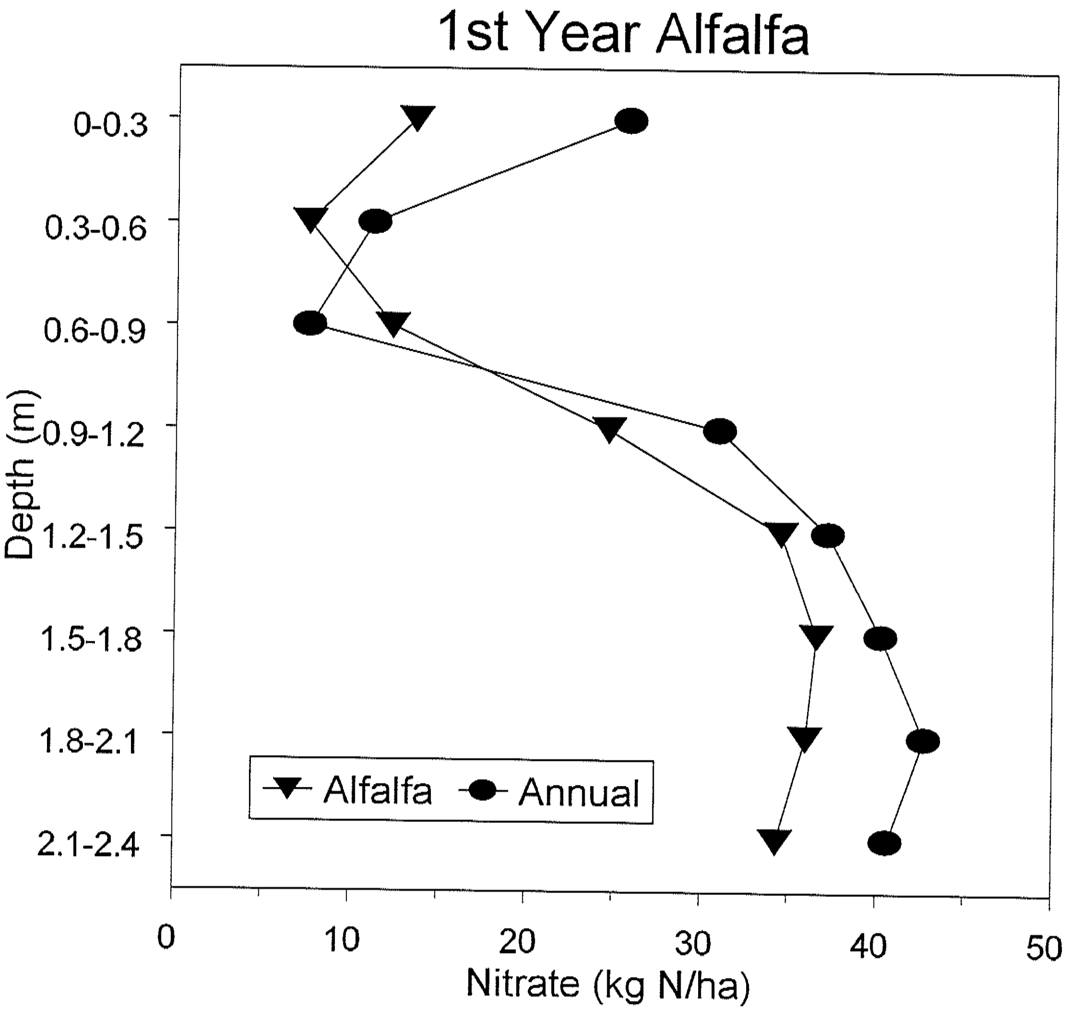

Figure D1. Nitrate extraction by first year alfalfa relative to an annual crop control at Winnipeg in the fall of 1990. Analysis of variance proved non-significant at each depth.

