This is to certify that the

dissertation entitled

Nitrogen Cycling in Animal-, Legume-, and Fertilizer-Based Cropping Systems

presented by

Glendon Hamilton Harris, Jr.

has been accepted towards fulfillment
of the requirements for

PhD degree in **Crop and Soil Sciences**

Alan B. Hoteman
Major professor

Date **February 5, 1993**

NITROGEN CYCLING IN ANIMAL-, LEGUME-, AND FERTILIZER-BASED CROPPING SYSTEMS

By

Glendon Hamilton Harris, Jr.

A DISSERTATION

Submitted to
Michigan State University
in partial fulfillment of the requirements
for the degree of

DOCTOR OF PHILOSOPHY

Department of Crop and Soil Sciences

1993

ABSTRACT

NITROGEN CYCLING IN ANIMAL-, LEGUME-, AND FERTILIZER-BASED CROPPING SYSTEMS

By

Glendon Hamilton Harris, Jr.

Managing nitrogen (N) for maximum crop use efficiency and minimum environmental impact in agricultural systems that use organic versus fertilizer N inputs requires more information on N cycling in these systems. The overall objective of this research was to compare N cycling in animal-, legume-, and fertilizer-based cropping systems with an emphasis on evaluating each system for the its potential to lose N to the environment. Specific objectives were 1) to trace the fate of red clover (*trifolium pratense* L.) and ammonium sulphate ^{15}N into crops plus soil, and calculate loss by subtraction, 2) measure the soil N supplying capacity using long-term laboratory incubations, 3) measure NO_3-N leaching using lysimeters, and 4) measure the effect of mineralization-immobilization turnover (MIT) on recovery of legume and fertilizer ^{15}N by corn using a modified ^{15}N method.

^{15}N-labeled red clover and ammonium sulphate were applied to microplots in the long-term Rodale farming systems trial located in eastcentral Pennsylvania in 1987

and 1988 and traced into first-year corn (*Zea mays* L.), second-year barley (*Hordeum distichum* L.) and soil after each crop. Over a 2-y period, more fertilizer N was contributed to crops (40 vs. 17 % of input), more legume N was contributed to soil (47 vs. 17 % of input) and a similar amount of N from both sources was lost from the cropping systems (39 % of input).

Long-term (200+ d), aerobic, incubations of soil sampled from the Rodale farming systems in 1988 showed that the N supplying capacity of the animal-based cropping system was greater than the legume- and fertilizer-based systems which were similar. A double exponential model was used to estimate the size of the active fraction of soil organic matter N, which was greater in the animal-than legume-based system, which in turn was greater than in the fertilizer-based system.

Nitrate leaching from the animal-, legume-, and fertilizer-based cropping systems at Rodale was measured over a 2-y period starting in Fall 1990 using 0.76 m dia. by 1.0 m deep intact, natural drainage lysimeters. Most NO_3-N leaching occurred during winter months from all cropping systems and certain crop sequence/N input/climate combinations in each system caused large amounts of NO_3-N to be lost by leaching. Most of the NO_3-N leached was derived from soil organic matter and not from the applied legume and fertilizer N sources as determined using ^{15}N.

^{15}N-labeled fertilizer and legume plant material were applied to unlabeled soil, and vice versa, in field microplots at two locations in Michigan in 1991. It was determined that MIT had no effect on recovery of alfalfa (*Medicago sativa* L.), red clover, hairy vetch (*Vicia villosa* Roth.), or ammonium sulphate ^{15}N by corn.

To my father, Glendon Hamilton Harris, Sr., for always believing in me.

ACKNOWLEDGMENTS

I owe thanks to many people who helped me during my eight year tenure at Michigan State. To Dr. Oran Hesterman, for putting up with me and having great patience while I completed my second degree under his tutelage. To Dr. Eldor Paul, for teaching me everything I know about soil microbiology and for giving me the opportunity to harvest rabbits, pheasants and grouse when tired of harvesting forages and corn. I would also like to thank the other members of my guidance committee, Drs. Boyd Ellis and Tom C. Voice for their assistance.

A special thanks to the C.S. Mott Foundation and Sustainable Agriculture Chair, Dr. Richard R. Harwood, for financial assistance to this project and supporting me with a sustainable agriculture fellowship.

Thanks go to Rodale collaborators Rhonda R. Janke, Kim K. Kroll and Steve Peters; farm manager Jeff Moyer and the rest of his crew; and all the interns who helped me including Lou Saporito. Technical help at MSU from Brian Graff, Jim Bronson, Joe Paling, Dave Harris, Jon Dahl, and Chris Willet was greatly appreciated. Special recognition goes to Dr. T.S.Griffin at the University of Maine for his support and assistance.

Most importantly, I thank my wife, Mary, and my daughter, Michasia: this degree is as much theirs as mine.

PREFACE

Chapters 1, 3, and 4 of this dissertation are written in the style required for publication in the *Agronomy Journal*. Chapter 2 is in the style required for publication in the *Soil Science Society Journal*.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

INTRODUCTION

THE LONG-TERM CROPPING SYSTEMS EXPERIMENT AT THE RODALE INSTITUTE RESEARCH CENTER

The overall objective of the research reported in this dissertation was to compare nitrogen (N) cycling in cropping systems that use animal manure, forage legumes, or inorganic fertilizer as N sources. The potential for animal-, legume-, and fertilizer-based cropping systems to pollute the environment with N was also evaluated. Included in all chapters of this dissertation are results from nitrogen cycling research in a long-term cropping systems experiment located at the Rodale Institute Research Center in eastcentral Pennsylvania. A site description and other pertinent background information regarding management practices, experimental design, and results for that experiment are provided here. Additional information can be found in the cited references and selected bibliography listed at the end of this chapter.

In response to a report on organic farming published by USDA (1980), agronomists at Rodale initiated a long-term cropping systems experiment in 1981 to study the transition from conventional to low-input farming methods. The site is a 6.1 ha field located at the Rodale Institute Research Center in eastcentral Pennsylvania. The soil is primarily a Comly silt loam (fine-loamy, mixed, mesic, Typic Fragiudalf) with a 3 percent south-facing slope. Soil properties at the initiation

of the study were reported by Liebhardt et al. (1989) and Doran et al. (1987) and are listed in Table I.1. Climate at the site was described by Doran et al. (1987) as humid, continental with mean annual air temperature of 12.4 °C and mean annual precipitation of 108 cm.

Prior to 1981, the site was farmed conventionally, i.e. commercial fertilizers and chemical pesticides were used to produce corn and wheat crops. Three cropping systems, each consisting of a five-yr rotation, were initiated in 1981. System 1, called low-input/livestock (LIP-LS) or low-input/animal (LIP-A), included red clover (*Trifolium pratense* L.)/alfalfa (*Medicago sativa* L.) hay, oat (*Avena sativa* L.), winter wheat (*Triticum aestivum* L.), corn (*Zea mays* L.) grain, corn silage and soybean (*Glycine max* Merr.) grain crops. Nitrogen was provided by imported animal (usually cattle) manure plus N in roots and spring regrowth of hay crops incorporated prior to each corn crop. System 2, called low-input cash grain (LIP-CG), produced a cash grain crop of either corn, soybean, oat, winter wheat or spring barley (*Hordeum distichum* L.) each year. Nitrogen was supplied by short-term legume hay or green manure crops [alfalfa, red clover or hairy vetch (*Vicia villosa* Roth.)] plowed down prior to each corn crop. System 3, called conventional (CONV), was a corn-soybean rotation using commercial fertilizers and chemical pesticides (herbicides and insecticides) as recommended by the Pennsylvania State University. Nitrogen fertilizer (usually urea) was applied to corn but not soybean in this system.

Conventional tillage (moldboard plow plus secondary tillage) was used in all three cropping systems, either in spring or fall following winter wheat. Lime was applied in all three systems in December, 1982 (8968 kg ha^{-1}) and potassium sulphate

Table I.1. Initial soil properties in the long-term cropping systems experiment at Rodale.

Soil Property	Value	Depth of sample	Time of sampling	Reference
pH	6.7	0-15 cm	March 1981	Liebhardt et al. (1989)
OM (g kg^{-1})	24	"	"	"
K (cmol$_c$ kg^{-1})	0.56	"	"	"
Ca (" ")	7.6	"	"	"
Mg (" ")	1.4	"	"	"
CEC (" ")	11.8	"	"	"
Bray$_1$ P (kg ha^{-1})	362	0-20 cm	July 1981	"
Carbon (")	32 500	0-15 cm	November 1981	Doran et al. (1987)
Nitrogen (")	2 600	"	"	"

was applied in March, 1989 (336 kg ha^{-1}). No fertilizer N or chemical pesticides were applied to either of the low-input systems. Mechanical (rotary hoe and cultivation) and cultural (crop rotation and relay cropping) methods were used to control weeds in the low input systems. These same cultural methods also aided in controlling insect and disease pests.

At the start of the experiment, each cropping system was initiated at three different phases or entry points of its rotation to obtain information about the effect of the starting crop on the conversion process. The experimental design was a split-plot randomized block with eight replications. Cropping systems were main plots and measure 18.3 by 91.5 m. Crop rotation entry points were subplots and measure 6.1 m by 91.5 m. The cropping sequence for all nine treatments (three cropping systems x three entry points) for 1981-1992 is shown in Table I.2.

Corn grain yields obtained in the low-input systems were 75 % of yields obtained in the conventional system for the first four years of the study, 1981-1984 (Liebhardt et al., 1989). Weed competition and insufficient N were reported as the major factors limiting corn yields during this period. In 1985, corn grain yields were comparable in both the LIP-CG and CONV systems. Soybean yields in the low-input systems were equal to or greater than yields in the conventional system for the 1981-1985 period. Small grain and hay crops were produced only in the low-input systems and were comparable to local county averages each year. Liebhardt et al. (1989) concluded that a favorable transition from conventional to low-input systems is feasible, but only if the conversion is started with crops that demand less N and/or are competitive with weeds (i.e. small grains, soybean, forage legumes).

Table 1.2. Cropping sequence for the low-input/cash grain (LIP-CG), conventional (CONV) and low-input/with animals (LIP-A) rotation treatments of the long-term Rodale experiment for 1981-1992.

Table I.2. Cropping sequence for the low-input/cash grain (LIP-CG), conventional (CONV) and low-input/with animals (LIP-A) rotation treatments of the long-term Rodale experiment for 1981-1992.

Cropping system	Rotation entry point	Year											
		1981	1982	1983	1984	1985	1986	1987	1988	1989	1990	1991	1992
		Crop †											
LIP-CG	1	O/RC	C	O/RC	C	S	O/RC	C	SB/W	W/RC	C	S/W	W/HV
	2	S	O/RC	C	W/HV	C	SB/S/W	W/RC	C	SB/S	O/RC	W/HV	C
	3	C	S	O/RC	C	O/RC	C/W	W/S	O/RC	C	SB/S	C/R	S
CONV	1	C	C	S	C	S	C	C	S	C	S	C	C
	2	S	C	C	S	C	S	C	C	S	C	S	C
	3	C	S	C	C	S	C	S	C	C	S	C	S
LIP-A	1	O/RC	RC	C	S	CS/W	W/RC	RC	C	S	CS/W	W/RC	RC
	2	C	S	CS	W/RC	RC	C	S	CS/W	W/RC	RC	C/R	S
	3	CS	W/RC	RC	C	S	CS/W	W/RC	RC	C	S/R	CS/W	W/RC

† C = corn grain, CS = corn silage, HV = hairy vetch, S = soybean, SB = spring barley, O = oat, R = ryegrass, RC = red clover, W = winter wheat

Originally called the "conversion project", the study was renamed the "farming systems trial " in 1986, and was continued in order to assess the long-term reliability, sustainability, and environmental impact of the low-input systems. Results from the second five-year rotation period have been reported by Peters et al. (1992). On average, corn grain yields in the low-input systems were equal to the conventional system for 1986-1990. In 1988, when a drought-period occurred from early June to mid-July, corn yields were higher in the low-input systems than in the CONV system. Average soybean yields in the LIP-A and CONV systems were equal for the 1986-1990 period. Soybean in the LIP-CG system were relay cropped with spring barley and as a result yielded 85 % of the CONV system.

Soil nitrate-N levels in corn plots during the 1986-1990 period were higher in the LIP-A system than in either the LIP-CG or CONV systems which were similar. Total soil N levels measured in 1981 and again 1991 showed a slight increase in the LIP-A system, no change in the LIP-CG system and a slight decrease in the CONV system. Soil organic matter levels were measured periodically between 1981 and 1989 and showed an increase in all three systems with the greatest increase in the low-input systems.

The gradual increase, then leveling off, of corn grain yields in the low-input systems (illustrated in Figure I.1) was cited as evidence that a new equilibrium between soil processes and plant growth -- where N is not limiting -- was reached in these systems by 1985 (Janke et al., 1991, Peters et al. 1992). It was also hypothesized that repeated applications of animal and legume green manures resulted in an enhancement of the active soil organic matter fraction and greater internal N

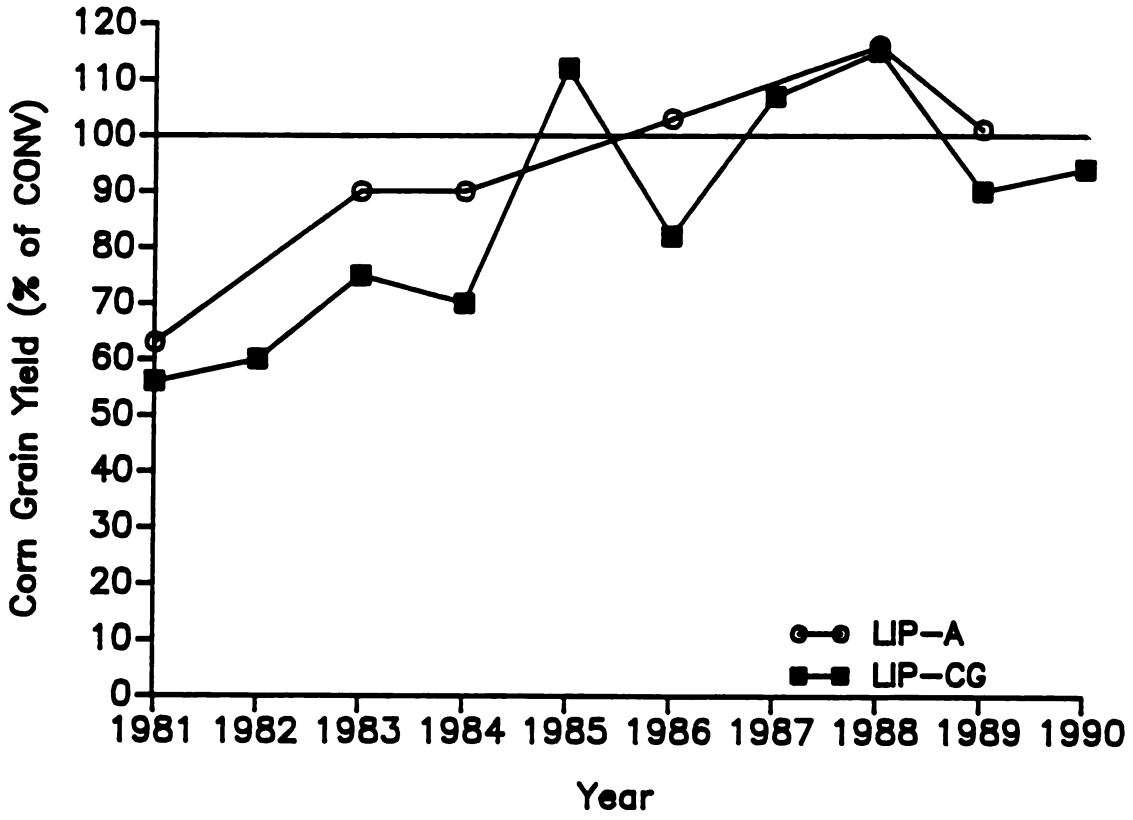

Figure I.1. Corn grain yields in the low-input/cash grain (LIP-CG) and low-input/animal (LIP-A) cropping systems compared to the conventional (CONV) cropping system of the long-term experiment at Rodale for 1981-1990.

Source of data: Liebhardt et al., 1987; Peters et al., 1992.

cycling in the low-input systems (Radke et al., 1988). The ^{15}N experiments reported in the following chapters of this dissertation were designed to test the above hypotheses, as well as to assess the environmental impact of animal-, legume-, and fertilizer-based cropping systems.

REFERENCES

Doran, J.W., D.G. Fraser, M.W. Culik, and W.C. Liebhardt. 1987. Influence of alternative and conventional agricultural management on soil microbial processes and nitrogen availability. Am. J. Alt. Agric. 2:99-106.

Liebhardt, W.C., R.W. Andrews, M.N. Culik, R.R. Harwood, R.R. Janke, J.K. Radke, and S.L. Rieger-Schwartz. 1989. Crop production during conversion from conventional to low-input methods. Agron. J. 81:150-159.

Peters, S.E., R. Janke, and M. Bohlke. 1992. Rodale's farming systems trial 1986-1990. Rodale Institute, Rodale Press, Emmaus, PA.

Janke, R.R., J. Mt.Pleasant, S.E. Peters, and M. Bohlke. 1991. Long-term, low-input cropping systems research. p. 291-317 *In* Sustainable agriculture field research and education: a proceedings. National Research Council, National Academy Press, Washington, D.C.

Radke, J.K., R.W. Andrews, R.R. Janke, and S.E. Peters. 1988. Low-input cropping systems and efficiency of water and nitrogen use. p. 193-218 *In* W.L. Hargrove (ed.) Cropping strategies for efficient use of water and nitrogen. ASA Spec. Publ. 51.

U.S. Department of Agriculture. 1980. Report and recommendations on organic farming. Washington, D.C.: U.S. Department of Agriculture.

SELECTED BIBLIOGRAPHY

Andrews, R.W., S.E. Peters, R.R. Janke, and W.W. Sahs. 1990. Converting to sustainable farming systems. p. 281-313 *In* C.A. Francis, C.B. Flora, and L.D. King (ed.) Sustainable Agriculture in Temperate Zones. John Wiley & Sons:New York.

Culik, M.N. 1983. The conversion experiment:reducing farming costs. J. Soil Water Conserv. 38:333-335.

Faeth, P.R., R. Reppetto, K.Kroll, Q. Dai, and G. Helmers. 1991. Paying the farm bill: U.S. agricultural policy and transition to sustainable agriculture. World Resources Institute, Washington, D.C.

Harwood, R.R. 1984. Organic farming research at the Rodale Research Center. p. 1-17 *In* D.F. Bezdicek, J.F. Power, D.R. Keeney, M.J. Wright, D.M. Kral, and S.L. Hawkins (ed.) Organic farming:current technology and its role in sustainable agriculture. ASA Spec. Publ. 46.

Harwood, R.R. 1985. The integration efficiencies of cropping systems. p. 64-75 *In* T.C. Edens, C. Frigeon, and S.L. Battenfield (ed.) Sustainable agriculture and integrated farming systems: 1984 Conf. Proc. Michigan State Univ. Press, East Lansing, MI.

Werner, M.R., and D.L. Dindal. 1990. Effects of conversion to organic agricultural practices on soil biota. Am. J. Alt. Agric. 5:24-32.

[This page left blank intentionally.]

CHAPTER ONE

FATE AND BEHAVIOR OF LEGUME AND FERTILIZER ^{15}N IN A LONG-TERM CROPPING SYSTEMS EXPERIMENT

ABSTRACT

An ^{15}N tracer study was conducted within a long-term cropping systems experiment to compare the fate and behavior of applied legume and fertilizer nitrogen (N) inputs. ^{15}N-labeled red clover (*Trifolium pratense* L.) and ammonium sulphate were applied to microplots within the low-input/cash grain and conventional cropping systems of the Farming Systems Trial at the Rodale Institute Research Center located in Kutztown, PA. The ^{15}N was applied to soil and traced into corn (*Zea mays* L.) in 1987 and 1988. Residual ^{15}N was also traced into second-year spring barley (*Hordeum distichum* L.) crops in 1988 and 1989. Legume and fertilizer ^{15}N remaining in soil after each crop was measured and loss of N was calculated by subtraction. Overall, more fertilizer than legume N was recovered by crops (40 vs. 17 % of input), more legume than fertilizer N was retained in soil (47 vs. 17 % of input), and similar amounts of N from both sources were lost from the cropping systems (39 % of input). More fertilizer than legume N was lost during the year of application (38 vs. 18 % of input), but more legume than fertilizer N was lost the year following application (17 vs. 4 % of input). Corn uptake and loss of legume N were not affected by the drought conditions of 1988 (when results were compared to

1987). However, recovery of fertilizer N by corn decreased (from 49 to 29 % of input) and loss of fertilizer N increased (from 30 to 46 % of input) in 1988 compared to 1987. Residual fertilizer and legume ^{15}N was distributed similarly among soil fractions (mostly into organic forms) and largely unavailable for uptake by second-year barley. Soil microbial biomass was larger in the legume-based than in the fertilizer-based system, but specific activity (C and N mineralization divided by biomass C and N) was the same. It was concluded that 1) significant amounts of legume N can be lost from cropping systems, especially during the year following incorporation, and therefore must be managed year-round, 2) legume-based systems are more drought tolerant than are fertilizer-based systems in terms of N loss, and 3) a larger, but not necessarily more active, soil microbial biomass is responsible for the greater soil N supplying capacity in the legume- compared to fertilizer-based system.

INTRODUCTION

Managing nitrogen (N) inputs to achieve agronomic, economic, and environmental sustainability is a major challenge facing modern agriculture. Relying less on commercial fertilizer N and more on N fixed biologically by legumes has been suggested as a way to meet this challenge (Keeney, 1982; NAS, 1989). Experiments comparing the fate and behavior of fertilizer versus legume N, and especially quantifying N losses, are needed (Smith et al., 1990; Smith et al., 1987; Peterson and Russelle, 1991;, NAS, 1989; Rosswall and Paustian, 1984).

^{15}N tracer methodology is recognized as a valuable tool for determining the fate and behavior of N applied in the environment (Hauck, 1971, 1982; L'Annunziata and Legg, 1984). Hundreds of agriculturally-related laboratory, greenhouse, and field experiments using ^{15}N have been reported. Most ^{15}N field experiments have studied the recovery of fertilizer N by crops. It is well documented that the use efficiency of fertilizer N by crops varies due to a number of factors, including timing and method of N application, tillage method, and climate. A well-managed, first-year, single-harvested crop recovers between 50 and 70 % of applied fertilizer N (Allison, 1966; Stanford, 1973). In addition, 10 to 40 % of applied fertilizer N may remain in soil, 5 to 10 % may be lost by leaching, and 10 to 30 % may be lost to the atmosphere in gaseous forms (Kundler, 1970; Westerman et al., 1972).

Until recently, there were limited data available on the fate of ^{15}N from

legume residues decomposing under field conditions. Studies using different legume species and recovery crops in various parts of the world have now been reported and show that: 1) less than 30 % of legume N is recovered by a subsequent non-legume crop, 2) large amounts of legume N are retained in soil, mostly in organic forms, 3) total recovery of legume N in crops and soils after one year averages 70 to 90 %, and 4) less than 5 % of legume N from the original application is recovered by a second non-legume crop (Harris and Hesterman, 1990; Ladd et al., 1983; Muller and Sundman, 1988; Ta and Faris, 1990).

Direct comparisons of fertilizers versus legumes as an N source for crops using ^{15}N in field experiments are rare, and none have been reported for cropping systems common to the northcentral and northeastern regions of the U.S. Westcott and Mikkelson (1987) compared vetch (*Vicia beghalensis* L.) and ammonium sulphate as N sources for flooded rice (*Oryza sativa* L.) using ^{15}N in the field. They found that rice recovered twice as much fertilizer N as vetch N when both sources were applied at either 60 kg ha^{-1} (18 vs. 9 % recovery) or at 120 kg ha^{-1} (52. vs 26 % recovery). Greater recovery of vetch N in soil resulted in similar total ^{15}N recoveries of legume and fertilizer N in the soil-plant system. Janzen et al. (1990) compared the annual legumes Tangier flatpea (*Lathyrus tingitanus* L.) and lentil (*Lens culinaris* L.) to ammonium sulphate fertilizer as an N source for wheat at three locations in Canada. Wheat recovered an average of 14 % of applied legume N and 36 % of fertilizer N. More legume N than fertilizer N was retained in soil and total recovery of legume N was often, but not always, greater than total recovery of fertilizer N. Ladd and Amato (1986) compared medic (*Medicago littoralis* L.) to three different

fertilizers as a N source for wheat in Australia. They also reported greater recovery of fertilizer N than legume N by wheat (46 vs. 17 %), greater retainment of legume N than fertilizer N in soil (62 vs. 29 %), and a similar total recovery of each N source in the soil-plant system.

The similar values for total ^{15}N recovery (in crops plus soil) reported in the studies above indicate that similar amounts of legume and fertilizer N were lost to the environment. This contradicts the belief by some that legume N is less susceptible than fertilizer N to losses (Bezdicek and Granastein, 1989; Papendick, 1987; USDA, 1980). Since loss of N from either source represents both an economic loss and potential pollution of the environment, better estimates of N loss from legume and fertilizer-based cropping systems are needed.

Despite reports of greater recovery of N from fertilizer sources than from legume sources by wheat (Ladd and Amato, 1986; Ta and Faris, 1990), little difference in wheat yields or total N uptake was observed. Lower use efficiency of legume N by wheat was thus associated with greater uptake of soil N. Bolton et al. (1985) suggested that greater soil N supplying capacities in legume-based than fertilizer-based systems, due to larger and more active microbial populations, can compensate for low crop use efficiency of legume N. Legume N inputs may also contribute more than fertilizer N inputs to long-term soil fertility through buildup of organic N reserves (Frye et al., 1985; Harris and Hesterman, 1990; Janzen et al., 1990; Ladd et al., 1981). However, Ladd and Amato (1986) and Ta and Faris (1990) both reported that little residual ^{15}N from either the fertilizer or legume sources was taken up by a second wheat crop. The true value of legume N inputs may therefore

be to affect conservation and cycling of soil N rather than providing a direct N source in the short- or long-term.

In a recent report on alternative agricultural systems (NAS, 1989), a need was expressed for nutrient cycling research in established cropping systems, emphasizing leguminous crops and potential for nutrient loss. The goal of the research reported herein was to compare the fate and behavior of legume and fertilizer N in a long-term experiment with cropping systems common to the northeastern and northcentral regions of the U.S. Specific objectives included: 1) to follow red clover and ammonium sulphate ^{15}N applied to established legume- and fertilizer-based cropping systems into crops and soil, and to calculate N loss during two growing seasons; 2) to measure and compare the recovery and distribution of legume and fertilizer N in three soil fractions (inorganic, microbial biomass, and non-biomass organic); and 3) to measure and compare the size and activity of the microbial biomass in soils from the legume- and fertilizer-based systems.

MATERIALS AND METHODS

The long-term experiment. The long-term experiment used for this ^{15}N tracer study was the "farming systems trial" located at the Rodale Institute Research Center in eastcentral Pennsylvania. Originally called the "conversion project", the experiment was established in 1981 to investigate yield-limiting factors when converting from conventional to low-input farming methods. A description and results of this study have been reported by Liebhardt et al. (1989) for the first five years (1981-1985), and by Peters et al. (1992) for the second five years (1986-1990). Two low-input cropping systems, one with an animal component and the other a cash grain rotation, were compared to a conventional corn-soybean rotation. The main N source for each cropping system was animal manure for the low-input/animal (LIP-A), legume green manure for the low-input/cash grain (LIP-CG), and inorganic fertilizer for the conventional (CONV). The N was applied before each corn crop in all systems. Chemical herbicides and insecticides were used in the conventional but not in the low-input systems. Each cropping system was originally initiated at three different phases of its rotation, or "entry points", in order to assess the effect of the starting crop on the conversion process. Experimental design was a split-plot, randomized complete block with eight replications. Cropping systems were main plots and rotation entry points were subplots. Each subplot measured 6.1 x 91.5 m. The soil was a Comly silt loam (fine-loamy, mixed, mesic, Typic Fragiudalf).

Climate was humid continental with annual precipitation of 108 cm and an annual mean temperature of 12.4 °C.

Two of the three entry points for the LIP-CG and CONV systems were used in this study. The cropping sequence for treatments used, up to and including the crop grown the year of ^{15}N application, are shown in Table 1.1. Soil properties for the LIP-CG and CONV systems were measured in 1987 and reported by Peters et al. (1992). Average pH was 6.8, organic matter was 24 g kg^{-1}, Bray P1 was 338 kg ha^{-1}, and potassium, calcium, magnesium, and CEC were 0.32, 6.4, 1.9 and 9.9 $cmol_{(+)}$ kg^{-1}, respectively.

^{15}N application. On 6 May 1987, ^{15}N-enriched red clover shoots (5.5 % a. e., C:N = 15:1) and ammonium sulphate fertilizer (10 % a.e.) were applied to microplots in Entry Point 1 subplots of the LIP-CG and CONV cropping systems, respectively. Legume N was applied at a rate equivalent to 165 kg ha^{-1} and fertilizer N was applied at a rate of 124 kg N ha^{-1}.

Microplots consisted of 61 cm-diameter by 45 cm-deep undisturbed soil columns enclosed by open-ended sheet metal cylinders that extended 5 cm above the soil surface. The cylinders were installed in four of the eight replications of the long-term experiment using a gas-powered post-hole digger as described by Harris and Hesterman (1990). Soil in microplots receiving ^{15}N red clover was excavated to 15 cm, existing clover roots were removed from the soil by hand, and labeled red clover material was mixed into the soil and returned to the microplot. To produce the labeled legume material used in this experiment, medium red clover was grown in a sand-filled bench in the greenhouse and fertilized weekly with a nutrient solution

Table 1.1. Cropping systems and rotation entry points for the low-input/cash grain (LIP-CG) and conventional (CONV) cropping systems of the long-term Rodale experiment used in this experiment.

Cropping System	Rotation Entry Point	1981	1982	1983	1984	1985	1986	1987	1988
		Crop							
LIP-CG	1	Oat Red Clover	Corn	Oat Red Clover	Corn	Soybean	Oat Red Clover	Corn	-
	2	Soybean	Oat Red Clover	Corn	Wheat Hairy Vetch	Corn	Barley Soybean Wheat	Wheat Red Clover	Corn
CONV	1	Corn	Corn	Soybean	Corn	Soybean	Corn	Corn	-
	2	Soybean	Corn	Corn	Soybean	Corn	Soybean	Corn	Corn

containing $(^{15}NH_4)_2SO_4$ (10 % a.e.). Shoot material was clipped from the bench, cut to 7 cm lengths with scissors, mixed thoroughly, then dried at 60 °C for 4 d before application to microplots. Soil in microplots of the CONV system was also excavated to a 15 cm depth, mixed, then returned to simulate spring plowing. Ammonium sulphate fertilizer was then applied in granular form directly to the microplots and incorporated into the top 5 cm of soil using a small hand cultivator.

