UNIVERSITY OF CAPE COAST

NUTRIENT DYNAMICS IN A COASTAL SAVANNA SOIL AMENDED WITH NEEM (*AZADIRACHTA INDICA*) LEAVES AND ANIMAL MANURE

BY
KOFI AGYARKO

A THESIS SUBMITTED TO THE DEPARTMENT OF SOIL SCIENCE, UNIVERSITY OF CAPE COAST, IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR THE AWARD OF
DOCTOR OF PHILOSOPHY (LAND USE AND EVIRONMENTAL SCIENCE) DEGREE

DECEMBER, 2004

CANDIDATE'S DECLARATION

I hereby declare that this is the result of my own original research and that no part has been presented for another degree in this University or elsewhere.

..... 14/12/2004

Kofi Agyarko Date

SUPERVISORS' DECLARATION

We hereby declare that the preparation and presentation of this thesis were supervised in accordance with the guidelines on supervision of thesis laid down by the University of Cape Coast.

..... 20/12/2004

Professor P. K. Kwakye Date

(Principal Supervisor)

..... 20/12/2004

Professor Mensah Bonsu Date

(Supervisor)

....

Dr B. A. Osei Date

(Supervisor)

DEDICATED TO THE SUPREME GOD THE FATHER, THE SON AND THE HOLY GHOST

ACKNOWLEDGEMENT

My sincere gratitude goes to my supervisors, Professor P. K. Kwakye, Rev. Fr. Professor Mensah Bonsu and Dr. B. A. Osei of the Department of Soil Science – University of Cape Coast, whose cooperation and guidance have made this research a success.

I equally thank the University of Education, Winneba and the Centre for School and Community Science and Technology Studies (SACOST) – University of Education, Winneba, for the financial assistance offered towards this research.

Professor Murray Isman of the University of British Columbia, Canada, needs to be appreciated for providing free of charge the pure analytical grade of Azadirachtin A which served as a catalyst for this research to take off.

Further compliments go to the entire staff of the Department Soil Science, University of Cape Coast and Mr. John Sackey Asante of the Nematology Department, CSIR, Fumesua, Kumasi for their technical support in the course of the research.

The hospitality of the staff of School of Agriculture, University of Cape Coast, is much appreciated.

I extend my heartfelt appreciation to my wife, Mrs. Agnes Agyarko, who in the pursuance of this research study took pains to look after my family.

I also acknowledge my parents, brothers and sisters for their moral support.

My final and sincere thanks go to the Supreme God the Father, the Son and the Holy Ghost for the divine guidance provided throughout the studies.

ABSTRACT

Sustainability of agriculture with the ever-increasing trend in population calls for urgent steps to make lands available for agricultural production. On these lands maintenance of soil fertility through the use of environmentally friendly diverse organic materials is imminent. Neem leaves and animal manure, particularly poultry and cow dung manure abound all year round and the potentials of these organic material are explored in the research study.

The study sought to investigate the performance of a soil amendment, the materials of which are various inclusion rates of neem leaves, poultry manure and cow dung applied to a Haplic Acrisol (Benya series), from a coastal savanna ecological zone in Ghana. Field and pot experiments were carried out at the Technology Village of the University of Cape Coast, from September 2002 to October 2003.

A proposition that an active ingredient in the neem leaves (azadirachtin A) might have a vital role to play in the amendment was investigated using Gas Chromatography to study its breakdown in the soil. Also, that the neem leaves in combination with animal manure could boost up the nutrient content of the soil as well as influencing the population dynamics of nematodes within the soil were studied. Above all, the efficiency of the soil amendment in achieving the afore-mentioned properties was tested by monitoring the performance of a test crop (carrot) under controlled conditions using pots.

Degradation of azadirachtin A in the soil followed the first order reaction kinetics. Degradation was fastest in the amendment which had 100 g neem leaves/kg soil. The inclusions of 5 g and 10 g poultry manure and 10 g cow dung to the neem leaves hastened the degradation of azadirachtin A. The interaction between poultry manure and the neem leaves enhanced the release of nutrients in the soil.

The peak of release of most of the soil nutrients occurred two weeks after incorporation of the neem leaves and poultry manure. The amount of CO_2 – C evolved relatively corresponded with the quantity of neem leaves and poultry manure added to the soil. The release of NH_4^+ – N, available P, exchangeable K, Ca and Mg was a mirror image of CO_2 – C evolved and the quantities also corresponded with the quantity of neem leaves and poultry manure incorporated into the soil. The amount of NO_3^--N released, however, was an exception to the above observed trend; the lower inclusion rates of the neem leaves and poultry manure released more nitrate than the higher levels at a point in time. This exception was assigned to the nitrification inhibitory role played by the neem leaves in the amended soil.

The 50 g neem leaves + 5 g poultry manure/kg soil proved to be the most effective in the control of root-knot nematodes on carrot roots. Generally, with the apparent increase in soil nutrients and the reduced root-knot nematodes of carrot the yield of the crop improved significantly.

Neem leaves and poultry manure may be used in place of synthetic compounds to provide soil nutrients and control plant parasitic nematodes, and thus, improve the growth and yield of carrots. Neem leaves may also be used to slow down the release of nitrates in the soil and thus reduce the ultimate pollution caused by nitrate leaching.

TABLE OF CONTENTS

		Page
	Candidate's and Supervisors' declaration	i
	Dedication	ii
	Acknowledgement	iii
	Abstract	iv
CHAPTER 1	Introduction	1
CHAPTER 2	Literature Review	6
	2.1 Organic manure and crop production	6
	2.2 Nutrients components of some manure materials	8
	2.3 Organic amendments and soil fertility	10
	2.3.1 Carbon dynamics	10
	2.3.2 Nitrogen dynamics	11
	2.3.3 C:N Ratio	13
	2.3.4 Phosphorus dynamics	14
	2.3.5 Exchangeable cations (K, Ca and Mg)	15
	2.3.6 Soil reaction and Al toxicity	16
	2.3.7 Cation Exchange Capacity (CEC)	17
	2.4 Reaction kinetics	18
	2.5 Soil physical properties and moisture retention	19
	2.6 Organic amendments and soil microorganisms	20
	2.7 Amendment with animal manure and nematode control	22

2.8 Neem extracts on nematode control 25

2.8.1 Active ingredients of neem 26

2.8.2 Degradation of azadirachtin in the environment 31

2.9 Conclusions 33

CHAPTER 3 Materials and methods 34

3.1 Experimental site 34

3.2 Preparation of treatment materials 34

3.2.1 Neem leaves, poultry manure and cow dung 34

3.2.2 Soil 35

3.2.3 Summary for treatments combinations 36

CHAPTER 4 The influence of soil amendment on the breakdown of azadirachtin A and population dynamics of nematode 37

4.1 Introduction 37

4.2 Materials and methods 39

4.2.1 Treatments and experimental design 39

4.2.2 Extraction, counting and identification of nematodes 40

4.2.3 Extraction of azadirachtin and Gas Chromatography (GC) analysis of extracts 41

4.2.4 Measurement of soil moisture and pH 42

4.2.5 Data analysis 42

4.3 Results and discussion 43

4.3.1 Effect of the soil amendment on nematode populations 43

4.3.1.1 Plant parasitic-nematodes 43

4.3.1.2 Non plant-parasitic nematodes 47

4.3.2 Effect of the soil amendment on azadirachtin A breakdown 49

4.3.2.1 GC chromatographs of azadirachtin A 49

4.3.2.2 Azadirachtin A concentration in neem leaves and recovery from soil 52

4.3.2.3 Azadirachtin A degradation in soil 52

4.4 Summary and conclusion 59

CHAPTER 5 Studies into the influence of soil amended with neem leaves and poultry manure on nutrient dynamics 61

5.1 Introduction 61

5.2 Materials and methods 62

5.2.1 Treatments and experimental design 62

5.2.2 Nutrient analyses 63

5.2.2.1 Organic carbon 63

5.2.2.2 Carbon dioxide 63

5.2.2.3 Total nitrogen 63

5.2.2.4 Total phosphorus 64

5.2.2.5 Inorganic NH_4^+- N and NO_3^- - N 64

5.2.2.6 Available phosphorus 65

5.2.2.7 Exchangeable bases 65

5.2.2.8 Exchangeable acidity 66

5.2.2.9 pH 66

5.2.2.10 Gravimetric water content 67

5.2.3 Data analysis 67

5.3 Results and discussion 68

5.3.1 Chemical composition of neem leaves and poultry manure, and the initial C:N ratio of treatments 68

5.3.2 Carbon dynamics 70

5.3.3 Nitrogen dynamics 73

5.3.4 Available phosphorus dynamics 87

5.3.5 Exchangeable potassium, calcium and magnesium 89

5.3.6 Basic Cation Ratio (BCR) of amended soil 92

5.3.7 Soil pH 94

5.3.8 Moisture contents of amendments 96

5.4 Summary and conclusions 98

CHAPTER 6	To test the validity of the soil amendment by investigating its impact on growth characteristics of carrot using pot experiment	99
	6.1. Introduction	99
	6.2. Materials and methods	101
	6.2.1 Treatments and experimental design	101
	6.2.2 Sowing of carrot seeds	102
	6.2.3 Sampling of soil and carrot for analyses	102
	6.3. Results and Discussion	104
	6.3.1. Nutrient status of the soil at harvest of carrot	104
	6.3.2 Nutrient contents of carrot at harvest	105
	6.3.3. Soil amendment and the control of root-knot nematodes of carrot	111
	6.3.4. Yield characteristics of carrot	113
	6.4 Summary and conclusions	117
CHAPTER 7	Summary, conclusions and recommendations	118
	7.1 Summary	118
	7.2. Conclusions	120
	7.3 Recommendations	121
	References	123
	Appendix A	149
	Appendix B	158

LIST OF TABLES

			Page
TABLE	2.1	Total N, P and K in some organic materials	9
	2.2	Nutrient content of neem seed cake and leaves	10
	2.3	Concentration of azadirachtin A in samples of seed kernel, bark, root, leaves and stem parts obtained from Kanthayapalayam, south India	27
	2.4	Efficacy of pure neem constituents to larvae of *Spodoptera litura* and nymphs of *Myzus persicae*	29
	3.1	Some physical and chemical properties of soil at the experimental site	35
	3.2	Treatment combinations	36
	4.1	Table showing treatments used in Chapter 4	39
	4.2	Effect of soil amendment on total plant parasitic nematode populations.	44
	4.3	Effect of soil amendment on non-parasitic nematode populations	48
	4.4	Percentage recovery of azadirachtin A extracted from soil immediately after amendment	53 51
	4.5	Half lives of azadirachtin A in soil amended with neem leaves and animal manure	56
	4.6	Moisture content and pH of amended soil	58

5.1	Table showing treatments used in Chapter 5	62
5.2	Some nutrient constituents of neem leaves and poultry manure	69
5.3	The initial C:N ratio of treatment materials	69
5.4	Net percentage nitrogen and carbon mineralized (at week 12) and rate of mineralization	76
5.5	Changes in C:N ratio of unamended soil and soil amended with neem leaves and poultry manure over the incubation period	86
5.6	Basic cation ratio (BCR) of treatments at the 12th week of incubation	93
6.1	Treatments applied to pots	101
6.2	Nutrient contents and pH of unamended and amended soil at harvest of carrot	106
6.3	Nutrient and ash contents in carrot leaves and roots at harvest	107
6.4	Impact of soil amendment on root-knot index of carrot	112
6.5	Effect of soil amended with neem leaves (NL) and poultry manure (PM) on yield characteristics of carrot	116

LIST OF FIGURES AND PLATES

			Page
FIGURE	4.1	GC Chromatographs of pure azadirachtin A (**A**), unamended soil/soil amended with animal manure only (**B**) and soil amended with neem leaves (**C**)	50
	4.2	Azadiractin degradation in one kilogram of soil amended with 50 g neem leaves	53
	4.3	Azadiractin degradation in one kilogram of soil amended with 100 g neem leaves	54
	5.1	Changes in CO_2 evolution in soil amended with neem leaves and poultry manure	73
	5.2	Organic carbon in soil amended with neem leaves and poultry manure	74
	5.3	Net cumulative CO_2-C mineralized from neem leaves and poultry manure	75
	5.4	Changes in $NH^+_4 - N$ in soil amended with neem leaves and poultry manure	82
	5.5	Changes in $NO^-_3 - N$ in soil amended with neem leaves and poultry manure	83

5.6	Net cumulative N mineralized from neem leaves and poultry manure	84
5.7	Changes in total nitrogen in soil amended with neem leaves and poultry manure	85
5.8	Changes in available phosphorus in soil amended with neem leaves and poultry manure	88
5.9	Changes in exchangeable potassium in soil amended with neem leaves and poultry manure	90
5.10	Changes in exchangeable calcium in soil amended with neem leaves and poultry manure	91
5.11	Changes in exchangeable magnesium in soil amended with neem leaves and poultry manure	92
5.12	Changes in pH of soil amended with neem leaves and poultry manure as affected by incubation period	96
5.13	Influence of incorporated neem leaves and poultry manure on moisture content of soil	97
6.1	Relationships between %N in soil and %N in carrot leaves and roots	109
6.2	Relationships between %K in soil and %K in carrot leaves and roots	109

6.3 Relationships between %P in soil and %P in leaves and roots of carrot 110

PLATES

1 Carrot with root-knots from unamended soil 114

2 Carrot with root-knots from soil amended with 5 g PM/kg soil 114

3 Carrot with root-knots from soil amended with 10 g NL/kg soil 114

4 Carrot with root-knots from soil amended with 10 g NL + 5 g PM/kg soil 114

5 Carrot with few root-knots from soil amended with 30 g NL + 5 g PM/kg soil 115

6 Carrot with no root-knots from soil amended with 50 g NL + 5 g PM/kg soil 115

CHAPTER ONE

1.0 INTRODUCTION

Man depends on agriculture in one way or the other for survival. In addition, many other sectors of the economy, for example, trade, services and the manufacturing sectors depend to a large extent on agriculture. It is mainly through agricultural development that the growing rural population can maintain itself and at the same time contribute to the growth of the national economy. However, efforts by man to boost agricultural production are always hampered by problems such as declining soil fertility, pest and disease infestation.

The decline in soil fertility is a major problem in agricultural production and concerns have been raised about the sustainability of agriculture to feed a world population expected to exceed 7.5 billion by the year 2020, under the present decreasing soil fertility as a result of limited availability of additional land for crop production (Gruhn *et al.*, 2000).

Inorganic and organic sources of fertilizer are normally used by farmers to improve low soil nutrient problems. Farmers, however, find it easier to regulate precisely the amounts of various nutrients added to the soil by using inorganic fertilizers. These are also easier and less time - and labour-intensive to use than organic sources and the yield gains tend to be more immediate than for organics, which take time to decompose and release nutrients (Muir, 2002).

Problems are, however, encountered in the use of inorganic fertilizers.

Muir (2002) has identified a series of interconnected changes occurring as farmers rely more and more on inorganic inputs and less and less on organics; these include, decrease in humus, lowered water holding capacity and more runoff of water, decreased aeration - as the soil loses the structure aided by organic material, decrease in the efficiency of free-living nitrogen fixers in the soil and increase in soil acidity. In addition, the high prices of inorganic fertilizer and transportation costs due to poor infrastructure have prevented most African farmers from the use of these synthetic products (Gruhn *et al.*, 2000).

As an alternative to the use of synthetic fertilizers to off-set the above problems, more attention needs to be given to the use of organic fertilizers which are more environmentally friendly. Organic manures are the primary sources of crop nutrients in many African farming systems (Drechsel and Reck, 1997). The incorporation of organic manures in soil improves the quality of the soil and provides needed nutrients for crop development. When properly handled and applied to the land, organic manure improves the structure of the soil (Kettering, 1992). Good soil fertility and high soil organic matter levels stimulate the growth of beneficial soil organisms, suppress disease-causing soil organisms and help to reduce the severity of pest damage to crops (Kettering, 1992; Dunn, 1994; Donald, 1999). The use of organic manure, therefore, would also help to reduce the usage of synthetic pesticide which leave unwanted residues in food, water and the

environment, affect non-target species and lead to the build-up of pesticide resistance in the target species (FAO, 1989).

The seeds and leaves of neem, a multipurpose plant, have been used as soil manure to provide nutrients to crops (Pandey *et al.*, 1991; Khan and Saxena, 1997; Tilander and Bonzi, 1997). The use of neem cake manure, for example, has been credited with a 19 and 37% increase in yields of paddy rice and cotton respectively, and has proved superior to castor, mahua or cow dung as a fertilizer for sugarcane (CERES, 1980). In addition the neem has proven to better control soil pests, especially soil nematodes than many soil organic manures (Khan and Saxena, 1997; Akhtar, 1999; Chakrabarti, 2000). The neem seed cake and the powder used as soil amendment have also been found to inhibit nitrification, and thus controlling excessive nitrate release in the soil (Neem Foundation, 1997; Lalljee *et al.*, 1999; Deepanjan *et al.*, 2000; Shah and Faheem, 2000).

The availability of neem leaves all year round, and poultry manure which has proved to be a good nutrient provider and a nematicide (Reddy *et al.*, 1993; Abdel Magid *et al.*, 1995; Mondini *et al.*, 1996; Nyakatawa and Reddy, 2000), gives hope to the farmer to solve the dual problem of low levels of soil nutrients and nematode damage, and also control nitrate release in the soil, when the two organic materials are combined in a soil amendment.

Though much research has been done using neem (seeds and leaves) in a soil amendment and in the control of plant diseases, a few grey areas still need to be looked at. The release pattern of nutrients from neem leaves and poultry manure as soil amendment has not been studied and needs attention. There is also lack of information on how the active ingredient (azadirachtin) of the neem degrades in the soil

The general objective of this research therefore was to investigate the dynamics of soil nutrients, the breakdown of azadirachtin A and parasitic nematode populations of a soil amendment consisting of neem leaves and animal manure.

The specific objectives of the research were to study:

i. **a)** the impact of a soil amendment with neem leaves, poultry manure and cow dung on soil nematodes.

   b) the degradation of azadirachtin A in the above amended soil using a Gas Chromatography (GC).

ii. the impact of neem leaves and poultry manure soil amendments on soil nutrient dynamics and

iii. the impact of the amendments on nutrient changes in the soil, root-knots of carrot and the growth of carrot.

The above objectives were based on the hypotheses that neem leaves added alone or in combination with organic manure will not have any significant effect on:

1. the population dynamics of plant and non plant-parasitic nematodes,
2. the breakdown of azadirachtin A, the active ingredient in neem leaves, in the soil,
3. the content of soil nutrients before and after the amendment,
4. the mineralization of organic carbon and nitrogen in the soil, and
5. the characteristics (growth, yield and root-knot infestation) of carrots.

CHAPTER TWO

2.0 LITERATURE REVIEW

The review covers the impact of organic manures on the soil environment with emphasis on soil nutrients and soil micro-organisms especially nematodes. Neem as a manure and the breakdown of its active ingredient, azadirachtin A, in the soil is also reviewed.

2.1. Organic manure and crop production

The application of organic materials into soil improves the physico-chemical properties of the soils and control pests which finally positively affect the performance of crops. Soil amendment with neem products does not only control pests but also provides nutrients to crops and thereby improv es their growth and yield (Pandey *et al.*, 1991; Reddy et al., 1993; Khan and Saxena, 1997; Neem Foundation, 1997).

Field experiments were carried out under centre-pivot irrigation in central Saudi Arabia during the winter to examine the yield and quality responses of wheat cv. Yecora Rojo to application of 0, 4.1, 8.25, 16.5 and 33.0 t chicken manure /ha. Grain yield, grain quality and straw yield were increased with increasing rate of chicken manure. The greatest economic return was given by 8.25 t /ha; application of higher rates were not profitable (Abdel Magid et al., 1995).

A study compared the efficacy of livestock manure, crop residue and fertilizer amendments to a desurfaced soil, cropped with spring wheat. The manures and crop materials were incorporated into the soil at 20 t /ha dry-weight equivalent. The over all best amendments were pig manure, poultry manure and *Medicago sativa* hay. Nitrate-N concentration in the 0 to 60 cm soil depth explained 71% of the variation in restorative ability of the amendments, while extractable P concentration in the 0 to 15 cm depth explained 16% of this variation. Yields from desurfaced plots amended with pig or poultry manure were not significantly different from plots with no top soil removal (Larney and Janzen, 1996).

The effects of different application rates of organic and inorganic fertilizers on soil physical properties and maize production in a severely degraded soil in Nigeria were studied . Poultry manure application significantly improved average maize height and average maize grain yield. The soil organic matter content was highly correlated with yield (r=0.86) (Obi and Ebo, 1995).

Four organic manures and NPK were assessed under field conditions for their comparative effects on tomato growth and yield. Fruit yields were best with swine or poultry manure applied at 10 t/ha. Very high manure application (30 t/ha) depressed growth and yield irrespective of the manure source (Oikeh and Asiegbu, 1993). Like all other crops, organic manure improved the performance of carrots, however, a lumpy soil or one that contains fresh manure led to forked, hairy or deformed roots (McCollum, 1975; Williams *et al.*, 1991).

2.2. Nutrient composition of some manure materials

The nutrient contents of organic manures are far below the inorganic fertilizers (e.g. Urea = 46%N) as could be observed from some examples in Tables 2.1 and 2.2. The application of larger quantities of the organic manure will be needed per hectare as compared to inorganic fertilizers. initially, however, the continuous application of organic manure to the soil will reduce the quantity needed for subsequent applications (WSARE, 1995). Application of organic manure will also improve the soil physically and biologically which the inorganic fertilizers cannot do.

The use of two or more sources of organic manure as soil amendment would be beneficial to ensure a more balanced nutrient source because of the differences in the nutrient content.

Table 2.1. Total N, P and K in some organic materials

Organic source	N	P	K
		(%)	
Poultry manure	1.50 – 3.00	0.50 – 1.50	0.60*
Pig, horse, cow manure	0.30 - 0.60	0.20*	0.50*
Green manure	1.50 – 5.00	0.20 – 0.50	2.00 – 5.00
Compost	0.50 – 2.00	0.20 – 0.50	0.50 – 2.00
Seaweed meal	2.00 – 3.00	------	2.00 – 3.00
Sewage sludge	1.00 – 5.00	0.40 – 2.50	------
Fish waste	4.00 – 10.00	5.00 – 10.00	------
Blood (slaughter house)	10.00 – 12.00	------	------
Human urine/Night soil	1.00 – 1.50	------	------

Sources: Caplan (1992); Nick and Bradley (1994); Hue (1995)

*Kettering (1992)

Table 2.2. Nutrient contents of neem seed cake and leaves

Nutrient (Total) (%)	[1] Neem seed cake	[2] Neem leaves
Nitrogen	3.56	2.10
Phosphorus	0.83	0.10
Potassium	1.67	1.30
Calcium	0.77	1.80
Magnesium	0.75	0.47

Sources: 1 Neem Foundation (1997)

2 Tilander and Bonzi (1997)

2.3. Organic amendments and soil fertility

Addition of organic material in any form helps to maintain the organic matter and the fertility level of the soil. The type of organic material added, however, influences considerably the rate of decomposition as well as the consequent chemical changes brought into the soil (Sarmah and Bordoloi, 1994).

2.3.1. Carbon dynamics

Carbon dioxide – C (CO_2 – C) released in amended soil is relatively proportional to the rate of the organic material applied (Abdel Magid *et al.*, 1993). The CO_2 - C is concomitantly released with nutrient elements and may be monitored as an indication of mineralization rate.

The CO_2 - C released in an aerobic incubation experiment with soils of different organic matter contents has been found to be highest during the first week of incubation and is proportional to the quantity of organic matter added (Sarmah and Bordoloi, 1994). The initial high rate of CO_2 - C evolution has been assigned to the initial higher biological activities (Pocknee and Sumner, 1997). Flushes of C mineralization is said to be the result of rapid decomposition of dead microbial biomass (West *et al.*, 1989). The rate of CO_2 - C release decreases with time as a result of the progressive depletion of labile C (Robertson and Morgan, 1995). The total mass of organic C decreases with time of incubation because of the conversion to CO_2 - C (Tiquia and Tam, 2000).

2.3.2 Nitrogen dynamics

As in the case of carbon, the release of mineral nitrogen was found to be higher for soil amended with higher organic matter (Sarmah and Bordoloi, 1994; Mbagwu *et al.*, 1994). The greatest mineral N released from decomposing legume occurred 2 to 5 weeks after soil incorporation (Sarrantonio and Scott, 1988). Similarly, 59% of total N was released within 2 weeks when legumes were used as soil amendment (Rubaduka *et al.*, 1991).

In other studies, Thonnissen *et al.* (2000) found the peak of N release to occur between 2 and 6 weeks in two study areas and between 5 and 8 weeks in another area when legume green manure was used as soil amendment.

Addition of legume hedgerow prunings to soil significantly increased the total N, NO_3^- - N and NH_4^+ - N (Okeleye and Adetunji, 1999). Hong *et al.*, (2000) also found an increase in the total N when soils were amended with cattle and hog manures.

As the initial total N concentration of amended soils increases, the N mineralization also increases (van Kessel *et al.*, 2000). During composting of poultry manure it was found that the conversion of organic N to NH_4^+ - N (ammonification) was greater under anaerobic stabilization, and the net rate of conversion of NH_4^+ - N to NO_3^- - N (nitrification) was greater under aerobic stabilization (Mahimairaja *et al.*, 1994).

Nitrogen content of organic manure declined by nearly 50% during the first 34 days of decomposition in soil; this large loss of N was thought to be associated with NO_3^- - N leaching (Schomberg and Steiner, 1999). Losses of N in a composting process of chicken litter were attributed mainly to NH_3 (ammonia) volatilization (Tiquia and Tam, 2000). Mahimairaja *et al.*, (1994) in a similar experiment also found the volatilization of NH_3 denitrification – which is a microbial reduction of nitrate and nitrite with the liberation of nitrous oxide (N_2O) and molecular nitrogen (N_2), to account for the loss of N in a composting process.

Nitrification inhibitors have been found to reduce N_2O or NO_2 emissions and NO_3^- - N loss from soils. Nitrogen loss from soils amended with urea coated with neem

(powdered *Azadirachta indica* seeds) and nimin (commercial derivative of neem) were significantly lower than the sole urea application (Deepanjan *et al.*, 2000).

Neem cake was mixed at the rates of 10, 20 and 30 kg/ha with an Oxisol to which $(NH_4)_2SO_4$ was added at 700 kg/ha. The neem cake at all levels depressed nitrification significantly compared to the control (Lalljee *et al.*, 1999). Similar nitrification inhibition properties of neem (*Azadirachta indica*) cakes, bakain (*Melia azedarach*) and arend (*Ricinus communis*) have been reported (Gnanavelrajah and Kumaragamage, 1999; Shah and Faheem, 2000).

2.3.3. C:N ratio

Mineralization and immobilization of N are governed by C:N ratios of the decomposing organic matter. Traditionally, it has been suggested that a C:N ratio greater than 30:1 will immobilize N, a ratio less than 20:1 will mineralize N, and a ratio between 20:1 and 30:1 will produce no net changes in N availability (Tisdale *et al.*, 1993). At a C:N ratio of 12:1, populations of decay bacteria are considered stable (Sullivan, 1999). A number of studies using various crop residues suggest that decomposition dynamics may be too complex to solely rely on the C:N ratio rule of thumb (Chandler *et al.*, 1980; Hatiori and Mukai, 1986; Henry, 1991). Release or demand for N depends not only on the C:N ratio but also on the types of organic compounds in the residue and how long the breakdown of these compounds has been occurring.

2.3.4. Phosphorus dynamics

Mineralized P was found to be higher in soils treated with organic matter. The highest levels were recorded for farmyard manure treatments (Sarmah and Bordoloi, 1994). Mbagwu *et al.*, (1994) found soils treated with increasing rates of organic matter to have a corresponding increasing contents of P. Similar increases in P with the additions of organic matter have been reported in other studies (More, 1994; Okeleye and Adetunji, 1999; Pool *et al.*, 2000; Hong *et al.*, 2000).

It is hypothesized that the application of leucaena, manure 'miombo' litter resulted in immobilization of P. Leucaena which is rich in N but low in P might result in P immobilization (Nyathi and Campbell, 1995).

Many factors influence the total quantity of P mineralized in soil. The total organic P is highly correlated with soil organic C, thus, P mineralization increases with increasing organic C (Tisdale *et al.*, 1993). As the ratio of soil organic C/P increases, P immobilization increases. The C/P ratio of decomposing residues regulates P immobilization or mineralization, as C/N ratio also regulates N mineralization/immobilization (Tisdale *et al.*, 1993).

According to Berg and McClaugherty (1989), P and N which are both important for microbial growth, behave similarly during decomposition.

2.3.5. *Exchangeable Cations (K, Ca, Mg and Na)*

Soil organic amendment with animal or plant sources has brought significant increases in soil nutrients, with K, Ca, Mg and Na being no exception. For example, increasing rates of dehydrated swine waste added to two soil types in Nigeria progressively increased the exchangeable Ca, Mg and K contents of the soils to between 21 to 32 % (Mbagwu *et al.*, 1994). Patiram (1994) also found these nutrient levels in soil to increase to more than 25 % after amending a sandy loam soil with organic matter.

Soil amended with 0, 10, 25, 50 and 75 tonnes/ha of manure compost increased the total K, Ca, Mg and Na to levels of 8, 17 and 10 % respectively in soil according to the rate of compost application (Wong *et al.*, 1999). The incorporation of legume hedgerow prunings of pigeon pea (*Cajanus cajan*) and *Leucaena* in a degraded soil in Ogun State in Nigeria over a period of 60 weeks increased the contents of the total K, Ca and Mg of the soil to over 15 % (Okeleye and Adetunji, 1999). Increases in total K, Ca and Mg of 21, 32 and 17 % respectively were recorded when a soil was amended with 10t/ha poultry manure (Pool *et al.*, 2000). Hong *et al.*(2000) also noted significant increases between 13 and 27 % in exchangeable K, Ca and Mg in a soil amended with animal waste.