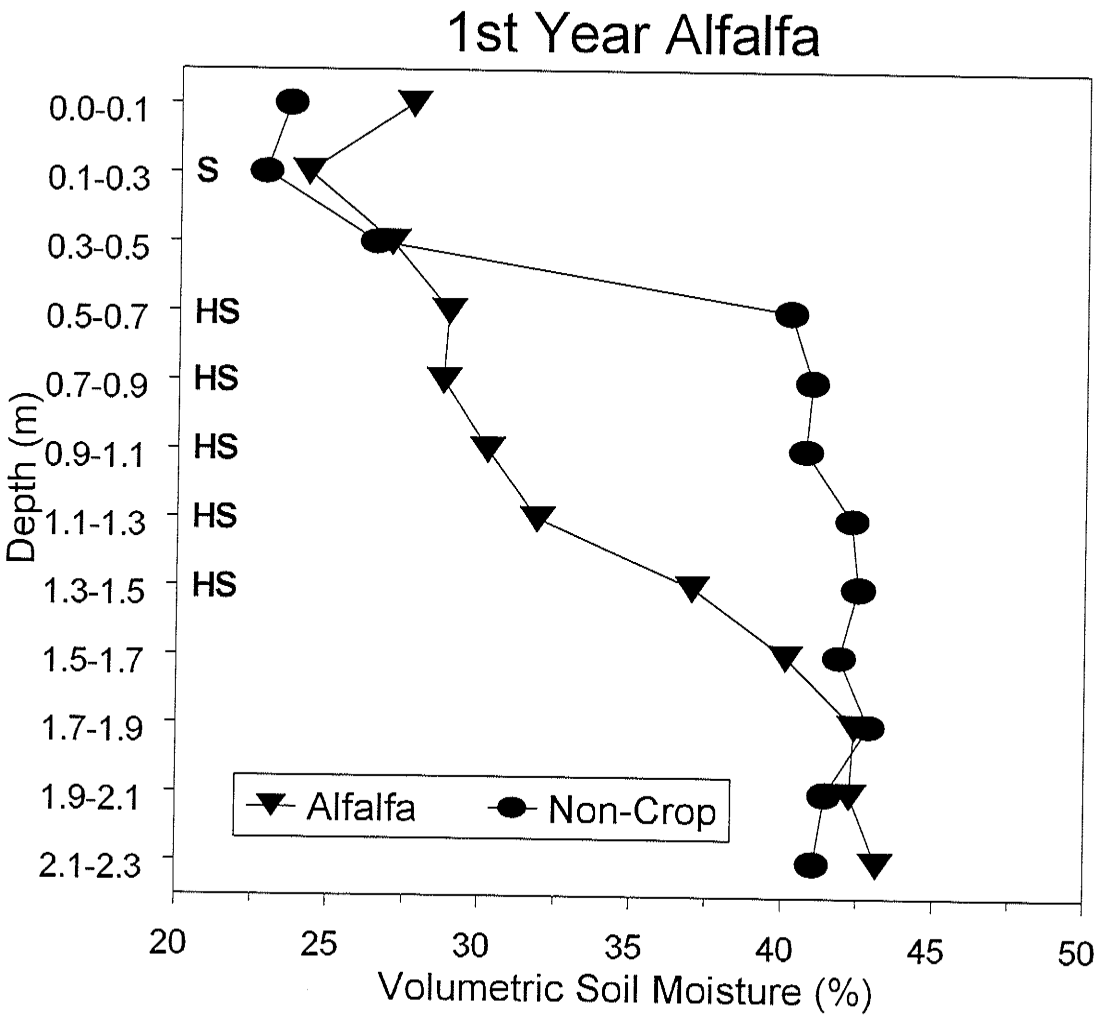

Figure D2. Water extraction by first year alfalfa relative to a non-crop control at Winnipeg in the fall of 1990. S, HS - analysis of variance significant at $P < 0.05$ and 0.01, respectively.

