IMPACT OF LEACHING AND DENITRIFICATION ON TEMPORAL DISTRIBUTION OF NITRATE IN SEVERAL MANITOBA SOILS

BY

SHERI L. GRIFT

A Thesis
Submitted to the Faculty of Graduate Studies
in Partial Fulfilment of the Requirements
for the Degree of

MASTER OF SCIENCE

Department of Soil Science
University of Manitoba
Winnipeg, Manitoba

National Library of Canada
Acquisitions and Bibliographic Services
395 Wellington Street
Ottawa ON K1A 0N4
Canada

Bibliothèque nationale du Canada
Acquisitions et services bibliographiques
395, rue Wellington
Ottawa ON K1A 0N4
Canada

Your file Votre référence

Our file Notre référence

0-612-62741-1

Canada

THE UNIVERSITY OF MANITOBA

FACULTY OF GRADUATE STUDIES

*****

COPYRIGHT PERMISSION

IMPACT OF LEACHING AND DENITRIFICATION ON TEMPORAL DISTRIBUTION OF NITRATE IN SEVERAL MANITOBA SOILS

BY

Sheri L. Grift

A Thesis/Practicum submitted to the Faculty of Graduate Studies of The University of

Manitoba in partial fulfillment of the requirement of the degree

of

MASTER OF SCIENCE

ABSTRACT

Grift, Sheri L. M. Sc., The University of Manitoba, March, 2001. Impact of Leaching and Denitrification on Temporal Variation in Profile Distribution of Nitrate in Several Manitoba Soils. Major Professor; Dr. C. M. Cho.

Leaching and denitrification are governed by several factors including soil management, moisture, aeration, organic carbon and soil temperature. The interaction of these factors and their effects on the processes of denitrification and leaching influence the temporal and spatial distribution of nitrate in soils. Nitrate leaching in the field was monitored on an Orthic Black loamy fine sand (Stockton soil), an Orthic Black loam (Wellwood soil) and a Gleyed Rego Black Almasippi sand. Four treatments were examined: fertilizer applied (100 kg ha^{-1} N as NH_4NO_3) on fallow, fertilizer applied and seeded to wheat, no fertilizer applied on fallow and no fertilizer applied on wheat. Samples were taken in 15 cm increments to a depth of 120 cm and analyzed for moisture and nitrate content, and groundwater samples were analyzed for nitrate content. Laboratory studies of nitrate disappearance rate were conducted on the Stockton and Almasippi soils using soil slurries incubated on a shaker at one of five temperatures: 5, 11, 16.5, 20.5, and 26.5°C. The amounts of NO_3-N remaining in the slurries after set incubation periods were graphed as a function of time.

Although nitrates moved mainly with soil water, soil nitrate contents of planted and fallow treatments were significantly different, while fertilizer application had no significant effects on nitrate distribution and profile content at the application rates used. Laboratory studies on denitrification rates indicate that for the Stockton profile, the biological activity decreased exponentially with depth and increased with increasing temperature according to the Arrhenius equation. The Almasippi profile exhibited two rates of biological activity: one for the 0-15 cm depth and another for the 15-120 cm depth, presumably due to the rapid decline in soil organic matter content below 15 cm in this profile.

ACKNOWLEDGEMENTS

I would like to express my sincere thanks to Dr. Cho for his patience, knowledge and guidance, and to Dr. Burton and Dr. Vessey for all of their suggestions, assistance and encouragement.

I would also like to thank Gregg Morden, Gloria Gingera and Darren Clouston, for their friendship, their assistance both in the laboratory and in the field, and particularly for their companionship on the long car trips to and from Carberry.

My gratitude also goes out to the staff and students of the Department of Soil Science, and to the federal and provincial "Soil Survey" staff, for all the friendship and support that has been extended to me throughout the years. In particular, I would like to thank Dr. Racz, John Fitzmaurice, Arnie Waddell, Michele Fitzgerald, Michelle Harland and Curtis Cavers for their heartfelt encouragement and for never letting me forget that I wanted to do this.

Finally, I would like to thank all of my family and friends, who were afraid to ask how my thesis was going, but were always there to listen when I needed to talk. I would especially like to thank my wonderful husband Parry LePage, without whose love and support this degree would never have been completed, and my beautiful daughter Adara, whose entrance into the world gave me the impetus I needed to finish.

TABLE OF CONTENTS

Page

ABSTRACT ii

ACKNOWLEDGEMENTS iv

LIST OF TABLES vii

LIST OF FIGURES viii

LIST OF ABBREVIATIONS xi

1. INTRODUCTION 1

2. LITERATURE REVIEW 5
    - 2.1 Fates of Nitrate in Soil 5
    - 2.2 Nitrate Leaching 8
        - 2.2.1 Environmental Implications 9
        - 2.2.2 Factors Influencing Nitrate Leaching 10
        - 2.2.3 Spatial and Temporal Variations in Nitrate Leaching 11
        - 2.2.4 Methods of Monitoring Nitrate Leaching 13
    - 2.3 Denitrification 14
        - 2.3.1 Environmental Implications of Denitrification 15
        - 2.3.2 Factors Controlling Denitrification 16
            - 2.3.2.1 Temperature 16
            - 2.3.2.2 Oxygen Status 19
            - 2.3.2.3 Moisture Content 21
            - 2.3.2.4 Amount of Available Carbon 22
            - 2.3.2.5 Soil pH 27
            - 2.3.2.6 Amount of Nitrate 28
        - 2.3.3 Temporal and Spatial Variations in Denitrification in the Field 30
            - 2.3.3.1 Spatial Variations in Denitrification 30
            - 2.3.3.2 Temporal Variations in Denitrification 32

3. MATERIALS AND METHODS 34
    - 3.1 Description of Sites and Soils 34
    - 3.2 Field Studies 37
        - 3.2.1 Site Preparation 37
        - 3.2.2 Samples and Analysis 40
            - 3.2.2.1 Soil Samples 40

3.2.2.2 Groundwater Samples 41
3.3 Laboratory Studies of Nitrate-N Disappearance 42
3.4 Statistical Analyses of Data........ 44

4. RESULTS AND DISCUSSION........ 47
4.1 Temporal and Spatial Variations in Nitrate Distribution in 1991 47
4.2 Temporal and Spatial Variations in Nitrate Distribution in 1992 61
4.3 Fluctuations in Total Profile Nitrate Concentrations 80
4.4 Denitrification Studies........ 92
4.4.1 Nitrate Disappearance Rate for the Stockton Soil........ 96
4.4.2 Nitrate Disappearance Rate of the Almasippi Soil 104

5. GENERAL DISCUSSION 111

6. SUMMARY AND CONCLUSIONS 123

7. CONTRIBUTION TO KNOWLEDGE........ 127

8. REFERENCES 128

9. APPENDICES 138
I. Mean soil temperatures for different profile depths of Stockton soil - Plot 1- for all sampling dates in 1991. 139
II. Mean soil temperatures for different profile depths of Wellwood soil -Plot 2- for all sampling dates in 1991. 140
III. Mean soil temperatures for different profile depths of Almasippi soil - Plot 3- for all sampling dates in 1991........ 141
IV. Soil temperatures for different profile depths of Stockton soil - Plot 1- for all sampling dates in 1992. 142
V. Soil temperatures for different profile depths of Wellwood soil -Plot 2- for all sampling dates in 1992. 143
VI. Soil temperatures for different profile depths of Almasippi soil- Plot 3- for all sampling dates in 1992. 144
VII. Oxygen flux of Almasippi soil in 1991 under two cropping treatments. 145
VIII. Oxygen flux of Stockton soil for two field seasons. 145
IX. Oxygen flux of Almasippi soil in 1992 under two cropping treatments........ 146
X. Equations for rates of nitrate disappearance for the Almasippi and Stockton soil profiles (0-120 cm depth). 147
XI. Arrhenius equations for the effect of temperature on rates of nitrate disappearance for the Almasippi and Stockton soil profiles (5-26°C). 147

LIST OF TABLES

Table ....Page

3.1 Description of soil properties of the sites. 36
4.1 Nitrate-N concentration of groundwater at plot 1 in 1991.. 49
4.2 Nitrate-N concentration of groundwater at plot 3 in 1991.. 50
4.3 Nitrate-N concentration of groundwater at plot 1 in 1992.. 64
4.4 Nitrate-N concentration of groundwater at plot 3 in 1992.. 65
4.5 Total nitrate-nitrogen in the Stockton soil profile over two years- plot 1................. 81
4.6 Total nitrate-nitrogen in the Wellwood soil profile over two years- plot 2. 82
4.7 Total nitrate-nitrogen in the Almasippi soil profile over two years - plot 3. 83
4.8 Effect of treatment on profile nitrate-nitrogen content - summary of response to ANOVA using all three plots. 85
4.9 Effect of treatment on profile nitrate-nitrogen content - summary of response to ANOVA using plots 1 and 2. 86
4.10 Total chloride content and nitrate-nitrogen: chloride ratios in the fertilized subplots of plots 1 and 3 to a depth of 105 cm. 89
4.11 Rates of nitrate disappearance on Stockton soil for all depths at various temperatures. 98
4.12 Activation energy and Q_{10} values for Stockton soil. 102
4.13 Rates of nitrate disappearance on Almasippi soil for all depths at various temperatures. 108
4.14 Activation energy and Q_{10} values for Almasippi soil... 110
5.1. Maximum nitrate attenuation potential of Almasippi profile in 1991. 118
5.2. Maximum nitrate attenuation potential of Almasippi profile in 1992. 119
5.3. Maximum nitrate attenuation potential of Stockton profile in 1991....................... 120
5.4. Maximum nitrate attenuation potential of Stockton profile in 1992....................... 121

LIST OF FIGURES

Page

Figure 3.1 Plot layout for the three replicate sites. 38

Figure 4.1 Amount of precipitation recorded for plot 1 during the 1991 growing season. 48

Figure 4.2 Amount of precipitation recorded for plot 3 during the 1991 growing season. 48

Figure 4.3 Distribution of nitrate in the soil profile as influenced by treatment for June 11-13, 1991. 52

Figure 4.4 Distribution of moisture in the soil profile under different treatments for June 11-13, 1991. 52

Figure 4.5 Distribution of nitrate in the soil profile as influenced by treatment for June 27, 1991 54

Figure 4.6 Distribution of nitrate in the soil profile as influenced by treatment for July 10-11, 1991 54

Figure 4.7 Distribution of nitrate in the soil profile as influenced by treatment for July 22-23, 1991 56

Figure 4.8 Distribution of nitrate in the soil profile as influenced by treatment for August 7-8, 1991. 56

Figure 4.9 Distribution of moisture in the soil profile under different treatments for August 7-8, 1991 58

Figure 4.10 Distribution of nitrate in the soil profile as influenced by treatment for August 19-22, 1991 58

Figure 4.11 Distribution of moisture in the soil profile under different treatments for August 19-22, 1991 60

Figure 4.12 Distribution of nitrate in the soil profile as influenced by treatment for

September 11, 1991. 60

Figure 4.13 Distribution of nitrate in the soil profile as influenced by treatment for September 25, 1991. 62

Figure 4.14 Distribution of nitrate in the soil profile as influenced by treatment for May 12-13, 1992. 62

Figure 4.15 Amount of precipitation recorded for plot 1 in 1992 66

Figure 4.16 Amount of precipitation recorded for plot 3 in 1992 66

Figure 4.17 Distribution of nitrate in the soil profile as influenced by treatment for May 25-27, 1992. 68

Figure 4.18 Distribution of nitrate in the soil profile as influenced by treatment for June 8, 1992 68

Figure 4.19 Distribution of chloride and nitrate under seeded and fallow conditions for June 8, 1992. 69

Figure 4.20 Distribution of moisture in the soil profile under different treatments for June 8, 1992 69

Figure 4.21 Distribution of nitrate in the soil profile as influenced by treatment for June 22, 1992 71

Figure 4.22 Distribution of chloride and nitrate under seeded and fallow conditions for June 22, 1992. 71

Figure 4.23 Distribution of nitrate in the soil profile as influenced by treatment for July 21, 1992 72

Figure 4.24 Distribution of chloride and nitrate under seeded and fallow conditions for July 21, 1992. 72

Figure 4.25 Distribution of moisture in the soil profile under different treatments for July 2 21, 1992 74

Figure 4.26 Distribution of nitrate in the soil profile as influenced by treatment for August 5-6, 1992 74

Figure 4.27 Distribution of moisture in the soil profile under different treatments for August 5-6, 1992 75

Figure 4.28 Distribution of chloride and nitrate under seeded and fallow conditions for August 6, 1992. 75

Figure 4.29 Distribution of nitrate in the soil profile as influenced by treatment for August 28-19, 1992. 76

Figure 4.30 Distribution of nitrate in the soil profile as influenced by treatment for August 31-September 2, 1992. 76

Figure 4.31 Distribution of nitrate in the soil profile as influenced by treatment for September 17, 1992. 78

Figure 4.32 Distribution of moisture in the soil profile under different treatments for August 18-19, 1992. 78

Figure 4.33 Distribution of chloride and nitrate under seeded and fallow conditions for August 18, 1992. 79

Figure 4.34 Distribution of chloride and nitrate under seeded and fallow conditions for August 31, 1992. 79

Figure 4.35 Nitrate disappearance rate of Stockton sandy loam at 5°C for 30-45 cm soil depth. 93

Figure 4.36 Semi-log plot of nitrate disappearance rate of the Stockton loamy fine sand profile at 5°C. 93

Figure 4.37 Semi-log plot of nitrate disappearance rate for Stockton loamy fine sand profile at 20.5°C. 97

Figure 4.38 Effect of temperature on nitrate disappearance rate at different depths of Stockton loamy fine sand profile. 100

Figure 4.39 Arrhenius plot of Stockton loamy fine sand at 0-15 cm depth: Effect of temperature on rate of nitrate disappearance. 100

Figure 4.40 Two rates of nitrate disappearance of the Almasippi sand as affected by soil depth at 26°C. 105

Figure 4.41 Two separate rate expressions for the Almasippi sand are combined into a single equation in order to express NDR for the entire profile at 26°C. 105

Figure 4.42 Effect of temperature on nitrate disappearance rates at different profile depths of the Almasippi sand. 107

LIST OF ABBREVIATIONS

DNI	Denitrification intensity
NDR	Nitrate disappearance rate
ODR	Oxygen diffusion rate
NFNW	No fertilizer, no wheat treatment
NFW	No fertilizer, wheat treatment
FNW	Fertilizer, no wheat treatment
FW	Fertilizer with wheat treatment
Ea	Activation energy
ppm	Parts per million

1. INTRODUCTION

Denitrification has been a subject of intense study since the early 1900's. However, it was believed that the process was of no importance under normal field conditions, and consequently early research on some factors influencing denitrification was limited (Panganiban, 1925). On the prairies, nitrate leaching did not appear to become a concern until well after commercial fertilizers became available in the 1930's (Hedlin, 1995), and then concerns were centred on the leaching of nitrate mineralized under cultivated conditions (Doughty et al.., 1954).

Currently, there is an abundance of information on the processes of denitrification and nitrate leaching, and research in these areas has been steady over the latter part of the 20th century. Initially, the major concern was economic, as any nitrate nitrogen removed from the root zone was nitrogen lost to the crop (Doughty et al.., 1954). However, over the years, the environmental impacts of nitrate leaching and denitrification have assumed much greater importance. Due to the high use of nitrogen fertilizers for crop maintenance, the possibility of groundwater contamination by nitrate is an important issue. High nitrate contents in drinking water can have adverse effects on human health (White, 1996) and a level of 10 mg L^{-1} has been set as a maximum allowable level of nitrate for potable water in Canada (CCREM, 1995).

Often, in the case of sandy soils, where large pore spaces result in the rapid movement of water, the maximum allowable limit of 10 mg NO_3-N L^{-1} may be exceeded in the groundwater to the point of doubling (Kladivko et al.., 1991). Leaching of soil nitrate is influenced not only by soil texture but also by other soil characteristics (Simmons and Baker, 1993), soil management (Toth and Fox, 1998; Levanon et al..., 1993; Roth and Fox, 1990), and frequency and duration of rainfall events (Izaurralde et al.., 1993). Under field conditions, variations in amounts of nitrate leaching may be both spatial (Pennock et al., 1992; Lund, 1982; Cameron et al., 1979) and temporal (Owens et al., 1999; Magdoff, 1992; Watts et al., 1991; Roth and Fox, 1990).

Denitrification is the process whereby facultative anaerobes use nitrate as the primary electron acceptor in the absence of oxygen. For this reason, denitrification is often seen as a method to reduce or prevent excess nitrate in the soil and groundwater (Schipper and Vojvodić-Vuković, 1998; Groffman et al., 1992; Obenhuber and Lowrance, 1991). However, several factors influence the occurrence and degree of denitrification including temperature, oxygen status, moisture content, available carbon, soil pH, amount of nitrate (Paul and Clark, 1989) and soil management (Watson and Mills, 1998). If these factors are optimum, significant losses of nitrate may occur via this pathway (Luo et al., 1998; Mahli et al., 1990). Like nitrate leaching, denitrification may also vary both across a field (Parkin, 1987; Folorunso and Rolston, 1984) and over time (Henault et al., 1998; Groffman and Tiedje, 1991).

While there is a good understanding of the factors influencing nitrate leaching and denitrification, the interactions between these factors within each process and between the processes themselves, are less clearly defined under field conditions (Parsons et al., 1991). Quite often, rates of denitrification as determined in the literature appear to be almost specific to soil type, and a great deal of research has been concentrated on studying denitrification of regional soils (Weier et al., 1993; Peterjohn, 1991; Peterjohn and Schlesinger, 1991; Hixson et al., 1990; Gilliam et al., 1977).

In Manitoba, potato production has been increasing, and optimum locations are coarse textured soils in proximity to a readily accessible water source for irrigation. This water source is usually an aquifer. The high hydraulic conductivity of the sandy soils located above these aquifers, coupled with the high levels of N fertilization recommended for optimum potato production, substantially increases the risk of nitrate groundwater contamination in these areas. The primary objective of this project was to determine the fate of surface applied nitrogen in three medium to coarse textured Manitoba soils: a Stockton loamy fine sand, a Wellwood loam and an Almasippi sand. These soils, three examples of medium to coarse textured soils representative of agro-Manitoba, were treated as field replicates in this study. All were located above aquifers to allow for groundwater sampling for nitrate content throughout the growing season. Nitrate was monitored in both the soil profiles and groundwater under field conditions in order to demonstrate the dynamics of nitrate movement through the soil profile in fertilized and unfertilized, planted and fallow conditions on these soils.

In order to determine the extent of the effects of biological activity on temporal and spatial nitrate distribution in these soils, laboratory studies of denitrification intensity

(DNI) were performed under saturated, anaerobic, nitrate-rich conditions using a range of soil temperatures encountered in the field during spring, summer and fall. This permitted the determination of the denitrification capacity in these soils to a depth of 120 cm. Parameters calculated for the denitrification capacity of these soils at different profile depths as influenced by soil temperature could be used to model the maximum nitrate attenuation of each soil over the growing season. This combination of field observations and laboratory studies allowed for an examination of nitrate dynamics in the different soil types, and a better understanding of the temporal and spatial distributions of nitrate within a field.

2. LITERATURE REVIEW

2.1 Fates of Nitrate in Soil

Nitrates in soil are subject to various possible fates, some of which result in the removal of nitrogen from the soil system. The fate of this nutrient is dependent on both soil microbial activity and soil physical properties. Processes to which nitrate is subject include denitrification, assimilatory nitrate reduction, dissimilatory reduction to ammonium, leaching from the soil profile, crop uptake, and off-site transport by wind or water erosion (Paul and Clark, 1989). Nitrate that is not physically removed from the profile or transformed by microbial processes may accumulate at some depth below the rooting zone, generally under arid conditions, where soil moisture is insufficient for leaching or sustained microbial reduction. This nitrate will be subject to leaching and/or microbial processes when soil moisture penetrates to that depth.

Paul and Clark (1989) report the following range of average values for the contributions made by various processes involved in removal of fertilizer nitrogen from the soil:

crop uptake	0-60%
immobilization	0-40%
gaseous loss	0-30%
erosion	0-15%
leaching	0-10%.

Others believe that after uptake, the largest fertilizer nitrogen removals occur via denitrification, leaching and erosion, in descending order (Lowrance, 1992). In fact, amounts of fertilizer N lost through each of the various processes may be higher than the averages shown above depending upon soil conditions. For example, in a loamy sand studied by Lowrance (1992), N losses to denitrification and erosion combined were only 20% of the total N lost to leaching. Of the total N lost to leaching as accounted for in the subsurface flow in Lowrance's study, 97% was in the form of NO_3-N, indicating that leaching was a major avenue of loss for NO_3-N on this coarse textured soil.

Crop uptake of nitrate is largely dependent on the amount of available ammonium in the soil. Plants will preferentially absorb NH_4^+ over NO_3^- because NO_3^- must first be reduced by the plant before it can be used to synthesize amino acids and proteins (Paul and Clark, 1989). In spite of the extra energy the plant must expend in absorbing nitrate, this is often the dominant form of N taken up by crops (Tisdale et al.., 1985). This is due largely to the free movement of NO_3^- in soil. Nitrate is transported to the roots by diffusion and mass flow, while NH_4^+ may become adsorbed by soil colloids (Paul and Clark, 1989).

Assimilatory reduction is the method used by microorganisms to incorporate nitrogen for cell growth. This is commonly referred to as immobilization of nitrogen as N incorporated in this way is not available to plants. Generally, reduction of nitrate in this manner will only occur if insufficient ammonium exists in the soil, as microorganisms are capable of incorporating ammonium without expenditure of additional energy required for reduction. This may be a major source of nitrate loss in the soil under certain conditions, particularly when high carbon, low nitrogen residues such as wheat straw are incorporated into the soil. C:N ratios of stable organic matter in soil are generally between 9 and 12 (Tisdale et al., 1985). If the straw does not provide adequate N to meet its carbon supply, microorganisms will assimilate mineral N from the soil to maintain this ratio. It is not a large source of loss where soil carbon inputs are low.

Dissimilatory reduction of nitrate occurs under anaerobic conditions and involves the reduction of nitrate to ammonium. The likely reaction is thought to be as follows (Paul and Clark, 1989):

$2HNO_3 + 4H \text{ -----}{>} 2H_2O + 2HNO_2$

$2HNO_2 \text{ ---}{>}?\text{---}{>} [2NH_2OH] + 6H \text{---}{>} 2H_2O + 2NH_4$

The extent of this occurrence in nature is unknown (Paul and Clark, 1989), but it is encouraged by the presence of large amounts of readily available carbon. Stanford et al.. (1975) found that soil amended with glucose at rates of 0.48 mg C g^{-1} or greater produced increasingly higher amounts of labelled $^{15}NH_4$-N with increasing glucose additions. Under unamended conditions, other researchers looked at seventeen different surface soils and found that NH_4-N production accounted for less than 2% of NO_3-N consumption, with the exception of one soil (6%) (Burford and Bremner, 1975).

Nitrate may be removed offsite via water and wind erosion. In the case of wind erosion, nitrate removed is in association with soil particles. Often, arid periods precede this type of erosion, and nitrate may accumulate near the surface through capillary rise. This may result in significant nitrate removal with erosion of soil by wind (Paul and

Clark, 1989). During runoff events, 99% of the NO_3-N lost due to water erosion is associated with the sediment, and can account for fertilizer losses as large as 160 kg N ha^{-1} y^{-1} under corn or fallow (Hargrave and Shaykewich, 1997).

Of the various avenues of nitrate loss, two are of most environmental concern: denitrification and nitrate leaching. Denitrification releases N_2O gas, a known contributor to ozone depletion. Nitrate leaching may result in contamination of groundwater and possibly of surface waters, with adverse health affects to those who use the water as a drinking source. Due to the extent of their impact on the environment, these two methods of nitrate removal are addressed in greater detail in further sections.

2.2 Nitrate Leaching

Nitrate is a concern in the soil profile because it moves with the soil water. This movement has often been explained by the following equation, which assumes movement by convection and diffusion:

$$\partial c/\partial t = D(\partial^2 c/\partial z^2) - V_o(\partial c/\partial z)$$

where c is the concentration of NO_3 (mg L^{-1}), t is time (days), D is the apparent mean diffusion coefficient (cm^2 day^{-1}), z is the linear distance in direction of flow (cm) and V_o is the average pore velocity (cm day^{-1}) (Paul and Clark, 1989). This equation implies that flow rate is proportional to leaching volume and inversely proportional to the volumetric water content; it is considered valid for homogeneous soil conditions (Paul and Clark, 1989).

In dry seasons, nitrate may concentrate at the surface as capillary rise increases, but in wet seasons it is leached from the soil surface (Tisdale et. al, 1985). Nitrate may move deep into the soil profile, where it is out of the reach of plant root systems, or it may be removed completely from the profile and enter the ground water (Paul and Clark, 1989). This is cause for concern as such removal of nitrogen from the rhizosphere is uneconomical for crop producers and can be an environmental hazard once it reaches the ground water.

2.2.1 Environmental Implications

The Canadian Water Quality Guidelines (CCREM, 1992) recommend a maximum allowable concentration of NO_3-N in drinking water of 10 mg L^{-1}. Levels higher than this can lead to a condition called methaemoglobinaemia in infants. High NO_3-N levels have also been shown to have adverse effects on reproduction, and have been associated with miscarriages and gastric cancer (White, 1996). Unfortunately, such levels of nitrates (>10 mg L^{-1}) are often found in ground water, particularly under coarse textured soils, which are subject to high nitrate leaching.

Roth and Fox (1990) discovered substantial movement of NO_3^- to the 90 - 120 cm profile depth within one year of application. Owens et al., (1991) studied well drained loam soils and found that the response time of aquifers to soil management effects was 3 years, but noted that this would be much faster under sandier soils with shallower depth to water table. They found that the ground water nitrate levels had reached 10 mg L^{-1} by the tenth year of the study (Owens et al., 1991). Similarly, researchers in Maryland (Weil, Weismiller and Turner, 1990) discovered that the nitrate concentration of ground water under four treatment fields was greater than 10 mg L^{-1} at N application rates common to the area (240-800 kg N ha^{-1} yr^{-1}). The soils used in this particular study ranged in texture from very fine sand to gravelly coarse sand. Kladivko et al.. (1991) examined subsurface flow of a silt loam soil by installing subsurface drains at 75 cm depth and reported NO_3-N levels averaging 20-30 mg L^{-1}, and generally above 10 mg L^{-1}.

Nitrate leaching is not only an agricultural problem. Significant nitrate leaching has been determined to occur from urban lawns. Exner et al. (1991) studied leaching conditions under a simulated urban lawn and found a particular problem with fall fertilizer application. They discovered that up to 95% of fall applied fertilizer was leached from the root zone (15 cm) within 34 days. Watts et al. (1991) also found that irrigated grass on sandy soils creates a hazard for ground water pollution.

2.2.2 Factors Influencing Nitrate Leaching

The amount of nitrate leached from a soil is dependent on soil texture, land management, and frequency and duration of rainfall events. Water movement, and hence nitrate movement is greater in sandier soils, a fact recognized (and studied) by various researchers. Previous fertilizer history must also be taken into consideration as nitrate accumulation can vary. Roth and Fox (1990) found that previously manured sites tend to accumulate greater amounts of nitrate (295 kg NO_3-N ha^{-1}) than unmanured sites (36 kg NO_3-N ha^{-1}). The type of tillage employed on agricultural land may also affect nitrate leaching. Nitrate leaching is generally higher under partial tillage than under zero tillage (Levanon et al., 1993). This is ascribed to higher nitrogen mineralization under partial tillage, while zero tilled soils have higher rates of immobilization and denitrification, resulting in less nitrate in the soil profile. Izaurralde et al. (1993) found that nitrate leaching was related to soil management, but stressed that rapid drainage events caused by heavy precipitation were also a factor in nitrate leaching.

2.2.3 Spatial and Temporal Variations in Nitrate Leaching

There may be several non-microbial reasons for spatial variation in soil nitrate concentrations. Variability is influenced by variable fertilizer application, micro topography and soil physical properties (Cameron et al., 1979). Cameron et al. (1979) studied spatial variability of anions and discovered that initially, variability can be accounted for by non-uniform fertilization; in cases of high fertilization rates, it accounted for more than 98% of the total variation. Differential leaching may occur after rainfall if there is variable rain catchment in micro relief areas; this can greatly influence leaching patterns (Cameron et al., 1979). Lund (1982) also found considerable spatial variations of nitrate in four study areas. Macropore or bypass flow may also be a factor in preferential nitrate leaching, particularly in medium soils with good aggregate formation (Misra and Mishra, 1977; Levanon, 1993; Shuford et al., 1977; Simmons and Baker, 1993). Simmons and Baker (1993) determined that macropore flow had a significant effect on leachate concentration, which was generally lower than the nitrate concentration in the soil matrix. Tyler and Thomas (1977) found similar results but speculated that macropore flow could be a significant problem with surface applied fertilizer in May and June. In this case, the nutrients will be carried with water from the surface through fissures and possibly out of the rooting zone and into the ground water.

Nitrate adsorption may also be a factor in spatial nitrate variability and reduced nitrate leaching in some soils. Nitrate adsorption has generally been found to occur mainly in tropical soils, but recent work in Fraser Valley suggests that nitrate adsorption should be incorporated when modelling nitrate leaching in acidic soils (Kowalenko and Yu, 1996).

Temporal variation in nitrate leaching has been studied by various researchers. Many have found that soil nitrate concentrations in fall were much higher than spring concentrations in the same area (Watts et al., 1991; Roth and Fox, 1990; Magdoff, 1992). Magdoff (1992) found no significant nitrate leaching in summer, which was attributed to evapotranspiration being greater than precipitation, but discovered that leaching may be considerable in fall if the soil nitrate content is high. Fall nitrate accumulation increases with increasing nitrogen application rates, and nitrates tend to accumulate at the surface, whereas spring nitrate accumulation occurs at a depth of 60-90 cm (Roth and Fox, 1990). Watts et al. (1991) studied grass production and also found higher nitrate concentrations in the fall than in the spring, which they ascribed to mineralization in the fall following the break up of sod. They determined that leaching during the winter months was a big source of nitrate loss (Watts et al., 1991). Owens et al. (1999) also found that nitrate leaching was most significant in the late winter and early spring under fertilized forages.

2.2.4 Methods of Monitoring Nitrate Leaching

Leaching has been studied *in situ*, under manipulated field conditions (irrigation, flooding) (Misra and Mishra, 1976), and in laboratory columns (Kowalenko and Yu, 1996), to measure rates of nitrate movement. The study of nitrate leaching usually involves soil and/or ground water sampling and one or more of the following monitoring methods:

1) Lysimeters or porous cups
2) ^{15}N labelled fertilizers
3) A nonreactive tracer (generally Cl or Br).

Various methods have been used to collect water from the soil profile during a leaching event. These include the use of various types of lysimeters (Toth and Fox, 1998; Tyler and Thomas, 1977; Owens et al., 1992), porous suctions cups (Misra and Mishra, 1976), vacuum operated extractors (Watts et al., 1991), and zero tension samplers (Simmons and Baker, 1993).

Nitrogen is subject to microbial manipulation, and following nitrate movement in the soil profile does not guarantee that all nitrate present is there as a result of the fertilizer added, or that all of the nitrate will be subject to leaching. Nonreactive tracers such as chloride or bromide are used because these anions move similarly to nitrate with water but are not subject to microbial activity (Cameron et al., 1979). Chloride is the anion generally used (Lund, 1982; Cameron et al., 1979; Tyler and Thomas, 1977; Shuford et al., 1977; Misra and Mishra, 1976; Endelman et al., 1974), but bromide has also been used, particularly where soil background Cl levels are high (Levanon et al., 1993). ^{15}N labelled fertilizers may also be used when a nitrogen budget is being established or they may also be used as a tracer in determining the amount of fertilizer N that is being lost to leaching (Mahli et al.., 1996) or involved in nitrification, mineralization, and immobilization (Watson and Mills, 1998).

2.3 Denitrification

Denitrification is the process whereby facultative anaerobes use nitrate as the primary electron acceptor in the absence of oxygen. The most probable pathway for this biological action is as follows:

$$NO_3^- \rightarrow NO_2^- \rightarrow (NO) \rightarrow N_2O \rightarrow N_2$$

Nitric oxide (NO) may or may not be an intermediate of this process. It tends to persist mainly under acidic conditions (Nagele and Conrade, 1990a; Cady andBartholomew, 1960), but is not readily observed in all situations. More recently, Gödde and Conrad (1998) ivestigated the relative contributions of denitrification and nitrification to production of NO in four different soils and with the exception of an acid forest soil, determined that in fact nitrification was the process governing NO production. Both N_2O and N_2 are gaseous in nature and either or both may be end products of denitrification if the N_2O escapes from the soil before it is further reduced to N_2. Nitrogen gas (N_2) is not subject to further reduction and will be released from the soil matrix, if the system is not closed.

Denitrifying populations are mainly composed of bacteria (Paul and Clark, 1989), although some fungi have also been found to be capable of denitrification. Many organisms that participate in denitrification lack the enzymes necessary to contribute to the entire pathway, so that complete denitrification requires a consortium of microorganisms. The microbial composition can also affect the outcome of nitrate reduction as proposed by Chèneby et al. (1998). They studied two soils and determined that although the soils had similar numbers of nitrate reducing organisms, the soils had different numbers of organisms capable of carrying denitrification through to the formation of N_2O and N_2. It has also been noted that even if all conditions for denitrification are similar between soils, the denitrifying enzyme activities of the soils may differ (Henault et al., 1998).

Large populations of denitrifiers can exist in the rooting zones of arable soils, with estimated numbers greater than one million organisms per gram of soil (Alexander, 1977). The presence of large numbers of denitrifying organisms generally indicates the denitrification potential of a soil, assuming that conditions conducive to denitrification were to exist (Alexander, 1977). Nitrous oxide emissions have been significantly correlated (r^2=0.92) directly to microbial activity as measured in some soils (Paramasivan et al., 1998).

2.3.1 Environmental Implications of Denitrification

The process of denitrification has both positive and negative implications for the environment. Nitrous oxide (N_2O) is an intermediate of denitrification, and may be a possible end product if it is released from the system prior to further reduction to N_2. Production of N_2O is detrimental to the environment as it contributes to the greenhouse gas effect (Government of Canada, 1991). However, denitrification may also be a sink for nitrous oxide, in that it can be reduced to N_2 if it enters the soil system. In addition, denitrification can significantly reduce the impacts of nitrogen loading on groundwater. Weil et al., (1990) found that denitrification was significant in preventing NO_3^- contamination of groundwater if enough carbon were present in anaerobic zones within the soil. However, Lowrance (1992) discovered that if environmental conditions were unfavourable for the reduction of nitrate, nitrogen loss via denitrification and surface runoff combined was only 20% of the nitrogen removed by leaching. Because the rate and occurrence of denitrification is governed by various environmental factors, results regarding denitrification are often largely site and/or soil specific.

2.3.2 Factors Controlling Denitrification

2.3.2.1 Temperature. The temperature range in which denitrification occurs has been given as 5-75 °C, with the upper range occupied by thermophillic bacteria (Paul and Clark, 1989), but other sources state that the range of temperatures in which denitrification can occur is actually 2-60 °C (Tisdale et al., 1985). Sources generally agree that denitrification rates are very slow at 5°C (Stanford et al., 1975) and may not be significant unless there is an adequate supply of an available energy source (Gilliam and Gambrell, 1978; Bailey, 1976). Jacobson and Alexander (1980) found no denitrification in soils maintained anaerobically at 1 °C for seven days, but found some nitrate reduction in the samples when they were incubated at 7 °C. Cho et al. (1979) determined that 2.75°C was the lower temperature threshold for denitrification, and found a linear relationship between temperature and DNI up to 20°C, while Pelletier et al. (1999) determined that there was still significant denitrifying enzyme activity at 2°C. Chemical denitrification, whereby NO_2^- is reduced to N_2, has been determined to occur under frozen conditions (Christianson and Cho, 1983).

Mahli et al. (1990) attempted to determine if the optimum temperatures for denitrification were lower in prairie soils than in the soils of warmer climates. They found an increase in the denitrification rate of 3.9 times in magnitude as the temperature was raised from 4-10 °C, and a slightly lower increase in rate of 3.5 times in magnitude as the temperature increased from 10-40 °C. At 60 °C, they found that the denitrification rate decreased in two of the three soils studied. This is comparable to the findings of previous researchers, who have found that denitrification rates change very little from 35-45 °C (Mangaraja and Misra, 1978; Stanford et al.., 1975).

A Q_{10} value signifies the magnitude that the rate of a reaction will increase for every 10°C increase in temperature. Within the temperature range of 15-35 °C, the Q_{10} value has often been cited as approximately 2. Cooper and Smith (1963) found a Q_{10} value of 2 for the temperature range of 20-30 °C. Stanford et al. (1975) used two soils of different textures and also found a Q_{10} of 2, although in this case the temperature range in question was 15-35 °C. In order to determine the effect of carbon amendments on the Q_{10} value, they ran a second experiment with identical conditions but with no glucose amendment and found that the Q_{10} value of 2 did not change from that of the unamended samples for eight different soils. Mangaraja and Misra (1978) reported similar results for a temperature range of 25-35 °C: soil samples with no carbon amendment had a Q_{10} of 1.9, while identical samples with sucrose added had a Q_{10} of 1.8. These Q_{10} values differ from those reported by Gilliam and Gambrell (1978) who obtained Q_{10} values of 4-6 for 15-25 °C, and a Q_{10} of 2 for the temperature range of 5-15 °C. Keeney et al. (1979) found that the total ($N_2 + N_2O$) -N evolved did not increase much between 7-15°C, which differs from the earlier findings of Stanford et al. (1975) who reported a tenfold increase in the rate of denitrification when the temperature was increased from 10 to 15°C.