2.3.6. *Soil reaction and Al toxicity*

Conflicting accounts of the effect of organic matter on soil pH exist. Organic matter additions to soil have been reported to both increase and decrease soil pH. The application of 10t/ha poultry manure into a Mexican soil increased the soil pH from 4.5 to 5.3 (Pool *et al.*, 2000). Increases of about 25 % in soil pH were also recorded by Hong *et al.* (2000) after incorporating cattle and hog manures into soil. Legume hedgerow prunings added to a degraded soil increased the soil pH from 6 to 7 for the first 30 weeks after incorporation, and thereafter remained fairly constant for the next 12 weeks (Okeleye and Adetunji, 1999). Increase in pH from 5.2 to 5.8 followed by pH stabilization was also observed by Mahimairaja *et al.*, (1994) during composting of poultry manure with different amendments. Manure additions to an acidic soil increased the pH significantly by 20 % (Patiram, 1994). Other examples of pH increases of between 20 and 25 % after incorporation of manure into soil are reported by Ashgar and Kanehiro (1980) and Bessho and Bell (1992).

Decreases in soil pH of between 10 and 28 % were recorded when farm wastes and organic manures were incorporated into a sodic Vertisol in India (More, 1994). The application of chicken manure to soil has been found to also decrease the pH from 7.4 to 5.5 (Kara and Erel, 1999). More examples of pH decreases with manure additions into soils are known (Bevacqua and Mellano, 1994).

It was observed that individual treatments of lime and organic residue (especially chicken manure) decreased the exchangeable Al in the soil, diminishing therefore both the soil acidity and Al saturation percentage. The latter parameter decreased from 72 to 4.2%. The combination of lime and chicken manure led to a decrease of exchangeable Al to undetectable levels (Rojas *et al.*, 2001).

Since addition of organic residues to soils often results in an initial increase in soil pH, a decrease in the concentration of exchangeable Al would be expected to occur. Indeed, several workers have measured an increase in soil pH with a concomitant decrease in exchangeable Al and Al saturation during the decomposition of organic residues in soils (Hoyt and Turner, 1975; Bessho and Bell, 1992; Noble *et al.*, 1996; Wong *et al.*, 1998a, b). Adsorption of Al onto decomposing organic residues would also tend to reduce exchangeable Al levels (Hoyt and Turner, 1975). Research using unamended soils suggests that increases in OM caused by additions of organic residues to soils tends to decrease exchangeable Al through complexation by the newly added or formed OM (Haynes and Mokolobate, 2001).

2.3.7. Cation Exchange Capacity (CEC)

Cation Exchange Capacity (CEC) represents the sites in the soil that can hold positively charged nutrients like calcium (Ca^{2+}), magnesium (Mg^{2+}) and potassium (K^{+}). If CEC is increased, the soil can hold more nutrients and release them for plant growth. To increase CEC, organic matter has to be increased (Griffin, 2002). Humus is an important product of organic matter decomposition. As well as producing a certain quantity of nutrients that are taken up by higher plants, humus

is important for a number of other reasons. The surface area of colloidal humus particles (micelles) is high. This contributes to its high cation exchange capacity (CEC), water holding capacity (WHC) and is important in aggregate formation and stability (Brady, 1990). The CEC of micelles may be 2-30 times higher than for mineral colloids, and this may account for as much as 20-90% of the adsorption of cations by mineral soils (Graves *et al.*, 2001). Organic matter accounts for as much as one third of cation exchange capacity of surface soils and is responsible, perhaps more than any other single factor, for the stability of soil aggregates (Brady, 1990).

2.4. Reaction kinetics

The dynamics of mineralization of nutrients of organic manures in soil have been described with the first-order reaction kinetics (Chae and Tabatabai, 1986; Chescheir *et al.*, 1986) or set of orders of reaction (Gale and Gilmour, 1986). Biomass loss data for soybean and indigofera were found to follow the first-order reaction kinetics (Thonnisen *et al.*, 2000) described for litter decomposition by Wieder and Lang (1982). The first-order reaction equation was used to calculate the mineralization rate constant of dehydrated swine waste in soil amendment (Mbagwu *et al.*, 1994) as was proposed by Gilmour *et al.*, (1977). Carbon mineralization has been found to initially follow the first-order reaction kinetics, while decomposition over long periods of time followed the zero-order/linear reaction kinetics (Seyfried and Rao, 1988). Similarly van Kessel *et al.* (2000) found CO_2-C evolution from soil amended with manure to initially follow the first-order

reaction kinetics, and in some cases the first-order phase was followed by a linear response of the CO_2-C evolution. Abdel Magid *et al.* (1993) found the rate of CO_2 release from decomposing organic material to follow the zero-order reaction kinetics. Cumulative N mineralization has been found to follow a linear course of production (Murwira and Kirchmann, 1991). Nordmeyer and Richter (1985) detected N mineralization in sandy soils to follow the zero order reaction kinetics. Cumulative amounts and the rate of loss of NH_3 during aerobic and anaerobic incubation of poultry manure with different soil amendment levels obeyed the linear/zero-order reaction kinetics (Mahimairaja *et al.*, 1994).

2.5. Soil physical properties and moisture retention

Organic matter loosens the soil, which increases the amount of pore space. This has several important effects. The density of the soil reduces (it becomes less compacted) and the soil structure improves. This means that the sand, silt and clay particles in the soil stick together, forming aggregates or crumbs. Because there is more pore space, the soil is able to hold more water and more air (Griffin, 2002). Manure additions in large amounts tend to lower the surface bulk density of mineral soils because of the addition of low bulk density material and the consequent promotion of soil aggregation (Jones, 2002). The addition of organic material improves soil structure or "workability" immensely and vastly improves the water-holding capacities of sandy soils, a distinct advantage in arid climates (Williams, 2002).

Humus, the least decomposable material from organic matter, increases the water-holding capacity of soil, improves soil aeration and increases infiltration of water into the soil (versus run off of water), because of its porosity (Muir, 2002).

Increases in soil moisture content of upon organic matter application are well known. Mbagwu *et al.* (1994) found increased moisture retention capacities of about 18 % in soils amended with dehydrated swine waste. A degraded Ultisol in southern Nigeria had its moisture retention status improved by 10 % when 10t/ha poultry manure was added to the soil (Obi and Ebo, 1995). The water holding capacity of a soil was improved by 13 % with the addition of poultry manure (El Nadi *et al.*, 1995). Other organic wastes have been found to improve soil physical properties including the moisture status (Hafez, 1974; Khaleel *et al.*, 1981; Sweeten and Mathers, 1985; Mbagwu, 1989).

2.6. Organic amendments and soil microorganisms

The addition of organic manure has been reported to both increase and decrease microbial populations in the soil. Chicken manure was found to harbour a heavy population of pathogenic nematodes and harmful fungal flora, thereby acting as a major source of infestation (Grewal *et al.*, 1989).

Two types of organic amendments were used at the rate of 4 t/ha: decomposed chicken manure and compost made of sugarcane bagasse, sawdust and ashes at

2:1:1 to control foot rot in sweet pepper caused by *Phytophthora capsici.* Disease incidence was reduced to 30% and 65% (67% and 26% in control) by chicken manure and compost, respectively, when they were incorporated into the plant bed (Corrales *et al.*, 1990).

Two field experiments were conducted in the San Joaquin Valley, California, USA, during 1991-92 to determine the effects of commercial chicken compost, ammonium phosphate fertilizer and solarization, alone or combined, on several soilborne pathogens and the growth and yield of lettuce. *Pythium ultimum* was controlled by solarization alone and in combination with chicken compost (Gamliel and Stapleton, 1993).

The incidence of Sclerotinia soft rot of lettuce and survival rate of sclerotia of *S. sclerotiorum* were reduced by the addition of organic amendments to field plots at the Frankston Research Station, Seaford, Victoria, Australia. Of the 6 materials

tested, stable manure, fowl manure and lucerne hay were the best, and all except brown coal significantly reduced the disease compared with the control (Asirifi *et al.*, 1994).

During 1989-92 the biological control of root-rot of mandarins caused by *Phytophthora nicotianae* var. parasitica and *Phytophthora colocasiae* was studied. *Trichoderma harzianum*, *T. viride* and *Gliocladium virens* were widely distributed

in Citrus orchards in Kodagu, Karnataka, India. Seventeen isolates were highly antagonistic to both *Phytophthora* spp. in vitro. For large-scale fungal multiplication, local waste (coffee-cherry husk, fruit skin and berry mucilage, poultry manure and mushroom-grown waste) was a suitable substrate with 20-30 million Colony Forming Units per gram (c.f.u/g). Pot trials amended with coffee-cherry husk and poultry manure in a 1:2 ratio decreased feeder root-rot and increased seedling growth (Sawant *et al.*, 1995).

Wheat straw or poultry pinus-sawdust litter added to *Pinus pinaster* forest acid soil in Spain increased soil microbial populations, with the poultry litter addition being more marked than the straw addition, the increase in numbers followed the order, bacteria > fungi > actinomycetes (Acea and Carballas, 1996).

2.7. Amendment with animal manure and nematode control

Soil incorporation of neem controls nematodes as has been mentioned in section '2.4'. Other soil amendments like poultry litter also suppress nematodes development. A significant reduction in numbers of galls, egg masses and nematode populations was recorded when poultry manure and mustard cake were applied at 1 or 2 t/ha to control *Meloidogyne incognita* on *Vigna radiata* in India (Borah and Phukan, 1992).

Organic amendments of pressmud (25 t/ha), farmyard manure (20 t/ha), poultry manure (20 t/ha), coconut fibre (25 t/ha) and sugarcane bagasse (25 t/ha) applied to sugarcane in the field in India controlled plant parasitic nematodes to various

degrees (Jonathan *et al.*, 1991). The application of silkworm faeces, compost or cattle manure to soil heavily infested with *Meloidogyne javanica* and planted with *Vigna radiata* gave 41, 59 and 83% reduction in nematode numbers respectively (Toida *et al.*, 1991).

The effects of chicken litter on *Meloidogyne arenaria* in tomato plants cv Rutgers seedlings were determined in greenhouse. After 10 days of inoculation of the nematodes, the total number of nematodes in the roots decreased with increasing rates of chicken litter. After 46 days, egg numbers also decreased with increasing litter rates (Kaplan and Noe, 1993).

The efficacy of nematicides, phorate or carbofuran at 1 kg a.i./ha, and organic soil amendments, oil seed cakes of mustard, *Azadirachta indica* or *Ricinus communis* and poultry manure on nematode population and growth characteristics of *Curcuma longa* L. was assessed. All the treatments reduced the nematode population and amount of damage to the crop (Haidar *et al.*, 1998).

Though organic amendment has registered reduction in nematode population, field studies in Warsaw, Poland , resulted in increased densities of native population of *Steinernema feltiae* (Bednarek and Gaugler, 1997).

Studies of the effects of poultry manure, cattle manure, horse manure, fruit canning factory waste and burnt township refuse on some plant parasitic nematodes

affecting tomatoes were carried out at Ibadan, Nigeria. Root gall rating at the end of the trial was significantly reduced by all the organic manures. Population of all nematodes fell immediately after application of soil amendment but increased gradually thereafter (Babatola, 1989).

Five organic manures: cattle manure, poultry manure, horse manure, burnt township refuse and citrus wastes were applied at rates 10-20 t/ha to soil of known nematodes (*Meloidogyne incognita, Pratylenchus brachyurus, Helicotylenchus* spp *and* 10 *Xiphinema* spp) infestation. Burnt township refuse, poultry and cattle manures significantly increased leaf yield of *Celosia argentea* and reduced nematode infestation (Babatola and Oyedunmade, 1992).

Three applications of half, normal and supernormal doses of nitrogen, 50, 100 and 200 kg/ha respectively in 8 organic amendments (groundnut, cotton seed and soybean oil cakes, poultry manure, sheep manure, cowdung, raw sewage sludge and cassava peelings) were tested for their effect on the development of *Pratylenchus brachyurus* and on the growth and yield of okra in a greenhouse at 30-33 °C. All the amendments brought about significant decrease in nematode developmental rate and corresponding increase in plant growth and yield (Khan, 1994). McSorley and Frederick (1999) studied the population densities of nematodes in field soil without plants for ten months following the application of organic amendments to pots in a greenhouse. Treatments consisted of homogenous crop residues of maize (*Zea mays*), texas panicum (*Panicum texanum*), velvet bean

(*Mucuna pruriens*) and control without any amendment. Plant-parasitic nematodes declined in all the treatments due to the absence of a food source. However, the populations of bacterivore and fungivore nematodes increased, and were higher than the initial nematodal populations.

2.8. Neem extracts on nematode control

Chopped leaves of neem 50 g/kg infested soil evaluated on papaya under greenhouse conditions significantly reduced root galling (Reddy *et al.*, 1993).

Soil amendment with neem cake controlled nematode populations of sugarcane and green gram in separate experiments in India (Jonathan et al., 1991; Pandey *et al.*, 1991). The application and incorporation of neem cake in soil management of root-knot nematode (*Meloidogyne javanica)* significantly reduced root galling on tomato (Khan and Saxena, 1997). The populations of plant parasitic nematodes, *Meloidogyne incognita, Rotylenchulus reniformis, Tylenchorhynchus brassicae* and *Helicotylenchus indicus* on mung bean (*Phaseolus aureus)* and chickpea were significantly reduced with the application of neem cake (Tiyagi and Alam, 1995).

Neem cake at 5 g/kg of nursery soil mixture and 1 kg/plant in the main field proved effective as treatment against *Meloidogyne incognita* in papaya (Ramakrishnan and Rajendran, 1998). The impact of neem oil cake at 2 tonnes/ha/year on the control of root-knot nematode disease of mulberry was studied on 10 farmers' fields in India; the disease severity was reduced by 66.7 –

73.0% and leaf yield was increased by 18.8% over the control plots (Sharma *et al.*, 1998).

Seed coating of okra with neem based products (Achook, neem cake and powdered leaf extracts) and *Melia azedarach* powdered leaf, each at 100% (w/w), 50% (w/w) or 25% (w/w), were tested against *Meloidogyne incognita* in microplots. Neem cake at 100%, Achook at 50% and neem powdered leaf at 100% were effective against the nematodes (Deka and Rahman, 1998).

2.8.1. Active ingredients of neem

In nature, plant chemical defense against pests does not depend on a single compound, but instead on mixtures of compounds. Neem protects itself from multitudes of pests with multitudes of pesticidal ingredients belonging to a class called triterpenes or limonoids (BOSTID, 1992). New limonoids are still being discovered in neem but azadirachtin, salannin, meliantriol and nimbin are the best known, with azadirachtin proving to be the tree's main agent for battling pests (BOSTID, 1992).

The concentration of azadirachtin varies in different parts of the neem plant. Greater amount of the compound is found in the neem seed with very smaller amounts in the leaves, bark, root and stem of the plant (Table 2.3).

Table 2.3. Concentration of azadirachtin A in samples of seed kernel, bark, root, leaves and stem parts obtained from Kanthayapalayam, south India

Plant parts	Moisture content (% w/w)	Azadirachtin conc. (mg/100g wt with moisture)
Seed kernel	25	24.85
Bark	17	0.42
Leaves	35	0.59
Root	15	0.24
Stem	20	0.15

Source: Sundaram (1996)

Azadirachtin and azadirachtin-containing neem seed extracts act as antifeedants, growth regulators and sterilants in insects (Schmutterer, 1988). The antifeedant effect of azadirachtin at a concentration of 0.4% on *Nodostoma pubicolle*, a pest of pear, decreased the feeding of the beetle (Durairaj *et al.*, 1991). Application of various azadirachtin doses (0.5 to 10 µg/larva) in methanol to the last instar spinning stage larvae of rice moth (*Corcyra cephalonica)* inhibited the development of the pest (Sharma, 1992). Azadirachtin (0.3%) had strong ovicidal effect on eggs of *Helicoverpa armigera*, the potency was as strong as the synthetic pesticide, endosulfan (Patel and Patel, 1997).

Koul *et al.* (1996) studied the effect of neem allelochemicals, azadirachtin, salannin, nimbin and nimbinene on nutritional physiology of larval *Spodoptera*

litura. Nutritional analyses revealed strong antifeedant and growth regulatory effects of azadirachtin which were independent of each other. While salannin and nimbinene induced concentration dependent feeding deterrence only nimbin was inactive at the 10 ppm level. Salannin and nimbin did not interfere with the trypsin activity of the gut. These results and those from nutritional studies suggest that salannin and nimbinene have no toxicity mediated effects on *Spodoptera litura* larvae, and antifeedant activity is a result of the effects on deterrent and other chemoreceptors.

The efficacy of pure neem constituents on the larvae of *Spodoptera litura* and nymphs of *Myzus persicae* was carried out by Isman *et al.*, (1996) (Table 2.4).

From Table 2.4, in *S. litura*, the dietary EC_{50} value for salannin and nimbin are 75 and 120 times respectively higher than that for azadirachtin. For the 2nd instar *Myzus persicae*, the LC_{50} values for salannin and nimbin are 295 and 385 times respectively higher than that for azadirachtin. At least in these two species, azadirachtin is almost several times more effective than the other major limonoids from neem. Isman *et al.*, (1996) found that larvae of *S. litura* can rapidly habituate (i.e. become desensitized) to the antifeedant effects of azadirachtin upon repeated or continuous exposure. In contrast, when exposed to neem producing the same concentration of azadirachtin, habituation does not take place and the caterpillars remain continuously sensitive to the antifeedant action, suggesting that the other limonoids have important consequences for the practical use of neem for insect control

Table 2.4. Efficacy of pure neem constituents on larvae of *Spodoptera litura* and nymphs of *Myzus persicae*

Compound	*S. litura* larval growth dietary EC_{50} (ppm) (day 10)	*M. persicae* mortality LC_{50} (m insolution) (day 6)
Azadirachtin	0.21	1.3
3-Tigloyl-azadirachtol	0.29	-
Salannin	15.7	383
Deacetyl-salannin	>25	-
Nimbin	>25	>500
Deacetyl-nimbin	>25	>500

Source: Isman *et al.*, (1996

The importance of other limonoids in pest control is also revealed in an experiment carried out by Stark and Walter (1995). They performed series of toxicity studies with *Acyrthosiphon pisum* and the neem extract formulations Margosan-O (MO), MO devoid of neem oil, Azatin, RH-9999, Azatin with 5% neem oil, RH-9999 with 5% neem oil and neem oil (5%). It was found that the addition of neem oil from MO increased the efficacy of neem insecticides that did not contain the oil, while removal of neem oil from MO reduced its efficacy by 62%. Six limonoids

(nimbandiol, deacetylnimbin, 6-acetylnimbandiol, deacetylsalannin, nimbon, and salannin) and two unidentified chemicals believed to be limonoids, were identified in neem oil. It can be concluded that neem oil with these limonoids play a role in increasing the efficacy of neem insecticides.

According to BOSTID (1992) compounds in neem especially, azadirachtin, do not kill insects - at least not immediately. Instead they both repel and disrupt their growth and reproduction. Research over the past 20 years has shown that azadirachtin is one of the most potent growth regulators and feeding deterrents ever assayed. It will repel or reduce the feeding of many species of pest insects as well as some nematodes. In fact, it is so potent that a mere trace of its presence prevents some insects from even touching plants. Azadirachtin is structurally similar to insect hormones called "ecdysones," which control the process of metamorphosis as the insects pass from larva to pupa to adult. It affects the corpus cardiacum, an organ similar to the human pituitary, which controls the secretion of hormones. Metamorphosis requires the careful synchrony of many hormones and other physiological changes to be successful, and azadirachtin seems to be an "ecdysone blocker." It blocks the insect's production and release of these vital hormones. Insects then will not molt. This of course breaks their life cycle.

2.8.2. Degradation of azadirachtin in the environment

With the exception of azadirachtin, no information has been found on the degradation of the other limonoids of neem. Schmutterer (1988) stated that under

tropical field conditions, the residual action of neem averages about five days. Results from a study of neem seed extracts applied to beets to control cotton leaf worm, *Spodoptera littoralis*, indicate that leaves taken from neem-sprayed plants retained full antifeedant and growth-regulating activities against the leaf worm for three days, and partial activities for five days after application (Meisner *et al.*, 1983).

Azadirachtin degrades in the presence of ultraviolet light (including sunlight), with half-life (pure compound on glass surface or in solution) of approximately 24 hours (Barnby *et al.*, 1989). However, degradation of azadirachtin in neem oil under solar simulating conditions may be considerably delayed (Isman *et al.*, 1991).

Azadirachtin hydrolysed readily at 35 ^{0}C. and its disappearance followed simple pseudo-first-order kinetics; the rate constants ranged from 2.48 x 10^{-3} to 67.7 x 10^{-3}/h and were faster in basic than in acidic media (Szeto and Wan, 1996). Formulated azadirachtin A degradation in gellan gum-based minimal medium and vermiculite system followed peudo-first-order kinetics with half-lives of 44.4 and 13.2 to 46.2 days respectively (Wan *et al.*, 1997).

The initial concentrations of AZ-A found on fir and oak foliage in Canada when Margosan-O formulation was applied at 7, 15 and 40 g ai/ha were 4.3, 12.8 and 28.8 µg /g (fresh weight) respectively in fir needles, and 10.5, 31.4 and 96.2 µg /g (fresh weight) respectively in oak foliage. In spite of the vast differences in

geometry and initial concentrations, the DT_{50} values for all the samples were nearly the same and ranged from 17 to 22 hours, and the rate constants ranged from 0.0311 to 0.0414, the values were not markedly different (Sundaram, 1996).

Sundaram (1996) studied the degradation of AZ-A in nursery soil (autoclaved and non autoclaved); the DT_{50} values (in days) for the autoclaved and non autoclaved soils under the experimental conditions used were 37.65 and 25.77 respectively. The lower DT_{50} value obtained for the non autoclaved soil indicated the role of microbes in the degradation of AZ-A. Autoclaving destroyed the soil microbes responsible partially for the degradation of AZ-A. In a similar experiment in the United States, Stark and Walter (1995) studied the effects of temperature (15 and 25 ^{O}C) and microbial activity on the persistence of AZ-A and B in soil. The DT_{50} for AZ-A was 43.9 and 19.8 days for non autoclaved soil kept at 15 and 25 ^{O}C respectively, and that for AZ-B was 59.2 and 20.8 days for non autoclaved soil kept at 15 and 25 ^{O}C, respectively. Microbial activity was also responsible for faster degradation because DT_{50}s for autoclaved soil were much longer than for non autoclaved soil. The DT_{50}s for AZ-A in autoclaved soil were 91.2 (15 ^{O}C) and 31.5 days (25^{O}C). DT_{50}s for AZ-B in autoclaved soil were 115.5 (15 ^{O}C) and 42.3 days (25 ^{O}C).

2.9. Conclusions

Though much research has been done on neem as soil amendment in the area of soil nutrition and protection, the literature is silent on:

1. the release pattern(s) of nutrients when neem leaves and poultry manure have been used in a soil amendment and
2. the degradation in soil of the active ingredient (azadirachtin) in neem.

CHAPTER THREE

3.0 MATERIALS AND METHODS

This section presents materials and methods that were common for the three experiments conducted in this study. Detailed descriptions of the experiments are presented in subsequent chapters.

3.1. Experimental site

The experiment was conducted at the Technology Village of the University of Cape Coast (5.07 ^{O}N, 1.14 ^{O}W). The area has a bimodal pattern of rainfall distribution, with an annual rainfall ranging between 900 and 1000 mm. The main rainy season is from April to July with a peak in June, and a minor rainy season from September to November with a peak in October. Mean annual minimum temperature is about 23.3 ^{O}C and the maximum about 30 ^{O}C (Meteorological Station, Cape Coast, 1989). The site is used for conducting field research on vegetables and other crops.

3.2. Preparations of treatment materials

3.2.1. Neem leaves, poultry manure and cow dung

Neem leaves were harvested around the experimental site, air dried for one week (check Table 5.2 for moisture content) and ground to pass through 2 mm mesh. Three months old poultry manure packed in fiber bags and fresh cow dung were collected from the University of Cape Coast Research Farm, air dried for three weeks and also sieved through a 2 mm mesh.

3.2.2. Soil

The soils at the experimental site were developed over Sekondian rocks, which are mainly sandstone, shales and conglomerates. The soils, which are at 30 – 35 m above sea level and occur on sloping to gently undulating land, consist of the Edina, Atabadzi, Benya and Udu association. The soils are highly weathered, leached of their bases and are therefore acidic. They are dominated by low activity clays (Asamoa, 1973). Samples were taken from the Benya Series, classified as a Haplic Acrisol (FAO/UNESCO, 1988) at a depth of 0 – 15 cm and bulked. The soil was sieved through a 2 mm mesh immediately after sampling.

Table 3.1. Some physical and chemical properties of soil at the experimental site

Property		Value
pH		4.6
Organic carbon (%)		0.85
Total N (%)		0.06
Total K (%)		0.06
Total P (%)		0.04
NO_3^--N (mg N/kg)		18.17
NH_4^+ - N (mg N/kg)		5.79
Available P (mg/kg)		7.12
ECEC ($cmol_c$/kg)		2.42
Exch. Acidity ($cmol_c$/kg)		0.20
Sand (%)	Sandy Loam	63.30
Silt (%)		20.50
Clay (%)		16.20

3.3. Summary of the treatment combinations

Table 3.2. Treatment combinations

CHAPTER 4 Treatment (per kg soil)	CHAPTER 5 Treatment (per kg soil)	CHAPTER 6 Treatment (per kg soil)
Control (No amendment)	Control (No amendment)	Control (No amendment)
10 g C	5 g PM	5 g PM
5 g PM	10 g NL	10 g NL
10 g PM	10 g NL + 5 g PM	10 g NL + 5 g PM
50 g NL	20 g NL + 5 g PM	30 g NL + 5 g PM
100 g NL	30 g NL + 5 g PM	50 g NL + 5 g PM
50 g NL + 10 g C	40 g NL + 5 g PM	
100 g NL + 10 g C	50 g NL + 5 g PM	
50 g NL + 5 g PM		
50 g NL + 10 g PM		
100 g NL + 5 g PM		
100 g NL + 10 g PM		

C = Cow dung

PM = Poultry manure

NL = Neem leaves

The details about how each treatment was prepared can be found in the respective chapters.

CHAPTER 4

4.0 The influence of soil amendment on the breakdown of azadirachtin A and population dynamics of nematodes

4.1. Introduction

Synthetic pesticides have been used to control soil pests leading to positive gains in agricultural production (Johnston *et al.*, 1995; Sharma and Sharma, 1995; Sultan *et al.*, 1995), however, this has negative implication on the environment as mentioned in Chapter 1.

Generally in developing countries, people suffer from short term exposure (including that resulting from suicide), and chronic effect of long term exposure to synthetic pesticides (W H O, 1990; Fening, 1999). These negative effects of synthetic pesticides on the environment have led to finding alternative means of pest control (Powers *et al.*, 1993; Johnson *et al.*, 1995; Kerry and Bourne, 1996: Sarathchandra *et al.*, 1996). Aside the application of any kind of pesticide to control pests, good soil fertility and high soil organic matter levels help to reduce the severity of pest damage to crops (Nellum, 1985; Dunn, 1994; Donald, 1999).

The products of the neem tree, *Azadirachta indica* when used as soil amendment have been found to enrich and control parasitic soil nematodes (Chapter 1). The neem leaves, for example, which are found all year round have been used as soil amendment to effectively control root-knot nematodes of papaya and to improve the growth characteristics of the plant at a rate of 50 g neem leaves/ kg soil under glasshouse conditions (Reddy *et al.*, 1993).

The neem's chemical broadside is a mixture of 3 or 4 related compounds or limonoids, and it backs these up with 20 or more others that are minor but nonetheless active; and of these limonoids, azadirachtin has proved to be the main agent for battling pests (BOSTID, 1992). Azadirachtin A and B account for about 99% of the array of azadirachtins in the neem plant with the azadirachtin A being the most dominant compound (Isman *et al.*, 1996).

The present study was to investigate the effect of soil amended with neem leaves, poultry manure and cow dung manure on the populations of plant and non plant-parasitic nematodes. Poultry manure and cow dung have been reported to have nematicidal properties (Babatola, 1989; Babatola and Oyedunmade, 1992; Ali, 1995; Nahar *et al.*, 1996).

In the quest for controlling soil pests, the study of the behaviour of azadirachtin A as it breaks down in the soil is imperative. Studies of the degradation of azadirachtin A in the soil using High Performance Liquid Chromatography (HPLC) have been carried out under controlled laboratory conditions (Stark and Walter, 1995) and in the greenhouse (Sundaram, 1996), using different neem formulations as the source of the active ingredient. In addition to the nematodal study, therefore, the breakdown of azadirachtin A in the amended soil was also assessed, using Gas Chromatography (GC).

4.2. Materials and methods

4.2.1. Treatments and experimental design

Treatments consisted of soil + neem leaves , soil + cow dung , soil + poultry manure and combinations of these in different proportions as listed below (Table 4.1). Soil sample weight of 10 kg was used as base for the preparation of each treatment.

Table 4.1. Table showing treatments used in Chapter 4

Treatment (per kg soil)
No amendment (Control)
10 g C
5 g PM
10 g PM
50 g NL
100 g NL
50 g NL + 10 g C
100 g NL + 10 g C
50 g NL + 5 g PM
50 g NL + 10 g PM
100 g NL + 5 g PM
100 g NL + 10 g PM

C= Cow dung
PM= Poultry Manure
NL= Neem Leaf

4.2. Materials and methods

4.2.1. Treatments and experimental design

Treatments consisted of soil + neem leaves , soil + cow dung , soil + poultry manure and combinations of these in different proportions as listed below (Table 4.1). Soil sample weight of 10 kg was used as base for the preparation of each treatment.