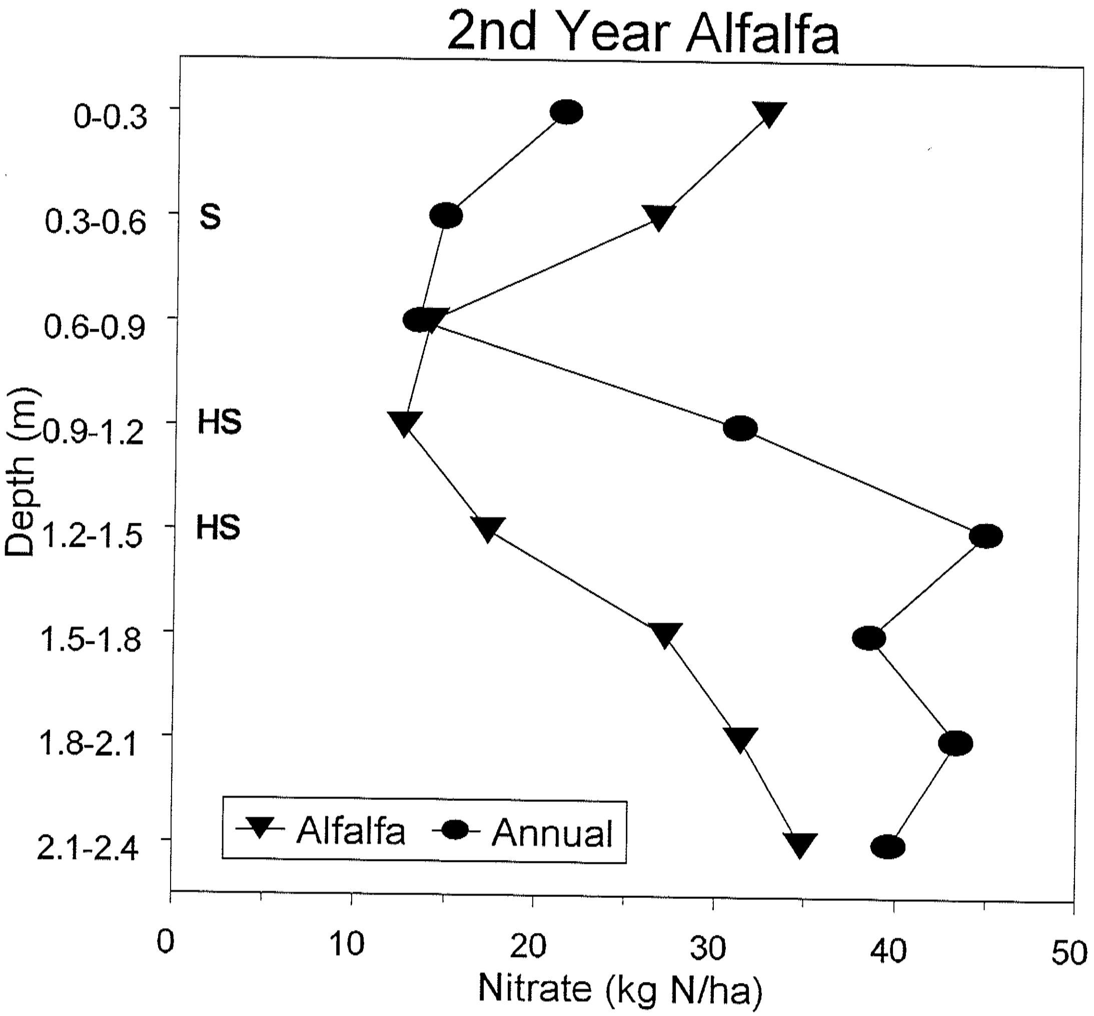

Figure D3. Nitrate extraction by second year alfalfa relative to an annual crop control at Winnipeg in the fall of 1991. S, HS - analysis of variance significant at $P < 0.05$ and 0.01, respectively.