Temperature has both direct and indirect effects on the rate of denitrification. Directly, denitrification rate (k, mol L^{-1}) increases as temperature (T, °K) increases as indicated by the Arrhenius equation:

$$k = Ae^{-Ea/(RT)}$$

or in its logarithmic form as

$$\ln k = -Ea/(RT) + \ln A$$

whereby Ea is the activation energy (kJ mol^{-1}), A is the intercept (mol L^{-1} s^{-1}), and R is the gas constant (8.314 J K^{-1} mol^{-1}) (Barrow, 1988). A graph of ln k vs. 1/T would show a linear relationship for a particular soil sample, provided the treatment of the soil sample, i.e. presence and level of amendments, remained the same under all temperatures studied. Such an equation could provide a useful estimate or indication of the rate of denitrification in a particular soil at any temperature within the temperature range studied. Under field conditions, temperature affects the distribution and movement of NO_3^- and O_2 in the soil profile, and will affect the amount of moisture lost from the soil through evaporation or plant uptake. Pang et al. (1977) applied urea to the centre of 12 cm columns of clay loam and studied the movement of NO_3^- under isothermal and gradient conditions. They discovered that not only did NO_3^- accumulate at the warmer end of the column, but also NO_3^- travelled farther from the site of application under a temperature gradient. This would affect the nitrate availability for denitrification in the field, particularly in the warmer summer months, as the nitrate would likely travel from the deeper more anaerobic soil zones further up the soil profile where denitrification would be more limited by aeration. In fact, in a field study by Parsons et al. (1991), they found no correlation between temperature and denitrification rate. They attributed this to the inverse relationship in the field between temperature and water filled pore space. Under laboratory conditions, soil temperature prior to the anaerobic incubation does not appear to affect the denitrification rate under incubation conditions (Smid and Beauchamp, 1976).

2.3.2.2 Oxygen Status. The effect of oxygen on denitrification has been seen to be twofold: the presence of oxygen inhibits denitrification, but if oxygen were not present, nitrification would not occur and denitrification would be limited by a lack of nitrate substrate (Alexander, 1977). Patrick and Reddy (1976) investigated the fate of ammonium in flooded soils in oxygenated atmospheres and determined that NH_4^+ in the flooded layer above the soil would be nitrified to nitrate, which would diffuse back into the anaerobic layer and be subject to denitrification. They state that the ammonium gradient created by this process would result in further diffusion of NH_4^+ into the flooded layer, continuing the production and eventual reduction of nitrate.

Various studies have been conducted concerning the upper level of oxygen that may be present in a soil system before denitrification can occur. Burton and Beauchamp (1985) found that, under field conditions, oxygen repressed denitrification at air filled porosities greater than 35%. Studies executed on denitrification within soil aggregates concluded that there was no significant denitrification within the aggregates unless the pore oxygen concentration was less than 0.5%, and denitrification rates did not approach the potential rates until the pore oxygen concentration was less than 0.3% (Parkin and Tiedje, 1984). The maximum concentration of oxygen in solution equilibrated with normal air is 0.26 µmole mL^{-1}, and Trevors (1985) suggests an upper limit of 0.20 µmole mL^{-1} oxygen in solution before denitrification will occur. He concluded that denitrifiers may begin reducing nitrate at low oxygen concentrations (Trevors, 1985). Kroeckel and Stolp (1985), found complete reduction of nitrate at $pO_2 \leq 6.7$ kPa, and complete recovery of nitrate at $pO_2 \geq 40$ kPa, and both aerobic respiration and denitrification at limited rates at pO_2 values between 6.7 kPa and 13.3 kPa. More recent studies investigating the relationship between denitrification and air filled porosity found that clay soils exhibited denitrification at higher air filled porosities than loam soils, and sandy soils showed no significant relationship between air filled porosity and denitrification (Groffman and Tiedje, 1991). They suggest that this is likely a result of the influence of soil texture on the availability of oxygen. Therefore, availability of oxygen may be of greater importance than the total amount of oxygen present in the soil system. This is particularly true of field conditions, where anaerobic microsites may occur wherever oxygen consumption is greater than its supply by diffusion (Alexander, 1977).

However, unless conditions within the soil system are conducive to denitrification, an absence of oxygen does not ensure that denitrification will occur. Sexstone et al. (1985) studied O_2 profiles in aggregates with a moisture content greater than 28%. They found that not all aggregates with anaerobic microsites exhibited denitrification, although all aggregates that did exhibit denitrification possessed anaerobic microsites. They suggested that the aggregates that had anaerobic microsites in which no denitrification occurred may have been limited by a lack of carbon and/or nitrogen substrate.

The availability or amount of oxygen present in a soil system may also influence the end product of denitrification. Firestone et al. (1979) found that a small addition of oxygen to anaerobic soil slurries, aside from causing a significant decrease in denitrification, also resulted in an increasing ratio of N_2O:N_2 produced. They suggested that denitrification was so slow in such systems that N_2O was able to diffuse from the site before it could be further reduced.

2.3.2.3 Moisture Content. It is well established that denitrification increases at high soil moisture contents. Kroeckel and Stolp (1986) determined that for a soil with a bulk density of 1.33 Mg m^{-3}, the lowest gravimetric water content at which denitrification occurred was 0.30 (approximately 0.40 volumetric water content). This value is in close agreement with the results of Grundmann et al. (1988), who found a field N_2O flux of near zero when the volumetric water content was decreased from 0.38 to 0.33. Other researchers have also found that denitrification rate increases with increasing moisture content (Christensen et al., 1990a), with peaks in denitrification occurring at water filled pore space levels of > 60% (Parsons et al., 1991). Denitrification may occur at substantial rates at simulated field capacity levels (Abbasi and Adams, 1998), but N_2O reduction is not always stimulated by addition of water (Jordan et al., 1998).

The reason often cited to explain the effect of moisture on the denitrification rate of a soil is the constraint placed on oxygen diffusion as the soil moisture increases. In addition, soil moisture may also play a more direct role on the rate of denitrification, and be instrumental in the determination of the end products that may form. Cady and Bartholomew (1960) performed a study in which soils at gravimetric moisture contents of 10%, 12.5% and 15% were incubated under anaerobic conditions. They discovered that the higher the moisture content of the soil, the more rapidly NO_3^- was reduced. After nine days incubation, N_2O had accumulated in the systems with the two lower soil moisture contents, and only N_2 could be found in the system with 15% moisture content.

2.3.2.4 Amount of Available Carbon. Generally, denitrification rates are lower in soils with low carbon contents. Several attempts have been made to quantify this relationship, the relationship between carbon content and soil respiration, and the relationship between soil respiration rate and denitrification rate. Burford and Bremner (1975) studied seventeen surface soils and discovered that the denitrification capacities of the soils were directly correlated to their total organic carbon (r=0.77), water soluble organic carbon (r=0.99) and mineralizable carbon (r=0.99) contents. They used the following equation to describe the relationship:

$$4(CH_2O) + 4NO_3^- + 4H^+ = 4CO_2 + 2N_2O + 6H_2O.$$

This indicates that 1 μg of available carbon is required for the production of 1.17 μg of N as N_2O (Burford and Bremner, 1975).

Katz et al. (1985) also found a good correlation (r=0.93) between denitrification capacity, soluble organic carbon, and oxygen consumption at various depths in five soils. Abbasi and Adams (1998) determined that nitrate disappearance rate was positively correlated (r^2=0.88) to organic matter content at all soil depths studied. Soil respiration as measured by CO_2 evolution has been shown to be strongly correlated to the denitrification rate (Parsons et al.., 1991). Drury et al.. (1991) looked at background denitrification (no carbon or nitrogen amendments), and found that it was strongly correlated to biomass carbon (r = 0.854) and organic carbon (r = 0.901). In earlier unamended studies, de Catazzaro and Beauchamp (1985) found that mineralizable carbon was a good predictor of denitrification in the first four days of the process, but found a lack of correlation later in the process which they attributed to nitrate depletion.

Stanford et al. (1975) determined the rate of nitrate disappearance (k, hr^{-1}) for several soils with organic carbon contents ranging from 5.3 to 30.2 mg g^{-1}, and noted that the rate of nitrate disappearance was highest at higher organic carbon contents. They added incrementally larger amounts of glucose to these soils, and found that k increased as the amounts of glucose added were increased for all soils until further additions had no more effect, and k became constant.

Burford and Bremner (1975) measured N_2O and N_2 evolution on several incubated, N amended soils and discovered that the rate of production slowed after five days, and was considerably slower after seven days. When subsamples of those soils with low denitrification rates were amended with 2 mg glucose C g^{-1} and subjected to the same incubation conditions, all nitrate was gone from the system within seven days.

A study by de Cantazzaro and Beauchamp (1985) compared the effects on denitrification rate of different carbon amendments: straw, alfalfa, sucrose, glucose, glycerol and mannitol. They found that denitrification rates were similar for all soluble carbon treatments. The actual ranking from lowest to highest denitrification rates was as follows: samples with no carbon amendments, straw amended samples, soluble carbon amended samples and alfalfa amended samples. Unamended samples had significantly lower denitrification rates.

Wheatley et al. (1991) monitored a potato crop to which they applied four separate treatments: no fertilizer, fertilizer (NH_4 and NO_3), fertilizer and sucrose, and fertilizer and straw. They determined the denitrification potential with and without added glucose for soils taken from each treatment and incubated at 100% moisture content anaerobically for 72 hours. At the beginning of the season, the potential rates were significantly greater only for the sucrose treatment, but by the end of the season, they were significantly greater for the straw amendment as well. The soils with glucose added showed similar trends to the soils with no added glucose but the rates of denitrification were higher.

Similarly, glucose added to unplanted and planted soils increased the denitrification rate in both soils, but in this case glucose additions decreased the rate difference between the planted and unplanted soils, although trends remained the same (Smith and Tiedje, 1979). In fact, carbon additions have been shown to increase the denitrification potential to five times (Drury et al., 1991) to ten times (Jordan et al., 1998) the background (no carbon or nitrogen amendments) denitrification level. When introduced in the preincubation stage, carbon amendments have increased denitrification capacity and also the total microbial biomass, which may be due to an increase in the size of the denitrifier biomass (Myrold and Teidje, 1985). Perhaps, most interesting is the fact that with carbon addition to soil samples, N_2O: N_2 ratios of denitrification products in the laboratory generally decrease at higher available carbon amendments (Weier et al., 1993). In this case, the denitrification rate is likely so rapid with the addition of carbon that N_2O is unable to diffuse away from the site of denitrification and is therefore reduced to N_2.

Carbon additions have also been examined as a possible alleviation of NO_3^- contaminated groundwater through their stimulation of denitrification. Obenhuber and Lowrance (1991) used microcosms containing aquifer material and approximated conditions under which the aquifer existed. They found that adding 10 mg C L^{-1} water per day increased the amount of N_2O dissolved and the most probable number of microorganisms, and decreased the NO_3:Cl ratio as well as the oxygen levels in the ground water. Therefore, they surmised that the addition of carbon to an aquifer promoted denitrification with existing microbial populations, which increased from 4% of the total population to 35-60%. Less than 2 % of the nitrate loss was attributed to assimilatory nitrate reduction.

When studying background denitrification, an important limitation could therefore be considered available carbon. Under field situations, the amount of carbon in the soil profile generally decreases with depth. Therefore, some effect of soil depth on denitrification could be expected to be observed. Luo et al. (1998) reported that denitrification in the field was highest at the soil surface 0-5 cm and generally decreased exponentially with depth. They determined that the carbon was limiting to denitrification at all profile depths, particularly in the subsurface soils. In spite of this, some subsoils have been found to have potential denitrification levels considered significant enough to reduce the hazard of nitrate contamination of groundwater (Castle et al., 1998). Smith and Tiedje (1979) studied denitrification rates in rhizosphere soil and found that rates were highest to lowest in this order: rhizosphere> non rhizosphere planted soil> unplanted soil. Wheatley et al. (1991) studied plants over the growing season and found that roots had a seasonal effect on denitrification as well as a soil depth effect. They found that the denitrification rate of soils increased with root emergence and again with senescence, and dropped as roots disappeared. They attributed this effect to carbon inputs by roots.

In a different approach, Katz et al. (1985) found a higher denitrification rate (mean NO_3 disappearance in μg day $^{-1}$ g $^{-1}$ of soil) in organic than in mineral soils. They concluded that there was a higher rate of carbon solubilization from organic matter than carbon consumed in denitrification, and that this restricted denitrification rates in some soils. Similarly, Lind and Eiland (1989) sampled soils to 20 m depth and found a potential for denitrification at all depths that was dependent on the presence of available organic carbon. McCarty and Bremner (1992) looked at surface (0-25 cm) and subsurface (150-200 cm) soils and discovered that although the number of denitrifiers decreased with depth, they were still significant at 150-200 cm depth (100-600 per g of soil). In fact, glucose amended subsurface soils showed high denitrification rates after seven days incubation. They concluded that the water-soluble carbon was decomposed in surface soils before it could reach the subsoil (water in tile drains was found to have a carbon content of < 2.9 μg C mL $^{-1}$). Later, they added extracts of surface soils to subsoils and found stimulatory effects on denitrification that were correlated to the organic carbon contents of the surface soil amendments (McCarty and Bremner, 1993).

2.3.2.5 Soil pH. Generally, acid soils are believed to have smaller populations of denitrifiers than soils of higher pH (Alexander, 1977). The preferred pH for denitrifiers is 6-8, with denitrification proceeding slowly below pH 5, and very little or no denitrification occurring below pH 4 (Paul and Clark, 1989). The products which accumulate during the denitrification process also differ as the pH changes: the dominant product is N_2O at pH 6.0-6.5, NO at $pH < 5.5$ and N_2 at $pH > 6.5$ (Tisdale et al., 1985).

As early as 1960 (Cady and Bartholomew, 1960), researchers studied the effect of pH on denitrification rate. In this particular study, they looked at a sandy loam soil and observed the denitrification rates when the soil was at a pH of 7.5 and when it was 4.5. They observed a much faster denitrification rate at a pH of 7.5, but observed no NO production. They proposed that NO is an equilibrium product of some reaction governed by pH. However, Nagele and Conrad (1990a) found that autoclaving a soil resulted in no NO production, although the un-autoclaved incubated soil produced NO, even at acidic pH. This led them to conclude that NO production must be a result of microbial activity.

A study by Blackmer and Bremner (1978) showed that as soil pH decreased, the inhibitory effect of NO_3^- on N_2O reduction increased. However, when nitrate was removed from the system, N_2O was reduced equally as rapidly in neutral, acid and calcareous soils. This effect of pH on N_2O was also observed by Nagele and Conrad (1990a) in a study which compared changing the soil pH in both an acid forest soil (pH was raised from 4 to 7) and an agricultural soil (pH was decreased from 7.8 to 6.5). They found that a low soil pH resulted in an increase in N_2O and NO released from the soil, in both the agricultural and forest soils. More recently, Ellis et al. (1998) noted that pH significantly influenced the number of denitrifiers in the soil, enzyme synthesis rates, and the actual kinetics of nitrate reduction.

Waring and Gilliam (1983) examined soils in the pH range of 3.2-7.3 and determined that soil pH did not strongly correlate with soil nitrate reduction but did correlate strongly with denitrification. They attributed this to the fact that the conversion of NO_3^- to organic N was high in acidic soils (14-18% of NO_3^-) and low in soils with a pH >4 (only 0.5-1.9%). Therefore, they believed that much of the pH effect was actually due to the presence of soluble carbon. In another study (Parkin et al., 1985), two soils with pH's of 4.08 and 6.02 were compared. Each soil had an optimum pH for denitrification near their natural pH (3.9 for the acid soil, 6.3 for the neutral soil), which appeared to indicate the presence of two different denitrifier populations in the two soils. Therefore, pH was not limiting at either site, although the natural denitrification rates at the more acidic site was one third the rate at the neutral site. These results enforced the findings of Muller et al. (1980), who determined that soils of like pH had similar denitrification rates, regardless of differences in vegetation.

2.3.2.6 Amount of Nitrate. Generally, higher NO_3^- concentrations may result in an increased rate of denitrification, and certainly will result in increased production of $N_2O:N_2$ (Firestone et al.., 1979; Weier et al.., 1993). Specifically, results have indicated that if $[NO_3^-] > 20\ \mu g\ mL^{-1}$ in solution, zero order kinetics will prevail, and the reaction will be independent of the nitrate concentration (Myrold and Tiedje, 1985). Conversely, at low nitrate concentrations, the reaction will occur according to first order kinetics and the rate of reaction will increase as the nitrate concentration increases (Firestone et al., 1979; Paul and Clark, 1989). This may be due to the low rate of diffusion to denitrification sites under low nitrate concentrations (Paul and Clark, 1989). Cho and Sakdinan (1978) noted that under flooded conditions, NO_3^- would be the preferred electron acceptor and N_2O would not be reduced until NO_3^- was removed from the system, provided that the system was shaken. This would provide a constant supply of electron acceptors to the site of denitrification, and would ensure the rate of nitrate disappearance was independent of the original nitrate concentration (Cho and Sakdinan, 1978).

As early as 1963, Cooper and Smith (1963) compared rates of denitrification (total time required to reduce all NO_3^- to N_2) at initial nitrate concentrations of 300 ppm, 150 ppm, 75 ppm and 37.5 ppm and found no change in the overall rates of denitrification at these initial nitrate concentrations. They did note an overall reduction in the maximum amounts of N_2O in the atmosphere when initial nitrate concentration was reduced. Others have also confirmed that NO_3^- has an inhibitory effect on N_2O reduction which increased with initial nitrate concentration (Blackmer and Bremner, 1978; Weir et al.., 1993) and decreasing pH (Blackmer and Bremner, 1978). However, research has also indicated that this effect is lessened in soils that are subjected to prolonged flooding (Terry and Tate, 1980).

In a field study by Ryden (1983), NH_4NO_3 was applied at a rate of 0, 250 and 500 kg N ha^{-1}. It was found that the rate of denitrification was not dependent on nitrate concentration at levels greater than 5-10 µg g^{-1}, although rates decreased with nitrate concentration at nitrate concentrations below 5 µg g^{-1}. Both the control and 250 kg N ha^{-1}

treatment plots acted as sinks for N_2O when soil moisture was greater than 20% by weight, nitrate content was less than 1 $\mu g\ g^{-1}$ and soil temperature was above 5-8°C. It should be noted that Mahli et al.. (1990) did report an increase in the rate of nitrate loss with increasing nitrate concentrations from 50 to 500 mg kg^{-1}. This appears to contrast with other available information for laboratory studies. However, Jordan et al. (1998) found an increase in nitrous oxide production of up to 10 times the pre-treatment rate when nitrate was added to field studies of denitrification in riparian forests. Others have also noted that NO_3-N limited denitrification at some soil depths, notably at the soil surface in the spring when nitrate concentration is low (Luo et al., 1998).

2.3.3 Temporal and Spatial Variations in Denitrification in the Field

2.3.3.1 Spatial Variations in Denitrification. Spatial variations in denitrification under field conditions are large (Folorunso and Rolston, 1984; Parkin, 1987). Researchers have found that spatial variability exists in fields with no differences between soil types, and suggest that while the measured characteristics of bulk soil may be a good predictor of temporal variations in denitrification rate, they will not accurately predict spatial variations (Parsons et al., 1991). Parkin (1987) believes that this spatial variation is due to the distribution of particulate organic carbon material, which influences the magnitude of denitrification rates and the spatial variability under natural conditions.

Parkin (1987) adopted the term "hot-spots" to describe those areas in the field where natural denitrification rates are significantly higher than the rates exhibited by the bulk of the field. It is thought that at these particular locations, oxygen consumption can lead to the presence of anaerobic microsites. That is, these sites may be a result of high microbial activity that may in fact be due to the presence of high levels of particulate organic matter (Parkin, 1987). Similar conclusions were reached by other researchers, who determined that these hot spots were not nitrate limiting (Christensen et al.., 1990b). Christensen et al.. (1990a) found hot spot activity in moist soil but determined that denitrification occurred throughout the soil if flooded conditions prevailed. Those microsites that were extremely active were assumed to approximate the denitrification potential of the soils, and addition of succinate failed to stimulate activity in these cores to the degree it did in low activity cores. They concluded that a removal of limitations to denitrification by reducing oxygen, adding nitrate and soluble carbon would reduce the spatial variations in denitrification. This would make any variation still present dependent on the distribution of the denitrifier population alone (Parkin, 1987).

Folorunso and Rolston (1985) examined spatial phase spectra and frequency patterns of various cycles in soil and found that both N_2O and CO_2 fluxes were in phase, meaning that total activity was high in any hot spots that were found. It was determined by other researchers that this flux did not correlate directly to the soil water content, water soluble carbon content, or soil solution nitrate content (Grundmann et al.., 1988). They attributed this lack of apparent relationship to the interaction between the various factors controlling denitrification rate, but did note that the soil water content would contribute to the variability in these parameters (such as nitrate and oxygen contents). Burton and Beauchamp (1985) did find some evidence of a relationship between N_2O production and air filled porosity and carbon supply, but found few significant relationships between N_2O

production and other parameters. Grundmann et al.. (1988) agreed with Parkin's (1987) conclusions that hot spots may be a result of particulate organic matter.

Seech and Beauchamp (1988) examined the denitrification rate in soil aggregates and found that denitrification and overall microbial activity was greater in small than in large aggregates. They attributed this to carbon availability, which was higher in small aggregates. Elliot et al.. (1991) reported that earthworm casts could be significant microsites in which denitrification could occur, contributing in part to spatial variation.

2.3.3.2 Temporal Variations in Denitrification. While there are seasonal variations in denitrification in the field, there is some temporal stability in spatial variations of denitrification. This conclusion was reached by Christensen et al..(1990b), who determined that at conditions of field capacity or greater moisture and unlimited nitrate supply, some locations in the field had consistently higher denitrification rates than other locations throughout the field season. On an overall field scale, denitrification is seasonal, with increased activity in the spring, and was very low or immeasurable in summer and autumn (Parsons et al.., 1991). Groffman and Tiedje (1991) obtained similar results when comparing spring microbial activity to that in summer, which they interpreted as a negative relationship between denitrification activity and temperature under field conditions. This may perhaps be more indicative of a positive relationship between moisture content and denitrification activity.

Along with a seasonal response, there may also be temporal variations in denitrification in the field as a result of single events, such as rainfall. Sexstone et al..

(1985) studied denitrification rates of a sandy soil as it was affected by rainfall events. They discovered that the denitrification rate was constant for the first 1-3 hours following the event, then increased for the next 3-6 hours, at which point the rate was ten times higher the rate before the soil was wetted. Within 12 hours of the initial event, rates had returned to their original levels. Ryden (1983) also observed that denitrification increased rapidly after rainfall events, particularly after fertilizer had recently been applied. In this case, the highest rates of denitrification (2.0 kg N ha^{-1} day^{-1}) occurred after N application in the spring when the soil water content was 30% by weight.

3. MATERIALS AND METHODS

3.1 Description of Sites and Soils

The continued use of high levels of nitrogen fertilizers has lead to an increased risk of groundwater contamination by nitrates, particularly on sandy soils that have a low water holding capacity. A field study was conducted to monitor nitrate movement through coarse to medium soils to determine the extent of groundwater contamination under these conditions. Three sites were chosen as replicates for this study based on soil texture and the presence of attainable groundwater. Two of these sites were located near the town of Carberry and will be called Carberry A (legal location: NE 25-10-15W) and Carberry B (SW 22-10-14W). The third site was located near the town of St. Claude at NE 10-9-8W. Topography at all sites was level. The two soils near Carberry were well drained Orthic Black soils. Carberry A was determined to be a Stockton soil with a sandy loam surface texture while the Carberry B site (Wellwood soil) exhibited a clay loam surface texture. The soil at the St. Claude site was an imperfectly drained Gleyed Rego Black soil of the Almasippi series. The depth to groundwater was considered approximately 6 m at the Carberry A site, and 1.5 m at the St. Claude site.

The physical and chemical properties of the soils are given in Table 3.1. Analyses were performed as follows:

A. Soil Texture

Particle size analysis was performed on all three soils using the hydrometer method after pre-treatment for removal of organic matter with 30% H_2O_2. These methods are outlined by Sheldrick and Wang (1993).

B. Bulk Density

Bulk densities were determined in the field to a depth of 120 cm for both the Carberry A and St. Claude sites. A modified core method from Culley (1993) was used, and three replicate cores were taken for each depth. Three sub samples from each replicate were analyzed gravimetrically for water content in order to calculate the oven dry weight of each soil core.

C. Organic Matter Content

Organic matter contents were determined for 15 cm increments to a depth of 120 cm for both the Carberry A and St. Claude sites. A wet oxidation-redox titration method was used, similar to that described by Tiessen and Moir (1993).

D. pH and Electrical Conductivity

Both the pH and EC measurements were based on a 1:1 mixture of soil and water that was stirred periodically for 30 minutes and then let to stand for one hour. At this point, readings were taken using an Accumet 25G pH meter with a combined calomel electrode. The suspensions were then filtered and a conductivity meter was used to obtain estimates of solute concentration. Values were obtained to a depth of 120 cm for the Carberry A and St. Claude sites.

Table 3.1 Description of soil properties of the sites.

Site and Depth	pH	Electrical Conductivity	Bulk Density	Organic Matter
St. Claude		**dS m^{-1}**	**Mg m^{-3}**	**%**
0-15 cm	8.03	0.147	1.16	1.84
15-30 cm	8.32	0.112	1.50	0.88
30-45 cm	8.34	0.200	1.56	0.66
45-60 cm	8.46	0.160	1.47	0.40
60-75 cm	8.48	0.170	1.51	0.18
75-90 cm	8.55	0.150	1.61	0.32
90-105 cm	8.45	0.175	-	0.25
105-120 cm	8.47	0.170	-	0.18
Carberry A				
0-15 cm	6.39	0.300	1.04	4.29
15-30 cm	6.84	0.220	1.12	2.31
30-45 cm	7.91	0.315	1.12	1.19
45-60 cm	7.62	0.450	1.14	1.19
60-75 cm	7.99	0.500	1.09	0.78
75-90 cm	8.03	0.500	1.08	0.52
90-105 cm	8.21	0.505	1.07	0.64
105-120 cm	8.21	0.460	-	0.19

3.2 Field Studies

3.2.1 Site Preparation

All replicated plots were 50 m x 50 m in size (Fig.3.1). These plots were divided into two halves and one half of the plot was seeded to wheat while the other half was left fallow. Central subplots measuring 20 m X 20 m were created at Carberry A and St. Claude and these subplots were fertilized with 100 kg ha^{-1} of N as NH_4NO_3 and 100 kg ha^{-1} K as KCl. The central subplot created at the Carberry B site was slightly larger (30 m x 30 m), but was also fertilized with 100 kg ha^{-1} N and K. This allowed for the establishment of four different treatments under which nitrate movement could be observed: soil under wheat with and without added nitrogen, and soil left fallow with and without added nitrogen.

Wells were installed to enable sampling of the groundwater for nitrates, and to allow the groundwater depth to be monitored. Two wells were installed at the Carberry A plot, one on the fertilized subplot, and the other on the unfertilized section of the plot. Four wells were placed at the St. Claude plot, one in the middle of the fertilized subplot and the remaining three at the edges of the unfertilized plot. No wells were installed at the Carberry B plot. Wells at all sites consisted of PVC pipe 10 cm in diameter, open at the bottom, with holes drilled into the sides at periodic intervals. The pipes extended to five meters in depth at the Carberry A plot and to only two meters in depth at St. Claude to reflect the estimated depth to water table.

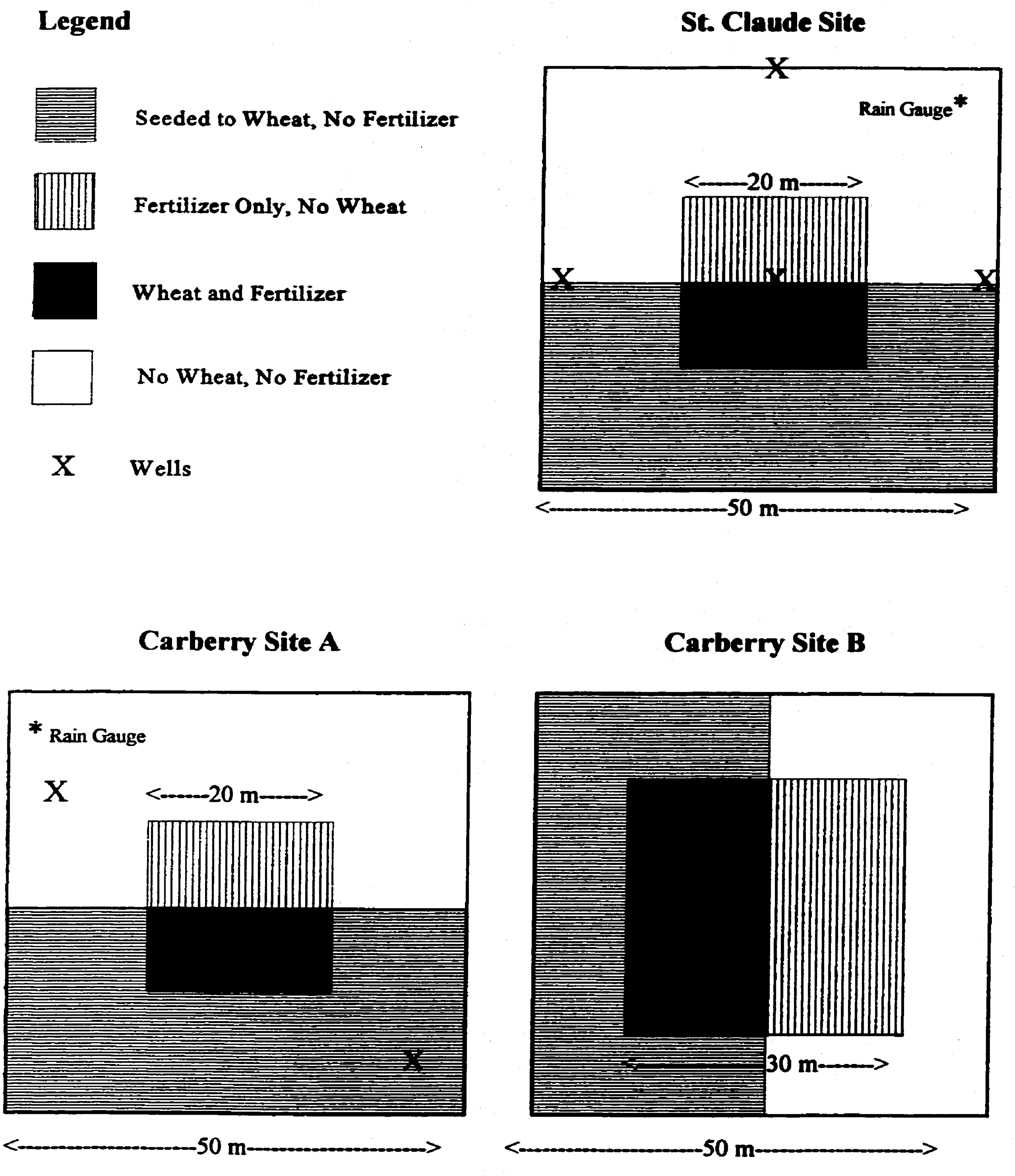

Figure 3.1 Plot layout for the three replicate sites.

Since temperature affects the rate and amount of denitrification (and hence NO_3-N removal from the soil profile) that will occur, soil temperature was monitored at all sites. Three temperature probes were installed at each of the three plots to allow temperature to be monitored to a depth of 120 cm. Temperature was taken at the soil surface, and at 2.5, 5, 10, 30, 60, 90 and 120 cm depth (Appendices I-VI).

Precipitation will obviously increase soil moisture and may result in nitrate leaching from the profile and in increased denitrification. Rain gauges were set up at the St. Claude and Carberry A plots to monitor precipitation in order to illustrate the relationship between rainfall and soil nitrate transport.

Denitrification occurs under anaerobic conditions and it was therefore desirable to measure soil aeration. Platinum microelectrodes were inserted at the Carberry A and St. Claude plots to allow the oxygen diffusion rate to be monitored in the soil at various depths. Two sets of microelectrodes were placed at St. Claude, in the northeast and northwest corners of the plot. These allowed for monitoring of the oxygen diffusion rate (ODR) in the soil at depths of 10, 20, 30, 50, 75 and 100 cm (Appendices VII and IX). One set of electrodes was installed at Carberry A to measure the ODR at depths of 10, 20, 30, 50 and 60 cm (Appendices VIII). The microelectrodes were left in the soil for the duration of the first field season, then reinstalled in the second field season. Measurements were made using a Model D Oxygen Diffusion Ratemeter from Jensen Instruments.

Soil samples were obtained for all four treatments at each of the three plots in 15 cm increments to a depth of 120 cm. The St. Claude plot was sampled on a weekly basis, while the Carberry plots were sampled every two weeks. Soil temperature, amount of precipitation and ODR were also measured at this time. The depth to groundwater was measured, and if possible, a groundwater sample was obtained from each of the wells at the Carberry A and St. Claude plots. Two existing wells near the Carberry A plot were also sampled. This included an irrigation well north of the plot and a well located in a shed adjacent to the plot. Effluent from a tile drain approximately 200 m from the St. Claude plot was also collected regularly.

3.2.2 Samples and Analyses

3.2.2.1 Soil Samples. An auger was used to obtain soil samples weekly to every two weeks from all four subplots at the three replicate plots from May to September in 1991 and 1992. These samples were taken in 15 cm increments to a depth of 120 cm. Each sample was sealed in a plastic bag and placed in a cooler with ice packs for transport to the laboratory. All sealed samples were placed in a freezer at a temperature of -4°C until NO_3-N extractions could be performed.

Prior to analysis, the soil samples were removed from the freezer and allowed to thaw at room temperature for approximately two hours. Two grams of field moist soil were placed into a 50 mL plastic centrifuge tube with 20 mL of 2 mol L^{-1} KCl. These centrifuge tubes were placed on a shaker for a period of 30 min and then centrifuged for 10 min. The supernatant was collected and placed in 15 mL plastic centrifuge tubes for storage. These tubes were stored in the refrigerator at 4°C and were analyzed for nitrate nitrogen content within one week. In order to determine the dilution factor for NO_3-N and the oven dry weight of soil used, the moisture content of the soil samples was determined at the time the nitrates were extracted, based on 5 g samples. This is a modified version of the extraction method outlined by Maynard and Kalra (1993).

A Tecator II autoanalyzer was used to determine the nitrate content of the samples based on the cadmium reduction procedure given by Maynard and Kalra (1993). The standards used in the laboratory were 0.5, 1.0, 2.0, 3.0, 4.0 and 5.0 µg NO_3-N mL^{-1}. Sets of repeats and standards were analyzed after every 24-sample run to ensure precision of the results to within 1 %. If NO_3-N readings were greater than 4.0 µg NO_3-N mL^{-1}, samples were diluted with 2 mol L^{-1} KCl and re-analyzed. This method involves the use of a cadmium converter column that reduces NO_3 to NO_2. The resulting NO_2-N is measured colorimetrically by the autoanalyzer. Periodically, a set of samples was run without the cadmium converter column to ensure that there was no NO_2-N in the samples, as this could lead to a higher reported level of NO_3-N than was actually present.

Following nitrate analysis, the original soil samples were air dried and stored in plastic bags. Soil samples obtained in 1991 were not analyzed for chloride content. However, air-dried soil samples obtained from the fertilized treatments of the St. Claude and Carberry A plots in 1992 were analyzed for chloride content at the end of the season. A mercury II thiocyanate method was used for colorimetric determination of chloride concentration as measured by a spectrophotometer (Fixen et al., 1988).

3.2.2.2 Groundwater Samples. Groundwater samples were obtained using a Masterflex peristaltic hand pump and collected in a 500 mL plastic jar with a screw top lid. These samples were immediately placed in a cooler with ice packs to be transported to the laboratory. The water samples were then placed in a refrigerator at a temperature of 4°C until they could be analyzed (less than one week).

At time of analysis, a 1 mL aliquot of groundwater was diluted to 20 mL with 2 mol L^{-1} KCl. The procedure for analysis for NO_3-N content of the groundwater samples is otherwise the same as that outlined for the soil samples above.

3.3 Laboratory Studies of Nitrate-N Disappearance

Profile samples from two plots were selected for the nitrate disappearance study: Carberry A and St. Claude. The second Carberry plot was not used in the study. Soil samples previously analyzed for NO_3-N in the first field season were air dried and sealed in labelled plastic bags for use in this experiment. For each plot, samples of equal depths were combined from all four subplots. This soil was thoroughly mixed and passed through a 2 mm sieve. The result was eight homogeneous soil samples (15 cm incremental depths to 120 cm) for each of the two profiles.

Destructive sampling was used to determine the rate of NO_3-N disappearance. If the amount of NO_3-N remaining in each tube after its designated incubation time were graphed as a function of time, the resulting slope would be the rate of nitrate disappearance of each soil depth at the particular temperature under study. This required that 6-8 samples be used for each soil depth at each temperature studied. Each sample represented one incubation time for one plot at a particular temperature. These samples consisted of 12.5 g of air-dry soil, which was placed in an airtight container and saturated, with 25 mL of KNO_3 solution containing 25 μg NO_3-N L^{-1}. After the samples were agitated to ensure removal of air trapped in the soil layer, oxygen was evacuated from all containers and argon was added to an internal pressure of 88 kPa. This pressure was chosen because it was less than atmospheric pressure (101.3 kPa) and would allow for an increase in the internal pressure of the sample tubes resulting from the accumulation of CO_2 and N_2O produced during the incubation. The samples were placed on a shaker and incubated for a period ranging from 6 hours to 12 days, to allow for up to 8 separate points for determination of the NO_3-N disappearance rate.

The samples were continuously shaken throughout the incubation to ensure that the N_2O produced could diffuse away from active sites of denitrification and NO_3 could be preferentially reduced, so that the rate of nitrate reduction would be an estimate of the rate of denitrification (Cho and Sakdinan, 1978). This procedure was repeated for each depth of soil from both sites. All samples (from both plots at all depths) were incubated at one of the temperatures under examination before repeating the procedure at another temperature. This ensured that the temperature remained consistent for all samples at all depths. Nitrate disappearance rates for the two soils were studied at the following temperatures: 5, 11, 16.5, 20.5, and 26.5°C.

After incubation for the designated period for each sample (ranging from 6 hours to 12 days), samples were removed from the shaker. The containers were examined for evidence of air leakage prior to opening and any that appeared to have been contaminated by oxygen were not used; these incubation times were later repeated with fresh samples.

If the tube appeared to be undisturbed, it was opened and the soil slurry was poured into a 50 mL plastic centrifuge tube. A 0.1 mL aliquot of concentrated HCl was added to each tube to lower the pH of the slurry in order to stop denitrification, and then the tubes were centrifuged for 10 minutes. The supernatant was decanted into 15 mL plastic centrifuge tubes and stored in a refrigerator at 4°C.

All refrigerated samples were analyzed for NO_3-N within 48 hours using the Tecator II autoanalyzer. The method used was as previously outlined, with the exception that a 1 mL solution sample was diluted to 10 mL with 2 mol L^{-1} KCl prior to being analyzed.