Table 4.1. Table showing treatments used in Chapter 4

Treatment (per kg soil)
No amendment (Control)
10 g C
5 g PM
10 g PM
50 g NL
100 g NL
50 g NL + 10 g C
100 g NL + 10 g C
50 g NL + 5 g PM
50 g NL + 10 g PM
100 g NL + 5 g PM
100 g NL + 10 g PM

C= Cow dung
PM= Poultry Manure
NL= Neem Leaf

The treatments mixture were transferred into polythene bags of 4.8 cm diameter and 15 cm long leaving one centimeter of space at the top. Perforations were made at the base of the bags. There were three replications. A replicate represented a batch of 10 bags, each weighing 300 g of the treatment. Batches of replicates were placed in the soil on the 16th of September, 2002 using the Completely Randomized Design (CRD). The polythene bags were placed in the soil to a depth of 14 cm. There was enough rainfall to keep the treatments moist throughout the experiment.

Sampling was immediately done for nematode and azadirachtin A extraction. Subsequent sampling followed 14, 28, 42, 56, 70 and 84 days after placement of bags in the field. The polythene bags were collected one at a time as representative samples for replicates during sampling. The contents of the polythene bags were well shaken to ensure thorough mixing.

4.2.2. Extraction, Counting and Identification of nematodes

This part of the experiment was conducted at the Department of Nematology – CSIR, Fumesua, Kumasi.

A modified Baermann's method (Dropkin, 1989) for nematodes extraction was used. Nematodes in 100g soil samples were determined by migration through a double-ply tissue sheet into water for 24 hours. A subsequent direct counting of nematodes in the extracts was made using a stereomicroscope and x40

magnification (Opperman *et al.*, 1993). External morphological features were used to identify the nematodes (Mai *et al.*, 1968; Willmott *et al.*, 1977).

4.2.3. Extraction of Azadirachtin and Gas Chromatography (GC) analysis of extracts

This part of the experiment was conducted at the Chemistry Department of the Kwame Nkrumah University of Science and Technology, Kumasi Ghana.

Amended soil samples were air dried for 48 hours. Fifty grams (50 g) of each soil sample was weighed and defatted with ether, and azadirachtin extracted with 200 ml methanol for 6hours using a soxhlet apparatus. Extracts were filtered with Whatman No. 1 filter paper and reduced to just dryness using a rotary evaporator at 20 °C. Reconstitution of extracts was made with 10 ml of GC grade methanol, followed by the quantification of azadirachtin A with the GC.

A Perkin Elmer Gas Chromatograph model 1022 Plus, equipped with Flame Ionic Detector (FID) was operated at an oven, injection and detector temperatures of 180, 255 and 26 °C respectively. The column, 0-17, was used for the separation of the azadirachtin. A flow rate of 15 ml per minute was operated for the carrier gas (nitrogen), with sample injection volume of 5 µl. Pure analytical grade of azadirachtin A was used as the standard to identify and quantify the unknown azadirachtin A in the samples. The azadirachtin concentration in the neem leaves before soil amendment was also determined through the same processes of extraction and analysis.

4.2.4. Measurement of soil moisture and pH

Ten grams of samples placed in 100 ml beakers were oven dried overnight a 105 ^{0}C. After this period the samples were cooled in a desicator and reweighed. The moisture content of the samples were then calculated through the difference of the two readings (Rowell, 1994).

Samples of 10 g were weighed into 50 ml centrifuge tubes, 25 ml of distilled water was added to each sample. Tubes were capped and shaken by hand for 15 minutes, and the pH measured with a pH meter (Rowell, 1994).

4.2.5. Data Analysis

The half lives (time required for 50% loss) of azadirachtin in the samples were calculated using the following equation (Stark and Walter, 1995):

$$t_{\frac{1}{2}} = \frac{0.6931}{k}$$

where $t_{\frac{1}{2}}$ = half life

k = rate constant/slope obtained from the plot of natural log azadirachtin A concentration against incubation period.

The nematode numbers, half lives, pH and moisture content of samples were compared statistically using the MSTATC statistical software package (Russell, 1990). Minitab version 11.21 was used for the Fitted Line Plots of Figures 4.2 and 4.3. The ANOVA tables for all the statistical calculations are found in Appendix B

4.3. Results and discussion

4.3.1. Effect of the soil amendments on nematode populations

4.3.1.1. Plant Parasitic Nematodes

Plant parasitic nematode numbers for the soil treatments under the sampling dates are presented in Table 4.2. The total numbers of plant-parasitic nematodes decreased with time in all the treatments. The individual parasitic nematodes (*Meloidogyne, Scutellonema, Pratylenchus, Paratricodorus, Tricodorus, Helicotylenchus, Criconemella* and *Rotylenchus)* also followed similar decreasing pattern (Appendices A1 – A8). The amended soils had significantly lower numbers of the nematodes than the unamended soil throughout the study period. Similar trends showing nematode numbers reducing after application of organic soil amendments have been reported by other workers (Muller and Gooch, 1982; Rodriguez-Kabana, 1986; Babatola, 1989; Stirling, 1991; McSorley and Frederick, 1999).

Among the amendments, neem and its combinations registered zero nematode numbers immediately after the application of the organic matter. Some nematodes, however, were detected during the second sampling two weeks later (Sept. 30). A greater proportion of the nematodes might have been destroyed by the neem compounds as nematode numbers at that time were significantly lower than the

Table 4.2. Effect of soil amendment on total plant parasitic nematode populations.

Amendment (per kg of soil)	Nematodes per 100 g of soil							
	Sep. 16	Sep. 30	Oct. 14	Oct. 28	Nov. 11	Nov. 25	Dec. 9	Mean
No amendment	219 a	226 a	159 a	88 a	85 a	63 a	63 a	129 a
10 g C	132 b	102 b	99 b	53 b	41 b	33 b	28 b	70 b
5 g PM	107 c	88 c	68 c	44 c	40 b	29 b	16 c	56 c
10 g PM	90 d	68 d	62 d	25 d	31 c	24 c	9 d	44 d
50 g NL	0 e	27 f	28 f	20 e	10 fg	11 d	4 f	14 ef
100 g NL	0 e	20 g	21 g	4 h	2 i	4 efg	0 g	7 gh
50 g NL + 10 g C	0 e	30 f	36 e	21 e	11 ef	4 efg	8 d	16 e
100 g NL + 10 g C	0 e	22 g	12 h	8 g	12 e	5 ef	1 g	9 gh
50 g NL + 5 g PM	0 e	30 f	24 fg	13 f	1 i	8 de	4 f	11 fg
50 g NL + 10 g PM	0 e	35 e	22 g	14 f	18 d	4 efg	6 e	14 ef
100 g NL + 5 g PM	0 e	19 g	22 g	11 fg	9 g	0 g	1 g	9 gh
100 g NL + 10 g PM	0 e	13 h	14 h	1 h	4 h	2 fg	0 g	5 gh

Means followed by the same letter in each column do not differ significantly ($P \leq 0.05$)
C = Cow dung
PM = Poultry Manure
NL= Neem Leaf

ANOVA Tables – Appendices B1-B8 (pp 158 – 160)

poultry manure and cow dung amendments and the control. Nematode numbers for the rest of the sampling dates, especially the last sampling day and the mean numbers of nematodes in the soil amended with neem were fewer than the other treatments. Virtually no recovery of nematodes was recorded for the neem amended soils throughout the study for *Helicotylenchus, Criconemella* and *Rotylenchus* (Appendices A6 – A8). The inclusion of neem seemed to have adversely affected these nematodes, however, these nematodes were very low in numbers even in the unamended soil as compared to the others especially *Meloidogyne* which is known to be abundant in the tropics (Dropkin, 1989).

Poultry manure at 10 g / kg soil had significantly lower nematode numbers than the 10 g cow dung and 5 g poultry manure (Table 4.2). The higher rate of the poultry manure was significantly more effective in the nematode control than the lower level and the cow dung. Both the poultry manure and the cow dung have been found to be effective in controlling parasitic nematodes, with the effectiveness increasing with increasing levels of manure and also the kind of manure applied (Toida *et al.*, 1991; Babatola and Oyedunmade, 1992; Kaplan and Noe, 1993; Riegel *et al.*, 1996).

The suppressive effects of organic amendments on plant-parasitic nematodes have been attributed to the enhancement of soil microbial populations by the amendments and the accompanied chemical by-products, which may have antagonistic effect on the parasites (Rodriguez-Kabana, 1986; Dunn, 1994).

At the dosage levels applied, the neem leaves and its combinations proved to be better than the sole poultry manure and the cow dung in controlling the plant-parasitic nematodes. The neem leaves apparently possess higher nematicidal potency than the poultry manure and the cow dung. No significant differences were noticed among the treatments containing neem amendments. The high quantities of the neem leaves might have overshadowed any clear changes that might have been brought by the mixture of the poultry manure and the cow dung.

Plant-parasitic nematodes were found to decline in all the treatments, this was attributed to the absence of a food source for the parasites, as also observed by McSorley and Frederick (1999). However, the populations of bacterivore and fungivore, non-parasitic nematodes increased, and were higher than the initial nematodal populations, as could be observed in the next section.

Though plant parasitic nematode populations tend to decline under fallow conditions, amending the fallow soils with organic matter especially neem leaves might prove better in drastically controlling the parasites.

4.3.1.2. Non plant-parasitic nematodes

Table 4.3 shows the population of non-parasitic nematodes for the different dates of sampling under the various soil treatments.

The application of the amendments reduced the numbers of the non plant-parasitic nematodes initially (zero day of incubation), as was the case for the parasitic plant nematodes at the same day of incubation (Table 4.2). The neem leaves amendments were the most suppressive. However, the nematode numbers in the amended soils rose to levels significantly higher than the unamended soil which was the opposite for the plant parasitic nematodes (Table 4.2 and Appendices A1 - A8). For the subsequent sampling dates the numbers of the non plant-parasitic nematodes continued to decline in the unamended soil whilst a rise in nematodes numbers was the case for the amended soils (Table 4.3). Number of nematodes 70 days after incubation and also the mean number of nematodes of the amended soils were significantly higher than the unamended soil. The higher dose of the neem leaves (100 g/kg soil) and their combinations recorded the highest nematodes populations. McSorley and Frederick (1999) in a similar research also found the application of different organic materials to the soil to increase the populations of non plant-parasitic nematodes as bacterivores, fungivores and omnivores.

The probable increase in numbers of nematodes antagonistic to the plant-parasitic nematodes might have contributed to the suppressive action of the amendments against the plant-parasitic nematodes (Akhtar and Malik, 2000).

Probable slight changes of conditions in treatments and experimental error might have accounted for the inconsistencies in nematode numbers measured.

Table 4.3. Effect of soil amendment on non-parasitic nematode populations.

Amendment (per kg of soil)	Nematodes per 100g of soil							
	Sep. 16	Sep. 30	Oct. 14	Oct. 28	Nov. 11	Nov. 25	Dec. 9	Mean
No amendment	113 a	72 h	88 h	78 k	60 k	55 j	26 l	70 j
10 g C	101 b	220 g	80 h	208 j	25 l	370 i	250 j	180 i
5 g PM	96 bc	790 d	320 g	352 i	150 j	344 i	300 i	336 h
10 g PM	93 c	1020 bc	340 g	744 e	200 i	1120 e	195 k	530 g
50 g NL	2 d	990 c	670 f	472 g	356 h	960 g	830 h	611 f
100 g NL	4 d	1104 b	1300 d	2152 b	1636 b	1480 d	2436 b	1444 b
50 g NL + 10 g C	2 d	656 e	2300 b	364 i	852 g	556 h	1028 g	822 d
100 g NL + 10 g C	3 d	800 d	2496 a	2076 c	1408 c	2448 b	2744 a	1711 a
50 g NL + 5 g PM	6 d	960 c	1035 e	435 h	880 f	1104 e	1548 e	853 d
50 g NL + 10 g PM	2 d	1238 a	320 g	600 f	900 e	1024 f	1090 f	739 e
100 g NL + 5 g PM	0 d	444 f	1712 c	2788 a	2372 a	3010 a	1724 c	1721 a
100 g NL+ 10 g PM	0 d	760 d	1075 e	1696 d	1338 d	2312 c	1668 d	1264 c

Means followed by the same letter in each column do not differ significantly ($P \leq 0.05$)
C = Cow dung
PM = Poultry Manure
NL = Neem Leaf

ANOVA Tables – Appendices B79-B86 (pp 184 – 186)

4.3.2. Effect of the soil amendments on the breakdown of azadirachtin A

4.3.2.1. Gas Chromatography (GC) Chromatograph of azadirachtin A

Figure 4.1A shows the graphical presentation of the retention time of the standard analytical azadirachtin A which occurred at 7.9 minutes. The unamended soil and soil amended with only the animal manure showed no peak in the chromatograph (Figure 4.1B). The peaks observed in the chromatograph of the neem amended soil chromatograph (Figure 4.1C) therefore are the results of the compounds in the neem leaves. The other peaks alongside azadirachtin A (Figure 4.1C) which might be some of the limonoids of neem were, however, not identified.

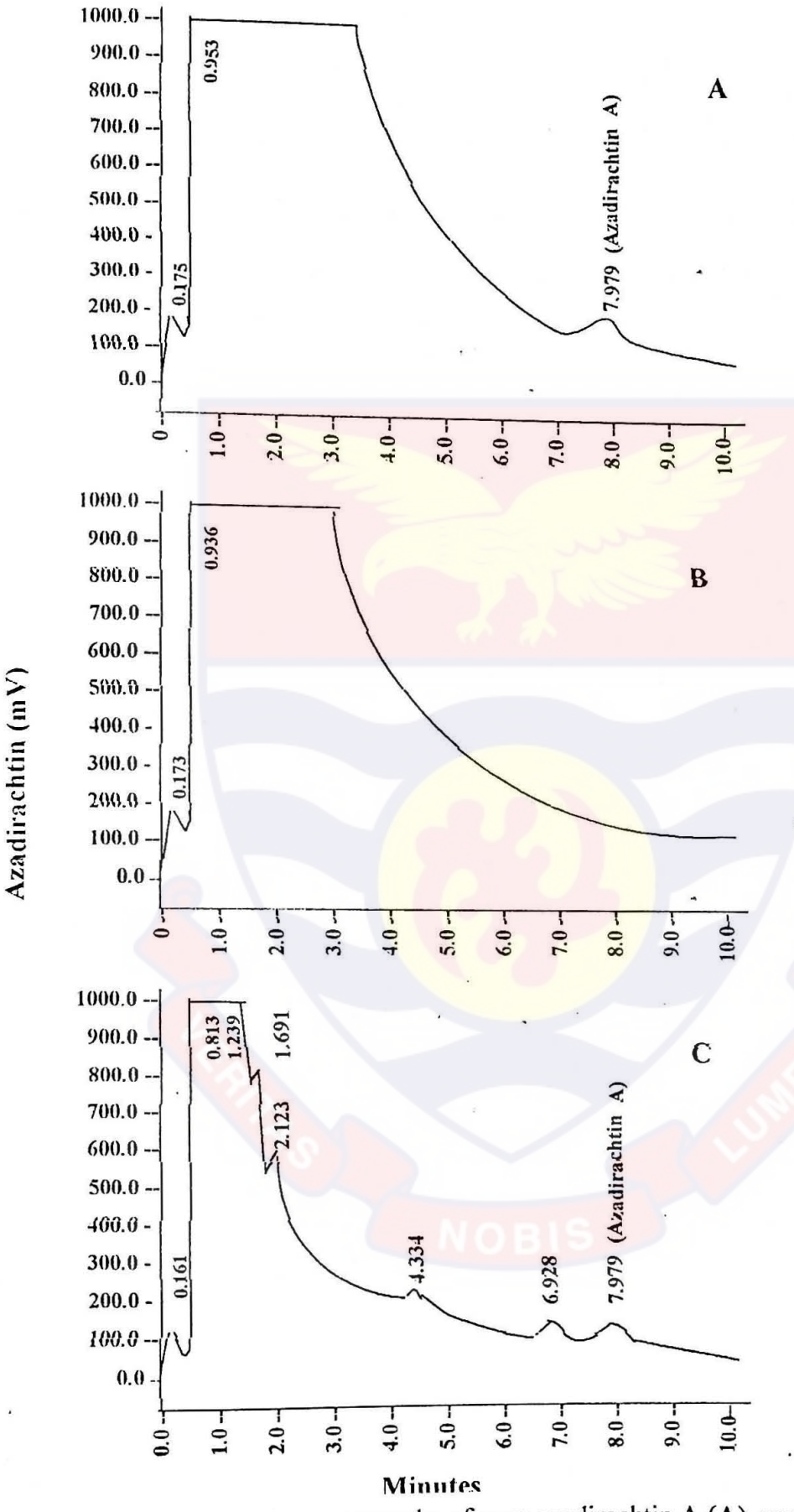

Figure 4.1. GC Chromatographs of pure azadirachtin A (**A**), unamended soil/soil amended with animal manure only (**B**) and soil amended with neem leaves (**C**).

4.3.2.2. Concentration of azadirachtin in neem leaves and recovery from soil

The concentration of azadirachtin A in the neem leaves was 3.31 µg/g (15.43% moisture). Sundaram (1996) reported the concentration of azadirachtin A in neem leaves from India to be 5.90 µg/g which was higher than the concentration obtained in the present study. The difference may be attributed to the different ecosystems of the neem sources, such as soil, climate and rainfall, and also the collection, processing and storage conditions of the neem leaves (Sundaram, 1996). The different analytical procedures of the current study (Gas Chromatography procedure) and that of Sundaram (High Performance Liquid Chromatography procedure) might have also contributed to the difference in concentration of azadirachtin A in the leaves picked by the two procedures.

Based on the concentration (3.31 µg/g) of azadirachtin A in the neem leaves, the percentage recovery of the compound in the various treatments was calculated (Table 4.4). The percentage recovery ranged between 60 and 95% with no specific pattern observed among the treatments.

The percentage recovery of azadirachtin A in soils obtained by Stark and Walter (1995) was 80.4 ± 12.12% and was considered to be acceptable for analysis. The present recovery compared favourably with that of Stark and Walter (1995).

Table 4.4. Percentage recovery of azadirachtin A extracted from soil immediately after amendment

Treatment	Expected recovery µg Aza/g soil	Amount recovered µg Aza/g soil	Percentage recovery
50 g NL	0.1504	0.0898	60
50 g NL + 5 g PM	"	0.1423	95
50 g NL + 10 g C	"	0.1275	85
50 g NL + 10 g PM	"	0.1287	86
100 g NL	0.3008	0.2299	76
100 g NL + 5g PM	"	0.2446	81
100 g NL + 10 g C	"	0.2618	87
100 g NL+ 10 g PM	"	0.2393	80

NL = Neem leaves
PM = Poultry Manure
C = Cow dung

4.3.2.3. Degradation of azadirachtin A in soil

The breakdown of azadirachtin A followed the first order kinetics (Stark and Walter, 1995; Wan *et al.*, 1997). Figures 4.2 and 4.3 represent the first order degradation curves of azadirachtin A in the various soil amendments. The days of incubation had strong relationship with the concentration of azadirachtin A in the soil amendments as could be observed from the R^2 values which ranged between 0.7551 to 0.9038. Soil amendments receiving 100 g neem leaves had steeper slopes than those receiving 50g neem leaves.

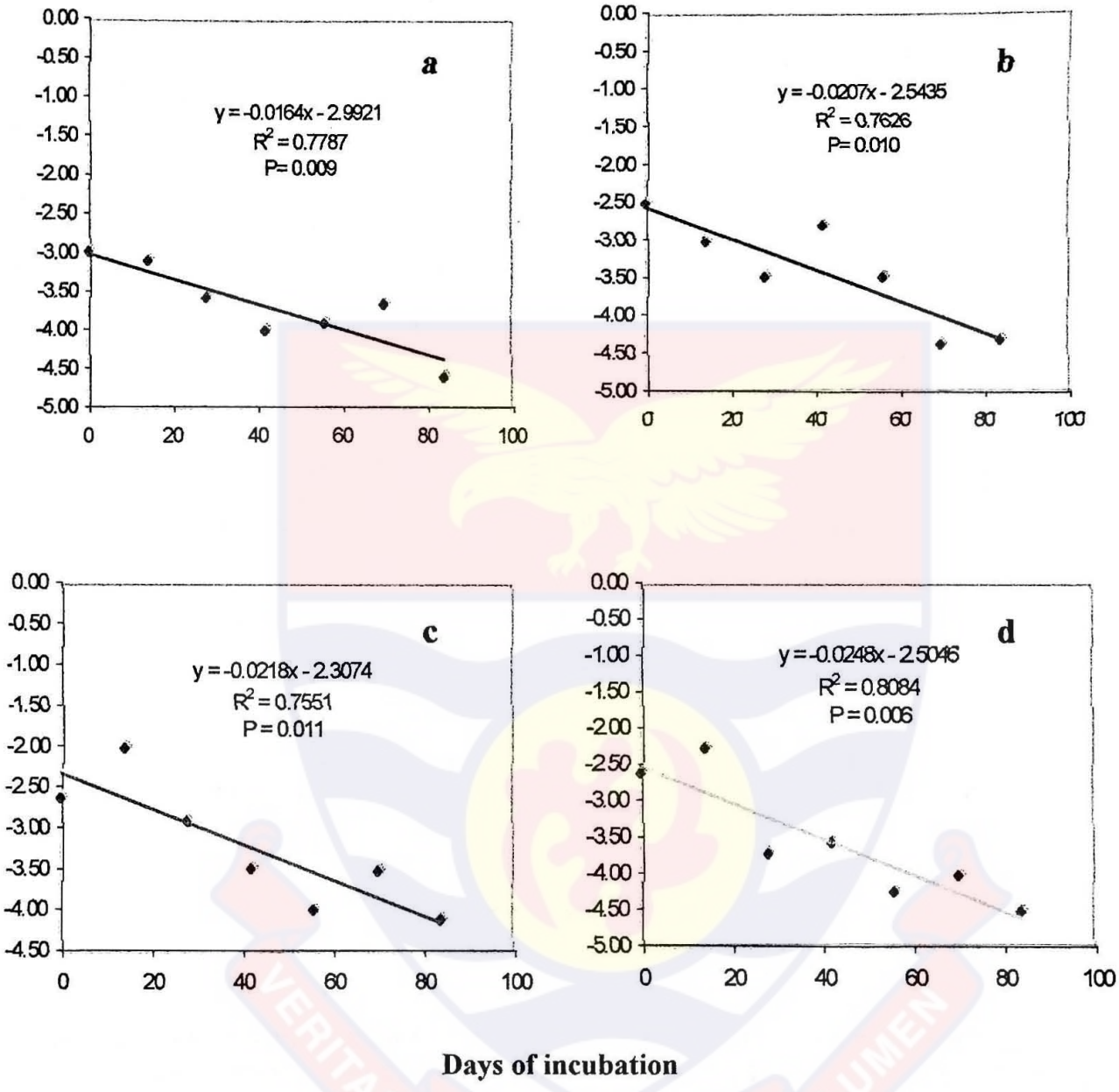

Figure 4.2. Azadiractin degradation in one kilogram of soil amended with (a) 50 g neem leaves, (b) 50 g neem leaves + 5 g poultry manure, (c) 50 g neem leaves + 10 g cow dung, and (d) 50 g neem leaves + 10 g poultry manure.

ANOVA Tables for the Fitted line Plots – Appendices B90-B93 (pp 187 – 188)

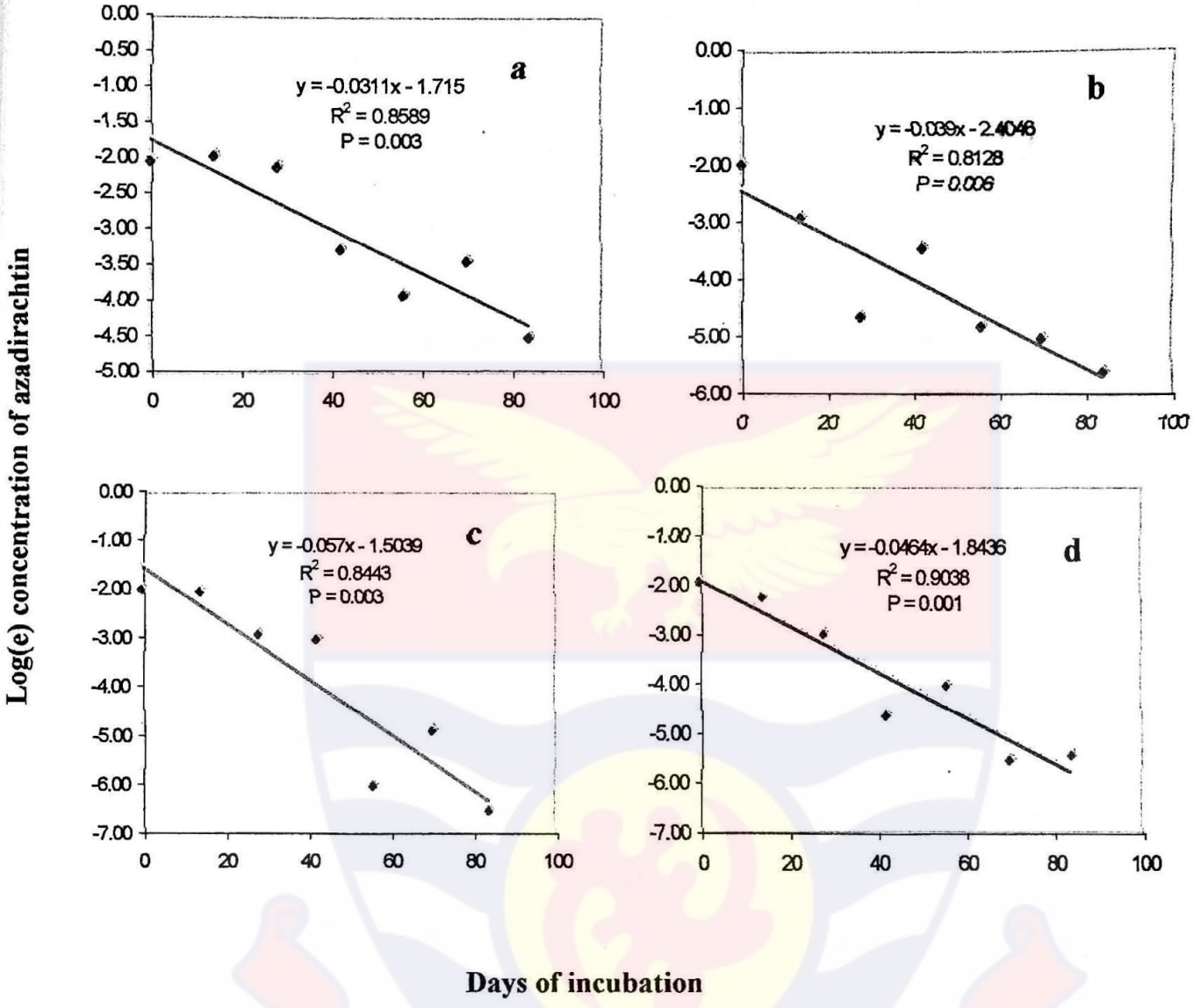

Figure 4.3. Azadirachtin degradation in one kilogram of soil amended with (a) 100 g neem leaves, (b) 100 g neem leaves + 5 g poultry manure, (c) 100 g neem leaves + 10g cow dung, and (d) 100 g neem leaves + 10 g poultry manure

ANOVA Tables for the Fitted line Plots – Appendices B94-B97 (pp 189 – 190)

The corresponding half lives are presented in Table 4.5. The half life is inversely proportional to the rate constant and therefore the steeper the slope of the curve or the more negative the slope the smaller the half life and vice versa. Treatments with higher neem amendment levels of 100 g/kg soil (Figures 4.3) showed steeper slopes with shorter half lives of azadirachtin A ranging between 12.19 and 22.29 days (Table 4.5) as compared to the lower amendment levels of 50 g neem leaves/kg soil with half lives between 28.63 and 42.26 days. Within these two groups of neem amendments the addition of the poultry manure and the cow dung gave significantly shorter half lives of the azadirachtin than the sole neem amendments.

The kind of animal manure and the quantity used did not bring any significant differences in the half lives of azadirahtin A in the soil among the neem leaf and the animal amendments (Table 4.5). However, the half lives followed in this order, 100 g NL + 10 g PM (12.19d) < 100 g NL + 10 g C (14.32d) < 100 g NL + 5 g PM (17.79d) and 50 g NL + 10 g PM (28.63d) < 50 g NL + 10 g C (31.79d) < 50 g NL + 5 g PM (33.48d), indicating poultry manure must have enhanced faster breakdown of azadirachtin A in the soil than cow dung at the same soil amendment level. The different rates of the organic amendments in the current study might be the major factor in the differences of the half lives of azadirachtin A. The breakdown of organic compounds in soils is known to be affected by the quantity of the organic matter in the soil (Morril *et al.*, 1982). Organic matter is food for many soil microbes (fungi, actinomycetes and bacteria) and promotes

Table 4.5. Half lives of azadirachtin A in soil amended with neem leaves and animal manure

Amendment (per kg of soil)	Half life of azadirachtin A (days)
50 g NL	42.26 a
50 g NL + 5 g PM	33.48 b
50 g NL + 10 g C	31.79 b
50 g NL + 10 g PM	28.63 bc
100 g NL	22.29 cd
100 g NL + 5 g PM	17.79 de
100 g NL + 10 g C	14.32 e
100 g NL + 10 g PM	12.19 e

Half lives with the same letters are not significantly different from each other($P \leq 0.05$).

NL Neem leaf
PM Poultry manure
C Cow dung

ANOVA Table Appendix B87 (pp 186)

their proliferation in the soil (Acea and Carballas, 1996) in proportion to the amount added. The breakdown of azadirachtin A in the soil is enhanced by soil microbes (Stark and Walter, 1995; Sundaram, 1996) and would be affected differently under the different soil amendment conditions as has been observed in the current study.