Plant and soil analyses, Year 1. Six corn seeds were planted in each microplot on 6 May 1987 after ^{15}N application. Soon after emergence plants were thinned to three per microplot. The plants so thinned were left on the surface of the microplot. The remaining three corn plants were grown to physiological maturity and harvested on 5 Oct 1987. Corn grain, stover and roots were separated then weighed, ground and analyzed for total N and ^{15}N on a Europa Scientific Tracermass mass spectrometer after conversion of sample N to N_2 by Dumas combustion in a Roboprep CN analyzer (Europa Scientific LTd., Crewe, England)(Preston and Owens, 1983). Roots were handpicked from the top 15 cm soil layer only, and washed with tap water before drying.

After corn harvest, entire soil layers from 0-15, 15-30 and 30-45 cm were excavated, weighed and then mixed thoroughly before sampling. A subsample from each layer was dried and analyzed for total N, inorganic N, and ^{15}N in each fraction. Soil total N and ^{15}N were measured by the same method used for corn samples. Soil inorganic N ($NO_3^- + NH_4^+$) in filtered KCl extracts (100 mL 2 M KCl: 20 g dry soil, shaken 1 h) was measured colorimetrically on a Lachat flow-injector analyzer using Lachat QuikChem Method no. 12-107-04-1-A. Soil inorganic ^{15}N in KCl extracts was

released as NH_3 following reduction of NO_3-N with Devarda's alloy and addition of MgO. The released NH_3 was trapped on acidified filter disks (Whatman no. 3, 6 mm dia) as NH_4-N (Brooks et al., 1989). The $^{15}NH_4$-N was analyzed on the Europa Scientific Tracermass after combustion of the filter disks in the Roboprep analyzer.

Microbial biomass C and N in field-moist sample from the 0- to 15-cm soil layer of each microplot were determined using the chloroform fumigation incubation method (Jenkinson and Powlson, 1976). Soils were not reinoculated after fumigation; biomass C = C_f/K_c, where C_f=CO_2-C evolved from the fumigated sample, and K_c=0.41; and biomass N = N_f/K_n, where N_f = NH_4-N released during incubation and K_n = (-0.014 x C_f/N_f) + 0.39) (Voroney and Paul, 1984). Carbon respired from unfumigated control soils during the 10 to 20 day period was used as a measure of microbial activity and to calculate specific respiratory activity (CO_2-C respired divided by microbial biomass C) (Schnurer et al., 1985). Similarly, N mineralization during the 0 to 20 day period in unfumigated control soils was used as a measure of N availability and to calculate a specific mineralization activity (N mineralized divided by microbial biomass N). Soil excavated from the microplots after corn harvest was returned by layer after sampling. ^{15}N-labeled corn stover and roots, however, were not returned to the microplots.

Plant and soil analyses, Year 2. Spring barley was planted in microplots of both the LIP-CG and CONV systems on 29 March 1988 to measure recovery of residual legume and fertilizer ^{15}N by a second non-legume crop. Seeding rate was 108 kg ha^{-1} and no additional N was applied to either system. Spring barley was planted in the CONV system microplots, even though the surrounding subplot was

planted to soybean (according to the original cropping sequence), in order to a have a common second crop in both systems for comparative purposes. The mature barley plants were harvested on 24 July 1988, separated into grain, straw and roots and analyzed for total N and ^{15}N according to the same procedures used for corn. Soil was sampled after barley harvest and analyzed for the same parameters using the same methods as sampling after corn harvest.

The second 2-yr cycle (Entry Point 2). To account for year-to-year variability, the same experiment as described above was repeated starting in May 1988, using Entry Point 2 subplots of the LIP-CG and CONV systems. This provided a second 2-year cycle with corn in 1988 and spring barley in 1989. All N application, planting, harvesting, sampling and laboratory procedures were identical to those used for the 1987-1988 cycle (Entry Point 1), with the exception that the ^{15}N red clover plant material and ammonium sulphate fertilizer were both applied at a rate of 124 kg N ha^{-1}. We applied ^{15}N and planted corn on 3 May 1988, harvested corn and sampled soil on 14 Oct 1988, planted barley on 10 April 1989, and harvested barley and sampled soil on 26 July 1989.

^{15}N calculations and experimental design. Recovery of ^{15}N from red clover and ammonium sulphate by first-year corn, second-year barley and soil after each crop was calculated using the same equations used by Harris and Hesterman (1990). Loss of ^{15}N was estimated at two points during the cropping sequence of each cropping system (after corn harvest and after barley harvest) by subtraction, i.e. applied N not accounted for in crops and soil. All data were analyzed by 2-way analysis of variance for a randomized complete block with four replications.

RESULTS

Crop Dry Matter Yields and Total N Uptake

Corn (grain and whole plant) dry matter (DM) yield and total N uptake were significantly higher in the CONV than in the LIP-CG system microplots in 1987, but were not significantly different between the LIP-CG and CONV systems in 1988 (Table 1.2). Very low corn yields in two replications of the CONV system, where crop rooting depth is known to be shallow, may have attributed to the high coefficients of variation and lack of significance differences in 1988.

No significant differences were measured between the fertilizer- and legume-based systems for yields or total N uptake by second-year spring barley in 1988. Spring barley yields and total N uptake were significantly higher in the LIP-CG than in the CONV system in 1989.

Fate of Legume and Fertilizer ^{15}N

Entry Point 1 (1987 Corn/1988 Barley). Corn recovered 15 % of the red clover N applied to the LIP-CG and 49 % of the ammonium sulphate N applied to the CONV system (Table 1.3). Over three times more legume N than fertilizer N was recovered in soil sampled after corn harvest. Loss of applied N from the legume- and fertilizer-based systems was not significantly different, and based on application rates was equivalent to 34 and 38 kg N ha^{-1} for red clover and ammonium sulphate, respectively.

[This page left blank intentionally.]

Table 1.2. Crop grain and whole plant dry matter (DM) yields and total N uptake (TNUP) in microplots of the low-input/cash grain (LIP-CG) and conventional (CONV) cropping systems of the long-term Rodale Experiment.

	Year 1 (Corn)			Year 2 (Barley)		
	Grain DM Yield	Whole Plant DM Yield	Whole Plant TNUP	Grain DM Yield	Whole Plant DM Yield	Whole Plant TNUP
	g plot^{-1}					
Entry Point 1†						
LIP-CG	298	715	4.5	107	237	2.1
CONV	388	868	7.2	104	244	2.1
Significance	*	*	*	NS	NS	NS
CV (%)	8	6	16	24	28	24
Entry Point 2‡						
LIP-CG	241	534	4.9	32	89	1.2
CONV	173	401	4.0	10	69	0.8
Significance	NS	NS	NS	*	**	**
CV (%)	40	29	35	33	5	6

*, **, *** Significant at the 0.05, 0.01 and 0.001 probability levels, respectively; NS = not significant at P = 0.05.
†Year 1 = 1987; Year 2 = 1988.
‡Year 1 = 1988; Year 2 = 1989.

Table 1.3. Fate of red clover ^{15}N in the low input /cash grain (LIP-CG) and fate of ammonium sulphate ^{15}N in the conventional (CONV) cropping system of the long-term Rodale experiment.

	Year 1			Year 2			Total (Year 1 + 2)		
Cropping System	Corn	Soil	Loss	Barley	Soil	Loss	Crops	Soil	Loss
	% of input								
Entry Point 1†									
LIP-CG	14.7	72.1	13.2	2.7	57.1	12.2	17.4	57.1	25.4
CONV	49.3	20.5	30.2	1.3	19.2	0.0	50.6	19.2	30.4
Significance	***	***	NS	NS	***	NS	**	***	NS
CV (%)	11	13	41	22	6	6	11	6	9
Entry Point 2‡									
LIP-CG	16.4	60.2	23.4	1.1	37.4	21.7	17.5	37.4	45.1
CONV	29.4	24.7	45.9	0.6	15.1	9.0	30.0	15.1	54.9
Significance	NS	*	**	*	***	NS	NS	***	NS
CV (%)	38	7	14	21	4	55	36	4	17

*, **, *** Significant at the 0.05, 0.01 and 0.001 levels, respectively; NS = not significant at P = 0.05.
†Year 1 = 1987; Year 2 = 1988.
‡Year 1 = 1988; Year 2 = 1989.

The second-year spring barley crop recovered small amounts of residual legume and fertilizer ^{15}N in 1988 (Table 1.3). Since there was more legume ^{15}N left in soil after corn harvest, however, the legume N contribution to the second crop was significantly higher than the fertilizer N contribution on a kg N ha^{-1} basis (4.4 vs. 1.6, respectively - data not shown).

More legume N than fertilizer N was left in soil after barley harvest in 1988 (57 vs. 19 % of input). However, the decline in the amount of fertilizer N left in soil between corn harvest in 1987 and barley harvest in 1988 was equal to the amount of residual fertilizer N taken up by the barley crop. This means there was no additional loss of fertilizer N from the CONV cropping system during this period. On the other hand, the decline in the amount of legume N in soil during this same period (Fall 1987 to Summer 1988) exceeded the amount taken up by the barley crop. Twelve percent of the legume N originally applied, or the equivalent of 20 kg N ha^{-1}, was lost from the LIP-CG between corn harvest in 1987 and barley harvest in 1988.

Totals after two growing seasons (corn in 1987 and barley in 1988) showed more fertilizer N than legume N recovered by crops (51 vs. 17 % of input), more legume N than fertilizer N remaining in soil (57 vs. 19 % of input) and similar amounts of N from each source lost from the cropping systems (25 vs. 30 % of input, or 41 vs. 37 kg ha-1, from legume vs. fertilizer, respectively.)

Entry Point 2 (1988 Corn/1989 Barley). Corn recovered 16 % of the legume N applied to the LIP-CG and 29 % of fertilizer N applied to the CONV system in 1988 (Table 1.3). Sixty percent of the legume N applied, or the equivalent of 74 kg N ha^{-1}, remained in soil of the LIP-CG system after corn harvest. This was

significantly higher than the amount of fertilizer N left in soil after corn harvest (25 % of input or 31 kg N ha^{-1}).

Loss of fertilizer ^{15}N from the conventional system during the 1988 corn growing season was significantly higher (over twice as high in terms of % of input) than loss of red clover ^{15}N from the LIP-CG system. Based on the application rate of 124 kg N ha^{-1}, the 23 and 46 % loss of legume and fertilizer N applied represent losses equivalent to 27 and 57 kg N ha^{-1}, respectively. Recovery of residual soil ^{15}N by a second-year barley crop grown in 1989 was significantly higher in the LIP-CG than in the CONV system, although values were small for both legume and fertilizer N sources (Table 1.3).

There was more red clover N than fertilizer N left in soil after barley harvest in 1989. Since the decline in ^{15}N in soil between corn harvest in 1988 and barley harvest in 1989 exceeded recovery of ^{15}N by barley in both systems, there was loss of both legume and fertilizer N during this period (equivalent to 28 and 12 kg N ha^{-1}, respectively). Expressed as a percentage of residual ^{15}N left in soil after corn harvest, the loss of both legume and fertilizer N during Year 2 was the same (36 %).

Totals after two growing seasons (corn in 1988 and barley in 1989) showed that the amounts of fertilizer N and legume N recovered by crops were not significantly different, more legume N than fertilizer N remained in soil, and similar amounts of N from each source were lost from the cropping systems (45 vs 55 % of input or 56 vs. 68 kg ha^{-1} from legume and fertilizer, respectively).

Recovery and Distribution of Legume and Fertilizer N in Soil

More legume N than fertilizer N was recovered in the inorganic, microbial biomass and non-biomass organic soil fractions at each soil sampling, except for the inorganic fraction after corn harvest in 1988 when no difference in recovery was detected (Table 1.4). Only small amounts of either legume N or fertilizer N were recovered in the inorganic fraction at each sampling (less than 5 % of input). Overall, an average of 16 % of the applied legume N was recovered in the microbial biomass compared to 4 % recovery of fertilizer N in this fraction. The non-biomass organic fraction contained the most N from the applied sources, averaging 38 % of legume N applied and 14 % of fertilizer N applied. The amounts of both legume and fertilizer N in each fraction declined between Year 1 and Year 2 for both entry points.

Although there was more legume N than fertilizer N recovered in each of the three soil fractions measured as shown in Table 1.4, distribution of total N recovered in each system among the three soil fractions was often similar (Table 1.5). For example, there was no significant difference in distribution of fertilizer N and legume N in soil after corn in 1987 (Entry Point 1) where an average of 7 % of the ^{15}N recovered was found in the inorganic fraction, 28 % in the microbial biomass and 65 % in the non-biomass organic. The distribution of ^{15}N in soil after corn in 1988 (Entry Point 2) showed a larger proportion of fertilizer N than legume N in the inorganic fraction, a greater proportion of legume N than fertilizer N in microbial biomass and no significant difference between sources for the proportion in the non-biomass organic fraction (Table 1.5). Although there were significant differences in

Table 1.4. Recovery in three soil fractions of ^{15}N applied to microplots in the long-term cropping systems experiment at Rodale.

Cropping System	Year 1 (Corn)			Year 2 (Barley)		
	Inorganic	Microbial Biomass	Non-biomass Organic	Inorganic	Microbial Biomass	Non-biomass Organic
	% of input					
Entry Point 1†						
LIP-CG	4.4	19.8	47.9	1.2	15.6	40.3
CONV	1.7	5.8	12.7	0.4	4.5	14.3
Significance	*	***	**	**	***	**
CV (%)	31	3	19	10	7	13
Entry Point 2‡						
LIP-CG	3.4	16.0	40.8	1.8	11.2	24.4
CONV	4.3	4.4	16.0	0.9	3.1	11.1
Significance	NS	**	*	**	***	*
CV (%)	75	18	18	10	3	36

*, **, *** Significant at the 0.05, 0.01 and 0.001 levels, respectively; NS = not significant at P = 0.05 level.
†Year 1 = 1987; Year 2 = 1988.
‡Year 1 = 1988; Year 2 = 1989.

Table 1.5. Distribution among three soil fractions of ^{15}N applied to microplots in long-term cropping systems experiment at Rodale. [Values are expressed as a percentage of total ^{15}N recovered in soil.]

Cropping System	Year 1 (Corn)			Year 2 (Barley)		
	Inorganic	Microbial Biomass	Non-biomass Organic	Inorganic	Microbial Biomass	Non-biomass Organic
	% of total ^{15}N recovered in soil					
Entry Point 1†						
LIP/CG	6.1	27.5	66.4	2.1	27.3	70.6
CONV	8.4	28.7	62.9	2.1	23.4	74.5
Significance	NS	NS	NS	NS	**	**
CV (%)	19	8	4	13	3	1
Entry Point 2‡						
LIP/CG	5.6	26.6	67.8	4.8	29.9	65.3
CONV	17.4	17.4	64.8	6.0	20.5	73.5
Significance	NS	**	NS	*	**	**
CV (%)	84	8	14	7	10	4

*, **, *** Significant at the 0.05, 0.01 and 0.001 levels, respectively; NS = not significant at P = 0.05 level.
†Year 1 = 1987; Year 2 = 1988.
‡Year 1 = 1988; Year 2 = 1989.

the distribution of legume and fertilizer ^{15}N into particular soil fractions measured after second-year barley, in general, the distribution was similar with 4 % in the inorganic fraction, 25 % in the microbial biomass, and 71 % in the non-biomass organic fraction.

Microbial Biomass Pool Sizes and Activity

Carbon mineralization (respiration) was greater in soils from the LIP-CG than from the CONV system for each sampling date (Table 1.6). The amounts of C respired from soil sampled after corn and after barley were fairly similar within each cropping system. Carbon respiration in both systems was at least twice as high, however, in soils from Entry Point 2 than in soils from Entry Point 1. The size of the microbial biomass C pool was significantly greater in the LIP-CG than CONV soils at each sampling. The biomass C : total soil C ratio was also greater for the LIP-CG than CONV system at each sampling. No difference was detected in specific respiration activity between systems at any soil sampling.

As with C, N mineralization and the size of the biomass N pool was greater in the LIP-CG than in the CONV systems at each sampling date (Table 1.7). However, the amount of N mineralized in soils from both systems and both entry points was lower after barley (Year 2) than after corn (Year 1). Specific N mineralization activity was greater in soil from the LIP-CG than from the CONV system sampled after corn in 1987 (Entry Point 1), but there was no significant difference between systems for the other three sampling times. The biomass N : soil N ratio was greater in soil from the legume- than fertilizer-based system at every sampling.

Table 1.6. Soil microbial biomass activity and pool size parameters in the low-input/cash grain (LIP-CG) and conventional (CONV) cropping systems in the long-term experiment at Rodale -- Carbon.

Cropping system	Year 1 (Corn)				Year 2 (Barley)			
	CO_2-C Respiration	Biomass C	Specific Respiration Activity†	Biomass C : Soil C Ratio	CO_2-C Respiration	Biomass C	Specific Respiration Activity	Biomass C : Soil C Ratio
	------- (ug g^{-1}) -------				-------- (ug g^{-1})--------			
Entry Point 1 ‡								
LIP-CG	81	929	0.089	0.042	66	777	0.087	0.035
CONV	34	658	0.050	0.033	45	574	0.080	0.029
Significance	*	*	NS	*	**	*	NS	NS
CV (%)	34	10	33	10	8	11	11	11
Entry Point 2 §								
LIP-CG	186	758	0.217	0.034	185	675	0.275	0.030
CONV	86	403	0.228	0.020	116	412	0.287	0.021
Significance	**	***	NS	***	**	**	NS	*
CV (%)	3	6	7	5	7	10	14	11

*, **, and *** Significant at the 0.05, 0.01 and 0.001 levels, respectively; NS = not significant at P = 0.05 level.
† CO_2-C respiration/Biomass C.
‡Year 1 = 1987; Year 2 = 1988.
§Year 1 = 1988; Year 2 = 1989.

Table 1.7. Soil microbial biomass activity and pool size parameters in the low-iput/cash grain (LIP-CG) and conventional (CONV) cropping systems in the long-term experiment at Rodale - Nitrogen.

Cropping system	Year 1 (Corn) N Mineralization	Biomass N	Specific Mineralization Activity	Biomass N : Soil N Ratio	Year 2 (Barley) N Mineralization	Biomass N	Specific Mineralization Activity	Biomass N : Soil N Ratio
	------- (ug g^{-1}) --------				-------- (ug g^{-1}) --------			
Entry Point 1 ‡								
LIP-CG	13.6	182	0.075	0.051	6.2	186	0.034	0.054
CONV	8.2	133	0.060	0.044	4.6	132	0.035	0.042
Significance	***	*	**	*	*	*	NS	*
CV (%)	7	8	5	4	11	9	9	10
Entry Point 2 §								
LIP-CG	11.6	186	0.063	0.052	4.6	188	0.025	0.057
CONV	5.4	86	0.064	0.030	2.6	103	0.026	0.035
Significance	*	**	NS	**	NS	**	NS	**
CV (%)	25	3	33	7	26	9	26	9

*, **, and *** Significant at the 0.05, 0.01 and 0.001 levels, respectively; NS = not significant at P = 0.05.
†N mineralization/Biomass N.
‡Year 1 = 1987; Year 2 = 1988.
§Year 1 = 1988; Year 2 = 1989.

DISCUSSION

Weather. Bi-monthly precipitation data for the Rodale site during the 1986-1990 growing seasons (1 Apr to 31 Oct) were reported by Peters et al. (1992). The weather pattern for each of the three years of our ^{15}N study was very different and likely affected the results. Rainfall in 1987 was close to average and evenly distributed in what was considered a "normal" year. In 1988, an extremely dry period from the first of June to mid-July caused 1988 to be known as a "drought" year. The dry period, however, was followed by twice the monthly average rainfall in the second half of July and normal precipitation for the remainder of the growing season. Above average rainfall was recorded during May, June and July of 1989, which was categorized as a "wet" year.

Crop yields and total N uptake. Corn yields in both cropping systems were negatively affected by the drought-conditions of 1988 (compared to 1987). However, the yield reduction in the CONV system was more severe than in the LIP-CG. This observation supports the claim by other researchers that although corn productivity may be higher in conventional than organic systems under favorable growing conditions, under adverse conditions, particularly droughts, the reverse occurs (Sahs and Lesoing, 1985; Bezdicek and Granastein, 1989; Lockertz et al., 1980; Rickerl and Smolik, 1990). Total N uptake by corn followed the same trend as yields, with greater N uptake in the CONV system in 1987 and greater N uptake in the LIP-CG

system in 1988.

Fate of Legume and Fertilizer ^{15}N

Twice as much fertilizer N was lost from the CONV system as legume N from the LIP-CG system during the corn growing season (Year 1) in both 1987 and 1988. Loss of both N sources was greater during the drought year of 1988 than during 1987. Loss of fertilizer N was especially high in 1988 where almost 50 percent of the applied N was lost. High N losses during a drought year might not seem possible. However, it is possible that both fertilizer and legume N not taken up by corn, stressed during the dry period of June and the first half of July, was susceptible to loss by denitrification or leaching during the wet period that followed in the second half of July and up until corn harvest.

The similar recovery of legume vs. fertilizer N by the second-year spring barley crops may be related to recovery and distribution of applied N sources among the three soil fractions measured. For example, larger amounts of legume N were retained in each soil fraction after corn harvest in 1988 but the distribution of recovered N into the different fractions was similar for both sources. In addition, over 90 % of the residual legume and fertilizer N was in organic forms. Therefore, mineralization of small amounts of the ^{15}N in the organic fractions may have led to the low recovery of both sources by barley, and the slightly higher recovery of legume N may be related to the larger pool sizes of residual legume ^{15}N.

The drought conditions of 1988 had little affect on distribution of legume N among soil fractions after corn harvest (compared to after corn in 1987). On the

other hand, more fertilizer N was distributed to the soil inorganic fraction and less was distributed to the microbial biomass in 1988 than in 1987. We thought this might result in more fertilizer uptake by barley in 1989. This did not occur, however, and may be due to loss and/or stabilization of the inorganic N into organic N pools over winter and prior to seeding barley. No soil samples were taken in Spring 1989 so this can not be confirmed.

Larger amounts of legume N than fertilizer N were lost between corn and barley harvest for both entry points. Whether most of the residual N from either source was lost during the overwinter period or during the barley growing season is not known. However, loss of residual legume N in 1988 (Entry Point 1) probably occurred before the drought period started in the beginning of June. Also, the higher loss of legume N in 1989 compared to 1988 was probably related to the above average rainfall that occurred for most of the barley growing season in 1989. Loss of residual fertilizer N in 1989 may have also been due to the precipitation patterns that year, or to the fact that more fertilizer N was left in the inorganic fraction after the drought year of 1988. Since the distribution of residual legume and fertilizer N in soil after corn was generally similar, the greater loss of legume N than fertilizer N during Year 2 may have been due to the larger pool size of residual legume N. The fact that the same proportion of residual legume and fertilizer N was lost in 1989 supports this theory.

Microbial Biomass Pool Sizes and Activity

Due to its critical role as source/sink and transformer of soil N, the size of the microbial biomass is thought to be a good indicator of soil quality. The size of the microbial biomass pool is also thought to be a better index for assessing treatment effects since it is more responsive to management practices such as tillage, crop rotation, and fertilization than total soil organic matter. In addition, the ratios of microbial biomass C : soil C and microbial biomass N :soil N may be even more dynamic than the amount of microbial biomass C or N alone (McGill et al., 1988). Microbial biomass activity is also sensitive to management effects and is usually measured by quantifying the amount of C mineralized (respired) from an unfumigated soil sample. However, relating C mineralization to the size of the microbial biomass C pool (calculating specific activity) is now thought to be a better or more effective way of characterizing microbial activity (Anderson and Domsch, 1986; Campbell et al., 1991).

Microbial biomass C and N pool sizes were significantly larger in the LIP-CG than in the CONV system of this study at every soil sampling. The microbial biomass C : soil C and microbial biomass N : soil N ratios were also always greater in the LIP-CG than in the CONV system. This indicates that the pool sizes were not only greater, but that a greater portion of the soil C and N was made up of biomass C and N in the LIP-CG system.

Based on C mineralization, soil microbial activity was greater in the LIP-CG system than in the CONV system. However, the specific respiratory activity was very similar between the LIP-CG and CONV systems. Since the biomass C pool was

significantly greater in the LIP-CG system but there was no difference between systems in specific respiratory activity, and since C mineralization is the product of biomass C times specific respiratory activity, the greater microbial activity in the LIP-CG based on C mineralization was really due to the larger pool size of microbial biomass C.

As with C, N mineralization in soil from the LIP-CG was also greater than from the CONV cropping system at each sampling. This is not surprising since the C and N processes are highly related. The specific N mineralization activity, like specific C mineralization activity, was also similar for both systems. This suggests that N mineralization -- or the N supplying power -- of soil in the LIP-CG system was greater than in the CONV system primarily due to the larger pool size of microbial biomass N.

SUMMARY

The long-term cropping systems experiment at the Rodale Institute Research Center has demonstrated that adequate plant nutrition and crop productivity can be achieved when replacing fertilizer N with legume N inputs. Our ^{15}N study revealed clues as to why this is possible and also revealed some important differences and similarities in the cycling of N in legume- and fertilizer-based cropping systems.

The lower recovery of applied legume N by a subsequent non-legume crop was compensated for by greater uptake of soil N. A larger and more active soil microbial biomass was measured in the legume-based system and is thought to be responsible for this compensation. Specific C and N mineralization activity (mineralization divided by microbial pool size) was actually similar between systems. This suggests that the greater microbial activity in the legume-based system as measured simply by C and N mineralization, was really due to the larger pool sizes of microbial biomass C and N.

More legume N than fertilizer N was retained in soil, but the distribution of the N sources among different soil fractions was similar. In addition, most of the residual N from both sources was recovered in organic forms. This may explain why only small amounts of legume and fertilizer N were available for uptake by a second crop.

Substantial amounts of both legume and fertilizer N were lost during the year

of application, although loss of fertilizer N was generally higher. More legume N than fertilizer N was lost the year after application and is thought to be related to the larger amount of legume N left in soil after Year 1. Similar amounts of legume and fertilizer N were lost over the course of two growing seasons. Methods to minimize loss of N from both legume and fertilizer sources therefore should be investigated.

Different precipitation patterns during the years of this study seemed to affect the fate and behavior of applied N sources. In particular, the drought conditions of 1988 caused corn yields in the conventional system to be drastically reduced and subsequently, almost half of the fertilizer N applied was lost. Recovery by corn and loss of legume N was affected less by the drought. Therefore, legume-based cropping systems may be more "drought-proof" not only in terms of crop yields but also N loss.

Finally, loss of applied N in this study was calculated by subtraction. Mechanisms of N loss, such as nitrate leaching or denitrification, for both the applied N sources and indigenous soil N should be studied at this site. Also, better estimates of C and N mineralization potential, and the role of microbial biomass in cycling of soil N in these systems using long-term soil incubations would be valuable.

REFERENCES

Allison, F.E. 1966. The fate of nitrogen applied to soils. Adv. Agron. 18:219-258.

Anderson, Traute-Heidi, and K.H. Domsch. 1986. Carbon assimilation and microbial activity in soil. Z. Pflanzenernaehr. Bodenk. 149:457-468.

Bezdicek, D.F, and D. Granastein. 1989. Crop rotation efficiencies and biological diversity in farming systems. Am. J. Alt. Agric. 4:111-119.

Bolton, H., Jr., L.F. Elliot, R.I. Papendick, and D.F. Bezdicek. 1985. Soil microbial biomass and selected soil enzyme activities: Effect of fertilization and cropping practices. Soil Biol. Biochem. 17:297-302.

Brooks, P.D., J.M. Stark, B.B. McInteer, and T. Preston. 1989. Diffusion method to prepare soil KCl extracts for automated N-15 analysis. Soil Sci. Soc. Am. J. 53:1707-1711.

Bruulsema, T.W. and R.B. Christie. 1987. Nitrogen contribution to succeeding corn from alfalfa and red clover. Agron. J. 79:96-100.

Campbell, C.A., V.O. Biederbeck, R.P. Zentner, and G.P. Lafond. 1991. Effect of crop rotations and cultural practices on soil organic matter, microbial biomass and respiration in a thin Black Chernozem. Can. J. Soil Sci. 71:363-376.

Doran, J.W., D.G. Fraser, M.N. Culik, and W.C. Liebhardt. 1987. Influence of alternative and conventional agricultural management on soil microbial processes and nitrogen availability. Am. J. Alt. Ag. 2:99-106.

Frye, W.W, W.G. Smith, and R.J. Williams. 1985. Economics of winter cover crops as a source of nitrogen for no-till corn. J. Soil Water Conserv. 40:246-258.

Harris, G.H. and O.B. Hesterman. 1990. Quantifying the nitrogen contribution from alfalfa to soil and two succeeding crops using nitrogen-15. Agron. J. 82:129-134.

Hauck, R.D. 1971. Quantitative estimates of nitrogen-cycle processes. p. 65-80. *In* Nitrogen-15 in soil-plant studies. Int. Atomic Energy Agency, Vienna.

Hauck, R.D. 1982. Nitrogen-isotope-ratio analysis. p. 735-779. *In* A.L. Page et al. (ed.) Methods of soil analysis. Part 2. 2nd ed. Agron. Monogr. 9. ASA and SSSA, Madison, WI.

Janzen, H.H., J.B. Bole, V.O. Biederbeck, and A.E. Slinkard. 1990. Fate of N applied as green manure or ammonium sulphate fertilizer to soil subsequently cropped with spring wheat in three sites in Western Canada. Can. J. Soil Sci. 70:313-323.

Jenkinson,D.S., and D.S. Powlson. 1976. The effects of biocidal treatments on metabolism in soil -- V. A method for measuring soil biomass. Soil Biol. Biochem. 8:209-213.

Keeney, D.R. 1982. Nitrogen management for maximum efficiency and minimum pollution. p. 605-649 *In* F.J. Stevenson (ed.) Nitrogen in agricultural soil. Agron. Monogr. 22. ASA, Madison, WI.

Kundler, P. 1970. Utilization, fixation, and loss of fertilizer nitrogen (Review of international results of the last 10 years of basic research). Albrecht-Thaer-Arch. 14:191-210.

Ladd, J.N., and M. Amato. 1986. The fate of nitrogen from legume and fertilizer sources in soils successively cropped with wheat under field conditions. Soil Biol. Biochem. 18:417-425

Ladd, J.N., M. Amato, R.B. Jackson, and J.H.A. Butler. 1983. Utilization by wheat crops of nitrogen from legume residues decomposing in the field. Soil Biol. Biochem. 15:231-238.