3.4 Statistical Analyses of Data

The plots were set up with a central fertilized subplot to enable determination of fertilizer effects on the groundwater concentration of nitrate. Each of the three plots was at a different site, but sites were chosen to be replicates: all had medium to coarse soils and water tables at depths shallow enough to allow for sampling using wells and a hand pump. Although same day sampling was often impractical, the plots were sampled within one to two days. However, it became apparent during the first year of the study that plot 3 was less than ideal as a replicate, as the extremely shallow water table at this site strongly influenced the movement of nitrate in this soil profile. For this reason, the means and standard errors for the three replicate plots were not determined. Instead, graphical illustrations using individual data points obtained from each of the plots were used to demonstrate the variation in nitrate distribution within the soil profiles over time.

In order to determine the effects of treatment on the nitrate distribution within the soil profiles, the total amounts of nitrate were calculated for the soil profile for each of the four treatments over all three plots. Amounts were calculated on a basis of kg NO_3-N ha^{-1} using a profile depth of 105 cm and soil bulk densities as previously presented in Table 3.1. The full 120 cm sampling depth was not used, as soil bulk densities for the 105-120 cm depth could not be obtained for the profiles. For plot 3, soil bulk densities were only obtained for the top 90 cm, but the value of bulk density obtained for the 75-90 cm depth at this plot was used for the 90-105 cm depth as an approximate value for bulk density. The actual value could not be obtained at this plot as the soil texture and close proximity to the water table made it difficult to remove a core sample for bulk density measurement below the 90 cm soil depth. For plot 2, bulk density values were estimated from plot 1 measured values.

Analysis of variance (ANOVA) was performed on the replicate data (as dictated by the common sampling dates) using JMP IN® software from SAS institute, and the effects of crop, fertilizer and crop x fertilizer interactions were examined for each year's data set. As the proximity to the water table at the third plot had profound effects on the nitrate distribution, and ultimately nitrate content of that soil profile, it was not considered a good replicate. The analyses were, therefore, run a second time for each year, this time without considering the data from plot 3.

Linear regression was used to describe the change in nitrate concentration over time in the incubation studies, the semi-log linear relationship between nitrate disappearance rate and soil depth and the semi-log linear Arrhenius graphs. Coefficients of determination (r^2 or r) were calculated and tested for significance via t-tests and standard errors of slope were determined using Microsoft Excel 97. Microsoft Excel 97 was also used to determine the effects of temperature on the nitrate disappearance rate at various soil depths. In this case, the relationship which best fit was the power relationship ($y=cx^b$, where c and b are constants) provided that the temperature of 26.5°C was included in the calculation. Coefficients of determination (r^2) were determined with Microsoft Excel 97 software, which uses a transformed regression model for determination of coefficient to describe the power relationship.

4. RESULTS AND DISCUSSION

4.1 Temporal and Spatial Variations in Nitrate Distribution in 1991

Plots 1 and 2 were seeded to wheat and fertilized on May 23,1991 with NH_4NO_3 and muriate of potash at rates of 100 kg N ha^{-1} and 100 kg K ha^{-1}, respectively. At the time of fertilizer application on these plots, the wheat had emerged and was approximately 5 cm tall. The precipitation gauge was installed at plot 1 on this date, and precipitation was monitored for the remainder of the season (Figure 4.1). Two wells were installed at this plot, and nitrate-nitrogen water concentrations were recorded for these wells, along with an irrigation well and an indoor well, for the remainder of the sampling season (Table 4.1). The third plot (plot 3) was seeded and fertilized at the same rates as for the first two plots on May 28, 1991. Three wells, the middle, east and west wells, were also installed at plot 3 on that date. The remaining well (north well) was installed on June 27, 1991 along with the precipitation gauge, and groundwater nitrate levels (Table 4.2) and precipitation (Figure 4.2) were monitored at this plot for the remainder of the season. No wells or precipitation gauges were installed at plot 2.

Sampling intensities of the plots varied, with plots 1 and 2 being sampled every two weeks, while plot 3 was sampled weekly. During periods of heavy rainfall activity, changes in the distribution of nitrate in the soil profile could occur rapidly, and it was

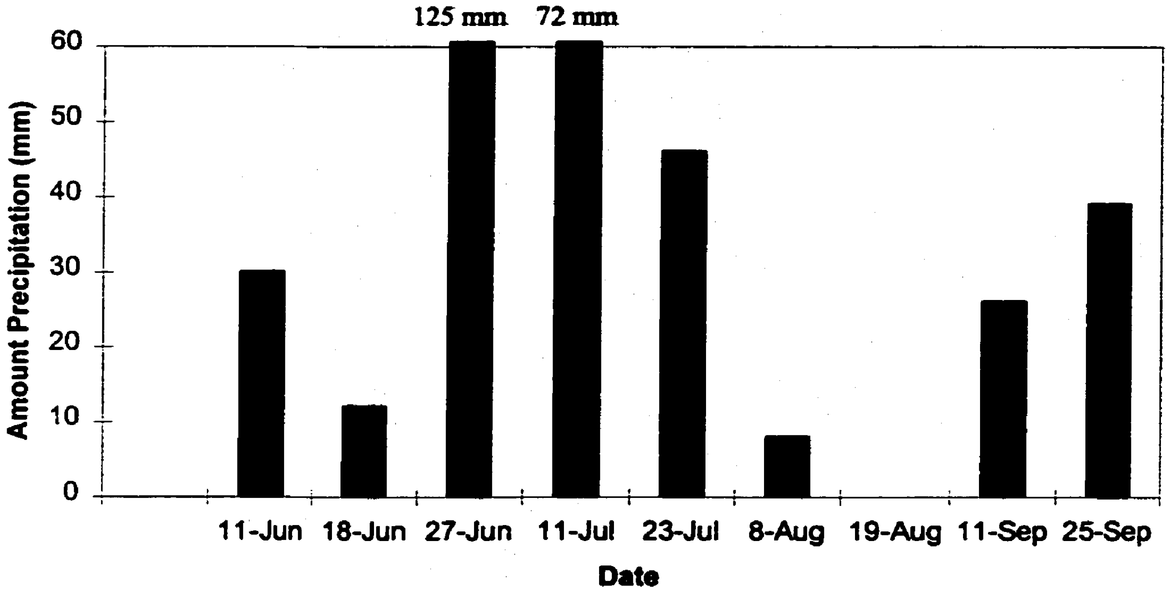

Figure 4.1 Amount of precipitation recorded for plot 1 during the 1991 growing season.

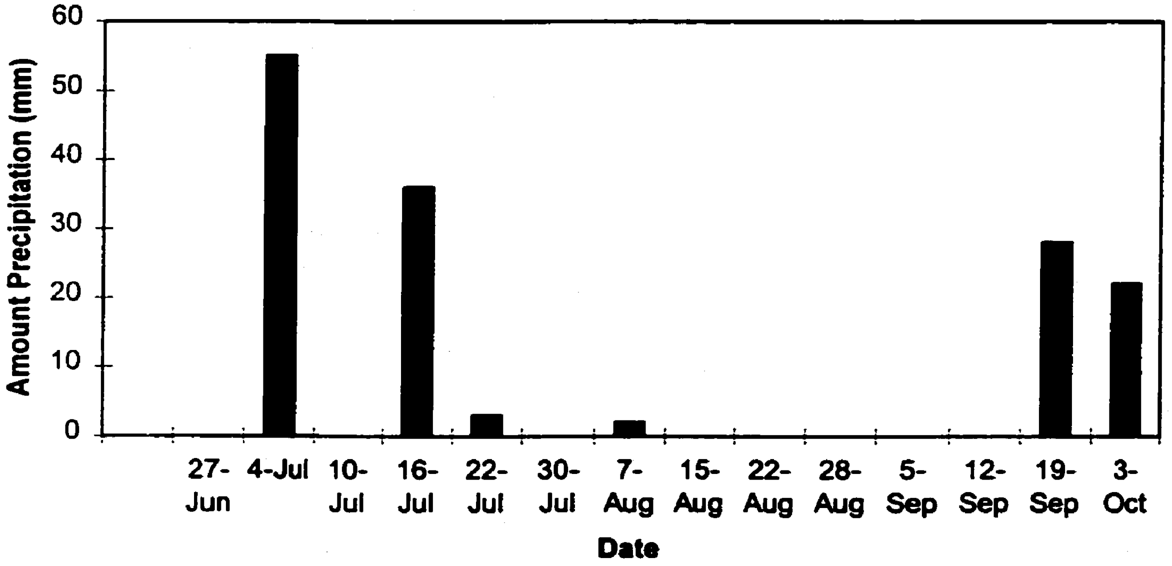

Figure 4.2 Amount of precipitation recorded for plot 3 during the 1991 growing season.

Table 4.1 Nitrate-N concentration of groundwater at plot 1 in 1991.

Date	Irrigation Well	Well for Other Use	Well of Unfertilized Subplot	Well of Fertilized Subplot
		$\mu g\ NO_3\text{-}N\ mL^{-1}$		
June 18	6.6	0.4	*	*
June 27	6.0	1.0	*	*
July 11	7.6	3.0	8.8	11.0
July 23	4.6	0	5.2	6.6
August 8	7.2	0	4.6	6.2
August 19	10.6	0	-	-

* Well not installed.
- Water table dropped below well depth.

believed that weekly monitoring of plot 3 might better illustrate the temporal variations in nitrate distribution. In addition, this plot had the shallowest water table, so impacts on groundwater health would be experienced at this plot more quickly than at the others. During the weeks when all three plots were visited, every effort was made to sample the plots within one to two days, so that these samples could be considered replicates. Due to the distance between the first two plots and the third plot, same-day sampling was often impractical. It became apparent during the first year of the study that plot 3 was a poor replicate, as the shallow water table in this area proved to have a distinct impact on nitrate distribution within the soil profile. Results have, therefore, been presented as individual points on the graph, rather than as means with error bars, in order to provide the reader with a clearer picture of the actual nitrate-nitrogen distributions in the profiles.

Table 4.2 Nitrate-N concentration of groundwater at plot 3 in 1991.

Date	**East Well**	**Centre Well**	**West Well**	**North Well**	**Tile Drain**
			$\mu g\ NO_3\text{-}N\ mL^{-1}$		
May 31	33.0	-	-	-	-
June 13	31.8	82.4	-	-	-
June 27	15.6	72.8	125.4	45.6	7.4
July 4	28.8	66.8	78.2	51.0	8.2
July 10	37.0	98.8	84.6	84.6	13.0
July 16	50.8	71.8	41.6	60.8	11.0
July 22	41.4	63.4	31.4	49.6	11.2
July 30	55.4	82.4	-	34.8	-
August 7	59.4	-	-	58.6	38.6
August 15	65.8	90.2	50.6	37.6	-
August 22	-	80.0	40.6	39.4	-

- No water in well.

The first round of soil samples were obtained post fertilization during the period of June 11-13, 1991. At this point, the fertilizer application was readily apparent in the high concentration of nitrate nitrogen at the soil surface (0-15 cm) of the fertilized subplots of all plots (Figure 4.3). Over 30 mm of precipitation was recorded at plot 1, and although there was no marked downward movement of nitrates, the increased soil moisture may have added to the nitrate load of the profile through nitrification of applied ammonium. It is interesting to note that the nitrate nitrogen distribution patterns were quite similar for all three replicates with the exception of the 105-120 cm nitrate concentrations. Values for all treatements at this depth were more widely divergent, particularly for the unfertilized treatments of plot 1, which expressed the highest profile nitrate nitrogen concentrations at this depth. This may be a relic of cropping history, as this field had been planted to potatoes the previous year, and applied nitrogen may not have been completely removed by the crop, and may have leached to this depth in the soil profile.

In fact, in a study conducted by Errebhi et al.. (1998), four application rates of nitrogen were examined on potatoes in a loamy sand, and it was determined that residual soil NO_3-N contents after harvest were at background levels in the top 90 cm, regardless of nitrogen treatment. However, there was a significant difference in nitrate levels between treatments at depths below 140 cm (Errebhi et al., 1998).

Soil moisture distribution patterns within the profiles (0-120 cm) were similar for all three plots (Figure 4.4), but generally moisture levels appear to highest for plot 2 (loam), then plot 1 (loamy fine sand) and lowest for plot 3 (sand), which is a reflection of their soil textures. This basic trend is exhibited through the entire season, with some exceptions for plot 3 as detailed later.

By June 27, 1991, 135 mm of precipitation was recorded. The area around plot 2 was completely inundated, and the field could not be sampled. Plot 3 was extremely wet, but samples could still be obtained to 120 cm. The initial high surface nitrate levels for

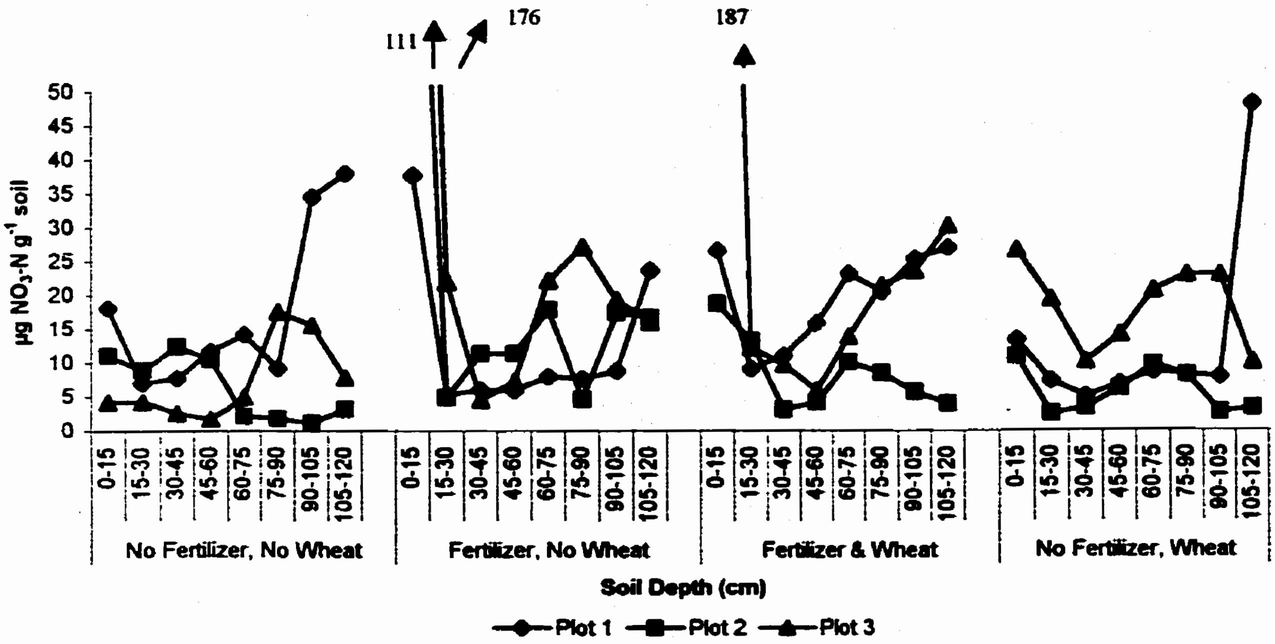

Figure 4.3 Distribution of nitrate in the soil profile as influenced by treatment for June 11-13, 1991.

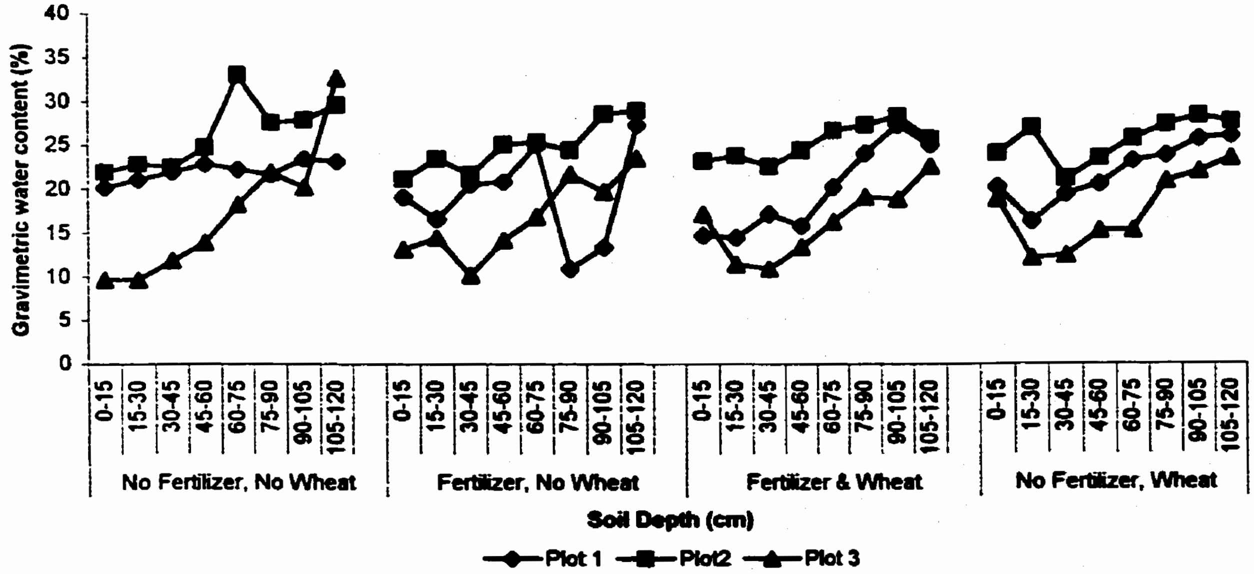

Figure 4.4 Distribution of moisture in the soil profile under different treatments for June 11-13, 1991.

plots 1 and 3 had dissipated and the nitrate appeared to be concentrating at some depth in the profile, likely as a result of leaching due to the heavy rainfall (Figure 4.5). This was also reflected by the nitrate concentrations in the ground water at plot 3, where nitrate levels in all wells were extremely high (Table 4.2). The groundwater nitrate concentrations were not above 10 $\mu g\ NO_3\text{-}N\ g^{-1}$ at plot 1, but as the depth to ground water at this plot was still greater than 4.5 m, the groundwater would not rapidly show the effects of a single event of nitrate leaching.

By the sampling dates of July 10 and 11, further precipitation had been recorded (Figures 4.1 and 4.2), and although plot 2 was no longer out of reach, plot 3 was completely saturated, and samples could only be obtained to a depth of 60 cm. The depth to water table, as measured from the west well at plot 3 was only 45 cm, and less than 4.5 m at plot 1. While the tile drain near plot 3 had been freely running since June 27, this date was the first during which the nitrate-nitrogen concentrations in the tile drain effluent climbed above 10 $\mu g\ NO_3\text{-}N\ mL^{-1}$ (Table 4.2). It was also the first date that a sample could be obtained from the well located on the fertilized subplot of plot 1, where nitrate concentration in the ground water beneath the site was 11 $\mu g\ NO_3\text{-}N\ mL^{-1}$ (Table 4.1).

Soil nitrate levels were similar for plots 1 and 2 for the seeded treatments (Figure 4.6). This was likely a result of uptake of nitrogen by the wheat as the crop at plot 1 was 90 cm tall and at the heading stage, and approximately half of the plot showed lodging. However, for the unseeded treatments, plot 1 had high nitrate spikes below 60-75 cm depth, while plot 2 did not. Given the inundation experienced at the second plot two weeks prior, it is entirely plausible that the nitrate either leached below the 120 cm

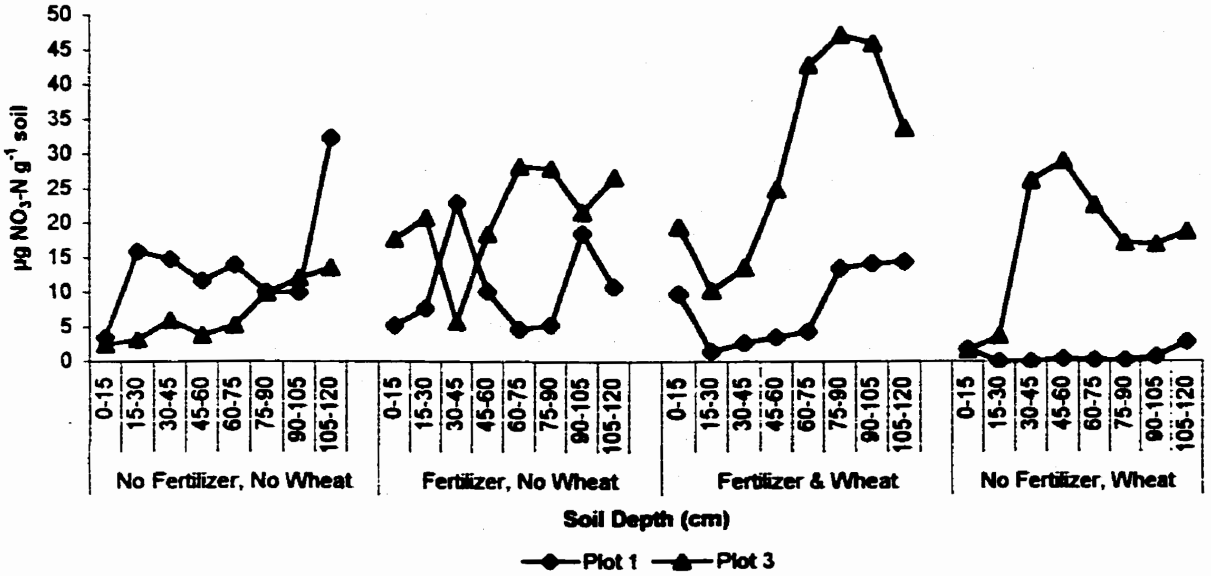

Figure 4.5 Distribution of nitrate in the soil profile as influenced by treatment for June 27, 1991.

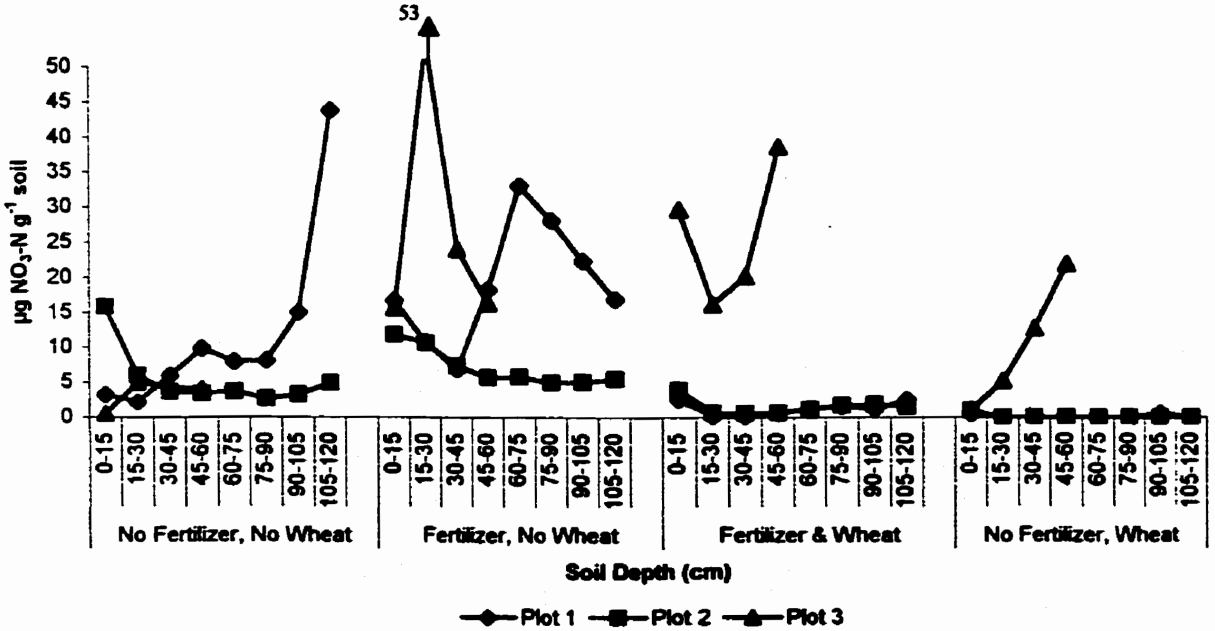

Figure 4.6 Distribution of nitrate in the soil profile as influenced by treatment for July 10-11, 1991.

sampling depth, or was subject to denitrification, or both.

Plot 3 was completely different from the other two plots on this date. Although low levels of nitrate were measured under the NFNW treatment, nitrate levels under the fertilized subplot were quite high, and for both of the wheat subplots, soil nitrate concentrations appeared to be climbing with depth. Interestingly, this is particularly true for the NFW treatment, which is following the pattern of distribution determined for the previous two sampling dates (Figures 4.4 and 4.5), and nitrate levels are actually climbing with depth. Since there was some ponding in the field beyond the NFW subplot, and the water table was highest in the west well, it seems that a discharge area existed immediately to the west of the field. It is therefore likely that the increased nitrate levels exhibited by the NFW treatment at depth were due to movement in the groundwater from below the fertilized treatments. The proximity of the groundwater table to the soil surface had a profound effect on soil nitrate distribution in this plot.

On the following sampling dates in the third week of July, nitrate distribution patterns in the soil were similar for all three plots within each of the four treatments with two exceptions. Plot 2 had a fairly high level of nitrates evenly distributed throughout the top 120 cm of soil for the NFNW subplot, and plot 3 still showed evidence of elevated soil nitrates due to lateral flow of nitrates in the groundwater (Figure 4.7). Of the four treatments, the NFW treatment seemed to have the lowest soil nitrate levels for plots 1 and 2. Since this is the treatment receiving no nitrogen amendments but still supporting a crop of wheat, this is hardly surprising. The uptake of nitrogen by the wheat was becoming evident in the soil nitrate distribution patterns at this point, and the low levels

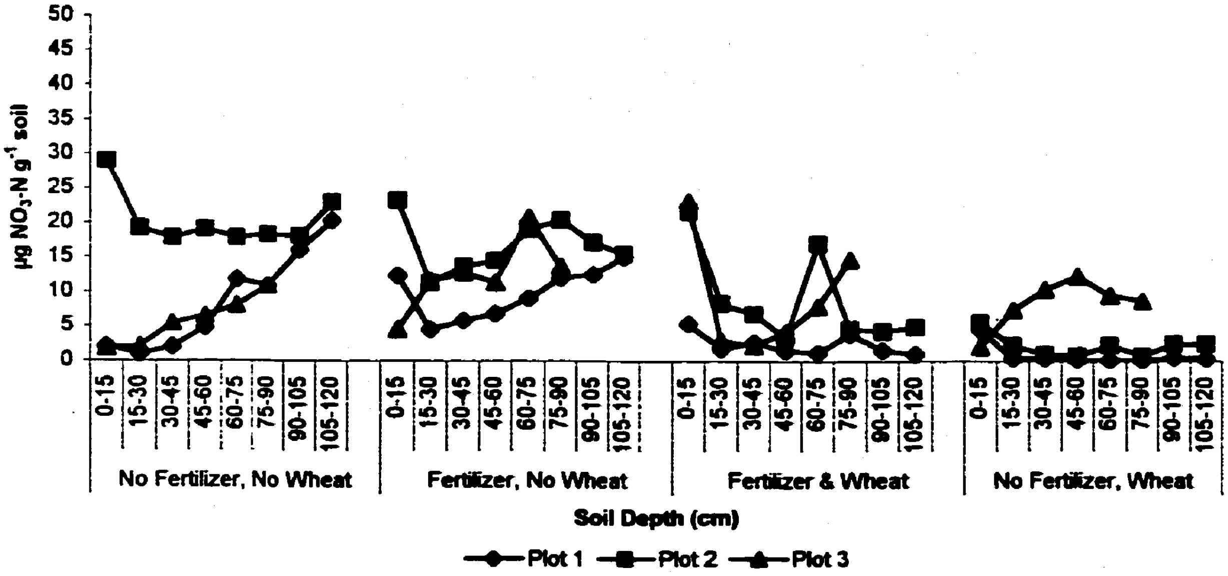

Figure 4.7 Distribution of nitrate in the soil profile as influenced by treatment for July 22-23, 1991.

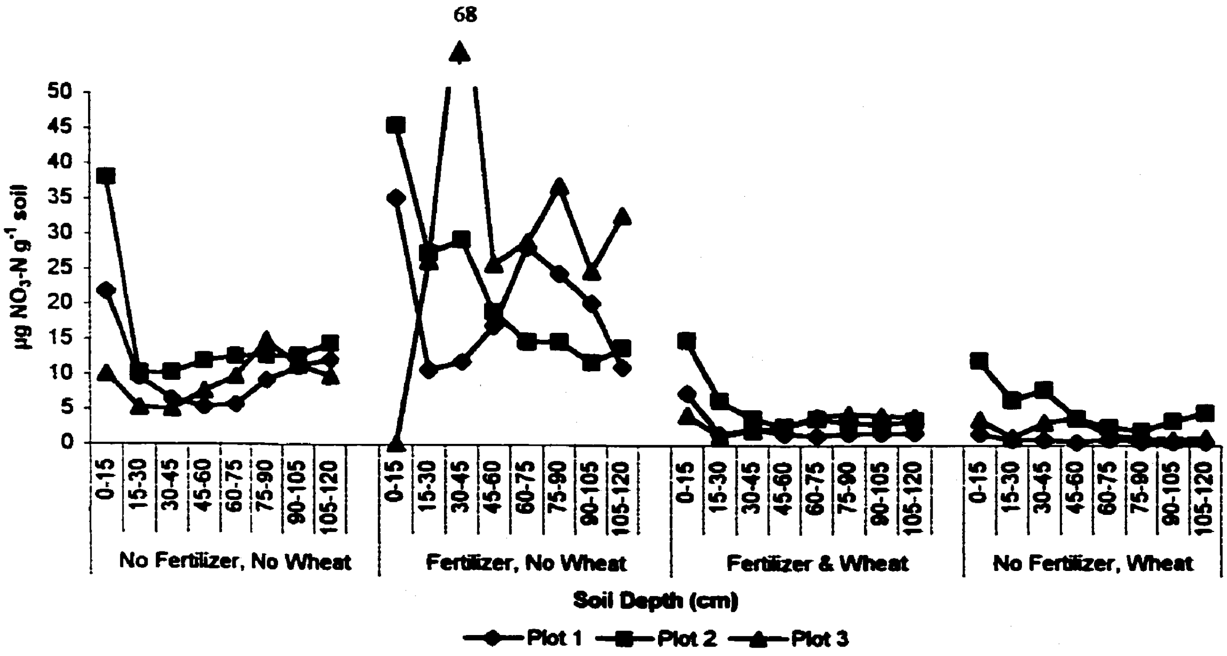

Figure 4.8 Distribution of nitrate in the soil profile as influenced by treatment for August 7-8, 1991.

of nitrates under the seeded treatments continued for plots one and two for the remainder of the growing season. It is important also to note that denitrification could have been occurring at optimum rates in these soils based on the soil moisture levels and high soil temperatures (Appendices I, II and III) for these dates in July. This could contribute to the removal of nitrate under the seeded treatments, as planted soil has generally been shown to have higher denitrfication rates than unplanted soil (Smith and Tiedje, 1979).

In the first week of August, nitrate levels were low for all plots under the seeded treatments (Figure 4.8), again likely due to crop uptake. The water table had receded somewhat at plot 1 and had dropped to 0.75 m at plot 3, but nitrate levels in the groundwater of plot 3 were still very high in those wells that still accessed the water table. There was no water flowing from the tile drain and the moisture content of the surface soil was fairly low (Figure 4.9). However, as the soil nitrate was generally highest in the surface 0-15 cm of all treatments for all plots, it appeared that the nitrate may have been moving up the soil profile with evaporation and upward movement of soil water through capillary rise.

On August 19-22, 1991, accumulation of nitrate at the soil surface continued and it became apparent that although upward movement of nitrates may have made a contribution to this phenomenon, nitrification was and had been occurring for all three plots on both dates. This was particularly evident under the FNW treatment (Figures 4.8 and 4.10), where the total amount of nitrate in the upper part of the soil profile was higher than what could be accounted for solely by redistribution. Certainly soil temperatures and moisture levels would have been favourable for this process (Appendices I, II and

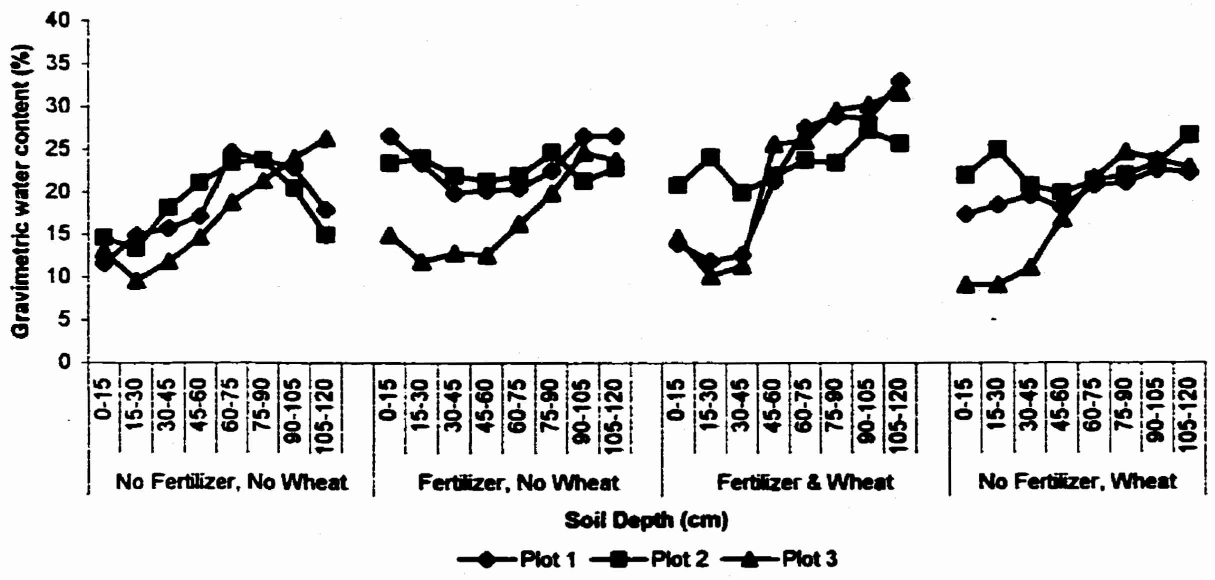

Figure 4.9 Distribution of moisture in the soil profile under different treatments for August 7-8, 1991.

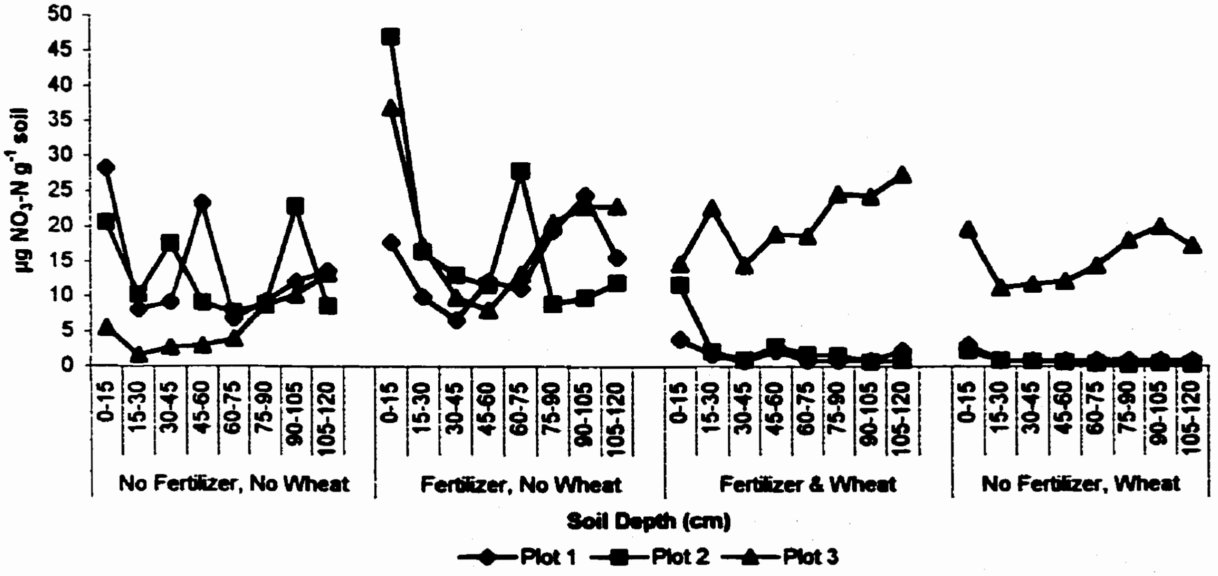

Figure 4.10 Distribution of nitrate in the soil profile under different treatments for August 19-22, 1991.

III). Profile nitrate distributions under seeded treatments of plot 3 had changed from the previous sampling date, and high levels of nitrates were expressed throughout these soil profiles (Figure 4.10). Once again, the lateral movement of nitrates with ground water at the third plot was masking treatment effects. Water levels were lower near the soil surface for all plots (Figure 4.11) and upward movement of water in response to evapotranspiration would have brought the nitrates in the shallow water table at plot 3 into the soil profile itself. It appears that very little, if any removal of nitrates through denitrification was occurring at plot 3. Even with crop uptake, there was simply too much nitrate in the soil profile as a result of nitrates in the groundwater for the nutrient to be completely removed from the soil. It is also worth noting that the rooting depth of the wheat at this plot was less than at plots 1 and 2, due to the saturated status of the soil for much of the earlier growing season, and crop uptake from this plot would likely be less.