Moisture as a factor promoting the breakdown of organic compounds in the soil (Morril *et a.l*, 1982) might have contributed to the pattern of the breakdown of

azadirachtin A breakdown in the soil as the higher levels of soil amendments (100 g NL) significantly showed higher moisture content than the lower level (50 g NL) (Table 4.6).

The contribution of pH in the breakdown of azadirachtin A could not be considered as a factor in this study as the pH values of the amendments were not significantly different from each other (Table 4.6).

Samples were exposed to the same varying environmental field temperature conditions, and thus eliminating differences in the azadirachtin A breakdown resulting from temperature differences (Stark and Walter, 1995; Szeto and Wan, 1996).

The breakdown of azadirachtin A in soil amended with neem leaves could be affected by the quantity of the neem leaves applied and the additions of animal manure. The potency of azadirachtin A to control pests in the soil is likely to be reduced at a faster rate under higher rates of applied neem leaves, and re-infestation of pests therefore is also likely be higher when larger amount of the material is applied to the soil.

Table 4.6. Moisture content and pH of amended soils

Amendment (per kg of soil)	pH	Moisture content (%)
50 g NL	6.3 a	11.67 b
50 g NL + 5 g PM	6.6 a	10.45 b
50 g NL + 10 g C	6.4 a	11.16 b
50 g NL + 10 g PM	6.6 a	12.10 b
100 g NL	6.5 a	15.11 a
100 g NL + 5 g PM	6.4 a	16.90 a
100 g NL + 10 g C	6.3 a	16.01 a
100 g NL + 10 g PM	6.6 a	16.00 a

Moisture contents with the same letters are not significantly different from each other ($P \leq 0.05$).

NL Neem leaf
PM Poultry manure
C Cow dung

ANOVA Tables – Appendices B88-B89 (pp 187)

4.4. Summary and conclusions

Soil amendment with neem leaves, poultry manure and cow dung significantly reduced plant-parasitic numbers and increased non plant-parasitic nematode populations.

The 50 g neem leaves/kg soil and the 100 g neem leaves/ kg soil and their combinations with the poultry manure and the cow dung were not significantly different in the control of plant-parasitic nematodes. The neem and the neem based amendments were more effective than the sole poultry manure and the cow dung amendments in the suppression of the plant-parasitic nematodes.

The breakdown of azadirachtin A in the soil varied with the different inclusion rates of the neem leaves and the animal manure. The higher level of the neem leaves (100 g) soil amendment and its combinations recorded shorter half lives of azadirachtin A than the lower level of neem leaves (50 g) and its combinations. The use of Gas Chromatography (GC) for quantification of azadirachtin A in the soil and in the neem leaves has been made possible in the current study. The GC therefore can be used as an alternative to the High Pressure Liquid Chromatography (HPLC).

From the above it would be more economical to use the lower level 50 g neem leaves in a soil amendment to control soil parasitic nematodes than the higher level 100 g neem leaves. The effects are significantly not different from each other in

performance. In addition, the active ingredient of neem, azadirachtin A would stay longer in the 50 g neem leaves soil amendment.

CHAPTER FIVE

5.0 Studies into the influence of soil amended with neem leaves and poultry manure on nutrient dynamics.

5.1. Introduction

Aside the application of neem to control pests as revealed in the previous chapter, neem products also enrich the soil (Khan and Saxena, 1997; Akhtar, 1999; Chakrabarti, 2000). This is imperative, for the farmer needs not only to control soil pests but also to improve the nutrient status of his soil for optimum crop yield using organic substances.

Appropriate methods of farming using organic substances which are less expensive and do not disturb nature, would have key roles to play in ensuring food security, improving human health and conserving the environment. The dual activity of neem as fertilizer and pest repellent has made it a favoured traditional organic source for soil amendment in India (Neem Foundation, 1997).

The neem tree is commonly found in the tropical regions with the leaves evergreen throughout the year. The use of the neem leaves which is readily available combined with poultry manure, an effective nutrient provider (Abdel Magid *et al.*, 1995, Riegel *et al.*, 1996, Nyakatawa and Reddy, 2000) as soil amendment would help farmers to solve the dual problem of low soil fertility and soil pest control especially nematodes.

The present study seeks to compare the nutrient content of a soil amended with neem leaves and poultry manure before and after the amendment, and to particularly observe the patterns in the mineralization of carbon and nitrogen.

5.2. Materials and Methods

5.2.1. Treatments and experimental design

The experiment was set up on 17th of February 2003. The treatment combinations (Table 5.1) used in the experiment, the quantity of soil used and the description under this section were the same as in Chapter 4.

Table 5.1. Table showing treatments used in Chapter 5

Treatment (per kg soil)
No amendment (Control)
5 g PM
10 g NL
10 g NL + 5 g PM
20 g NL + 5 g PM
30 g NL + 5 g PM
40 g NL + 5 g PM
50 g NL + 5 g PM

PM = Poultry manure
NL = Neem leaves

5.2.2. Nutrient analyses

5.2.2.1. Organic carbon

Organic carbon in the samples was determined by the dichromate oxidation method (IITA, 1985). Ten millilitres of a normal solution of $K_2Cr_2O_7$ was added to 0.5 g of treatment sample in a 500 ml conical flask. This was followed by the addition of 20 ml concentrated H_2SO_4. After 30 minutes 200 ml of distilled water was added to the contents of the flask. An amount of 0.2 g of NaF followed by 1.0 ml of diphenylamine indicator were then added. The organic carbon of the sample was determined by titration with freshly prepared 0.5 *M* ferrous sulphate solution after carrying out a blank titration.

5.2.2.2. Carbon dioxide

Carbon dioxide was determined by the modified method of Rowell (1994). Carbon dioxide produced by respiration from 50 g soil sample placed in a 250 ml conical flask was trapped by absorption into 10 ml 0.3 *M* NaOH solution for 6 hours. The NaOH was placed in a vial suspended in the flask from a rubber bung which was used to seal the flask. After the 6^{th} hour the CO_2 trapped was determined by titration with 0.1 *M* HCl after adding 10 ml of 1 *M* $BaCl_2$.

5.2.2.3. Total nitrogen

Total nitrogen in the soil, neem leaves and poultry manure was determined by the Kjeldahl oxidation method. This involved digestion of 0.2 g sample at 360 ^{O}C for

2 hours with a digestion mixture (selenium powder, lithium sulphate, hydrogen peroxide and concentrated sulphuric acid) followed by steam distillation and titration with 1/140 *M* HCl (Anderson and Ingram, 1989).

5.2.2.4. Total phosphorus

Aliquot from the digest above was used for the determination of total phosphorus colorimetrically (Anderson and Ingram, 1989). One millilitre of sample was pipetted into a test tube with the addition of 4.0 ml ascorbic acid solution and 3.0 ml molybdate reagent. The content of the test tube was mixed well and allowed to stand for about 1 hr for colour development. The absorbance was determined with a spectrophotometer at 880 nm. The total phosphorus in the sample was determined from a graph of absorbance against standard concentrations of phosphorus.

5.2.2.5. Inorganic NH_4^+ - N and NO_3^- - N

Inorganic NH_4^+ - N and NO_3^- - N in the soil were determined after extraction of 40 g sample with 2 *M* KCl on a mechanical shaker for 1 hour. It was followed by steam distillation with MgO and Devarda's Alloy for NH_4^+-N and NO_3^--N respectively with boric acid solution as an indicator in the receiving flasks. Distillates were titrated with 0.01 *M* HCl (Rowell, 1994).

5.2.2.6. Available phosphorus

For the analysis of available phosphorus , 1 g soil sample was mixed and shaken with 7 ml Bray No. 1 solution (15ml of 1 *M* NH_4F and 25 ml of 0.5 *M* HCl in 460 ml distilled water) in centrifuge tubes for 1 minute and filtered through Whatman No. 1 filter paper. Aliquots of 2ml of sample extracts were used to determine the phosphorus through the ascorbic acid method (IITA, 1985).

5.2.2.7. Exchangeable bases

Exchangeable calcium, magnesium and potassium were extracted by mixing 5 g of soil sample with 20 ml 1 *M* ammonium acetate solution overnight in 100 ml volumetric flask. The suspension was transferred to a funnel fitted with Whatman No 1 filter paper and filtered into a 100 ml volumetric flask The sample in the funnel was leached with 4 successive 20 ml volumes of the acetate solution (IITA, 1985). Aliquots of the extract were used for the subsequent determination of the exchangeable bases. The concentration of exchangeable potassium was determined using a Flame photometer (IITA, 1985). Calcium + magnesium were determined by the EDTA Titrimetry method. A 15 ml of buffer solution (NH_4Cl + NH_4OH) was added to 25 ml aliquot from the sample above in 250 ml conical flask Distilled water was added to the flask to the 150 ml mark. Ten drops each of KCN, $NH_2OH.HCl$, $K_4Fe(CN)_6$ and triethanolamine were added to the content. Ten drops of Eriochrome Black T indicator were added to the flask content and the solution titrated with 0.005 *M* EDTA to a blue end point for the determination of both cations (Page *et al.*, 1982). Calcium alone was determined through the same

process as calcium + magnesium, however, enough 10% NaOH solution was added in place of the buffer solution to raise the pH to 12, and 5 drops of Calcon indicator was used instead of the Eriochrome Black T indicator (Page *et al.*, 1982). Magnesium was estimated from the difference of the two titrations.

5.2.2.8. Exchangeable acidity

Exchangeable acidity was extracted by adding 3 successive 30 ml 1 *M* KCl to 5 g sample in a 45 ml centrifuge tube, the content of the tube was shaken for 1 hour on a shaker and centrifuged at 2000 rpm for 15 minutes at each addition of the KCl. After each addition, the clear supernatant was decanted into a 100ml volumetric flask. The volume was made up to the mark with 1 *M* KCl. An amount of 25 ml of the KCl extract was titrated with 0.05 *M* NaOH in the presence of a phenolphthalein indicator to determine the exchangeable acidity (IITA, 1985).

5.2.2.9. pH

Measurement of pH was made by making a suspension of 10 g sample in a 50 ml centrifuge tube with 25 ml distilled water. The content of the tube was shaken for 15 minutes. Electrodes of a pH meter were inserted in the suspension and the pH recorded after 30 seconds (Rowell, 1994).

5.2.2.10. Gravimetric water content

Moisture content (gravimetric water content) was determined by weighing 10 g sample into a pre-weighed 100 ml beaker and heating in an oven for 24 hours at 105 ^{O}C The sample was reweighed after heating. The percentage moisture was calculated from the difference of the two weighing (Rowell, 1994).

$$\text{Moisture content of sample} = \frac{\text{Soil sample before drying (g)} - \text{Soil sample after drying (g)}}{\text{Soil sample after drying (g)}}$$

5.2.3. Data analysis

The cumulative amounts of the net CO_2 – C released and the net mineral N produced from the neem leaves and the poultry manure were plotted against time of incubation; a linear model/zero order reaction equation (equation 1) described the relationships better than the first order reaction equation (equation 2)

$$m = a + kt \qquad (1)$$

$$m = a(1 - \exp^{-kt}) \qquad (2)$$

where m = amount of nutrient at time ' t' , while 'a' and 'k' are constants.

The net rates and the net amounts of mineralization of organic carbon and nitrogen and the final nutrient contents of treatments at the end of the incubation period were subjected to the analysis of variance (ANOVA) and the Duncan's Multiple Range Test using the MSTAT-C statistical software (Russell, 1990).

All relevant ANOVA Tables are found in Appendix B.

5.3. Results and Discussion

5.3.1. Chemical composition of neem leaves and poultry manure, and the initial C:N ratio of treatments

The total N, P, Ca, and Mg contents were higher in the poultry manure than in the neem leaves, while that of K and Organic carbon were higher in the neem leaves (Table 5.2). The quantity of P and Mg in the poultry manure were about 9 and 3 times respectively higher than in the neem leaves. The chemical composition is comparable to reported data in Tables 2.1 and 2.2.

The treatments with higher inclusions had wider initial C:N ratios (Table 5.3). Mineralization and immobilization are affected by the C:N ratio of the decomposing organic matter. A C:N ratio greater than 30:1 will immobilize N, a ratio less than 20:1 will mineralize N, and a ratio between 20:1 and 30:1 will produce no net changes in N availability (Tisdale et al., 1993). Based on these assumptions the C:N ratios of the treatments were expected to initially favour mineralization.

Table 5. 2. Chemical composition of neem leaves and poultry manure

	N	P	K	Ca	Mg	OC	C:N	Moisture
	(%)							Content (%)
Poultry manure	3.55	1.29	0.95	1.16	0.96	36.97	10.41	18.77
Neem leaves	2.50	0.14	1.19	1.13	0.31	49.46	19.78	16.05

Table 5. 3. The initial C:N ratio of treatment materials

Treatment	Organic Carbon (%)	Total N (%)	C:N
Control (unamended soil)	0.85	0.05	17.00
5 g PM/kg soil	1.02	0.07	14.57
10 g NL/kg soil	1.27	0.08	15.88
10 g NL + 5 g PM /kg soil	1.59	0.10	15.90
20 g NL + 5 g PM /kg soil	2.05	0.12	17.08
30 g NL + 5 g PM /kg soil	2.45	0.13	18.87
40 g NL + 5 g PM /kg soil	3.53	0.18	19.61
50 g NL + 5 g PM /kg soil	3.68	0.19	19.25

5.3.2. Carbon dynamics

With the exception of the unamended soil (control) which had a peak of CO_2 – C production at the second week of incubation, the rest of the amended soil recorded their peaks on the first day of incubation (Figure 5.1). The CO_2 – C production continued to decline after the peaks. The period of continuous reduction of CO_2 – C, up to the attainment of almost constant production of the gas varied widely among the treatments. The unamended soil and the lower levels of the neem and the poultry manure inclusions (5 g poultry manure/kg soil, 10 g neem leaves/kg soil, 10 g neem leaves + 5 g poultry manure/kg soil and 20 g neem leaves + 5 g poultry manure/kg soil) had this period up to the 4th week of incubation.

The higher levels of the amendments (30 g neem leaves + 5 g poultry manure/kg, 40 g neem leaves + 5 g poultry manure/kg and 50 g neem leaves + 5 g poultry manure/kg), however, had an extension of the period of CO_2 – C decline to the 6th week of incubation.

A slight second peak of CO_2 – C production was recorded for the highest level of amendment at the 8th week of incubation. Soil moisture content of treatments rose to a maximum on the 8th week of incubation (Figure 5.13) and this might have accounted for the slight second flush of CO_2 – C production observed for the highest treatment level of the amendments. The changes in the soil moisture conditions might have boosted microbial biomass activities leading to the production of the second flush of the CO_2 – C production (Franzluebbers *et al.*, 1994; Robertson and Morgan (1995). The organic carbon content of the other

treatments might not be enough for this second flush of CO_2 evolution to be observed.

The observed peaks/flushes of the CO_2 – C probably indicate active biological reactions taking place in the soil media after the additions of the organic matter. Such an observation was also recognised by West *et al.* (1989) and Pocknee and Sumner (1997) using a different organic amendment source of soil incorporation. With the loss of degradable organic carbon as CO_2 – C, the organic carbon content of treatments relatively declined (Figure 5.2).

The differences in the flushes of CO_2 – C production or the differences in the decline of the CO_2 - C production may be assigned to the differences in the organic carbon contents of the amendments (Table 5.3). The amount of CO_2 – C production relatively corresponded with the quantity of organic carbon in the treatment.

The net cumulative CO_2 – C produced by the various amendments is presented in Figure 5.3. The rate of production of CO_2 – C (Table 5.4) was calculated from the net cumulative graph by fitting the linear equation, $m = a + bt$ (zero order reaction equation). The rate of release of nutrients in soils in previous studies followed the zero-order reaction kinetics (Nordmeyer and Richter, 1985; Seyfried and Rao, 1988; Murwira and Kirchmann, 1991; Abdel Magid *et al.*, 1993; Mahimairaja *et al.*, 1994).

Treatments with higher organic carbon produced significantly higher net cumulative $CO_2 - C$, rate of production of the $CO_2 - C$ (Table 5.4) and also higher net percent of mineralized organic carbon at the 12th week of incubation (Table 5.4). However, the 5 g PM with low organic carbon of 1.02 % (Table 5.3) had higher net cumulative $CO_2 - C$, rate of $CO_2 - C$ production and higher net per cent organic carbon mineralization than the 10 g NL. This might have resulted from the high levels of microbes, both amonifiers and nitrifiers in the poultry manure (Bacharach, 1957).

The mineralization of organic carbon ranged between 33.33% for the 10 g NL and 70% for the 50 g NL + 5 g PM. The higher the rate of the $CO_2 - C$ released the higher the net percent of the organic carbon mineralized and vice versa. The combination of 10 g NL and 5 g PM resulted in higher rate of $CO_2 - C$ production and higher net percent organic carbon mineralization than the individual materials (Table 5.4). The higher microbial content of the poultry manure might have increased the biological activities in the 10 g neem leaves + 5 g poultry manure/kg soil, and thus resulting in the higher rate of $CO_2 - C$ production and higher net percent organic carbon mineralization than the sole 10 g NL and 5 g PM. The addition of the poultry manure to other treatments might have played a similar role.

Organic carbon mineralization is a mirror image of the release patterns of other nutrients in the soil. The larger the C evolved as $CO_2 - C$ the higher the likelihood of release of the other soil nutrients (Abdel Magid *et al.*, 1993; Atalla *et al.*, 1995).

5.3.3. Nitrogen dynamics

Nitrogen changes in the treatments are presented in Figures 5.4, 5.5 and 5.6 for NH_4^+ - N, NO_3^- - N and total nitrogen respectively. Two conspicuous peaks/flushes of NH_4^+ - N production were observed, the highest peak of production occurred at the second week and the lowest peak at week eight. At these peaks the higher amendment levels also produced higher amounts of the NH_4^+ - N (Figure 5.1).

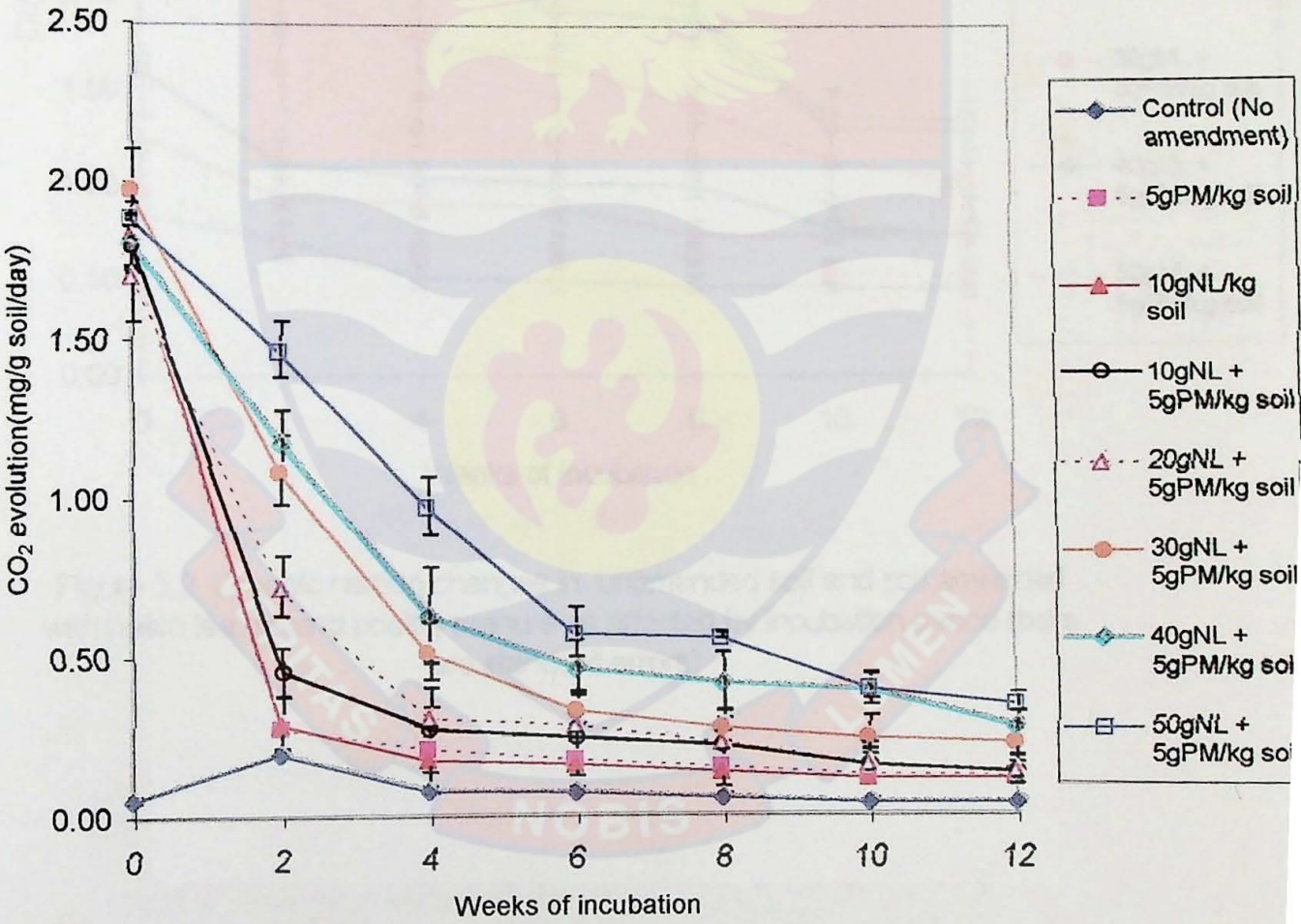

Figure 5.1. Changes in CO_2 evolution in unamended soil and soil amended with neem leaves and poultry manure as affected by incubation period (bars are standard errors)

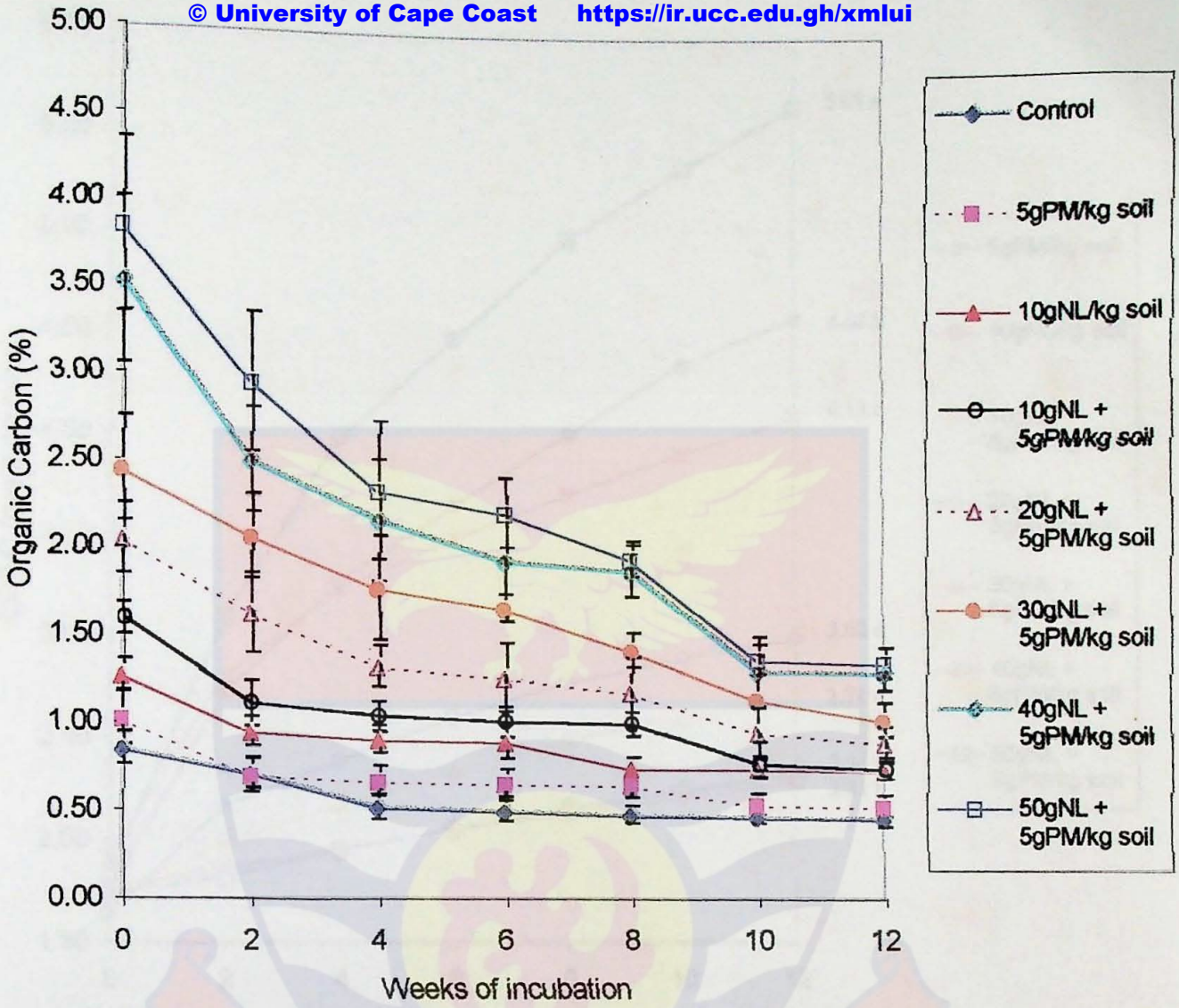

Figure 5.2. Organic carbon changes in unamended soil and soil amended with neem leaves and poultry manure as affected by incubation period (bars are standard errors)

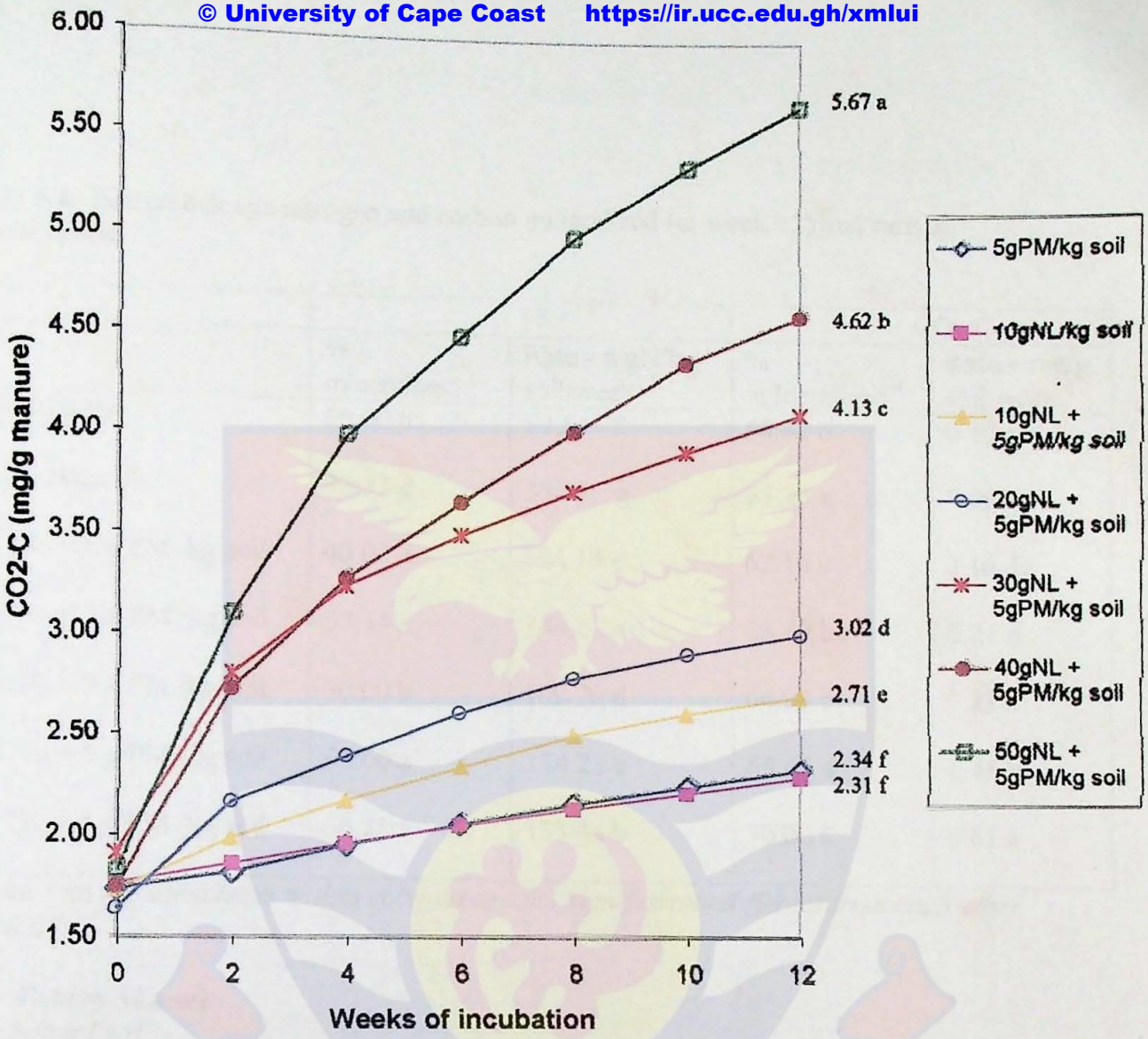

Figure 5.3. Net cumulative CO_2-C mineralized from neem leaves and poultry manure

Letters show differences in cumulative figures at the 12th week of incubation($P \leq 0.05$)

ANOVA Table – Appendix B98 (pp 190)

Table 5.4. Net percentage nitrogen and carbon mineralized (at week 12) and rate of mineralization

	N		CO_2-C	
	% mineralized [1]	Rate - mgN/kg soil/week [2]	% mineralized [1]	Rate – mg/g soil/week [2]
5 g PM/kg soil	50.00 b	59.54 f	58.82 d	0.10 e
10 g NL/kg soil	33.33 d	38.53 g	33.33 e	0.09 e
10 g NL + 5 g PM /kg soil	40.00 c	104.18 e	62.16 c	0.16 de
20 g NL + 5 g PM /kg soil	57.14 a	144.62 a	63.33 bc	0.21 d
30 g NL + 5 g PM /kg soil	50.00 b	109.70 d	64.38 b	0.33 c
40 g NL + 5 g PM /kg soil	56.00 a	114.21 c	68.66 a	0.45 b
50 g NL + 5 g PM /kg soil	56.25 a	133.48 b	70.00 a	0.61 a

Means with the same letter within columns are not significantly different from each other ($P \leq 0.05$).