Ladd, J.N., J.M. Oades, and M. Amato. 1981. Distribution and recovery of nitrogen from legume residues decomposing in soil sown to wheat in the field. Soil Biol. Biochem. 13:251-256.

L'Annunziata, M.F., and J.O. Legg.(ed.). 1984. Isotopes and radiation in agricultural sciences. Academic Press, London.

Legg, J.O., and J.J. Meisinger. 1982. Soil nitrogen budgets. p. 503-557 *In* F.J. Stevenson (ed.) Nitrogen in agricultural soils. Agron. Monogr. 22.

Liebhardt, W.C., R.W. Andrews, M.N. Culik, R.R. Harwood, R.R. Janke, J.K. Radke, and S.L. Rieger-Schwartz. 1989. Crop production during conversion from conventional to low-input methods. Agron. J. 81:150-159.

Lockertz, W., G. Shearer, S. Sweeney, G. Kuepper, D. Wanner, and D.H. Kohl. 1980. Maize yields and soil nutrient levels with and without pesticides and standard commercial fertilizers. Agron J. 72:65-72.

McGill, W.B., J.E. Dormaar, and E. Reinl-Dwyer. 1988. New perspectives on soil organic matter quality, quantity and dynamics on the Canadian Prairies. *In* Land degradation and conservation tillage. Proc. 34th annual CSSS/AIC meeting, Calgary, AB.

Muller, M.M., and V. Sundman. 1988. The fate of nitrogen (^{15}N) released from different plant materials during decomposition under field conditions. Plant Soil 105:133-139.

National Academy of Sciences (NAS). 1989. Alternative agriculture. National Academy Press, Washington, D.C.

Papendick, R.I., L.F. Elliot, and J.F. Power. 1987. Alternative production systems to reduce nitrates in groundwater. Am. J. Alt. Agric. 2:19-24.

Peters, S., R. Janke, and M. Bohlke. 1992. Rodale's farming systems trial 1986-1990. Rodale Press. Emmaus, PA.

Peterson, T.A., and M.P. Russelle. 1991. Alfalfa and the nitrogen cycle in the corn belt. J. Soil Water. Conserv. 46:229-235.

Rickerl, D.H., and J.D. Smolik. 1990. Farming systems' influences on soil properties and crop yields. J Soil Water Cons. Jan-Feb: 121-125.

Rosswall, T., and K. Paustian. 1984. Cycling of nitrogen in modern agricultural systems. Plant Soil. 76:3-21.

Sahs, W.W. and G. Lesoing. 1985. Crop rotations and manure versus agricultural chemicals in dryland grain production. J. Soil Water Conserv. 40:511-516.

Schnurer, J., M. Clarholm, and T.Rosswall. 1985. Microbial biomass and activity in an agricultural soil with different organic matter contents. Soil Biol. Biochem. 17:611-618.

Shen, S.M., P.B.S. Hart, D.S. Powlson, and D.S. Jenkinson. 1989. The nitrogen cycle in the Broadbalk wheat experiment: ^{15}N-labelled fertilizer residues in the soil and in the soil microbial biomass. Soil Biol. Biochem. 21:529-533.

Smith, M.S., W.W. Frye, and J.J. Varco. 1987. Legume winter cover crops. Adv. Soil Sci. 7:95-139.

Smith, S.J., J.S. Schepers, and L.K. Porter. 1990. Assessing and managing agricultural nitrogen losses to the environment. Adv. Soil Sci. 14:1-43.

Stanford, G. 1973. Rationale for optimum nitrogen fertilization in corn production. J. Environ. Qual. 2:159-166.

Ta, T.C., and M.A. Faris. 1990. Availability of N from ^{15}N-labeled alfalfa residues to three succeeding barley crops under field conditions. Soil Biol. Biochem. 22:835-838.

United States Department Of Agriculture (USDA). 1980. Report and recommendations on organic farming. U.S. government printing office, Washington, DC.

Voroney, R.P., and E.A. Paul. 1984. Determination of K_C and K_N in situ for calibration of the chloroform fumigation-incubation method. Soil Biol. Biochem. 16:9-14.

Westcott, M.P., and D.S. Mikkelsen. 1987. Comparison of organic and inorganic nitrogen sources for rice. Agron J. 79:937-943.

Westerman, R.L., L.T. Kurtz, and R.D. Hauck. 1972. Recovery of ^{15}N-labeled fertilizers in field experiments. Soil Sci. Soc. Amer. Proc. 36:82-86.

CHAPTER TWO

THE SOIL NITROGEN SUPPLYING CAPACITY OF ANIMAL-, LEGUME-, AND FERTILIZER-BASED CROPPING SYSTEMS

ABSTRACT

Two long-term incubation experiments were conducted on soils sampled from different cropping systems of the long-term Rodale experiment located in eastcentral PA. The nitrogen (N) supplying capacity of soil from an animal-based system was greater than the N supplying capacity of soil from legume- and fertilizer-based systems, which were similar. Microbial biomass carbon (C) and N levels declined 50, 40 and 30 % during the incubation in soils from the animal-, legume- and fertilizer-based systems, respectively. Carbon mineralization was also measured and paralleled N mineralization at a near constant ratio of 14:1 throughout the incubation for all three soils. The traditional single exponential model overestimated the mineralization potential, N_o, and underestimated the rate constant, k, compared to a double exponential model. It was concluded that the single exponential model does not adequately describe the C and N mineralization processes in soil and should not be used. In addition, differences in the active soil C and N pools as estimated by the double exponential model were more sensitive to cropping system effects. Recently-added legume and fertilizer N inputs contributed only 10 % to total N mineralized

during an incubation, but were highly equilibrated with the microbial biomass and mineralizable N pools. Soil from the 15-45 cm layer of the profile contributed a significant amount of potentially available N, but below 45 cm, net immobilization occurred. Sampling time and previous crop also affected C and N mineralization.

INTRODUCTION

As predicted by Keeney (1982), accurate assessment of soil nitrogen (N) availability has become an important component of developing cost-effective, environmentally-sound agricultural systems. Efficient use of not only fertilizer N, but also organic N sources such as legume green manures and animal manures, depends on knowing the N supplying capacity of soil. Estimates of N mineralization potential using short-term incubations (7 to 14 d) are dependent on soil sampling and pretreatment conditions and do not necessarily reflect the potential, long-term N supplying capacities of soils (Stanford and Smith, 1972). Chemical methods of estimating soil organic N availability have also proven unreliable, because they fail to extract biologically meaningful fractions of soil N. Although more laborious and time-consuming, long-term aerobic incubations have proven to be an effective way to determine the N mineralization potential of soils (Keeney, 1982). Even though nitrogen cycling in soil is known to be tightly linked with C transformations and microbial biomass, these parameters are not always measured or reported for long-term incubation studies. Also, studies of N mineralization potential as a function of soil depth are needed to improve estimates of the N supplying capacity of soil throughout the entire root zone (Hadas et al., 1986).

Stanford and Smith (1972) are credited with developing the original long-term (30 wk) incubation procedure. The method involves mixing a dried and sieved soil

sample with sand, preleaching initial mineral N, then periodic leaching of accumulated mineral N with a weak salt and "minus-N" solution and suction to remove excess water. Bremner (1965), in a discussion of aerobic incubations as tests of soil N availability, questioned the efficacy of preleaching, the use of amendments to improve leachability and aeration, the adjustment of water content by suction, and the addition of nutrient solutions. In addition, filters used with the leaching procedure are subject to clogging in the later stages of the incubation, and ignoring the leaching of organic N can lead to errors in determining N mineralization potentials (Smith et al., 1980).

In their original paper, Stanford and Smith (1972) discouraged the use of continuous incubation of soil without leaching as a method of predicting long-term N mineralization capacities. Cumulative inhibitory effects, such as drops in pH and accumulation of unspecified toxins, were reported to adversely affect the N mineralization-time curves. In a later article, Stanford (1982) restated that the reliability of results from extended incubations in static, closed systems is "questionable". A year later, however, in a study comparing open (leaching) and closed (static) techniques, Maynard et al. (1983) reported that although the pattern of N release was slightly different, the final amount of mineralized N was independent of the method employed. Since this report, at least two other long-term incubation experiments using the static technique have been published, with seemingly normal N mineralization curves (Azam et al., 1989; Hadas et al., 1986).

A number of mathematical models can be used to describe cumulative net N mineralization as a function of incubation time (Juma et al., 1984; Deans et al.,

1986). Although a linear (zero order) relationship has been reported by a few researchers, N mineralization with time normally follows a curvilinear function and is best explained by first order exponential kinetics. According to the original procedure (Stanford and Smith, 1972), the cumulative amount of inorganic N mineralized (N_m) at time (t) during the incubation is used to estimate the potentially mineralizable N, N_o, and a rate constant, k, by the simple first order equation:

$$Nm = N_o(1-e^{-kt}) \qquad [1]$$

Since nonlinear fitting programs were not easily accessible in 1972, N_o was first approximated using a hyperbolic or double reciprocal of a hyperbolic equation followed by iterative plotting to estimate N_o and k. It was later discovered that estimating N_o and k by nonlinear least squares (NLLS) regression analysis was more precise, more accurate and less ambiguous than the linear approximation/iterative method (Smith et al., 1980).

The simple first order equation ([1]) mathematically defines N_o as a single, readily-mineralizable pool of organic N that is mineralized at a rate proportional to its pool size. The existence of N_o as a single, discrete, and homogenous organic N pool, however, is thought to be unlikely (Juma et al., 1984). This may explain why several researchers have reported that a double exponential model with two mineralizable N pools, each with its own rate constant, fits experimental data from long-term N mineralization studies better than the single exponential model (Boyle and Paul, 1989; Deans et al., 1986; Molina et al., 1980; Nordmeyer and Richter, 1985). The double exponential model can be written as:

$$Nm = N_a(1-e^{-k1t}) + N_r(1-e^{-k2t}) \qquad [2]$$

where N_a and N_r are more available and resistant fractions of N_o, respectively, and k1 and k2 are the corresponding first order rate constants. A simplified version of the double exponential model where the second term is reduced to the linear (zero order) expression, k2t, has also been used to describe C and N mineralization during long-term soil incubations (Bonde et al., 1988; Bonde and Rosswall, 1987; Hauot et al., 1989; Blet-Charaudeau et al., 1990). In the mixed order (first/zero) equation, only three, rather than four, parameters need to be estimated. However, if the Nr pool of the double exponential model is constrained to a measured pool size, such as half of the organic soil N, $N_{1/2}$, Nr can be expressed as $N_{1/2}$ - Na, and the double exponential model is also reduced to a three parameter model.

Estimates of N_o from long-term incubations of soils that received fertilizer, legume, and animal manure N sources in various combinations have been reported (Beauchamp et al., 1986; Bonde and Roswall, 1987; Bonde et al., 1988; Boyle and Paul, 1989; Campbell et al, 1991; El-Haris, 1983; Hadas et al., 1986). None of these studies, however, involved soils where the fertilizer, legume or animal manure was used as the sole source of N for cropping systems that are known to be comparable in crop productivity and not limited by available N. In addition, C mineralization and changes in microbial biomass C and N during the incubation were measured in only a few of these studies (Bonde et al., 1988; Boyle and Paul, 1989; Campbell et al., 1991). The objectives of this research were to compare C and N mineralization, as well as concomitant changes in microbial biomass C and N, during long-term incubations of soils from established fertilizer-, legume-, and animal-based cropping systems. Incubations were also conducted for ^{15}N-labeled soil and soil from

different depths of the fertilizer- and legume-based cropping systems to evaluate N mineralization derived from recently-added N sources and from lower depths in the soil profile, respectively.

MATERIALS AND METHODS

Two separate long-term laboratory incubation experiments were conducted with soils taken from a long-term cropping system experiment located at the Rodale Institute Research Center in southeastern Pennsylvania. The long-term experiment, described in detail by Liebhardt et al. (1989) and Peters et al. (1992), was established in 1981 to compare a low-input/cash grain (LIP-CG) system where legume green manure was the primary N source, and a low-input/animal (LIP-A) system where the primary N source was animal manure, to a conventional (CONV) corn-soybean rotation where N fertilizer and pesticides are used. Soil at the site was primarily a Comly silt loam (fine-loamy, mixed, mesic Typic Fragiudalf). By 1985, prior to when soils were sampled for the long-term incubation experiments reported in this study, N did not appear to limit crop productivity and corn yields were comparable for the three cropping systems (Liebhardt et al., 1989).

Experiment 1.

Soil was sampled from the LIP-CG, LIP-A and CONV main plots in Oct 1988. The previous crop in all three systems was corn (*Zea mays* L.). Five, five cm dia soil cores were taken to a 15 cm depth from four of the eight field replications. Soil was also sampled from microplots located in the LIP-CG and CONV systems which had received ^{15}N-labeled red clover and ammonium sulphate, respectively, in

May of 1988 (Chapter 1). The ^{15}N-labeled soil (0-15 cm layer) had been excavated from the 0.6 m dia microplots and mixed thoroughly before sampling. All samples were sieved (2 mm) and kept at field moisture in a 4 °C cooler until being weighed for incubation.

A small sample (approximately 20 g fresh weight) of each soil was dried for 24 hours at 105 °C to determine moisture content. A separate sample was dried at 60 °C for 48 hours, ground finely, and then analyzed for total N and ^{15}N on a Europa Tracermass Mass spectrometer. The ^{15}N content of the unlabeled soils provided background ^{15}N levels. Seven portions (20 g dry weight equivalent) of each field-moist soil were then weighed into separate 90 ml plastic specimen cups to measure the amount of mineral N present on day 0, 11, 21, 41, 71, 131 and 211 of the incubation. An equivalent sample of each soil was also weighed into seven glass erlenmeyer flasks to measure microbial biomass C and N by the chloroform fumigation incubation method (CFIM, Jenkinson and Powlson, 1976) on day 0, 10, 30, 60, 120 and 200. Soils were not reinoculated after fumigation, no C control was subtracted, $K_c = .41$ and $K_n = (-0.014 \times C_f/N_f) + 0.39$) (Voroney and Paul, 1984). All soils were brought to 55 % of water holding capacity and placed in a growth chamber at 25 °C and 80 % relative humidity.

At each sampling date, a set of soils in specimen cups was used to provide a 0 to 10 day unfumigated control for the microbial biomass C measurement (hence the 11 day difference in sampling dates for N mineralization and microbial biomass). For example, on day 30, a set of soils in glass flasks was removed from the growth chamber and fumigated with alcohol-free chloroform for 24 hours. Then, after

evacuation of the chloroform, the flasks were placed in air-tight glass canning jars (0.9 L) fitted with rubber septums and placed back in the growth chamber. At this time a set of soils in specimen cups was also placed in glass canning jars. Ten days later (day 41), all jars were sampled for CO_2 concentration using a 1 ml gas sample (sampled with a syringe) and a Varian Aerograph Model 920 gas chromatograph. Soil mineral N (NO_3^- + NH_4^+) for both the fumigated and unfumigated soils was then measured colorimetrically on a flow-injection analyzer after extraction with 100 mL 2 M KCl (shaken 1 hr) and filtering. The ^{15}N content of the mineral N (NO_3^- + NH_4^+) for the unfumigated samples was measured after diffusion (Brooks et al., 1989) by mass spectrometry. For the fumigated samples, NH_4^+only was analyzed for ^{15}N content.

Cumulative CO_2 was measured by placing the set of specimen cups to be sampled for mineral N on day 211 in canning jars and measuring CO_2-C as described above on day 11, 21, 41, 71, 131, 171, 198, and 211. All other soils were left uncapped (specimen cups) or unstoppered (glass flask) in the growth chamber except for when being measured for microbial biomass C. Soil moisture content of all samples was checked periodically and maintained at 55 % of water holding capacity.

Results were analyzed separately using a two-way ANOVA. Experimental design was a randomized complete block with four replications. If a significant treatment effect was observed for the unlabeled soils, the means were further separated by an LSD test using an 0.05 alpha level.

Experiment 2.

Soils were sampled from the LIP-CG and CONV systems main plots in May 1991. The previous crop was a spring barley (*Hordeum distichum* L.)/soybean (*Glycine max* Merr.) relay crop in the LIP-CG system and soybean in the CONV system. Three, five cm dia soil cores were taken from four replications in the vicinity of leaching lysimeters installed for another experiment. Soil was sampled to a depth of 105 cm and partitioned into layers of 0-15, 15-30, 30-45 and 45-105 cm. Pretreatment of soils and determination of moisture and total N content were the same as in Experiment 1.

Seven portions (20 g dry weight equivalent) of each field-moist soil were weighed into separate 90 ml plastic specimen cups to be sampled for mineral N on day 0, 15, 30, 60, 120 and 379. Six equivalent portions of soil from the 0-15 cm layer of both cropping systems were weighed into separate 125 ml glass erlenmeyer flasks for the determination of microbial biomass C and N by the CFIM on day 0, 15, 30, 60, 120, and 379. Microbial biomass C and N was measured for the 15-30, 30-45 and 45-105 cm soil layers of each cropping system on day 0, 60 and 120. All samples were placed in a growth chamber and kept at 55 % water holding capacity and 22 °C. A lid with a small hole was placed on all specimen cups and parafilm was placed over the flasks to reduce the rate of moisture loss.

Mineral N in fumigated and unfumigated soils was measured as in Experiment 1. Microbial biomass C and N was also measured as in Experiment 1, except CO_2 evolved was trapped in vials containing 1 mL of 1 M NaOH and was measured by titration. Also, the incubation time was 15 d and soils in specimen cups were not

used to measure 0 to 10 day control carbon. Instead, an additional set of soil was weighed into specimen cups and placed in canning jars with base traps to provide both 0-10 day unfumigated controls for microbial biomass C measurements and cumulative CO_2. Experimental design was a randomized complete block with four replications. Soil depths were analyzed separately using a 2-way ANOVA.

RESULTS AND DISCUSSION

Experiment 1

The initial combustible (total) N levels as measured on day zero were significantly higher for the low-input soils that received organic N inputs (legumes and animal manure) compared to the CONV soil that received inorganic fertilizer N (Table 2.1). Extractable (inorganic) N levels at the commencement of the experiment were not different between soils from the low-input and conventional systems for either the unlabeled or labeled soils.

Cumulative C and N mineralized in the unlabeled soils were significantly higher in the animal-based than in the legume- or fertilizer-based cropping systems at each sampling during the incubation (Table 2.2). Applications of organic materials (animal manure and sludge) have been shown to increase the C and N mineralization potential of soils (Bonde et al., 1988, Griffin and Laine, 1983). No significant difference in C mineralization was observed between the legume- and fertilizer-based systems. N mineralization, however, was significantly greater in the legume- compared to fertilizer-based system at every sampling except at 131 days. This result is similar to that reported by Bonde and Rosswall (1987), where more N was mineralized from a non-fertilized alfalfa ley compared to fertilized barley.

An average of 27 % of the N mineralized from the three cropping systems by the end of the incubation period was accumulated in the first three weeks. Stanford

Table 2.1. Initial combustible (total) and extractable (inorganic) soil† N levels for Experiment 1.

Cropping system	Combustible (total) N	Extractable (inorganic) N
	---------- $\mu g\ g^{-1}$ soil ----------	
Unlabeled (main plots)		
LIP-A	3650a‡	13.5a
LIP-CG	3340a	12.5a
CONV	2650b	17.3a
Labeled (microplots)		
LIP-CG	3410	16.4
CONV	2620	14.4
Significance	**	NS
CV (%)	5	58

*,**,*** Significant at the 0.05, 0.01, and 0.001 probability levels, respectively; NS = not significant at the 0.05 level.
†depth of sampling = 15 cm.
‡Values within a column followed by the same letter are not significantly different by an $LSD_{(0.05)}$ test.

Table 2.2. Cumulative C mineralization, N mineralization and the C mineralization : N mineralization ratio for unlabeled soil in Experiment 1.

Cropping System	Day 11	21	41	71	131	170	198	211
	Cumulative CO_2-C mineralized ($\mu g\ g^{-1}$ soil)							
LIP-A	237a†	500a	134a	1372a	1858a	2134a	2319a	2392a
LIP-CG	118b	244b	636b	745b	1090b	1309b	1419b	1488b
CONV	100b	200b	494b	594b	885b	1039b	1148b	1214b
	Cumulative N mineralized ($\mu g\ g^{-1}$ soil)							
LIP-A	16a	46a	72a	102a	133a	--	--	179a
LIP-CG	9b	31b	49b	57b	81b	--	--	104b
CONV	5c	23c	36c	46c	65b	--	--	89c
	C mineralization : N mineralization							
LIP-A	14.6a	10.8a	14.7a	13.4a	14.7a	--	--	13.7a
LIP-CG	14.9a	7.9a	13.0a	13.3a	13.5a	--	--	14.3a
CONV	18.6a	8.9a	13.9a	13.1a	13.5a	--	--	13.6a

†Values for within a column followed by the same letter are not significantly different by an LSD(0.05) test.

and Smith (1972) and Beauchamp et al. (1986) reported that 25 % of N_o was mineralized in the first two weeks of a long-term incubation. The ratio of C mineralized : N mineralized was fairly constant at 14:1 for all three cropping systems throughout the incubation. This is slightly higher than the ratio of 11:1 reported by Carter and Rennie (1982) and Robertson et al. (1988), but supports the suggestion by Robertson et al. (1988) that the labile soil N pool could be estimated by simply measuring mineralization of soil C and adjusting to a fixed C:N ratio.

Soil microbial biomass C and N levels at each sampling date during the incubation were significantly higher in the animal-based system than the legume- and fertilizer-based systems which were similar (Table 2.3). The microbial biomass C:N ratio, like the mineralized C:N ratio, was basically the same for all three cropping systems and constant throughout the incubation. Overall, the microbial biomass C:N ratio averaged about 5:1, which is lower than the 8:1 value reported for most soils.

The size of the microbial biomass C and N pools declined during the incubation for all three soils (Table 2.3). The observed pattern of initial rapid loss followed by a steady decline was similar to that reported by Bonde et al. (1988), Boyle and Paul (1989), Marumoto et al. (1982) and Robertson et al. (1988). Attempts to fit the decline of microbial biomass C and N to single or double exponential decay function models, as was done by Bonde et al. (1988) and Boyle and Paul (1989), however, were unsuccessful. This may have been due to having only six observations during the incubation for these parameters. The net decrease in soil microbial biomass C and N pools (final-initial) was greater for the LIP-A than for the LIP-CG and CONV systems (Table 2.4). The proportional decrease in microbial

Table 2.3. Microbial biomass C, N and the microbial biomass C:N ratio for unlabeled soils in Experiment 1.

Cropping System	Day 0	10	30	60	120	200
	Microbial Biomass Carbon ($\mu g\ g^{-1}$ soil)					
LIP-A	885a†	757a	629a	688a	543a	456a
LIP-CG	627b	490b	445b	445b	462b	396b
CONV	456c	365b	368b	382b	351b	324c
	Microbial Biomass N ($\mu g\ g^{-1}$ soil)					
LIP-A	195a	199a	148a	166a	114a	88a
LIP-CG	122b	105b	105b	99b	86b	72b
CONV	94b	88b	84b	78b	70c	64b
	Microbial biomass C : N					
LIP-A	4.6a	3.8a	4.3a	4.1a	4.8a	5.2a
LIP-CG	5.1a	4.6a	4.2a	4.5a	5.4a	5.5a
CONV	4.9a	4.2a	4.4a	4.9a	5.0a	5.1a

†Values for within a column followed by the same letter are not significantly different by an LSD(0.05) test.

Table 2.4. Microbial biomass C and N net decrease, proportional decrease, and contribution to net C and N mineralization for unlabeled soils in Experiment 1.

Cropping System	Net decrease† ($\mu g\ g^{-1}$)	Proportional decrease‡	Contribution to net mineralization§
	---------- Microbial Biomass Carbon ----------		
LIP-A	430a¶	0.52c	0.18a
LIP-CG	232b	0.63b	0.16ab
CONV	132c	0.72a	0.11b
	---------- Microbial Biomass Nitrogen ----------		
LIP-A	107a	0.46b	0.60a
LIP-CG	50b	0.59a	0.48ab
CONV	30b	0.68a	0.34b

†final-initial
‡final/initial
§(final-initial)/cumulative mineralization at 200 d
¶Values within a column followed by the same letter are not significantly different by an LSD(0.05) test.

biomass C and N pools (final/initial) was also greater for the animal-based than for the legume- and fertilizer-based systems. A little over 50 % of the soil microbial biomass C and N was mineralized over the course of the incubation for the LIP-A system compared to 40 % for the LIP-CG and 30 % for the CONV. Boyle and Paul (1989) reported a 50 % decrease in microbial biomass C by the end of a 20 week incubation of sludge amended soils. This value compares well with the decline in microbial biomass C in the LIP-A soil in our study which also received large amounts of C input (in manure applications). Robertson et al. (1988) reported a 35 % decline in microbial biomass N during long-term incubation of a soil that had been cropped to N-fertilized cereals. This result is comparable to the observed decline in microbial biomass N in soil from the CONV system, which also received fertilizer N inputs. The contribution of microbial biomass C and N to mineralized C and N (final-initial/mineralized C or N at 200 day) in soils from the three cropping systems followed the same ranking as the decline in microbial biomass C and N. Between 11 and 18 % of the mineralized C was accounted for by the decline in biomass C and between 34 and 60 % of the mineralized N was from biomass N. Although low estimates of microbial biomass contribution to mineralized N during incubations have been reported (e.g. 15-25% : Juma and Paul, 1984), our results agree more with higher estimates such as reported by Boyle and Paul (1989), Marumoto et al. (1982), and Robertson et al. (1988).

Carbon and N mineralization data for the unlabeled soils were fitted to single exponential, double exponential, and mixed order (first/zero) models. The Nr parameter in the double exponential was defined as half the organic N (as reported in

Table 2.1) minus Na. Likewise, the Cr pool was defined as half the total C pool minus Ca. Justification for these definitions comes from extensive acid hydrolyses and carbon dating research which show that 50 % of the soil C is highly recalcitrant with a mean residence time of at least 500 years (Campbell et al., 1967; Martel and Paul, 1974). Nitrogen, being closely associated with C, is therefore thought to mineralize from the highly resistant pool at approximately the same rate (VanVeen et al., 1981). Although a good fit to the data was obtained by all three types of models (as indicated by r^2s greater than 0.900 - data not shown), the residual sum of squares for the double and mixed order models were consistently similar and less than the single exponential model. Since the double order model provides more information (Nr and k2 are separated) than the mixed order (Nr and k2 are lumped), C and N pool size and rate constant estimates generated from the single and double exponential models for all three soils were compared (Table 2.5). The most striking result from this curve fitting exercise is the difference between N_o and k of the single order model compared to Na and k2 of the double exponential model. The N mineralization potential (N_o) from the single exponential model has been considered the "active fraction" of soil organic matter. However, the active fraction as estimated by Na in the double exponential model was less than half of the value of N_o for all soils. This indicates that over half of the C or N mineralized in our 200 day incubation was contributed from a pool with a half life of around 13 years. Further, our results show that No was overestimated and K was underestimated when using the single instead of the double exponential model. This finding is the opposite to that reported by Deans et al. (1986). The difference between the models is depicted graphically for C and N

Table 2.5. Pool sizes and rate constants for the single and double exponential models used to describe the C and N mineralization for unlabeled soils in Experiment 1. [Pool sizes are in $\mu g/g$ soil and rate constants are in d^{-1}.]

Cropping system	Model					
	Single Exponential		Double exponential			
Nitrogen	$Nm = No(1-e^{-kt})$		$Nm = Na(1-e^{-k1t})+(Nr(1-e^{-k2t})$			
	No	K	Na	k1	Nr	k2
LIP-A	190	0.0109	70	0.0327	1747	0.000297
LIP-CG	109	0.0119	41	0.0377	1623	0.000185
CONV	103	0.0087	28	0.0350	1288	0.000228
Carbon	$Cm = Co(1-e^{-kt})$		$Cm = Ca(1-e^{-k1t}) + Cr(1-e^{-k2t})$			
	Co	K	Ca	k1	Cr	k2
LIP-A	2571	0.0110	1131	0.0249	9569	0.000664
LIP-CG	1770	0.0080	540	0.0240	10610	0.000439
CONV	1480	0.0075	386	0.0260	9514	0.000423

of the LIP-A soil in Figures 2.1 and 2.2, respectively. At first glance the models appear similar. However, the double exponential model seems to give a better fit than the single exponential model during the early part of the incubation. In addition, the linear trajectory near the end of the incubation (around 200 days) predicted by the double model, compared to the leveling off to an asymptote predicted by the single model, also fits the data better, makes more sense biologically, and illustrates an important difference between the models.

Both the N_o of the single model and Na of the double model ranked highest for the LIP-A, lowest for the CONV system, with the LIP-CG in between. The difference among systems was much more pronounced for Na than for N_o, however. This indicates Na as provided by the double model may be a more sensitive indicator of the active fraction than N_o. Carbon and N mineralization curves for the three cropping systems according to the double exponential model are compared in Figures 2.3 and 2.4, respectively.

Comparing the net decline in microbial biomass C and N (from Table 2.4) to the Ca and Na values of Table 2.5 reveals that about 50 % of the active C fraction was derived from biomass C whereas 100% of the active N fraction was derived from microbial biomass N (i.e the microbial biomass N pool was the active N fraction).