In September, most of the soil profile nitrate was concentrated at the soil surface (Figure 4.12). This was either a function of movement within the soil solution, or as an expression of mineralization and nitrification following harvesting. Twenty millimetres of precipitation were recorded between August 19 and September 11, 1991, so mineralization and nitrification likely played a role in this increased nitrate due to a flush of biological activity following the addition of moisture to a warm, dry soil. Distribution patterns were similar for all plots on the unseeded treatments and for plots 1 and 2 on the seeded treatments. Nitrate levels were quite high under the seeded treatments for plot 3, which again was a result of the high nitrate concentrations in the ground water. Plot 3 was not sampled on September 25, 1991, but nitrate distribution within the soil profile of

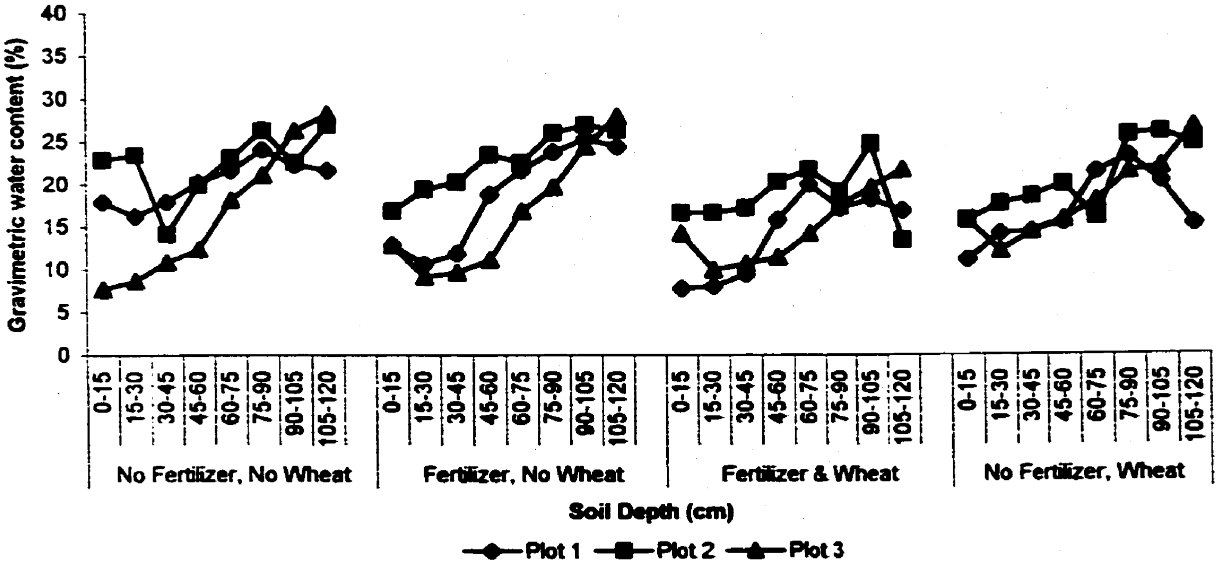

Figure 4.11 Distribution of moisture in the soil profile under different treatments for August 19-22, 1991.

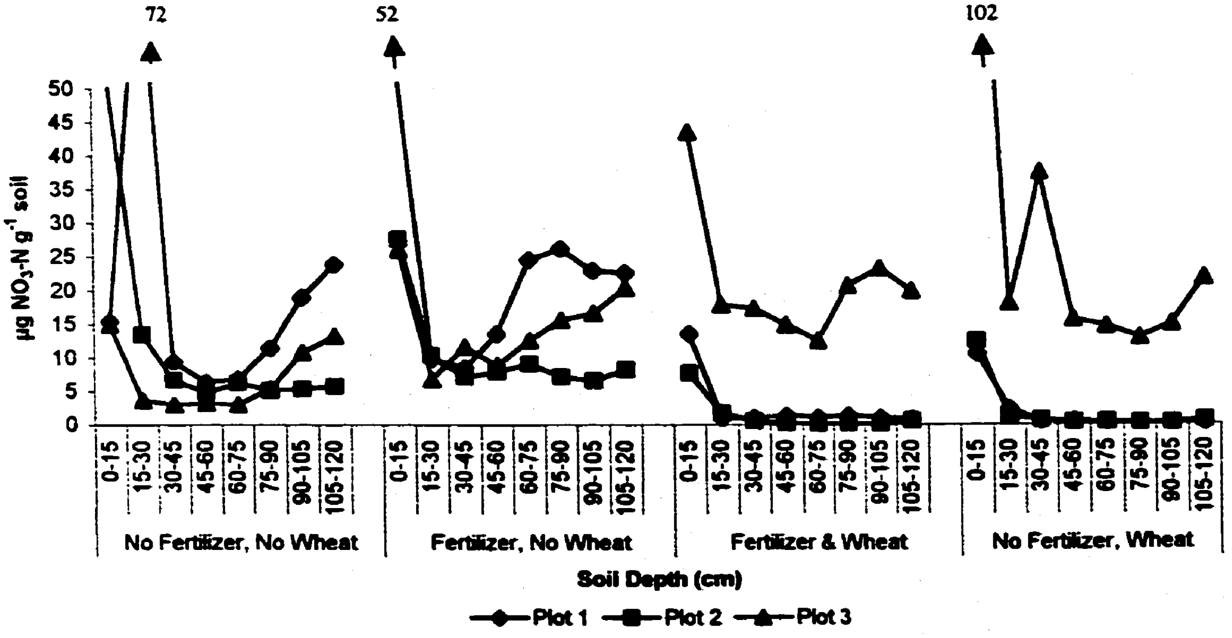

Figure 4.12 Distribution of nitrate in the soil profile as influenced by treatment for September 11, 1991.

plots 1 and 2 appear to be similar to the observations from two weeks prior (Figure 4.13), with accelerated nitrification near the soil surface following an additional 40 mm of precipitation.

4.2 Temporal and Spatial Variations in Nitrate Distribution in 1992.

Chloride in the form of KCl was applied to the fertilized subplot as a nonreactive tracer of nitrate movement in the soil profile. As previously mentioned, chloride moves with water similarly to nitrate, but it is not subject to microbial activity (Cameron et al, 1979). Therefore, the distribution of chloride within the soil profile is basically a result of the movement of the applied chloride with water in the soil profile, whereas nitrate distribution is a factor of both microbial activity (denitrification and nitrification) and leaching. Thus chloride distribution within the soil profile will provide confirmation as to the extent of leaching that occurred.

Although chlorides were applied to all three plots in both years at a NO_3-N: Cl^- ratio of 0.55, chloride concentrations in the soil profile were not analyzed for 1991 and were only analyzed for the fertilized treatments in 1992. Chloride concentrations in the soil profile of plot 2 were not analyzed for either year, as this plot did not appear to be significantly different in nitrate distribution from plot 1. Residual NO_3-N and Cl^- were present in the soil profiles from the previous season and nitrogen as NH_4^+ was also applied to the plot in both years. Since these background nutrient levels and any transformations of NH_4^+ to nitrate were not accounted for, it is difficult to establish the

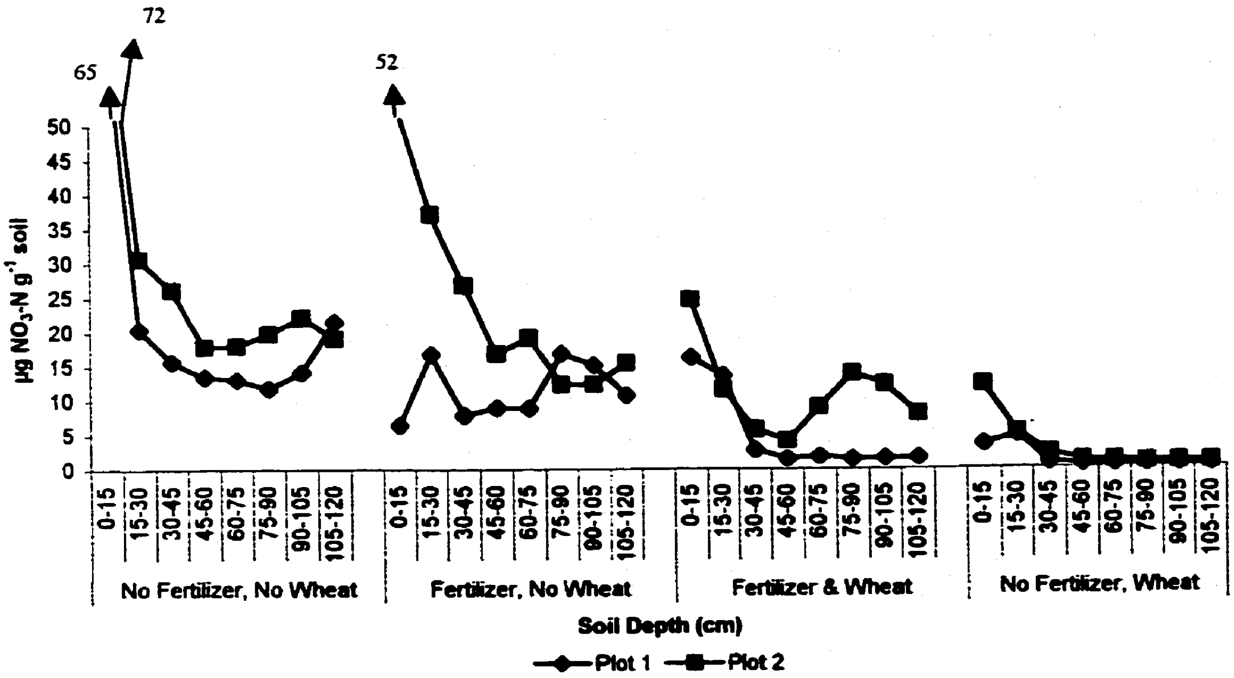

Figure 4.13 Distribution of nitrate in the soil profile under different treatments for September 25, 1991.

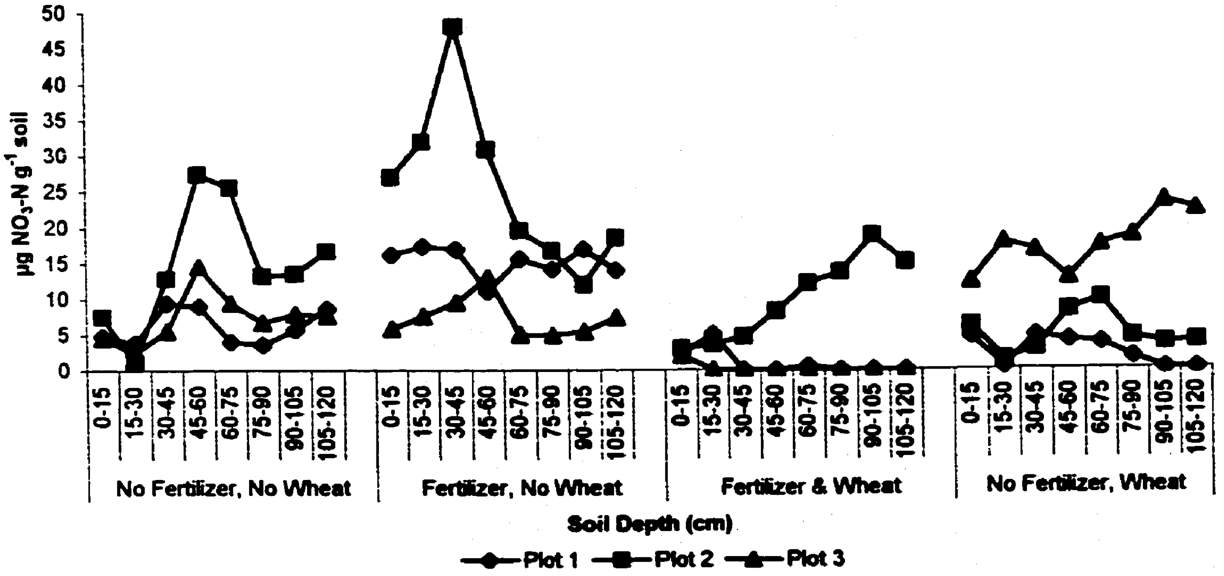

Figure 4.14 Distribution of nitrate in the soil profile under different treatments for May 12-13, 1992.

actual ratio at which the nutrients would be present in the soil at the start of the season. However, Cl^- and NO_3-N concentration as they changed with depth over time were graphed for use as an indicator of whether or not temporal variations in NO_3^- distribution were due to physical movement or biological transformations.

Prior to fertilization and seeding for the second season of the study, the three plots were sampled on May 12 and 13, 1992 (Figure 4.14). Nitrate distribution patterns for all plots were similar, and nitrate concentrations in the soil profile peaked at 45-60 cm depth for plots 2 and 3 for the treatments that had no crop the previous year. Generally, the nitrate levels were lower, and concentrated deeper in the profile than they had been in the last sampling of late September, 1991 (Figure 4.13). This indicated that with snow melt and early spring precipitation, nitrate appeared to have moved down the soil profile since the fall, and may have been subject to denitrification. These observations reflect the experimental results of Owen et al. (1999), who found the greatest amounts of nitrate were transported via leaching during the late winter and early spring and Pelletier et al. (1999), who determined that denitrification enzyme activity varied seasonally, with highest levels in November and May-June.

For plot 1, nitrate levels in both plot wells and the outside source were below 10 μg NO_3-N mL^{-1}, and the water table was at 4.2 m depth (Table 4.3). The water table at plot 3 was only at 60 cm depth, and water table effects present in 1991 were still evident in the high nitrate content of the profile of plot 3 under the NFNW treatment. Recall that the high nitrate levels in the ground water were reflected in the soil profiles with the shallowest depths to ground water. Ground water flux resulted in nitrate deposition in the soil profiles at depths that would normally have had low levels of nitrate at this point in the season. All four wells on the plot had very high groundwater nitrate contents (Table 4.4) and effluent from the tile drain was found to have a nitrate concentration of 12.6 μg NO_3-N mL^{-1}. Therefore leaching was likely significant under plot 3 for the overwintering period of 1991-1992. Rain guages for plots 1 and 3 were installed on May 12 and May 6, 1992, respectively and precipitation was monitored for both of these plots for the remainder of the growing season (Figures 4.15 and 4.16).

Table 4.3 Nitrate-N concentration of groundwater at plot 1 in 1992.

Date	**Irrigation Well**	**Well for Other Use**	**Well of Unfertilised Subplot**	**Well of Fertilized Subplot**
		μg NO_3-N mL^{-1}		
May 12	7.8	-	4.4	7.2
May 27	7.8	-	5.8	8.6
June 8	7.0	-	5.0	7.2
June 22	5.4	-	4.0	5.2
July 7	3.2	0	2.2	7.8
July 21	3.0	0	2.0	6.2
August 6	4.6	0	1.8	12.2
August 18	10.2	0	2.4	12.8
August 31	9.8	0	2.0	5.6
September 17	10.2	-	-	-

- No sample could be obtained.

Table 4.4 Nitrate-N concentration of groundwater at plot 3 in 1992.

Date	North Well	West Well	Centre Well	East Well	Tile Drain
			$\mu g\ NO_3\text{-}N\ mL^{-1}$		
May 6	25.0	33.6	51.4	51.2	7.8
May 13	32.0	36.2	57.6	46.2	12.6
May 20	29.2	35.8	61.2	39.8	9.8
May 25	27.0	41.4	60.4	40.4	9.2
June 2	26.2	38.0	61.0	44.8	-
June 8	29.0	40.2	61.2	44.6	-
June 16	-	39.4	65.4	49.0	-
June 22	32.4	36.6	97.4	53.0	-
June 29	37.2	34.6	87.8	54.2	-
July 7	36.6	36.2	94.8	52.4	-
July 14	-	36.2	84.2	49.8	-
July 21	-	34.6	77.6	53.2	5.0
July 28	33.8	33.8	75.0	56.2	-
August 5	35.0	33.2	71.8	63.4	-
August 11	-	35.2	73.0	65.0	-
August 19	-	36.0	72.0	65.6	-
August 26	32.6	37.8	63.2	55.6	-
Sept. 2	33.0	37.6	47.8	58.8	4.6
Sept. 10	29.6	37.0	41.0	55.0	-
Sept. 22	33.2	37.4	42.6	54.8	3.0

- No water in well.

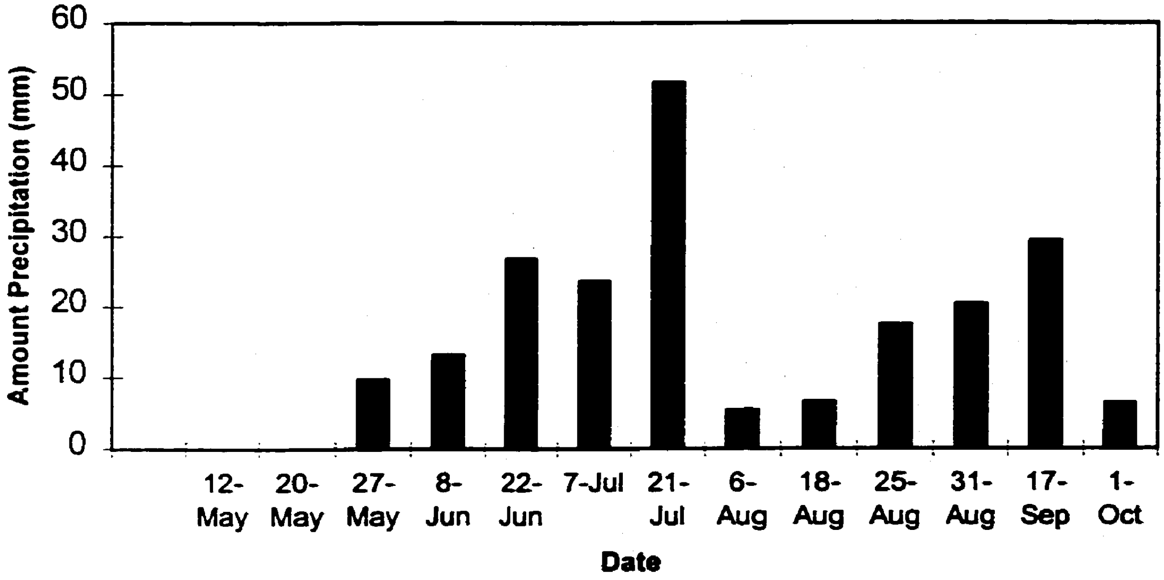

Figure 4.15 Amount of precipitation recorded for plot 1 in 1992.

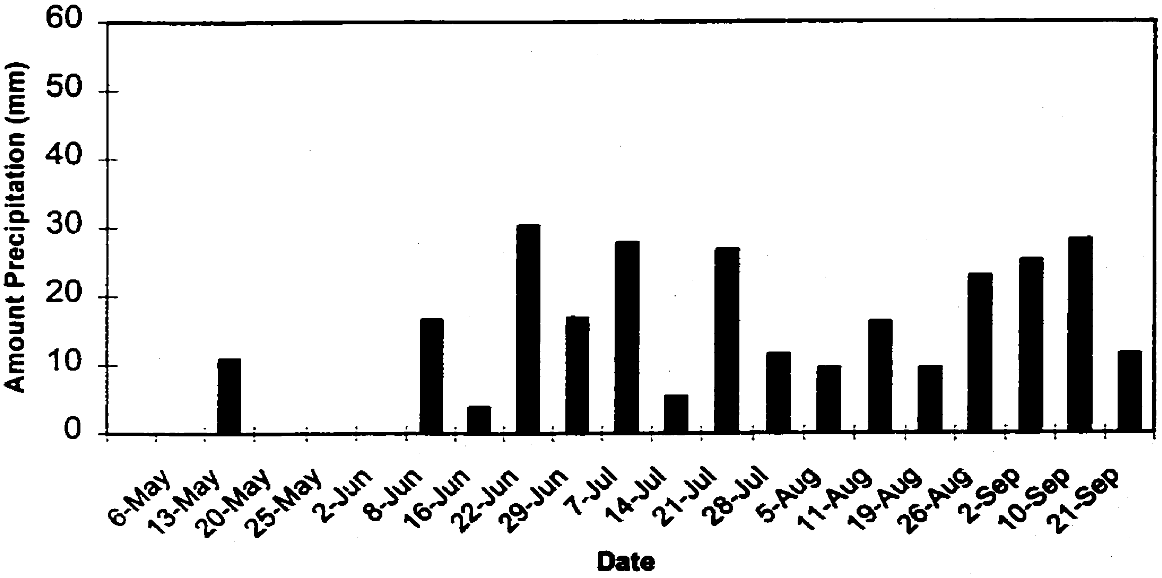

Figure 4.16 Amount of precipitation recorded for plot 3 in 1992.

The plots were seeded to wheat on May 19, 1992 and the last sampling dates prior to fertilization occurred on May 25 & 27, 1992. Soil profile nitrate distributions were similar for plots 1 and 3, but plot 2 had higher nitrate levels for the NFNW treatment, and distribution appeared to be erratic below the unseeded treatments at this plot (Figure 4.17). The levels of nitrate in the soil profile of plot 2 on the NFNW treatment remained higher than the other two plots for much of the growing season, and was likely a result of less leaching within this soil profile. After sampling on May 27, 1992, NH_4NO_3 and muriate of potash were applied to the central subplot of all three plots at rates of 100 kg N ha^{-1} and 100 kg K ha^{-1}, respectively.

By the next sampling date of June 8, 1992, 12 mm of precipitation was recorded for plot 1, and 17 mm was recorded for plot 3. Some of the fertilizer nitrate had already leached down into the soil profiles, particularly for plot 3, as expressed by the accumulation of soil nitrate with depth (Figure 4.18). However, much of the nitrate in the plot 1 fertilized subplots still remained at the soil surface, as did the chloride in the fertilized subplots (Figure 4.19), indicating that extensive leaching had not occurred. It is interesting to note that plot 2 does not appear to be following the trends indicated by plots 1 and 3 for this season. A quick examination of the soil moisture distribution in these three profiles from this date (Figure 4.20) indicates that although the moisture content of plots 1 and 3 was increasing with depth, soil moisture at plot 2 was relatively evenly distributed throughout the soil profile. The overall nitrate contents in the surface 0-120 cm of this soil are also higher, particularly under the NFNW treatment. It seems apparent that at this time, very little leaching of nitrates had occurred in this profile from the

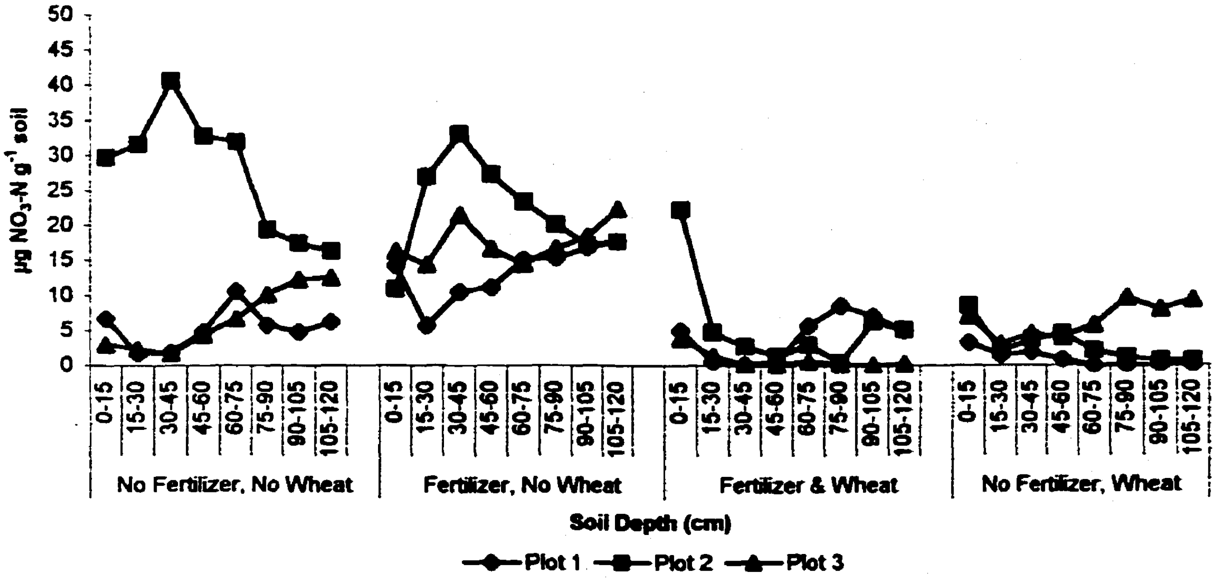

Figure 4.17 Distribution of nitrate in the soil profile under different treatments for May 25-27, 1992.

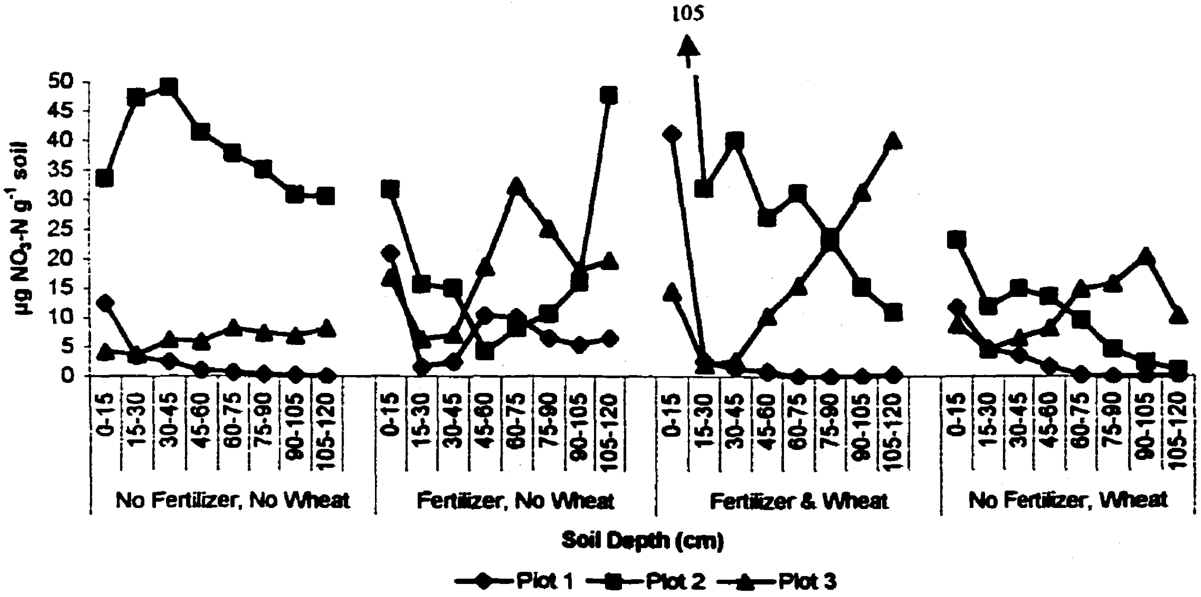

Figure 4.18 Distribution of nitrate in the soil profile as influenced by treatment for June 8, 1992.

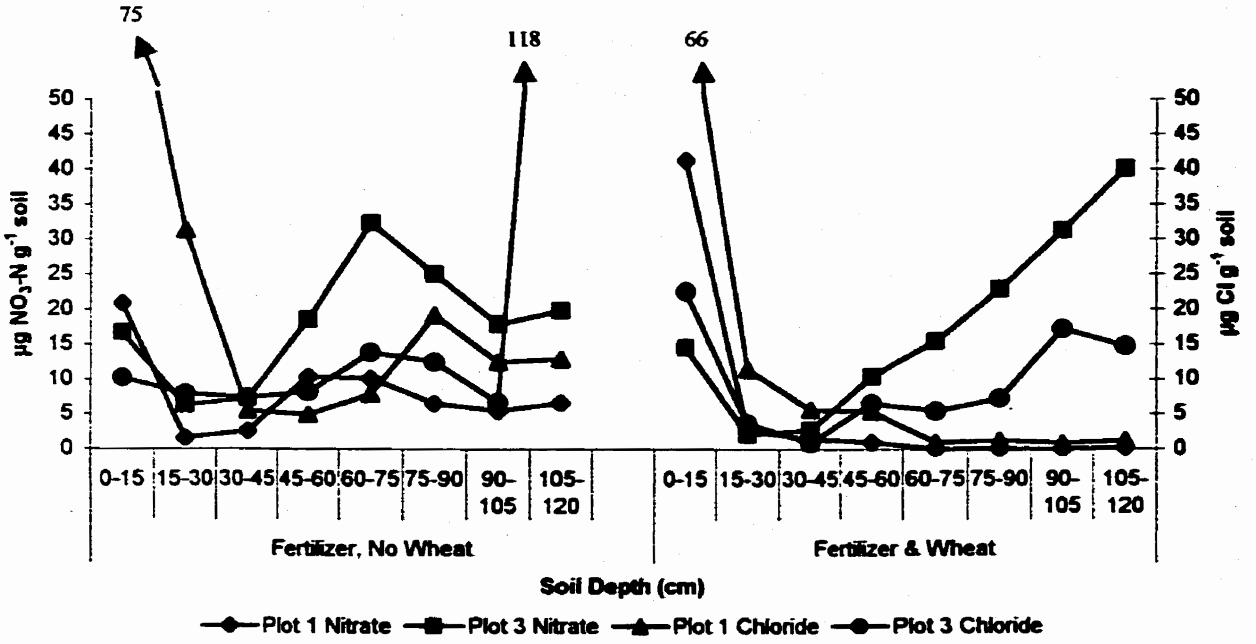

Figure 4.19 Distribution of chloride and nitrate under seeded and fallow conditions for June 8, 1992.

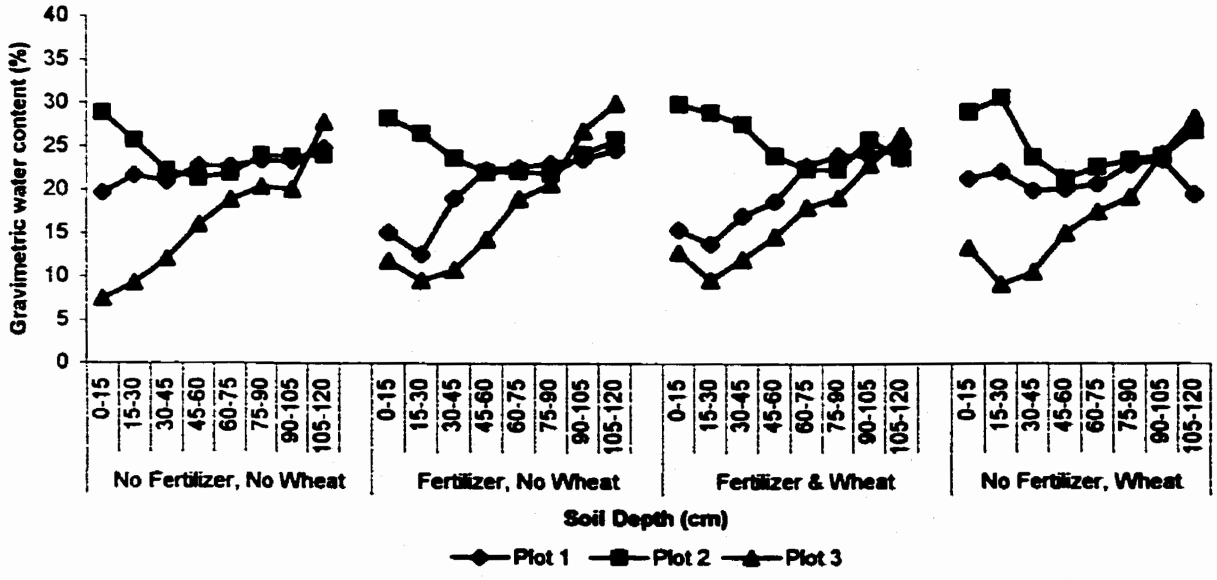

Figure 4.20 Distribution of moisture in the soil profile under different treatments for June 8, 1992.

previous fall.

Nitrate distribution patterns were similar for all plots by June 22, 1992, although actual levels of nitrates were still higher for plot 2 under the uncropped treatments (Figure 4.21). Crop uptake of nitrate resulted in the depletion of nitrate from all soil profile levels below 15 cm in the seeded treatments, with the exception of plot 3 under the NFW treatment. As in 1991, water table influences at this plot (the water table was at 120 cm depth, and the nitrate levels in the ground water were very high) had a profound effect on the distribution of nitrate in the unfertilized plots due to lateral movement of nitrate in the ground water. Chloride and nitrate distribution patterns were relatively similar for the fertilized subplots of plots 1 and 3 (Figure 4.22) with the exception of the 0-15 cm depth for plot 3 under the FW treatment. This may have been a result of variability caused by non-uniform fertilizer application (Cameron et al., 1979).

By July 21, nitrate levels in the soil profile had decreased for all treatments, but were still highest for plot 2 under the fallow treatments (Figure 4.23). Once again, there were nitrates at depth for plot 3 under the cropped treatments. Given the high nitrate concentration in the groundwater and the high water table, this was not unexpected, and in fact repeats trends exhibited by this plot in 1991. Chloride distribution for plots 1 and 3 largely mimic the nitrate distribution in the soil profile, with the exception of high levels of chlorides at the surface of the FW profiles for both plots (Figure 4.24). It is unclear as to why this would be the case. If considered individually, the high chloride levels at the surface of the FW treatment would not be so unexpected, as uptake of nitrate by wheat is higher than that of chlorides. However, the chloride levels

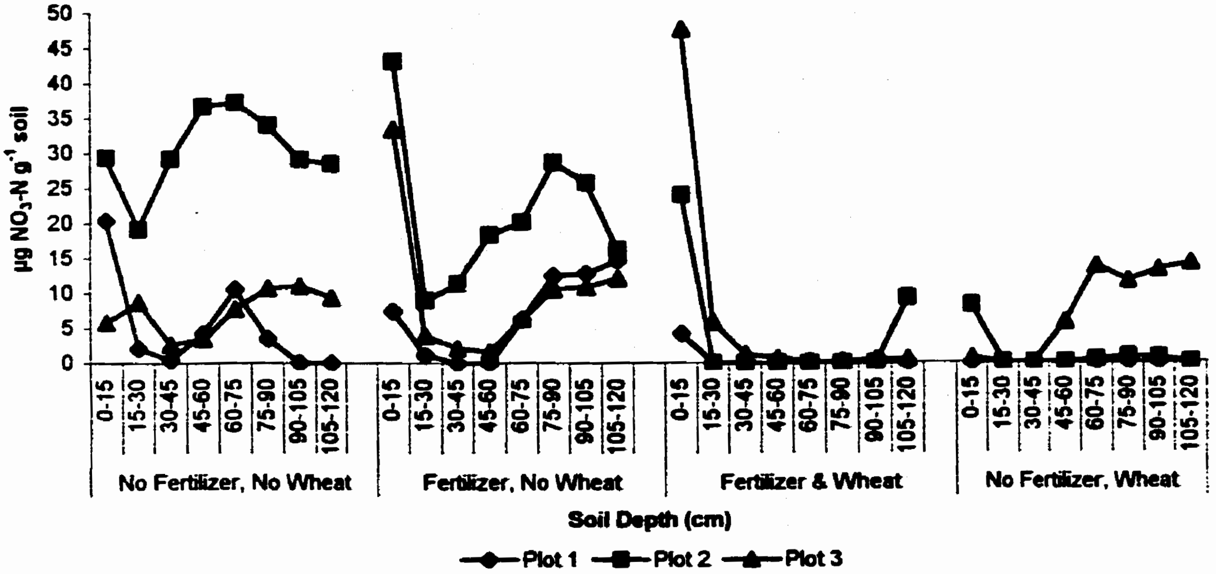

Figure 4.21 Distribution of nitrate in the soil profile as influenced by treatment for June 22, 1992.

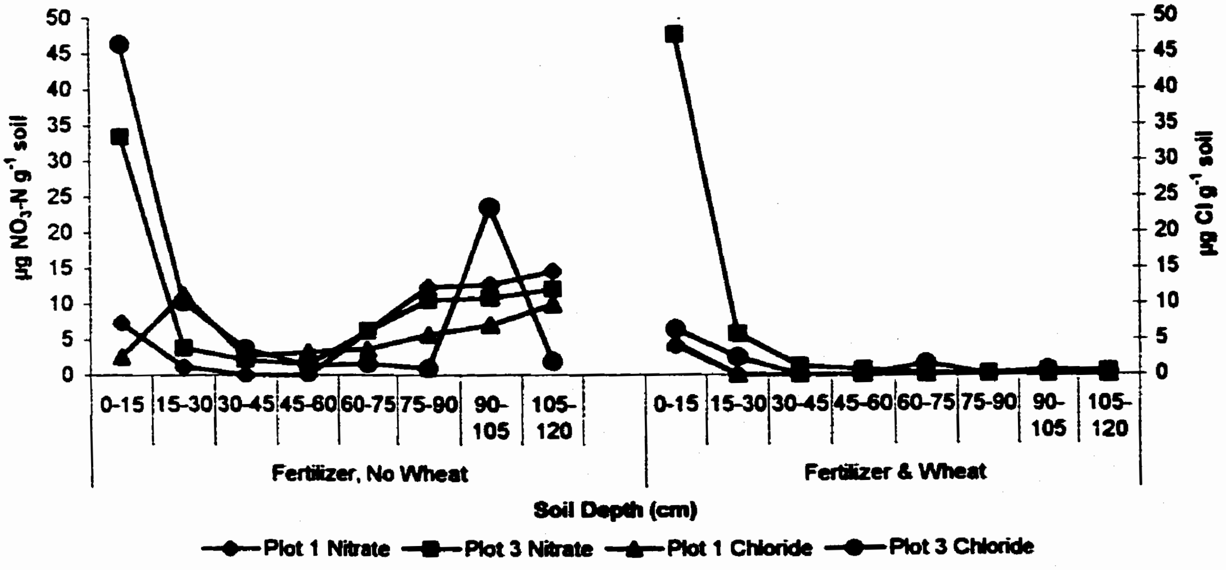

Figure 4.22 Distribution of chloride and nitrate under seeded and fallow conditions for June 22, 1992.

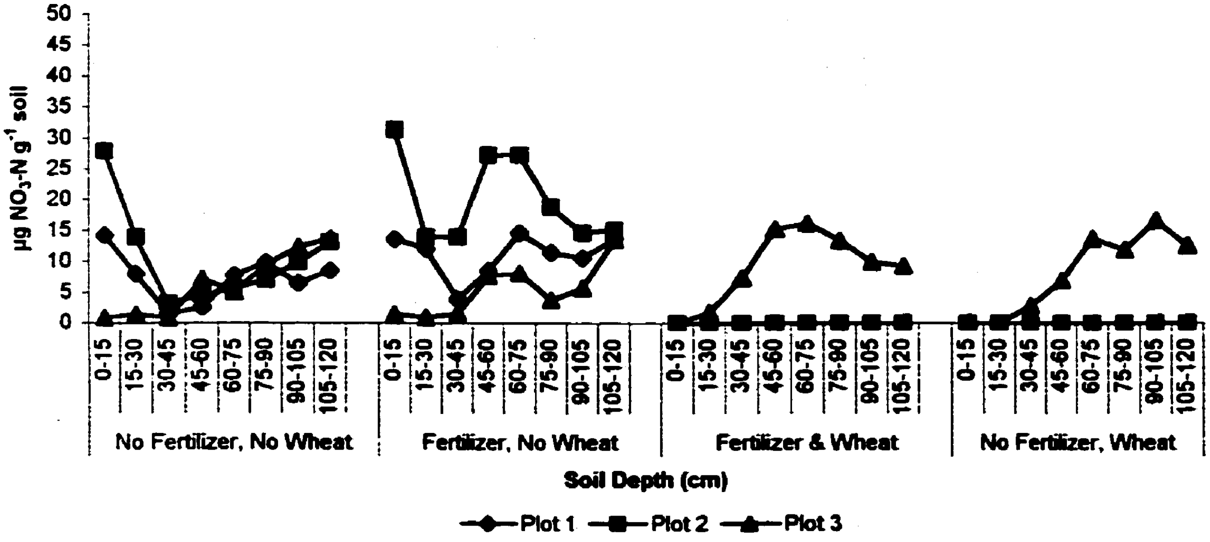

Figure 4.23 Distribution of nitrate in the soil profile as influenced by treatment for July 21, 1992.