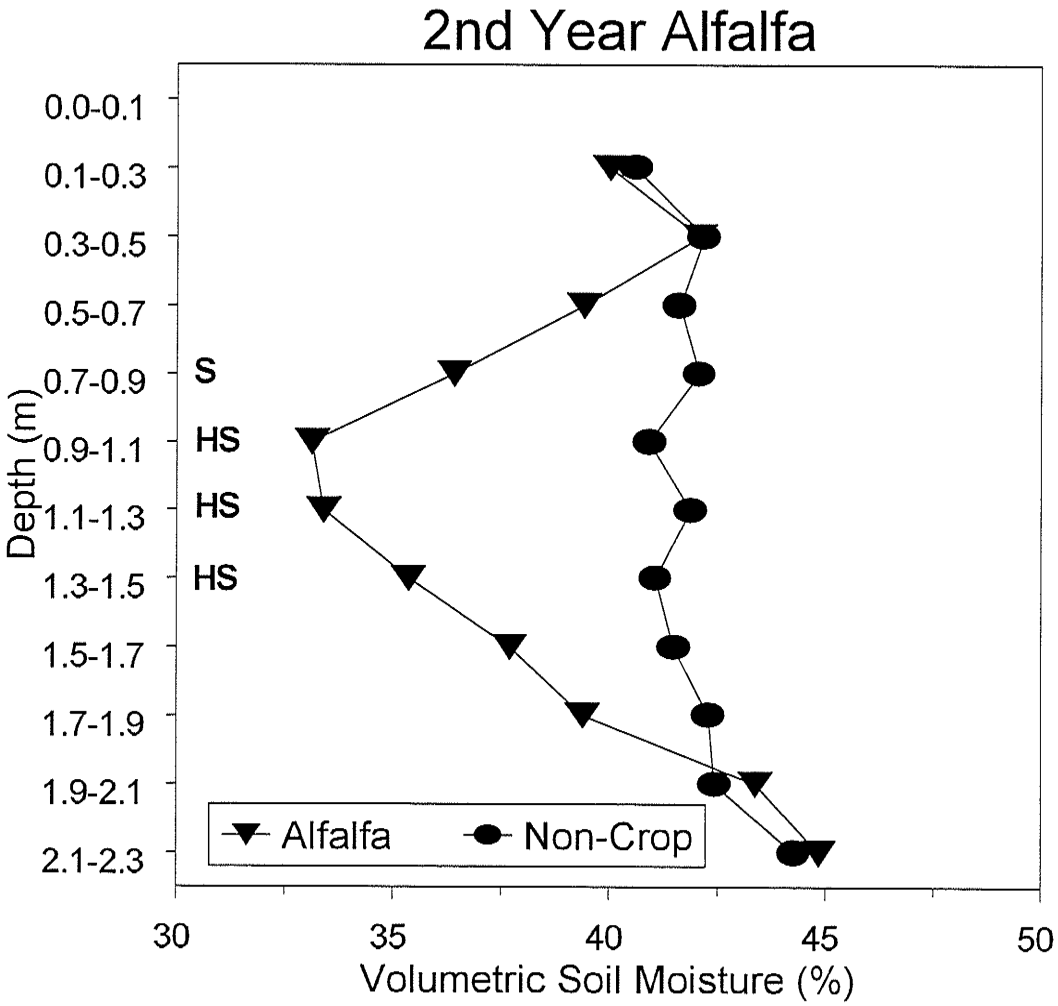

Figure D4. Water extraction by second year alfalfa relative to a non-crop control at Winnipeg in the fall of 1991. S, HS analysis of variance significant at $P < 0.05$ and 0.01, respectively.