The ^{15}N labeled soils were included in this experiment to compare how recent additions of legume and fertilizer N to the LIP-CG and CONV systems mineralize in compared to native soil N. Mineralization of labeled N (lower two curves) accounted for about 10 % of the total N (labeled + unlabeled) mineralized for both systems (Figure 2.5). The mineralization is clearly dominated by N from older pools. These

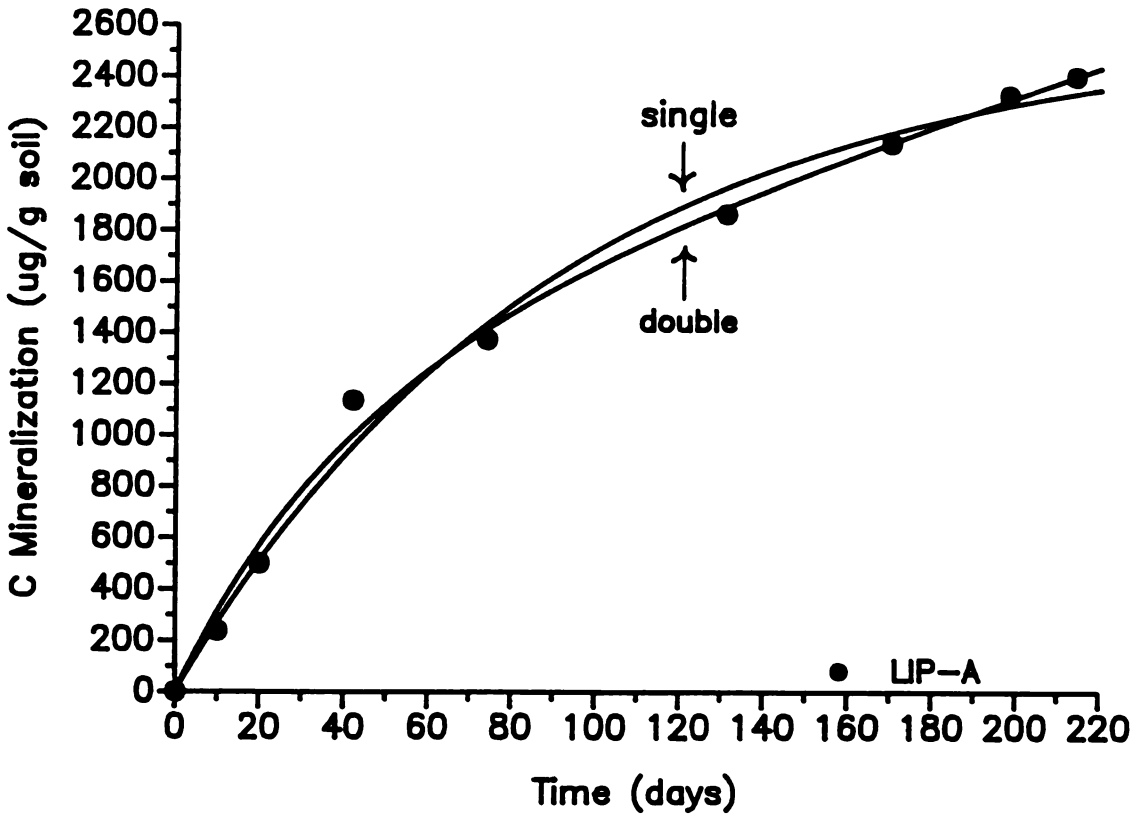

Figure 2.1. Carbon mineralization in soil from the low-input/with animal (LIP-A) cropping systems treatment of Experiment 1 as modeled by single and double exponential models. [See Table 2.5 for equations; points on the graph are means from 4 replications.]

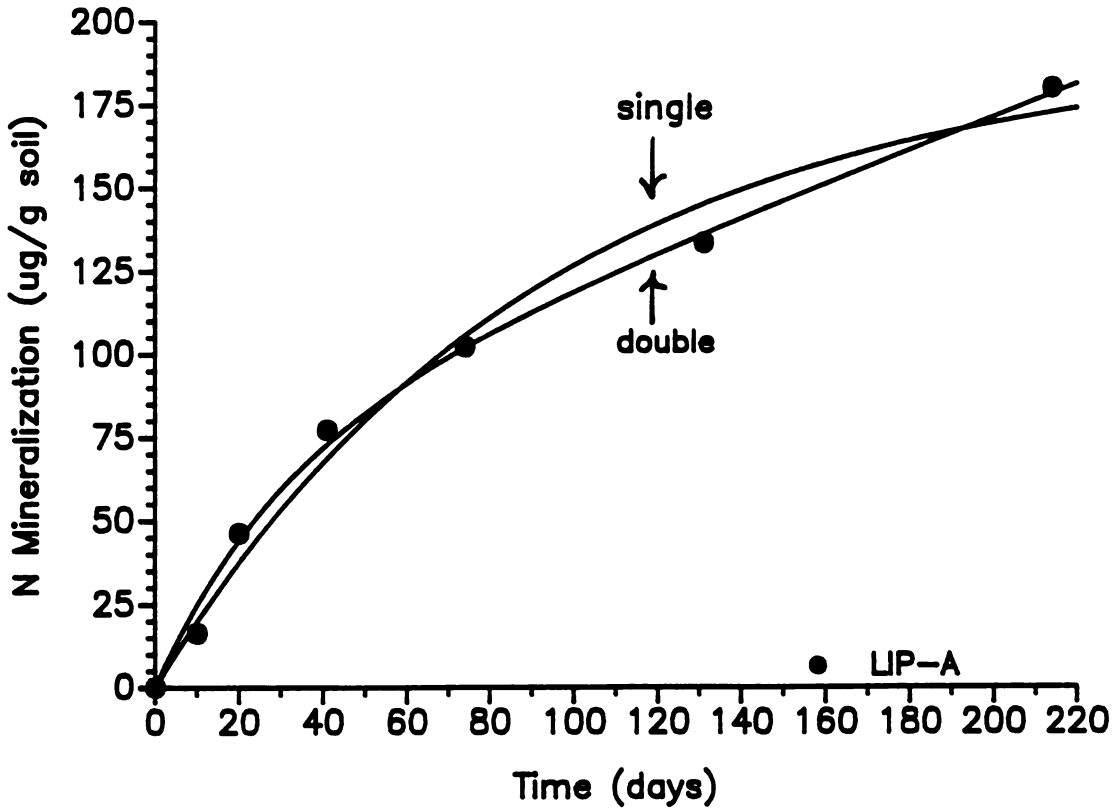

Figure 2.2. Nitrogen mineralization in soil from the low-input/with animal (LIP-A) cropping systems treatment of Experiment 1 as modeled by single and double exponential models. [See Table 2.5 for equations; points on the graph are means from 4 replications.]

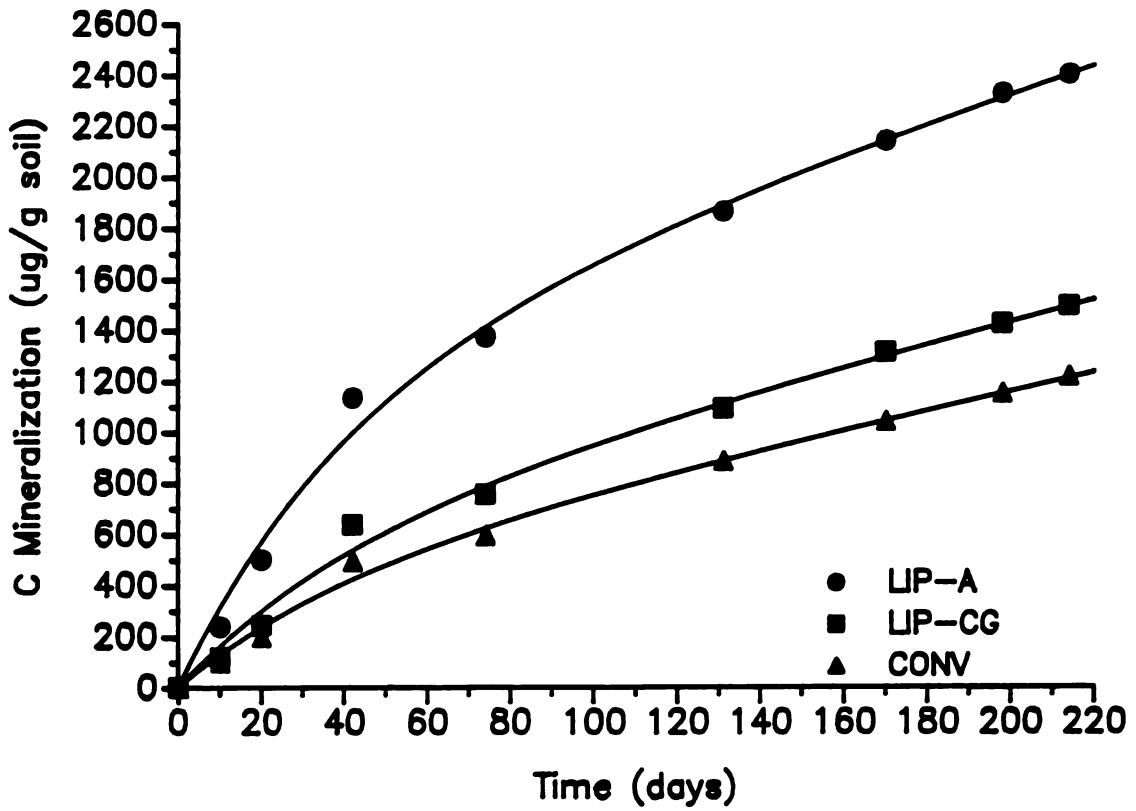

Figure 2.3. Carbon mineralization in soil from the low-input/with animal (LIP-A), low-input/cash grain (LIP-CG), and conventional (CONV) cropping systems treatments in Experiment 1 as described by the double exponential model. [See Table 2.5 for equations; points on the graph are means from 4 replications.]

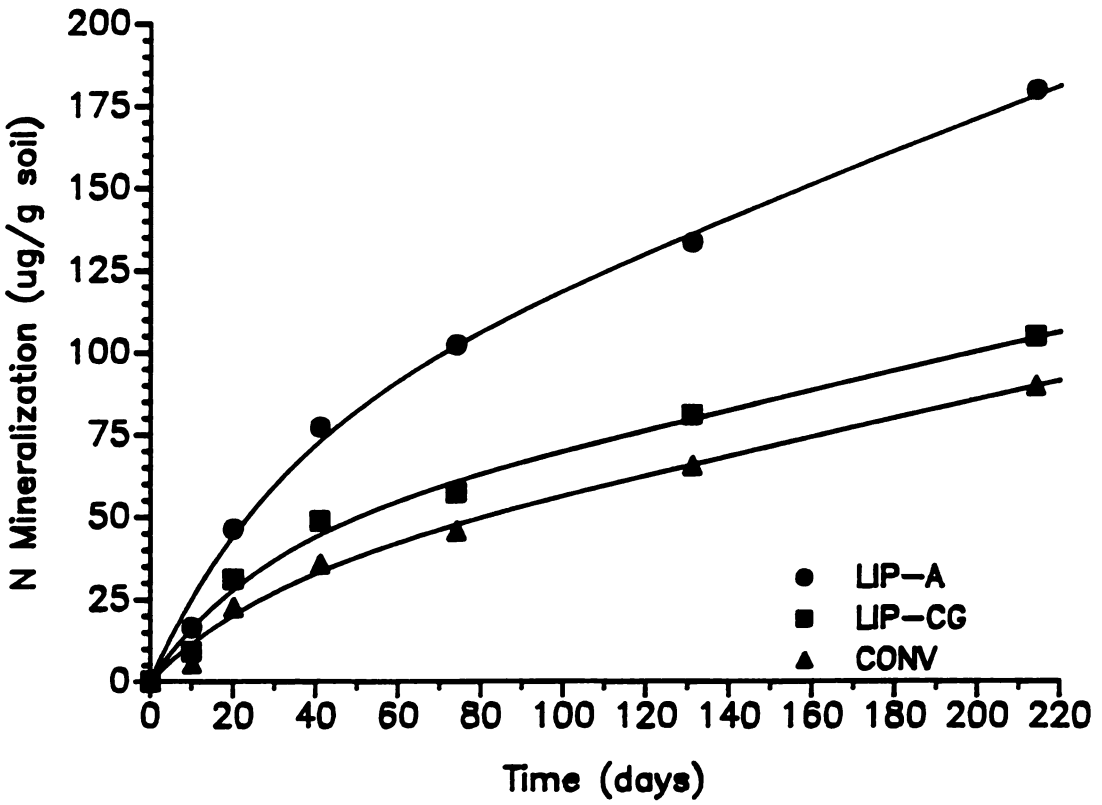

Figure 2.4. Nitrogen mineralization in soil from the low-input/with animal (LIP-A), low-input/cash grain (LIP-CG), and conventional (CONV) cropping systems treatments in Experiment 1 as described by the double exponential model. [See Table 2.5 for equations; points on the graph are means from 4 replications.]

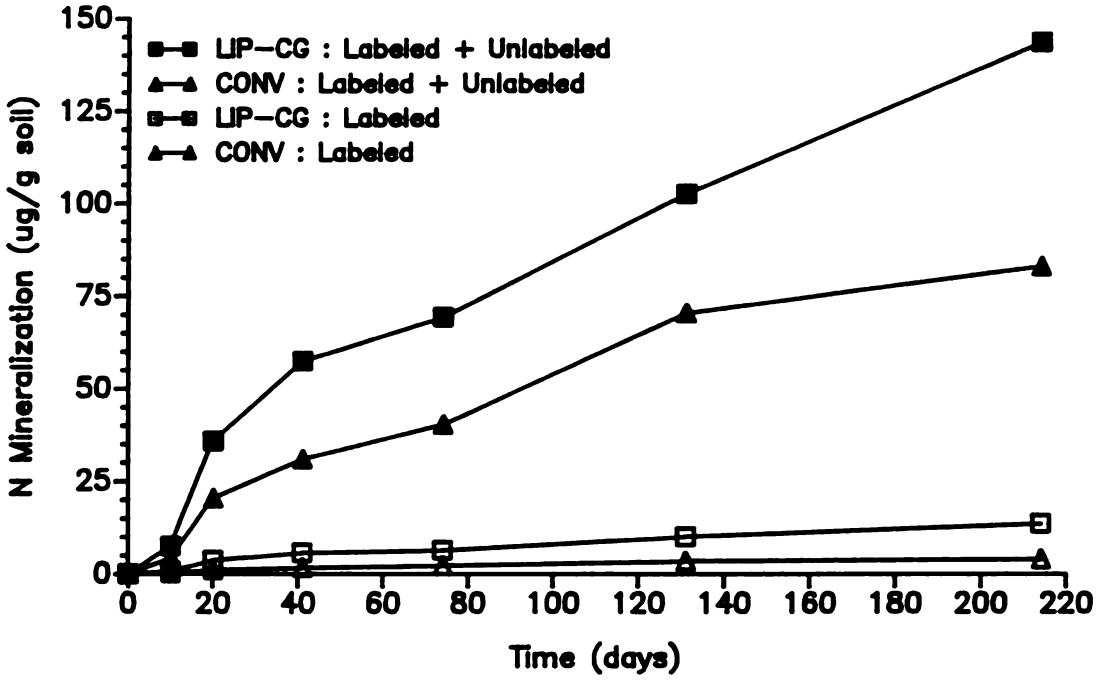

Figure 2.5. Mineralization of recently-applied red clover and ammonium sulphate ^{15}N in soils from the low-input/cash grain (LIP-CG) and conventional (CONV) cropping systems treatments in Experiment 1 (bottom curves) compared to mineralization of total (labeled and unlabeled) N (top curves).

curves were not fit to any models and the points on the graph are means for 4 observations (replications).

The ^{15}N contents of mineralized N and microbial biomass N for soil from the LIP-CG system were both similar and remained constant (i.e. did not decline) during the incubation (Figure 2.6). The same observation was made for the CONV system in replications (reps) 1 and 2 but not reps 3 and 4 (the data were presented in this manner to illustrate this result). A large amount of fertilizer N (^{15}N) was left in soils from microplots of reps 3 and 4 due to a drought period prior to sampling in Summer 1988. As a result, the mineralized N early in the incubation of the CONV soil consisted of this residual fertilizer ^{15}N and declined during the incubation. The ^{15}N level however did not decline to the same level of the microbial biomass N indicating that these two pools were still not in equilibrium by the end of the incubation. The lack of a decline in the ^{15}N enrichment for all other curves (mineralized N and biomass N of LIP-CG and biomass N of CONV) is hard to explain. The ^{15}N levels should have declined steadily during the incubation if a pool of unlabeled N was being mineralized. The constant level observed leads one to believe the recently added ^{15}N equilibrated with the more resistant native organic N pool. There are some reasons to believe this is possible. For one, the legume and fertilizer ^{15}N had actually been in the soil in the field for 6 months before the incubation. By this time 26 and 17 % of the residual legume and fertilizer ^{15}N, respectively, was in the microbial biomass and at least another 60 % was in the non-biomass organic pool (Chapter 1). Another explanation resides in our incubation methodology. Since the soils were not leached during the incubation, a large ^{15}N signal from early in the incubation may not have

Table 2.6. Initial combustible (total) and extractable (inorganic) soil† N levels for Experiment 2.

Cropping system	Combustible (total) N	Extractable (inorganic) N
	---------- $\mu g\ g^{-1}$ soil ----------	
	0-15 cm	
LIP-CG	3200	35.9
CONV	3040	19.4
Significance	NS	NS
CV(%)	7	77
	15-30 cm	
LIP-CG	2510	13.6
CONV	2370	11.6
Significance	NS	NS
CV (%)	10	21
	30-45 cm	
LIP-CG	1860	10.2
CONV	1980	8.6
Significance	NS	NS
CV(%)	6	19
	45-105 cm	
LIP-CG	2107	7.6
CONV	2338	8.4
Significance	NS	NS
CV(%)	28	14

NS = not significant at the 0.05 level of probability.
†depth of sampling = 15 cm.

been diluted by the smaller amounts mineralized later in the incubation. A third explanation is that denitrification of accumulated NO_3-N led to an increase in ^{15}N enrichment and masked the effect of mineralized ^{14}N. In any case, the strong similarity between ^{15}N enrichment of the microbial biomass N and mineralized N argues for a total turnover of a mature microbial population rather than an immature population which would not have come into equilibrium with the labeled N.

Experiment 2

The purpose of this experiment was to measure the mineralization potential of different soil depths in the LIP-CG and CONV cropping systems. Unlike in Experiment 1, the initial combustible (total) N pool sizes for the 0-15 cm soil layer were similar for the LIP-CG and CONV systems (Table 2.6). The combustible (total) N pool for the other layers and the extractable (inorganic) N pool size for all layers was similar for the two systems. Net immobilization (approximately 2 ug/g) was observed for the 45 -105 cm soil depth for both cropping systems (data not shown). Thus, no soil N from this lower layer would have been available for plant uptake. The mineralization curves for the other three layers indicate that little N was contributed from the 30-45 cm layer, but when combined with the 15-30 cm layer, these two layers combined contributed approximately 50 % of the total N mineralized from both systems (Figure 2.7). Unlike in Experiment 1, there was no significant difference in N mineralization for the 0-15 cm layer between the LIP-CG and CONV systems. Also, the amount of N mineralized from the 0-15 cm layer of both systems was greater than in Experiment 1. These results may have been due to sampling the

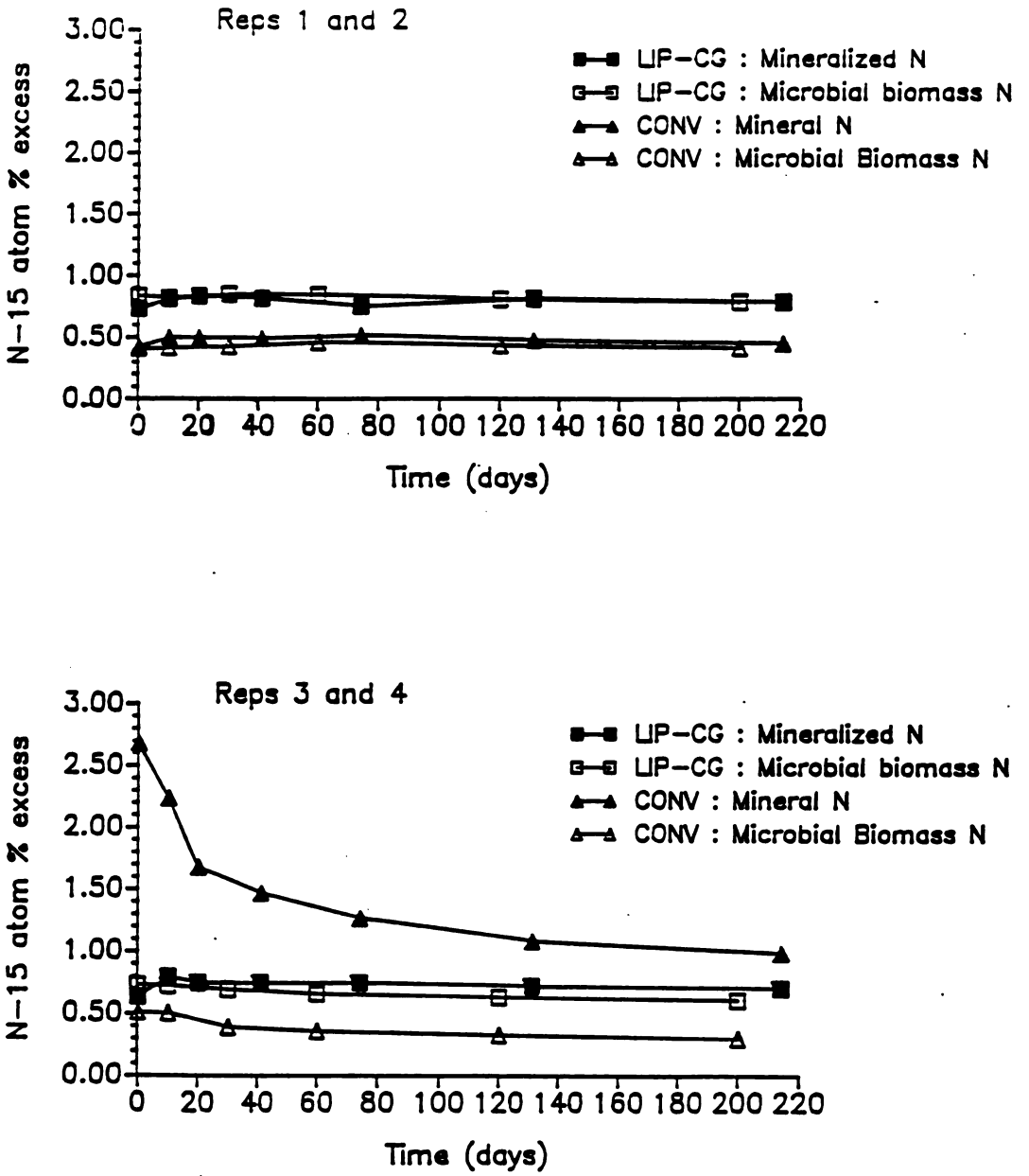

Figure 2.6. ^{15}N enrichment levels of mineralized and microbial biomass N in labeled soils from the low-input/cash grain (LIP-CG) and conventional (CONV) systems during the incubation in Experiment 1. [Results are shown for replications 1 and 2 vs. 3 and 4, separately.]

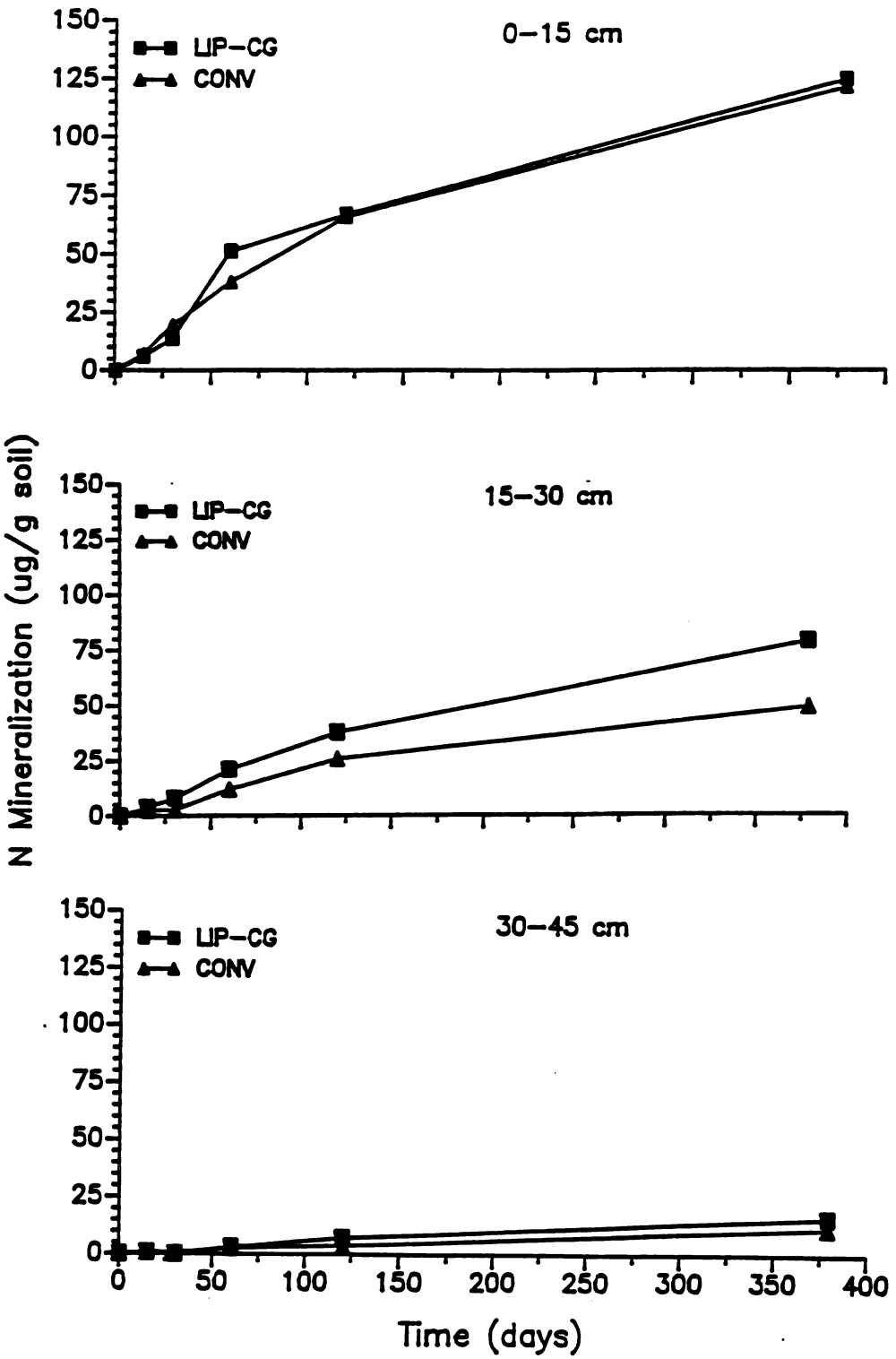

Figure 2.7. Nitrogen mineralization in soil from the 0-15, 15-30, and 30-45 cm depths of the low-input/cash grain (LIP-CG) and conventional (CONV) cropping systems treatments in Experiment 2.

soil in spring for this experiment compared to sampling in fall in Experiment 1. El-Haris et al. (1983) also observed greater N mineralization in spring vs. fall sampled soils. The difference may also have been due to sampling soil following soybean (Experiment 2) vs after corn (Experiment 1). Carbon mineralization with soil depth (Figure 2.8) also paralleled the N mineralization curves in this experiment (Experiment 2) illustrating that the constant C mineralized : N mineralized ratio holds even for different soil depths.

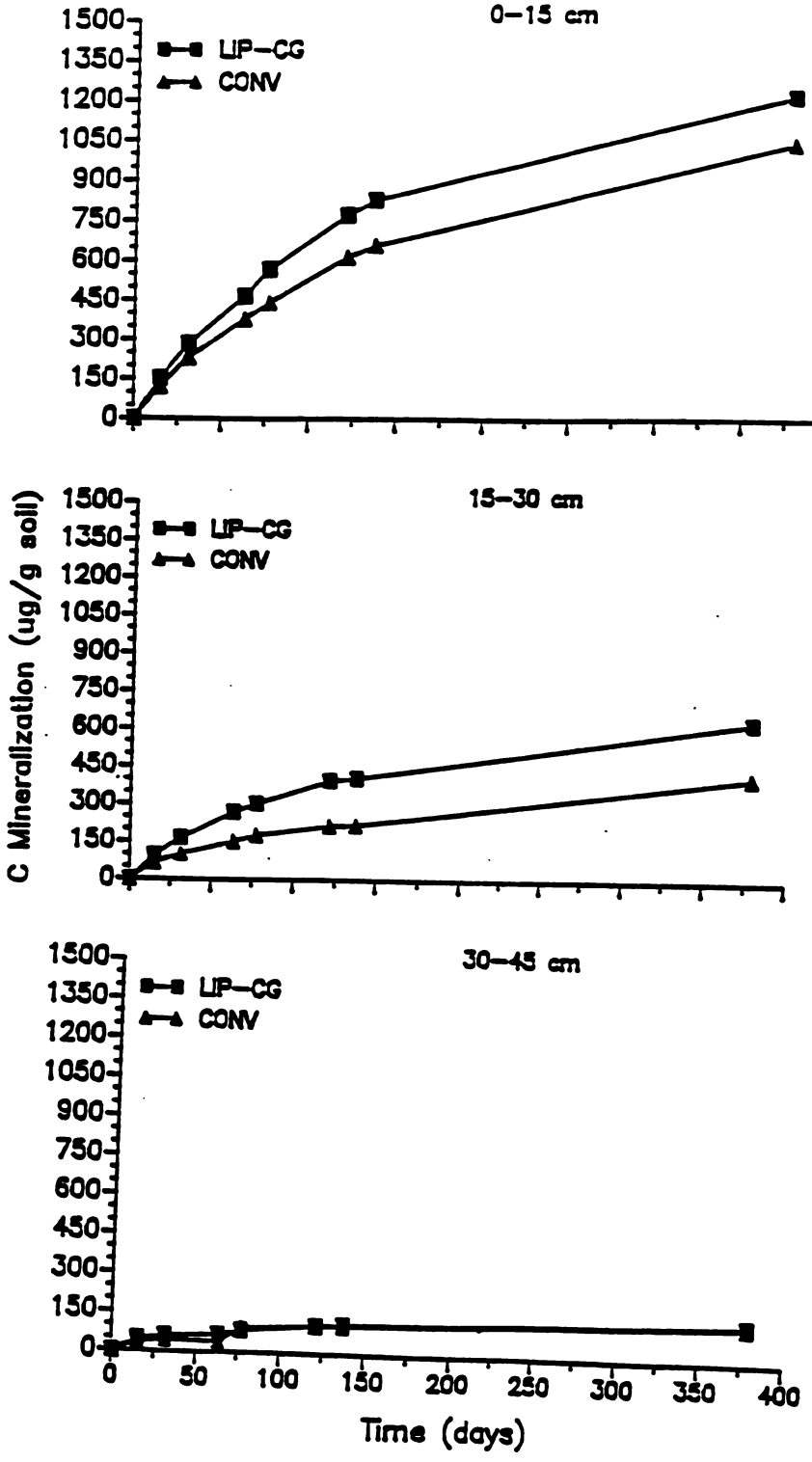

Figure 2.8. Carbon mineralization in soil from the 0-15, 15-30, and 30-45 cm depths of the low-input/cash grain (LIP-CG) and conventional (CONV) cropping systems treatments in Experiment 2.

CONCLUSIONS

The N supplying capacity of the animal-based cropping system in the long-term Rodale experiment was clearly greater than the N supplying capacity of the legume- and fertilizer-based systems as determined by long-term soil incubations in this study. Therefore, the animal-based system may have the greatest potential for N loss and environmental pollution. The similar N supplying capacity of the legume- and fertilizer-based systems indicates that both of these systems may be operating at optimum levels for maximum crop use efficiency and minimum N loss.

The closed system (static) incubation technique appeared to yield normal N mineralization curves similar to what is expected with the traditional leaching procedure. In addition, the highly correlated C mineralization curves indicate that it may be possible to predict N mineralization during incubations by simply measuring cumulative CO_2-C respired from soils.