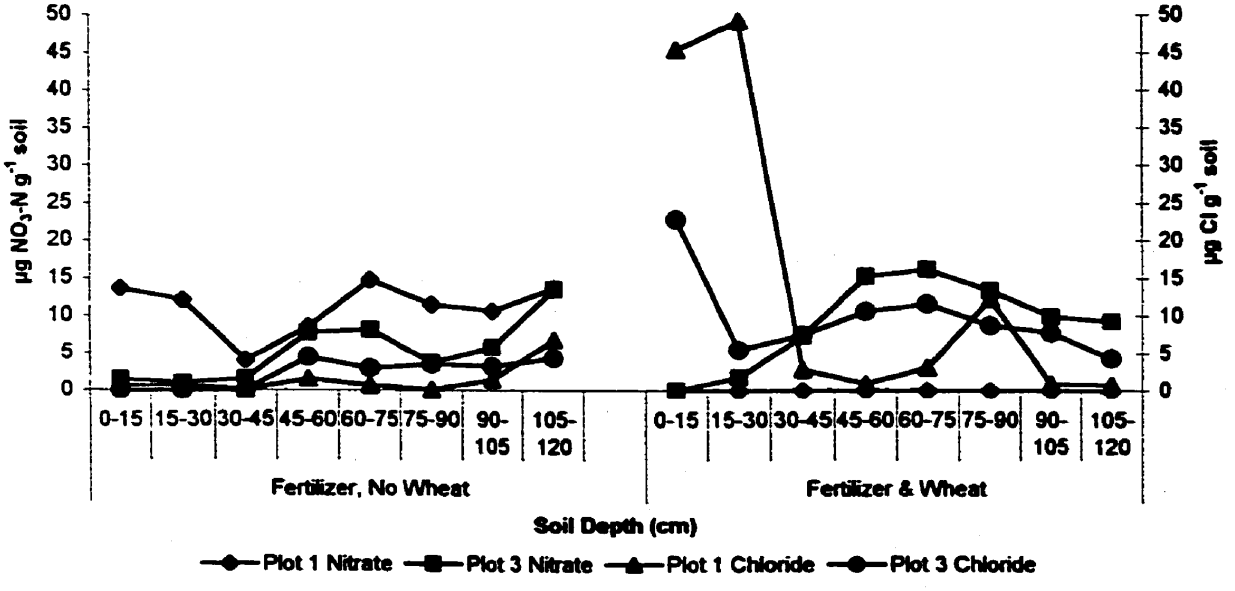

Figure 4.24 Distribution of chloride and nitrate under seeded and fallow conditions for July 21, 1992.

should then be at least as high for the FNW treatment, where there would be no uptake, and this was not the case. It is possible that the removal of soil water by the growing wheat created a localized moisture deficit, and the resulting capillary rise of soil water carried chlorides (and likely nitrates) near the surface horizons, where differential rates of nutrient uptake would result in residual chloride. However, this does not appear to be well reflected in the soil moisture distribution data (Figure 4.25).

For the sampling dates of August 5 & 6, 1992, nitrate concentrations in the fallow treatments had increased, particularly in the surface horizons (Figure 4.26). Although this could be an artifact of nitrate movement up the soil profile, the nitrate distribution patterns in the soil profiles do not match the moisture distribution patterns (Figure 4.27) or the chloride distribution patterns (Figure 4.28). It is therefore most likely that the cause of the nitrate increase near the soil surface was nitrification, following the most recent precipitation event (5 mm at plot 1 and 21 mm at plot 3). This echoes results for 1991, where nitrification also occurred in the mid summer months on the fallow treatments. A study conducted by Watson and Mills (1998), determined that there was substantial nitrification in soils receiving NH_4NO_3 fertilizer, particularly on those soils that had a history of high N inputs. During the incubation time of the study (21 days), the soil receiving NH_4NO_3 at the same rate as our plots (100 kg N ha^{-1}) had gross nitrification production equivalent to 44.0 kg N ha^{-1} (Watson and Mills, 1998). This pattern of increasing nitrate accumulation near the surface of the soil profile continued for plots 1 and 2 for the remainder of the growing season until the final sampling date of September 17, 1992 (Figure 4.29, 4.30 and 4.31). Plot 3 did not particularly follow this pattern,

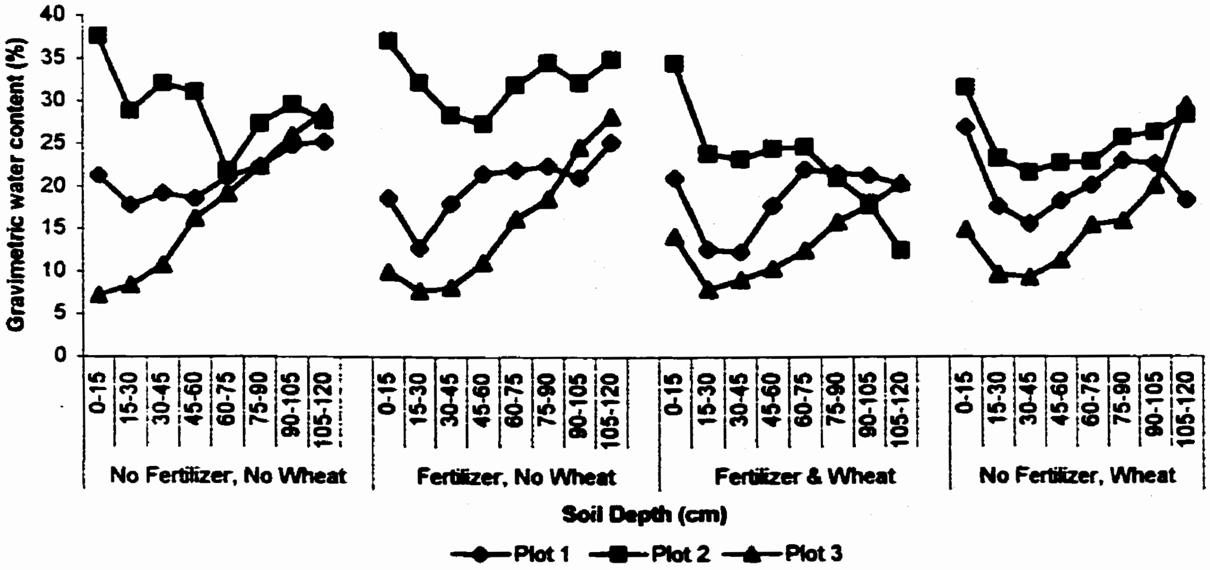

Figure 4.25 Distribution of moisture in the soil profile under different treatments for July 21, 1992.

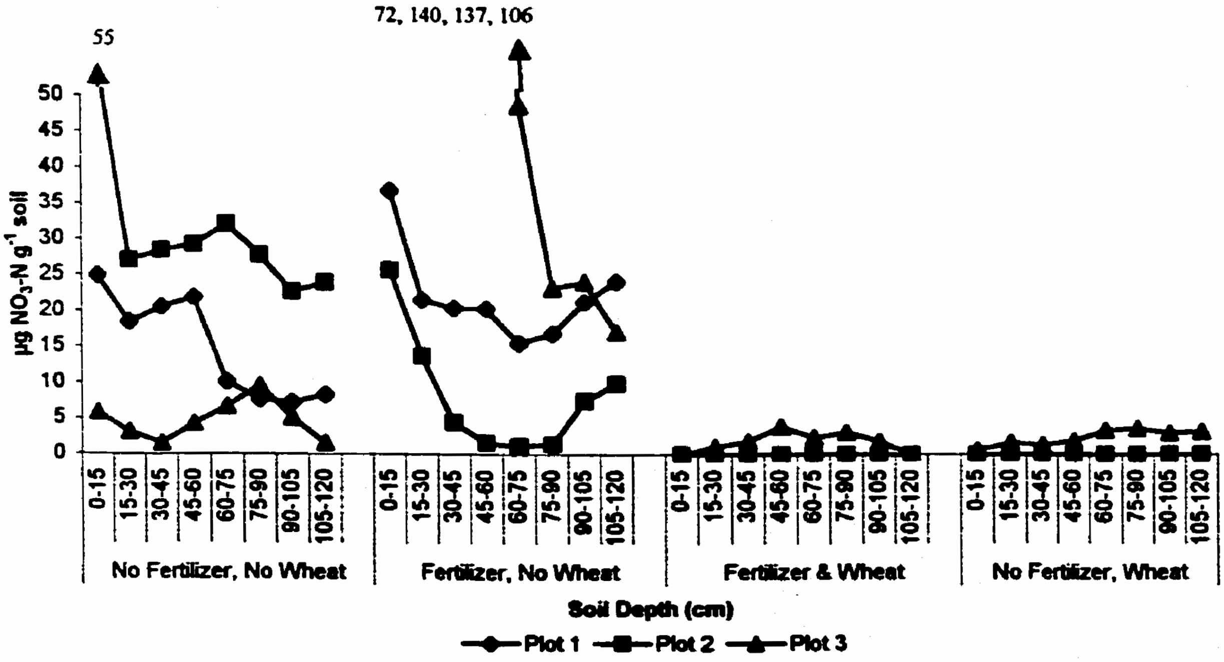

Figure 4.26 Distribution of nitrate in the soil profile as influenced by treatment for August 5-6, 1992.

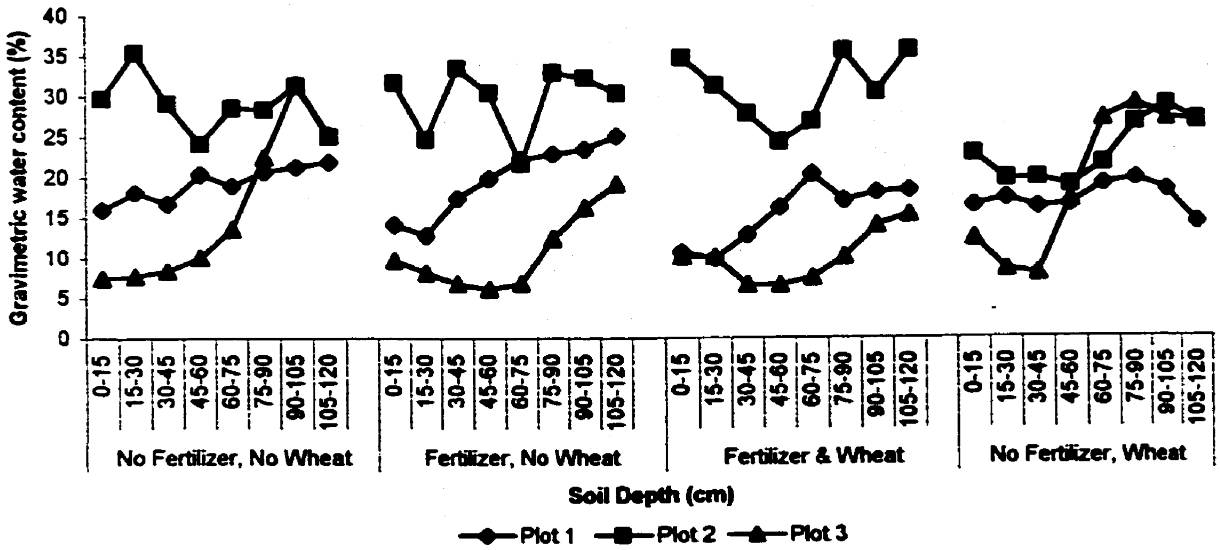

Figure 4.27 Distribution of moisture in the soil profile under different treatments for August 5-6, 1992.

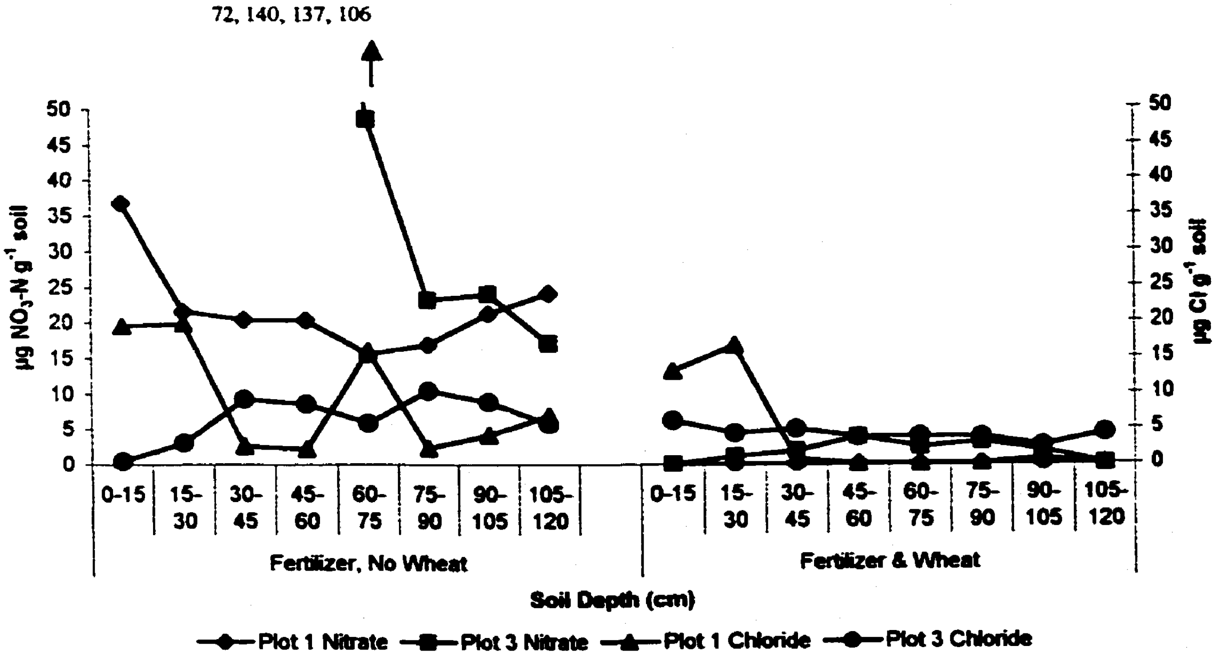

Figure 4.28 Distribution of chloride and nitrate under seeded and fallow conditions for August 6, 1992.

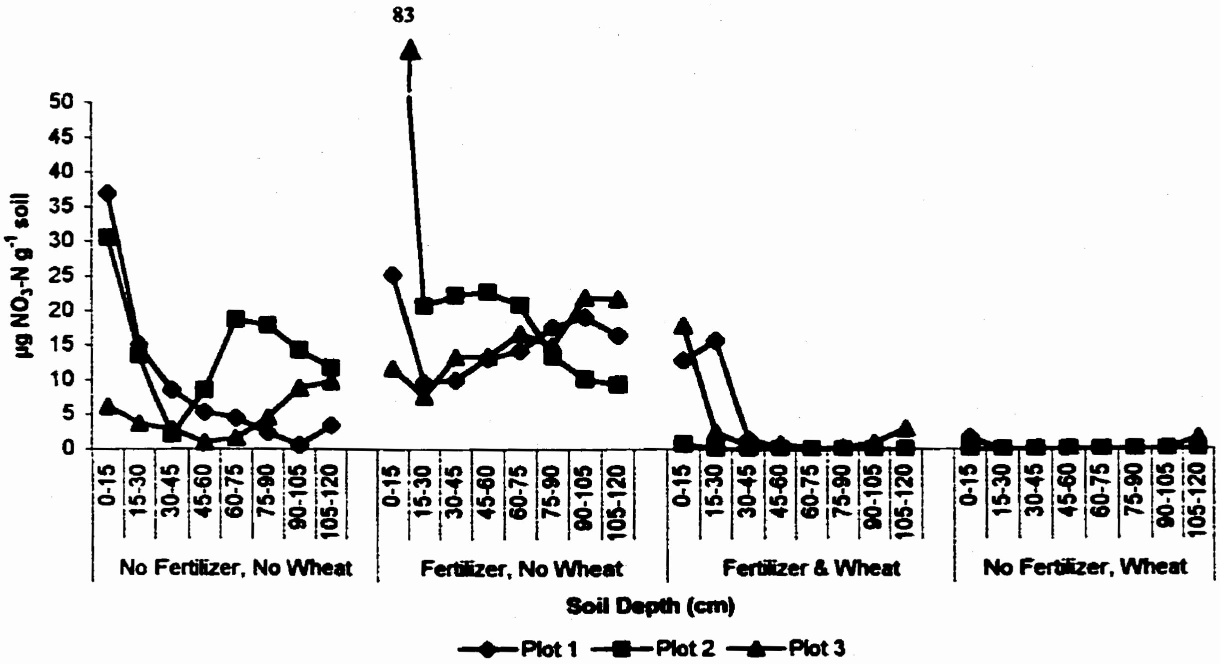

Figure 4.29 Distribution of nitrate in the soil profile as influenced by treatment for August 18-19, 1992.

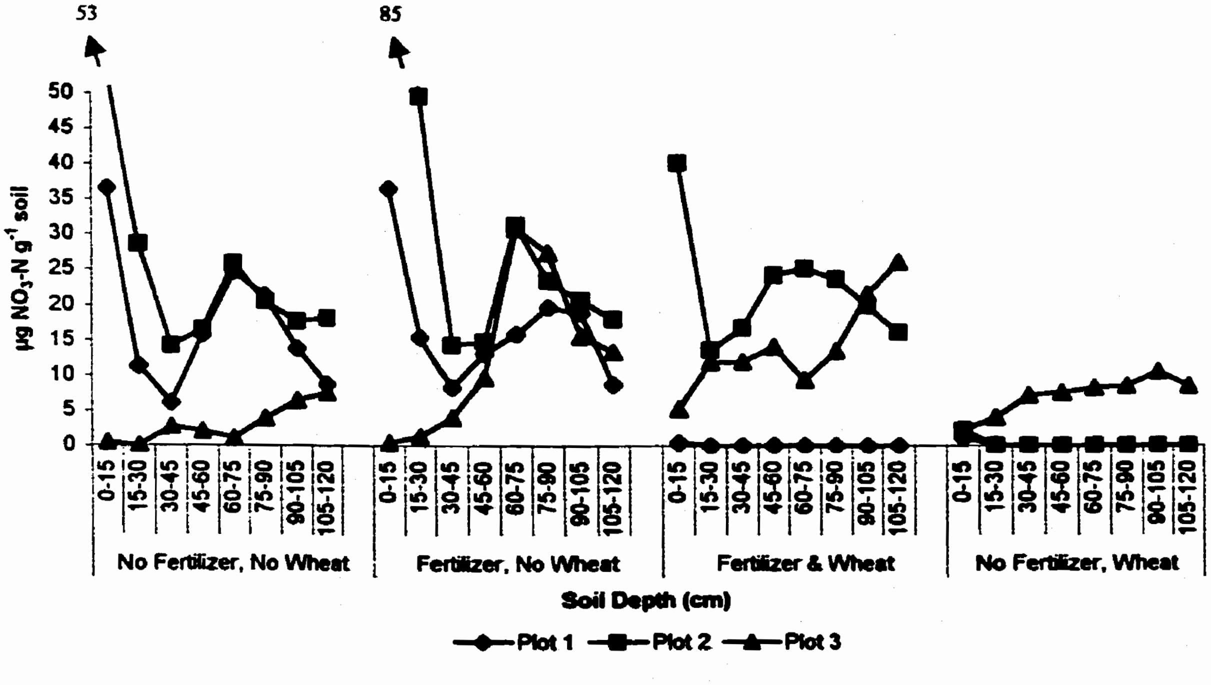

Figure 4.30 Distribution of nitrate in the soil profile as influenced by treatment for August 31-September 2, 1992.

although it did in the previous field year. This change in seasonal response may be due to the very low moisture content of the surface 15 cm of plot 3, which may have been less than optimum for nitrification in this particular soil (Figure 4.32). Examination of the chloride data for August 18 (Figure 4.33) and 31 (Figure 4.34) shows close relationships between chloride and nitrate distributions in soil, indicating that at least some of the increased nitrate near the soil surface is a result of upward movement of soil nutrients with ground water. The large flush in nitrate between the August 18 and August 31 sampling dates followed approximately 40 mm of precipitation, which likely stimulated nitrification under the uncropped treatments. This pattern of nitrate distribution for plots 1 and 2 is also seen on September 17, 1992, following a further 30 mm of precipitation. Plot 3 was not included for this final sampling date, as the plot was not sampled within a 2 day time period. Chloride data was also not evaluated for this date, so some effect of movement of nitrates in the profile can not be completely eliminated as a factor influencing the increase of nitrate near the surface of the soil profile.

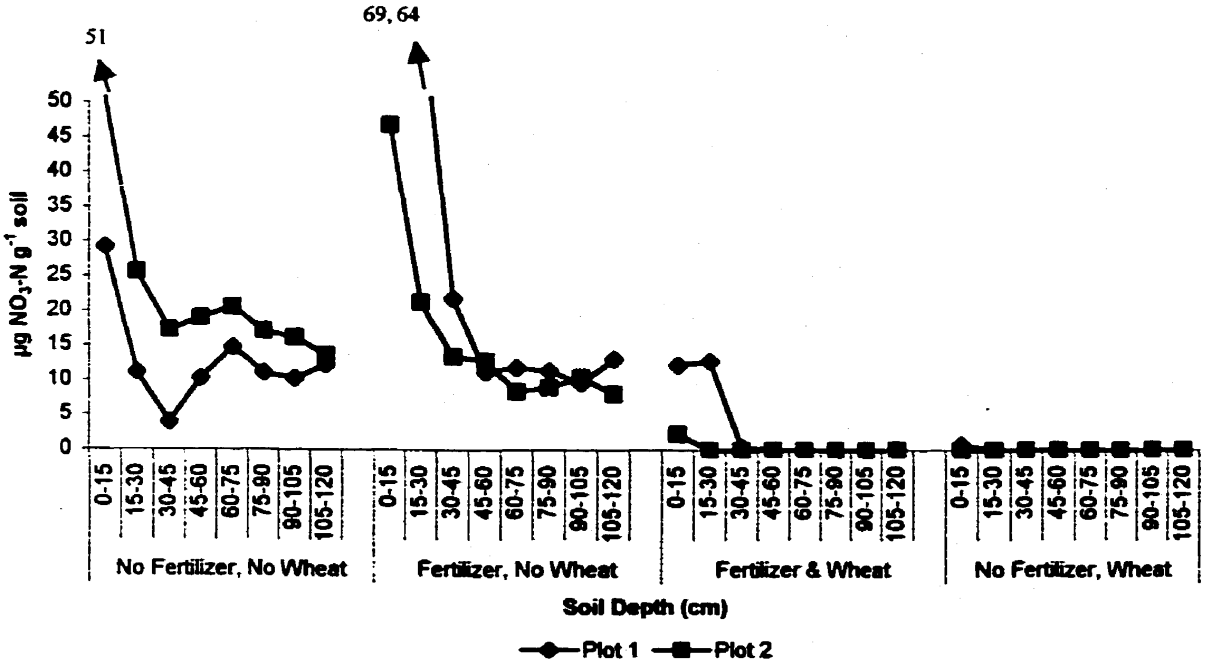

Figure 4.31 Distribution of nitrate in the soil profile under different treatments for September 17, 1992.

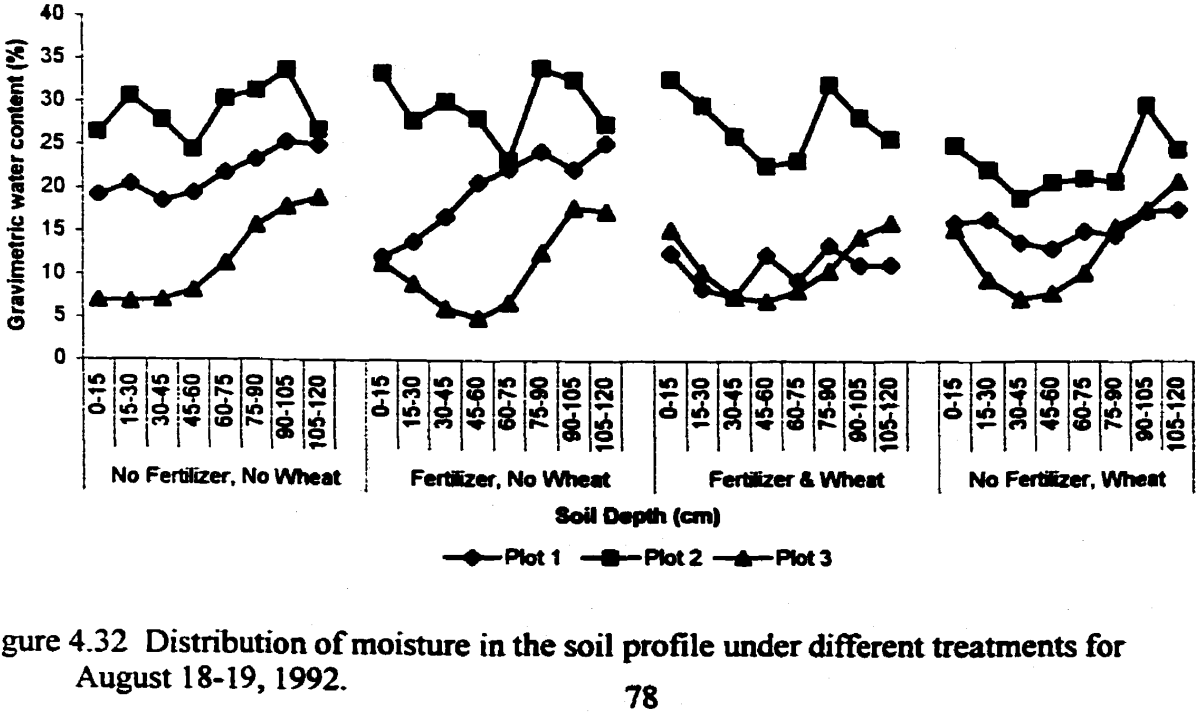

Figure 4.32 Distribution of moisture in the soil profile under different treatments for August 18-19, 1992.

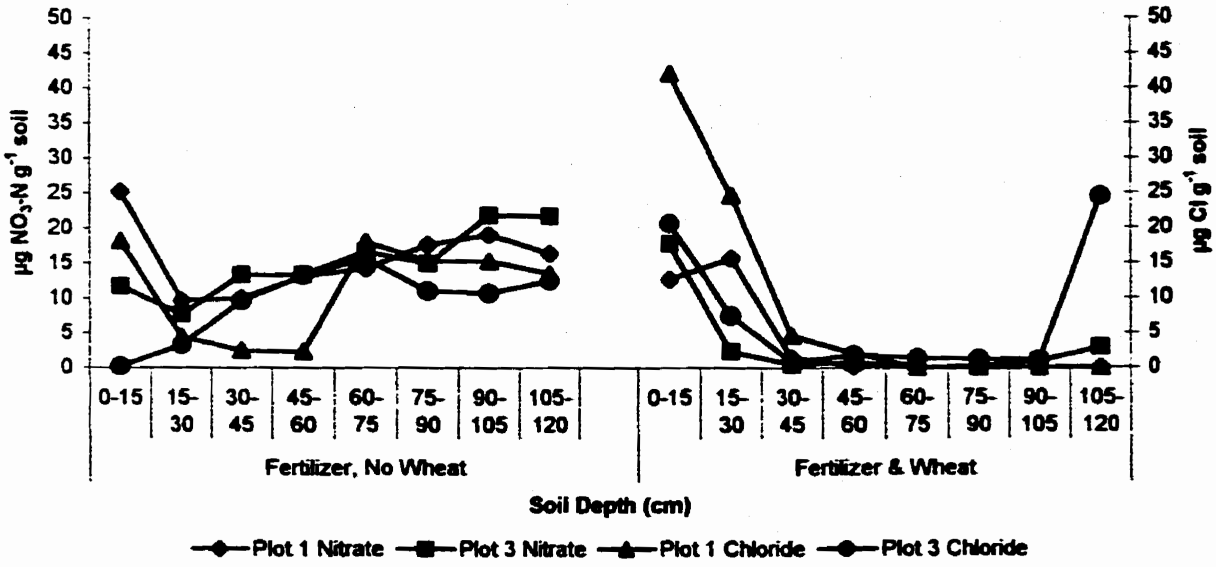

Figure 4.33 Distribution of chloride and nitrate under seeded and fallow conditions for August 18, 1992.

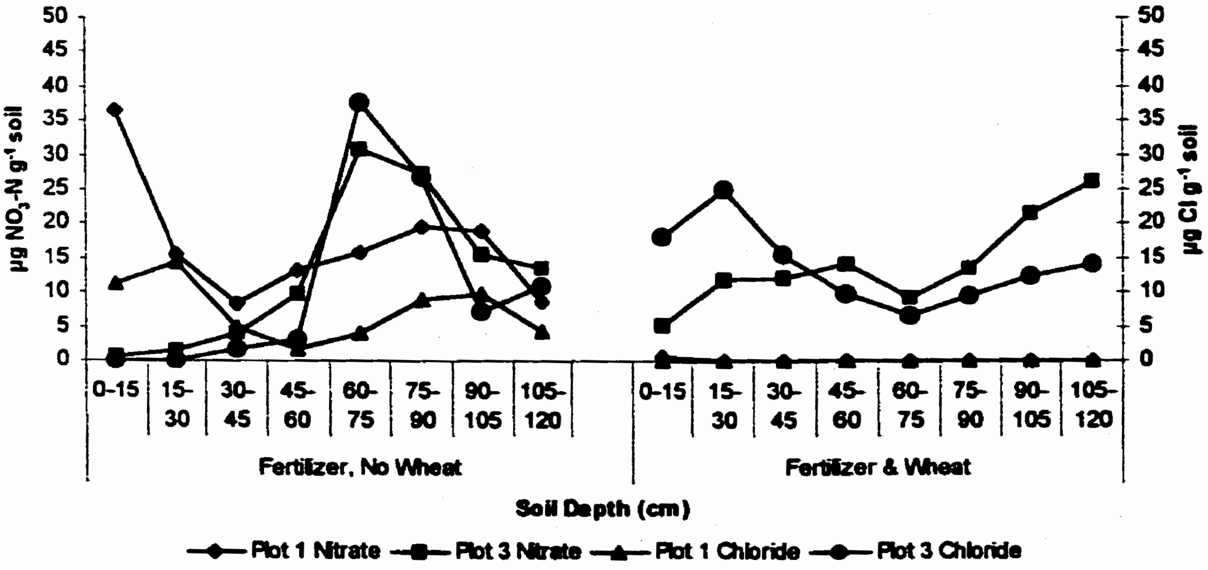

Figure 4.34 Distribution of chloride and nitrate under seeded and fallow conditions for August 31, 1992.

4.3 Fluctuations in Total Profile Nitrate Concentrations.

In an effort to determine whether there was a net loss or gain of nitrate nitrogen in the soil profile over the course of the growing season, the total amounts of nitrate were calculated for the soil profile for each of the four treatments over all three plots. Amounts were calculated on a basis of kg NO_3-N ha^{-1} using a profile depth of 105 cm and soil bulk densities as previously presented in Table 3.1. The full 120 cm sampling depth was not used, as soil bulk densities for the 105-120 cm depth could not be obtained for the profiles. For plot 3, soil bulk densities were only obtained for the top 90 cm, but the value of bulk density obtained for the 75-90 cm depth at this plot was used for the 90-105 cm depth as an approximate value for bulk density. The actual value could not be obtained at this plot as the soil texture and close proximity to the water table made it difficult to remove a core sample for bulk density measurement below the 90 cm soil depth. For plot 2, bulk density values were estimated from plot 1 measured values, as the soil type was the same, and these had not been measured in the field.

The results for all three plots for both years are presented in three separate tables (Tables 4.5, 4.6 and 4.7). Analysis of variance (ANOVA) was performed on the replicate data (as dictated by the common sampling dates) using JMP IN® software from SAS institute, and the effects of crop, fertilizer and crop x fertilizer interactions were examined for each year's data set. When all three plots were considered, there were no significant

Table 4.5 Total nitrate-nitrogen in the Stockton soil profile over two years- plot 1.

Date & Code[*]	No Fertilizer, No Wheat	Fertilizer, No Wheat	Fertilizer & Wheat	No Fertilizer & Wheat
	kg NO_3-N ha^{-1}			
S01 06/11/91	166.67	127.59	214.65	93.48
S02 06/27/91	132.17	122.54	79.52	5.37
S03 07/11/91	86.44	222.50	12.31	1.81
S04 07/23/91	79.49	102.94	29.43	10.39
S05 08/08/91	112.43	240.42	29.06	9.30
S06 08/19/91	158.92	165.35	18.06	13.09
S07 09/11/91	231.80	254.12	32.61	23.81
S08 09/25/91	247.73	133.08	63.16	17.91
S09 05/12/92	66.81	177.28	12.74	31.39
S10 05/27/92	59.05	144.94	44.35	12.05
S11 06/08/92	33.68	92.95	72.89	35.70
S12 06/22/92	66.15	64.20	6.53	0.00
S13 07/07/92	93.33	207.78	0.00	3.10
S14 07/21/92	81.46	122.36	0.00	0.00
S15 08/06/92	182.61	249.77	0.00	0.00
S16 08/18/92	118.65	177.12	48.89	2.57
S17 08/31/92	210.11	207.19	0.73	1.90
S18 09/17/92	147.09	323.77	41.21	1.15

[*] The Date Code indicates which sampling dates in Tables 4.5, 4.6 and 4.7 were used to statistically analyze treatment effects. Only dates which were assigned a code were used, and dates with the same code were considered to be replicates.

Table 4.6 Total nitrate-nitrogen in the Wellwood soil profile over two years- plot 2.

Date & Code*		No Fertilizer, No Wheat	Fertilizer, No Wheat	Fertilizer & Wheat	No Fertilizer & Wheat
		kg NO_3-N ha^{-1}			
S01	06/11/91	79.51	284.80	103.52	70.56
S03	07/11/91	62.88	84.51	17.06	1.52
S04	07/23/91	228.18	194.93	106.70	24.25
S05	08/08/91	176.34	265.84	61.55	63.34
S06	08/19/91	157.83	218.30	34.75	8.77
S07	09/11/91	147.49	123.35	17.22	24.76
S08	09/25/91	335.47	288.20	131.32	37.16
S09	05/12/92	166.94	307.26	104.55	59.86
S10	05/27/92	334.84	262.48	64.94	37.00
S11	06/08/92	453.85	164.64	445.30	131.56
S12	06/22/92	352.38	253.32	37.44	15.44
S13	07/07/92	254.50	57.79	4.74	0.00
S14	07/21/92	116.93	241.04	0.00	0.00
S15	08/06/92	363.42	89.53	0.00	0.00
S16	08/18/92	170.67	312.85	1.19	0.00
S17	08/31/92	287.20	387.45	265.93	3.45
S18	09/17/92	270.84	198.13	3.62	0.00

*The Date Code indicates which sampling dates in Tables 4.5, 4.6 and 4.7 were used to statistically analyze treatment effects. Only dates which were assigned a code were used, and dates with the same code were considered to be replicates.

Table 4.7 Total nitrate-nitrogen in the Almasippi soil profile over two years - plot 3.

Date & Code[*]	No Fertilizer, No Wheat	Fertilizer, No Wheat	Fertilizer & Wheat	No Fertilizer & Wheat
	kg NO_3-N ha^{-1}			
S01 06/13/91	137.02	582.96	603.03	326.92
S02 06/27/91	131.31	378.50	545.70	314.15
07/04/91[a]	38.76	792.16	768.71	93.26
07/10/91[a]	31.57	239.65	220.20	91.51
07/16/91[b]	110.77	178.81	408.57	186.02
07/22/91[c]	79.92	168.73	114.59	113.93
07/30/91	77.23	295.57	351.03	324.72
S05 08/07/91	168.17	569.77	61.98	36.23
08/15/91	125.46	395.37	157.14	183.26
S06 08/22/91	113.48	334.01	378.06	279.18
08/28/91	132.48	277.39	679.06	296.07
09/05/91	145.79	299.62	380.78	215.71
S07 09/12/91	126.95	263.16	372.30	493.16
09/19/91	115.04	475.08	205.79	356.71
10/03/91	150.42	345.67	253.84	200.23
05/06/92	198.57	319.23	271.96	203.89
S09 05/13/92	133.40	132.08	3.42	323.46
S10 05/25/92	123.31	319.29	12.80	116.52
06/02/92	150.04	495.62	58.33	175.57
S11 06/08/92	117.61	325.68	321.85	205.14
06/16/92	93.80	169.26	273.69	330.16
S12 06/22/92	135.56	170.09	103.50	138.72
06/29/92	84.59	219.65	31.98	250.97
S13 07/07/92	149.55	674.58	198.63	213.26
07/14/92	150.00	398.83	149.21	129.88
S14 07/21/92	119.95	99.19	170.14	152.19
07/28/92	77.33	296.01	29.02	27.06
S15 08/05/92	84.73	1258.02	32.39	43.51
08/11/92	90.99	319.11	94.51	28.05
S16 08/19/92	87.76	277.33	48.70	6.30
08/26/92	130.89	280.14	16.73	15.22
S17 09/02/92	58.03	240.22	263.30	131.40
09/10/92	97.22	173.31	81.30	12.23
09/22/92	86.40	393.51	46.96	73.57

[*] The Date Code indicates which sampling dates in Tables 4.5, 4.6 and 4.7 were used to statistically analyze treatment effects. Only dates that were assigned a code were used, and dates with the same code are replicates.

a - based on 0-60 cm soil depth
b - based on 0-75 cm soil depth
c - based on 0-90 cm soil depth

interaction effects, and only one date on which there was a significant difference ($p < 0.05$) between the fertilized and unfertilized treatments (August 18, 1992) (Table 4.8). Under crop effects, there were only four dates that were significantly different at the $p<0.05$ level: July 11 and August 8 in 1991, and May 27 and August 18 in 1992. As the proximity to the water table at the third plot had profound effects on the nitrate distribution, and ultimately nitrate content of that soil profile, it was not considered a good replicate. The analyses were, therefore, run a second time for each year, this time without considering the data from plot 3, and these results are presented in Table 4.9. Once again, there were no significant effects ($p> 0.05$) of crop x fertilizer interactions on total nitrate in the soil profile for either year, and only one date in 1992 (August 18) that showed significance ($p<0.05$) for fertilizer effects. For 1991, there was a marked effect of crop on total nitrate content in the soil profile that was expressed later in the season. In this case, profile nitrate contents from July 11 to September 25, 1991, were significantly lower under the treatments with crop, regardless of whether the subplot received a fertilizer application. For 1992, the May 12 and 27 sampling dates, which occurred pre-fertilization for that year, showed significant crop effects at the $p<0.10$ level, which were likely a reflection of the previous year's treatments. Sampling dates from July 21, 1992 to September 17, 1992 had significantly higher soil nitrate concentrations in the uncropped treatments than in the crop treatments at the level of $p<0.05$. This echoed the results from 1991.