PM= Poultry Manure
NL= Neem Leaf

1. Values were calculated based on the difference between the initial net total % of N/C and the total % N/C at the 12th week of incubation

2. Values were calculated from the net cumulative mineralization graphs (CO_2 – C and N) by fitting the linear equation $m = a + bt$

ANOVA Tables – Appendices B134-B137 (pp 202 – 203)

Generally, the NH_4^+ - N production drastically declined by the 4th week of incubation. The 2nd small flush of NH_4^+ - N production at the 8th week might result from ammonification of organic nitrogen and immobilization of NO_3^- - N. Moisture content of treatments were highest at the 8th week (Figure 5.13) and thus created an anaerobic condition which favoured the ammonification and the immobilization processes (Mahimairaja *et al.*, 1994). The NH_4^+ - N production thereafter fell even below the initial recorded values, probably due to higher rate of nitrification and slower rate of ammonification .

The 5 g poultry manure amendment released higher amount of NH_4^+ - N than the 10 g neem leaves/kg (Figure 5.4) though the two amendment materials have similar total N and C:N ratio in the soil (Table 5.3). This variation may be expected as the poultry manure contains easily decomposable compounds (Abdel Magid *et al.*, 1993) and high levels of microbes to speed up mineralization (Bacharach, 1957).

The 10 g neem leaves + 5 g poultry manure/kg soil had higher release of NH_4^+ - N than the 10 g neem leaves/kg soil, indicating an enhancement of the release of NH_4^+ - N by the addition of the poultry manure to the neem leaves.

The addition of poultry manure to neem leaves in any soil amendment would be important to enhance the release of NH_4^+ - N, and thus improve the available N status in the soil for the growth and development of crop.

The peak of NO_3^- - N production occurred around the 4th week of incubation (Figure 5.5), two weeks after the 1st peak of NH_4^+ - N production. Soil amendment

with poultry manure alone and the unamended soil, however, had this peak at the 2nd week of incubation. Such trends of NH_4^+ - N production peaks giving way to NO_3^- - N production peaks are bound to happen in an incubation process as NH_4^+ - N production from organic compound (ammonification) is a prerequisite in nitrogen mineralization for the production of NO_3^- - N (nitrification).

The NO_3^- - N production reduced to a very low level after the peak of production for the various treatments between the 6th and 10th weeks of incubation, which corresponded with the period of the 2nd peak of NH_4^+ - N production (Figure 5.4). Moisture content of treatments was highest at this period of incubation and thus probably created an anaerobic condition which might have favoured the conversion of NO_3^- - N to NH_4^+ - N, leading to decreased levels of NO_3^- - N between the 6th and the 10th weeks. The NO_3^- - N production levels increased again but did not reach the original level because the moisture contents of the samples were still high.

The higher levels of the neem leaves and the poultry manure were supposed to release higher amounts of NO_3^- - N in relation to the CO_2 – C (Figure 5.1) and NH_4^+ - N (Figure 5.4), but this was not so. However, the lower levels of amendments (10 g NL + 5 g PM /kg soil, 20 g NL + 5 g PM /kg soil and 30 g NL + 5 g PM /kg soil) rather released higher NO_3^- - N than the higher levels of the neem leaves and the poultry manure (40 g NL + 5 g PM /kg soil and 50 g NL + 5 g PM /kg soil) until the 8th week of incubation (Figure 5.5). It was after this period

that the higher levels of the neem leaves 40 g NL and 50 g NL and the poultry manure tended to release higher amounts of NO_3^- - N. The higher treatment levels of the neem leaves apparently inhibited the nitrification process which resulted in reduced NO_3^- - N production.

Table 5.5 shows the C:N ratios of treatments measured at the various incubation periods. The higher amendment levels with higher organic carbon levels had wider C:N ratios, however, the differences were in most cases not significant within the incubation periods. The C:N ratios in all the treatments decreased with time of incubation as organic carbon was mineralised. The C:N ratios at the end of the incubation were around 12:1 where populations of decay bacteria were considered stable (Sullivan, 1999). The C:N ratios throughout the experiment favoured mineralization . The release or demand for N depends not only on the C:N ratio but also on the types of organic compounds in the residue (Chandler *et al.*, 1980; Hatiori and Mukai, 1986; Henry, 1991).

The net cumulative N (NH_4^+- N and NO_3^- - N) mineralized from the neem leaves and the poultry manure in the amendments are indicated in Figure 5.6. The 50 g NL + 5 g PM and the 20 g NL + 5 g PM produced the highest net cumulative N with the 10 g NL and the 5 g PM releasing the lowest amount of the nutrient. The net rates of production of N, calculated from the net cumulative graph of N as done for the carbon above, and the net per cent of N mineralized at the end of the incubation period are shown in Table 5.4.

The fastest rate of N mineralization and the highest amount of net percent N mineralized was from the 20gNL + 5gPM combination, which was greater than the higher levels of the other combinations. Barbarika et al. (1985) reported N mineralization to be negatively affected when the C:N ratios were increased from 2.4 to 16. There was no conspicuous effect of the C:N ratio on N mineralization in the present study. This may be due to the insignificant differences in the C:N ratios in Table 5.5 among the neem leaves amendments measured during most of the incubation periods (Table).

Neem products such as the seed powder and the seed cake have been reported to be nitrification inhibitors because of azadirachtin, the active ingredient of the neem plant (Neem Foundation, 1997; Lalljee *et al.*, 1999; Deepanjan *et al.*, 2000; Shah and Faheem, 2000). The higher levels of neem leaf with corresponding high amounts of azadirachtin might have played a similar nitrification inhibitory role in the present study up to the 8th week of incubation (Figure 5.5). This eventually might have had an impact on the calculated net cumulative and rate of N mineralization. The inhibitory potency of the azadirachtin seemed to have reduced after the 8th week as a result of degradation or the NO_3^- - N release potential of the lower rates of the neem leaves might have decreased because of the initial faster release of NO_3^- - N (Figure 5.5).

The suppression of NO_3^- - N release in the soil with the use of neem leaves in a soil amendment would be important in controlling the gradual release of NO_3^- - N for crop growth, and the prevention of excessive NO_3^- - N from leaching and causing pollution (Gnanavelrajah and Kumaragamage, 1999).

The total nitrogen in the various treatments at the end of the study was lower than the initial concentrations (Figure 5.7). The decrease in the nitrogen level may be attributed to volatilisation of NH_3, nitrous oxide (NO_2) and molecular nitrogen (N_2) (Nodar *et al.*, 1990; Mahimairaja *et al.*, 1994). Generally, the total nitrogen and NH_4^+- N contents were significantly higher in higher amendment levels (Appendix A9). The NO_3^- - N production did not follow this pattern because of the probable inhibition of nitrification.

For the efficient use of N from the neem leaves and the poultry manure soil amendments, the crop's N requirement should be timed closely to the nitrogen release patterns (NH_4^+- N and NO_3^- - N) of the amendments.

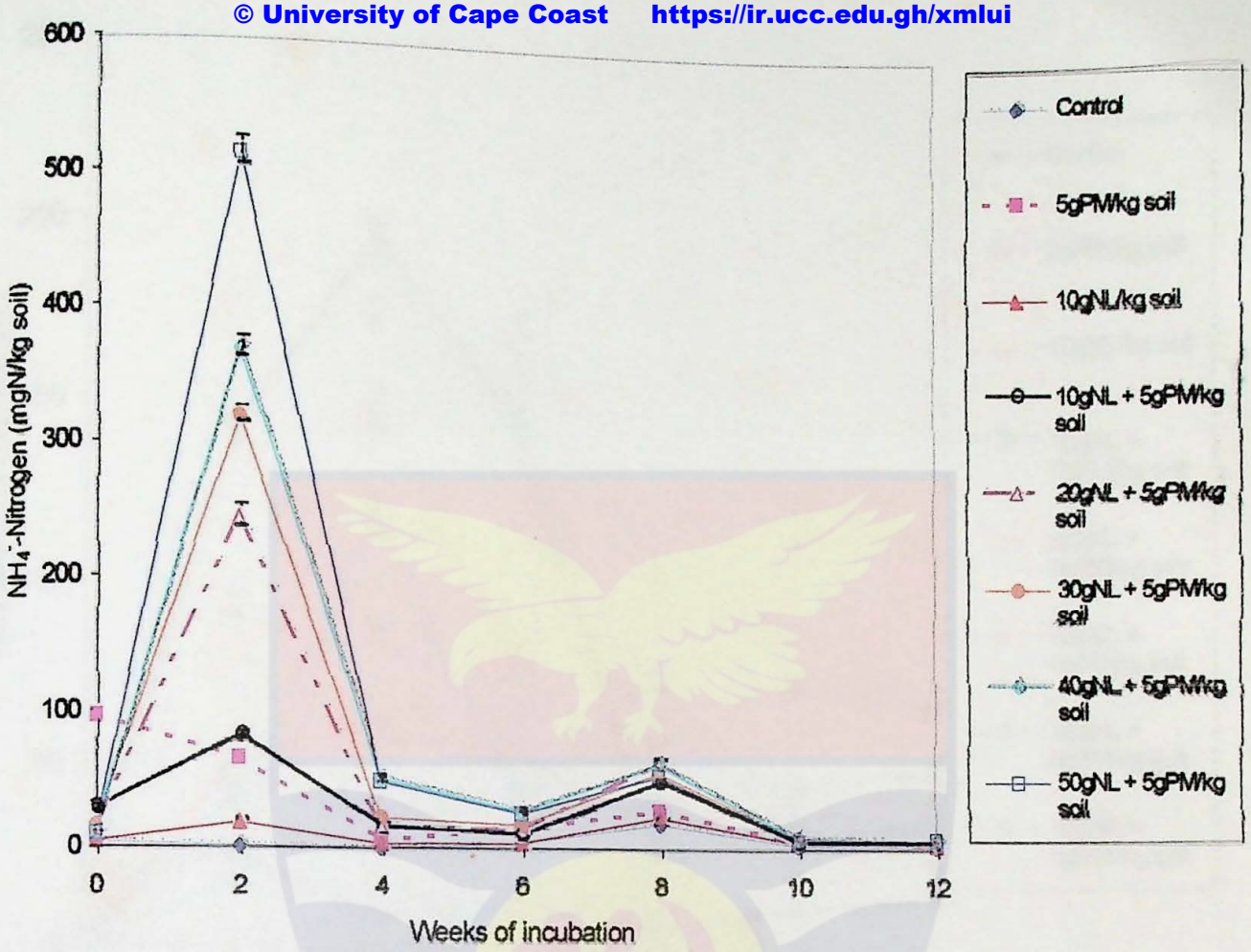

Figure 5.4. Changes in NH_4^+- Nitrogen released in unamended soil and soil amended with neem leaves and poultry manure as affected by incubation period (bars are standard errors)

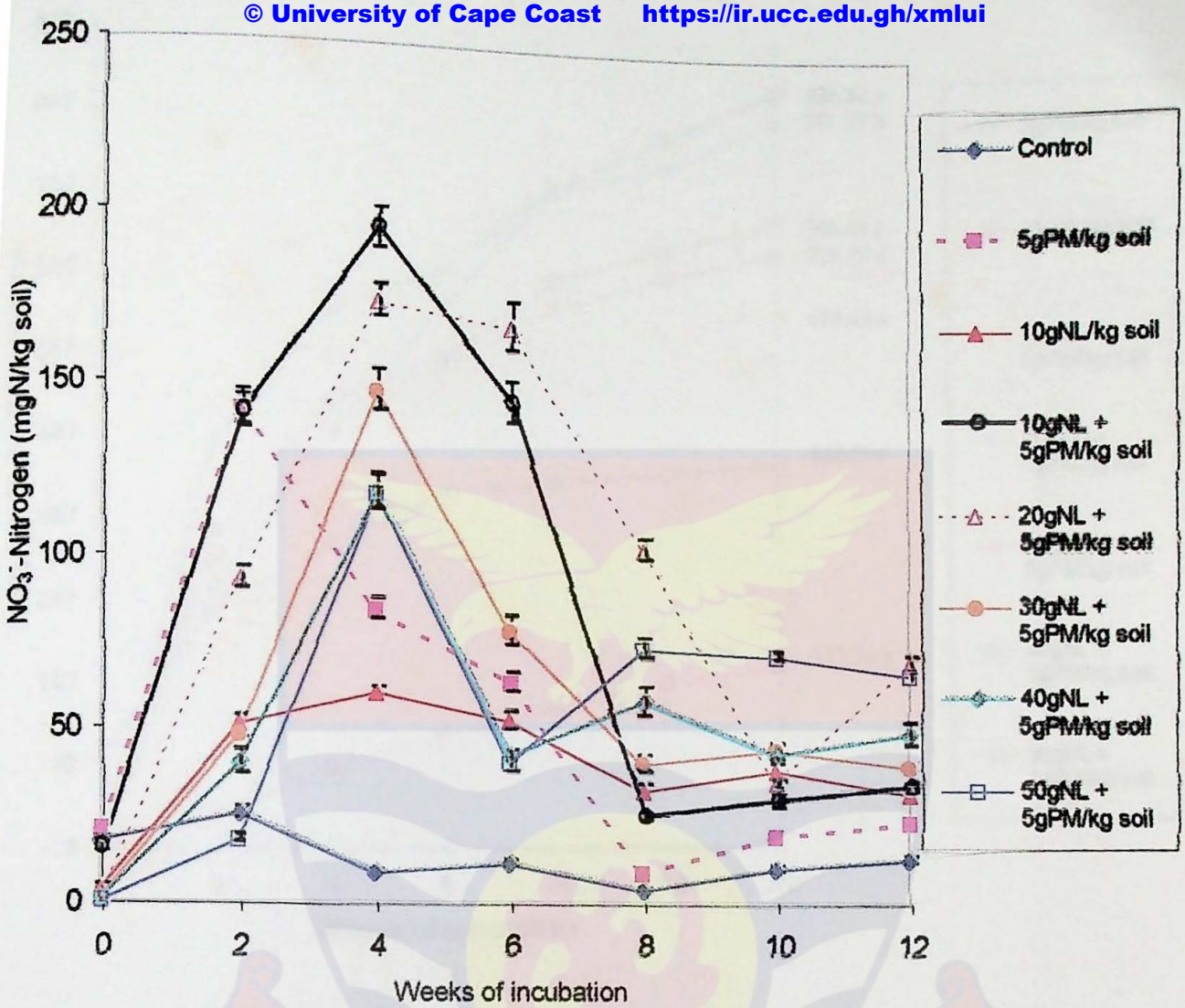

Figure 5.5. Changes in NO_3^--Nitrogen in unamended soil and soil amended with neem leaves and poultry manure as affected by incubation period (bars are standard errors)

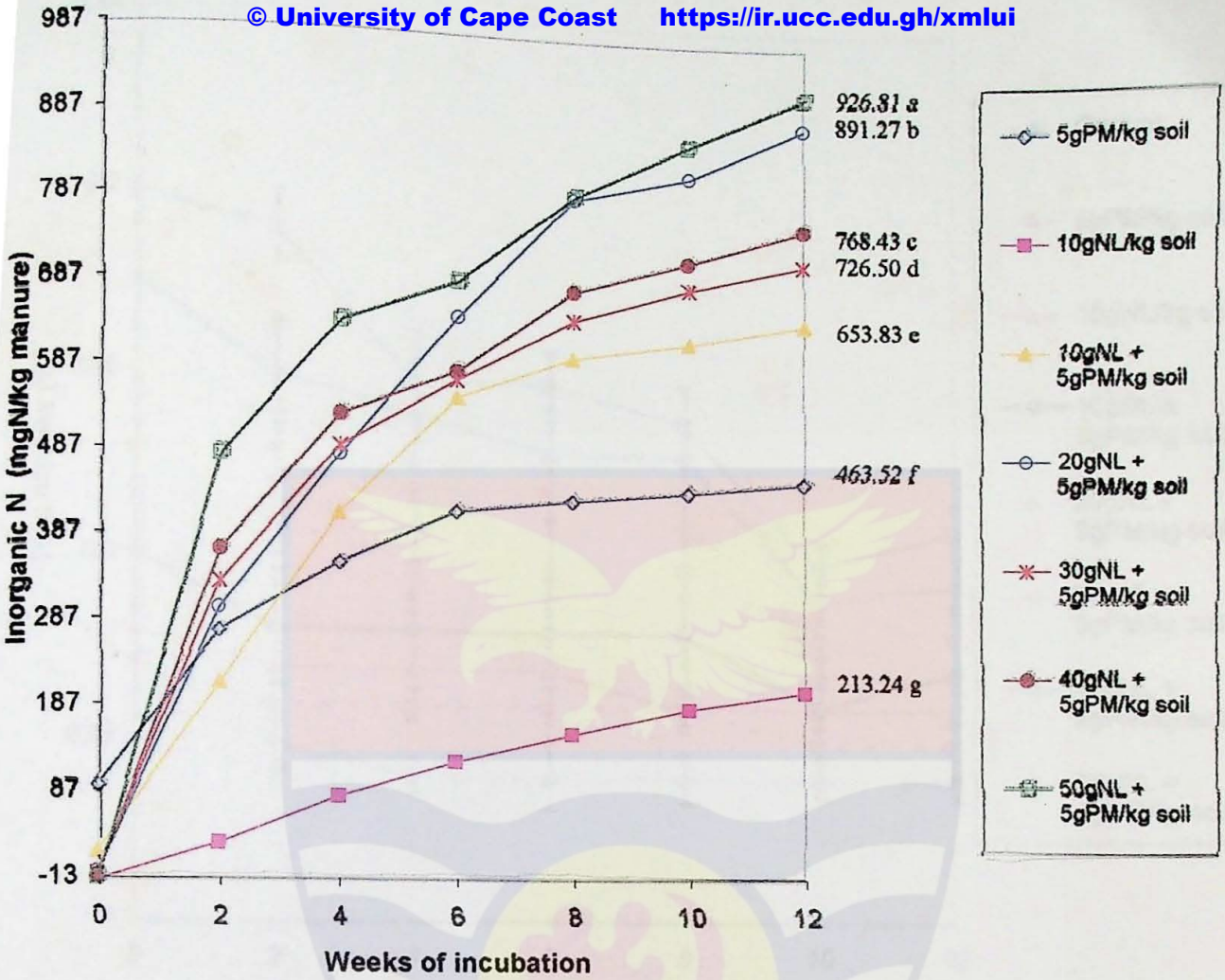

Figure 5.6. Net cumulative N mineralized from neem leaves and poultry manure

Letters show differences in cumulative figures at the 12th week of incubation($P \leq 0.05$)

ANOVA Table – Appendix B99 (pp 190)

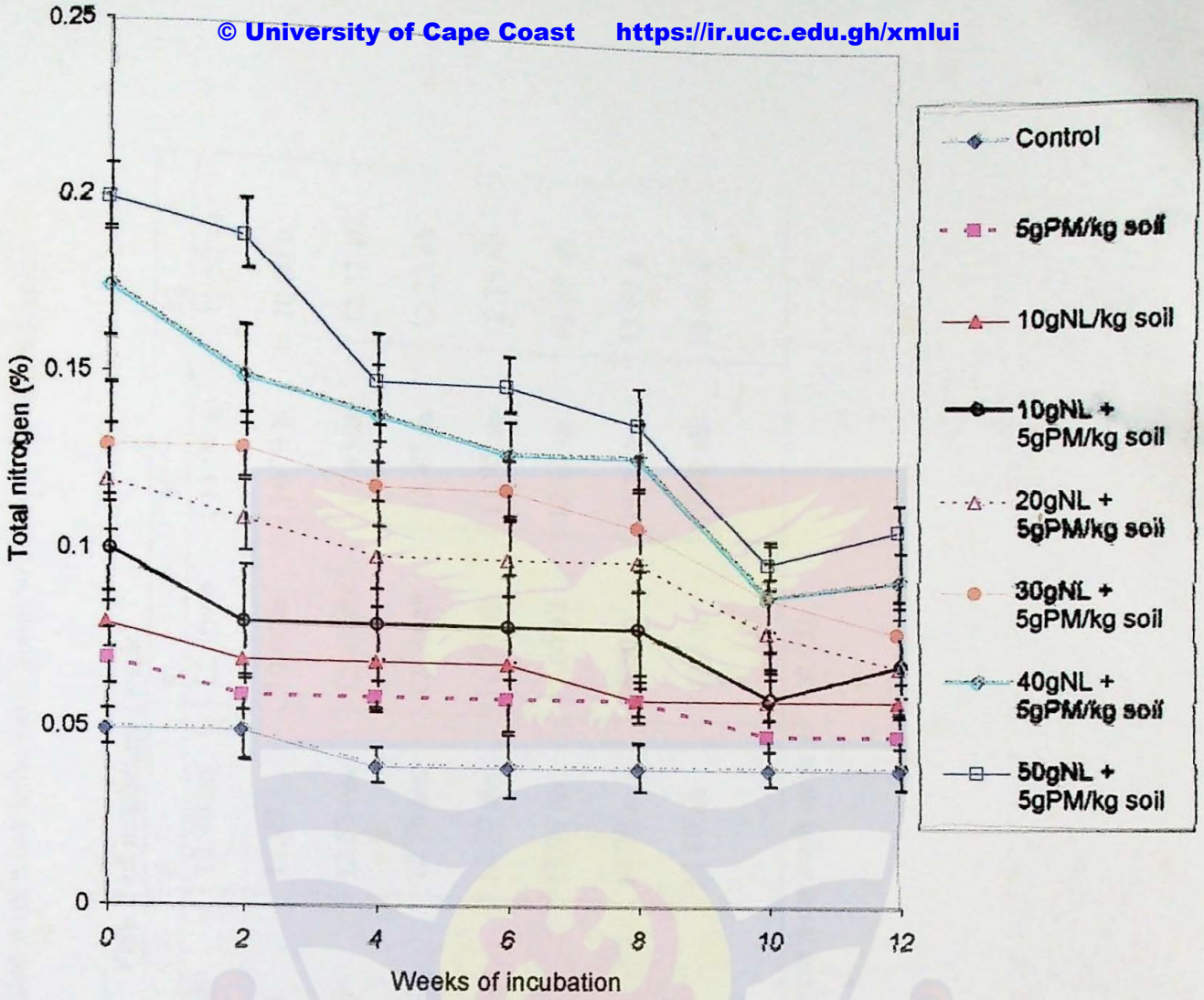

Figure 5.7. Changes in total N in unamended soil and soil amended with neem leaves and poultry manure as affected by incubation period (bars are standard errors)

Table 5.5. Changes of C:N ratio of unamended soil and soil amended with neem leaves and poultry manure over the incubation period .

Treatment	Period of incubation (week)						
	0	2	4	6	8	10	12
No amendment (Control)	17.00 abc	14.10 ab	12.90 ab	12.40 bc	12.10 ab	11.90 ab	11.50 ab
5 g PM/kg soil	14.57 c	11.67 b	11.10 b	11.00 c	10.90 b	10.70 b	10.60 b
10 g NL/kg soil	15.88 bc	13.60 ab	13.03 ab	12.85 abc	12.49 ab	12.45 ab	12.33 ab
10 g NL + 5 g PM /kg soil	15.90 bc	13.90 ab	13.00 ab	12.58 abc	12.50 ab	12.40 ab	12.33 ab
20 g NL + 5 g PM /kg soil	17.08 abc	14.80 a	13.33 ab	12.70 abc	11.90 ab	11.95 ab	12.85 ab
30 g NL + 5 g PM /kg soil	18.85 ab	15.90 a	14.80 a	13.90 ab	13.00 ab	12.80 ab	12.88 ab
40 g NL + 5 g PM /kg soil	19.61 a	16.70 a	15.50 a	14.90 a	14.60 a	14.50 a	13.68 a
50 g NL + 5 g PM /kg soil	19.25 a	15.50 a	15.60 a	14.80 ab	14.01 a	13.80 ab	13.60 a

Means with the same letter within columns are not significantly different from each other ($P \leq 0.05$).

PM= Poultry Manure
NL= Neem Leaf

ANOVA Tables – Appendices B100-B106 (pp 191 – 193)

5.3.4. Available phosphorus dynamics

Figure 5.8 shows the pattern of available phosphorus produced from the treatments. Nitrogen and phosphorus have been found to behave similarly during organic matter decomposition (Berg and McClaugherty, 1989), however, the pattern of available phosphorus released did not have the same pattern as NH_4^+ - N and NO_3^- - N (Figures 5.4 and 5.5 respectively). The peaks of available phosphorus production occurred at the 2nd week of incubation, and the concentration of the phosphorus remained constant almost at that level for all the treatments for the rest of study period. The peaks on the other hand conformed with the peak of production of NO_3^- - N for the sole poultry manure treatment.

The total phosphorus concentration in the neem leaves was far lower than that in the poultry manure (Table 5.2). The pattern of available phosphorus observed in Figure 5.8 was probably influenced by the addition of the poultry manure. This could be deduced from the differences between the phosphorus levels in the 10 g neem leaves/kg soil and 10 g neem leaves + 5 g poultry manure /kg soil treatments. The addition of poultry manure to the neem leaves at this level caused conspicuous rise in the available phosphorus level .

The poultry manure might have enhanced the release of phosphorus from the neem leaves, as poultry manure is known to have a variety of aerobic and anaerobic

bacteria for the breakdown of organic compounds (Bacharach, 1957). Neem leaves alone used as soil amendment did not bring any significant increase in the available phosphorus levels just as with the control at the end of the incubation period (Appendix A9).

In the application of neem leaves as soil amendment in crop production it is therefore advisable to include poultry manure to boost the level of available phosphorus in the soil.

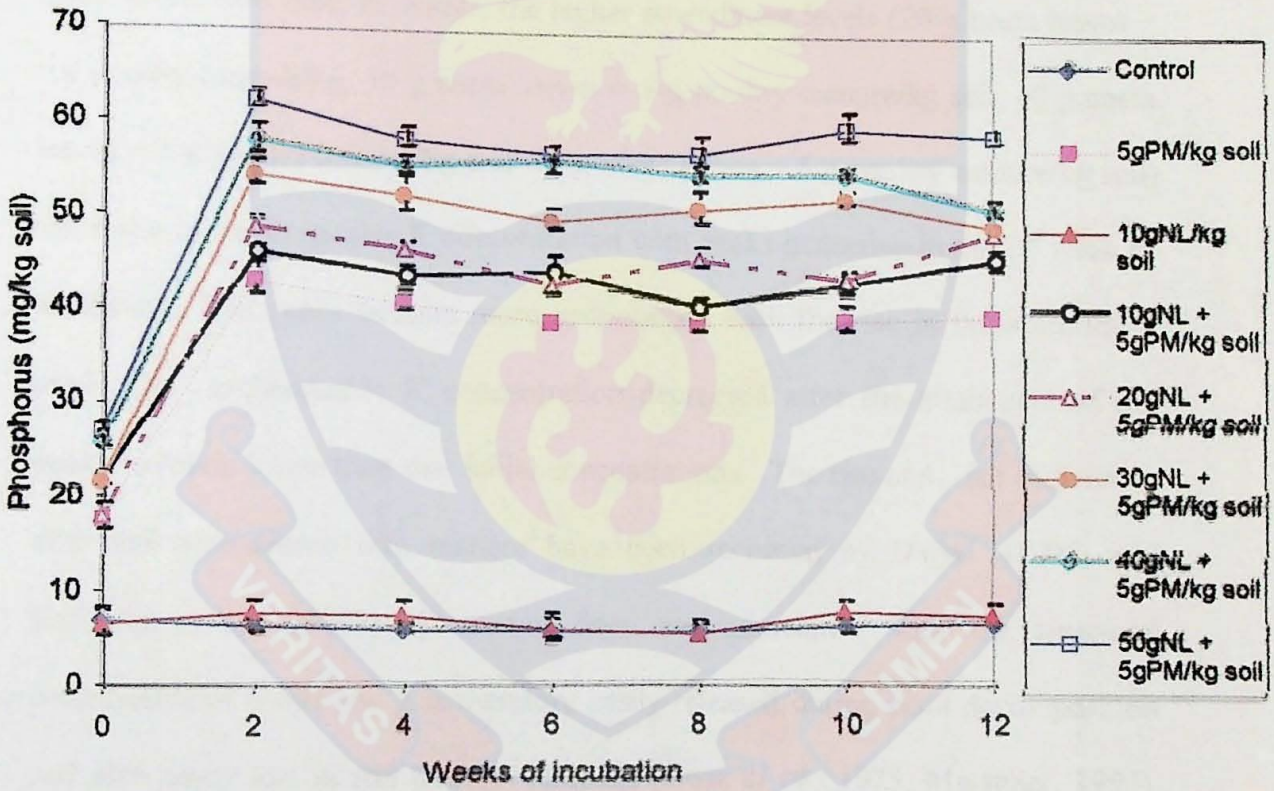

Figure 5.8. Changes in available phosphorus in unamended soil and soil amended with neem leaves and poultry manure as affected by incubation period (bars are standard errors)

5.3.5. Exchangeable potassium, calcium and magnesium

The production trends of K, Ca and Mg are presented in Figures 5.9, 5.10 and 5.11 respectively. Exchangeable K concentrations were higher for the amended soil than the control/unamended soil (Figure 5.9). The exchangeable K of the unamended soil remained at almost the same concentration throughout the study. For the lower levels of amendments (5 g poultry manure/kg soil, 10 g neem leaves/kg soil and 10 g neem leaves + 5 g poultry manure/kg soil), the concentration of exchangeable K fell gradually from the initial higher values to lower levels with time. However, the higher amendment levels (20 g neem leaves + 5 g poultry manure/kg, 30 g neem leaves + 5 g poultry manure/kg soil, 40 g neem leaves + 5 g poultry manure/kg and 50 g neem leaves + 5 g poultry manure/kg soil) had a rise in exchangeable K concentration with peaks occurring in the 2nd week of incubation. The peaks became more pronounced with the rise in the amendment level. The exchangeable K concentration decreased after the attainment of the peaks to levels lower than the initial concentrations. The rise and fall of K levels after soil amendment with manure have been recorded by Datta (1996) and Kalburtji *et al.*, (1997). Potassium does not associate with the structural components of plants and it is therefore easily released during plant decomposition and also easily lost in soil through leaching (Gosz *et al.*, 1973; Marshner, 1995), and thus probably accounted for the observed trends in Figure 5.9.