Measuring the change in microbial biomass C and N pool sizes during the incubation revealed that the biomass was the primary source for greater C and N mineralization potential in the animal-based system compared to the legume- and fertilizer-based systems.

Curve fitting our C and N mineralization data showed that the traditional single exponential model grossly overestimated the N mineralization potential (No) and underestimated the rate constant (k) compared to the double exponential model.

It is our opinion, that because the single exponential model treats No as a single, discrete pool and fails to predict continued mineralization in later stages of the incubation, it does not make sense biologically and should not be used to predict the N mineralization potential of soils. The double exponential model better describes C and N mineralization during soil incubations and, in addition, it provides estimates active soil organic matter fractions (C and N) that appear sensitive to management practices. Our results also showed that the soil microbial biomass accounted for half of the active C fraction and all of the active N fraction in soil from all three (animal-, legume-, and fertilizer-based) cropping systems.

Recently-added legume and fertilizer N sources (after one corn growing season) contributed little to the soil N mineralization potential. This supports our previous finding that little residual N is made available for a second-year crop in the field (Chapter 1). The apparent equilibration of the residual fertilizer and legume ^{15}N with the microbial biomass indicated that significant internal cycling of the recent N inputs, and possibly a complete turnover of microbial biomass N, occurred during the incubation.

Incubation of soil from different depths in the soil profile showed that the contribution of lower depths to mineralizable N can be significant and should not be ignored. However, a depth existed below which net immobilization occurred and no available soil N was made available (that depth was 45 cm for our soils). Sampling time (e.g. fall vs. spring) and previous crop (i.e. corn vs. soybean) seemed to affect the C and N mineralization results of this study and should be considered when planning long-term soil incubations.

REFERENCES

Azam, F., R.L. Mulvaney, and F.J. Stevenson. 1989. Transformation of ^{15}N-labelled leguminous plant material in three contrasting soils. Biol. Fertl. Soils. 8:54-60.

Beuchamp, E.G., W.D. Reynolds, D. Brasche-Villeneuve, and K. Kirby. 1986. Nitrogen mineralization kinetics with different soil pretreatments and cropping histories. Soil Sci. Soc. Am. J. 50:1478-1483.

Blet-Charaudeau, C., J. Muller, and H. Laudelout. 1990. Kinetics of carbon dioxide evolution in relation to microbial biomass and temperature. Soil Sci. Soc. Am. J. 54:1324-1328.

Bonde, T.A., and T. Rosswall. 1987. Seasonal variation of potentially mineralizable nitrogen in four cropping systems. Soil Sci. Soc. Am. J. 51:1508-1514.

Bonde, T.A., J. Schnurer, and T. Rosswall. 1988. Microbial biomass as a fraction of potentially mineralizable nitrogen in soils from long-term experiments. Soil Biol. Biochem. 20:447-452.

Boyle, M., and E.A. Paul. 1989. Carbon and nitrogen mineralization kinetics in soil previously amended with sewage sludge. Soil Sci. Soc. Am. J. 53:99-103.

Bremner, J.M. 1965. Nitrogen availability indexes. p. 1324-1345 *In* C.A. Black (ed.) Methods of soil analysis, Part 2. Agron. Monogr. 9. ASA and SSSA, Madison, WI.

Brooks, P.D., J.M. Stark, B.B. McInteer, and T. Preston. 1989. Diffusion method to prepare soil extracts for automated nitrogen-15 analysis. Soil Sci. Soc. Am. J. 53:1707-1711.

Campbell, C.A., V.O. Biederbeck, R.P. Zentner, and G.P. Lafond. 1991. Effect of crop rotation and cultural practices on soil organic matter, microbial biomass and respiration in a thin Black Chernozem. Can. J. Soil Sci. 71:363-376.

Campbell, C.A., E.A. Paul, D.A. Rennie, and K.J. McCallum. 1967. Applicability of the carbon-dating method of analysis to soil humus studies. Soil Sci. 104:217-224.

Deans, J.R., J.A.E. Molina, and C.E. Clapp. 1986. Models for predicting potentially mineralizable nitrogen and decomposition rate constants. Soil Sci. Soc. Am. J. 50:323-326.

El-Haris, M.K., V.L. Cochran, L.F. Elliott, and D.F. Bezdicek. 1983. Effect of tillage, cropping, and fertilizer management on soil nitrogen mineralization potential. Soil Sci. Soc. Am. J. 47:1157-1161.

Griffin, G.F., and A.F. Laine. 1983. Nitrogen mineralization in soils previously amended with organic wastes. Agron. J. 75:124-129.

Hadas, A., S. Feiggenbaum, A. Feigin, and R. Portnoy. 1986. Nitrogen mineralization in profiles of differently managed soil types. Soil Sci. Soc. Am. J. 50:314-319.

Houot, S., J.A. E. Molina, R. Chaussod, and C.E. Clapp. 1989. Simulation by NCSOIL of net mineralization in soils from the Deherain and 36 Parcelles fields at Grignon. Soil Sci. Soc. Am. J. 53:451-455.

Juma, N.G., and E.A. Paul. 1984. Mineralizable soil nitrogen: amounts and extractability ratios. Soil Sci. Am. J. 48:76-80.

Juma, N.G., E.A. Paul, and B. Mary. 1984. Kinetic analysis of net nitrogen mineralization in soil. Soil Sci. Soc. Am. J. 48:753-757.

Keeney, D.R. 1982. Nitrogen -- availability indices. p. 711-733 *In* A.L. Page (ed.) Methods of soil analysis. Part 2. 2nd ed. Agron. Monogr. 9. ASA and SSSA, Madison, WI.

Liebhardt, W.C., R.W. Andrews, M.N. Culik, R.R. Harwood, R.R. Janke, J.K. Radke, and S.L. Reiger-Schwartz. 1989. Crop production during conversion from conventional to low-input methods. Agron. J. 81:150-159

Martel, Y.A., and E.A. Paul. 1974. Effects of cultivation on the organic matter grassland soils as determined by fractionation and radiocarbon dating. Can. J. Soil Sci. 54:419-426.

Marumoto, T, J.P.E. Anderson, and K.H. Domsch. 1982. Mineralization of nutrients from soil microbial biomass. Soil Biol. Biochem. 14:469-475.

Maynard, D.G., J.W.B. Stewart, and J.R. Brittany. 1983. Sulphur and nitrogen mineralization in soils compared using two incubation techniques. Soil Biol. Biochem. 15:251-256.

Nordmeyer, H., and J. Richter. 1985. Incubations experiments on nitrogen mineralization in loess and sandy soils. Plant Soil. 83:433-445.

Peters, S.E., R. Janke, and M. Bohlke. 1992. Rodale's farming systems trial 1986-1990. Rodale Institute, Rodale Press, Emmaus, PA.

Robertson, K., J. Schnurer, M. Chlarholm, T.A. Bonde, and T. Rosswall. 1988. Microbial biomass in relation to C and N mineralization during laboratory incubations. Soil Biol. Biochem. 20:281-286.

Smith, J.L., R.R. Schnabel, B.L. McNeal, and G.S. Campbell. 1980. Potential errors in the first-order model for estimating soil nitrogen mineralization potentials. Soil Sci. Soc. Am. J. 44:996-1000.

Stanford, G. 1982. Assessment of soil nitrogen availability. p. 651-688 *In* F.J. Stevenson (ed.) Nitrogen in agricultural soils. Agron. Monogr. 22. ASA and SSSA, Madison, WI.

Stanford, G. and S.J. Smith. 1972. Nitrogen mineralization potentials of soils. Soil Sci. Soc. Am. Proc. 36:465-472.

VanVeen, J.A., W.B. McGill, H.W. Hunt, M.J. Frissel, and C.V. Cole. 1981. Simulation models of the terrestrial nitrogen cycle. in F.E. Clark and T. Rosswall (eds.). Terrestrial nitrogen cycles. Ecol. Bull. (Stockholm) 33:25-48.

CHAPTER THREE

FERTILIZER AND LEGUME NITROGEN CONTRIBUTIONS TO CORN: COMPARISON OF ISOTOPIC AND NONISOTOPIC METHODS

ABSTRACT

Mineralization-immobilization turnover (MIT) is often cited as the cause of low apparent recovery of fertilizer and legume nitrogen (N) by crops when using ^{15}N methodology. The objectives of this research were to 1) develop and test a modified ^{15}N method of measuring legume and fertilizer N contribution to corn (*Zea mays* L.) that accounts for MIT effects, and 2) compare results to those obtained using traditional nonisotopic methods. Short-term (2-y) crop rotations were established in Spring 1990 at two Michigan locations: on a Capac Loam at the Michigan State University Agronomy Farm in East Lansing (EL) and on a Kalamazoo loam at the Kellogg Biological Station (KBS) in Hickory Corners. First-year crops included alfalfa (*Medicago sativa* L.), red clover (*Trifolium pratense* L), hairy vetch (*Vicia villosa* Roth.), and corn. A non-N_2-fixing alfalfa cultivar was also included to provide a control for N_2-fixation measurements and for estimating non-N rotation effects for the legumes. The modified ^{15}N method was conducted in 1991 by applying ^{15}N labeled fertilizer or legume plant material to unlabeled soil in microplots, and vice versa, then measuring ^{15}N recovery by corn. Corn was also grown in main plots with

split fertilizer N rates (0, 75, 150, and 225 kg ha^{-1}) to measure fertilizer N contribution by the difference method and legume N contribution by the fertilizer replacement value (FRV) method. Using the modified ^{15}N method, it was determined that mineralization-immobilization turnover had no effect on recovery of fertilizer and legume ^{15}N by corn in our study. The difference method overestimated fertilizer N contribution to corn due to increased soil N uptake with increased fertilizer N rates. This increase was not due to a "priming effect" however, as indicated by results from the modified ^{15}N method. Estimates of legume N contributions to corn were greater using the FRV method than when using the ^{15}N method. One reason for this discrepancy was that the FRV method measured both N and non-N rotation effects. Non-N effects accounted for over 80 % of the total rotation effect (i.e. increased corn yield) of alfalfa at KBS as determined using the non-N_2-fixing alfalfa cultivar. Another reason for the discrepancy was the faulty assumption used in the FRV method that fertilizer N use efficiency is 100 %. When the fertilizer N use efficiency measured in our study was taken into account, the FRV and ^{15}N methods gave similar estimates of legume N contribution to corn. It was concluded that MIT effects on recovery of fertilizer and legume N should be measured using modified ^{15}N methods in order to estimate the true N contribution from these sources.

INTRODUCTION

Increasing crop nitrogen (N) use efficiency continues to be a major goal for agriculture. Reaching this goal requires accurate measurements of quantities of N absorbed by crops from different sources. With the current interest in substituting N fixed biologically by forage legumes grown in crop rotations for fertilizer N inputs, measuring and comparing the N contribution from these two sources has become even more important. Both isotopic (^{15}N) and nonisotopic methods have been used in field experiments to measure the recovery of legume and fertilizer N by crops. Large discrepancies in results using these methods have led to considerable discussion and controversy over which methodology is more accurate. In addition, none of the methods so far used to estimate legume N contributions adequately distinguishes either between N derived from N_2-fixation vs. N derived from soil, or between N vs. non-N rotation effects.

Fertilizer N Contributions

Two methods commonly used to quantify crop uptake of applied fertilizer N are the difference method and the ^{15}N method. In the difference method, the amount of N absorbed by plants in unfertilized control plots is subtracted from the amount of N absorbed by plants to which N fertilizer had been applied. In the ^{15}N method, fertilizer labeled with the ^{15}N isotope is applied to plots and the ^{15}N content of the

crop growing on those plots is measured. The difference method normally gives higher estimates than the ^{15}N method of fertilizer N recovery (Harmsen and Kolenbrander, 1965; Jansson and Persson, 1982). Some researchers (Hauck, 1971; Westerman and Kurtz, 1974) have stated that the difference method overestimates fertilizer N uptake compared to the ^{15}N method because it erroneously assumes that mineralization, immobilization, and other N transformations are the same in both the fertilized and unfertilized soils. Specifically, the "priming effect", described as an increase in mineralization of soil N due to the addition of fertilizer N (Hauck and Bremner, 1976), has been cited as a problem leading to inaccuracy with the difference method. Other researchers (Jansson, 1971; Harmsen and Moraghan, 1988) have stated that mineralization-immobilization turnover (MIT) of soil N leads to an underestimate of fertilizer N recovery when using the ^{15}N method. This is because MIT is thought to cause both 1) a pool substitution wherein labeled fertilizer N "stands proxy" for unlabeled inorganic soil N that would have been immobilized, and 2) a displacement reaction wherein labeled fertilizer N is immobilized at the same time unlabeled organic N is mineralized (Jenkinson et al., 1985). Jansson (1958) has claimed that MIT could lead to misinterpretation of experiments with ^{15}N fertilizers, and Paul (1984) has stated that acceptance of significant internal cycling (MIT) would result in recalculation of some ^{15}N results.

Allison (1966) emphasized that the difference and the ^{15}N methods "do not determine exactly the same thing". The ^{15}N method is a direct method which measures recovery of ^{15}N fertilizer by a crop, distinguishing between fertilizer and soil N uptake. The difference method is an indirect method that measures a crop

response to fertilizer and does not distinguish between crop N absorption from fertilizer and from soil organic matter. Jenkinson et al. (1985) pointed out that neither the difference nor the ^{15}N method can provide unequivocal results. Priming effects can cause the difference method to be in error and MIT can cause the ^{15}N method to be in error. Jansson and Persson (1982) concluded that fertilizer N use efficiency experiments should encompass both the difference and ^{15}N methods.

The contribution of fertilizer N to crops can also be estimated based on N use efficiency by crops derived from 1) linear regression of total N uptake by a crop on fertilizer N applied (nonisotopic), and 2) linear regression of labeled (^{15}N) fertilizer N uptake by a crop on fertilizer ^{15}N applied. These methods are not as popular as the difference and ^{15}N methods and have the disadvantage that fertilizer N contribution is based on a single value for fertilizer N use efficiency, determined as the slope of the regression line, for all fertilizer N rates. Westerman and Kurtz (1974) used all four of the above methods to determine the N contribution from fertilizer N to sorghum-sudangrass (*Sorghum sudanese* L.) in the field. They found that the difference method gave higher estimates of N recovery than did the ^{15}N method, which gave higher estimates than the nonisotopic linear regression method, which in turn gave higher estimates than the isotopic linear regression method.

Legume N Contributions

Hesterman (1988) described three methods commonly used to measure the contribution of legume N to nonlegumes in cropping systems: 1) total N in legume biomass (TNLB), 2) the fertilizer replacement value (FRV), and 3) the ^{15}N method.

In addition, a fourth method, the difference method (difference in total N uptake by crops in legume-amended vs. non-amended soils) has also been used to estimate recovery of legume N. The TNLB method is simplest (i.e. least time-consuming and laborious) but has the disadvantage that it represents only the amount of legume N potentially available for uptake by subsequent crops, i.e. it reveals nothing about the availability of the legume N to crops. With the FRV method, a curve is generated for a nonlegume monoculture response to fertilizer N. The yield of a nonlegume crop following a legume with no fertilizer N applied is then compared to yield from the response curve. The FRV is defined as the quantity of fertilizer N required to produce a yield in a crop that does not follow a legume that is identical to that produced by incorporation of the legume (Hesterman, 1988). With the ^{15}N method, ^{15}N-labeled legume plant material is incorporated into soil and recovery of legume N is measured based on the ^{15}N content of the recovery crop and the amount of ^{15}N applied.

Similar to when comparing the difference and ^{15}N methods of measuring fertilizer N uptake, the nonisotopic FRV method often gives much higher estimates of legume N contribution than does the ^{15}N method. For example, using the ^{15}N method, Harris and Hesterman (1990) reported an N contribution from alfalfa to corn of 24 kg ha^{-1}. This value was considerably lower than typical alfalfa N contribution estimates based on FRV methodology which typically exceed 100 kg ha^{-1} (e.g. Bruulsema and Christie, 1987, Fox and Piekielek, 1988). The ^{15}N method is often considered to underestimate legume N recovery for the same reasons given for fertilizer N recovery, i.e. pool substitution or displacement due to MIT. However,

no field studies have adequately assessed the affect of MIT on recovery of legume N.

Two reasons for the large discrepancy between the FRV and ^{15}N methods of measuring legume N contribution were offered by Harris and Hesterman (1990). One reason, which was similar to the reason given by Allison (1966), was that the FRV and ^{15}N methods measure different effects. The ^{15}N method measures solely the N contribution from the legume plant material while the FRV measures the overall response of a crop to a previous legume in terms of fertilizer N application. The overall response measured by the FRV method can include both N and non-N rotation effects. Non-N rotation effects are yield-enhancing effects not directly associated with N, such as improvement in soil structure, reduction of disease and weed pressure, and production of growth-promoting substances. Non-N rotation effects can be estimated using N response curves and are represented by the yield differential between crops following legumes and non-legumes at the highest fertilizer N rate (Pierce and Rice, 1988). Baldock et al. (1981) estimated that 25 % of the corn yield increase in rotations was due to non-N rotation effects. The second reason given for the discrepancy between the ^{15}N and FRV methods of estimating legume N contributions was that the FRV method does not take into account typical fertilizer N use efficiencies of less than 100 percent. Norman et al. (1990) compared the FRV and ^{15}N methods for estimating the recovery of N from rice (*Oryza sativa* L.), soybean (*Glycine max* L.) and wheat (*Triticum aestivum* L.) residues by a subsequent rice crop. They concluded that if the use efficiency of fertilizer N (50 % in their study) was taken into account, the two methods gave comparable results.

Fox et al. (1990) compared the ^{15}N and difference methods of measuring

legume N uptake in a greenhouse study. They found that the difference method gave estimates 20 % higher than the ^{15}N method for recovery of N from snail medic (*Medicago scutellata* L.) and vigna (*Vigna trilobata* L. var verde) residues by sorghum-sudangrass. They speculated that the ^{15}N method was in error due to MIT and stated that all results using the ^{15}N method should be questioned until validated with the difference method, which they consider preferable.

In the same comprehensive review on mineralization and immobilization of soil N in which Jansson and Persson (1982) recommended using both the ^{15}N and difference methods to measure fertilizer N uptake, they also recommended the use of crosswise- tagged systems to elucidate the effect of MIT. With crosswise-tagged systems, an inorganic soil N pool is initially labeled with ^{15}N while an organic pool is not, and vice versa. The ^{15}N content of the inorganic and organic N pools is then measured throughout the experiment in both systems.

The objective of this study was to determine fertilizer and legume N contributions to corn using a modified ^{15}N method that accounts for any effects due to MIT, and to compare these results to those obtained using nonisotopic methods, which included: 1) the difference and linear regression of total N uptake methods (for fertilizer), and 2) the TNLB, FRV, and difference methods (for legumes). We also measured N_2-fixation by the legumes in order to estimate the contribution of fixed-N from the legumes to corn. In addition, we measured the non-N rotation effect for corn following alfalfa using a non-N_2-fixing legume cultivar and compared the result to estimates based on N response curves.

MATERIALS AND METHODS

Short-term (2-yr) crop rotations were established in Spring 1990 at two locations in Michigan: at the Michigan State University Agronomy Farm in East Lansing (EL), and at the Kellogg Biological Station in Hickory Corners, MI (KBS). Soil types were Capac loam (fine-loamy, mixed, mesic, Aeric Ochraqualf) at EL and Kalamazoo loam (fine-loamy, mixed, mesic, Typic Hapludalf) at KBS. Selected soil characteristics for the sites are presented in Table 3.1.

Management of First-Year crops,1990. First-year crops were established in 3.8 m by 38.1 m plots at both locations in Spring 1990, and included: 'Saranac' alfalfa, medium red clover, 'common' hairy vetch, 'Ineffective Saranac' alfalfa, and 'Pioneer 3772' corn. Legumes were seeded on 3 May at EL and 2 May at KBS using a 19-row grain drill with 0.2 m row spacings. Seeding rate for the alfalfas and red clover was 15 kg ha^{-1}. Hairy vetch was seeded at 50 kg ha^{-1}. 'Saranac' alfalfa, red clover and hairy vetch were inoculated with appropriate *Rhizobia* spp. at recommended rates. Ineffective Saranac alfalfa seed, obtained from the University of Minnesota, was not inoculated and served as a non-N_2-fixing control. The N_2-fixing activity of Ineffective Saranac is reportedly 2 % or less of the effective parental cultivar (Barnes et al., 1990) and its usefulness as a non-N_2-fixing control has been demonstrated in field experiments by the ^{15}N isotopic dilution method (Boller and Heichel, 1983). Corn was planted on 23 May at EL and 22 May at KBS using a

Table 3.1 Initialt soil characteristics at East Lansing (EL) and Kellogg Biological Station (KBS).

Location	Bray-1 P	Exchangeable K	Ca	Total N	Organic matter	CEC	pH
	---------- kg ha^{-1} ---------			----- g kg^{-1} ------		cmol kg^{-1}	
EL	134	206	3494	17	24	10	6.9
KBS	143	278	2240	9	16	6	6.1

tsoil sampled Oct 1989.

2-row planter with 0.76 m row spacing. Seeding rate for corn was 55 000 seeds ha^{-1}. No starter fertilizer N or herbicides were used for legume or corn establishment. Alfalfa and red clover plots at both locations received an application of 1.12 kg ha^{-1} a.i. 2,4-DB [4-(2,4-dichlorophenoxy) butoic acid] on 11 June to control broadleaf weeds. Corn plots were cultivated on 26 Jun at EL and 3 Jul at KBS to control weeds. Alfalfa and red clover plots were also sprayed with chlorpyrifos [O,O-diethyl O-(3,5,6-trichloro-2-pyridinyl) phosphorothioate] on 26 Jun at EL and 3 Jul at KBS to control potato leafhopper.

Alfalfa and red clover plots were harvested three times during the seeding year at both locations (11 Jul, 27 Aug and 26 Oct at MSU and 25 Jul, 31 Aug and 24 Oct at KBS). On each harvest date, a 1.2 m by 7.6 m strip from the center of each plot was chopped with a flail-type harvester and weighed. A subsample (approximately 500 g) was taken and dried at 60 C for 4 d to determine dry matter content. Subsamples were used to determine total N content by micro-kjeldahl digestion of 0.1 g ground (1 mm) sample in 4 ml of 12 M H_2SO_4 (with 1.5 g K_2SO_4 and 0.75 g Se catalyst) followed by colorimetric determination of $NH4^+$ on a Lachat flow-injector analyzer (Quik-Chem method no. 10-107-06-2-E, Lachat Chemicals Inc., Mequon, WI). On the last harvest date in Oct (after a killing frost at both locations), hairy vetch herbage was clipped at ground level from two 0.35 m by 0.35 m quadrats per plot to determine dry matter (DM) yield and N content. Dry matter yield and N content of roots/crowns for alfalfa and red clover were determined in Oct by excavating two 0.35 m by 0.35 m quadrats to plow depth (0.25 m). Roots/crowns were washed with tap water to remove soil, weighed, and analyzed for N by the same

micro-kjeldahl procedure used for shoot material. Hairy vetch roots/crowns were not sampled from the main plots.

N-fixation by legumes during the seeding year was measured using the ^{15}N isotopic dilution method (McAuliffe et al., 1958). Microplots consisting of 0.38 m by 0.38 m by 0.20 m tall sheet metal boxes were inserted to a 0.15 m depth in all legume plots on 14 May at EL and 22 May at KBS (soon after emergence of the legumes). Microplot boxes were installed with minimum disturbance to the growing legumes and were located in the center and approximately 1.5 m from the end of each plot. A small amount (equivalent to 5 kg N ha^{-1}) of highly labeled (78 % ^{15}N) ammonium sulphate was then applied evenly to each microplot in a solution with a C:N ratio of 20:1 (half of the carbon supplied from D-glucose and half from sodium acetate). On each date that the alfalfa and red clover plots were harvested, shoots from the microplots were clipped, dried, weighed, ground and analyzed for total N and ^{15}N on a Europa Scientific CN analyzer/mass spectrometer. On the last harvest date (Oct), hairy vetch shoots were also sampled from microplots for total N and ^{15}N determination. The hairy vetch was destructively sampled at this time since it had already begun to senesce and was expected to winterkill. The percent of legume N derived from fixation was calculated by the equation:

$$\% \ N_2 \text{ fixed} = [1 - {}^{15}N \text{ a.e. FC} / {}^{15}N \text{ a.e NFC}] \times 100 \qquad [1]$$

where FC = N_2-fixing crop and NFC = non-N_2-fixing crop (Fried and Middelboe, 1977). Ineffective Saranac alfalfa (alfalfa-) was used as the non-N_2-fixing reference

crop for Saranac alfalfa (alfalfa+), red clover and hairy vetch.

Results for alfalfa and red clover DM, total N, and fixed-N harvested in 1990 were analyzed using ANOVA for a randomized complete block design with 4 replications. First-year corn grain and stover yields were not measured. Corn ears were, however, removed from the plots and the stover was chopped and left on the soil surface.

Spring 1991 Legume Sampling. Immediately prior to moldboard plowing all plots on 9 May at EL and 10 May at KBS, both shoots and roots/crowns were sampled from the alfalfa and red clover main plots. Two 0.35 by 0.35 quadrats per plot were sampled and analyzed for dry matter and N content using methods similar to the previous year. Hairy vetch shoots (in main plots seeded to hairy vetch in 1990) that had completely winterkilled were also sampled at this time. Roots/crowns were again not sampled from the hairy vetch main plots. Both shoots and roots/crowns were sampled from the alfalfa and red clover N-fixation microplots at this time to be analyzed for total N and ^{15}N. The sheet metal boxes were removed from the plots after sampling.

Corn Main Plots, 1991. Corn ('Pioneer 3753') was planted on all plots on 13 May at both locations. Each corn plot was 4 rows wide and planted with a 4-row planter at EL and a 2-row planter at KBS. Seeding rate was 55 000 seeds ha^{-1} and row spacing was 0.76 m at both locations. Main plots (3.8 m by 24.4 m) were split into four subplots (3.8 m by 6.1 m) and fertilized with 0, 75, 150, and 225 kg N ha^{-1} as ammonium sulphate broadcast on 29 May at EL and 1 Jun at KBS. A 3 m long section at the end of each replication where the N-fixation microplots were located

was avoided when establishing the fertilized subplots. In addition, a 11.7 m long section at the opposite end of each replication was reserved (i.e. not plowed or planted with a corn planter) for the modified ^{15}N method.

An unconfined microplot (0.76 m by 1.0 m, centered over a non-yield row) was established in each subplot and fertilized with labeled (1.4 % ^{15}N) ammonium sulphate. Metal catch pans were placed over these microplots when subplots were fertilized with unlabeled N. The pans were then removed and a wooden frame was placed on the soil surface surrounding the microplot. The ^{15}N fertilizer was applied evenly to the microplot in 4 L dH_2O with a watering can and the frame was then removed.

Weed control was accomplished using 2.24 kg ha^{-1} a.i. metolachlor [2-chloro-N-(2-ethyl-6-methylphenyl)-N-(2-methoxy-1-methyl-ethyl) acetamide] and 1.96 kg ha^{-1} a.i. cyanazine [2[[4-chloro-6-(ethlamino)-S-tiazin-2-yl]amino]-2-methylpropionitrile] preemergence herbicides at both locations plus 1.12 kg h^{-1} a.i. betazon [3-(1-methylethyl)-1 H-2,1,3-benzothiadiazin-4(3H)-one 2,2-dioxide] postemergence herbicide at EL. Alleys were cut between fertilized subplots with a 0.76 m wide flail-type harvester on 5 Aug at EL and 6 Aug at KBS to aid in corn harvest. Corn was picked by hand from the center two rows of each 4-row subplot on 25 Sep at EL and 8 Oct at KBS. Grain was shelled with a mechanical sheller, weighed and sampled for dry matter and N content. Corn stover from the center two rows of each subplot was harvested with a flail-type harvester, weighed and sampled for DM and N content. Total N was determined on each sample by the micro-kjeldahl procedure described previously. Grain and stover from a single corn plant located in the center

of each unconfined ^{15}N microplot was harvested by hand, dried, weighed, ground (0.5 mm) and analyzed for total N and ^{15}N.

Results for corn grain and aboveground DM yield and total N uptake were analyzed by ANOVA procedures for a randomized complete block, split-plot design with 4 replications. Main plots were first-year crops and subplots were fertilizer N rates. The same data were also analyzed within fertilizer N rate and, if significant, means were separated by an LSD test. The effects of fertilizer N on corn yields were also analyzed within each first-year crop. Significant corn yield responses to N within first-year crops were further analyzed using single degree of freedom orthogonal contrasts (linear and quadratic) and the appropriate regression equations were calculated.

The fertilizer N contribution to corn in this study was calculated by the difference, ^{15}N, and nonisotopic and isotopic linear regression methods as described in the introduction. The unconfined microplots that received ^{15}N fertilizer were used to calculate fertilizer uptake by the ^{15}N method. The linear regression methods of measuring fertilizer N contribution were calculated in this study as described by Westerman and Kurtz (1974), using the basic equation: $y = a + bx$, where :

Nonisotopic

y = total N in corn

a = intercept (total N in corn with no N fertilizer N applied)

b = regression coefficient ($b \times 100$ = fertilizer N use efficiency in %)

x = rate of applied fertilizer N

Isotopic

y = fertilizer ^{15}N in corn

a = intercept (theoretically = 0, i.e. no ^{15}N in corn when none applied)

b = regression coefficient (b x 100 = fertilizer N use efficiency in %)

x = rate of applied fertilizer ^{15}N

Since the uptake of ^{15}N fertilizer at the zero ^{15}N rate is theoretically zero, the 0,0 data point was used in each first-year crop data set and the regressions were forced through the origin.

The legume N contribution to corn in this study was calculated by the TNLB, FRV, and difference methods as described in the introduction. Total N in legume biomass (shoots + roots/crowns) from the Spring 1991 sampling was used to calculate the legume N contribution by the TNLB method. Fertilizer replacement values of the legumes were calculated on grain and aboveground biomass DM yield response to fertilizer N for corn following corn. Total (N + non-N) and non-N rotation effects of all legumes on corn were calculated using N response curves (N response method) as described in the introduction. The non-N rotation effect of alfalfa on corn using the non-N_2-fixing alfalfa (non-N_2-fixing method) was calculated by the yield differential between unfertilized corn following corn and following Ineffective Saranac alfalfa, if the difference was significantly different by an LSD test.

Modified ^{15}N Method

Legume and fertilizer N (at the 150 kg ha^{-1} rate) contributions to corn were measured using a modified ^{15}N method. Three microplots, consisting of 0.38 m by 0.38 m by 0.20 m open-ended sheet metal boxes, were installed in each main plot (except 'Ineffective Saranac') at EL on 15 May and at KBS on 16 May 1991. The boxes were inserted to a 0.15 m depth with minimum disturbance to soil inside the microplots. The three microplots in each main plot were spaced 2 m apart and established in an undisturbed (not plowed) area at one end of each replication.