Table 4.8 Effect of treatment on profile nitrate-nitrogen content - summary of response to ANOVA using the means of all three plots.

Date Code	Wheat			Fertilizer			Wheat* Fertilizer
	+	-	Effects	+	-	Effects	Effects
	Mean — kg NO_3-N ha^{-1} —			Mean — kg NO_3-N ha^{-1} —			
S01	235±204	230±186	NS	319±222	146±96	NS	NS
S02[a]	236±245	191±125	NS	282±220	146±127	NS	NS
S03	8±8	114±73	*	84±98	38±43	NS	NS
S04	43±43	151±71	**	109±68	86±100	NS	NS
S05	66±55	255±163	*	205±205	116±68	**	NS
S06	94±144	191±77	NS	191±149	94±68	NS	NS
S07	100±140	191±65	NS	177±142	114±79	NS	NS
S08[a]	62±50	251±86	*	154±95	160±157	NS	NS
S09	58±52	164±80	NS	123±113	99±53	NS	NS
S10	49±42	207±114	*	141±125	115±117	NS	NS
S11	187±160	198±160	NS	237±149	148±156	NS	NS
S12	50±56	174±113	**	106±92	118±129	NS	NS
S13	59±90	240±225	NS	191±254	108±98	NS	NS
S14	48±77	130±57	NS	105±95	73±58	NS	NS
S15	20±34	371±447	**	272±492	119±137	NS	NS
S16	32±36	191±88	*	144±131	78±67	*	NS
S17	99±130	232±108	NS	227±127	103±118	NS	NS
S18[a]	11±20	235±78	*	142±148	105±130	NS	NS

a - based on two plots only; .
** - significant at $p<0.10$;
NS - not significant at $p<0.10$;
* - significant at $p<0.05$.

Table 4.9 Effect of treatment on profile nitrate-nitrogen content - summary of response to ANOVA using plots 1 and 2.

Date Code	Wheat			Fertilizer			Wheat* Fertilizer
	+	-	Effects	+	-	Effects	Effects
	Mean —— kg NO_3-N ha^{-1} ——			Mean —— kg NO_3-N ha^{-1} ——			
S01	121±64	165±88	NS	183±83	103±62	NS	NS
S03	8±8	114±73	*	84±98	38±42	NS	NS
S04	43±43	151±71	**	109±68	86±107	NS	NS
S05	41±26	199±69	*	149±121	90±78	**	**
S06	19±11	175±29	*	109±98	85±74	NS	NS
S07	25±6	189±64	*	107±109	107±109	NS	NS
S08	62±50	251±86	*	154±95	160±158	NS	NS
S09	52±40	180±99	**	150±124	81±66	NS	NS
S10	40±22	200±122	**	129±99	111±143	NS	NS
S11	171±187	186±186	NS	194±172	164±213	NS	NS
S12	15±16	184±143	NS	90±111	108±143	NS	NS
S13	2±2	153±93	**	68±97	88±117	NS	NS
S14	0±0	140±69	*	91±116	50±58	NS	NS
S15	0±0	221±115	*	85±118	137±174	NS	NS
S16	13±24	195±83	*	135±140	73±86	NS	NS
S17	68±132	273±85	**	215±162	126±146	NS	NS
S18	11±20	235±78	*	142±148	105±129	NS	NS

NS - not significant at $p<0.10$. * - significant at $p<0.05$. **- significant at $p<0.10$.

The effects of crop on nitrate content in the soil profile may be simply an expression of nutrient uptake by the crop, as wheat generally will remove approximately 95 kg N ha^{-1} from the soil (Soil Fertility Guide, Manitoba Agriculture, 2000). However, this should still result in a difference in nitrate levels at the end of the growing season between fertilized and unfertilized subplots, unless the wheat took up all of the additional nitrogen added, or denitrification was occurring. There appeared to be enough residual nitrate in the soil at the beginning of the growing season to support a crop on the NFW treatment, and there were no significant differences between the fertilized and unfertilized subplots at the end of the growing season.

Given the very high levels of nitrate in the NWNF treatments at the end of the growing season, it appears that nitrification played a significant role in supplying additional nitrate to the soil profile under uncropped conditions. This is not particularly surprising since the NH_4^+ applied to the soil for these treatments would not have been taken up by plants, and was therefore likely available for nitrification. The fact that nitrification was occurring under the uncropped systems suggests that given the proper moisture, aeration and temperature conditions, denitrification was likely occurring to some degree under all treatments, particularly in the cropped treatments where carbon additions from root exudates would stimulate denitrification (Bailey, 1976). Therefore, crop uptake would not be solely responsible for nitrate depletion under the cropped treatments. Both of the cropped treatments for plots 1 and 2 differed in 1992 in that nitrate was undetectable in the two profiles on numerous sampling dates, whereas the nitrate was never completely depleted from the soil profiles on any date in 1991. This would be expected on the unfertilized subplot, where nitrate had not been added for two years, but slightly unexpected for the fertilized subplot, which would have received just enough nitrogen to support the crop for the duration of the growing season. Considered with the residual nitrate in the profile from the previous season, there should have been sufficient nitrogen in the soil profile to support the crop without going to depletion. This may, therefore, be an indication of the occurrence of denitrification under the cropped treatments.

Analysis of variance (ANOVA) was performed on the replicate chloride data for plots 1 and 3 (as dictated by the common sampling dates in 1992) using JMP IN® software from SAS institute, and the effects of crop on the soil chloride contents of the fertilized subplots were examined. Although plot 3 was not considered an ideal replicate due to the influence of the high water table, chloride contents were not measured for plot 2, so the replicates used were plots 1 and 3 (Table 4.10). Unlike the case for the nitrate contents of the profiles, where there were distinct crop effects, there were no significant differences ($p>0.10$) in profile chloride contents between the no wheat and the wheat subplots. This was expected, as chloride is not subject to biological changes, and spring wheat will typically only remove 5.6 kg Cl^- ha^{-1} per growing season (Soil Fertility Guide, Manitoba Agriculture, 2000), while the application rate of Cl^- was 90 kg ha^{-1}. The fact that chloride was present at high levels throughout the soil profile illustrates the extent of leaching that occurred in the soils used in this study.

Table 4.10 Total chloride content and nitrate-nitrogen: chloride ratios in the fertilized subplots of plots 1 and 3 to a depth of 105 cm.

Plot	**Sample date**	**Fertilizer, No Wheat**	**Fertilizer & Wheat**	**Fertilizer, No Wheat**	**Fertilizer & Wheat**
		kg Cl ha^{-1}		**NO_3-N:Cl**	
1*	27/05/92	108.48	81.01	1.34	0.55
1	08/06/92	250.38	145.61	0.37	0.50
1	22/06/92	59.69	7.01	1.08	0.93
1	07/07/92	0.0	77.46	-	0.00
1	21/07/92	8.28	186.31	14.78	0.00
1	06/08/92	108.80	50.76	2.30	0.00
1	18/08/92	122.79	118.51	1.44	0.41
1	31/08/92	89.46	0.14	2.32	5.20
1	17/09/92	48.33	183.05	6.70	0.23
		kg Cl ha^{-1}		**NO_3-N:Cl**	
3**	02/06/92	579.44	291.57	0.86	0.20
3	08/06/92	432.42	170.30	0.75	1.89
3	16/06/92	332.60	135.28	0.51	2.02
3	22/06/92	182.99	22.82	0.93	4.53
3	29/06/92	63.23	83.68	3.47	0.38
3	07/07/92	176.85	240.76	3.81	0.83
3	14/07/92	82.03	164.90	4.86	0.90
3	21/07/92	43.26	169.57	2.29	1.00
3	28/07/92	131.03	64.62	2.26	0.45
3	05/08/92	122.69	75.00	10.25	0.43
3	11/08/92	148.69	78.61	2.15	1.20
3	19/08/92	177.67	126.54	1.56	0.38
3	26/08/92	166.57	107.31	1.68	0.16
3	02/09/92	204.79	247.71	1.17	1.06
3	10/09/92	131.11	88.79	1.32	0.92
3	22/09/92	219.22	70.91	1.80	0.66

* Fertilizer was applied at a rate of 90 kg Cl^- ha^{-1} after sampling on May 27, 1992.

** Fertilizer was applied at a rate of 90 kg Cl^- ha^{-1} after sampling on May 25, 1992.

Often, NO_3-N: Cl^- ratios are used to qualitatively determine if leaching, nitrification or denitrification is occurring in the soil profile (Weil et al., 1990; Misra and Mishra, 1976). In our study, the application ratio of nitrogen to chloride was approximately 1.1, while the application of nitrate-nitrogen:chloride was 0.55. If we assume that the ammonium applied as fertilizer eventually nitrified, then the ratio that we would use as our standard would be approximately one. Therefore, one could assume that if the ratio remains the same, or close to 1, there is little biological activity occurring involving nitrate as the distribution of chloride and nitrate within the soil profile is similar to the application rates. If the ratio is less than 1, it could be hypothesized that denitrification is likely occurring, or in the case of the FW treatment, that nitrogen uptake into the plant has occurred. (Wheat uptake of nitrate is much greater than chloride, approximately 17 to 1 as estimated using data in the Soil Fertility Guide, Manitoba Agriculture, 2000). If the ratio is greater than 1, nitrification and mineralization are likely contributing nitrate to the soil system.

Although not statistically significant, trends in the data could be observed (Table 4.10). Nitrate-chloride ratios were greater than one for the FNW treatment after June 22, 1992 for plot 3 and on all sampling dates except for June 8, 1992 for plot 1. This indicates that net nitrification may be occurring under this treatment beginning at least by June, 1992. For the FW treatment on plot 3, the ratios were rarely greater than 1.1, and only once after the June 22 sampling date. At plot 1, the ratios were greater than one only once under the FW treatment, on August 31, 1992. These values could be expected where nitrogen uptake and/or denitrification would be occurring. Therefore, although not statistically significant, differences in the nitrate-chloride ratios appear to support the seasonal nitrate fluctuations.

The actual total concentrations of Cl^- within the soil profiles fluctuated over time, and did not appear to differ between cropped and uncropped treatments, except for shortly after the fertilization date (Table 4.10). These differences in Cl^- concentrations within the soil profiles may in part be due to spatial variability in fertilizer application, or nonuniform leaching (Cameron et al., 1979). Lund (1982) concluded that spatial variations in nitrate and chloride distributions were related to field and soil characteristics in only a few cases, and often had no relation to these characteristics at all. The variation may also be due to movement of chloride in and out of the sampling zone, so that if chloride had moved below 105 cm, the profile Cl^- content would be low for a particular sampling date. If, by a subsequent date, chloride had moved back up the profile with soil water, the chloride would again be present in the soil profile depths used to calculate total Cl^- profile concentration. Therefore, temporal variations of total Cl^- and NO_3-N would be more prominent on those soils sampled to a lesser depth (Lund, 1982). In addition, differential anion uptake by plants could play a role in both temporal and spatial variations in Cl^- profile contents, as there can be a negative interaction between Cl^- and NO_3-N uptake: as the nitrate content of the soil water decreases, the uptake of chloride by the plant will increase (Tisdale et al., 1985). Furthermore, application of chlorides to soils at rates of 50 kg Cl^- ha^{-1} can reduce uptake of nitrate by wheat (Mohr, 1992), which also complicates the use of ratios under seeded treatments.

4.4 Denitrification Studies

For each of the field sampling depths for the Stockton loamy fine sand (plot 1) and the Almasippi sand (plot 3) profiles, five to eight sub samples were incubated for periods varying from six hours to fourteen days, depending upon the incubation temperature. For instance, incubation times at 5°C ranged from 2 to 14 days, whereas incubation times at 26.5°C ran from 6 hours to 4 or 6 days, depending on the sample depth. This was done to ensure that carbon did not become limiting to the denitrification process for soil depths where more rapid rates of denitrification could be expected. Under lower temperatures, lag times for increase in biological activity following rewetting of the air dried soil seemed to persist beyond the generally held expectation of 4-6 hours (Smith and Tiedje, 1979; Dendooven and Anderson, 1994), as evidenced by a slight increase in nitrate-nitrogen in the closed system for up to 24 hours following onset of incubation.

For example, for the Stockton soil, 30-45 cm depth at 5°C, six soil slurries were placed on the shaker in the incubation chamber. One sample was removed at each of 2, 4, 6, 8, 10 and 14 days and destructively sampled. The nitrate concentration of each soil slurry was determined and plotted as a function of time for the construction of one graph (Figure 4.35). This step was performed for every profile depth at each temperature studied. The slope of each graph ($\mu g\ NO_3\text{-}N\ g^{-1}\ d^{-1}$) gave the rate of nitrate disappearance

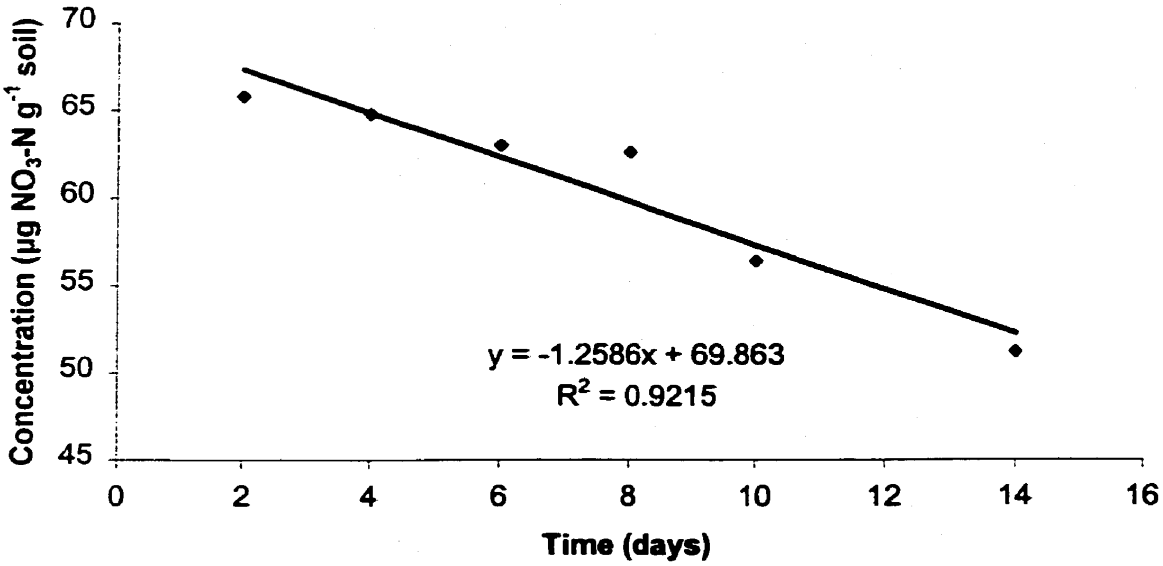

Figure 4.35 Nitrate disappearance rate of Stockton sandy loam at 5°C for 30-45 cm soil depth.

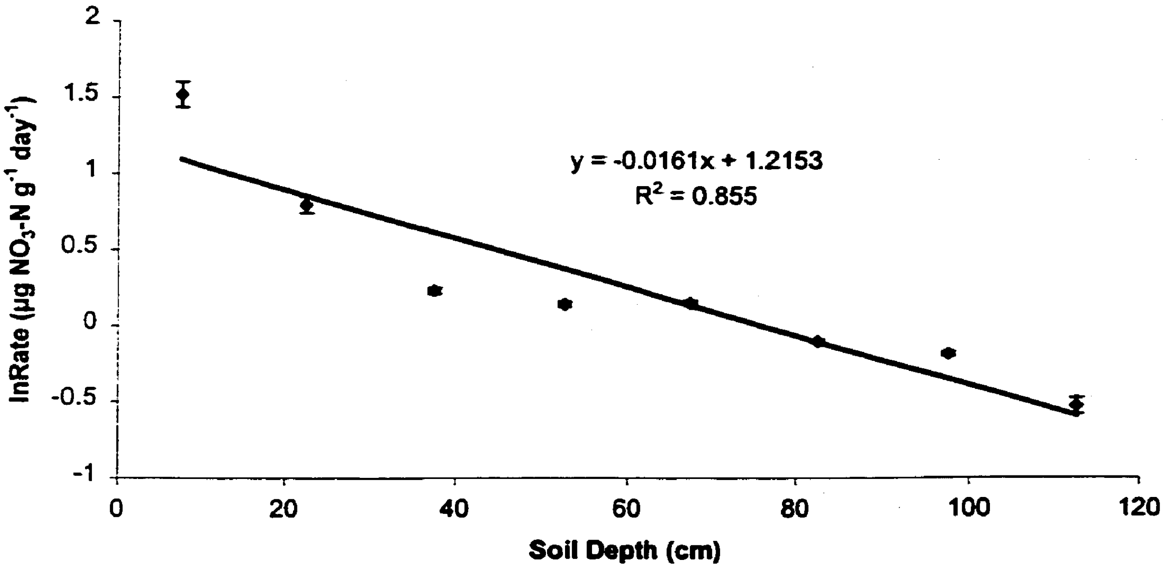

Figure 4.36 Semi-log plot of nitrate disappearance rate of the Stockton loamy fine sand profile at 5°C.

(NDR) at that particular soil depth and temperature. For the specific example of the 30-45 cm Stockton profile depth at 5°C, the rate of nitrate disappearance was calculated to be 1.26 μg NO_3-N g^{-1} soil day^{-1}. Determination of the nitrate disappearance (NDR) rate of the soil profile at each of the temperatures under investigation required the creation of 80 individual graphs (5 temperatures x 8 profile depths x 2 soils). Combined, these graphs represent the nitrate-nitrogen concentrations of more than 500 individual soil slurries (6-8 individual samples per graph). The rates of nitrate disappearance (slopes of the graphs), which were temperature and depth dependent, were then plotted in a semi-log linear manner as a function of soil depth for each temperature under study (Figure 4.36). This procedure was performed for both the Almasippi and Stockton soils at five temperatures: 5°C, 11°C, 16.5°C, 20.5°C and 26.5°C (Appendix X).

In this study, nitrate disappearance rate was chosen to provide a measure of denitrification rate in the soils because it was a less complex and faster method than others in use. Although this method is not as widely used as nitrous oxide production measurements, it is still a valid determination of denitrification potential in soils (Abbasi and Adams, 1998), provided that conditions for preferrential reduction of nitrate are supplied (Cho and Sakdinan, 1978). To elaborate, NO_3^- has been shown to be reduced preferentially over N_2O under reducing conditions, provided that adequate NO_3^- exists in the system, and N_2O is able to diffuse away from the site of denitrification once it is formed (Cho and Sakdinan, 1978). These conditions can be adequately met by providing sufficient nitrate substrate for the reaction to occur, and shaking soil slurries to ensure a steady supply of nitrate to sites of reduction (Cho and Sakdinan, 1978). Under these conditions, nitrous oxide would not be reduced until all nitrate was removed from the system. Nitrate disappearance would then be a reasonable estimate of denitrification occurring in the soil slurry.

Background denitrification potential is generally considered to be the maximum possible denitrification rate that could be obtained on a particular soil with no C or N amendments (Drury et al., 1991). Although nitrates were added to the soil slurries in this study, it should be noted that the rates of nitrate disappearance determined for soils in this study are in actual fact, the background denitrification potentials of these soils. While the presence of nitrate in a system may stimulate denitrification and is certainly necessary for denitrification to proceed, the rate of denitrification is not affected by the level of nitrates present in the system, provided that the nitrate substrate is not limiting (Cho and Sakdinan, 1978). In other words, there should be enough nitrate in the system for the reduction of nitrate to proceed, but additional nitrate will not increase the rate of nitrate disappearance (Ryden, 1983). This is not true for carbon amendments. The addition of readily available carbon sources can result in denitrification rate increases of up to 5-10 times the levels prior to carbon additions (Jordan et al., 1998; Drury et al., 1991). Therefore, no carbon amendments were used in this study, in order to more accurately determine in-field nitrate disappearance rates. As the rates in this study are not being amplified in any way, values determined for nitrate disappearance rates in this study could therefore be considered to be potential background denitrification rates. Hence, these rates are reasonable estimates of denitrification rates in the two soil profiles studied under saturated conditions.

If a readily available carbon source had been added to this system, the resulting nitrate disappearance rate would have been a determination of denitrification potential of the soil. The denitrification potential has often been defined as the maximum possible rate of denitrification that a soil can attain with the assistance of amendments. The added presence of carbon could also increase the dissimilatory reduction of nitrate to ammonium as described earlier in the literature review. If that were to occur, a measure of nitrate disappearance rate under these conditions would not simply be a measure of denitrification and may result in an over estimation of denitrification rate in the soils. However, even when dissimilatory reduction of NH_4^+ is believed to be a factor, it accounts for only minor amounts of nitrate loss (Abbasi and Adams, 1998). As well, while nitrate is added under field conditions, carbon is not usually added, unless a zero-till situation is being considered. Therefore, the situation in the laboratory was constructed to more closely resemble conventional till field situations. Sampling began after at least six hours to allow the system to equilibrate following the flush caused by rewetting the air-dried soil.

4.4.1 Nitrate Disappearance Rate for the Stockton Soil

It was determined that the nitrate disappearance rate (NDR) of the Stockton soil decreased exponentially with depth for all temperatures (Figure 4.37). Generally, correlation was very good for the upper 45 cm of the profile (r is generally >0.9), but not as consistent for the 75-120 cm depth, where correlation coefficients (r) ranged from

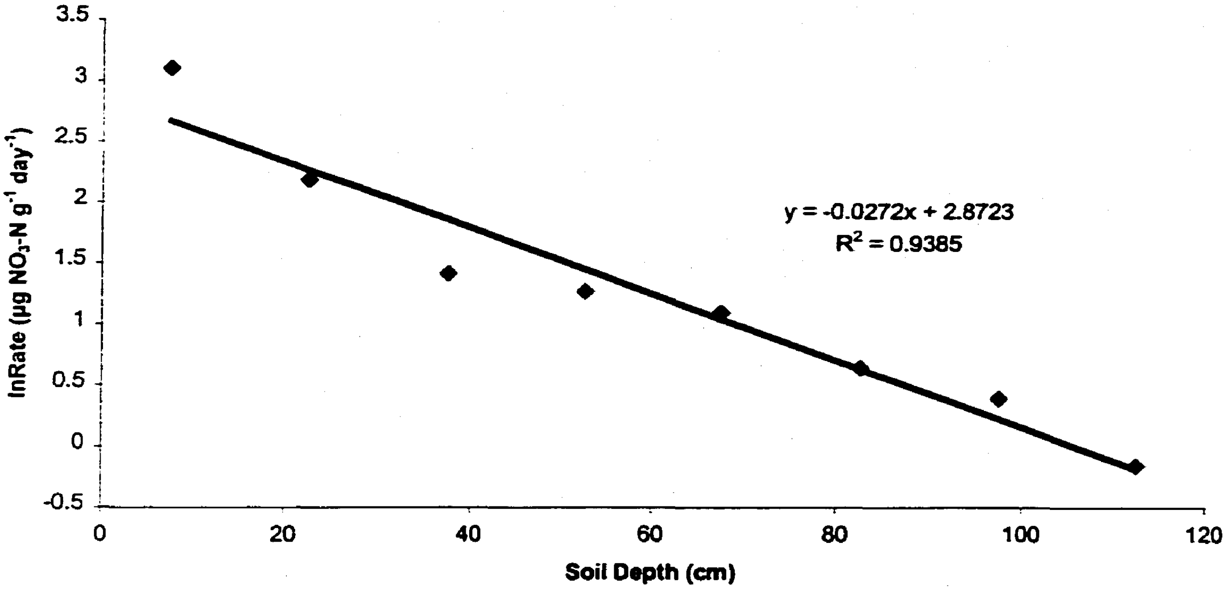

Figure 4.37 Semi-log plot of nitrate disappearance rate for Stockton loamy fine sand profile at 20.5°C.

0.117-0.981 (Table 4.11). Since the rate of nitrate disappearance is much slower at the deeper depths (particularly for 105-120 cm), and several different samples were used for determination, small differences between the individual samples would have a large effect on correlation.

Other researchers have studied the changes in nitrate disappearance rate with depth in soil at a single temperature (20°C) and determined that the amount and rate of nitrate reduction decreased significantly with depth (Abbasi and Adams, 1998). They performed regression analysis between the nitrate disappearance rate (NDR) and organic matter content as a function of soil depth down to 7.5 cm and determined that there was a positive correlation (r^2=0.88) between NO_3^- disappearance and organic matter content.

Table 4.11 Rates of nitrate disappearance on Stockton soil for all depths at various temperatures.

Temperature (°C)	**Soil Depth (cm)**	**Rate of NO_3-N Disappearance ($\mu g\ g^{-1}\ day^{-1}$)**	**Standard Error**	**Correlation Coefficient (r)**	**Level of Significance**
5	0-15	4.56	0.50	0.966	$p < 0.01$
	15-30	2.22	0.31	0.971	$p < 0.01$
	30-45	1.26	0.20	0.960	$p < 0.01$
	45-60	1.15	0.27	0.840	$p < 0.05$
	60-75	1.16	0.21	0.963	$p < 0.01$
	75-90	0.90	0.16	0.913	$p < 0.05$
	90-105	0.83	0.12	0.938	$p < 0.05$
	105-120	0.59	0.12	0.826	$p < 0.10$
11	0-15	10.38	0.55	0.993	$p < 0.01$
	15-30	5.42	0.28	0.992	$p < 0.01$
	30-45	3.62	0.38	0.969	$p < 0.01$
	45-60	3.04	0.25	0.980	$p < 0.01$
	60-75	1.33	0.45	0.770	$p < 0.05$
	75-90	1.42	0.29	0.895	$p < 0.01$
	90-105	1.25	0.32	0.849	$p < 0.01$
	105-120	0.78	0.28	0.780	$p < 0.05$
16.5	0-15	20.89	1.74	0.986	$p < 0.01$
	15-30	7.66	0.64	0.983	$p < 0.01$
	30-45	5.46	0.52	0.982	$p < 0.01$
	45-60	3.50	0.45	0.954	$p < 0.01$
	60-75	2.71	0.58	0.886	$p < 0.01$
	75-90	1.63	0.27	0.925	$p < 0.01$
	90-105	1.58	0.22	0.945	$p < 0.01$
	105-120	1.53	0.14	0.975	$p < 0.01$
20.5	0-15	22.27	4.79	0.901	$p < 0.01$
	15-30	8.90	1.05	0.973	$p < 0.01$
	30-45	4.09	0.59	0.951	$p < 0.01$
	45-60	3.54	0.46	0.952	$p < 0.01$
	60-75	2.98	0.26	0.978	$p < 0.01$
	75-90	1.90	0.15	0.981	$p < 0.01$
	90-105	1.48	0.25	0.927	$p < 0.01$
	105-120	0.86	0.39	0.668	$p < 0.10$
26	0-15	54.60	8.48	0.954	$p < 0.01$
	15-30	16.47	1.17	0.988	$p < 0.01$
	30-45	7.62	0.80	0.974	$p < 0.01$
	45-60	6.30	0.90	0.952	$p < 0.01$
	60-75	3.57	0.36	0.976	$p < 0.01$
	75-90	0.61	0.39	0.577	NS
	90-105	0.20	0.75	0.117	NS
	105-120	0.15	0.50	0.136	NS

Therefore, they attributed the reduced rates of denitrification at lower depths to lower organic matter contents. Luo et al., (1997) determined that denitrification activities in two soils were highest in the top 0-5 cm, and decreased exponentially with depth to 40 cm, independent of sampling sites and times. However, they attributed the reduced denitrification rates at lower depths to lower denitrifying populations deeper in the soil, rather than simply a limitation of available carbon.

Rates of denitrification in subsoil cores removed from 1-2 m and incubated under anaerobic conditions with no carbon amendments have exhibited denitrification capacities of 0.2 - 0.5 mg N kg^{-1} d^{-1} at a temperature of 12°C (Castle et al., 1998). Values determined in this study for the 1.05 - 1.2 m depth for the Stockton soil at 11°C were comparable at 0.78-1.25 mg N kg^{-1} d^{-1}. Castle et al. (1998) believed that this level of denitrification potential in the subsurface soils was sufficient to reduce leaching of nitrate to the groundwater and subsequent nitrate contamination of groundwater on these soils. The relatively low levels of nitrates in the water table below the Stockton soil profile throughout the growing season may be a reflection of this.

Nitrate disappearance rate increased exponentially as temperature increased (Figure 4.38). Rates at the 0-15 cm depth changed most dramatically with temperature, and rates at the lower depth changed much less. For example, the NDR of the 0-15 cm depth at 5°C was only 4.56 μg g^{-1} day^{-1}, but was measured as being 54.60 μg g^{-1} day^{-1} at 26°C, a twelve-fold increase, while the rates for the 45-60 cm depth increased by a factor of 5.5 from 1.15 to 6.30 μg g^{-1} day^{-1} at the same temperatures. This is likely a reflection

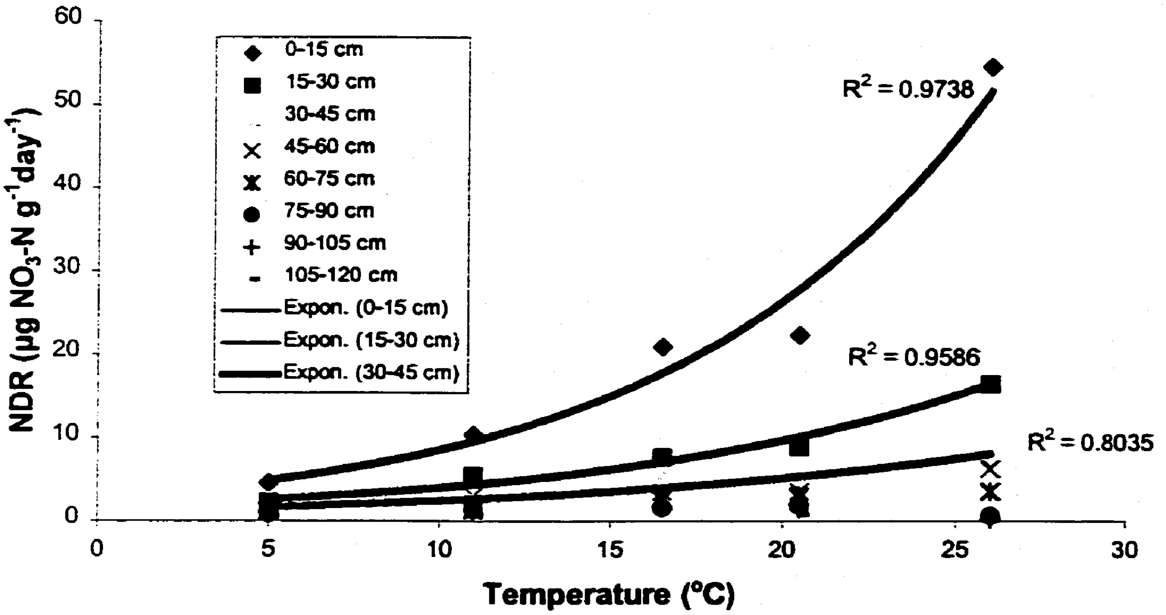

Figure 4.38 Effect of temperature on nitrate disappearance rate at different depths of Stockton loamy fine sand profile.

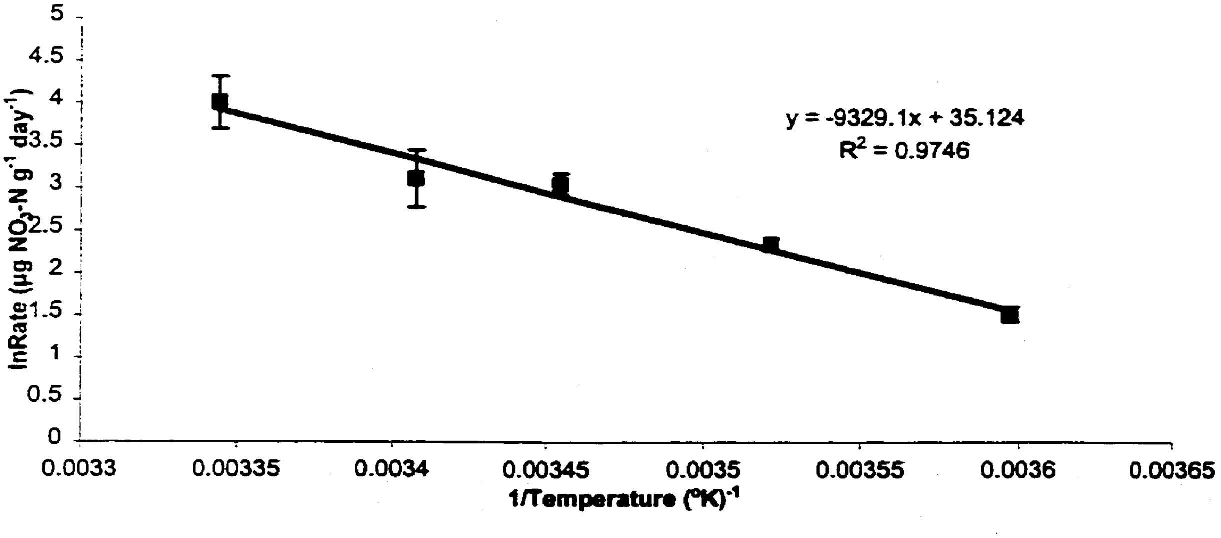

Figure 4.39 Arrhenius plot of Stockton loamy fine sand at 0-15 cm depth: Effect of temperature on rate of nitrate disappearance.

of the size of the microbial population as distributed within the soil profile.

Arrhenius plots of the rate of denitrification of all soil sample depths were calculated using all temperatures under investigation in order to describe the relationship between denitrification and temperature in the soil profile. Results for the 0-15 cm depth are presented in Figure 4.39, above. It was found that a good correlation could be obtained for the top 75 cm of the soil profile, but correlation for the remaining 75-120 cm depths was very poor (Table 4.12). When the rates obtained at 26°C for the 75-120 cm depths were dropped, correlation (r) improved significantly for the 75-90 cm and 90-105 cm depths (from 0.1292 to 0.9738 and from 0.4608 to 0.9230, respectively) but less improvement could be seen for the 105-120 cm depth. It is likely that 26°C is higher than the optimum temperature range for denitrification at these soil depths in the Stockton soil. In fact, soil temperatures for the 60 cm depth only climbed above 20°C once (August 19, 1991) in the two years that they were monitored at this site. Mean temperatures at the 120 cm depth never even reached 20°C during the field study, and the temperature of 20.5 °C used in the denitrification study may also be outside of the bounds of optimum temperature range for denitrification for this soil at the 105-120 cm depth. It may, therefore, be reasonable to assume that the microbial population of this soil profile does indeed change with depth, and the population within the subsoil is less heat tolerant.

According to the correlation coefficients for the Arrhenius equations obtained for the soil profile, the model can be considered a good representation of the effects of temperature on denitrification within the Stockton soil, with the exception of temperatures greater than 20°C at depths below 90-105 cm. Activation energy (Ea) as determined using

Table 4.12 Activation energy and Q_{10} values for Stockton soil.

Soil Depth	**Correlation Coefficient for Arrhenius Graph**[b]	**Activation Energy (Ea)**	**Q_{10} Values**
cm	**r**	**Joules**	
0-15	0.9872	77, 562.1	2.97
15-30	0.9812	61, 204.3	2.36
30-45	0.9024	51, 828.6	2.07
45-60	0.9359	48, 809.8	1.98
60-75	0.9586	41, 142.7	1.78
75-90	0.1292[c]	-	-
75-90[a]	0.9738	31, 419.4	1.55
90-105	0.4608[c]	-	-
90-105[a]	0.9230[d]	28, 862.5	1.46
105-120	0.4365[c]	-	-
105-120[a]	0.6459[c]	25, 968.8	1.44

[a] Data following was based on four temperatures only: 5°C, 11°C, 16.5°C and 20.5°C.
[b] Unless otherwise indicated, all coefficients for correlation (r) are significant at $p<0.05$.
[c] There is no significant linear relationship between temperature and depth at $p<0.1$.
[d] The coefficient for correlation (r) is significant at $p<0.1$.
- Not determined.

the Arrhenius equation is an indicator of the response of the denitrifying population to temperature. That is to say, the higher the activation energy, the greater the impact of temperature on the reaction. Activation energies were determined for each depth of the Stockton profile and were found to decrease with depth (Table 4.12). This is likely a reflection in the amount of readily available carbon and the size (and possibly composition) of the microbial population, both of which would be expected to decrease with depth. Values obtained for Ea are similar to those reported in literature of anywhere from 59 - 74 kJ mol^{-1} (Pelletier et al., 1999).

Q_{10} values, which signify the magnitude that the rate of denitrification will increase for every 10°C increase in temperature, were also calculated for all soil depths using the Arrhenius equations obtained from the graphs. These values ranged from 1.44 to 2.97, which is close to the Q_{10} values offered in the literature of 1.74, (Peterjohn, 1991) and 1.8 (Mangaraja and Misra, 1978). However, higher values (4-6) have also been given in the literature (Gilliam and Gambrell, 1978), and this may be somewhat dependent on the source of carbon used for denitrification (Pelletier et al., 1999). Most interestingly, the Q_{10} values decreased with depth, and were nearly half the rate exhibited by the surface 0-15 cm (2.97) by the 105-120 cm soil depth (1.44).

Therefore, the assumption that Q_{10} values have any universality is likely an inaccurate one, unless the depth effects exhibited here are simply another expression of carbon effects on the Q_{10} values as mentioned by Pelletier et al. (1999). If that were the case, then the change in Q_{10} values with depth would simply be a result of decreasing amounts of readily available organic carbon present in the soil with depth. However, Stanford et al. (1975) and Mangaraja and Misra (1978) found that Q_{10} values did not change at all or very little (0.1 difference), respectively, when soil samples were amended with carbon. It is therefore plausible that the change in Q_{10} values observed in this study is a result of the intrinsic characteristics of the microbial population as it changes with soil depth. This would also account for the drastic increase in the rate of denitrification with increases in temperature noted for the 0-15 cm depth, where the microbial population would be the largest.