As in the preceding discussions, higher levels of the amendments corresponded with higher exchangeable Ca production (Figure 5.10). The highest production of Ca for the treatments also occurred in the 2nd week of incubation after which the nutrient's concentration declined.

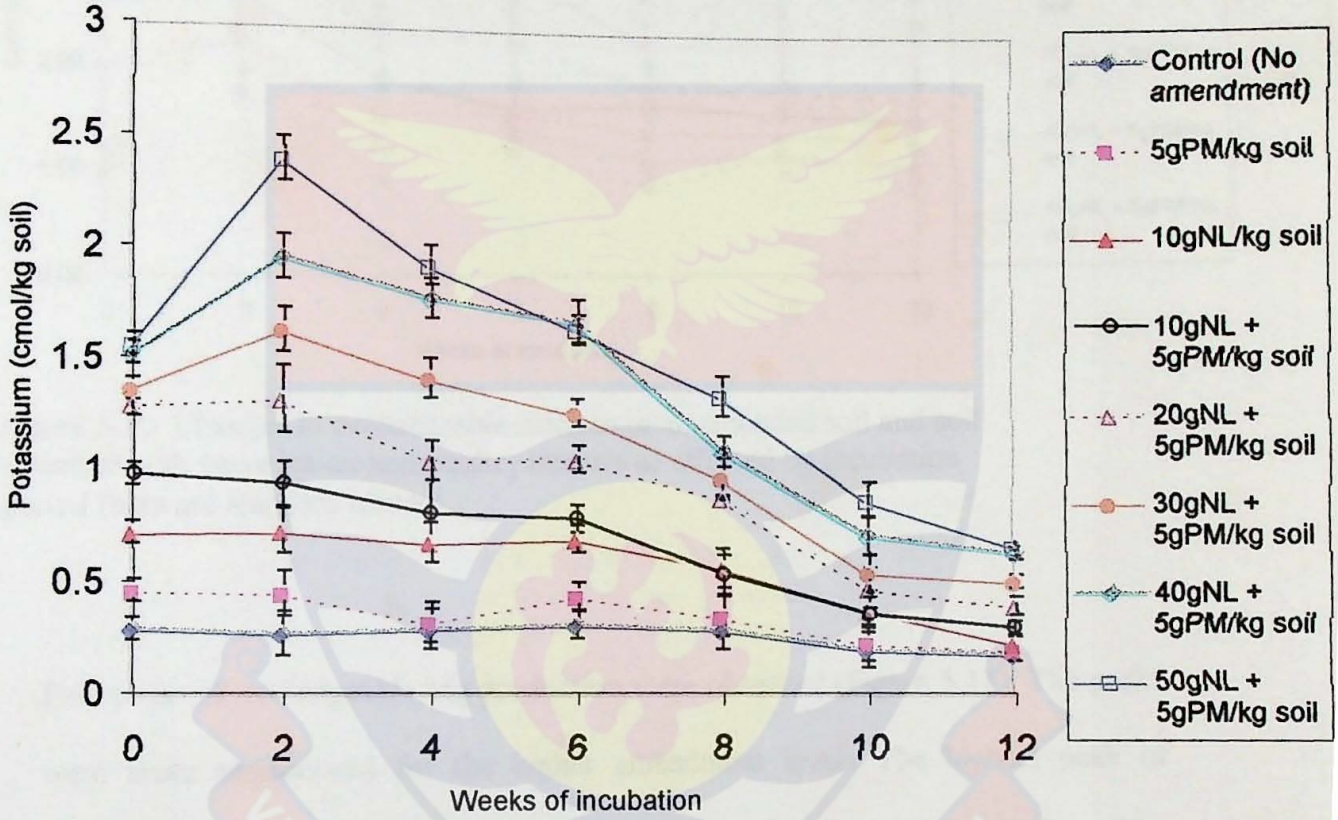

Figure 5.9. Changes in exhangeable potassium in unamended soil and soil amended with neem leaves and poultry manure as affected by incubation period (bars are standard errors)

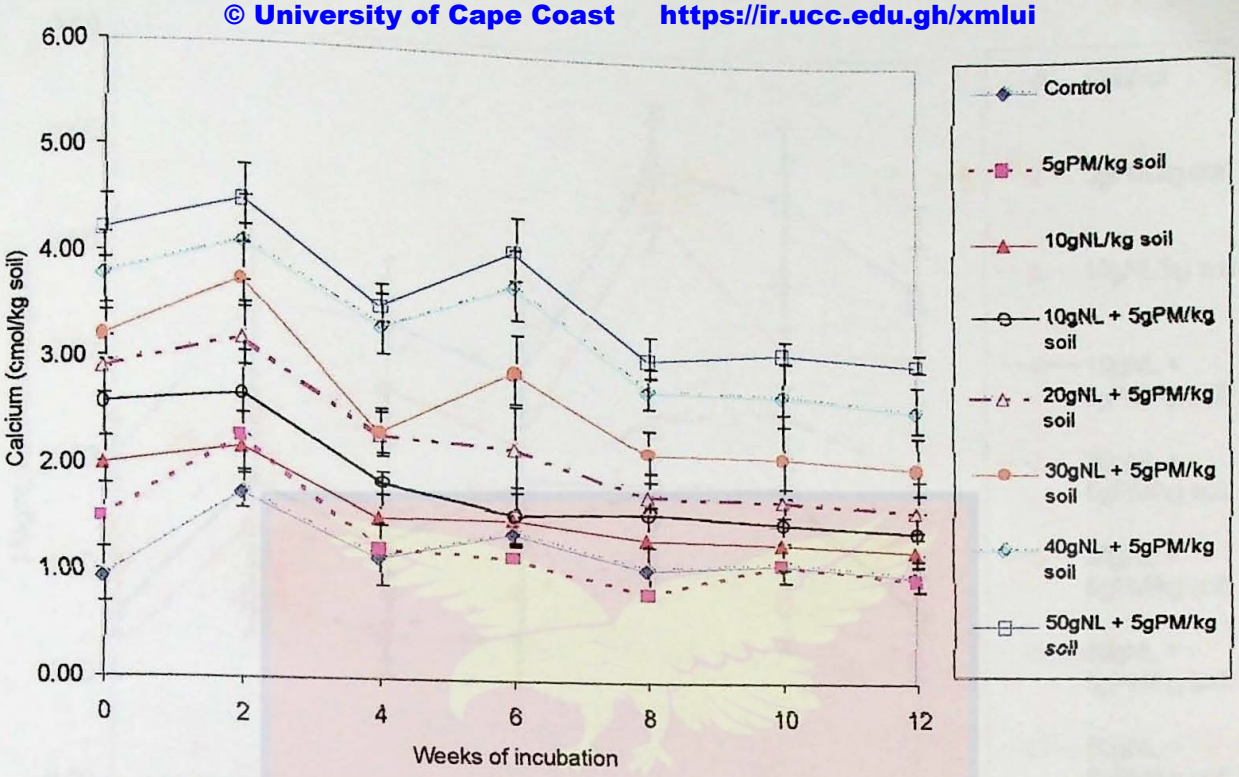

Figure 5.10. Changes in exchangeable calcium in unamended soil and soil amended with neem leaves and poultry manure as affected by incubation period (bars are standard errors)

Two peaks of exchangeable Mg production were observed (Figure 5.11). The peaks were more pronounced for the higher amendment levels. The highest peak of exchangeable Mg production occurred at the 8th week of incubation. The conditions pertaining at this week, described for NH_4^+ - N production, might have favoured maximum release for the Mg.

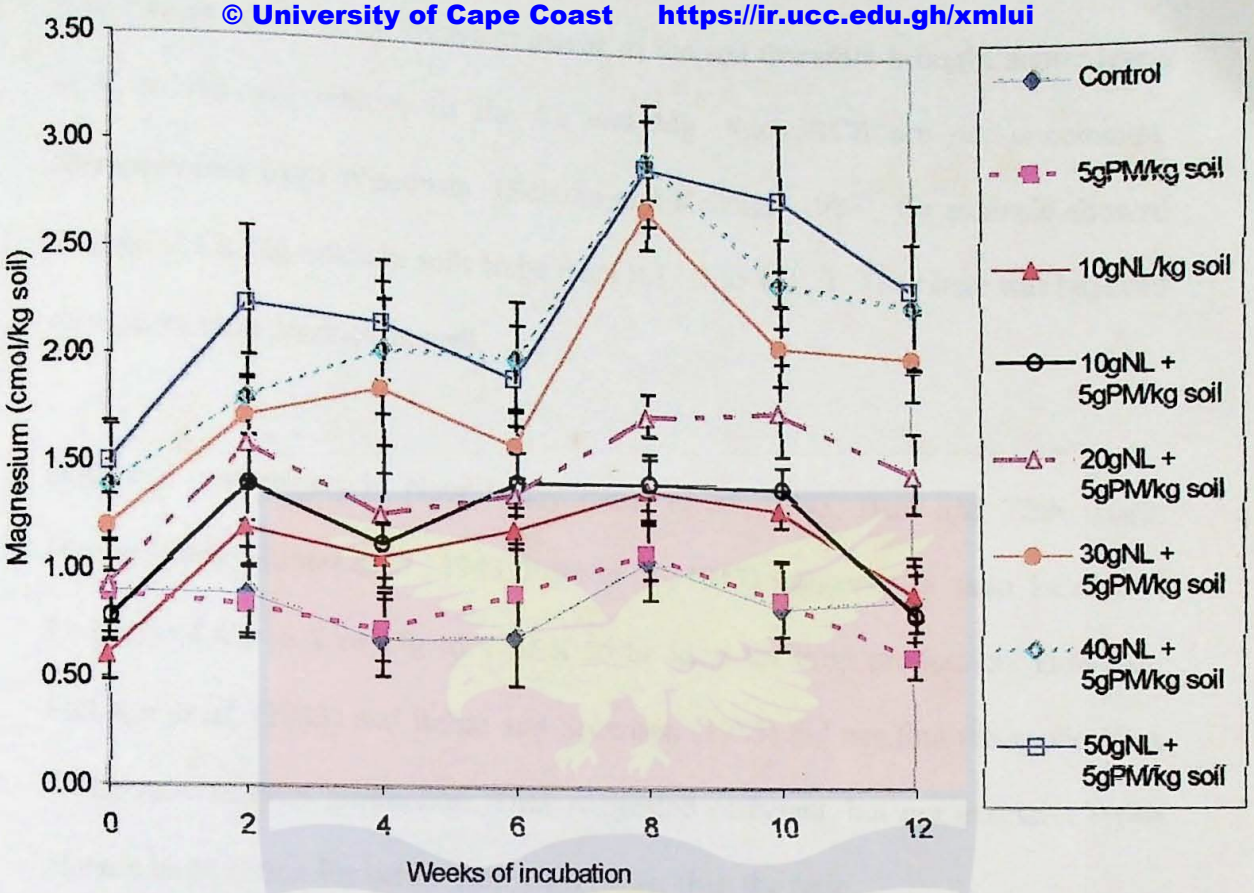

Figure 5.11. Changes in exchangeable magnesium in unamended soil and soil amended with neem leaves and poultry manure as affected by incubation period (bars are standard errors)

5.3.6. . Basic cation ratios (BCR) of amended soil

The neem leaves and the poultry manure in the amended soil significantly increased the Ca, Mg, and K levels in the soil (Appendix A9). The Ca, Mg, and K levels in the treatments at the end of the study are also shown in Table 5.6. The range the Ca:K ratio became closer as the amendment levels of the neem leaves increased. The K level in the neem leaves was found to be higher than the Ca and Mg (Table

5.2). More additions of the neem leaves to the soil therefore brought higher levels of K to the soil relative to the Ca and Mg. Such BCR are not uncommon. Measurements from Wisconsin (Schulte and Kelling, 1985), for example showed a range of Ca:Mg ratios in soils to be from 8.1 : 1 to 1.0 : 1. The range was believed to support crop production well.

Bear and co-workers in New Jersey (Bear *et al.*, 1945; Bear and Toth, 1948; Hunter, 1949; Hunter *et al.*, 1943; Price *et al.*, 1947) suggested a ratio balance of 13 parts of Ca to 2 of Mg to 1 of K to be ideal for crop production. However, McLean *et al.* (1983) and Rehm and Sorensen (1985) did not find the applicability of the ratio to crop production. They suggested sufficient, but not excessive levels of each basic cation for better crop yield rather than the ratio.

Table 5.6. Basic cation ratio (BCR) of treatments at the 12th week of incubation

Treatment	cmol$_c$ /kg soil			Ratio
	Ca	Mg	K	Ca : Mg : K
No amendment (Control)	1.04	0.90	0.18	5.8 : 5.0 : 1
5 g PM/kg soil	1.01	0.63	0.20	5.1 : 3.2 : 1
10 g NL/kg soil	1.29	0.93	0.23	5.6 : 4.0 : 1
10 g NL + 5 g PM /kg soil	1.44	0.82	0.30	4.8 : 2.7 : 1
20 g NL + 5g PM /kg soil	1.68	1.50	0.40	4.2 : 3.8 : 1
30 g NL + 5 g PM /kg soil	2.09	2.06	0.51	4.1 : 4.0 : 1
40 g NL + 5 g PM /kg soil	2.65	2.30	0.64	4.1 : 3.6 : 1
50 g NL + 5 g PM /kg soil	2.80	2.39	0.68	4.1 : 3.5 : 1

5.3.7. Soil pH

The pH changes in the various amendments are presented in Figure 5.12. The treatment levels of 30 g neem leaves + 5 g poultry manure/kg, 40 g neem leaves + 5 g poultry manure/kg and 50 g neem leaves + 5 g poultry manure/kg had a significant rise in pH above the unamended soil at the end of the incubation period (Appendix A9), with peaks occurring at the 2nd week of incubation. The pH fell thereafter with signs of stabilization from the 4th week onwards. The peaks corresponded with the peaks of NH_4^+ - N production and the peaks of production of the exchangeable cations. The high amounts of the basic cations and the NH_4^+ - N production at this period might have contributed to the rise in pH. The incorporation of organic matter with a subsequent rise in pH has been reported (Atallah *et al.*, 1995; Datta, 1996; Hong *et al.*, 2000; Pool *et al.*, 2000). No peaks

for pH were observed for the lower levels of amendment and the control, their pHs remained almost unchanged throughout the incubation period. However, a fall in pH even below the initial pH of the unamended soil was observed for the 5g poultry manure/kg soil, 10g neem leaves + 5g poultry manure/kg soil and 20g neem leaves + 5g poultry manure/kg soil at a point in time during the study. The fall in pH corresponded with the peak of NO_3^- - N production (Figure 5.5) due to the release of H^+ ions during the nitrification process.

The pattern of pH rise and fall may be attributed to the ammonification and nitrification of organic nitrogen in the treatments (Pocknee and Sumner, 1997). The release of cations at the lower levels of amendment might have been masked by the higher rate of nitrification which lowered the pH, with the reverse being the case for the higher levels of amendment.

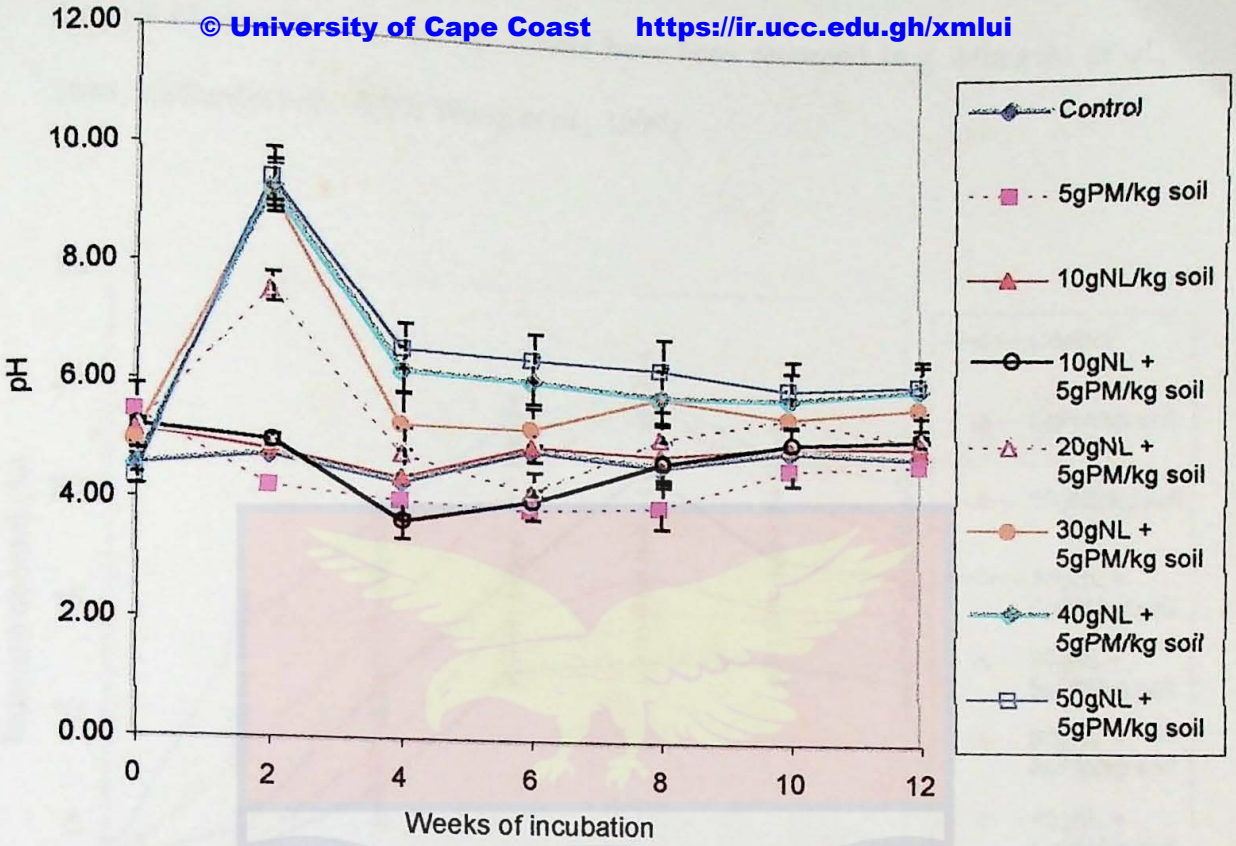

Figure 5.12. Changes in pH of soil amended with neem leaves and poultry manure as affected by incubation period (bars are standard errors)

5.3.8. Moisture contents of amended soil

The study started in February and ended in May 2003, a period with a rising level of rainfall in the study area. There was frequent rainfall at this time and no water was applied to treatments on the field. The moisture content increased with the increasing levels of amendment. All the amendments had higher moisture content above the control/unamended soil (Figure 5.13). The incorporation of the neem leaves and the poultry manure significantly increased the water holding status of

the soil (Appendix A3). Similar results have been recorded (e.g. Mbagwu *et al.*, 1994; Kalburtji *et al.*, 1997; Wong *et al.*, 1999).

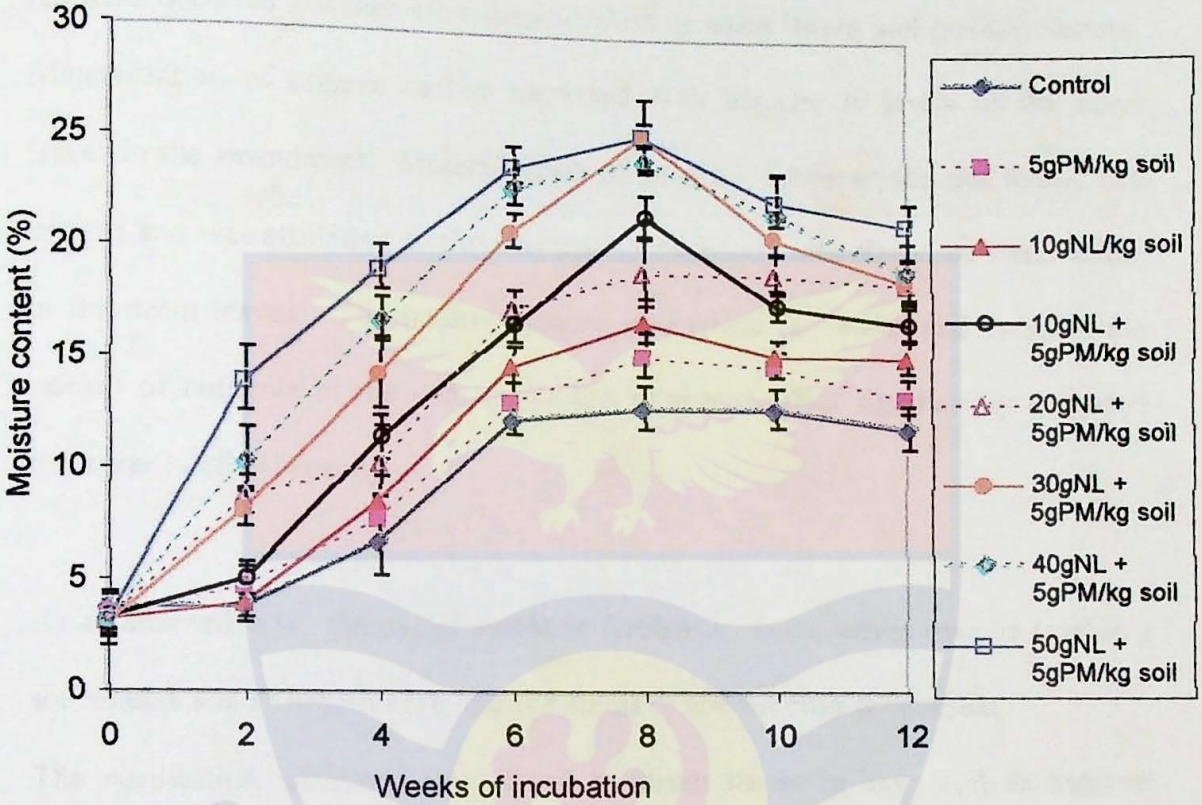

Figure 5.13. Influence of incorporated neem leaves and poultry manure on moisture content of soil (bars are standard errors)

5.4. Summary and conclusions

Soil nutrient levels increased with increasing levels of the neem leaves and the poultry manure added in a soil amendment. The highest levels of most of these soil nutrients occurred 2 weeks after incorporation of neem leaves and poultry manure. Mineralization of organic carbon increased with the rise in levels of the neem leaves in the amendment. Mineralization of nitrogen, however, did not follow this pattern and was attributed to the nitrification inhibitory role played by azadirachtin in the neem leaves. The poultry manure was found to boost and enhance the release of nutrients in the soil, whilst the combined effect significantly increased the water holding capacity.

As an alternative to the use of synthetic fertilizers, neem leaves may be used in a soil amendment to improve the nutrient status of soil for crop production.

The nitrification inhibitory role played by neem leaves in any soil amendment would most likely control the excessive release of nitrates in soil and thus prevent leaching and environmental pollution of underground waters.

The addition of poultry manure to the neem leaves in the amendment would enhance the release of the nutrients from the neem leaves, and also boost the nutrient levels in the soil especially phosphorus, which was found to be low in the neem leaves.

CHAPTER SIX

6.0 To test the validity of the soil amendment by investigating its impact on growth characteristics of a test crop (carrot) using pot experiment

6.1. Introduction

The use of neem leaves and poultry manure in a soil amendment has been observed in Chapters 4 and 5 to favourably suppress growth of nematodes and provide some plant nutrients. The adoption of such organic sources in place of synthetic pesticides and fertilizers would help check the degradation of the environment by these synthetic substances.

Though inorganic chemicals are known to be hazardous to the environment, they have brought positive gains in agriculture (Hemeng, 1980; Johnston *et al.*, 1994; Johnston *et al.*, 1995; Sharma and Sharma, 1995; Sultan *et al.*, 1995). Man is therefore faced with the critical dilemma of consistently obtaining high crop yields without polluting soil, air, and water, and without declining soil fertility.

Plant-parasitic nematodes, important pests in agriculture which cause loss of yield and quality of most food and fiber crops, have been controlled without the use of inorganic chemicals on certain crops but by the use of organic amendments (Khan *et al.*, 1974; Alam *et al.*, 1979; Mian and Rodriguez-Kabana, 1982; Sarathchandra *et al.*, 1996). The organic amendments in addition to the pest control also increase the fertility status of the soil.

Carrots are the most sensitive crop to root-knot nematodes, and yield reduction of 45% in commercial fields has been recorded in the United States (Widmer *et al.*, 2001). Other yield loses on this crop as a result of nematodes are also known (Greco *et al.*, 1993; Guyton *et al.*, 1989; Schiliro *et al.*, 1995). Inorganic pesticides have proved effective in the control of these nematodes on carrots (Johnston *et al.*, 1994; Johnston *et al.*, 1995), however, the associated problems attached to the use of these chemicals call for an alternative, that is, natural products which will not only control nematodes but also add nutrients to the soil to enhance the yield of carrot.

The objective of this current study therefore was to evaluate the impact of neem leaves and poultry manure in a soil amendment on soil nutrients, root-knot on carrot and growth of carrot. Some treatments (check section 6.2.1) of the previous study (Chapter 5) were also used for the current study.

6.2. Materials and methods

The experimental site, properties of the soil used for the experiment and preparation of treatment materials are the same as in Chapter 3.

6.2.1. Treatments and experimental design

Table 6.1. Treatments applied to pots

Treatment (per kg soil)
No amendment (Control)
5 g PM
10 g NL
10 g NL + 5 g PM
30 g NL + 5 g PM
50 g NL + 5 g PM

PM = Poultry manure
NL = Neem leaves

Treatments were placed in plastic buckets, with a carrying capacity of 7 kg of soil. Each treatment was replicated three times. The treatments were placed under partial shade, that is, on the veranda of the laboratory at the Technology Village of the University of Cape Coast. The completely randomized design (CRD) was used.

6.2.2. Sowing of carrot seeds

The carrot was sown on the 22nd of April, 2003, two weeks after incubation of treatments. Thinning to 15 plants per pot was done one and half weeks after germination.

6.2.3. Sampling of soil and carrot for analyses

The carrots were harvested three months after sowing. The carrot roots were carefully scooped out of the soil to avoid destruction of the rootlets. The soil attached to the root was gently washed off to expose the root-knots. The number of root-knots per plant was counted and the root-knot Index was calculated following the method of Taylor and Sasser (1978).

The roots and leaves of the carrot were oven dried at 40°C for 5 days, ground, and the total nitrogen, phosphorus and potassium determined (Check Chapter 5, section 5.2.2 for methods).

Soil from the pots were spread on paper sheet, samples were then taken through the quarter method. Care was taken to remove all plant materials. Samples were immediately used for analysis. The total nitrogen, ammonium-N, nitrate-N, total phosphorus, total potassium, available phosphorus and the exchangeable potassium of soil of the treatments were determined (Check Chapter 5, section 5.2.2 for methods).

The ash contents of roots and leaves of carrot of the individual treatments were determined by oven drying 1 g sample in crucibles at 105 °C over night, cooled in a desiccator, weighed and placed in a muffle furnace at 500 °C over night. The samples were again cooled in a desiccator and samples reweighed (Stewart *et al.*, 1974). The per cent ash was calculated expressed on oven dry weight.

The data were subjected to analysis of variance (ANOVA) and the Duncan's Multiple Range Test for the separation of means using the MSTAT- C statistical software (Russell, 1990). The line of best fit in Microsoft Excel Spread Sheet and Minitab version 11.21 were employed to find the relationships (R^2) between the soil nutrient content and that in the carrot. All relevant ANOVA Tables are found in Appendix B.

6.3. Results and Discussion

6.3.1. Nutrient status of the soil at harvest of carrot

All the amended soil samples recorded significantly higher nutrient levels than the unamended soil (Table 6.2). In most cases the nutrient levels also significantly corresponded with the inclusion rates of the neem leaves and poultry manure in the amendment. The pattern of the nutrient changes in the treatments was almost the same as in the previous study, since some of the same treatment combinations were used in this study. However, the nitrification inhibitory role supposed to be played by the neem leaves was not portrayed in this study, as the increasing levels of the neem leaves in the amendment had no reducing effect of the NO^{-}_{3} – N measured. This might be due to the time of analysis (about 14 weeks after incubation of treatments) of the treatments in the present study, which was far beyond the observable effect of nitrification inhibition noted before the 8^{th} week of incubation of treatments in the preceding study.

There were slight increments in pH (0.18 – 0.65) with increase in the inclusion rates, however, the increments were not significantly different (Table 6.2). This was also noted in the previous study. Soil amendment with organic materials are known to increase soil pH (Okeleye and Adetunji, 1999; Pool *et al.*, 2000)

The combination of 10 g NL/kg soil, 30 g NL/kg soil and 50 g NL/kg soil with a constant amount of poultry manure (5 g PM/kg soil) brought significant increases

in the total nitrogen, total phosphorus, NO_3 - N, NH_4^+ – N, available phosphorus and exchangeable potassium in the soil.