The equivalent of 10 kg N ha^{-1} as ammonium sulphate was applied to each microplot as an aqueous solution (4 L) using a watering can. The solution also contained the equivalent of 40 kg C ha^{-1} as glucose and sodium acetate (equal parts C) to give a final C:N ratio of 40:1. The ammonium sulphate applied to one microplot in each main plot was unlabeled (^{14}N), and the other two microplots received labeled (98 % ^{15}N) ammonium sulphate. Forty-eight hours after the solution was applied, the top 15 cm of soil from each microplot was excavated, mixed and sampled. Soil was also sampled (0-15 cm) from an area outside the microplots to provide an untreated control. All soil samples were analyzed for total N and ^{15}N, inorganic N and ^{15}N and microbial biomass C, N and ^{15}N according to methods described in Chapter 2.

After soil sampling, legume residues (in those plots that had been sown to legumes the previous year) were removed from the soil by hand. Legume plant material labeled with ^{15}N (ca. 5 % atom excess) was added to the microplot that had received unlabeled ammonium sulphate. Unlabeled plant material was added to one of the microplots that had received ^{15}N labeled ammonium sulphate. No plant

material was added to the other microplot that had received ^{15}N labeled ammonium sulphate. All legume plant material applied to the microplots was produced in the greenhouse (fertilized weekly with either labeled or unlabeled N), dried and ground (2mm). Alfalfa and red clover plant material was applied at a rate equivalent to 112 kg N ha^{-1} and hairy vetch was added at a rate equivalent to 68 kg N ha^{-1}.

After sampling soil from microplots in the main plots that had first-year corn in 1990, the soil was returned directly to the microplots. Ammonium sulphate fertilizer labeled with ^{15}N (10 % atom excess) was then added at a rate of 150 kg N ha^{-1} to the microplot that had received the initial unlabeled ammonium sulphate solution. Unlabeled ammonium sulphate was added at a rate of 150 kg ha^{-1} to one of the microplots that had received labeled ammonium sulphate initially. No ammonium sulphate fertilizer was added to the other microplot that received the initial ^{15}N solution. All fertilizer solutions were applied to the microplots evenly in 1 L dH_20 using a watering can. The treatments for this method were thus designated:

1) Unlabeled (^{14}N) soil / ^{15}N legume plant material or fertilizer
2) ^{15}N soil / Unlabeled (^{14}N) legume plant material or fertilizer
3) ^{15}N soil / No legume plant material or fertilizer.

Two corn seeds were planted in each microplot and subsequently thinned to one plant per microplot soon after emergence. The area surrounding the microplots was rototilled and corn was planted in this area by hand. During the growing season, the microplots and surrounding area were weeded by hand as needed. When corn

from the main plots was harvested at each location, corn grain, stover and roots were sampled from each microplot and analyzed for total N and ^{15}N. After corn harvest, soil from the microplots was excavated, mixed, sampled and analyzed by the same methods used at the beginning of the experiment.

The experimental design for this method was a randomized complete block, split-plot with 4 replications. First-year crops were main plots and microplot treatments (1, 2 and 3) were subplots. The effect of the initial N solution on soil microbial biomass C and N, inorganic N and total N pool sizes 48 hours after application was determined by single degree of freedom orthogonal contrast, comparing treatments 1 + 2 + 3 to the untreated control soil. The contribution of fertilizer and legume N to corn was calculated by:

^{15}N uptake by corn in treatment 1 + treatment 2 - treatment 3 [2]

Recovery of ^{15}N in treatment 1 alone represents the traditional ^{15}N method of estimating fertilizer and legume N contributions and was calculated as described in Harris and Hesterman (1990). Recovery of ^{15}N by corn in treatments 2 and 3 was used to correct for MIT and/or priming effects. Recovery of ^{15}N by corn in treatments 2 and 3 was adjusted (proportionally) to the size of the soil inorganic and microbial biomass pools where the ^{15}N originated before using equation [2]. For example, if the soil inorganic plus microbial biomass pools in treatment 2 or 3 was equivalent to 200 kg N ha^{-1} at the beginning of the experiment, and 10 % of the ^{15}N in these pools was recovered by corn at the end of the experiment, the value used in

the above equation would be 0.1 x 200 = 20 kg N ha^{-1}. If recovery of ^{15}N by corn in treatments 2 and 3 was not significantly different, as determined by an LSD test, the above equation reduces to treatment 1 alone , which is equivalent to the traditional ^{15}N method.

RESULTS AND DISCUSSION

The Modified ^{15}N Method

Forty-eight hours after applying the initial N solution, the soil microbial biomass C and N pools in treatments 1, 2 and 3 at both EL and KBS were approximately 10 % greater in size than in the untreated control soil (Table 3.2). In addition, the inorganic soil N pool in treatments 1, 2 and 3 at KBS was 20 % greater than in the untreated control. These results indicate that soil N transformations in treatments 1, 2 and 3 may not reflect transformations occurring in the main plots. There was, however, no difference in soil pool sizes among treatments 1, 2 and 3 within any first-year crop at either location (data not shown). This was expected, since the same amount of N was applied to each treatment. If the soil pool sizes were significantly different between treatments at this stage of the experiment, the final result using the above equation may not be valid.

The distribution of ^{15}N among microbial biomass, inorganic and total N pools was not different between treatments 2 and 3 measured 48 hours after the label was applied (Table 3.3). This was also expected since both treatments were the same at this point. If ^{15}N among soil fractions in treatments 2 and 3 was different at the beginning of the experiment, the adjustment for MIT effects using these treatments in the equation above would not be possible.

Table 3.2. Effect of nitrogen solution on soil microbial biomass C and N, inorganic N and total N pools 48 hours after application to microplots of the modified ^{15}N method. [Values are averaged over first-year crops and treatments 1, 2 and 3 and compared to untreated control soil).]

		Soil Pool			
Location	Treatment	Microbial biomass Carbon	Microbial biomass Nitrogen	Inorganic N	Total N
		---------- ug g^{-1} ----------			
EL	Avg. 1-3	504	100	14.7	1600
	Control	451	93	14.1	1590
	Significance	***	**	NS	NS
	CV(%)	7	8	17	9
KBS	Avg. 1-3	390	72	13.4	880
	Control	337	65	11.2	840
	Significance	**	**	**	NS
	CV(%)	14	11	21	7

*,**,*** Significant at the 0.05, 0.01 and 0.001 probability levels, repsectively;
NS = not significant at P = 0.05.

Table 3.3. Recovery of ^{15}N from solution applied for the modified ^{15}N method in soil microbial biomass, inorganic and total N pools 48 hours after application. [Values are averaged over first-year crops.]

Location	Treatment	Soil N Pool		
		Microbial biomass	Inorganic	Total
		---------- kg ha^{-1} ----------		
EL	2	2.06	0.621	3.64
	3	2.04	0.645	3.71
Significance		NS	NS	NS
CV(%)		26	33	29
KBS	2	2.21	0.853	3.60
	3	1.75	0.802	3.34
Significance		NS	NS	NS
CV(%)		34	40	28

*,**,*** Significant at the 0.05, 0.01, and 0.01 probability levels, repectively; NS = not significant at P = 0.05.

Table 3.4. Recovery of ^{15}N by corn grain and aboveground biomass from treatments 2 and 3 of the modified ^{15}N method. [Values are averaged over first-year crops.]

Location	Treatment	Grain	Aboveground biomass
		---------- kg ha^{-1} ----------	
EL	2	0.704	0.994
	3	0.643	0.893
Significance		NS	NS
CV(%)		17	17
KBS	2	0.657	1.104
	3	0.626	1.026
Significance		NS	NS
CV(%)		17	17

*,**, *** Significant at the 0.05, 0.01, and 0.001 probability levels, respectively; NS = not significant at P = 0.05.

Corn recovered similar amounts of ^{15}N from treatments 2 and 3 for all first-year crops at both EL and KBS (Table 3.4). Recovery of legume and fertilizer ^{15}N by corn was therefore not affected by any priming or MIT action. The equation for calculating legume and fertilizer N contributions by the modified ^{15}N method was reduced to treatment 1 alone, or the equivalent of the traditional ^{15}N method. Results by the traditional ^{15}N method, as represented by treatment 1 of the modified ^{15}N method for legumes and the unconfined microplots for fertilizer, were therefore used for comparisons to the nonisotopic methods in the following sections.

Fertilizer N Contributions

Difference Method vs. ^{15}N Method. Although not compared statistically, fertilizer N contribution estimates using the difference and ^{15}N methods are discussed here. Estimates of the fertilizer N contribution to corn grain and aboveground biomass following first-year corn were greater using the difference method compared to the ^{15}N method at the 75 kg ha^{-1} fertilizer N rate at EL and all three N rates at KBS (Table 3.5). The two methods gave similar results, however, for the 150 and 225 kg ha^{-1} N rates at EL. The ^{15}N method did not underestimate fertilizer N contributions to corn due to MIT effects as proven by results from the modified ^{15}N method discussed previously. Instead, the difference method overestimated the fertilizer N contribution to corn following corn at the 75 kg ha^{-1} N rate at EL and all three N rates at KBS. The cause of the overestimation was greater soil N uptake by corn in the fertilized plots compared to the unfertilized control (Table 3.6). This was not a priming effect, but rather an increase in soil N uptake proportional to the increase in fertilizer N

Table 3.5. Fertilizer N uptake by corn grain and aboveground biomass following different first-year crops as measured by the difference and ^{15}N methods at two Michigan locations.

Location	First-year crop	Difference Method			^{15}N Method		
		Fertilizer N rate (kg ha^{-1})			Fertilizer N rate (kg ha^{-1})		
		75	150	225	75	150	225
		---------- Fertilizer N uptake by grain (kg ha^{-1}) ----------					
EL	Alfalfa+	19	23	24	16	26	31
	Red clover	19	7	15	18	23	28
	Hairy vetch	20	16	16	12	32	34
	Alfalfa-	31	36	38	18	34	39
	Corn	28	36	55	18	32	51
	LSD(0.05)	11	13	9	5	8	14
	CV(%)	33	35	20	23	19	25
KBS	Alfalfa+	28	37	47	14	18	21
	Red clover	27	31	36	10	21	34
	Hairy vetch	27	40	47	11	23	32
	Alfalfa-	38	49	54	18	23	29
	Corn	26	59	58	16	28	35
	LSD(0.05)	10	19	12	NS	8	14
	CV(%)	21	29	16	39	25	30
		--- Fertilizer N uptake by aboveground biomass (kg ha^{-1}) ----					
EL	Alfalfa+	15	26	33	19	32	41
	Red clover	23	10	20	21	28	35
	Hairy vetch	25	28	26	15	42	42
	Alfalfa-	38	39	42	24	42	48
	Corn	31	47	69	22	41	64
	LSD(0.05)	13	18	17	6	9	18
	CV(%)	31	38	29	20	16	25
KBS	Alfalfa+	29	50	60	19	26	28
	Red clover	23	34	42	14	29	46
	Hairy vetch	42	47	59	15	32	43
	Alfalfa-	45	61	72	25	30	40
	Corn	36	58	78	22	38	46
	LSD(0.05)	14	25	17	NS	11	18
	CV(%)	26	31	17	41	24	29

Table 3.6. Soil N uptake by corn grain and aboveground biomass following first-year corn at El and KBS.

	Fertilizer N rate (kg ha^{-1})			
Location	0	75	150	225
	-------- Soil N uptake by grain (kg ha^{-1}) ----------			
EL	46	56	49	50
KBS	35	45	73	65
	---- Soil N uptake aboveground biomass (kg ha^{-1}) ----			
EL	62	69	60	61
KBS	52	60	96	94

uptake. Fried and Broeshart (1974) also made this distinction and explained that healthier plants (due to fertilizing with N when N is limiting) simply absorb more soil N. Stimulated root development and rhizosphere effects have also been cited as possible causes of increased soil N uptake by N-fertilized crops in the field (Harmsen and Moraghan, 1988; Jansson and Persson; 1982). The fact that the difference and ^{15}N methods gave similar estimates of fertilizer N contribution to corn at EL for the 150 and 225 kg ha^{-1} N rates -- when uptake of soil N by corn was similar to the unfertilized control -- further supports our conclusion that the difference method overestimated fertilizer N contribution to corn when soil N uptake by corn in the fertilized plots is greater than in the unfertilized control.

The difference and ^{15}N methods of estimating fertilizer N contributions were also compared for corn following first-year legume crops in this study (Table 3.5). At EL, the difference method gave similar or higher estimates than the ^{15}N method for fertilizer N contributions to corn following legumes at the 75 kg ha^{-1} N rate EL, and lower estimates at the 150 and 225 kg ha^{-1} N rates. At KBS, the difference method gave higher estimates of fertilizer N contributions to corn following legumes at all three N rates. The overall trend for estimating fertilizer N contributions to corn following legumes using the difference and ^{15}N methods was similar to the trend for corn following corn (especially at KBS). The difference method probably overestimated the fertilizer N contribution to corn following legumes. This could not be proven in this study since uptake of soil N by corn could not be distinguished from uptake of legume N in the fertilized plots. Uptake of soil N and legume N could be distinguished in fertilized systems if a crosswise tagging system had been used, i.e.

applying ^{15}N-labeled fertilizer and unlabeled legume N, and vice versa. This information would be useful to determine the most efficient and environmentally-sound fertilizer N rates for corn grown following legume crops.

Nonisotopic vs. Isotopic Linear Regression. The nonisotopic and isotopic linear regression methods gave similar estimates of fertilizer N contribution to corn following first-year corn and non-N_2-fixing alfalfa at all fertilizer N rates at EL (Table 3.7). At KBS, the nonisotopic method gave higher estimates than the isotopic method at all fertilizer N rates. There was no significant linear effect of fertilizer N applied on total N uptake by corn (nonisotopic linear regression) following legumes at EL. Thus, fertilizer N contribution to corn using this method at this location could not be calculated. Also, there was no clear trend comparing the nonisotopic and isotopic regression methods of estimating fertilizer N contributions to corn following legumes at KBS.

Disadvantages of the linear regression methods, as illustrated by our results, are 1) fertilizer N contribution can not be calculated if the linear effect of fertilizer N (or ^{15}N) on total N (or ^{15}N) uptake by crops is not significant, and 2) estimates of fertilizer N contributions using nonisotopic and isotopic linear regression methods may vary depending on first-year crops and location.

Non-N Rotation Effects of Legumes

Using the N response method, i.e. based on the yield difference between corn following corn and following legumes (both at the 225 kg ha^{-1} fertilizer N rate) non-N rotation effects of alfalfa, red clover and hairy vetch on corn ranged from 18 to 42 %

Table 3.7. Fertilizer N uptake by corn grain and aboveground biomass following different first-year crops as measured by the nonisotopic and isotopic regression methods.

Location	First-year crop	Nonisotopic regression Method			Isotopic regression Method		
		Fertilizer N rate (kg ha^{-1})			Fertilizer N rate (kg ha^{-1})		
		75	150	225	75	150	225
		------- Fertilizer N uptake by grain (kg ha^{-1}) -------					
EL	Alfalfa+	--	--	--	11	22	34
	Red clover	--	--	--	10	21	32
	Hairy vetch	--	--	--	14	28	43
	Alfalfa-	16	32	47	14	28	43
	Corn	17	34	52	16	33	50
KBS	Alfalfa+	14	28	43	8	16	25
	Red clover	9	18	27	10	21	32
	Hairy vetch	15	30	45	11	22	34
	Alfalfa-	18	36	54	10	21	32
	Corn	23	46	70	13	26	38
		--- Fertilizer N uptake by aboveground biomass (kg ha^{-1}) ---					
EL	Alfalfa+	--	--	--	15	30	45
	Red clover	--	--	--	13	26	38
	Hairy vetch	--	--	--	16	33	50
	Alfalfa-	18	36	54	18	36	54
	Corn	22	44	68	20	40	61
KBS	Alfalfa+	18	36	54	11	22	34
	Red clover	--	--	--	15	30	45
	Hairy vetch	17	34	52	15	30	45
	Alfalfa-	24	48	72	15	30	45
	Corn	31	62	92	16	33	50

of the total (N + non-N) rotation effect at EL, and from 0 to 31 % at KBS (Table 3.8). The response curves at EL and KBS are shown in Figures 3.1 and 3.2, respectively, and the equations used to generate the curves are listed in Table 3.9. Using the N response method, the non-N rotation effects of alfalfa on corn at EL and KBS were 31 and 9 % of the total effect, respectively based on grain yields and 42 and 0 % , respectively, based on aboveground biomass yields. Using the non-N_2-fixing method, i.e. the difference between corn yields following Ineffective Saranac alfalfa and following corn (both not fertilized), it was estimated that over 80 % of the total effect of alfalfa to corn was due to non-N rotation effects at KBS. The non-N rotation effect of alfalfa on corn could not be calculated at EL using the non-N_2-fixing legume method since corn yields were not significantly different when following either Ineffective Saranac alfalfa or corn with no N applied. The large discrepancy between the two methods for estimating the non-N rotation effect of alfalfa to corn at KBS may be due to a faulty assumption of the N response method. Russelle et al. (1987) pointed out that the assumption that rotation effects are constant and not affected by N fertilization when using the N response method has not been tested. In addition, since the non-N_2-fixing method is a more direct measurement that does not involve fertilization with N, it may be a more accurate assessment of non-N rotation effects.

Table 3.8. Total (N + non-N) and non-N rotation effects measured by the "N response" and "non-N_2-fixing methods at EL and KBS.

Method	Location	Corn Yield Parameter	First-yr Crop	Rotation Effects: Total (N + Non-N)	Rotation Effects: Non-N	Rotation Effects: % of Total Due To Non-N
				------ kg ha^{-1} ------		
N Response	EL	Grain	Alfalfa+	4444	1371	31
			Red clover	4851	862	18
			Hairy vetch	3824	954	25
	KBS		Alfalfa+	2786	246	9
			Red clover	3816	0	0
			Hairy vetch	3133	962	31
	EL	Aboveground Biomass	Alfalfa+	6487	2854	42
			Red clover	7095	1392	20
			Hairy vetch	5994	1675	28
	KBS		Alfalfa+	4605	0	0
			Red clover	5397	0	0
			Hairy vetch	4338	1053	24
Non-N_2-fixing	KBS	Grain	Alfalfa+	2047	-	-
			Alfalfa-	-	1700	83
		Aboveground Biomass	Alfalfa+	2657	-	-
			Alfalfa-	-	2507	94

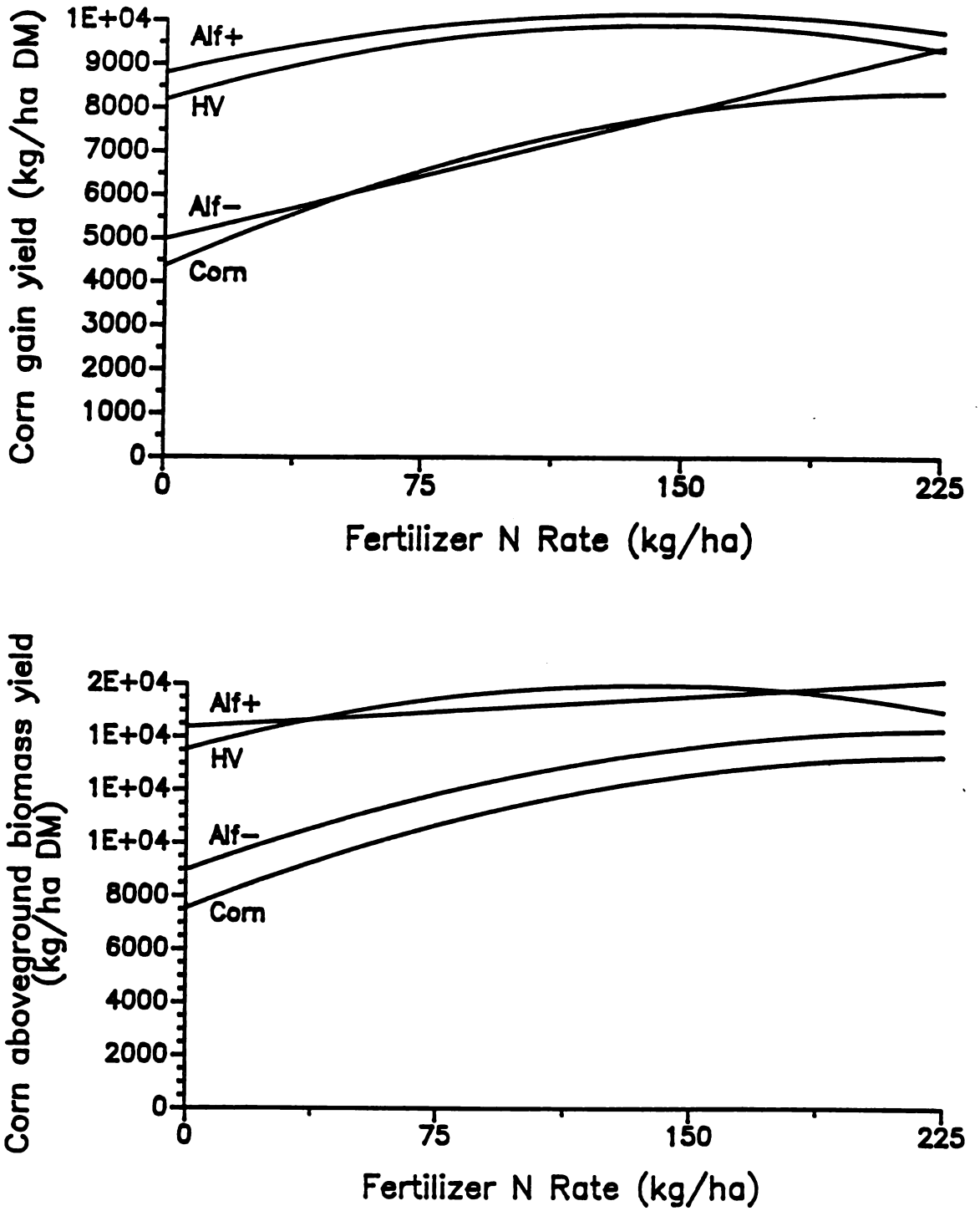

Figure 3.1. Effect of N fertilizer on corn grain (top) and aboveground biomass (bottom) yields following different first-year crops at EL.

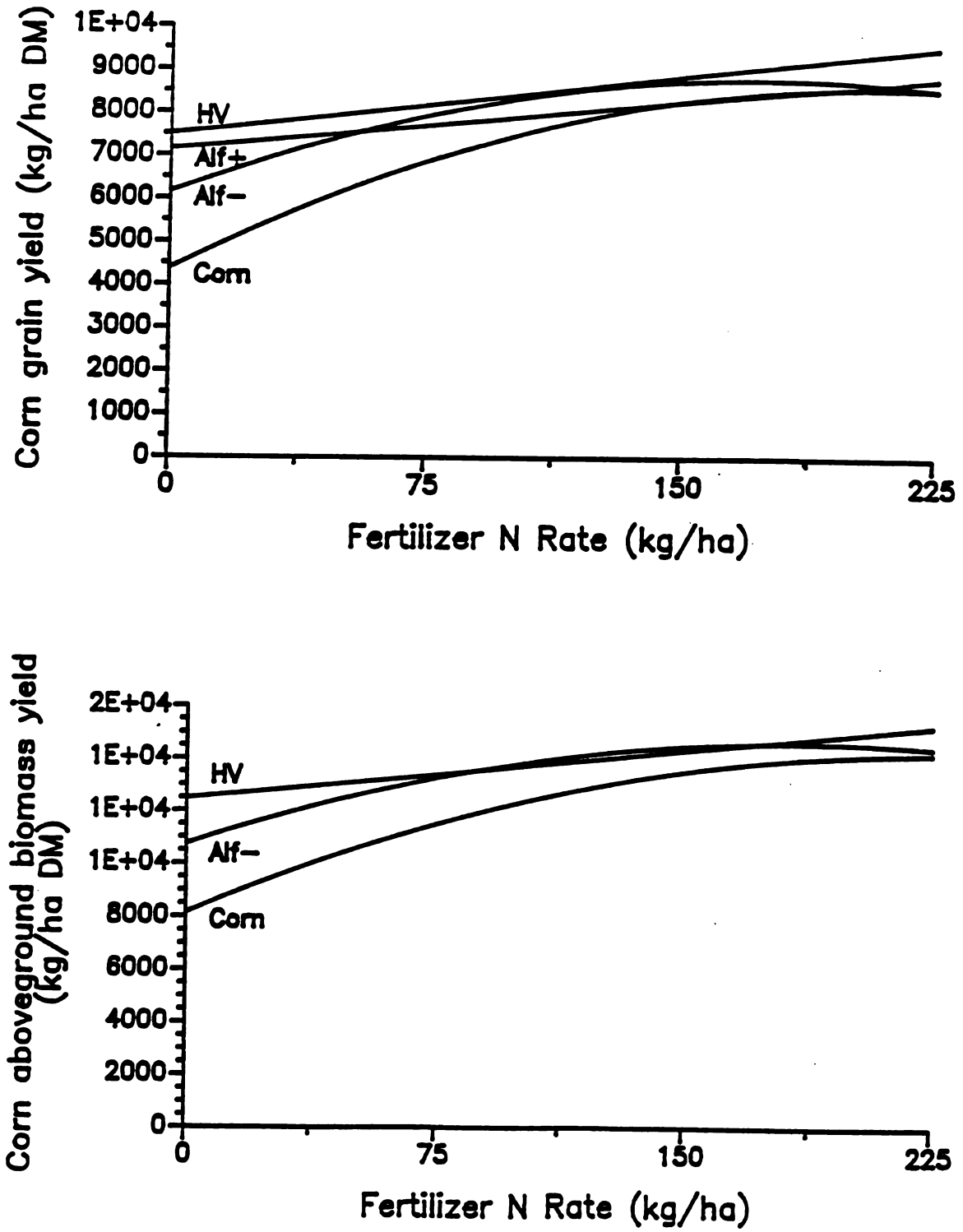

Figure 3.2. Effect of N fertilizer on corn grain (top) and aboveground biomass (bottom) yields following different first-year crops at KBS.

Table 3.9. Nitrogen response curve equations for corn grain and aboveground biomass dry matter yields following different first-year crops at EL and KBS. [Equations used for Figures 3.1. and 3.2.]

Response	Location	First-year crop	Equation†	r^2
Grain Yield	EL	Alfalfa+	$y = 8804 + 18.7x - 0.065x^2$	.879
		Hairy vetch	$y = 8184 + 24.1x - 0.085x^2$	.750
		Alfalfa-	$y = 4981 + 19.6x$	.950
		Corn	$y = 4360 + 35.5x - 0.079x^2$	.964
	KBS	Alfalfa+	$y = 7150 + 7.26x$	.432
		Hairy vetch	$y = 7497 + 8.9x$	.841
		Alfalfa-	$y = 6142 + 40.6x - 0.087x^2$	.980
		Corn	$y = 4364 + 40.6x - 0.098x^2$	.986
Aboveground biomass Yield	EL	Alfalfa+	$y = 14378 + 7.6x$	.860
		Hairy vetch	$y = 13525 + 35.4x - 0.13x^2$	.951
		Alfalfa-	$y = 8963 + 45.9x - 0.10x^2$	.987
		Corn	$y = 7531 + 50.1x - 0.11x^2$	.965
	KBS	Hairy vetch	$y = 12465 + 12.2x$	.812
		Alfalfa-	$y = 10740 + 42.2x - 0.12x^2$	.980
		Corn	$y = 8127 + 53.8x - 0.12x^2$	.989

†y=response, x=fertilizer N rate; All equations significant at the 0.05 probability level

Legume N Contributions

The TNLB method gave similar estimates of legume N contributions for each species at EL and KBS, averaging 138 kg ha^{-1} for alfalfa, 157 kg ha^{-1} for red clover and 80 kg ha^{-1} for hairy vetch (Table 3.10). These values, however, may be underestimated due to differences in the amount of N present in legumes in fall vs. spring. For alfalfa and red clover at EL, almost twice as much N was present in roots/crowns in Fall '90 compared to Spring '91 (Table 3.11). For example, 106 kg N ha^{-1} was present in alfalfa roots/crowns in Fall '90, but only 55 kg N ha^{-1} was present in Spring '91. The TNLB method therefore may have underestimated the N contribution by 51 kg ha^{-1}. Griffin and Hesterman (1991) recommended using values of root/crown N from fall samplings when estimating N contributions from legumes destroyed in spring to account for such differences. The fate of this "missing N" and its availability to subsequent crops, however, is not known. Also, in our study there was little difference in legume root/crown N content measured in Fall '90 and Spring '91 at KBS. Therefore, at KBS, the estimate of legume N contribution using the TNLB method would not differ whether legume root/crown N content values for the fall or the spring sampling were used. Estimates of N contribution to corn from hairy vetch using the TNLB method may be underestimated by more than 100 kg ha^{-1} at both locations in this study. This was concluded since, an average of 200 kg N ha^{-1} was measured in hairy vetch shoots at EL and KBS in Fall '90, but less than 100 kg N ha^{-1} was present in Spring '91. However, as with the decline in alfalfa and red clover root/crown N between fall and spring at EL, the fate and availability of this "missing" N is not known.

Table 3.10. Estimates of legume N contribution to corn grain (G) and aboveground biomass (AGB) using the total N in legume biomass (TNLB), fertilizer replacement value (FRV), ^{15}N, and difference methods.

			Method					
			FRV		^{15}N		Difference	
Location	Legume	TNLB	G	AGB	G	AGB	G	AGB
		kg N ha^{-1}						
EL	Alfalfa+	145	225	225	25	33	58	86
	Red clover	156	182	225	20	28	53	59
	Hairy Vetch	87	225	225	10	13	54	66
KBS	Alfalfa+	130	62	59	20	31	24	30
	Red clover	158	93	123	22	33	42	66
	Hairy vetch	73	87	90	15	23	34	36

Table 3.11. Total N content of legumes sampled in Fall '90 and Spring '91 at EL and KBS.

Location	Plant part	Legume	Fall '90	Spring '91
			-------- kg ha^{-1} -------	
EL	Root/crown	Alfalfa+	106	55
		Red clover	93	47
KBS	Root/crown	Alfalfa+	59	50
		Red clover	54	58
EL	Shoot	Hairy vetch	234	87
KBS	Shoot	Hairy vetch	173	73

Results using the ^{15}N isotopic dilution method of measuring N-fixation showed that at EL an average of 42 % of the total N in legumes (shoots + roots/crowns) measured in Spring '91 came from biological fixation (Table 3.12). Legumes sampled in Spring '91 at KBS derived more N by fixation than legumes at EL, accounting for 64 % of the total N content. These values represent the potential contribution of fixed-N by legumes to corn. The greater N_2-fixation by legumes at KBS is probably related to the lower N status of soil at that location.