It is important to note that these activation energy values (and for that matter, the Q_{10} values) are likely valid only for the range of temperatures observed in this study. It is apparent that temperatures greater than 26°C will not be well accounted for in this model for certain soil depths, and previous research (Pelletier et al., 1999) has indicated that the Arrhenius model is not ideal for describing the effects of temperature on denitrification at low temperatures (below 4°C) in cold climate soils. However, the temperatures under study encompass much of the temperature variation within a field season and therefore use of the Arrhenius model for describing the effect of temperature on denitrification in these soil profiles during the growing season is valid.

4.4.2 Nitrate Disappearance Rate of the Almasippi Soil.

On examination of the the Almasippi profile, it was discovered that a rate of biological activity for 0-15 cm depth existed separately from the rate of biological activity of the 15-120 cm depth. This is likely due to the rapid decrease in soil organic matter

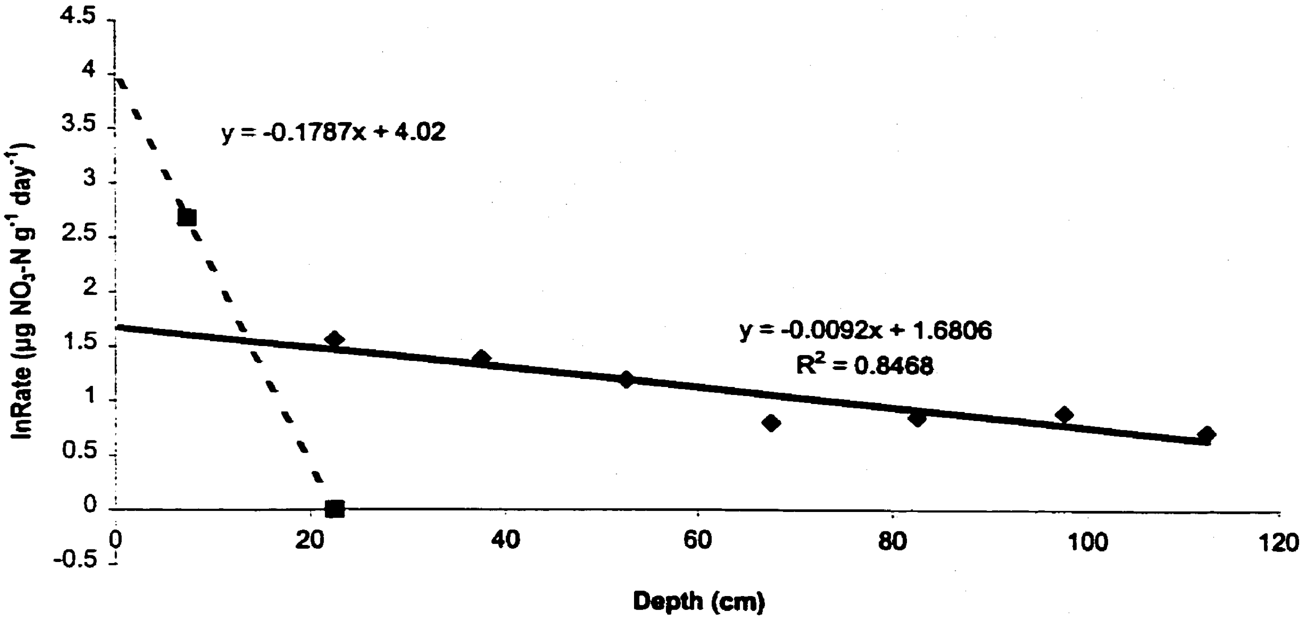

Figure 4.40 Two rates of nitrate disappearance of the Almasippi sand as affected by soil depth at 26°C.

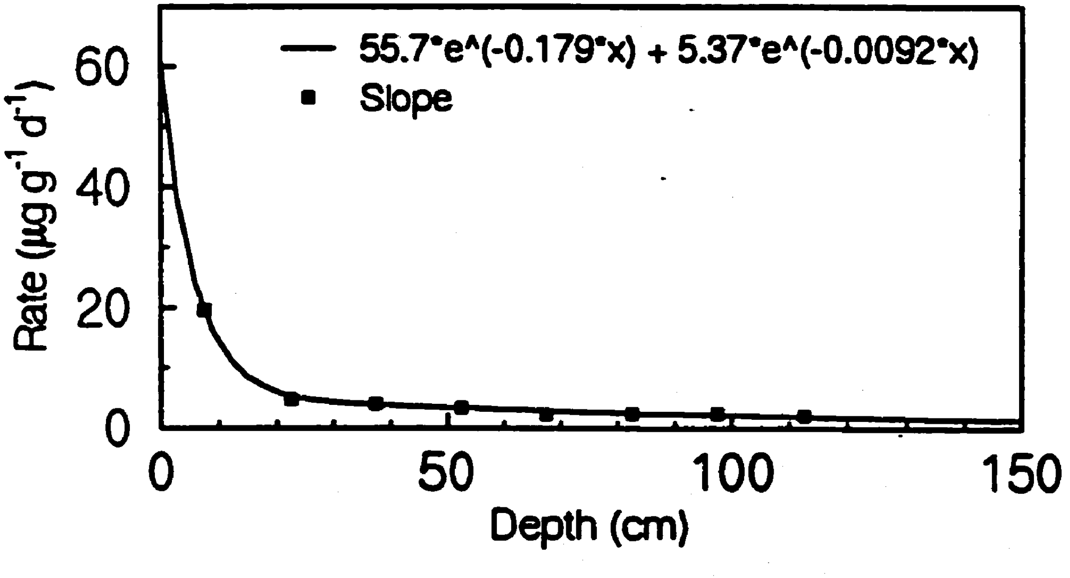

Figure 4.41 Two separate rate expressions for the Almasippi sand are combined into a single equation in order to express NDR for the entire profile at 26°C.

below 15 cm depth. Both rates were determined graphically and showed an exponential decrease in biological activity with depth (Figure 4.40).

The equation for the line representing 0-15 to 15-30 cm depths was determined by first calculating the equation for 15-120 cm when plotted in a semi-log linear manner. The value for y was then extrapolated from this equation at x = 7.5 cm (average of 0-15 cm depth). From y, which is the natural logarithm of the rate, the extrapolated rate could be obtained. This extrapolated rate was subtracted from the actual nitrate disappearance rate, and the natural logarithm of the difference was used to determine the slope of the line connecting 0-15 cm to 15-30 cm depths. The equation obtained for this line represents a rate of activity in the 0-15 cm depth beyond the overall rate of reaction for the entire profile as epitomized by the 15-120 cm depths. The equations for both rates were combined in a single expression to illustrate the overall rate of biological activity for the profile and a close fit was obtained (Figure 4.41).

As in the Stockton profile, nitrate disappearance rate increased exponentially as temperature increased (Figure 4.42). Again, rates at the 0-15 cm depth changed most dramatically with temperature, and rates at the lower depth changed much less. For example, the NDR of the 0-15 cm depth at 5°C was only 0.928 $\mu g\ g^{-1}\ day^{-1}$, but was measured as being 19.60 $\mu g\ g^{-1}\ day^{-1}$ at 26°C, a rate twenty-one times greater than that determined for the lower temperature. This is a larger change in rate than observed for the Stockton soil at the same temperatures, although the actual NDR of the Stockton soil at 26°C was much higher (54.60 $\mu g\ g^{-1}\ day^{-1}$). Correlation for the Almasippi soil was best for the 0-15 cm depth ($r>0.92$) for all temperatures, and was good for all other soil depths, with a level of significance of $p<0.10$ for all profile depths (Table 4.13).

Arrhenius graphs were plotted over the temperature range of the study and correlation was relatively good for all depths above 105 cm (Table 4.14). The 105-120 cm depth was not significantly correlated at $p=0.10$, possibly due to non-uniformity of the samples. There was a clay tongue that occasionally reached above the 120 cm depth at the St. Claude site, and consequently composed a portion of some of the soil samples taken at the 105-120 cm depth. When soil samples were bulked for this experiment, they were mixed and sieved, but were not ground. Therefore, some non-uniformity of samples may have persisted and could account for the varying rates of denitrification and poor correlation at this depth.

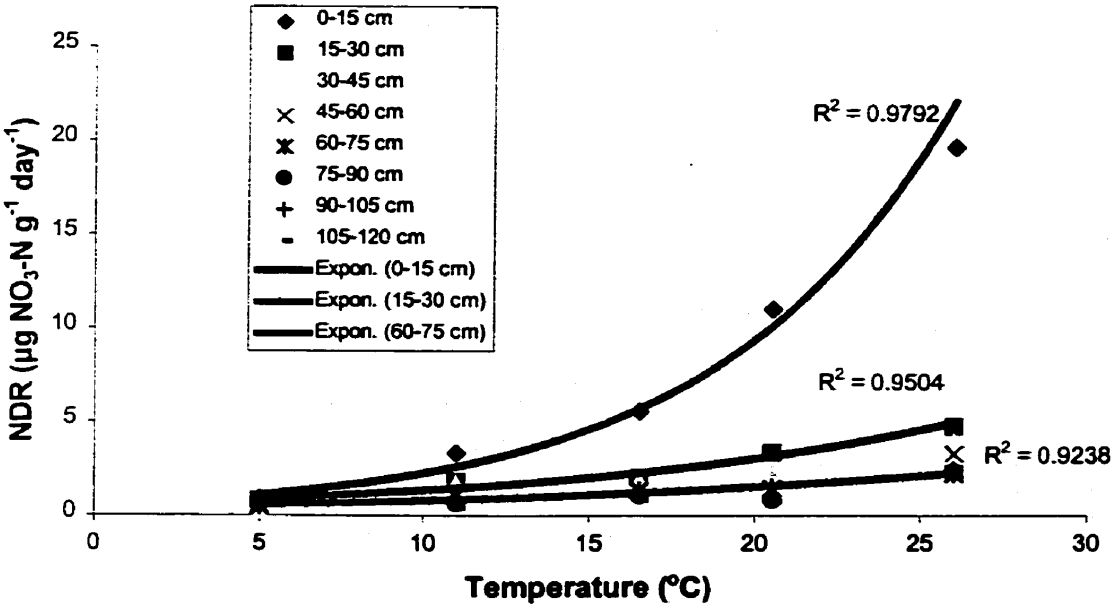

Figure 4.42 Effect of temperature on nitrate disappearance rates (NDR) at different profile depths of the Almasippi sand.

Table 4.13 Rates of nitrate disappearance on Almasippi soil for all depths at various temperatures.

Temperature (°C)	**Soil Depth (cm)**	**Rate of NO$_3$-N Disappearance (μg g^{-1} day^{-1})**	**Standard Error**	**Correlation (r)**	**Level of Significance**
5	0-15	0.93	0.11	0.920	$p<0.01$
	15-30	0.79	0.12	0.891	$p<0.05$
	30-45	0.26	0.11	0.817	$p<0.05$
	45-60	0.61	0.16	0.928	$p<0.05$
	60-75	0.50	0.10	0.920	$p<0.01$
	75-90	0.63	0.09	0.938	$p<0.10$
	90-105	0.55	0.12	0.959	$p<0.01$
	105-120	0.88	0.15	0.927	$p<0.10$
11	0-15	3.33	0.29	0.978	$p<0.01$
	15-30	1.86	0.36	0.905	$p<0.01$
	30-45	1.80	0.28	0.943	$p<0.01$
	45-60	1.31	0.18	0.953	$p<0.01$
	60-75	1.07	0.13	0.957	$p<0.01$
	75-90	0.65	0.10	0.936	$p<0.01$
	90-105	0.75	0.22	0.814	$p<0.05$
	105-120	0.43	0.17	0.709	$p<0.05$
16.5	0-15	5.56	0.82	0.959	$p<0.01$
	15-30	2.04	0.69	0.770	$p<0.05$
	30-45	1.96	0.36	0.911	$p<0.01$
	45-60	1.35	0.32	0.848	$p<0.01$
	60-75	1.33	0.25	0.909	$p<0.01$
	75-90	1.06	0.29	0.833	$p<0.05$
	90-105	1.28	0.10	0.979	$p<0.01$
	105-120	0.84	0.16	0.889	$p<0.01$
20.5	0-15	11.00	1.13	0.980	$p<0.01$
	15-30	3.37	0.34	0.975	$p<0.01$
	30-45	2.25	0.26	0.961	$p<0.01$
	45-60	1.29	0.23	0.928	$p<0.01$
	60-75	1.42	0.21	0.939	$p<0.01$
	75-90	0.84	0.30	0.748	$p<0.05$
	90-105	1.51	0.38	0.854	$p<0.01$
	105-120	0.75	0.21	0.820	$p<0.05$
26	0-15	19.60	3.78	0.933	$p<0.01$
	15-30	4.77	0.66	0.955	$p<0.01$
	30-45	4.01	0.50	0.963	$p<0.01$
	45-60	3.30	0.41	0.970	$p<0.01$
	60-75	2.24	0.23	0.974	$p<0.01$
	75-90	2.35	0.34	0.952	$p<0.01$
	90-105	2.44	0.39	0.943	$p<0.01$
	105-120	2.05	0.51	0.897	$p<0.05$

Unlike the Stockton soil, 26°C did not appear to be outside the range of optimum temperatures for denitrification at the lower depths for the Almasippi soil. This is despite the fact that the soil temperature at depths below 60 cm was less than 20°C on all sampling dates over the two years of the field study. It may be that the microbial populations of this soil were less depth specific than that of the Stockton soil. Field data seems to indicate that extensive leaching and water table fluctuations occurred with the Almassipi field site, which likely resulted in a mixing of populations from one depth to the next. As the water moved up and down the soil profile, so did the soil microorganisms. This would not be true for the Stockton soil, as that site appeared to have less leaching, and a lower water table. It should be noted that previous research by Luo et al.. (1997) concluded that heavy rainfall did not wash microorganisms down the soil profile of a well drained sandy loam.

Activation energies did decrease as the depth of soil dropped below 15 cm. However, the pattern here is also very different from that of the Stockton soil. In this case, as with the two different rates of biological activity for the Almasippi soil, the 0-15 cm depth has both activation energy and Q_{10} values distinct from the remaining 15-120 cm of the soil profile. The activation energy for the 0-15 cm depth is 98, 380J whereas the remainder of the profile has an average activation energy level of 48, 827J. The Q_{10} values for the entire soil profile are within the expected range outlined in the literature, but here, again, the value at 0-15 cm is 3.97 while the average value for the remainder of the profile is 2.02. (The 30-45 cm depth is somewhat of an anomaly in that it has a particularly high Ea of 78, 442 J and a Q_{10} of 2.99). This further supports the use of two separate equations to describe Almasippi soils.

Table 4.14 Activation energy and Q_{10} values for Almasippi soil.

Soil Depth	**Correlation Coefficient for Arrhenius Graph**[a]	**Activation Energy (Ea)**	**Q_{10} Values**
cm	**r**	**Joules**	
0-15	0.9917	98, 380	3.97
15-30	0.9767	56, 415	2.20
30-45	0.8988	78, 443	2.99
45-60	0.9001	45, 473	1.90
60-75	0.9645	45, 021	1.88
75-90	0.8450[b]	38, 534	1.70
90-105	0.9932	49, 457	2.00
105-120	0.5995[c]	28, 456	1.50

[a] Coefficients of correlation (r) are significant at p=0.05 unless otherwise indicated.
[b] Coefficient of correlation (r) is significant at p=0.10.
[c] There is no significant linear relationship between NDR and soil temperature at p=1.0.

5. GENERAL DISCUSSION

A seasonal pattern of nitrate distribution emerged for the two years of the study on these sandy and clay loam soils. In early May of the first season, nitrate levels were initially low, indicating that nitrates were not accumulating to any significant extent within these profiles prior to the period of study. By June, nitrate was quite mobile within the soil profile and leached in response to rainfall, and moved out of the profile via uptake of nutrients by plants. Increased crop uptake of nitrate in July appeared to be accompanied by a net removal of nitrate via denitrification under the seeded treatments, while there was a net increase of nitrate via mineralization and nitrification under the uncropped treatments. Although all three of the processes would likely be occurring under all treatments to some extent, increased carbon exudates from roots under the seeded treatments likely resulted in increased denitrification in anaerobic microsites (Bailey, 1976) as compared to the uncropped treatments. Under the uncropped treatments, lack of carbon inputs may have resulted in some mineralization of organic nitrogen within the soil. This increase in ammonium may in turn have resulted in increased nitrification, and a net gain in nitrate content in the soil profile over that in the seeded treatments.

In August, some nitrate appeared to move up the profile in response to surface evaporation, and this upward movement, plus nitrification in this month and into September resulted in very high nitrate levels in the top 120 cm of the unseeded soil profiles. This pattern was repeated for the second year of the study, and is similar to findings of Watts et al.. (1991), Roth and Fox (1990) and Magdoff (1992), who noted that soil nitrate concentrations in fall were much higher than spring concentrations in the same area. Since nitrate levels were so low in these soils in early May, it is likely that significant denitrification occurs in these soils in early spring when the soil is very wet and surface temperatures during the day may be quite high.

As previously indicated, there were significant differences in the amount of nitrate in the surface 0-105 cm of soil between the cropped and uncropped treatments. As the amount of nitrates in the uncropped profiles appeared to increase over the growing season, these cropping effects were not only a result of nitrogen uptake by the crop, but also of the large amount of mineralization and nitrification occurring on these soils under fallow conditions. Other researchers have reported net nitrification rates of 2.7 kg N ha^{-1} d^{-1} for soils receiving 100 kg N ha^{-1} from NH_4NO_3 fertilizers on an annual basis (Watson and Mills, 1998), indicating that nitrification was quite significant within these soils.

It should be noted that nitrate to chloride ratios support the theory that net nitrification was certainly occurring under the fallow plots. Ratios of nitrate nitrogen to chloride within the fallow subplots were much higher than the nitrogen to chloride application of 1.1 for the latter part of the growing season for plots 1 and 3. Therefore, even though the patterns (and rates) of biological activity (as expressed by denitrification in each soil profile) were different for both soils, there was a marked contribution of nitrate to the soil profile for both soils, regardless of their actual activity rates. The resulting ratios were much higher than 1.1 at some periods during the growing season. Since the application ratio of inorganic N to Cl^- was 1.1 (the ratio of NO_3-N to Cl^- applied was approximately 0.55), even if all the nitrogen applied as NH_4^+ nitrified, the ratio should still not be much higher than 1. However, nearer the end of the growing season, actual ratios were much greater than one, indicating that mineralization in this soil was also likely occurring. Watson and Mills (1998) determined that net mineralization of existing pools of organic N was not significant during the period of their study, but gross mineralization was occurring at a significant rate.

Under the fallow conditions imposed in this study, the limit of available carbon in the soil would support the mineralization of soil organic N, as there would be little additional available C for growth of the microorganisms under the fallow conditions. Generally, the soil nitrate levels in the top 1.2 m of the fallow subplots were often higher than the 168 kg NO_3-N ha^{-1} level provided by the Department of Agriculture and Food, Province of Manitoba as the level above which the soil is considered to be nitrate contaminated. These levels were surpassed at various times throughout the growing season regardless of whether or not any additional fertilizer was applied.

The seeded treatments had very different nitrate distribution patterns from the fallow treatments for all soils studied. Although the soil would have likely been well oxygenated in July, considerable denitrification appeared to be occurring on the seeded treatments, presumably in anaerobic microsites within the soil profile. This is indicated by the fact that the seeded treatments were both depleted of nitrogen for plots 1 and 2, regardless of whether fertilizer was applied. At the very least, it is likely that some denitrification was occurring on the FW treatment, as sufficient nitrogen for the growing season was applied, and still the nitrate was completely depleted from the soil profile by the end of the second growing season. This would be expected based on the study by Smith and Tiedje (1979), who found the highest rates of denitrification in rhizosphere soil. The timing of the denitrification period (July) reflects the seasonal effects discovered by Wheatley et al. (1991) whereby soil denitrification rates increased with root emergence. On the other hand, Groffman and Tiedje (1991) and Parsons et al. (1991) determined that while denitrification activity increased in spring, it was lower in summer. This may be an issue with soil moisture, which must be significant (as both a contributor to limited aeration and in and of itself) in order for denitrification to occur - a situation that is not necessarily present during the summer months.

It is worth noting that the above mentioned patterns of varying total nitrate nitrogen contents under the different treatments was not true for plot 3, certainly during the 1991 growing season. In this case, there appeared to be very little difference in the total amounts of nitrate in the soil profile under the different treatments. Given the high perched water table at plot 3, and the low rate of denitrification expressed by the Almasippi soil from this plot, this is not surprising. Levels of nitrate at this site were directly and rapidly affected by groundwater fluctuations. In 1991, with the groundwater reaching up to 45 cm from the soil surface, the soil surface was bog-like and our feet would sink in up to our ankles. At this point, with the level of mixing in the groundwater, the plot could not really be considered to have unfertilized and fertilized treatments.

The amounts of nitrates in the soil profiles under all four treatments for this soil in 1991 were very high across the board, likely because in this particular soil system, with its perched water table and low denitrification rates, the nitrates simply had no place to go. There was nowhere to be leached to, as the water table was so high that downward movement was effectively stopped. Although the soil temperatures were still quite warm at some of the sampling times, and likely the surface was moist enough to successfully support denitrification, very little reduction of nitrate occurred in this soil profile.

This is similar to results obtained by Klemedtsson et al. (1988) on a loam soil. They found only a slow increase in nitrous oxide production after a dry loam soil was rewetted, and surmised that this indicated a low overall biological activity of the soil. The researchers thought it would, therefore, be unlikely that the loam soil would have anaerobic microsites caused by high oxygen consumption at moisture levels below saturation. Christensen et al. (1990a) observed denitrification on a coarse textured soil with little aggregation and found only short periods of increased activity following irrigation. This led them to suspect that little denitrification occurred in that soil at field capacity. Results of the laboratory studies of denitrification in the Almassippi soil confirm that it is highly unlikely that denitrification would be responsible for a large amount of nitrate loss at the St. Claude site as rates are low even under saturated conditions.

Nitrate levels in the groundwater were generally low below the Stockton soil at plot 1, and were only higher than the maximum level of 10 $\mu g\ NO_3\text{-}N\ mL^{-1}$ on two occasions in 1991. Water from the well on the fertilized subplot on plot 1 registered 11.0

μg NO_3-N mL^{-1} on July 11, and water from the irrigation well had nitrate levels of 10.6 μg NO_3-N mL^{-1} on August 19, 1991. In 1992, the irrigation well had water nitrate levels of 10.2 μg NO_3-N mL^{-1} on both August 18 and September 17, while water in the well of the fertilized subplot had nitrate levels of 12.2 and 12.8 μg NO_3-N mL^{-1} on August 6 and 18, respectively. Nitrate levels of groundwater directly below the unfertilized subplot were never greater than 6 μg NO_3-N mL^{-1} in 1992. It is apparent from the data obtained that nitrate leaching at this site was quite limited, as the groundwater under the area of study where no nitrate was applied did not seem to be affected. In fact, the well that has been described as being for other use had consistently low to zero levels of nitrate present in the water in both years of the study.

Nitrate concentrations in the groundwater beneath the site at St. Claude were consistently very high (often greater than 30 μg NO_3-N mL^{-1}) and always greater than the maximum acceptable limit of 10 μg NO_3-N mL^{-1}. This was true from the first well sampled on May 31, 1991 to the last well sampled on September 22, 1992. Even the tile drain, which was located 50 m from the plot, occasionally had levels of nitrate greater than 10 μg NO_3-N mL^{-1} when it was flowing. Nitrate levels in the groundwater did not differ greatly between wells or years, but levels were highest in the Center and West wells in 1991 and in the Center well in 1992. As the Center well was located in the middle of the fertilized subplot, this is not unexpected. Seasonally, groundwater nitrate levels were highest in the West well from late June to early July in 1991, and for the Center well during the same period in 1992. No seasonal differences in nitrate contents of the groundwater were exhibited by any of the other wells in either of the years of the study.

Tile drain flow patterns differed from 1991 to 1992. It is unlikely based on the high levels of nitrate present in the groundwater that a significant level of denitrification was occurring in the aquifer.

The Arrhenius equations determined for both the Almasippi and Stockton profiles at each soil depth (Appendix XI) were used to calculate the maximum denitrification potential for each soil profile based on actual temperatures measured in the field (Appendices I, III, IV and VI). These results are presented in Tables 5.1, 5.2, 5.3 and 5.4. The potential denitrification rates were determined for the entire soil profile (to 120 cm) for both soils in both years of the study. Calculations were made based on the 0-120 cm rates, and then again on the 15-120 cm rates based both on the amount of NO_3-N denitrified per kg of soil and using the bulk densities of the soils as determined in the field (Table 3.1) on a per ha of land basis. The rates as determined are extremely high, particularly when the top 0-15 cm of soil is considered in the calculation, but seem to agree with rates maximum rates determined by others (Mahli et al., 1990).

The assumptions made in making these calculations include the following conditions: saturated soil, anaerobic conditions, unlimited nitrate and carbon supply, and no constraints on NO_3-N and N_2O diffusion. The existence of all of these conditions at the soil surface under the high temperatures measured is extremely unlikely. Therefore, the maximum attenuation was also calculated for only the 15-120 cm depth, to give a more reasonable estimate of potential denitrification rates. Again, all conditions as previously mentioned are assumed to exist. In reality, much of the denitrification that occurs within the soil profile will do so in anaerobic microsites and/or hot spots (Parkin,

Table 5.1. Maximum nitrate attenuation potential* of Almasippi profile in 1991.

Sample Date	Maximum denitrification potential*			
	0-120 cm	15-120 cm	0-120 cm	15-120 cm
	mg NO_3-N kg^{-1} d^{-1}		kg NO_3-N ha^{-1} d^{-1**}	
May 28, 1991	34.62	7.80	64.67	18.00
June 27, 1991	31.51	10.49	60.80	24.22
July 4, 1991	17.60	9.51	36.04	21.98
July 10, 1991	32.25	10.45	62.06	24.13
July 23, 1991	41.79	11.60	79.32	26.81
July 30, 1991	46.65	12.03	88.02	27.78
Aug 7, 1991	23.76	10.15	47.14	23.47
Aug 15, 1991	29.37	14.77	59.49	34.10
Aug 22, 1991	38.32	13.71	74.47	31.66
Aug 28, 1991	45.74	13.65	87.34	31.52
Sept 5, 1991	22.99	10.97	46.29	25.37
Sept 13, 1991	17.93	9.98	36.91	23.09
Sept 19, 1991	10.94	7.22	23.22	16.76
Oct 3, 1991	9.83	6.46	20.84	14.98
Seasonal Mean	28.81	10.63	56.19	24.56

* The maximum denitrification potential for the soil profile was calculated using actual soil temperatures measured for each soil depth, and Arrhenius equations determined for each soil depth over the temperature range of 5-26°C. This assumes that the soil is completely saturated and anaerobic and that the supply of both nitrate and carbon are not limiting.

**Calculations were made using bulk densities as determined in the field. Bulk densities for depths 90-105 and 105-120 cm were approximated from the bulk density measured for 75-90 cm.

Table 5.2. Maximum nitrate attenuation potential* of Almasippi profile in 1992.

Sample Date	Maximum denitrification potential*			
	0-120 cm	**15-120 cm**	**0-120 cm**	**15-120 cm**
	mg NO_3-N kg^{-1} d^{-1}		**kg NO_3-N ha^{-1} d^{-1}****	
May 6, 1992	26.94	6.67	50.66	15.39
May 13, 1992	10.54	5.55	21.52	12.84
May 20, 1992	46.82	7.89	85.94	18.19
May 25, 1992	13.02	5.72	25.94	13.24
June 2, 1992	31.54	9.37	60.19	21.62
June 16, 1992	14.95	10.32	31.87	23.82
June 22, 1992	31.34	8.20	59.22	18.96
June 29, 1992	22.81	9.66	45.19	22.31
July 7, 1992	34.85	9.07	65.82	20.95
July 14, 1992	41.83	11.17	79.12	25.77
July 21, 1992	23.62	9.45	46.49	21.84
July 28, 1992	18.81	10.28	38.63	23.78
Aug 5, 1992	28.49	10.21	55.40	23.59
Aug 19, 1992	22.32	10.61	44.89	24.51
Sept 2, 1992	15.31	8.41	31.46	19.45
Sept 10, 1992	10.83	7.81	23.34	18.08
Sept 21, 1992	8.30	6.74	18.33	15.61
Seasonal Mean	23.67	8.65	46.12	20.00

* The maximum denitrification potential for the soil profile was calculated using actual soil temperatures measured for each soil depth, and Arrhenius equations determined for each soil depth over the temperature range of 5-26°C. This assumes that the soil is completely saturated and anaerobic and that the supply of both nitrate and carbon are not limiting.

**Calculations were made using bulk densities as determined in the field. Bulk densities for depths 90-105 and 105-120 cm were approximated from the bulk density measured for 75-90 cm.

Table 5.3. Maximum nitrate attenuation potential* of Stockton profile in 1991.

Sample Date	Maximum denitrification potential*			
	0-120 cm	**15-120 cm**	**0-120 cm**	**15-120 cm**
	mg NO_3-N kg^{-1} d^{-1}		**kg NO_3-N ha^{-1} d^{-1}****	
May 16, 1991	78.38	17.92	124.22	29.90
May 23, 1991	96.20	24.22	152.76	40.47
June 11, 1991	85.55	25.66	136.28	42.85
June 18, 1991	65.89	25.70	105.60	42.91
June 27, 1991	68.48	26.54	109.70	44.30
July 23, 1991	98.84	29.45	157.42	49.17
Aug 8, 1991	100.53	28.85	159.99	48.18
Aug 19, 1991	110.79	29.48	176.06	49.22
Sept 11, 1991	47.60	21.98	76.60	36.64
Sept 25, 1991	25.50	14.67	41.28	24.39
Seasonal Mean	77.77	24.45	123.99	40.80

* The maximum denitrification potential for the soil profile was calculated using actual soil temperatures measured for each soil depth, and Arrhenius equations determined for each soil depth over the temperature range of 5-26°C. This assumes that the soil is completely saturated and anaerobic and that the supply of both nitrate and carbon are not limiting.

**Calculations were made using bulk densities as determined in the field. Bulk densities for 105-120 cm were approximated from the bulk density measured for 90-105 cm.

Table 5.4. Maximum nitrate attenuation potential* of Stockton profile in 1992.

Sample Date	Maximum denitrification potential*			
	0-120 cm	15-120 cm	0-120 cm	15-120 cm
	mg NO_3-N kg^{-1} d^{-1}		kg NO_3-N ha^{-1} d^{-1**}	
May 12, 1992	24.27	12.87	39.20	21.42
May 27, 1992	108.12	14.96	170.24	24.91
June 22, 1992	98.84	18.55	156.16	30.91
July 7, 1992	139.23	18.88	219.20	31.46
July 21, 1992	94.09	19.69	148.89	32.82
Aug 6, 1992	70.58	21.97	112.47	36.64
Aug 18, 1992	61.47	19.18	97.93	31.96
Aug 31, 1992	41.68	17.23	66.82	28.67
Seasonal Mean	79.79	17.92	126.36	29.85

* The maximum denitrification potential for the soil profile was calculated using actual soil temperatures measured for each soil depth, and Arrhenius equations determined for each soil depth over the temperature range of 5-26°C. This assumes that the soil is completely saturated and anaerobic and that the supply of both nitrate and carbon are not limiting.

**Calculations were made using bulk densities as determined in the field. Bulk densities for 105-120 cm were approximated from the bulk density measured for 90-105 cm.

1987) and the actual overall rates will therefore be much lower. In addition, carbon and/or nitrate supplies would become depleted quite quickly under these optimum conditions, and the full seasonal attenuation potential would never be reached.

It is interesting to note that the seasonal mean potential denitrification rates were quite similar within each soil profile over the two years of the study. Mean rates were 78 and 80 mg NO_3-N kg^{-1} d^{-1} for the 0-120 cm depths for the Stockton soil in 1991 and 1992, respectively (Tables 5.3 and 5.4) and 29 and 24 mg NO_3-N kg^{-1} d^{-1} for the Almasippi soil in 1991 and 1992, respectively (Tables 5.1 and 5.2). Therefore, it may be reasonable to assume that actual seasonal denitrification levels are similar within a soil type from year to year, provided that soil management remains the same. However, the two soils were distinctly different from one another, and potential denitrification was much higher for the Stockton soil than for the Almasippi soil. This further supports the results of the field studies, where it became apparent that while leaching played a large role in the fate of nitrate in the Almasippi profile, denitrification was a major influence on the fate of nitrate in the Stockton soil.

6. SUMMARY AND CONCLUSIONS

This study examined the fate of surface applied nitrate nitrogen in three medium to coarse textured soils located above aquifers. The two soils located near Carberry, one a Stockton loamy fine sand (plot 1), and the other a Wellwood loam (plot 2), had similar seasonal nitrate distribution patterns throughout the two years of the study. Generally, soil levels of nitrate were initially low in May, then high after fertilizer application. In June, nitrate accumulations occurred lower down in the soil profile, with low nitrate levels at all profile depths under the seeded treatments. In July, nitrate levels were lower under fertilized treatments, particularly on the seeded treatments, and disproportionately lower than the levels of chloride present. During August the nitrate in the soil profile tended to accumulate at or near the surface, and this trend continued into September, with the actual nitrate concentrations at the top of the profile increasing.

Treatment effects could be observed for the Stockton and Wellwood soil profiles, with the presence of a crop significantly reducing the amount of nitrates present in the soil, to the point of depletion, regardless of whether fertilizer was applied. Treatment effects were not as obvious for the Almasippi soil, as its shallow water table ensured that all treatments received nitrates from the ground water, and differences in nitrate contents in the soil profiles under the treatments could not be distinguished. Although there were no significant effects of fertilizer on the amount of nitrogen in the soil profile, levels applied in this study were relatively modest (100 kg N ha^{-1}) and higher application rates would likely have exhibited significant differences, particularly after the second field season. Results from this study clearly indicate that leaving a field fallow could have significant detrimental impacts to ground water quality as the amounts of nitrate that accumulated under the fallow treatments were higher than the excess soil nitrate limit determined by the Province of Manitoba (150 lb N ac^{-1} / upper 4 feet). This was true even for the treatment for which no fertilizer had been applied.

The groundwater at plot 2 was not monitored, but nitrate levels in the groundwater were generally low at plot 1, and were only higher than the maximum level of 10 µg NO_3-N mL^{-1} on two occasions in 1991. In 1992, the irrigation well had water nitrate levels of 10.2 µg NO_3-N mL^{-1} on both August 18 and September 17, while water in the well of the fertilized subplot had nitrate levels of 12.2 and 12.8 µg NO_3-N mL^{-1} on August 6 and 18, respectively. Nitrate levels of groundwater directly below the unfertilized subplot were never greater than 6 µg NO_3-N mL^{-1} in 1992. The well that has been described as being for other use had consistently low to zero levels of nitrate present in the water in both years of the study.

Although the Almasippi sandy soil located at the St. Claude site also had some seasonal distribution patterns, the distribution of nitrate in the soil profile at this site was a direct result of the water table, particularly in 1991. Throughout the1991 field season, both the soil moisture and nitrate distributions fluctuated dramatically, as did the depth to groundwater. At its highest point, the groundwater was located only 60 cm from the soil surface. Like the Stockton and Wellwood sites, the Almasippi profile distribution showed an accumulation of nitrate at or near the soil surface in August and September of 1991.

Nitrate concentrations in the groundwater of the Almasippi soil were consistently very high (often greater than 30 μg NO_3-N mL^{-1}) and always greater than the maximum acceptable limit of 10 μg mL^{-1}. The tile drain, which was located 50 m from the plot, occasionally had levels of nitrate greater than 10 μg mL^{-1} when it was flowing. Nitrate levels in the groundwater did not differ greatly between wells or years. Tile drain flow patterns differed from 1991 to 1992.

Laboratory studies on denitrification indicated that for the Stockton loamy sand profile, the rate of biological activity decreases exponentially with depth. The Almasippi profile exhibits two rates of activity: one for the 0-15 cm depth and another for the 15-120 cm depth. Arrhenius graphs of the Stockton soil indicate that nitrate disappearance rate increased with temperature, except at depths of 75-120 cm, which were more highly correlated if the temperature of 26°C were omitted from calculations. This was not true for the Almasippi soil, where Arrhenius graphs with all temperatures were highly correlated. Activation energies calculated for the Stockton soil decreased with depth, as did Q_{10}values. For the Almasippi soil, the activation energy and Q_{10} values were much greater for the 0-15 cm profile depth than for the remainder of the profile.

In conclusion, nitrate distribution patterns for the sites showed some seasonal effects, although nitrate distribution in the Almasippi soil is strongly influenced by soil water movement. Groundwater contamination was minimal beneath the Stockton soil at plot 1. Rates of nitrate disappearance for this soil were relatively high, and denitrification could be a significant avenue for nitrate loss on the Stockton soil provided that conditions for denitrification are favourable. Groundwater below the Almasippi profile was highly contaminated within the first year of the study. Nitrate disappearance rates calculated for this soil are quite low, even for the 0-15 cm depth under saturated conditions, and it is not expected that there will be much nitrate lost to denitrification at this site. Therefore, the St. Claude site has a very high potential for groundwater contamination.

7. CONTRIBUTION TO KNOWLEDGE

Although there is an abundance of information on denitrification, most studies are either done in situ, or if in the laboratory, consider only the surface soil horizon. It is interesting to note that in this study, Q_{10} values changed with depth. Therefore, the optimum temperature range for denitrification may not be the same for all profile depths within the same soil type. This study confirms that provided a system is shaken to ensure that preferential reduction of nitrate occurs, zero-order denitrification kinetics will prevail and NDR will decrease in a semi log linear manner with depth. Finally, this study provides actual values for potential background denitrification rates for two Manitoba soil profiles. These values can be used to provide an estimate of maximum expected denitrification within these two soil profiles under saturated conditions.

8. REFERENCES

Alexander, M. 1977. Chapter 17: Denitrification. Pages 272-285. *In* John Wiley and Sons, (Eds.). Introduction to Soil Microbiology. 2nd edition. New York.

Abbasi, M. K., and Adams, W. A. 1999. Assessment of the contribution of denitrification to N losses from compacted grassland soil by NO_3^- disappearance and N_2O production during anaerobic incubation. Can. J. Soil Sci. **79**: 57-64.

Bailey, L. D. 1976. Effects of temperature and root on denitrification in a soil. Can. J. Soil Sci. **56**: 79-87.