The sole 10 g NL/kg soil and the sole 5 g PM/kg soil treatments also increased the nutrient status of the soil, however, their combinations gave higher levels (Chapter 5). Poultry manure when used as soil amendment has had positive influence on soil fertility (Oikeh and Asiegbu, 1993; Abdel Magid *et al.*, 1995; Obi and Ebo, 1995; Larney and Janzen, 1996,). The neem leaf is not only pesticidal but also a good soil nutrient provider when incorporated into the soil. Increases in soil fertility status have been recorded with the use of neem leaves and other plant materials as soil conditioners in Burkina Faso with the neem leaves giving better results (Tilander and Bonzi, 1997). The neem leaves have been used by Indian farmers over the years to enrich the soil (Neem Foundation, 1997). The quantity of organic material used for soil amendment has been found to impact a proportional rise in the nutrient levels of the soil (e.g. Mbagwu *et al.*, 1994; Wong *et al.*, 1999) as has been observed in the current study.

6.3.2. Nutrient contents of carrot at harvest.

The total N, P, K and the % Ash content in both the carrot leaves and roots increased with increasing rates of the neem leaves and poultry manure in the amendment (Table 6.3). The differences in nutrient levels in both the carrot leaves and roots were significant among the treatments.

Table 6.2. Nutrient contents and pH of unamended and amended soil at harvest of carrot

Treatment / Nutrient	N	K	P	NH_4^+ – N	NO_3^- – N	AvaiL. P	Exch. K (cmol$_c$/kg soil)	pH
	%			Mg/kg soil				
No amendment (Control)	0.05 d	0.07 a	0.03 c	2.70 c	12.00 f	12.00 f	0.14 e	4.95 b
5 g PM/kg soil	0.08 c	0.07 a	0.06 ab	6.40 b	13.50 e	28.90 d	0.16 d	5.13 b
10 g NL/kg soil	0.08 c	0.08 a	0.05 b	7.15 a	17.00 d	15.30 e	0.18 c	5.20 b
10 g NL + 5 g PM/kg soil	0.08 c	0.08 a	0.05 b	6.15 b	23.50 c	42.50 c	0.20 b	5.20 b
30 g NL + 5 g PM/kg soil	0.11 b	0.08 a	0.06 ab	7.80 a	45.00 b	44.10 b	0.21 b	5.30 ab
50 g NL + 5 g PM/kg soil	0.14 a	0.09 a	0.07 a	7.65 a	61.12 a	59.60 a	0.31 a	5.60 a

Means within columns with the same letters are not significantly different ($P \leq 0.05$).
NL = Neem leaves
PM = Poultry manure

ANOVA Tables – Appendices B107-B114 (pp 193 – 195)

Table 6.3. Nutrient and ash contents in carrot leaves and roots at harvest

Treatment / Nutrient	Carrot Leaves				Carrot Roots			
	N	K	P	Ash	N	K	P	Ash
	%							
No amendment (Control)	1.52 c	1.18 c	0.19 e	12.08 d	0.44 e	0.72 e	0.15 e	4.15 e
5 g PM/kg soil	1.66 c	1.15 c	0.31 bc	12.29 d	0.49 e	0.71 e	0.26 b	4.40 d
10 g NL/kg soil	1.83 c	1.30 bc	0.24 de	13.77 c	0.57 d	0.74 d	0.22 d	4.96 c
10 g NL + 5 g PM/kg soil	2.10 bc	1.38 b	0.27 cd	14.57 b	0.73 c	0.85 c	0.24 c	5.15 c
30 g NL + 5 g PM/kg soil	2.70 ab	1.59 a	0.33 b	17.06 a	0.92 b	0.94 b	0.27 b	5.98 b
50 g NL + 5 g PM/kg soil	2.92 a	1.68 a	0.40 a	17.46 a	0.98 a	1.01 a	0.29 a	6.39 a

Means within columns with the same letters are not significantly different ($P \leq 0.05$).
NL = Neem leaves
PM = Poultry manure

ANOVA Tables – Appendices B115-B122 (pp 196 – 198)

There was a general strong positive relationship ($P<0.05$) between the nutrient content in the soil and in the carrot. Figures 6.1 – 6.3 show this relationship (R^2) – as the per cent nutrient in the soil increased with the increasing amendment levels (x-axis), the nutrient content in the carrot leaves/roots (y – axis) also relatively increased. Such observations of the quantity of nutrients in the soil having a positive linear impact on the amount in the plant have also been reported by Manson *et al.*, (2002). The accumulation of the nutrients was higher in the leaves than in the roots (Figures 6.1 – 6.3). Most nutrients are found in higher quantities in the leaves than in the other parts of the plant (Buwalda and Smith, 1987; Sanginga *et al.*, 1994; Li *et al.*, 1998; Harrison *et al.*, 2000). The leaves are the production centres of food (photosynthesis) for the plant and obviously their nutrient concentrations are expected to be higher than in all the other parts of the plant.

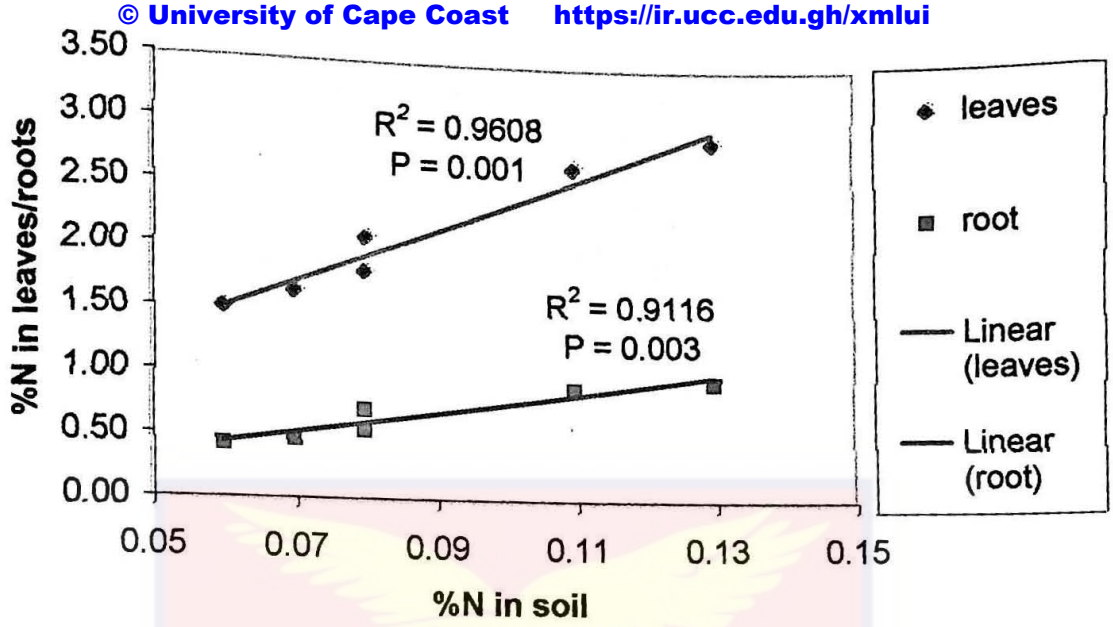

Figure 6.1. Relationship between %N in soil and %N in carrot leaves and roots

ANOVA Tables for the Fitted line Plots – Appendices B128-B129 (pp 200)

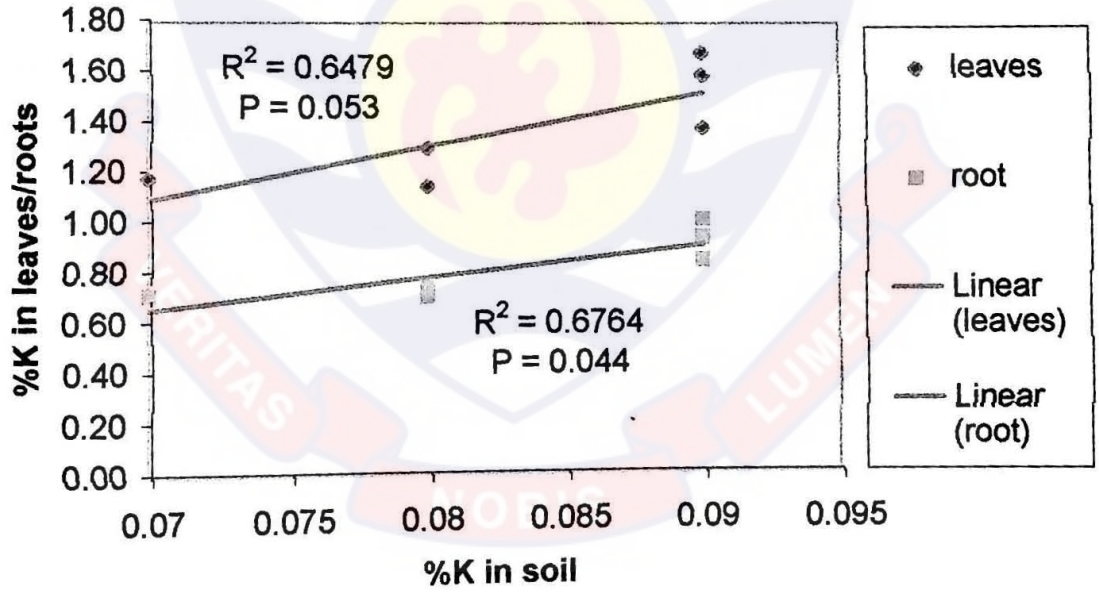

Figure 6.2. Relationship between %K in soil and %K in carrot leaves and roots

ANOVA Tables for the Fitted line Plots – Appendices B130-B131 (pp 201)

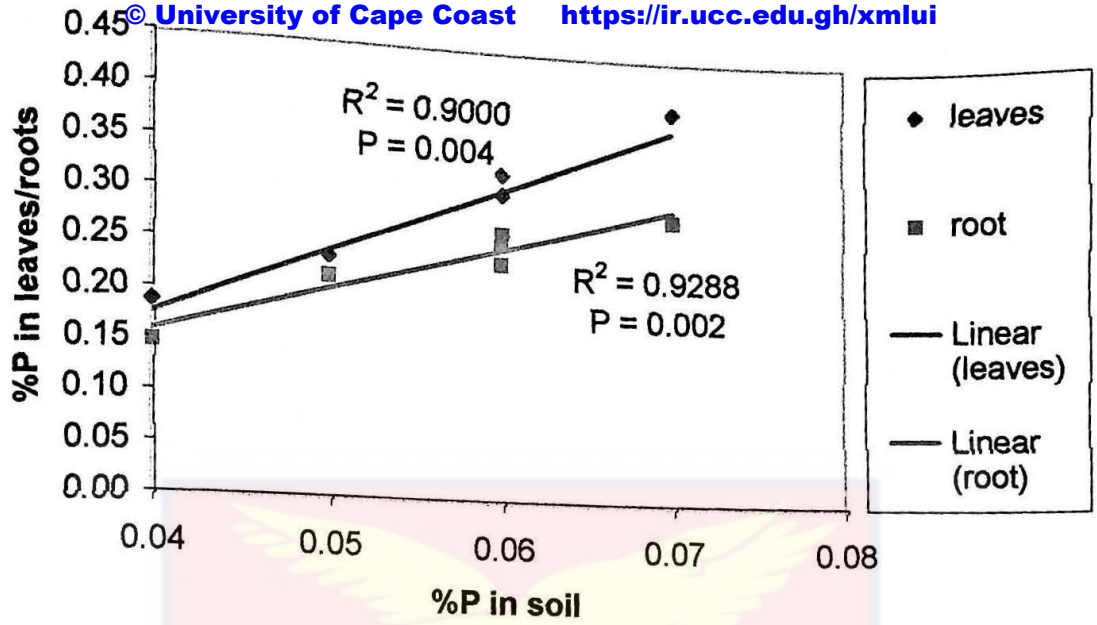

Figure 6.3. Relationship between %P in soil and %P in carrot leaves and roots

ANOVA Tables for the Fitted line Plots – Appendices B132-B133(pp 201)

Soil amendments with organic materials increase the level of nutrients in soils and correspondingly also increase the level of the nutrients in the plants growing on them (Hafner *et al.*, 1993; Schulthess *et al.*, 1997; Genda *et al.*, 2000; Singh and Bhati, 2003). The general nutrient levels of the carrot plant improved with the soil amendments as indicated by the higher ash contents in the carrot grown on the amended soil than on the unamended soil (Table 6.3).

6.3.3. Soil amendments and the control of root-knot nematodes of carrot

The soil amendments did not only provide soil nutrients with corresponding increases in the carrot plant but also had an impact on root-knot nematodes of the plant. Table 6.4 represents the root-knot index of carrot recorded under the soil treatments. The root-knot index reduced as the rate of neem leaves increased in the amendment. The carrot on the unamended soil and the soil amended with 10 g NL/kg soil gave the highest root-knot indices of 4.30 and 4.50 respectively, and were not significantly different from each other. The sole poultry manure treatment had a count of 2.30, and proved to be better in the control of the root-knot nematodes than the unamended soil, 10 g NL/kg soil and 10 g NL + 5 g PM/kg soil. Plates 1, 2, 3, 4, 5 and 6 give a picture of the effectiveness of the soil treatments on carrot root-knot nematodes. The effectiveness of poultry manure in controlling parasitic nematodes in other crops has been reported (Kaplan and Noe, 1993; Riegel *et al.*, 1996). The present study has also revealed the suppressive effect of the poultry manure on parasitic carrot nematodes. The ability of organic amendments especially poultry manure in the suppression of nematodes has been assigned to chemical by-products from the decomposing materials in the soil which are toxic to the nematodes (Dunn, 1994).

Table 6.4. Impact of soil amendment on root-knot index of carrot

Treatment	Root-knot index
No amendment (Control)	4.30 a
5 g PM/kg soil	2.30 c
10 g NL/kg soil	4.50 a
10 g NL + 5 g PM/kg soil	3.10 b
30 g NL + 5 g PM/kg soil	1.50 d
50 g NL + 5 g PM/kg soil	0.00 e

Means with same letters are not significantly different ($P \leq 0.05$).
NL = Neem leaves
PM = Poultry manure

ANOVA Tables – Appendices B123 (pp 198)

Neem products are noted for their effectiveness in controlling nematodes. The neem leaves have proved effective in previous studies on other crops (Wani, 1992; Reddy *et al.*, 1993; Deka and Rahman, 1998; Nanjegowda *et al.*, 1998). The 10 g NL/kg soil was not effective in controlling the nematodes, probably due to the low concentration of azadirachtin. The addition of the poultry manure to the 10gNL/kg soil was more effective in suppressing the nematodes. The subsequent drastic

decline of the root-knot index with increasing levels of the neem leaves combined with the constant amount of poultry manure in the soil treatment, may be assigned to the neem's nematicidal effect. The 50 g NL + 5 g PM/kg soil was the most effective soil amendment in the current study in controlling root-knot nematodes of carrot.

6.3.4. Yield characteristics of carrot

The mean root weight and shoot weight per plant varied among soil treatments (Table 6.5). The amended soils had heavier significant mean root weight and shoot weight per plant than the unamended soil. Higher levels of amendment gave corresponding higher mean root weight and shoot weight per plant. The mean root length per plant for the amended soils were significantly longer than the unamended soil. The mean root circumference per plant grown on the amended soils were also significantly larger than those grown on the unamended soil. Higher levels of amendments resulted in longer root length and larger root circumference of the carrot.

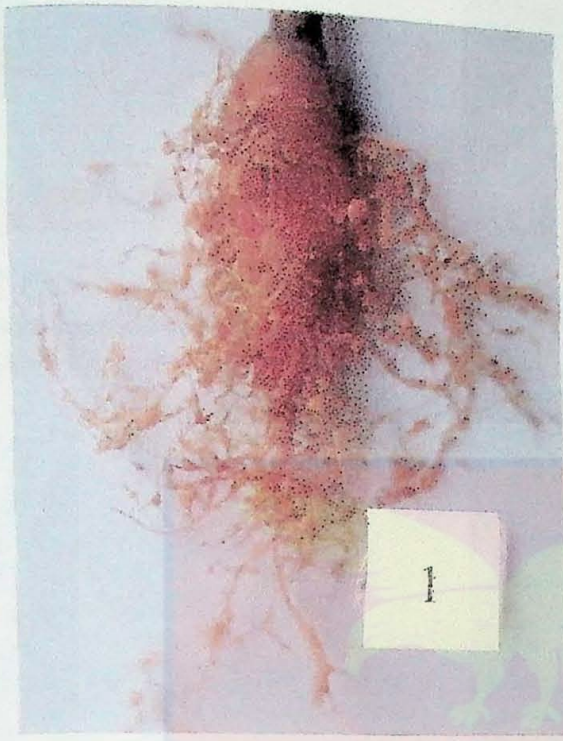

Plate 1. Carrot with root-knots from unamended soil

X 0.5

Plate 2. Carrot with root-knots from soil amended with 5 g PM/kg soil

X 0.5

Plate 3. Carrot with root-knots from soil amended with 10 g NL/kg soil

X 0.5

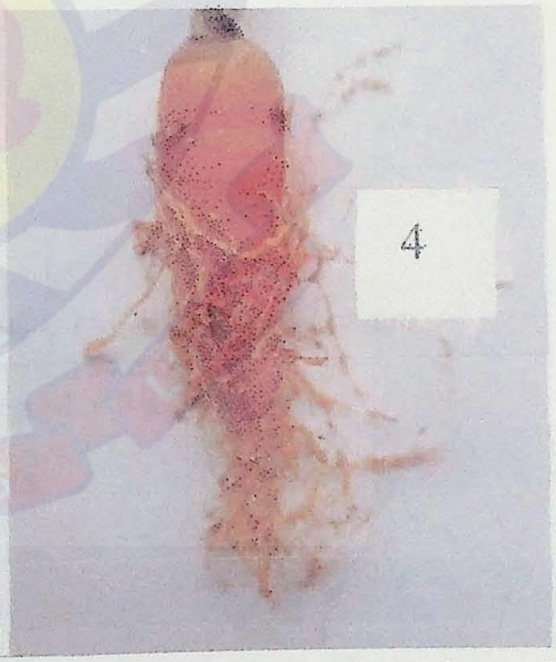

Plate 4. Carrot with root-knots from soil amended with 10 g NL + 5 g PM/kg soil

X 0.5

Plate 5. Carrot with few root-knots from soil amended with 30 g NL + 5 g PM/kg soil

X 0.5

Plate 6. Carrot with no root-knots from soil amended with 50 g NL + 5 g PM/kg soil

X 0.5

All the yield characteristics of the carrot improved relatively with the rising levels of the amendments, and these tended to follow the changes in nutrient level of the soil treatments and in the carrot plant (Tables 6.2 and 6.3). The control of nematode with the soil amendments might have also played a role in the improvement of the yield characteristics, however, the soil fertility impact of the amended soil seems to be stronger.

Table 6.5. Effect of soil amended with neem leaves (NL) and poultry manure (PM) on yield characteristics of carrot

Treatment / Yield Characteristics	Mean root length/plant (cm)	Mean circumference of root /plant (cm)	Mean root weight/plant (g)	Mean shoot weight/plant (g)
No amendment (Control)	7.48 d	6.60 d	8.90 f	4.00 f
5 g PM/kg soil	9.20 c	9.00 c	22.80 e	6.05 e
10 g NL/kg soil	8.96 c	9.80 b	28.70 d	9.90 d
10 g NL + 5 g PM/kg soil	9.88 b	10.15 b	29.80 c	11.30 c
30 g NL + 5 g PM/kg soil	10.10 b	10.80 a	32.50 b	13.30 b
50 g NL + 5 g PM/kg soil	10.94 a	11.25 a	34.20 a	15.20 a

Means with same letters are not significantly different ($P \leq 0.05$).
NL = Neem leaves
PM = Poultry manure

ANOVA Tables – Appendices B124-B127 (pp 199 – 200)

6.4. Summary and conclusions

Soil amended with neem leaves and poultry manure increased the nutrient status of the soil and correspondingly also improved the nutrient content of carrot growing in them.

The amendments had nematicidal effect on root-knot nematodes of carrot. Increasing the rate of the neem leaves in the amendment correspondingly reduced the root-knot nematodes of carrot. The poultry manure also showed high nematicidal effect on root-knot nematodes of carrot.

The yield of carrot improved significantly with the use of a soil amendment.

Neem leaves and poultry manure could be used in place of synthetic pesticides and fertilizers to improve the fertility status of the soil and the growth and yield of plants, as well as the ultimate reduction in environmental pollution as a result of the use of synthetic compounds.

CHAPTER 7

7.0. Summary, conclusions and recommendations

7.1 Summary

Field and pot experiments were carried out at the Technology Village of the University of Cape Coast. The site was of a coastal savanna environment, with soils belonging to the Haplic Acrisols.

It was found out that contrary to hypothesis '1', soil amended with neem leaves and the animal manure significantly reduced plant-parasitic nematodes. Populations of plant-parasitic nematodes, *Meloidogyne, Scutellonema, Pratylenchus, Paratricodorus* and *Tricodorus* in the soils amended with neem and the neem based materials were significantly lower than the control and the soils amended with sole poultry manure and cow dung treatments. The plant-parasitic nematodes, *Helicotylenchus, Criconemella* and *Rotylenchus* which were few in number in the control and the soils amended with sole poultry manure and cow dung were virtually absent in the neem and the neem based soils. The two inclusion rates of neem leaves, 50 g and 100 g/kg soil and their combinations with the poultry manure and the cow dung were not significantly different in the control of plant-parasitic nematodes. The neem and the neem based amendments were more effective than the sole poultry manure and the cow dung amendments in the control of the plant-parasitic nematodes. On the contrary, the populations of the non plant-

parasitic nematodes increased with the application of the amendments. Higher neem and neem based treatments promoted the proliferation of the non plant-parasitic nematodes.

The amendment significantly influenced the breakdown of azadirachtin A in the soil, rejecting hypothesis '2'. The half lives of azadirachtin A decreased with increasing rate of the neem leaves in the amendment. The addition of animal manure enhanced the breakdown of azadirachtin A in the soil. The Gas Chromatography (GC) was successfully used for the breakdown studies of the azadirachtin A in the soil.

Contrary to hypothesis '3', the nutrient levels in the soil were significantly increased by the neem leaves and the poultry manure application to soil. Soil chemical nutrients (organic carbon, total nitrogen, NH_4^+-N, NO_3^--N, available phosphorus, exchangeable K, Ca and Mg) increased with increasing levels of the neem leaves and the poultry manure in the soil. The combination of poultry manure and the neem leaves in the amendment enhanced the release of nutrients in the soil. The peak of mineralisation of most of the soil nutrients seemed to have occurred 2 weeks after incorporating neem leaves and poultry manure into the soil.

Mineralization of organic carbon increased with the rise in levels of the neem leaves in the amended soil, and thus contradicting hypothesis '4'. Mineralization

of nitrogen in the amended soil, however, did not follow the same pattern as the carbon. The higher rates of neem leaves rather seemed to inhibit nitrification.

Increasing the rate of neem leaves in the amendment correspondingly reduced the root-knot nematodes of carrot. The poultry manure also showed high nematicidal effect on root-knot nematodes of carrot. With the check of nematodes and the improved fertility levels of the soil, the growth of carrot was therefore, improved by the neem – poultry manure amendment, in contrast to hypothesis '5'.

7.2. Conclusions

The study has produced some results of practical significance which can contribute to soil fertility maintenance and nematode control. The following conclusions may therefore be drawn:

Neem leaves used as manure may provide valuable source of soil nutrients. The combination of poultry manure and neem leaves in a soil amendment may boost and enhance the release of nutrients and thus improve soil fertility. The peak of release of nutrients from such a soil amendment occurred after the 2nd week of incubation, and this may serve as a guide to plant crops in order to benefit from any such amendment.

The inhibitory effect of azadirachtin in the neem leaves on nitrification may lead to slow release of NO_3^-- N in the soil to satisfy the N requirement of crops. The

check of the excessive release of NO_3^- - N may slow down nitrate leaching and its associated environmental pollution of underground waters.

Neem leaves combined with poultry manure may effectively control plant-parasitic nematodes and prevent root-knot formation on carrot.

The active ingredient of neem, azadirachtin A, may stay shorter in the soil with higher application rate of neem leaves and the addition of animal manure.

The High Pressure Liquid Chromatography (HPLC) has been accepted so far as the only analytical method for the quantification of azadirachtin A, however, the current study has shown the possibility of using Gas Chromatography (GC) in analytical studies of azadirachtin A.

7.3. Recommendations

1. The breakdown of azadirachtin in the soil using seeds of neem instead of the leaves should be carried out for comparison.

2. The study of the breakdown of the other neem limonoids should also be carried out since they all support the pesticidal action of azadirachtin.

3 The impact of the soil amendment adopted in this study may be assessed on other vegetables.

4. Multi-locational studies are also recommended to assess the effect of different agro-ecological conditions on the selected parameters.

REFERENCES

Acea, M. J. and Carballas, T. (1996). Microbial response to organic amendments in a forest soil. *Bioresource Technology* **57**, 193-199.

Abdel Magid, H. M., Sabrah, R. E. A., Rabi, R. K., El Nadi, A. R. H. and Abdel Al, S. I. (1993). Biodegradation of municipal refuse and chicken manure in a winter-wheat ecosystem in Saudi Arabia. *Journal of Arid Environments* **25**, 411 – 419.

Abdel Magid, H.M., Abdel Aal, S. I., Rabie, R. K.and Sabrah, R. E.A. (1995). Chicken manure as a biofertilizer for wheat in the sandy soils of Saudi Arabia. *Journal of Arid Environment* **29**, 413-420.

Akhtar, M. (1999). Plant growth and nematode dynamics in response to soil amendments with neem products, urea and compost. *Bioresource Technology* **69**, 181 – 183.

Akhtar, M. and Malik, A. (2000). Roles of organic soil amendments and soil organisms in the biological control of plant parasitic nematodes. A review. *Bioresource Technology* **74**, 35 – 47.

Alam, M. M., Khan, A. M. and Saxena, S. K. (1979). Mechanism of control of plant parasitic nematodes as a result of the application of organic amendments to the soil. V-Role of phenolic compounds. *Indian Journal of Nematology* **9**, 136-142.

Ali, S. S. (1995). Management of *Meloidogyne javanica* through organic amendment in mungbean. *Indian Journal of Pulses Research* **8**, 204 – 205.

Anderson, J. M. and Ingram, J. S. I. (eds.) (1989). *Tropical soil biology and fertility*: A handbook of methods. CAB International, Wallingford, UK.

Asamoa, G. K. (1973). Soils of the proposed farm site of the University of Cape Coast. *Technical Report* **88**, 28.

Ashgar, M. and Kanehiro, Y. (1980). Effects of sugar cane trash and pineapple residues on soil pH, redox potential, extractable Al, Fe and Mn. *Trop. Agric. (Trinidad)* **57**, 245 – 258.

Asirifi, K. N., Morgan, W. C. and Parbery, D. G. (1994). Suppression of Sclerotinia soft rot of lettuce with organic soil amendments. *Australian Journal of Experimental Agriculture* **34**, 131-136.

Atallah, T., Andreux, F., Chone, T. and Grass, F. (1995). Effect of storage and composting on the properties and degradability of cattle manure. *Agriculture, Ecosystems and Environment* **54**, 203 – 213.

Babatola, J. O. (1989). Effects of some organic manures on nematodes in tomato cultivation. *Pakistan Journal of Nematology* **7**, 39-46.

Babatola, J. O. and Oyedunmade, E. E. A. (1992). Influence of organic manures and urea on nematode pests of *Celosia argentea*. *Nematologia Mediterranea* **20**, 237-239.

Bacharach, U. (1957). The aerobic breakdown of uric acid by certain *Pseudomonas*. *Journal of Gen. Microbiol.* **17**, 1 – 9.

Barbarika, A. Jr., Sikora, L. J. and Colacicco, D. (1985). Factors affecting the mineralization of nitrogen in sewage sludge applied to soils. *Soil Sci. Soc. Am. J.* **49**, 1403 – 1406.

Barnby, M. A., Yamaski, R. B. and Klocke, J. A. (1989). Biological activity of azadirachtin, three derivatives, and their ultraviolet radiation degradation products against tobacco budworm (Lepidoptera: Noctuidae) larvae. *J. Econ. Ent.* **82**, 58-63.

Bear, F.E., Prince, A.L. and Malcom, J.L. (1945). The potassium needs of New Jersey soils. New Jersey Agric. Exp. Stn. Bull. 721.

Bear, F.E., and Toth, L. (1948). Influence of calcium on availability of other soil cations. *Soil Science* **65**, 67-74.

Bednarek, A. and Gaugler, R. (1997). Compatibility of soil amendments with entomopathogenic nematodes. *Journal of Nematology* **29** 220-227.

Berg, B. and McClaugherty, C. (1989). Nitrogen and phosphorus release from decomposing litter in relation to the disappearance of lignin. *Canadian Journal of Botany* **67**, 1148 – 1156.

Bessho, T. and Bell, L. C. (1992). Soil solid and solution phase changes and mung bean response during amelioration of aluminium toxicity with organic matter. *Plant soil* **140**, 183 – 196.

Bevacqua, R. F. and Mellano, V. J. (1994). Cumulative effects of sludge compost on crop yield and soil properties. *Commun. Soil Sci. Plant Anal.* **25**, 395 – 406.

Board on Science and Technology for International Development (BOSTID) (1992). Neem: *A Tree for solving global problems*. Humanity Development Library. Document text. 6th edition .

Borah, A. and Phukan, P. N. (1992). Effect of different organic amendments for the management of *Meloidogyne incognita* on green gram. *Current Nematology* **3**, 49-50.

Brady, N. (1990). *The nature and properties of soils*. Macmillan Publ. Company, New York. 10th edition.

Buwalda, J. G. and Smith, G. S. (1987). Accumulation and partitioning of dry matter and mineral nutrients in developing kiwifruit vines. *Tree Physiology* **3**, 295 – 307.

Caplan, B. (1992). Organic gardening. Headline Book Publ., London, UK.