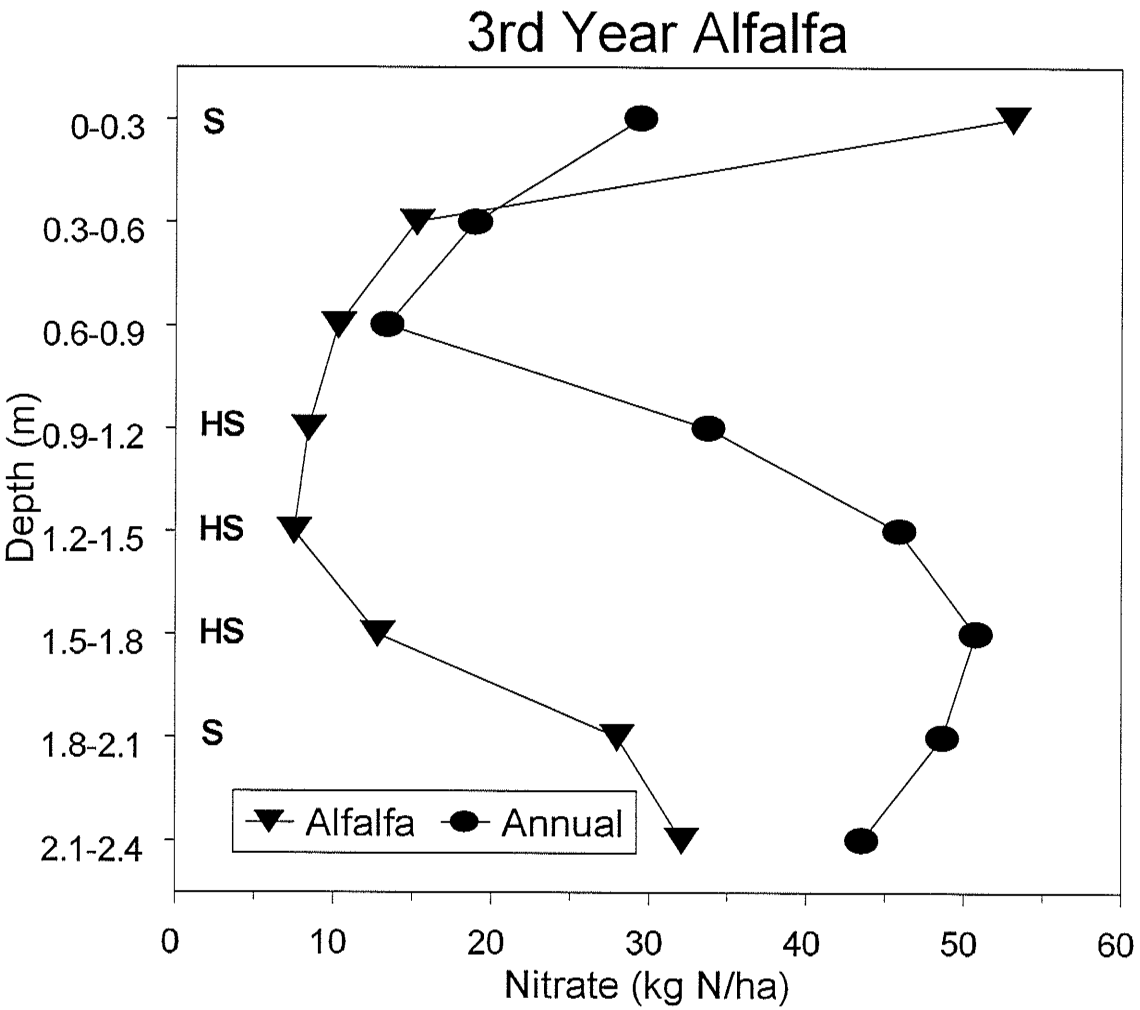

Figure D5. Nitrate extraction by third year alfalfa relative to an annual crop control at Winnipeg in the fall of 1992. S, HS - analysis of variance significant at $P < 0.05$ and 0.01, respectively.

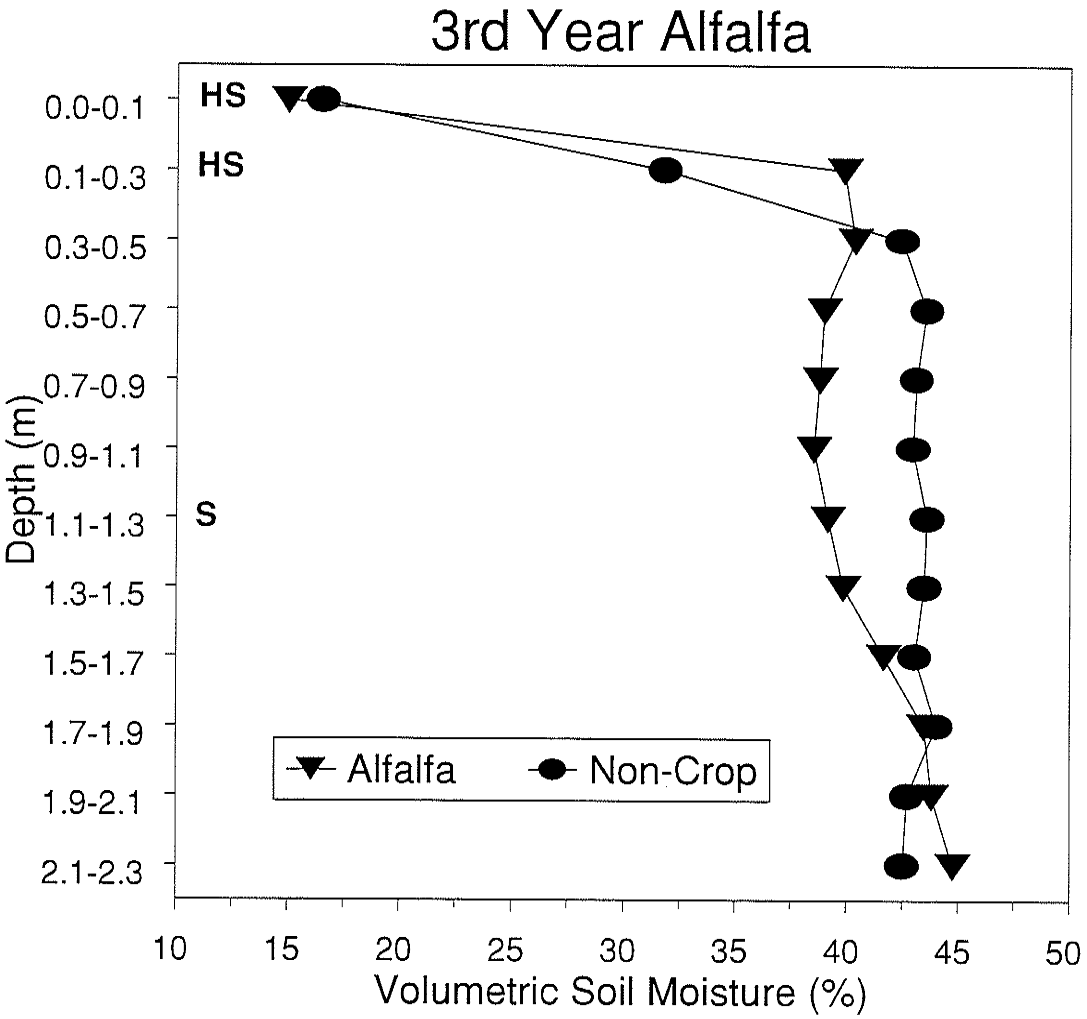

Figure D6. Water extraction by third year alfalfa relative to a non-crop control at Winnipeg in the fall of 1992. S, HS analysis of variance significant at $P < 0.05$ and 0.01, respectively.