Barrow, G. M. 1988. Section 18.6: Temperature and rates. Pages 710-712. *In* McGraw-Hill, Inc. (Eds.). Physical Chemistry. 5th edition. New York.

Blackmer, A. M. and Bremner, J. M. 1978. Inhibitory effect of nitrate on reduction on N_2O to N_2 by soil microorganisms. Soil Biol. Biochem. **10**: 187-191.

Burford, J. R. and Bremner, J. M. 1975. Relationships between the denitrification capacities of soils and total, water-soluble and readily decomposable soil organic matter. Soil Biol. Biochem. **7**: 389-394.

Burton, D. L. and Beauchamp, E. G. 1985. Denitrification rate relationships with soil parameters in the field. Commun. in Soil Sci. Plant Anal. **16**: 539-549.

Cady, F. B. and Bartholomew, W. V. 1960. Sequential products of anaerobic denitrification in Norfolk soil material. Soil Sci. Soc. Am. J. **??**: 477-482.

Cambardella, C. A., Moorman, T. B., Jaynes, D. B., Hatfield, J. L., Parkin, T. B., Simpkins, W. W., and Karlen, D. L. 1999. Water quality in Walnut Creek watershed: nitrate-nitrogen in soils, subsurface drainage water, and shallow groundwater. J. Environ. Qual. **28**: 25-34.

Cameron, D. R., Kowalenko, C. G. and Campbell, C. A. 1979. Factors affecting nitrate nitrogen and chloride leaching variability in a field plot. Soil Sci. Soc. Am. J. **43**: 455-460.

Castle, K., Arah, J. R. M., and Vinten, A. J. A. 1998. Denitrification in intact subsoil cores. Biol. Fertil. Soils **28**: 12-18.

CCREM. 1995. Canadian Water Quality Guidelines. March 1987. Canadian Council of Resource and Environment Ministers. Environment Canada, Ottawa.

Chèneby, D., Hartmann, A., Hénault, C., Topp, E. and Germon, J. C. 1998. Diversity of denitrifying microflora and ability to reduce N_2O in two soils. Biol. Fertil. Soils **28**: 19-26.

Cho, C. M., Sakdinan, L. and Chang, C. 1979. Denitrification Intensity and Capacity of Three Irrigated Alberta Soils. Soil Sci. Soc. Am. J. **43**:945-950.

Cho. C. M. and Sakdinan, L. 1978. Mass spectrometric investigation on denitrification. Can. J. Soil Sci. **58**: 443-457.

Christensen, S., Simkins, S. and Tiedje, J. M. 1990a. Spatial variation in denitrification: dependency of activity centers on the soil environment. Soil Sci. Soc. Am. J. **54**: 1608-1613.

Christensen, S., Simkins, S. and Tiedje, J. M. 1990b. Temporal patterns of soil denitrification: their stability and causes. Soil Sci. Am. J. **54**: 1614-1618.

Christianson, C. B. and Cho, C. M. 1983. Chemical denitrification of nitrite in frozen soils. Soil Sci. Soc. Am. J. **47**:38-42.

Cooper, G. S. and Smith, R. L. 1963. Sequence of products formed during denitrification in some diverse Western soils. Soil Sci. Soc. Am. J. ??: 659-662.

Culley, J. L. B. 1993. Chapter 50: Density and compressibility. Pages 529-540. *In* Lewis Publishers, CRC Press, Inc. Carter M. R. (ed.). Soil Sampling and Methods of Analysis. Boca Raton, Florida.

deCatanzaro, J. B. and Beauchamp, E. G. 1985. The effect of some carbon substrates on denitrification rates and carbon utilization in soil. Biol. Fertil. Soils **1**: 183-187.

Doughty, J. L., Cook, F. D., and Warder, F. G. 1954. Effect of cultivation on the organic matter and nitrogen of Brown soils. Can. J. Soil Sci. **34**: 406-411. (Cited in Hedlin, 1995).

Drury, C. F., McKenney, D. J. and Findlay, W. I. 1991. Relationships between denitrification, microbial biomass and indigenous soil properties. Soil Biol. Biochem. **23**: 751-755.

Elliot, P. W., Knight, D. and Anderson, J. M. 1991. Variables controlling denitrification from earthworm casts and soil in permanent pastures. Biol. Fertil. Soils 11: 24-29.

Ellis, S., Howe, M. T., Goulding, K. W. T., Mugglestone, M. A., and Dendooven, L. 1998. Carbon and nitrogen dynamics in a grassland soil with varying pH: effect of pH on the denitrification potential and dynamics of the reduction enzymes. Soil Biol. Biochem. **30**: 359-367.

Errebhi, M., Rosen, C. J., Gupta, S. C. and Birong, D. E. 1998. Potato yield response and nitrate leaching as influenced by nitrogen management. Agron. J. **90**: 10-15.

Exner, M. E., Burbach, M. E., Watts, D. G., Shearman, R. C. and Spalding, R. F. 1991. Deep nitrate movement in the unsaturated zone of a simulated urban lawn. J. Environ. Qual. **20**: 658-662.

Firestone, M. K., Smith, M. S., Firestone, R. B. and Tiedje, J. M. 1979. The influence of nitrate, nitrite, and oxygen on the composition of the gaseous products of denitrification in soil. Soil Sci. Soc. Am. J. **43**: 1140-1144.

Folorunso, O. A. and Rolston, D. E. 1984. Spatial variability of field-measured denitrification gas fluxes. Soil Sci. Soc. Am. J. **48**: 1214-1219.

Folorunso, O. A. and Rolston, D. E. 1985. Spatial and spectral relationships between field-measured denitrification gas fluxes and soil properties. Soil Sci. Am. J. **49**: 1087-1093.

Gilliam, J. W. and Gambrell, R. P. 1978. Temperature and pH as limiting factors in loss of nitrate from saturated Atlantic Coastal Plain soils. J. Environ. Qual. **7**: 526-532.

Gilliam, J. W., Dasberg, S., Lund L. J., and Focht, D. D. 1978. Denitrification in four California soils: Effect of Soil Profile Characteristics. Soil Sci. Soc. Am. J. **42**:61-66.

Gödde, Monika and Conrad, Ralf. 1998. Simultaneous measurement of nitric oxide production and consumption in soil using a simple static incubation system, and the effect of soil water content on the contribution of nitrification. Soil Biol. Biochem. **30**: 433-442.

Groffman, P. M. and Tiedje, J. M. 1991. Relationships between denitrification, CO_2 production and air-filled porosity in soils of different texture and drainage. Soil Biol. Biochem. **23**: 299-302.

Groffman, P. M., Gold, A. J. and Simmons, R. C. 1992. Nitrate dynamics in riparian forests: Microbial studies. J. Environ. Qual. **21**:666-671.

Grundmann, G. L., Rolston, D. E. and Kachanoski, R. G. 1988. Field soil properties influencing the variability of denitrification gas fluxes. Soil Sci. Soc. Am. J. **52**: 1351-1355.

Hargrave, A. P. and Shaykewich, C. F. 1997. Rainfall-induced nitrogen and phosphorous losses from Manitoba soils. Can. J. Soil Sci. **77**: 59-65.

Hatch, D. J., Jarvis, S. C. and Parkinson, R. J. 1998. Concurrent measurements of net mineralization, nitrification, denitrification and leaching from field incubated soil cores. Biol. Fertil. Soils **26**: 323-330.

Hedlin, R. A. 1995. Agriculture on the Canadian prairies: Retrospect and prospect. Can. J. Soil Sci. **75**: 253-259.

Henault, C., Devis, X., Page, S., Justes, E., Reau, R. and Germon, J. C. 1998. Biol. Fertil. Soils **26**: 199-207.

Hixson, S. E., Walker, R. F., and Skau, C. M. 1990. Soil denitrification rates in four subalpine plant communities of the Sierra Nevada. J. Environ. Qual. **19**:617-620.

Izaurralde, R. C., Feng, Y., Robertson, J. A., McGill, W. B., Juma, N. G., and Olson, B. M. 1995. Long-term influence of cropping systems, tillage methods, and N sources on nitrate leaching. Can. J. Soil Sci. **75**: 497-505.

Jacobson, S. N. and Alexander, M. 1980. Nitrate loss from soil in relation to temperature, carbon source and denitrifier populations. Soil Biol. Biochem. **12**: 501-505.

Jordan, T. E., Weller, D. E., and Correll, D. L. 1998. Denitrification in surface soils of a riparian forest: effects of water, nitrate and sucrose additions. Soil Biol. Biochem. **30**: 833-843.

Katz, R., Hagin, J. and Kurtz, L. T. 1985. Participation of soluble and oxidizable soil organic compounds in denitrification. Biol. Fertil. Soils **1**: 209-213.

Kladivko, E.J., Van Scoyoc, G. E., Monke, E. J., Oates, K. M. and Pask, W. 1991. Pesticide and nutrient movement into subsurface tile drains on a silt loam soil in Indiana. J. Environ. Qual. **20**: 264-270.

Klemedtsson, L., Svensson, B. H. and Rosswall, T. 1988. Relationships between soil moisture content and nitrous oxide production during nitrification and denitrification. Biol. Fertil. Soils **6**: 106-111.

Kowalenko, C. G. and Yu, S. 1996. Assessment of nitrate adsorption in soils by extraction, equilibration and column-leaching methods. Can. J. Soil Sci. **76**: 49-57.

Kroeckel, L. and Stolp, H. 1985. Influence of oxygen on denitrification and aerobic respiration in soil. Biol. Fertil. Soils **1**: 189-193.

Kroeckel, L. and Stolp, H. 1986. Influence of the water regime on denitrification and aerobic respiration in the soil. Biol. Fertil. Soils **2**: 15-21.

Levanon, D., Codling, E. E., Meisinger, J. J. and Starr, J. L. 1993. Mobility of agrochemicals through soil from two tillage systems. J. Environ. Qual. **22**: 155-161.

Lind, A. M. and Eiland, F. 1989. Microbiological characterization and nitrate reduction in subsurface soils. Biol. Fertil. Soils **8**: 197-203.

Lowrance, R. 1992. Nitrogen outputs from a field-size agricultural watershed. J. Environ. Qual. **21**: 602-607.

Lund, L. J. 1982. Variations in nitrate and chloride concentrations below selected agricultural fields. Soil Sci. Soc. Am. J. **46**: 1062-1066.

Luo, J., Tillman, R. W., White, R. E. and Ball, P. R. 1998. Variation in denitrification activity with soil depth under pasture. Soil Biol. Biochem. **30**: 897-903.

Magdoff, F. 1992. Minimizing nitrate leaching in agricultural production: how good can we get? Commun. Soil Sci. Plant Anal. **23**: 2103-2109.

Mahli, S. S., McGill, W. B. and Nyborg, M. 1990. Nitrate losses in soils: effect of temperature, moisture and substrate concentration. Soil Biol. Biochem. **22**: 733-737.

Mahli, S.S., Nyborg, M. and Solberg, E. D. 1996. Influence of source, method of placement and simulated rainfall on the recovery of ^{15}N-labelled fertilizers under zero tillage. Can. J. Soil Sci. **76**: 93-100.

Mangaraja, S. and Misra, C. 1978. Effect of temperature on denitrification. J. Indian Soc. Soil Sci. **26**: 401-402.

Maynard, D. G. and Kalra, Y. P. 1993. Chapter 4: Nitrate and exchangeable ammonium nitrogen. Pages 25-38. *In* Lewis Publishers, CRC Press, Inc. Carter M. R. (ed.). Soil Sampling and Methods of Analysis. Boca Raton, Florida.

McCarty, G. W. and Bremner, J. M. 1992. Availability of organic carbon for denitrification of nitrate in subsoils. Biol. Fertil. Soils **14**: 219-222.

McCarty, G. W. and Bremner, J. M. 1993. Factors affecting the availability of organic carbon for denitrification of nitrate in subsoils. Biol. Fertil. Soils **15**: 132-136.

Misra, C. and Mishra, B. K. 1977. Miscible dispacement of nitrate and chloride under field conditions. Soil Sci. Soc. Am. J. **41**: 496-499.

Müller, M. M., Sundman, V. and Skujinš, J. 1980. Denitrification in low pH Spodosols and peats determined with the acetylene inhibition method. Appl. Environ. Microbiol. **40**: 235-239.

Myrold, D. D. and Tiedje, J. M. 1985. Establishment of denitrification capacity in soil: effects of carbon, nitrate and moisture. Soil Biol. Biochem. **17**: 819-822.

Nägele, W. and Conrad, R. 1990a. Influence of pH on the release of NO and N_2O from fertilized and unfertilized soil. Biol. Fertil. Soils **10**: 139-144.

Nägele, W. and Conrad, R. 1990b. Influence of soil pH on the nitrate-reducing microbial populations and their potential to reduce nitrate to NO and N_2O. FEMS Microbiol. Ecol. **74**: 49-58.

Obenhuber, D. C. and Lowrance, R. 1991. Reduction of nitrate in aquifer microcosms by carbon additions. J. Environ. Qual. **20**: 255-258.

Owens, L. B., Edwards, W. M. and Van Keuren, R. W. 1992. Nitrate levels in shallow groundwater under pastures receiving ammonium nitrate or slow-release nitrogen fertilizer. J. Environ. Qual. **21**: 607-613.

Pang, P. C. K., Cho, C. M. and Hedlin, R. A. 1977. Distribution and transformation of band-applied urea in soil following incubation under isothermal and temperature gradient conditions. Can. J. Soil Sci. **57**: 409-416.

Panganiban, E. H. 1925. Temperature as a factor of nitrogen changes in the soil. J. Am. Soc. Agron. **17**: 1-31.

Paramasivam, S., Alva, A. K., Prakash, O. 1998. Denitrification from sandy soils treated with liquid or dry granular nitrogen form. Soil Science **163**: 686-693.

Parkin, T. B. 1987. Soil microsites as a source of denitrification variability. Soil Sci. Soc. Am. J. **51**: 1194-1199.

Parkin, T. B. and Tiedje, J. M. 1984. Application of a soil core method to investigate the effect of oxygen concentration on denitrification. Soil Biol. Biochem. **16**: 331-334.

Parkin, T. B., Sexstone, A. J. and Tiedje, J. M. 1985. Adaptation of denitrifying populations to low soil pH. Appl. Environ. Microbiol. **49**: 1053-1056.

Parsons, L. L., Murray, R. E. and Smith, M. S. 1991. Soil denitrification dynamics: spatial and temporal variations of enzyme activity, populations, and nitrogen gas loss. Soil Sci. Soc. Am. J. **55**: 90-95.

Patrick, W. H. Jr., Reddy, K. R. 1976. Nitrification-denitrification reactions in flooded soils and water bottoms: dependence on oxygen supply and ammonium diffusion. J. Environ. Qual. **5**: 469-472.

Paul, E. A. 1984. Dynamics of organic matter in soils. Plant and Soil. **76**: 275-285.

Paul, E. A. and Clark, F. E. 1989. Chapter 9: Reduction and transport of nitrogen. Pages 149-165. *In* Academy Press Inc. (Eds.). <u>Soil Microbiology and Biochemistry</u>. San Diego.

Pelletier, F., Prévost, D., Laliberté, G., and van Bochove, E. 1999. Seasonal response of denitrifiers to temperature in a Quebec cropped soil. Can. J. Soil Sci. **79**: 551-556.

Pennock, D. J., van Kessel, C., Farrell, R. E. and Sutherland, R. A. 1992. Landscape-scale variations in denitrification. **56**:770-776.

Peterjohn, W. T. 1991. Denitrification: Enzyme content and activity in desert soils. Soil Biol. Biochem. **23**:845-855.

Peterjohn, W. T. and Schlesinger, W. H. 1991. Factors controlling denitrification in a Chihuahuan desert ecosystem. Soil Sci. Soc. Am. J. **55**:1694-1701.

Roth, G. W. and Fox, R. H. 1990. Soil nitrate accumulations following nitrogen-fertilized corn in Pennsylvania. J. Environ. Qual. **19**: 243-248.

Ryden, J. C. 1983. Denitrification loss from a grassland soil in the field receiving different rates of nitrogen as ammonium. J. Soil Sci. **34**: 355-365.

Seech, A. G. and Beauchamp, E. G. 1988. Denitrification in soil aggregates of different sizes. Soil Sci. Soc. Am. J. **52**: 1616-1621.

Sexstone, A. J., Parkin, T. B. and Tiedje, J. M. 1985. Temporal response of soil denitrification rates to rainfall and irrigation. Soil Sci. Soc. Am. J. **49**: 99-103.

Sexstone, A. J., Revsbech, N. P., Parkin, T. B. and Tiedje, J. M. 1985. Direct measurement of oxygen profiles and denitrification rates in soil aggregates. Soil Sci. Am. J. **49**: 645-651.

Sheldrick, B. H. and Wang, C. 1993. Chapter 47: Particle size distribution. Pages 499-512. *In* Lewis Publishers, CRC Press, Inc. Carter M. R. (ed.). Soil Sampling and Methods of Analysis. Boca Raton, Florida.

Shuford, J. W., Fritton, D. D. and Baker, D. E. 1977. Nitrate-nitrogen and chloride movement through undisturbed field soil. J. Environ. Qual. **6**: 255-259.

Simmons, K. E. and Baker, D. E. 1993. A zero-tension sampler for the collection of soil water in macropore systems. J. Environ. Qual. **22**: 207-212.

Simmons, R. C., Gold, A. J. and Groffman, P. M. 1992. Nitrate dynamics in riparian forests: groundwater studies. J. Environ. Qual. **21**: 659-665.

Smid, A. E. and Beauchamp, E. G. 1976. Effects of temperature and organic matter on denitrification in soil. Can. J. Soil Sci. **56**: 385-391.

Smith, M. S. and Tiedje, J. M. 1979a. Phases of denitrification following oxygen depletion in soil. Soil Biol. Biochem. **11**: 261-267.

Smith, M. S. and Tiedje, J. M. 1979b. The effect of roots on soil denitrification. Soil Sci. Soc. Am. J. **43**: 951-955.

Smith, S. J. 1972. Relative rate of chloride movement in leaching of surface soils. Soil Science **114**: 259-263.

Stanford, G., Dzienia, S. and Vander Pol, R. A. 1975. Effect of temperature on denitrification rate in soils. Soil Sci. Soc. Am. Proc. **39**: 867-870.

Stanford, G., Legg, J. O., Dzienia, Stanislaw, and Simpson, E. C., Jr. 1975. Denitrification and associated nitrogen transformations in soils. Soil Sci. **120**: 147-152.

Stenberg, B., Johansson, M., Pell, M., Sjödahl-Svensson, K., Stenström, and Torstensson, L. 1998. Microbial biomass and activities in soil as affected by frozen and cold storage. Soil Biol. Biochem. **30**: 393-402.

Terry, R. E. and Tate, R. L. III. 1980. The effect of nitrate on nitrous oxide reduction in organic soils and sediments. Soil Sci. Soc. Am. J. **44**: 744-746.

Tiedje, J. M., Sexstone, A. J., Myrold, D. D. and Robinson, J. A. 1982. Denitrification: ecological niches, competition and survival. Antonie van Leeuwenhoek **48**: 569-583.

Tiessen, H. and Moir, J. O. 1993. Chapter 21: Total and organic carbon. Pages 187-200. *In* Lewis Publishers, CRC Press, Inc. Carter M. R. (ed.). Soil Sampling and Methods of Analysis. Boca Raton, Florida.

Tisdale, S. L., Nelson, W. L. and Beaton, J. D. 1985. Chapter 5: Soil and fertilizer nitrogen. Pages 112-188. *In* McMillan Publishing Co. (Eds.). Soil Fertility and Fertilizers. 4th edition. New York.

Toth, John D. and Fox, Richard H. 1998. Nitrate losses from a corn-alfalfa rotation: lysimeter measurement of nitrate leaching. J. Environ. Qual. **27**: 1027-1033.

Trevors, J. T. 1985. The influence of oxygen concentrations on denitrification in soil. Appl. Microbiol. Biotechnol. **23**: 152-155.

Tyler, D. D. and Thomas, G. W. 1977. Lysimeter measurements of nitrate and chloride losses from soil under conventional and no-tillage corn. J. Environ. Qual. **6**: 63-66.

Wang, F. L. and Alva, A. K. 1999. Transport of soluble organic and inorganic carbon in sandy soils under nitrogen fertilization. Can. J. Soil Sci. **79**: 303-310.

Waring, S. A. and Gilliam, J. W. 1983. The effect of acidity on nitrate reduction and denitrification in lower Coastal Plains soils. Soil Sci. Soc. Am. J. **47**: 246-251.

Watts, D. G., Hergert, G. W. and Nichols, J. T. 1991. Nitrogen leaching losses from irrigated orchardgrass on sandy soils. J. Environ. Qual. **20**: 355-362.

Weier, K. L., Doran, J. W., Power, J. F. and Walters, D. T. 1993. Denitrification and the dinitrogen/nitrous oxide ratio as affected by soil water, available carbon, and nitrate. Soil Sci. Soc. Am. J. **57**: 66-72.

Weier, K. L., MacRae, I. C. and Myers, R. J. K. 1993. Denitrification in a clay soil under pasture and annual crop: Estimation of potential losses using intact soil cores. **25**:991-997.

Weil, R. R., Weismiller, R. A. and Turner, R. S. 1990. Nitrate contamination of groundwater under irrigated Coastal Plain soils. J. Environ. Qual. **19**: 441-448.

Wheatley, R. E., Griffiths, B. S. and Ritz, K. 1991. Variations in the rates of nitrification and denitrification during growth of potatoes (*Solanum tuberosum* L.) in soil with different carbon inputs and the effect of these inputs on soil nitrogen and plant yield. Biol. Fertil. Soils **11**: 157-162.

White, V. 1996. Agriculture and drinking water supplies: removing nitrates from drinking water in Des Moines, Iowa. J. Soil and Water Cons. **51**: 454-456.

Williams, P. H., Jarvis, S. C., and Dixon, E. 1998. Emission of nitric oxide and nitrous oxide from soil under field and laboratory conditions. Soil Biol. Biochem. **30**: 1885-1893.

9. APPENDICES

I. Mean soil temperatures for different profile depths of Stockton soil-Plot 1- for all sampling dates in 1991.

Soil Depth (cm)	Mean Soil Temperature (°C)									
	May 16[1]	May 23	June 11	June 18	June 27	July 23	Aug 8	Aug 19[2]	Sept 11	Sept 25
0	29.4	37.2±0.9	33.8±2.1	26.8±2.8	27.8±1.0	35.4±2.1	36.3±5.9	34.7±1.3	23.5±1.8	13.9±0.8
2.5	28.0	32.4±0.6	30.7±2.9	25.1±1.5	25.6±1.7	32.2±3.0	32.4±3.5	33.5±0.7	20.6±2.8	13.0±2.3
5.0	27.9	25.3±7.3	23.0±5.6	21.7±3.9	22.1±3.6	26.3±6.4	26.4±8.1	30.4±2.4	18.4±3.1	11.3±2.2
10.0	25.6	22.8±1.5	23.1±3.1	21.6±1.9	21.4±1.3	22.3±1.3	22.4±1.9	23.9±0.3	16.0±1.0	9.4±0.4
30.0	16.2	20.9±6.4	21.2±6.1	20.6±2.8	21.0±2.5	22.4±4.1	22.3±4.6	21.7±2.0	17.0±0.9	10.5±0.7
60.0	12.2	14.6±0.5	16.2±0.8	18.0±1.0	18.4±1.0	19.9±1.4	19.0±1.5	21.5±0.8	17.7±1.3	12.4±0.6
90.0	8.0	11.0±0.6	14.4±0.8	15.7±0.9	16.6±1.0	18.2±1.1	17.9±1.5	20.0±1.2	17.2±0.6	13.8±0.8
120.0	4.8	8.4±0.6	12.3±1.0	13.9±0.6	14.9±0.8	16.6±0.8	17.4±1.5	18.7±1.4	17.4±1.5	14.6±1.0

[1] Temperatures are based on results from one probe.

[2] Mean temperatures are based on results from two probes.

II. Mean soil temperatures for different profile depths of Wellwood soil -Plot 2- for all sampling dates in 1991.

Soil Depth (cm)	**Mean Soil Temperature (°C)**							
	May 16[1]	May 23	June 11	July 23	Aug 8	Aug 19	Sept 11	Sept 25
0	28.0	35.9±3.0	33.2±4.1	35.3±5.2	42.7±6.1	46.8±2.8	30.0±2.4	14.6±2.5
2.5	28.1	29.7±0.5	30.6±3.0	33.4±1.9	37.9±2.2	43.9±0.7	28.0±2.5	14.6±0.5
5.0	28.0	26.8±1.1	28.1±4.3	31.7±4.8	33.9±3.7	39.2±4.9	24.6±2.3	14.0±1.7
10.0	22.6	22.7±1.1	23.0±3.7	26.0±5.4	26.7±4.3	27.3±4.2	18.9±1.6	11.2±1.1
30.0	14.1	15.8±0.3	16.3±0.8	19.9±1.8	18.4±1.0	20.1±2.1	16.9±0.7	11.5±0.2
60.0	9.8	12.2±0.5	14.3±0.9	18.7±1.8	17.3±1.1	19.1±2.1	17.6±1.2	13.0±0.3
90.0	6.1	8.6±0.4[2]	11.6±0.6[2]	16.2±1.8[2]	16.3±1.8[2]	17.5±2.5[2]	17.2±2.1[2]	13.9±0.8[2]
120.0	4.0	6.2±0.4	10.2±0.5	14.5±1.2	15.4±0.9	16.5±1.9	16.6±1.6	14.6±0.8

[1] Temperatures are based on results from one probe.
[2] Mean temperatures are based on results from two probes.

III. Mean soil temperatures for different profile depths of Almasippi soil - Plot 3- for all sampling dates in 1991.

Soil Depth (cm)	Mean Soil Temperature (°C)													
	May 28	June 27	July 4	July 10	July 23	July 30	Aug 7	Aug 15	Aug 22	Aug 28	Sept 5	Sept 13	Sept 19	Oct 3
0	35.5± 2.8[1]	29.0± 1.3	19.7± 1.0	30.8± 4.9	33.1± 2.6	36.1± 7.6	28.0± 4.6	22.2± 1.0	32.8± 4.2	33.9± 4.0	25.8± 2.4	23.3± 6.4	15.3± 1.3	13.9± 2.2
2.5	33.5[2]	27.7[2]	20.0± 1.4[1]	29.0± 2.4[1]	30.2± 3.6[1]	34.3± 5.3[1]	23.8± 2.7[1]	23.1± 0.6[1]	29.3± 6.2	31.1± 5.1	23.1± 3.1	20.1± 5.4	14.9± 2.5	13.7± 1.8
5.0	23.0± 3.0[1]	24.8± 1.0	18.2± 0.4	24.2± 2.3	26.6± 3.0	26.2± 2.7	20.7± 1.2	23.6± 0.1	24.6± 2.6	27.2± 2.8	20.4± 2.4	16.6± 2.2	12.6± 1.4	12.3± 1.2
10.0	18.8± 0.6[1]	21.9± 0.5	17.3± 0.1	20.5± 0.8	24.4± 1.7	22.0± 0.7	17.8± 0.7	23.6± 0.2	21.4± 0.6	24.0± 1.0	17.3± 0.8	14.6± 0.3	10.4± 0.6	10.6± 0.7
30.0	15.1± 0.2[1]	18.4± 0.7	16.6± 0.2	18.3± 0.3	19.4± 0.4	20.1± 0.0	17.0± 0.5	23.2± 0.3	21.6± 0.0	22.0± 0.7	17.2± 0.1	16.4± 0.2	10.0± 0.1	9.7± 0.4
60.0	13.2± 0.1[1]	17.2± 0.9	16.0± 0.3	17.1± 0.4	18.6± 0.3	19.1± 0.3	17.2± 0.5	21.3± 0.4	21.1± 0.3	20.5± 0.4	18.9± 0.2	17.2± 0.3	13.4± 0.2	11.4± 0.3
90.0	10.5± 0.1[1]	15.5± 1.1	15.0± 0.3	15.4± 0.4	17.6± 0.3	17.3± 0.2	16.6± 0.5	19.6± 0.9	19.4± 0.3	18.6± 0.5	18.7± 0.4	16.9± 0.3	14.9± 0.3	12.1± 0.6
120.0	8.2± 0.1[1]	13.5± 1.0	13.9± 0.3	14.1± 0.4	15.9± 0.3	15.9± 0.1	15.6± 0.7	17.4± 0.3	17.9± 0.1	17.4± 0.4	18.1± 0.4	16.4± 0.2	15.4± 0.2	12.5± 0.3

[1] Mean temperatures are based on results from two probes.

[2] Temperatures are based on results from one probe.

IV. Soil temperatures for different profile depths of Stockton soil - Plot 1- for all sampling dates in 1992.

Soil Depth (cm)	**Soil Temperature (°C)**							
	May 12	May 27	June 22	July 7	July 21	Aug 6	Aug 18	Aug 31
0	14.3	35.7	36.8	39.3	35.7	29.2	28.6	23.5
2.5	12.7	33.8	28.1	38.0	30.7	26.5	26.6	20.5
5.0	12.7	33.3	31.3	35.8	29.3	25.8	24.5	18.7
10.0	9.8	25.1	25.8	25.1	23.3	21.0	17.5	14.1
30.0	10.0	12.3	15.2	15.3	15.8	17.5	14.9	13.2
60.0	9.9	11.5	14.3	15.0	15.8	17.2	16.4	14.4
90.0	8.1	10.1	14.1	14.3	14.7	15.9	15.2	15.4
120.0	6.6	8.8	13.1	12.7	14.5	14.9	14.1	14.7

V. Soil temperatures for different profile depths of Wellwood soil -Plot 2- for all sampling dates in 1992.

Soil Depth (cm)	Soil Temperature (°C)								
	May 12	May 27	June 22	July 7	July 21	Aug 6	Aug 18	Aug 31	Sept 17
0	8.7	45.4	34.2	31.6	26.7	27.3	40.7	16.0	13.4
2.5	10.0	37.6	31.5	34.7	25.8	26.0	36.3	20.0	13.0
5.0	10.2	34.4	29.3	29.4	25.2	25.3	30.6	19.7	12.8
10.0	10.2	24.1	21.2	21.3	19.5	20.0	22.0	14.3	10.6
30.0	11.3	12.2	15.0	14.9	14.7	17.9	18.6	14.2	11.5
60.0	10.1	10.7	14.4	14.3	14.8	17.5	18.0	16.0	13.7
90.0	-	9.1	12.5	13.0	14.8	15.9	17.7	15.7	14.1
120.0	5.9	8.5	12.0	12.0	12.5	15.0	14.5	15.5	14.5

VI. Soil temperatures for different profile depths of Almasippi soil- Plot 3- for all sampling dates in 1992.

Soil Depth (cm)	**Soil Temperature (°C)**																
	May 6	**May 13**	**May 20**	**May 25**	**June 2**	**June 16**	**June 22**	**June 29**	**July 7**	**July 14**	**July 21**	**July 28**	**Aug 5**	**Aug 19**	**Sept 2**	**Sept 10**	**Sept 21**
0	**26.3**	**19.8**	**38.2**	**24.9**	**32.8**	**14.0**	**31.1**	**25.8**	**28.5**	**34.7**	**23.7**	**20.8**	**29.4**	**27.0**	**19.5**	**14.7**	**10.6**
2.5	**23.4**	**16.0**	**31.7**	**21.0**	**27.5**	**14.1**	**27.8**	**25.8**	**28.7**	**31.2**	**23.5**	**19.8**	**26.2**	**21.8**	**17.8**	**12.0**	**7.1**
5.0	**19.9**	**14.3**	**27.9**	**15.3**	**23.8**	**14.8**	**25.5**	**20.3**	**27.9**	**26.3**	**23.0**	**18.9**	**23.2**	**19.4**	**17.3**	**11.0**	**5.8**
10.0	**14.5**	**11.3**	**24.4**	**11.0**	**20.8**	**16.4**	**21.8**	**17.4**	**24.4**	**22.6**	**21.3**	**17.2**	**20.3**	**17.7**	**16.0**	**9.8**	**6.2**
30.0	**10.5**	**8.8**	**16.0**	**8.7**	**17.7**	**18.5**	**14.8**	**16.7**	**16.4**	**19.5**	**16.5**	**17.7**	**17.6**	**17.9**	**14.5**	**13.0**	**10.2**
60.0	**8.6**	**9.5**	**12.1**	**10.0**	**15.0**	**16.7**	**13.7**	**16.6**	**14.8**	**17.8**	**15.8**	**15.8**	**17.0**	**17.7**	**14.4**	**13.7**	**12.2**
90.0	**6.7**	**8.6**	**9.5**	**9.6**	**12.5**	**14.6**	**13.5**	**15.2**	**14.0**	**15.6**	**15.1**	**17.1**	**15.8**	**16.6**	**14.2**	**13.8**	**12.6**
120.0	**5.1**	**7.3**	**7.9**	**8.7**	**10.8**	**12.8**	**10.6**	**13.7**	**13.0**	**14.0**	**14.3**	**16.2**	**14.9**	**15.7**	**14.0**	**14.1**	**12.9**

VII. Oxygen flux of Almasippi soil in 1991 under two cropping treatments.

Depth (cm)	15/08/91	22/08/91	28/08/91	05/09/91	13/09/91	19/09/91	03/10/91
				Fallow			
				$\mu g\ O_2\ cm^{-2}\ min^{-1}$			
10	1.00	1.13	1.34	1.21	1.20	0.96	0.94
20	1.33	1.39	1.07	0.96	0.87	0.84	0.44
30	1.45	1.37	1.03	0.96	0.89	0.81	0.47
50	1.52	1.46	1.27	1.20	1.10	1.06	0.68
75	0.37	0.37	0.41	0.41	0.43	0.40	0.15
100	0.11	0.12	0.35	0.40	1.00	1.13	0.54
				Wheat			
				$\mu g\ O_2\ cm^{-2}\ min^{-1}$			
10	0.22	0.61	0.51	0.47	0.46	0.60	0.45
20	0.74	0.81	0.87	0.75	0.74	0.66	0.68
30	1.30	1.19	1.19	0.98	0.96	0.82	0.62
50	1.27	1.05	1.00	0.85	0.82	0.78	0.69
75	0.10	0.14	0.13	0.12	0.12	0.16	0.11
100	0.79	0.90	0.72	0.64	0.54	0.51	0.19

VIII. Oxygen flux of Stockton soil for two field seasons.

Depth (cm)	05/16/91	08/19/91	09/11/91	09/25/91
		$\mu g\ O_2\ cm^{-2}\ min^{-1}$		
10	0.03	0.86	1.08	1.19
20	0.92	1.00	1.20	0.66
30	1.10	1.04	1.15	0.93
50	1.14	1.20	1.45	0.68
50-60		0.57	0.61	0.24

Depth (cm)	06/22/92	07/07/92	07/21/92	08/06/92
		$\mu g\ O_2\ cm^{-2}\ min^{-1}$		
10	0.90	1.45	0.49	0.70
20	0.96	1.14	0.80	1.02
40	0.04	0.01	0.01	0.01
50	0.00	0.00	0.48	0.57
70	0.39	0.46	0.41	0.41

IX. Oxygen flux of Almasippi soil in 1992 under two cropping treatments.

Depth (cm)	16/06/92	22/06/92	29/06/92	07/07/92	21/07/92	28/07/92	05/08/92	26/08/92	02/09/92
					Fallow				
					$\mu g\ O_2\ cm^{-2}\ min^{-1}$				
10	1.06	1.76	1.33	1.26	1.58	0.92	1.43	1.27	1.20
20	1.46	1.82	1.38	1.21	0.75	0.67	0.21	0.00	0.00
30	1.40	1.09	0.79	0.76	0.43	0.25	0.12	0.02	0.00
50	1.63	1.28	1.23	1.16	1.18	1.11	0.06	0.69	0.57
75	0.08	0.08	0.05	0.06	0.03	0.04	0.11	0.80	0.77
100	0.13	0.09	0.19	0.20	0.05	0.00	0.00	0.04	0.04
					Wheat				
					$\mu g\ O_2\ cm^{-2}\ min^{-1}$				
10	1.15	1.08	1.16	1.39	0.97	0.71	0.86	0.67	0.84
20	1.39	1.23	1.24	1.29	1.21	1.04	0.03	0.02	0.03
30	1.53	1.32	1.36	1.04	1.03	1.00	0.03	0.02	0.03
50	1.53	1.32	1.36	1.30	1.26	1.24	0.06	1.00	0.96
75	0.18	0.14	0.17	0.15	0.11	0.06	0.06	1.22	1.42

Appendix X. Equations for rates of nitrate disappearance for the Almasippi and Stockton soil profiles (0-120 cm depth).

Soil Temperature (°C)	**Equations for nitrate disappearance (x = depth in cm)**	
	Almasippi profile	**Stockton profile**
	rate (µg NO_3-N g^{-1} day^{-1})	**ln rate (µg NO_3-N g^{-1} day^{-1})**
5		$y = 1.215 - 0.016x$
11	$y = 0.813e^{(-0.027x)} + 3.004e^{(-0.016x)}$	$y = 2.260 - 0.023x$
16.5	$y = 5.85e^{(-0.079x)} + 2.47e^{(-0.009x)}$	$y = 2.731 - 0.024x$
20.5	$y = 21.46e^{(-0.136x)} + 3.63e^{(-0.014x)}$	$y = 2.872 - 0.027x$
26	$y = 55.7e^{(-0.179x)} + 5.37e^{(-0.009x)}$	$y = 4.396 - 0.057x$

Appendix XI. Arrhenius equations for the effect of temperature on rates of nitrate disappearance for the Almasippi and Stockton soil profiles (5-26°C).

Soil Depth (cm)	**Arrhenius Equations (x = 1/temperature in °K^{-1})**	
	Almasippi profile	**Stockton profile**
	ln rate (µg NO_3-N g^{-1} day^{-1})	
0-15	$y = -11833x + 42.64$	$y = -9329x + 35.12$
15-30	$y = -6786x + 24.29$	$y = -7362x + 27.41$
30-45	$y = -9435x + 33.12$	$y = -6234x + 22.93$
45-60	$y = -5470x + 19.26$	$y = -5871x + 21.46$
60-75	$y = -5415x + 18.92$	$y = -4949x + 17.91$
75-90	$y = -4635x + 16.03$	$y = -3779x + 13.55$*
90-105	$y = -5949x + 20.74$	$y = -3231x + 11.51$*
105-120	$y = -3423x + 11.72$	$y = -3124x + 10.79$*

* Equations are based on four temperatures only: 5°C, 11°C, 16.5°C and 20.5°C.