Unlike with the TNLB method, the FRV method gave much different estimates of legume N contributions for each species at EL compared to KBS (Table 3.10). Corn yields following legumes with no fertilizer N at EL were often greater than when following corn at even the highest fertilizer N rate (Figure 3.1). In these cases, the legumes were assigned a FRV of 225 kg N ha^{-1} (the highest fertilizer N rate). The FRV for all legumes at EL averaged 218 kg N ha^{-1}. This is at the high end of the 13 (Fribourg and Bartholomew, 1956) to 200 kg ha^{-1} (Hesterman et al., 1986) range of FRVs for legume-corn systems, and twice as high as the more typical legume FRV of 100 kg N ha^{-1} (e.g. Bruulsema and Christie, 1987). The FRVs for legumes at KBS were much lower than at EL, averaging 60, 108, and 88 kg N ha^{-1} for alfalfa, red clover and hairy vetch, respectively. The reason for the higher estimates of legume N contribution to corn at EL compared to KBS using FRV methodology when estimates using the TNLB method were similar is unclear. However, Griffin and Hesterman (1989) also reported that legume N contribution estimates using the FRV method were not directly associated with the amount of N accumulated in legume biomass prior to plowdown.

Table 3.12. Total N and fixed-N content of legumes sampled in Spring '91 at EL and KBS.

Location	Legume	Shoots		Roots/crowns		Shoots + Roots/crowns	
		Total N	Fixed-N	Total N	Fixed-N	Total N	Fixed-N
		kg ha^{-1}					
EL	Alfalfa+	90	33	55	28	145	61
	Red clover	109	36	47	17	156	53
	Hairy vetch	87	44	0	0	87	44
KBS	Alfalfa+	80	50	50	36	130	86
	Red clover	100	62	58	42	158	104
	Hairy vetch	73	43	0	0	73	43

The FRV method grossly overestimated (by an average of 10 times) the N contribution from legumes to corn compared to the ^{15}N method at EL (Table 3.10). At KBS, the FRV method gave estimates of legume N contributions twice as high as the ^{15}N method for alfalfa, and 4 times as high for red clover and hairy vetch. The large discrepancies between the FRV and ^{15}N methods were not due to an underestimation of legume N contribution using the ^{15}N method due to MIT effects as proven by results from the modified ^{15}N method. The discrepancy is also not due to the FRV including non-N rotation effects at EL, since no rotation effects were measured at this location using the non-N_2-fixing method. Most of the discrepancy at EL therefore was probably related to differences in soil N uptake between treatments. Since non-N rotation effects accounted for over 80 % of the total rotation effect for alfalfa at KBS as measured by the non-N_2-fixing method, most of the discrepancy between the FRV and ^{15}N methods at KBS probably was due to the FRV including non-N rotation effects.

If the fertilizer N use efficiency as determined by the isotopic regression method is used to adjust legume FRVs in this study, like was done by Norman et al. (1990), the FRV estimates are only 4 times higher than estimates using the ^{15}N method at EL and are actually comparable at KBS (Table 3.13). These results support the conclusion by Norman et al. (1990) that the ^{15}N and FRV methods may compare favorably when the uptake efficiency of fertilizer N is taken into account. The unadjusted FRV estimates, however, still provide a good estimate of the overall response of a crop following a legume in terms of a fertilizer N application.

The difference method gave estimates of legume N contributions approximately

Table 3.13. Nitrogen contribution from three legume species to corn estimated by the fertilizer replacement value (FRV) method after adjusting for fertilizer N use efficiency.

Location	Legume	Corn Yield Response	
		Grain	Aboveground Biomass
		--------- Adjusted FRV (kg N ha^{-1}) --------	
EL	Alfalfa+	50	61
	Red clover	40	61
	Hairy vetch	50	61
KBS	Alfalfa+	10	13
	Red clover	16	27
	Hairy vetch	15	20

twice those from when using the ^{15}N method for all legumes at both locations (except alfalfa at KBS where results were similar). This is an even larger discrepancy than reported by Fox et al. (1990) where the difference and ^{15}N methods were measured in the greenhouse. Similar to with estimating fertilizer N contributions discussed earlier, the discrepancy between these two methods is probably due to overestimation by the difference method related to differences in soil N uptake with N fertilization. The discrepancy is not due to underestimations by the ^{15}N method due to MIT effects, again, as proven by results from the modified ^{15}N method.

CONCLUSIONS

The effect of mineralization-immobilization-turnover (MIT) on recovery of fertilizer and legume ^{15}N by crops should be measured using modified ^{15}N methods, such as the one used in this study, in order to estimate the true N contribution from these sources. Although we did not find a significant MIT effect using this method in short-term crop rotations, we did measure a MIT effect on recovery of fertilizer and legume ^{15}N by corn in the long-term cropping systems experiment at Rodale (data no shown). This indicated that it was possible to measure MIT effects on 15N recovery using our modified method. At Rodale, a fertilizer N contribution of 12 kg ha^{-1} and a legume N contribution of 23 kg ha^{-1} to corn due to MIT effects was measured and would be added to recovery of fertilizer and legume N by the traditional ^{15}N method to give a total N contribution due to applying the N source.

In our study, the difference method overestimated the fertilizer N contribution to corn due to increased soil N uptake by corn with increased fertilizer N rates. The modified ^{15}N method on the other hand accurately estimated the fertilizer N contribution to corn, proving it was not an underestimate due to MIT effects. In addition, results from the modified ^{15}N method indicated that the increase in soil N uptake by corn with increased fertilizer N rates was not due to "priming action". The linear regression methods (both isotopic and nonisotopic) of measuring fertilizer N uptake gave variable results and are not recommended for estimating fertilizer N contributions to crops.

Using a non-N_2-fixing legume to measure non-N rotation effects on corn yields gave much higher estimates than when using the more traditional N response method. Since the non-N_2-fixing legume should provide all the benefits of rotation except for those related to biologically fixed N, this method should be more accurate. In addition, the N response method of measuring non-N effects is probably subject to errors due to increased soil N uptake by crops with increased fertilizer rates similar to the difference method of measuring fertilizer N contributions. However, more data using this non-N_2-fixing legume method are needed to verify its usefulness in estimating non-N rotation effects.

A comparison of the FRV and ^{15}N methods of measuring legume N contributions revealed that FRV methodology gives higher estimates due to measuring N + non-N effects compared to the ^{15}N method which measures only N contributed from the legume plant material. The ^{15}N method gives an accurate measure of actual legume N recovered by crops, especially if priming and MIT effects are also measured. The FRV value method still gives a practical estimate of legume contribution in terms of a fertilizer N application, and if the use efficiency of fertilizer N is taken into account may give results comparable to the ^{15}N method. Measuring the amount of total N in legume biomass before incorporation may be useful to estimate total N contributed to cropping systems for purposes of calculating soil N balances but is probably not a good method of estimating actual amounts of legume N recovered by subsequent crops. Also, measuring N_2-fixation by legumes allows legume N contribution estimates to be based on N inputs from outside the system and not also soil N being recycled through the system.

REFERENCES

Allison, F.E. 1966. The fate of nitrogen applied to soils. Adv. Agron. 18:219-258.

Baldock, J.O., R.L. Higgs, W.H. Paulson, J.A. Jackobs, and W.D. Shrader. 1981. Legume and mineral N effects on crop yields in several crop sequences in the Upper Mississippi Valley. Agron. J. 73:885-890.

Barnes, D.K, G.H. Heichel, C.P. Vance, and R.N. Peadon. 1990. Registration of 'Ineffective Agate' and 'Ineffective Saranac' non-N_2-fixing alfalfa germplasms. Crop Sci. 30:752-753.

Boller, B.C., and G.H. Heichel. 1983. Photosynthate partitioning in relation to N_2-fixation capability of alfalfa. Crop Sci. 23:655-659.

Bruulsema, T.W., and R.B. Christie. 1987. Nitrogen contribution to succeeding corn from alfalfa and red clover. Agron. J. 79:96-100.

Fribourg, H.A., and W.V. Bartholomew. 1956. Availability of nitrogen from crop residues during the first and second season after application. Soil Sci. Soc. Amer. proc. 20:505-508.

Fried, M, and H. Broeshart. 1974. Priming effect of nitrogen fertilizers on soil nitrogen. Soil Sci. Soc. Amer. Proc. 38:858.

Fried, M, and V. Middelboe. 1977. Measurement of amount of nitrogen fixed by a legume crop. Plant Soil 47:713-715.

Fox, R.H., R.J.K. Meyers, and I. Vallis. 1990. The nitrogen mineralization rate of legume residues in soil as influenced by their polyphenol, lignin, and nitrogen contents. Plant Soil 129:251-259.

Fox, R.H., and W.P. Piekielek. 1988. Fertilizer N equivalence of alfalfa, birdsfoot trefoil, and red clover for succeeding corn crops. J. Prod. Agric. 1:313-317.

Griffin, T.S., and O.B. Hesterman. 1989. Effect of forage legume species and harvest management on nitrogen contributions in legume-potato rotations. p. 68-72. *In* Proc. Am. Forage and Grassland Council, Guelph, Ontario. 22-25 May 1989. Am. Forage and Grassland Council, Belleville, PA.

Griffin, T.S., and O.B. Hesterman. 1991. Potato response to legume and fertilizer nitrogen sources. Agron. J. 83:1004-1012.

Harmsen, G.W., and G.J. Kolenbrander. 1965. Soil inorganic nitrogen. *In* W.V. Bartholomew and F.E. Clark (ed.) Soil nitrogen. Agronomy Monograph No. 10:43-92. ASA, Madison, WI.

Harmsen, K., and J.T. Moraghan. 1988. A comparison of the isotope recovery and difference methods for determining nitrogen fertilizer efficiency. Plant Soil. 105:55-67.

Harris, G.H., and O.B. Hesterman. 1990. Quantifying the nitrogen contribution from alfalfa to soil and two succeeding crops using nitrogen-15. Agron J. 82:129-134.

Hauck, R.D. 1971. Quantitative estimates of nitrogen-cycle processes. Concepts and reviews. p. 65-80 *In* Nitrogen-15 in soil-plant studies. Vienna, Austria, International Atomic Energy Agency.

Hauck, R.D., and J.M. Bremner. 1976. Use of tracers for soil and fertilizer nitrogen research. Adv. Agron. 28:219-266.

Hesterman, O.B. 1988. Exploiting forage legumes for nitrogen contribution in cropping systems. p. 155-166 In J.F. Powers (ed.). Cropping strategies for efficient use of water and nitrogen. ASA Spec. Publ. 51.

Hesterman, O.B., T.S. Griffin, P.T. Williams, G.H. Harris, and D.R. Christenson. 1992. Forage legume-small grain intercrops: nitrogen production and response of subsequent corn. J. Prod. Agric. 5:340-348.

Hesterman, O.B., C.C. Sheaffer, D.K. Barnes, W.E. Leuschen, and J.H. Ford. 1986. Alfalfa dry matter and nitrogen production, and fertilizer nitrogen response in legume-corn rotations. Agron. J. 78:19-23.

Jansson, S.L. 1958. Tracer studies on nitrogen transformations in soil with special attention to mineralization-immobilization relationships. Kungl. Lantbrukshogskolans Ann. 24:101-361.

Jansson, S.L. 1971. Use of ^{15}N in studies of soil nitrogen. p. 129-166 *In* Soil A.D. McLaren and J. Skujins (ed.) Soil biochemistry, Vol. 2. New York, Marcel Dekker.

Jansson, S.L., and J. Persson. 1982. Mineralization and immobilization of soil nitrogen. p. 229-252 *In* Nitrogen in agricultural soils. ASA Agron. Monogr. 22. ASA. Madison, WI.

Jenkinson, D.S., R.H. Fox, and J.H. Rayner. 1985. Interactions between fertilizer nitrogen and soil nitrogen -- the so-called priming effect. J. Soil Sci. 36:425-444.

McAuliffe, C. D.S. Chamblee, H. Uribe-Arango, and W.W. Woodhouse. 1958. Influence of inorganic nitrogen on nitrogen fixation by legumes as revealed by ^{15}N. Agron. J. 50:334-337.

Norman, R.J., J.T. Gilmour, and B.R. Wells. 1990. Mineralization of nitrogen from nitrogen-15 labeled crop residues and utilization by rice. Soil Sci. Soc. Am. J. 54:1351-1356.

Pierce, F.J, and C.W. Rice. 1988. Crop rotation and its impact on efficiency of water and nitrogen use. p. 21-42 *In* W.L. Hargrove (ed.) Cropping strategies for efficient use of water and nitrogen. ASA Spec. Publ. No. 51. ASA, Madison, WI.

Russelle, M.P, O.B. Hesterman, C.C. Sheaffer, and G.H. Heichel. 1987. Estimating nitrogen and rotation effects in legume-corn rotations. p. 41-42 *In* J.F. Power (ed.) The role of legumes in conservation tillage systems. Proc. Soil Conserv. Soc. Am., Athens, GA. 27-29 Apr. 1987. Soil water Conserv. Soc. Am., Ankeny, IA.

Westerman, R.L., and L.T. Kurtz. 1974. Isotopic and nonisotopic estimations of fertilizer nitrogen uptake by sudangrass in field experiments. Soil Sci. Soc. Amer. Proc. 38:107-109.

CHAPTER FOUR

NITRATE LEACHING FROM ANIMAL-, LEGUME-, AND FERTILIZER-BASED CROPPING SYSTEMS

ABSTRACT

The strategy of replacing inorganic fertilizer nitrogen (N) inputs with organic N sources such as animal manures and forage legumes in sustainable agricultural systems has not been properly evaluated in respect to the potential for environmental pollution, particularly nitrate-N (NO_3-N) leaching. The objective of this research was to measure and compare NO_3-N leaching from animal-, legume-, and fertilizer-based cropping systems. Undisturbed, natural drainage lysimeters (0.76 dia. by 1.0 m deep) were installed in a long-term experiment located at the Rodale Institute Research Center in eastcentral Pennsylvania. Nitrate-N leaching was measured during a 2-yr period from cropping systems that used cattle manure or red clover (*Trifolium pratense* L.) green manure as N sources compared to a conventional system where commercial fertilizer N was used. Lysimeters (0.3 m dia. by 0.56 m deep) were also installed at two locations in Michigan to measure NO_3-N leaching under corn following alfalfa (*Medicago sativa* L.) hay, red clover hay, hairy vetch (*Vicia villosa* L.) green manure, and first-yr corn (*Zea mays* L.). ^{15}N-labeled legume and fertilizer materials were applied to lysimeters at all three locations to determine the amount of NO_3-N leached that was derived from these sources and from soil organic matter.

Most NO_3-N leaching occurred during winter months from all cropping systems at all locations. The NO_3-N concentrations in leachate collected at Rodale exceeded the 10 mg l^{-1} safety limit for certain crop/N input/climate combinations in each of the three systems studied. Large amounts (up to 34 kg ha^{-1} in one overwinter period) of NO_3-N were leached from the animal-based system after a manure application, but then small amounts of NO_3-N (less than 2 kg ha^{-1}) were leached after hay was established. Fertilizer N seemed most susceptible to loss during the wet growing season of 1992, when up to 32 kg NO_3-N ha^{-1} was leached. A 16 kg ha^{-1} "flush" of NO_3-N leached from the legume-based system after incorporating red clover in 1992. This value is still lower than other reports of NO_3-N leaching after incorporating legumes and less than the amount of NO_3-N leached from the fertilizer-based system during the same sampling period. Results using ^{15}N indicated that only small amounts of both legume and fertilizer N were leached during the first 12 months after application and most of the N leached in both of these systems was derived from soil organic matter. We concluded that animal manure, legume, and fertilizer N sources must all be managed carefully, year-round, with special attention to minimizing NO_3-N leaching during winter months. Avoiding excess NO_3-N in soil in autumn months and use of winter cover crops is recommended.

INTRODUCTION

Leaching of nitrate-nitrogen (NO_3-N) from agricultural soils is known to contribute to the degradation of groundwater quality (Goodrich, 1991; Hallberg, 1987, Keeney, 1986). Developing strategies that reduce or eliminate this potential source of pollution is a national research priority (USDA-ARS, 1988). Since excessive use of fertilizer N has been linked with NO_3-N levels in groundwater above the maximum limit considered safe for humans (Fletcher, 1991), improving the use efficiency of fertilizer N by crops to reduce NO_3-N leaching from this source is one strategy that has been well researched (EAS, 1988; IAEA, 1980).

Including legumes in crop rotations is another strategy recommended to reduce NO_3-N pollution of groundwater from agricultural systems (CAST, 1985; Papendick, 1987; Peterson and Russelle, 1991; Smith et al., 1990). Legume N is thought to be less susceptible than fertilizer N to leaching losses because, as an organic source, legume N is initially less readily available (USDA, 1980; Magdoff, 1991; Smith et al., 1987). Legumes, however, have not been properly evaluated with respect to their potential for environmental pollution, especially NO_3-N leaching (Keeney, 1982; NAS, 1989; Peterson and Russelle, 1990).

Limited information is available comparing the potential for NO_3-N leaching from fertilizer- and legume-based cropping systems. Based on NO_3-N accumulation in the soil profile, Hensler and Attoe (1970) reported greater leaching potential with

continuous corn than a corn-oat-meadow-meadow rotation. Using an ecosystems approach where leaching losses were measured directly, Long and Hall (1987) reported leaching losses of 18 kg N ha^{-1} y^{-1} for fertilized barley compared to 1 kg N ha^{-1} y^{-1} for unfertilized alfalfa. While this evidence suggests that little NO_3-N leaching occurs when legumes are actively growing, significant NO_3-N leaching losses have been measured when a legume stand is destroyed. Low and Armitage (1970) reported an average of 30 kg ha^{-1} y^{-1} NO_3-N leached from lysimeters during the growth of white clover, but 131 kg ha^{-1} y^{-1} after the clover winterkilled. Measuring NO_3-N leaching indirectly, based on soil nitrate levels, Adams and Pattinson (1985) estimated that 10 kg ha^{-1} y^{-1} of NO_3-N was lost by leaching during the growth of white clover, followed by 90 kg ha^{-1} y^{-1} when the legume stand was plowed and peas were planted. Robbins and Carter (1980), also using an indirect method, reported that unfertilized corn lost 60 and 17 kg NO_3-N ha^{-1} the first and second years, respectively, after plowing under alfalfa.

Mineralized soil N can contribute to nitrate contamination of groundwater in both fertilizer- and legume-based systems (Smith et al., 1990). Powlson (1988) reported on ^{15}N studies with winter wheat and showed that most of the nitrate subject to leaching overwinter is derived from mineralization of organic soil N rather than unused fertilizer nitrogen. By applying ^{15}N-labeled fertilizers and legume plant material to lysimeters and then analyzing the NO_3-N in the collected leachate for ^{15}N, the amount of leached N from these sources and N mineralized from soil organic matter can be distinguished. A number of lysimeter studies have been conducted to trace fertilizer ^{15}N into groundwater, usually when the fertilizer is applied at a rate

above the economic optimum. For example, Chichester and Smith (1978) traced the fate of ^{15}N labeled $Ca(NO_3)_2$ applied to lysimeters at a rate equivalent to 336 kg N ha^{-1}. After three years of continuous corn, they reported that 30 % of the fertilizer N had been leached out of the corn root zone. This accounted for 25 % of the total N leached, the other 75 % presumably came from mineralization of soil organic matter N. Muller (1987) applied ^{15}N-labeled subterranean clover plant material to lysimeters that were placed in the field and cropped to barley. After one year, 2 % of the added clover N was recovered in leachate, which accounted for 5 % of the total N leached.

Animal manure is also considered a large potential source of N contributing to the degradation of surface and groundwater quality (Keeney and Follet, 1991). While runoff of N from areas where animal manures are concentrated, such as feedlots, has been measured, NO_3-N leaching from animal-based cropping systems where animal manure is spread onto and often incorporated into the soil as an N source for subsequent crops needs to be investigated. Keeney (1982) pointed out that almost no data on leaching losses from organic systems, where legumes and/or animal manure are used as N sources instead of fertilizer, have been reported. Magdoff (1991) stated that although it is clear that high rates of either organic materials or synthetic N fertilizers have substantial potential for NO_3-N leaching losses, few experiments have been designed to compare the leaching potential of N from organic and synthetic sources when both supply optimal amounts of available N.

The objectives of this study were to measure NO_3-N leaching from 1) animal-, legume-, and fertilizer-based cropping systems in a long-term experiment and 2) from short-term (2-yr) legume- and fertilizer-based crop rotations. As a third objective, the

amount of NO_3-N leached from legume and fertilizer N sources in both the long- and short-term studies was measured using ^{15}N.

MATERIALS AND METHODS

The long-term experiment. Thirty-six intact, natural-drainage lysimeters were installed in the Rodale Farming Systems Trial experiment, located in eastcentral Pennsylvania, during the week of October 29, 1990. The long-term cropping experiment was established in 1981 to compare organic cropping systems (with or without animals) to a conventional corn-soybean rotation where chemical fertilizers and pesticides were used. Three cropping systems consisting of five-year rotations were compared. The low-input/cash grain (LIP-CG) system relied on forage legume (mainly red clover) green manure as an N source and produced a grain crop (either corn, soybean or a small grain) each year. The low-input/ animal (LIP-A) system had one plow-down of red clover hay plus approximately 200 kg N ha^{-1} in animal manure applied before each of two corn crops during the five-year cycle which also included soybean and small grain crops. The conventional system consisted of three corn and two soybean crops in five years. Each corn crop received 130 kg N ha^{-1} of inorganic fertilizer and no N was applied to the soybeans. Three entry points in the five-year rotation of each cropping system were included in the study to give different crop sequence/N application combinations each year. Management practices and results from 1981-1985 for this study have been reported by Liebhardt et al. (1989) and for 1986-1990 by Peters et al. (1992).

Lysimeters were installed in each of three entry points in four replications of

the low-input/animal (LIP-A), low-input/cash grain (LIP-CG), and conventional (CONV) cropping systems (4 replications x 3 entry points x 3 cropping systems = 36 lysimeters). Each lysimeter measured 0.76 m dia by 1.0 m deep and was constructed from 0.79 cm steel well casing. Lysimeters in two of the three entry points of the LIP-CG (Entry Points 2 and 3) and CONV (Entry Points 1 and 2) systems extended 5 cm above the soil surface to facilitate labeling with ^{15}N legume and fertilizer materials, respectively. All other lysimeters extend to 0.3 m below the soil surface to allow normal tillage operations. All lysimeters were inserted to a 1.0 m depth using a fabricated pile driver. Topsoil (0-30 cm) was excavated and placed aside before the insertion of lysimeters that did not extend to the soil surface. Little, if any, compaction of soil inside the lysimeters due to the insertion process was observed. After insertion, the lysimeters were pulled up using a track loader and placed on a 0.64 cm thick steel plate. The steel plate, previously fitted with a drain located in the center, was then welded to the bottom of the well casing. A small amount (approximately 2 kg) of clean #2 gravel was placed between the drain and the soil column before the plate was welded to the lysimeter to prevent the drain from becoming clogged with soil. Tubing (12.7 mm dia. food grade with 3.2 mm wall thickness) was connected to the drain and run to a 20 liter polyethylene carboy. An access tube that extended to the soil surface for leachate collection was also connected to the carboy and the lysimeter and carboy were lowered back into the ground. The carboy was guided into a hole dug adjacent to and lower than the bottom of the lysimeter to allow passive drainage of leachate from the lysimeter. Topsoil was placed back on top of lysimeters that did not extend to the soil surface after

installation.

All lysimeters were planted according to the normal cropping sequence (see Table I.2) and managed as the subplots of the main experiment. Tillage operations and the planting of crops in lysimeters that extended to the soil surface were done by hand. In May 1991, lysimeters in Entry Point 3 of the LIP-CG system and Entry Point 1 of the CONV system received ^{15}N labeled red clover and ammonium sulphate, respectively, both at a rate equivalent to 124 kg N ha^{-1}. Labeling procedures were identical to those described in Chapter 1. Leachate was collected from each lysimeter twice a month (near the beginning and middle) from November 1990 through October 1992 using an electric pump connected to the access tube. The leachate volume was recorded and a subsample was taken and stored at 4 °C until analyzed for NO_3-N. Nitrate-N concentration in the leachate was determined using a Lachat flow-injector autoanalyzer. The ^{15}N content of NO_3-N from labeled lysimeters was determined by mass spectrometry after diffusion onto a filter paper disk (Brooks et al., 1989).

The design for this experiment was a randomized complete block, split-plot with four replications. Cropping systems were main plots and rotation entry points were subplots. Results for the amount of leachate collected, flow-weighted NO_3-N concentration in leachate, and the amount of NO_3-N leached during the 1990-91 and 1991-92 overwinter periods (Nov through Apr) and the 1991 and 1992 growing seasons (May through Oct) were analyzed using ANOVA procedures. Based on the ^{15}N data, the percent of leachate N derived from fertilizer and legume plant material, the amount of fertilizer and legume N leached, and recovery of legume and fertilizer N in leachate were analyzed for the 1991 growing season and the subsequent

overwinter period (Nov '91 through Apr '92).

The short-term rotations. Thirty-two, natural drainage lysimeters were installed during the week of May 7, 1991 in the short-term crop rotation experiment conducted at EL and KBS described in Chapter 3. Lysimeters were installed in the same area where the modified ^{15}N experiment was conducted, following first-year crops of 'Saranac' alfalfa, medium red clover, 'common' hairy vetch, and corn grown in 1990 (4 first-year crops x 4 replications x 2 locations = 32 lysimeters). Each lysimeter measured 0.30 m dia by 0.56 deep and was made of schedule 40 polyvinylchloride (PVC). The lysimeters were inserted into the soil using a hydraulic cylinder fitted on the three-point hitch of a tractor and pulled up using a front-end loader. The lysimeters were then inverted and the bottom 5 cm of soil was removed and replaced with acid- washed pea stone. A false bottom, made of a 15 cm tall section of PVC with perforated (approximately 20, 4 mm dia holes) PVC sheeting at the top and solid sheeting on the bottom was then glued and caulked to the inverted lysimeter. A 1.0 cm hole was drilled into the side of the false bottom at the lowest possible level and a rigid elbow was inserted, glued and caulked. Rigid tubing was then connected to the elbow to extend to the soil surface for sampling access. The lysimeters, with false bottom and access tube attached, were then lowered back into the ground.

^{15}N-labeled alfalfa, red clover and hairy vetch were applied to lysimeters in plots that had been sown to these crops in 1990. ^{15}N-labeled ammonium sulphate was applied to lysimeters in plots where corn was grown in 1990. Alfalfa and red clover plant material was applied at a rate equivalent to 112 kg N ha^{-1}, hairy vetch was applied at a rate equivalent to 68 kg N ha^{-1}, and $(NH_4)_2SO_4$ fertilizer was applied at a

rate equivalent to 150 kg N ha^{-1}. Labeling procedures were again identical to those used in Chapter 1. Two corn seeds were planted in each lysimeter and subsequently thinned to one plant soon after emergence. Leachate was collected from each lysimeter once a month (at the end of each month) using an electric pump connected to the access tube. Leachate volume was recorded and a subsample was taken and analyzed by the same methods used for leachate from the long-term experiment described above.

The design for this experiment was a one-factor (first-year crop) randomized complete block, with four replications. Results for the amount of leachate collected, flow-weighted NO_3-N concentration in leachate, the amount of NO_3-N leached, and leaching of fertilizer and legume ^{15}N for the 1991 growing season and the 1991-92 overwinter period were analyzed using ANOVA procedures.

RESULTS

The Long-term Experiment

Leachate collected. There was no significant effect of cropping system or entry point on the amount of leachate collected from the lysimeters during any sampling period or the 2-yr total (Table 4.1). More leachate, however, was collected during the two overwinter periods (mean = 179 mm) compared to the two growing seasons (mean = 70 mm). More leachate was also collected from the lysimeters during the 1992 growing season (mean = 116 mm) compared to the 1991 growing season (mean = 24 mm). In addition, the amount of leachate collected from the lysimeters was more variable during the 1991 than the 1992 growing season, as indicated by the higher coefficient of variation (128 vs. 52 %). Variability in the amounts of leachate collected was lowest during the two overwinter periods (average CV = 42 %). The amounts of leachate collected accounted for 50 % of the amount of precipitation received during the 1990-91 overwinter, 4 % during the 1991 growing season, 40 % during the 1991-92 overwinter, and 15 % during the 1992 growing season (precipitation data not shown).

Leachate NO_3-N Concentrations Cropping systems had a significant effect on flow-weighted NO_3-N concentration in leachate collected from the lysimeters during the 1991 and 1992 growing seasons (Table 4.2). Averaged over entry points, the NO_3-N concentration in leachate from the LIP-A system during the 1991 growing

Table 4.1. The effect of cropping system and entry point on the amount of leachate collected from lysimeters in the long-term cropping systems experiment at Rodale during four sampling periods.

Cropping system (CS)	Entry point (EP)	Sampling period† 1990-91 OW	1991 GS	1991-92 OW	1992 GS
		---------- Leachate (mm) ----------			
LIP-A	1	199	63	157	53
	2	153	8	151	72
	3	267	32	132	139
LIP-CG	1	260	14	108	87
	2	231	45	200	160
	3	232	5	150	130
CONV	1	246	12	168	213
	2	161	24	160	126
	3	149	17	101	67
Significance					
CS		NS	NS	NS	NS
EP		NS	NS	NS	NS
CS x EP		NS	NS	NS	**
CV(%)		41	128	44	52

*,**,*** Significant at the 0.05, 0.01, and 0.001 probability levels, respectively; NS = not significant at P = 0.05.

†OW = overwinter (Nov - Apr), GS = growing season (May - Oct).

Table 4.2. The effect of cropping system and entry point on flow-weighted NO_3-N concentration in leachate collected from lysimeters in the long-term cropping systems experiment at Rodale during four sampling periods.

Cropping system (CS)	Entry point (EP)	Sampling Period† 1990-91 OW	1991 GS	1991-92 OW	1992 GS
		--------- NO_3-N Concentration (mg l^{-1}) ---------			
LIP-A	1	7.6	9.2	3.1	1.5
	2	8.0	10.7	9.8	10.6
	3	13.1	18.4	7.0	1.1
LIP-CG	1	3.1	4.0	2.8	1.8
	2	7.4	6.4	3.7	11.2
	3	6.1	3.1	7.2	7.3
CONV	1	7.3	5.3	6.8	13.4
	2	5.7	5.4	10.1	13.8
	3	3.5	9.6	21.6	20.6
Significance					
CS		*	*	*	*
EP		NS	NS	**	NS
CS x EP		**	NS	*	NS
CV(%)		32	53	52	71

*, **, *** Significant at the 0.05, 0.01, and 0.001 probability levels, respectively; NS = not significant at P = 0.05.

†OW = overwinter (Nov - Apr), GS = growing season (May - Oct).

season measured 12.8 mg l^{-1}. This value is above the 10 mg l^{-1} maximum concentration limit set by the U.S. Environmental Protection Agency (USEPA, 1986) and was significantly higher than the average concentrations from the LIP-CG and CONV systems which measured 4.5 and 6.8 mg l^{-1}, respectively (means separated by an LSD test, data not shown).

During the 1992 growing season, the flow-weighted NO_3-N concentration in leachate from the CONV system, averaged over entry points (15.9 mg l^{-1}), was above the USEPA safety limit and significantly greater than the average NO_3-N levels in the LIP-A and LIP-CG systems (4.4 and 6.7 mg l^{-1}, respectively). Nitrate-N concentrations in leachate were low (less than 2 mg l^{-1}) during this period in the LIP-A and LIP-CG systems when either red clover hay or a wheat/red clover interseeding was growing. The NO_3-N concentrations in leachate under corn or soybeans in the LIP-A and LIP-CG systems during the 1992 growing season were around the 10 mg l^{-1} standard safe limit and slightly lower than NO_3-N concentrations under corn or soybeans in the CONV system.

There was a significant cropping systems by entry point interaction for flow-weighted NO_3-N concentration in leachate collected during the two overwinter periods (Table 4.2). During the 1990-91 overwinter period, leachate NO_3-N concentrations ranged from 8 to 13 mg l^{-1} for the LIP-A system and from 3 to 7 mg l^{-1} for the LIP-CG and CONV systems.

During the 1991-92 overwinter period, leachate NO_3-N concentrations for the LIP-A and LIP-CG systems ranged from 1 to 11 mg l^{-1} whereas the CONV system measured from 13 to 21 mg l^{-1}. The NO_3-N concentration in Entry Point 3 of the

CONV system during this period (21 mg l^{-1}) was the highest observed for any given cropping system, entry point and sampling period combination.

Total NO_3-N Leached A cropping system by entry point interaction was indicated for the amount of NO_3-N leached during the 1990-91 overwinter period (Table 4.3) Between 14 and 34 kg NO_3-N ha^{-1} were leached from the LIP-A, 10 and 20 from the LIP-CG and 6 and 18 from the CONV during this period.

Only small amounts of NO_3-N were leached from the animal-, legume-, or fertilizer-based cropping systems during the 1991 growing season, ranging from 0.6 to 3.8 kg ha^{-1}. Like the amounts of leachate volume collected during this sampling period, the amounts of NO_3-N leached were small, highly variable (CV = 141) and not significantly different between cropping systems or entry points. The amounts of NO_3-N leached were greater during the 1991-1992 overwinter period (overall range = 3 to 25 kg ha^{-1}) than during the 1991 growing season but were also highly variable (CV = 92) and not significantly different between cropping systems and entry points.

The amounts of NO_3-N leached during the 1992 growing season varied significantly with cropping system and entry point. In the LIP-A system, NO_3-N leaching was low (2 kg ha^{-1} or less) when red clover hay or a wheat/hairy vetch interseeding were grown, but higher (10 kg ha^{-1}) under a soybean crop. In the LIP-CG system, NO_3-N was low under a wheat/hairy vetch interseeding (less than 3 kg ha^{-1}), but higher under soybean (11 kg ha^{-1}) and corn (16 kg ha^{-1}). Nitrate-N leaching during the 1992 growing season was greatest from the CONV system, ranging from 15 kg ha^{-1} under soybean to 32 kg ha^{-1} under corn.

Table 4.3. The effect of cropping system and entry point on the amount of NO_3-N leached from lysimeters in the long-term cropping systems experiment at Rodale during four sampling periods.

Cropping system (CS	Entry point (EP)	Sampling Period† 1990-91 OW	1991 GS	1991-92 OW	1992 GS
		---------- NO_3-N leached (kg ha^{-1}) ----------			
LIP-A	1	14.6	3.8	5.9	0.7
	2	14.4	0.6	15.5	9.7
	3	33.9	2.7	8.4	2.0
LIP-CG	1	10.3	0.9	3.4	2.6
	2	20.2	2.2	7.0	16.4
	3	13.1	0.1	12.5	10.8
CONV	1	18.0	0.8	11.8	32.3
	2	9.9	1.4	17.0	19.5
	3	5.8	1.9	24.6	15.4
Significance					
CS		NS	NS	NS	*
EP		NS	NS	NS	NS
CS x EP		***	NS	NS	**
CV(%)		40	141	79	92

*, **, *** Significant at the 0.05, 0.01, and 0.001 probability levels, respectively; NS = not significant at P = 0.05.

†OW = overwinter (Nov - Apr), GS = growing season (May - Oct).

Leaching of fertilizer and legume N. Although ^{15}N was detected in leachate collected during the 1991 growing season, negligible amounts of fertilizer or legume N were leached during this time period (Table 4.4). During the first overwinter period after the ^{15}N was applied (Nov '91- Apr '92), there was still very little N from legume or fertilizer leached. During the entire 12 month period after applying the legume and fertilizer ^{15}N, less than half a percent was recovered as NO_3-N in leachate, amounting to 0.5 kg ha^{-1} of legume and fertilizer N leached, and accounting for about 4 percent of the total N leached during this period.

The Short-term Rotations

First-year crop had no effect on leachate volume collected from lysimeters at EL (Table 4.5) or KBS (Table 4.6), although results were highly variable. Like in the long-term experiment, more percolate flowed through the lysimeters during the overwinter period than during the growing season. No leachate was collected from lysimeters in one replication at EL and two replications at KBS during the 1991 growing season. When this trend continued through the 1991-92 overwinter period, it was assumed that the lysimeters in these replications were either clogged or leaking. Therefore, the results for EL presented in Table 4.5 are based on data from 3 replications and results for KBS in Table 4.6 are based on 2 replications.

The flow-weighted NO_3-N concentration of leachate collected during the 1991-92 overwinter period at EL (Table 4.5) were significantly higher for corn following alfalfa than corn following red clover, hairy vetch, or corn (determined by an LSD test, data not shown). The level of NO_3-N in leachate following alfalfa during these

Table 4.4. Recovery of ^{15}N from red clover and ammonium sulphate sources in leachate collected from the LIP-CG and CONV systems of the long-term cropping systems experiment at Rodale during the 1991 growing season and 1991-92 overwinter period.

Cropping system	Entry point	Leachate-N derived from source (%)	Leachate-N derived from source (kg ha^{-1})	Recovery of ^{15}N in leachate (% of input)
		---------- 1991 growing season (May - Oct) ---------		
LIP-CG	3	3.0	0.003	0.002
CONV	1	1.2	0.018	0.015
	Significance	NS	NS	NS
	CV(%)	107	171	153
		---------- 1991-92 overwinter (Nov - Apr)----------		
LIP-CG	3	4.4	0.56	0.45
CONV	1	3.8	0.46	0.37
	Significance	NS	NS	NS
	CV(%)	50	32	33

*, **, *** Significant at the 0.05, 0.01, and 0.001 probability levels, respectively; NS = not significant at P = 0.05.

Table 4.5. The effect of first-year crop on the amount of leachate, flow-weighted NO_3-N concentration of leachate, and amount of NO_3-N leached from lysimeters in short-term rotations at East Lansing during two sampling periods.

First-year crop	Sampling period†	
	1991 growing season	1991-92 overwinter
	---------- Leachate (mm) ----------	
Alfalfa	11	305
Red clover	72	383
Hairy vetch	41	541
Corn	129	584
Significance	NS	NS
CV(%)	99	30
	-- Flow-weighted NO_3-N concentration (mg l^{-1}) --	
Alfalfa	4.1	13.5
Red clover	5.7	8.9
Hairy vetch	8.0	7.1
Corn	2.6	5.8
Significance	NS	*
CV(%)	113	25
	---------- NO_3-N Leached (kg ha^{-1}) ----------	
Alfalfa	0.3	40.7
Red clover	1.1	28.7
Hairy vetch	4.3	37.2
Corn	3.1	32.6
Significance	NS	NS
CV(%)	90	37

*, **, *** Significant at the 0.05, 0.01, and 0.001 probability levels, respectively; NS = not significant at P = 0.05.

† growing season = May - Oct, overwinter = Nov - Apr.

Table 4.6. The effect of first-year crop on the amount of leachate, flow-weighted NO_3-N concentration of leachate, and amount of NO_3-N leached from lysimeters in short-term rotations at the Kellogg Biological Station during two sampling periods.

First-year crop	Sampling period: 1991 growing season	Sampling period: 1991-92 overwinter
	Leachate (mm)	
Alfalfa	124	461
Red clover	106	457
Hairy vetch	80	382
Corn	312	331
Significance	NS	NS
CV(%)	124	65
	Flow-weighted NO_3-N concentration (mg l^{-1})	
Alfalfa	5.7	4.6
Red clover	3.3	5.0
Hairy vetch	1.6	3.5
Corn	1.9	0.9
Significance	NS	NS
CV(%)	30	30
	NO_3-N Leached (kg ha^{-1})	
Alfalfa	7.1	21.3
Red clover	3.1	7.1
Hairy vetch	1.1	13.8
Corn	4.4	4.0
Significance	NS	NS
CV(%)	83	72

*, **, *** Significant at the 0.05, 0.01, and 0.001 probability levels, respectively; NS = not significant at P = 0.05.

† growing season = May - Oct, overwinter = Nov - Apr.

periods (13 mg l^{-1}) exceeded the 10 mg l^{-1} safety limit. First-year crops had no effect on flow weighted NO_3-N concentrations in leachate collected during the 1991 growing season and 1991-92 overwinter period at KBS (Table 4.6).

The amounts of NO_3-N leached from the legume- and fertilizer-based rotations during the 1991 growing season and during the 1991-92 overwinter period at EL were not significantly different (Table 4.5). There was also no significant difference in the amounts of NO_3-N leached from the short-term legume- and fertilizer-based crop rotations at KBS (Table 4.6).

Negligible amounts of legume and fertilizer ^{15}N were leached from the short-term rotations during the first growing season at EL and KBS. Also, only small amounts of legume and fertilizer ^{15}N were recovered in leachate during the first overwinter period at both EL and KBS (Table 4.7). At EL, leachate-N derived from source (legume or fertilizer) during the overwinter period averaged 6 %, which amounts to about 2 kg N ha^{-1} leached and 2 % recovery of what was applied. During the overwinter period at KBS, 8 % of the NO_3-N leached was derived from legumes or fertilizer, 1 kg N ha^{-1} legume and fertilizer N was leached, and 0.8 % of the legume and fertilizer N applied was recovered in leachate.

Table 4.7. Recovery of legume and fertilizer ^{15}N in leachate collected from lysimeters in short-term rotations at East Lansing (EL) and the Kellogg Biological Station (KBS) during the 1991-92 overwinter period.

Location	First-year crop	Leachate-N derived from source	Leachate-N derived from source	Recovery of ^{15}N in leachate
		------ % ------	--- kg ha^{-1} ---	-- % of input --
EL	Alfalfa	6.5	2.4	2.0
	Red clover	5.9	1.5	1.2
	Hairy vetch	7.8	2.4	1.9
	Corn	4.0	1.2	0.8
	Significance	NS	NS	NS
	CV(%)	90	53	52
KBS	Alfalfa	9.8	2.0	1.3
	Red clover	0.0	0.8	0.6
	Hairy vetch	10.4	1.8	1.3
	Corn	2.7	0.1	0.03
	Significance	NS	NS	NS
	CV(%)	27	88	95

*, **, *** Significant at the 0.05, 0.01, and 0.001 probability levels, respectively; NS = not significant at P = 0.05.

DISCUSSION

The amount of NO_3-N leached from a cropping system is dependent on both the amount of water that percolates through the soil profile and the NO_3-N concentration of the soil solution (Legg and Meisinger, 1982). Water percolation alone, however, is sometimes used as an indicator of NO_3-N leaching potential (Williams and Kissel, 1991). In our experiments, the amount of leachate collected during each sampling period was similar among cropping systems. Therefore, based on water percolation alone, the NO_3-N leaching potential during each sampling period was similar for all cropping systems.

Based on water percolation alone, NO_3-N leaching is also more likely to take place during winter months than during summer months when evapotranspiration usually exceeds precipitation and plant uptake of water is high (Allison, 1973). Using monolith lysimeters, Chichester (1977) measured the highest NO_3-N leaching in winter months. We also collected more leachate during overwinter periods than growing seasons in our experiment. The NO_3-N leaching potential from all cropping systems in our experiment was therefore greater during winter than during the growing seasons.

Above-average rainfall during the 1992 growing season at the Rodale site probably led to the larger amounts of leachate collected from lysimeters during this sampling period compared to the 1991 growing season. Again, based on water

percolation alone, the NO_3-N leaching potential was greater during the 1992 than the 1991 growing season for all cropping systems at this site. The drier conditions of the 1991 growing season at Rodale also may have accentuated the variability in the amount of leachate collected from the lysimeters due to spatial variability. The low variability in the amounts of leachate collected at this site during the overwinter periods when larger amounts of leachate were collected supports this theory.

The amount of NO_3-N collected from lysimeters depends on both the volume and the NO_3-N concentration of the leachate collected. Since there was no difference in the amount of water collected from lysimeters in different cropping systems of our experiments during any given sampling period, any effect of cropping system on the amount of NO_3-N leached must have been due to differences in NO_3-N concentrations. Chichester (1977) reported similar results, where percolate volumes collected from lysimeters during the winter were essentially the same for different crop management practices and the amounts of NO_3-N leached depended on the NO_3-N concentration of the leachate.

Overall, nitrate-N concentrations in leachate collected from the long-term experiment at Rodale varied with cropping systems, entry points, and sampling periods. Originally, (during the first two sampling periods), NO_3-N concentrations were higher in leachate from the animal-based than the legume- or fertilizer-based cropping systems. An improperly-timed manure application in one of the entry points of the animal-based system is thought to have led to the average NO_3-N in leachate exceeding the EPA safety limit. Manure was applied in Entry Point 3 before corn silage in Spring 1989 following 2 years of red clover hay, and again in Spring 1991

before corn grain. When red clover hay was established in the animal-based cropping system during the next two sampling periods (1991-92 overwinter and 1992 growing season) the NO_3-N levels in leachate eventually dropped to less that 2 mg l^{-1}. A similar finding was reported by Owens (1990) where the NO_3-N concentration in leachate collected from lysimeters dropped from 15 mg l^{-1} or higher when corn was grown and fertilized with N to less than 5 mg l^{-1} under alfalfa with no fertilizer N applied. The NO_3-N concentration in leachate collected from the fertilizer-based system in our study, during the second two sampling periods, was greater than the other two systems and often exceeded the EPA limit. Leachate NO_3-N concentrations were especially high in the CONV systems during the growing season of 1992 and may have been related to the above-average precipitation received in May and June during this sampling period.

The NO_3-N concentration in leachate collected from the legume-based cropping system at Rodale exceeded the EPA limit during only one sampling period. This occurred during the 1992 growing season when red clover green manure was plowed down and corn was grown. In the short-term rotation experiment at KBS, concentrations of NO_3-N in leachate were higher in the legume- than fertilizer-based crop rotations. None of the NO_3-N concentrations however, exceeded the EPA limit. In the short-term rotations at EL, the NO_3-N concentrations in leachate following alfalfa did exceed the EPA limit and were higher than the other legume- or fertilizer-based cropping systems.

The amounts of NO_3-N leached from the cropping systems in our experiments followed the leaching potential predictions based on water percolation. More NO_3-N

was leached during the overwinter periods than growing seasons in both the short- and long-term studies, and more was leached during the 1992 than 1991 growing season at Rodale. Within sampling periods at Rodale, the amounts of NO_3-N leached also followed predictions based on NO_3-N concentrations in leachate. A large amount of NO_3-N (34 kg ha^{-1}) was leached during the 1990-91 overwinter period from the animal-based system entry point (Entry Point 3) that had the high NO_3-N concentration due possibly to mismanagement of manure N. During the 1992 growing season, 3 kg ha^{-1} or less NO_3-N was leached under legume hay or green manure crops in the animal- and legume-based systems and large amounts (15 to 32 kg ha^{-1}) were leached from the fertilizer-based system. The high NO_3-N concentration in leachate following the plowdown of red clover translated into 16 kg ha^{-1} NO_3-N leached from the legume-based system during the 1992 growing season. This value, however, may be higher than normally expected due to the abnormally wet conditions in 1992 and is still much lower than other reports of NO_3-N leaching following the plowdown of a legume stands (Adams and Pattinson, 1985; Low and Armitage, 1970; Robbins and Carter, 1980). In addition, this "flush" of NO_3-N leached following the plowdown of red clover in our study was the same or less than the amount of NO_3-N leached from the fertilizer-based system during the same sampling period.

Two percent or less of the legume and fertilizer ^{15}N applied to the short- and long-term cropping systems in this study were recovered in leachate collected during the first 12 months after ^{15}N application. Results from a previous ^{15}N study at the Rodale site revealed that up to 20 % of applied legume N and 45 % of applied fertilizer N was not accounted for in a corn crop or soil after 1 year (see Table 1.3).

This indicates that NO_3-N leaching is not the major mechanism of legume or fertilizer N loss in these cropping systems. Also, only 10 % or less of the NO_3-N collected in leachate during the first 12 months after ^{15}N application was derived from applied legume or fertilizer N. This result corroborates reports by others that most of the NO_3-N leached in fertilizer and legume-based cropping systems is derived from soil organic matter.

CONCLUSIONS

Certain crop sequence/N input/climate combinations in each of the three cropping systems at Rodale resulted in NO_3-N concentrations in leachate above the EPA maximum limit. Therefore, careful management of all three N sources, animal manure, legume, and fertilizer N, should be practiced. Management strategies similar to those used to minimize NO_3-N leaching from fertilizers, especially timing, amount, and method of application, may be useful for managing animal manure and legume N sources.

The guiding principle of best management practices for minimizing NO_3-N leaching from cropland is to avoid excess NO_3-N in soil at times when the NO_3-N is most vulnerable to leaching (Keeney, 1982). Our results support findings by others that most NO_3-N leaching occurs during winter months. Therefore, special attention should be paid to managing cropping systems year-round, using winter cereals, cover crops, or hay crops whenever possible.

REFERENCES

Adams, J.A. and J.M. Pattinson. 1985. Nitrate losses under a legume-based crop rotation in Central Canterbury, New Zealand. New Zealand J. Agric. Res. 28:101-107.

Allison, F.E. 1973. Soil organic matter and its role in crop production. Elsevier Scientific Publishing Co., New York.

Brooks, P.D., J.M. Stark, B.B. McInteer, and T. Preston. 1989. Diffusion method to prepare soil KCl extracts for automated N-15 analysis. Soil Sci. Soc. Am. J. 53:1707-1711.

Chichester. F.W. 1977. Effects of increased fertilizer rates on nitrogen content of runoff and percolate from monolith lysimeters. J. Environ. Qual. 6:211-233.

Chichester, F.W. and S.J. Smith. 1978. Disposition of ^{15}N-labeled fertilizer nitrate applied during corn culture in field lysimeters. J. Environ. Qual. 2:227-223.

Council for Agricultural Science and Technology. 1985. Agriculture and groundwater quality. Rpt. No. 103. Ames, Iowa.

Elsevier Applied Science. 1988. Nitrogen Efficiency in Agricultural Soils. D.S. Jenkinson and K.A. Smith (ed.).

Fletcher, D.A. 1991. A national perspective. p.9-17 *In* R.F. Follet et al. (ed.) Managing nitrogen for groundwater quality and farm profitability. SSSA, Inc. Madison, WI.

Goodrich, J.A., B.J. Lykins, Jr., and R.M. Clark. 1991. Drinking water from agriculturally contaminated groundwater. 20:707-717.

Hallberg, G.R. 1987. Agricultural chemicals in groundwater: extent and implications. Am. J. Altr. Agric. 2:3-15.

Hensler, R.F., and O.J. Attoe. 1970. Rural sources of nitrates in water. *In* V.L. Snoeyink (ed.) Nitrate and water supply: source and control. Proc. of the 12th Sanitary Engr. Conf. Urbana, Ill. 11-12 Feb. 1970. Univ. of Illinois.

International Atomic Energy Agency. 1980. Soil nitrogen as fertilizer or pollutant. Panel Proceedings Series. Brazil, 3-7 July 1988.

Keeney, D.R. 1982. Nitrogen management for maximum efficiency and minimum pollution. p. 605-649 *In* ASA Agron. Monogr. 22. ASA, Madison, WI.

Keeney, D. 1986. Sources of nitrate to groundwater. CRC Critical Resources in Environmental Control. 16:257-304.

Keeney, D.R., and R.F. Follet. 1991. Managing nitrogen for groundwater quality and farm profitability: overview and introduction. p. 1-7 *In* R.F. Follet, et al. (ed.) Managing nitrogen for groundwater quality and farm profitability. SSSA, Inc. Madison, WI.

Legg, J.O, and J.J. Meisinger. 1982. Soil nitrogen budgets. p. 503-566 *In* F.J. Stevenson (ed.) Nitrogen in agricultural soils. ASA Agron. Monogr. 22. ASA, Madison, WI.

Liebhardt, W.C., R.W. Andrews, M.N. Culik, R.R. Harwood, R.R. Janke, J.K. Radke, and S.L. Reiger-Schwartz. 1989 Crop production during conversion from conventional to low-input methods. Agron J. 81:150-159.

Long, S.P., and D.O. Hall. 1987. Nitrogen cycles in perspective. Nature. 329:584-585.

Low, A.J., and E.R. Armitage. 1970. The composition of leachate through cropped and uncropped soils in lysimeters to that of the rain. Plant Soil. 33:393-411.

Magdoff, F. 1991. Managing nitrogen for sustainable corn systems: problems and possibilities. Am. J. Altr. Agric. 6: 3-8.

Muller, M.M. 1987. Leaching of subterranean clover-derived N from a loam soil. Plant Soil. 102:185-191.

National Academy of Sciences. 1990. Alternative Agriculture. National Academy Press, Washington, D.C.

Olsen, R.J., R.F. Hensler, O.J. Attoe, S.A. Witzel, and L.A. Peterson. 1970. Fertilizer nitrogen and crop rotation in relation to movement of nitrate nitrogen through soil profiles. Soil Sci. Soc. Proc. 34:448-452.

Owens. L.B. 1990. Nitrate-nitrogen concentrations in percolate from lysimeters planted to a legume-grass mixture. J. Environ. Qual. 19:131-135.

Papendick, R.I., L.F. Elliot, and J.F. Power. 1987. Alternative production systems to reduce nitrates in groundwater. Am. J. Alter. Agric. 2:19-24.

Peters, S.E., R. Janke, and M. Bohlke. 1992. Rodale's farming systems trial 1986-1990. Rodale Institute, Rodale Press, Emmaus, PA.

Peterson, T.A. and M.P. Russelle. 1991. Alfalfa and the nitrogen cycle in the corn belt. J. Soil Water Conserv. 46:229-235.

Powlson, D.S. 1988. Measuring and minimizing losses of fertilizer nitrogen in arable agriculture. p. 231-245 *In* D.S. Jenkinson and K.A. Smith (ed.) Nitrogen efficiency in agricultural soils. Elsevier Applied Science. NY, NY.

Robbins, C.W. and D.L. Carter. 1980. Nitrate-nitrogen leached below the root zone during and following alfalfa. J. Environ. Qual. 9:447-450.

Smith, M.S., W.W. Frye, and J.J. Varco. 1987. Legume winter cover crops. Adv. Soil Sci. 7:95-139.

Smith, S.J., J.S. Shepers, and L.K. Porter. 1990. Assessing and managing agricultural nitrogen losses to the environment. Adv. Soil Sci. 14:1-43.

U.S. Department of Agriculture. 1980. Report and recommendations on organic farming. U.S. Government Printing Office, Washington, D.C.

U.S. Department of Agriculture, Agricultural Research Service. 1988. ARS Strategical groundwater plan. 2. Nitrate. Washington, D.C.

Williams, J.R., and D.E. Kissel. 1991. Water percolation: an indicator of nitrogen-leaching potential. p.59-83 *In* R.F. Follet et al. (ed.) Managing nitrogen for groundwater quality and farm profitability. SSSA, Inc. Madison, WI.

RECOMMENDATIONS

Overall conclusions from results of this dissertation are that N cycles differently in animal-, legume-, and fertilizer-based cropping systems, but these systems have near equal potential for polluting the environment with N. In addition, information gained on how animal manure, legume, and fertilizer N sources cycle differently in cropping systems can be used to adjust management practices in order to minimize adverse environmental impacts. For example, the high soil N supplying capacity of the animal-based system measured in Chapter 2, coupled with observations in Chapter 4 of high NO_3-N leaching from this system, suggests that applying manure immediately following legume hay crops should be avoided. For legume-based systems, although more legume N may be contributed to soil than to crops during the year of application, high losses of residual legume N in subsequent years (Chapter 1) indicates the need for strategies to retain this N in the system. Results in Chapters 1 and 4 indicate that fertilizer N is especially susceptible to loss during years with abnormal precipitation patterns. Minimizing fertilizer N loss during these years may be difficult, although applications of nitrification inhibitors based on better weather predictions is a possible strategy.

To reach the goal of maximizing crop N use efficiency while minimizing environmental pollution for systems that use animal manure, legume, fertilizer, or a combination of N sources, we recommend an approach that incorporates calculating a

soil nitrogen balance. Nitrogen balances for total soil N in cropping systems can indicate areas of management practice that need attention (e.g. handling of animal manure) and can also be used to judge the long-term sustainability of a particular cropping system (e.g. legume- vs. fertilizer based). Calculating nitrogen balances for labeled N (^{15}N) applied to cropping systems is not the same as for total soil N, but can provide valuable information concerning how N sources interact in the systems.

We calculated a N balance for total soil N in the short-term legume- and fertilizer-based crop rotation experiment at EL and KBS from Chapter 3 of this dissertation (Tables R.1 an R.2). The legume hay system was represented by an average for corn following alfalfa and red clover, the legume green manure system was corn following hairy vetch, and the fertilizer-based system was corn, fertilized with 150 kg N ha^{-1}, following corn. Except for N inputs by asymbiotic fixation and N outputs by losses other than leaching, all values in these tables were measured in the study. Symbiotic fixation by the legume hay and green manure crops was measured by ^{15}N isotopic dilution as explained in Chapter 3 and is a critical input necessary to calculate realistic N budgets in these systems. Atmospheric deposition was measured for wet and dry fall at a weather station at KBS and by a ^{15}N isotopic dilution experiment using sorghum-sudangrass growing in containers filled with sand in the field at both EL and KBS. Nitrate-N leaching was measured at El and KBS using lysimeters as described in Chapter 4, and NO_3-N leaching from applied sources and soil organic matter were measured separately using ^{15}N.

The N balance for the 2-year rotations at EL and KBS shows that the legume green manure system is accumulating soil N, more so than the fertilizer system, and

Table R.1. Nitrogen balance for the short-term (2-yr) legume- and fertilizer-based crop rotation experiment at EL.

	Crop rotation		
	Legume Hay	Legume Green Manure	Fertilizer
	---------- kg N ha^{-1} ----------		
Inputs			
1. Fertilizer	0	0	150
2. Symbiotic Fixation	200	234	0
3. Atmospheric deposition	20	20	20
5. Asymbiotic fixation	20	20	20
Outputs			
1. Crop harvest	340	100	80
2. Loss			
a) NO_3-N leaching	45	45	45
b) Other	20	30	50
Balance			
Inputs - Outputs	-165	+99	+15

Table R.2. Nitrogen balance for the short-term (2-yr) legume- and fertilizer-based crop rotation experiment at KBS.

	Crop rotation		
	Legume Hay	Legume Green Manure	Fertilizer
	---------- kg N ha^{-1} ----------		
Inputs			
1. Fertilizer	0	0	150
2. Symbiotic Fixation	170	175	0
3. Atmospheric deposition	20	20	20
5. Asymbiotic fixation	20	20	20
Outputs			
1. Crop harvest	230	70	100
2. Loss			
a) NO_3-N leaching	25	15	15
b) Other	10	15	25
Balance			
Inputs - Outputs	-55	+115	+50

that a net loss of soil N occurred in the legume hay system. If the sustainability of cropping systems is judged by the ability to maintain soil N fertility levels, the legume green manure system is more sustainable than the fertilizer system, which in turn is more sustainable than the legume hay system. These results indicate that the legume hay system would actually require supplemental N (e.g. from fertilizer or animal manures) in order to maintain the same soil N levels. However, these results are based on very short rotations and may not reflect the long-term effect of these system on soil productivity.

A soil N balance was also calculated for the animal-, legume-, and fertilizer-based cropping systems in the long-term experiment at Rodale (Table R.3). This balance is for a much longer time period than that used at EL and KBS (10-yr vs. 2-yr), but fewer N inputs and outputs were measured experimentally at this site. Fertilizer and animal manure N inputs, and crop N harvest and NO_3-N leaching outputs have been measured at Rodale. Symbiotic fixation by forage legumes (in the low-input systems) and soybean in all three cropping systems however needs to be measured at this site in order to make the balances more accurate. Also, atmospheric deposition could be a significant N input at this location considering its proximity to coal-burning industrial areas and should be measured. Losses of N by mechanism other than leaching, particularly denitrification, should have been estimated experimentally at this site, and for that matter at the EL and KBS sites also. An added advantage at the Rodale site is that the cropping systems have been in operation long enough to detect a significant change in total soil N. According to these numbers however, the calculated N balances do not reflect what is apparently

Table R.3. Nitrogen balance for the long-term Rodale cropping systems experiment: 1981-1990.

	Cropping system†		
	LIP-A	LIP-CG	CONV
	---------- kg N ha^{-1} ----------		
Inputs			
1. Fertilizer	0	0	840
2. Animal manure	856	0	0
3. Symbiotic Fixation			
a) red clover	546	548	0
b) soybean	347	268	566
4. Atmospheric deposition	100	100	100
5. Asymbiotic fixation	100	100	100
Outputs			
1. Crop harvest	1437	706	1020
2. Loss			
a) NO_3-N leaching	200	200	200
b) Other	400	100	300
Balance			
Inputs - Outputs	-88	+10	+86

† LIP-A = low-input with animal (animal-based), LIP-CG = low-input cash grain (legume-based), and CONV = conventional (fertilizer-based).

happening in the different cropping systems. The N balances indicate an accumulation of soil N in the fertilizer-based system, greater than in the legume-based system, and a decline in soil N in the animal-based system. This is contrary to what is indicated by the change in total soil N levels where an increase in the animal-based cropping system, more so than in the legume-based system, and a decline in the fertilizer-based system has been measured. This illustrates the importance of accurately measuring all N inputs and outputs for a given cropping system in order to construct an soil N balance that can be used to help maximize crop N use efficiency and minimize polluting the environment with N.