CERES (1980). FAO review on agriculture and development – A natural source for fertilizer and pests repellents. *CERES* No 075 (Volume 13 No. 3).

Chandler, J.A., Jewell, W.J., Glosett, J.M., Van Soest, P.J. and Robertson, J. B. (1980). Methane fermentation biodegradability. *Biotechnology and Bioengineering Symposium* No. 10. 93-107. John Wiley and Sons.

Chae, Y. M. and Tabatabai, M. A. (1986). Mineralization of nitrogen in soils amended with organic wastes. *Journal of Environmental Quality* **15**, 195 – 198.

Chakrabarti, S. (2000). An integrated approach to managing rice stem nematodes. *International Rice Research Notes* **25**, 16 – 17.

Chescheir, G. M., Westerman, P. W. and Safley, L. M. Jr. (1986). Laboratory methods for estimating available nitrogen in manures and sludges. *Agric. Wastes* **18**, 175 – 195.

Corrales, O., Vargas, E. and Moreira, M. A. (1990). The effect of organic matter on control of root rot in sweet pepper *Capsicum annum* caused by *Phytophthora capsici*. *Agronomia Costarricense* **14**, 9-13.

Datta, M. (1996). Potassium changes in soil upon incorporation of leaf prunings of multipurpose tree species in an acid soil of Tripura. *Journal of the Indian Society of Soil Science* **44**, 398 – 401.

Deepanjan, M., Sushil, K., Pathak, H., Jain, M. C., Upendra, K., Majumdar, D., Kumar, S. and Kumar, U. (2000). Reducing nitrous oxide emission from an irrigated rice field of North India with nitrification inhibitors. *Agriculture, Ecosystems and Environment* **81**, 163 – 169.

Deka, B. C. and Rahman, M. F. (1998). Management of *Meloidogyne incognita* infecting okra by using neem parts and products. *Journal of Agricultural Science Society of North East India* **11**, 124-126.

Donald, P. (1999). Managing Nematodes in Gardens. University Extension, University of Missouri – Columbia, *Agricultural Publication* G6204.

Drechsel, P. and Reck, B. (1997). Composted Shrub-pruning and other organic manures for smallholder farming systems in southern Rwanda. *Agroforestry Systems* **39**, 1-12.

Dropkin, V. H. (1989). *Introduction to plant nematology*. John Wiley and Sons, Inc. 56 – 70.

Dunn, R. A. (1994). Soil Organic Matter, Green Manures and Cover crops for Nematode Management. Institute of Food and Agricultural Sciences,

University of Florida, SS – ENY – 918. *Nematology Plant Protection Pointer* 38.

Durairaj, C., Soorianathasundaram, K. and Nambisan, K. M. P. (1991). Antifeedant effect of plant extracts on *Nodostoma pubicole* (Coleoptera: Eumolpidae), a pest on pear (*Pyrus communis*). *Indian Journal of Agricultural Sciences* **61**, 959-960.

El Nadi, A. H., Rabie, R. K., Magid, H. M. A., Sabrah, R. E. A. and Abdel Aal, S. I. (1995). Chemical, physico-chemical and microbiological examination of town refuse compost and chicken manure as organic fertilizers. *Journal of Arid Environments* **30**, 107 – 113.

Fening, J. O. (1999). Health hazards of pesticides usage in Ghana: The case of vegetable growers in the Akomadan district. *Journal of the Ghana Science Association* **1**, 1-3.

Food and Agriculture Organization (FAO)/ United Nations, Education, Science and Cultural Organization (UNESCO) (1988). FAO/UNESCO soil map of the world. Revised Legend.

Food and Agriculture Organisation (FAO) of UN (1989). The state of Food and Agriculture.

Franzluebber, K., Juo, A. S. R. and Manu, A. (1994). Decomposition of cowpea and millet amendments to sandy alfisol in Niger. *Plant and Soil* **167**, 255 – 265.

Gale, P. M. and Gilmore, J. T. (1986). Carbon and nitrogen mineralization kinetics of poultry litter. *Journal of Environmental Quality* **15**, 423 – 426.

Gamliel, A. and Stapleton, J. J. (1993). Effect of chicken compost or ammonium phosphate and solarization on pathogen control, rhizosphere microorganisms and lettuce growth. *Plant Disease* 77, 886-891.

Genda, S., Gupta, G. N., Kuppusamy, V. and Singh, G. (2000). Changes in soil properties and nutrient accumulation in neem under the canopy of *Acacia tortilis*. *International Tree Crops Journal* **10**, 237 – 246.

Gilmour, C. M., Broadbent, F. E. and Beeks, S. M. (1977). Recycling of carbon and nitrogen through land disposal of various wastes. *In: Soils for management of organic wastes and waste waters*, ed. L. F. Elliot and F. J. Stevenson. Soil Science Society of America, Madison, WI, 151 – 166.

Gnanavelrajah, N. and Kumaragamage, D. (1999). Nitrogen leaching losses and plant response to nitrogen fertilizers as influenced by application of neem (*Azadirachtin indica* A. Juss) materials. *Tropical Agricultural Research* **11**, 29 – 41.

Gosz, J. R., Likens, G. E. and Bormann, F. H. (1973). Nutrient release from decomposing leaf and branch litter in the Hubbard Brook Forest, New Hampshire. *Ecol. Monogr.* **43**, 173 – 191.

Graves, A., Matthews, R. and Walde, K. (2001). Soil organic matter – The forest agriculture interface review. Cranfield University – *Silsoe Technical Report* (R7560).

Greco, N., D'Addabo, T., Brandonisio, A. and Elia, F. (1993). Damage to Italian crops caused by cyst forming nematodes. *Journal of Nematology* **25**, 836-842.

Grewal, P. S., Soni, H. S. and Vijay, B. (1989). Cost effective pretreatment of chicken manure for controlling nematodes and fungi flora in synthetic compost used for the cultivation of *Agaricus bisporus* (Lange) Singer. *Indian Journal of Nematology* **18**, 22-26 .

Griffin, T. (2002). Know your soil – Soil organic matter. *University of Maine Cooperative Extension, Bulletin* No 2288.

Gruhn, P., Goletti, F. and Yudelman, M. (2000). Integrated nutrient management, soil fertility and sustainable agriculture: Current issues and future changes. *International Food Policy Research Institute*, 2020 Brief No 67.

Guyton, R. F., Thompson, J. R., Kimoto, R., Kratky, B. A., Holtzman, O. V., Thyr, B. D. and Miller, W. W. (1989).. Root-knot nematode populations and carrot yield following five forage legumes and continuous carrot. *Hort Science* **24**, 71-73.

Hafez, A. A. R. (1974). Comparative changes in soil physical properties induced by admixtures of manures from various domestic animals. *Soil Science* **118**, 53 – 59.

Hafner, H., George, E., Bationo, A. and Marschner, H. (1993). Effect of crop residues on root growth and phosphorus acquisition of pearl millet in acid soil in Niger. *Plant and Soil* **150**, 117 – 127.

Haidar, M. G., Jha, R. N. and Nath, R. P. (1998). Efficacy of some nematicides and organic amendments on nematode population and growth characteristics of turmeric *Curcuma longa* L. *Journal of Research, Birsa Agricultural University* **10**, 211-213.

Harrison, R. B., Reis, G. G., Reis, M. D. G. F., Bernardo, A. L. and Firme, D. J. (2000). Effect of spacing and age on nitrogen and phosphorus distribution in biomass of *Eucalyptus camaldulensis, Eucalyptus pellita and Eucalyptus urophylla* plantations in southern Brazil. *Forest Ecology and Management* **133**, 167 – 177.

Hatiori, H., and Mukai, S. (1986). Decomposition of sewage sludges in soil as affected by their organic matter composition. *Soil Sci. Plant Nutr.* **32**, 421-432.

Haynes, R. J. and Mokolobate, M. S. (2001.) Amelioration of Al toxicity and P deficiency in acid soils by additions of organic residues: A critical review of the phenomenon and mechanisms involved. *Nutrient Cycling in Agroecosystems* **59**, 47 – 63.

Hemeng, O. B. (1980). The Efficacy of selected nematicides for the control of root-knot nematodes (*Meloidogyne* sp) on tomato in Ghana. *Ghana Journal of Agricultural Science* **13**, 37-40.

Henry, C. (1991). Nitrogen dynamics of pulp and paper sludge to forest soils. *Wat. Sci. Tech.* **24**, 417-425.

Hong, K. Y., Hsu, F. H. and Lu, C. H. (2000). Effects of cattle hog manures on forage yield and quality of Napier grass and soil fertility. *Journal of Taiwan Livestock Research* **33**, 84 – 94.

Hoyt, P. B. and Turner, R. C. (1975). Effects of organic materials added to very acid soils on pH , aluminium, exchangeable NH_4 and crop yields. *Soil Science* **119**, 227 – 237.

Hue, N. V. (1995). Sewage sludge. *In* J. E. Rechcigl (ed.) Soil amendment and environmental quality. Lewis Publ. Boca Raton, FL.

Hunter, A. S., Toth, S. J. and Bear, F. E.(1943). Calcium-potassium ratios for alfalfa. *Soil Science* **55**, 6172.

Hunter, A.S. (1949). Yield and composition of alfalfa as affected by various calcium-magnesium ratios in the soil. *Soil Science* **67**, 53-62.

International Institute of Tropical Agriculture (IITA) (1985). Laboratory manual of selected methods for soil and plant analysis, IITA, Ibadan.

International Institute for Rural Reconstruction (IIRR) (1992). Environmentally sound technologies for women in agriculture. World Environmental Library – Humanity Library Project and GTZ/BMZ-WEL 1.1.

Isman, M. S., Koul, O., Arnason, J. T., Stewart, J. and Salloum, G. S. (1991). Development d'une insecticide a base de neem pour le Canada. *Mem. Ent. Soc. Can.* **159**, 39-46.

Isman, M. B., Matsuura, H., MacKinnon, S., Durst, T., Neil Towers, G. H. and Arnason, J. T. (1996). Phytochemistry of the Meliaceae – So many Terpenoids, So Few Insecticides. Phytochemical Diversity and Redundancy in Ecological Interactions, edited by Romeo et al., Plenum Press, New York. 155-178.

Johnson, A. W., Burton, G. W., Wilson, J. P. and Golden, A. M. (1995). Rotations with coastal bermudagrass and fallow for management of *Meloidogyne incognita* and soilborne fungi on vegetable crops. *Journal of Nematology* **27**, 457-464.

Johnston, S. A., Probasco, P. R. and Phillips, J. R. (1994). Evaluation of soil fumigants and nematicides for the control of root-knot nematodes on carrot, 1993. *Fungicide and Nematicide Test* **49**, 178 - 180.

Johnston, S. A., Probasco, P. R. and Phillips, J. R. (1995). Evaluation of soil fumigants and nematicides for the control of root-knot nematodes on carrot, 1994. *Fungicide and Nematicide Test* **50**, 187 - 191.

Jonathan, E. I., Krishnamoorthy, S. V., Mandharan, M. L. and Muthukrishnan, K. (1991). Effect of organic amendments on the control of sugarcane nematodes. *Bharatiya Sugar* **16**, 37-40.

Jones, T. (2002). Bulk and particle density. New Mexico State University, Soils Laboratory, *SOIL* 252L.

Kalburtji, K. L., Mamolos, A. P. and Kostopoulou, S. (1997). Nutrient release from decomposing *Lotus coniculatus* residues in relation to soil pH and nitrogen levels. *Agriculture, Ecosystems and Environment* **65**, 107 – 112.

Kaplan, M. and Noe, J. P. (1993). Effects of chicken excrement amendments on *Meloidogyne aranaria*. *Journal of Nematology* **25**, 71-77.

Kara, E. E. and Erel, A. (1999). Effect of chicken manure on some soil properties and dry matter content of oats. *Anadolu* **9**, 91 – 104.

Kerry, B. R. and Bourne, J. M. (1996). The importance of rhizosphere interactions in the biological control of plant parasitic nematodes – a case study using *Verticillium chlamydosporium*. *Pesticide Science* **47**, 69-75.

Kettering, M. (ed) (1992). Training and development – Intensive vegetable gardening for profit and self-sufficiency. *Peace Corps Information Collection and Exchange*, USA.

Khaleel, R., Reddy, K. R. and Overcash, M. R. (1981). Changes in soil physical properties due to organic waste applications: a review. *Journal of Environmental Quality* **10**, 133 – 141.

Khan, A. M., Alam, M. M. and Saxena, S. K. (1974). Mechanism of the control of plant parasitic nematodes as a result of application of oil cakes to the soil. *Indian Journal of Nematology* **4**, 93 – 96.

Khan, F. A. (1994). Effect of soil amendments on the development of *Pratylenchus brachyurus* and growth and yield of okra. *Afro-Asian Journal of Nematology* **4** 84-87.

Khan, T. A. and Saxena, S. K. (1997). Integrated management of root knot nematode *Meloidogyne javanica* infecting tomato using organic materials and *Paecilomyces lilacinus*. *Bioresource Technology* **61**, 247 – 250.

Koul, P., Shanker, J. S. and Kapil, R. S. (1996). The effect of neem allelochemicals on nutritional physiology of larval *Spodoptera litura*. *Entomologia Experimentalis et Applicata* **79** 43-50.

Lalljee, B., Singh, R. P. (ed) and Saxena, R. C. (1999). *Effect of neemcake on nitrification inhibition in an Oxisol of Mauritius. Azadirachta indica* A. Juss. 291 – 301. Science Publishers, Inc.; Enfield; USA.

Larney, F. J. and Janzen, H. H. (1996). Restoration of productivity to a desurfaced soil with livestock manure, crop residue, and fertilizer amendments. *Agronomy Journal* **88**, 921-927.

Li, R., Werger, M. J. A., During, H. J. and Zhong, Z. C. (1998). Carbon and nutrient dynamics in relation to growth rhythm in the giant bamboo *Phyllostachys pubescens*. *Plant and Soil* **201**, 113 – 123.

Mahimairaja, S., Bolan, N. S., Hedley, M. J. and Macgregor, A. N. (1994). Losses and transformation of nitrogen during composting of poultry manure with different amendments: An incubation experiment. *Bioresource Technology* **47**, 265 – 273.

Mai, W. F., Lyon, H. H. and Kruk, T. H. (1968). Pictorial Key to Genera of plant parasitic nematodes. Dept. of Plant pathology , New York State College of Agriculture. Unit of the State University of New York, Cornell University, Ithaca.

Manson, A. D., Miles, N., Robert, V. G. and Katusic, V. (2002). Plant analysis. Technical Publications, Soil Fertility: Course Notes, Kwazulu- Natal Department of Agriculture and Environmental Affairs.

Marshner, H. (1995). Mineral nutrition of higher plants. 2nd ed. Academic Press. San Diego. *In* Schomberg, H. H. and Steiner, J. L. (1999) Nutrient dynamics of crop residue decomposing on a fallow no-till soil surface. *Soil Science Soc. American Journal* **63**, 607 – 613.

Mbagwu, J. S. C. (1989). Effects of organic amendments on some physical properties of a tropical Ultisol. *Biological Wastes* **28**, 1 – 13.

Mbagwu, J. S. C., Unamba, O. I. and Nevoh, G. O. (1994). Physico-Chemical properties and productivity of two tropical soils amended with dehydrated swine waste. *Bioresource Technology* **49**, 163 – 171.

McCollum, J. P. (1975). Producing vegetable crops. Interstate Printers and Publishers, Inc.

McLean, E. O., Hartwig, R.C., Eckert, D.J. and Triplett, G.B. (1983). Basic cation saturation ratios as a basis for fertilizing and liming agronomic crops. Field studies. *Agronomy Journal* **75**, 635-639.

McSorley, R. and Frederick, J. J. (1999). Nematode population fluctuations during decomposition of specific organic amendments. *Journal of Nematology* **31**, 37-44.

Meisner, J., Ascher, K. R. S. and Zur, M. (1983). The residual effect of a neem seed kernel extract sprayed on fodder beet against larvae of *Spodoptera littoralis*. *Phytoparasitica* **11**, 51-54.

Meteorological Station, Cape Coast (1989). Weather report **16**, 62 – 64.

Mian, I. H. and Rodriguez-Kabana, R. (1982). Survey of the nematicidal properties of some organic materials available in Alabama as amendments to soil for control of *Meloidogyne arenaria*. *Nematropica* **12**, 235-246.

Mondini, C., Chiumenti, R., Da Borso, F., Leita, L. and De Nobili, M. (1996). Changes during process in the organic matter of composted and air-dried poultry manure. *Bioresource Technology* **55**, 243 – 249.

More, S. D. (1994). Effect of farm wastes and organic manures on soil properties, nutrient availability and yield of rice-wheat grown on sodic vertisol. *Journal of the Indian Society of Soil Science* **42**, 253 – 256.

Morrill, L. G., Mahilum, B. C. and Mohiuddin, S. H. (1982). *Organic compounds in soils: Sorption, degradation and persistence*. Ann Arbor Sc. Publ. Inc. 230 Collingwood, 180 – 185.

Muir, P. S. (2002). Consequences for organic matter in soil. Oregon State University: B1301 Human Impacts on Ecosystems.

Muller, R. and Gooch, P. S. (1982). Organic amendments in nematode control. An examination of the literature. *Nematropica* **12**, 319-326.

Murwira, H. and Kirchmann, H. (1991). Carbon and nitrogen mineralization of cattle manures, subjected to different treatments, in Zimbabwean and Swedish soils. *In* Proceedings of an International Symposium organized by the Laboratory of Soil Fertility and Soil Biology, Katholieke Universiteit Leuven and the International Institute of Tropical Agriculture (IITA), LEUVEN, Belgium, 4 – 6 November, 1991.

Nahar, M. S., Huq, M. I., Islam, M. S. and Hossain, M. H. (1996). Efficacy of organic soil amendment for the management of root-knot of tomato. *Bangladesh Journal of Plant Pathology* **12**, 17 – 20.

Nanjegowda, D., Naik, B. G., Ravi, K., Reddy, P. P. (ed.), Kumar, N. K. (ed.) and Verghese, A. (1998). Efficacy of neem products and a nematicide for the management of root-knot nematode *Meloidogyne incognita* in tomato nursery. Advances in IPM for horticultural crops. Proceedings of the First

National Symposium on Pest Management in Horticultural Crops: environmental implications and thrusts, Bangalore, India, 15 – 17 October 1997, 318 – 320.

Neem Foundation (1997). Neem for fertilizer management. www.neemfoundation.org/fert.htm

Nellum, A. L. (ed.) (1985). Training and development – Agricultural development workers. *Training Manual* Volume III Corps. Peace Corps Information Collection and Exchange, USA.

Nick, J. and Bradley, F. (1994). *Growing fruits and vegetables organically*. Rodale Press, Emmaus, PA.

Noble, A. D., Zenneck, I. and Randall, P. J. (1996). Leaf litter, ash, alkalinity and neutralization of soil acidity. *Plant Soil* **179**, 293 – 302.

Nodar, R., Acea, M. J. and Carballas, T. (1990). Microbial composition of poultry excreta. *Biological Wastes* **33**, 95 – 105.

Nordmeyer, H. and Richter, J. (1985). Incubation experiments on nitrogen mineralization in loess and sandy soils. *Plant and Soil* **83**, 433 – 445.

Nyathi, P. and Campbell, B. M. (1995). Interaction effect of tree leaf litter, manure and inorganic fertilizer on the performance of maize in Zimbabwe. *African Crop Science Journal* **3**, 451 – 456.

Nyakatawa, E. Z. and Reddy, K. C. (2000). Tillage, cover cropping and poultry litter effects on cotton: I. Germination and seedling growth. *Agronomy Journal* **95**, 992 – 999.

Obi, M. E. and Ebo, P. O. (1995). The effects of organic and inorganic amendments on soil physical properties and maize production in a severely degraded sandy soil in southern Nigeria. *Bioresource Technology* **51**, 117-123.

Oikeh, S. O. and Asiegbu, J. E. (1993). Growth and yield responses of tomatoes to sources and rates of organic manure in ferralitic soils. *Bioresource Technology* **45**, 21-25.

Okeleye, K. A. and Adetunji, M. T. (1999). The influence of legume hedgerow prunings on the properties of a degraded soil. *International Journal of Tropical Agriculture* **17**, 19 – 25.

Opperman, M. H., Wood, M., Harris, P. J. and Cherrett, C. P. (1993). Nematode and nitrate dynamics in soils treated with cattle slurry. *Soil Biol. Biochem.* **25**, 19 – 24.

Page, A. L., Miller, R. H. and Keeney, D. R. (ed.) (1982). *Methods of soil analysis: Part 2 – Chemical and microbial properties*. 2nd Edition. American Society of Agronomy and Soil Science Society of America, Inc, 677 South Segoe Road, Madison, Wisconsin, USA.

Pandey, G., Dwivedi, B. K. and Singh, R. B. (1991.) Effect of organic amendments on the root-knot disease of green gram and its rhizosphere microflora. *New Agriculturist* **2**, 203-206.

Patel, J. R., Patel, J. I., Mehta, D. M. and Shah, B. R. (1993). Integrated management of *Amsacta moorei* Butler with botanical pesticides. Botanical pesticides in integrated pest management: Proceedings of National

symposium held in January 21-22, 1990 at Central Tobacco Research Institute, Rajahmundry, India. 327-331.

Patel, U. G. and Patel, J. R. (1997). Ovicidal effect of botanical alone and in combination with synthetic insecticides on eggs of *Helicoverpa armigera*. *Indian Journal of Entomology* **59**, 326-329.

Patiram, I. (1994). Effect of organic manure and nitrogen on phosphate potential and its buffering capacity on an acid soil. *Journal of the Indian Society of Soil Science* **42**, 134 – 136.

Pocknee, S. and Sumner, M. E. (1997). Cation and nitrogen contents of organic matter determine its liming potential. *American Soil Science Society Journal* **61**, 86 – 92.

Pool, N. L., Trinidad, S. A., Etchevers, B. J. D., Perez, M. J. and Martinez, G. A. (2000). Improvement of soil fertility in hillside agriculture of Los Altos de Chiapas, Mexico. *Agrociencia* **34**, 251 – 259.

Powers, L. E. McSorley, R. and Dunn, R. A. (1993). Effects of mixed cropping on a soil nematode community in Honduras. *Journal of Nematology* **25**, 666-673.

Price, A.L., Zimmerman, M. and Bear, F.E. (1947). The magnesium-supplying power of 20 New Jersey soils. *Soil Science* **63**, 69-78.

Ramakrishnan, S. and Rajendran, G. (1998). Management of *Meloidogyne incognita* in papaya *Carica papaya* L. *Pest Management in Horticultural Ecosystems* **4**, 101-106.

Reddy, P. P., Khan, R. M., Rao, M. S., Chari, M. S. and Ramaprasad, G. (ed.) (1993). Management of root-knot nematodes infesting papaya by incorporation of some plant leaves. Botanical pesticides in integrated pest management: Proceedings of National symposium held in January 21-22, 1990 at Central Tobacco Research Institute, Rajahmundry, India. 421-423.

Rehm, G.W. and Sorensen, R.C. (1985). Effects of potassium and magnesium applied for corn grown on an irrigated sandy soil. *Soil Sci. Soc. Amer. J.* **49**, 1446-1450.

Riegel, C., Fernandez, F. A. and Noe, J. P. (1996). *Meloidogyne incognita* infested soil amended with chicken litter. *Journal of Nematology* **28**, 369-378.

Robertson, F. A. and Morgan, W. C. (1995). Mineralization of C and N in organic materials as affected by duration of composting. *Australian Journal of Soil Research* **33**, 511 – 524.

Rodriguez-Kabana, R. (1986). Organic and inorganic nitrogen amendments to soils as nematode suppressants. *Journal of Nematology* **18**, 129-135.

Rojas, L. A., Baquero, J. E., Ramirez, M., Rodriguez, F. and Roveda, G. (2001). Organic matter and its relation to maize crops on acid soils of Columbia. *In* W. J. Horst et al. (eds.) Plant nutrition – Food security and sustainability of agro-ecosystems. Kluwer Academy Publishers, Netherlands.

Rowell, D. L. (1994). *Soil Science: Methods and Applications*. Longman Group UK Limited.

Rubaduka, E. B., Cadish, G. and Giller, K. E. (1991). Mineralization of nitrogen in woody legume prunings and its recovery by maize. *In* Proceedings of an

International Symposium organized by the Laboratory of Soil Fertility and Soil Biology, Katholicke Universiteit Leuven and the International Institute of Tropical Agriculture (IITA), LEUVEN, Belgium, 4 – 6 November, 1991.

Russell, F. D. (1990). *Mstat crop directory*. Michigan State University, East Lansing, MI.

Sanginga, N., Danso, S. K. A , Zapat, F. and Bowen, G. D. (1994). Influence of pruning management on P & N distribution and use efficiency by N_2 fixing and non- N_2 fixing trees used in alley cropping systems. *Plant and Soil* **167,** 219 – 226.

Sarathchandra, S. U., Watson, R. N., Cox, N. R., DiMenna, M. E., Brown, J. A., Burch, G. and Neville, F. J. (1996). Effects of chitin amendment of soil on microorganisms, nematodes, and growth of white clover (*Trifolium ripens* L.) and perennial ryegrass (*Lolium perenne* L.). *Biol Fertil Soils* **22,** 221-226.

Sarmah, A. C. and Bordoloi, P. K. (1994). Decomposition of organic matter in soils in relation to mineralization of carbon and nutrient availability. *Journal of the Indian Society of Soil Science* **42**, 199 – 203.

Sarrantonio, M., and Scott, T. W. (1988). Tillage effects on availability of nitrogen to corn following a winter green manure crop. *Soil Science Society of American Journal* **52**, 1661 – 1668.

Sawant, I. S., Sawant, S. D. and Nanaya, K. A. (1995). Biological control of phytophthora root-rot of mandarin *Citrus reticulata* by *Trichoderma* sp grown on coffee waste. *Indian Journal of Sciences* **65**, 842-846.

Schiliro, E., Gampe, G., Colombo, A. and Boncorago, P. (1995). Severe damage to carrot caused by the nematode *Ditylenchus dispaci*. *Informatore-Agrario* **51**, 81-84.

Schmutterer, H. (1988). Potential of azadirachtin-containing pesticides for integrated pest control in developing and industrialized countries. *Journal of Insect Physiology* **34**, 713-719.

Schomberg, H. H. and Steiner, J. L. (1999). Nutrient dynamics of crop residues decomposing on a fallow no-till soil surface. *Soil Science Society of American Journal* **63**, 607 – 613.

Schulte, E.E., and Kelling, K.A. (1985). Soil calcium to magnesium ratios- should you be concerned? Bulletin G2986. University of Wisconsin Extension Service. Madison, Wisconsin.

Schulthess, F., Neuenschwander, P. and Gounou, S. (1997). Multi-trophic interactions in cassava, *Manihot esculenta*, cropping systems in the subhumid tropics of West Africa. *Agriculture, Ecosystems and Environment* **66**, 211 – 222.

Seyfried, M. S. and Rao, P. S. C. (1988). Kinetics of nitrogen mineralization in Costa Rican soils: Model evaluation and pre-treatment effects. *Zimbabwe Agric. Journal* **81**, 31 – 35.

Shah, Z. and Faheem, A. (2000). Evaluation of indigenous plant materials for the nitrification inhibition property in soil. *Sarhad Journal of Agriculture* **16**, 69 – 75.

Sharma, G. K. (1992). Growth-inhibiting activity of azadirachtin on *Corcyra cephalonica*. *Phytoparasitica (Israel)* **20**, 47-50.

Sharma, G. L. and Sharma, M. K. (1995). Management of root-knot nematodes *Meloidogyne incognita* and *Meloidogyne javanica* in tomato by bare root dip treatment. *Indian Journal of Nematology* **29**, 174-176.

Sharma, D.D., Chandrashekar, D. S., Srikantaswamy, K. and Govindaiah (1998). On-farm evaluation of cultural and chemical methods for the control of root-knot nematode disease of mulberry. *Indian Journal of Sericulture* **37**, 61-63.

Siddiqui, M. A. and Alam, M. M. (1997). Integrated control of plant parasitic nematodes with organic soil amendments/nematicides and ploughing of okra. Proceedings of the Indian National Science Academy, Part B, *Reviews and Tracts Biological Sciences* **63**, 545 – 550.

Simson, C.R., Corey, R.B. and Sumner, M.E. (1979). Effect of varying Ca:Mg ratios on yield and composition of corn and alfalfa. Commun. *Soil Sci. and Plant Anal.* **10**, 153-162.

Singh, G. and Bhati, M. (2003). Growth, biomass production and nutrient composition of Eucalyptus seedlings irrigated with municipal effluent in loamy sand soil of Indian desert. *Journal of Plant Nutrition* **26**, 2469 – 2488.

Stark, J. D. and Walter, J. F. (1995). Persistence of azadirachtin A and B in soil: Effects of Temperature and microbial activity. *J. Environ. Sci. Health B 30* **5**, 685-698.

Stewart, E. Allen (ed), Max Grimshaw, H. (ed), Parkinson, J. A. (ed) and Quarmby, C. (ed) (1974). *Chemical analysis of ecological materials.* Blackwell Scientific Publications, Osney Mead, Oxford.

Stirling, G. R. (1991). *Biological control of plant parasitic nematodes.* Wallingford, U.K.: CAB International.

Sullivan, P. (1999). Sustainable Soil Management- Soil System Guide. *Appropriate Technology Transfer for Rural Areas,* Fayetteville, AR

Sultan, M. S., Sharma, S. K., Singh, I. and Singh, Iderjit (1995.) Effect of some chemicals and late sowing on the disease intensity of root-knot nematode *Meloidogyne incognita* and yield of black gram. *Plant Disease Research* **1**, 86-88.

Sundaram, K. M. S. (1996). Azadirachtin Biopesticide: A review of studies conducted on its analytical chemistry, environmental behaviour and biological effects. *J. Environ. Sci. Health B* **4**, 913-948.

Sweeten, J. M. and Mathers, A. C. (1985). Improving soils with livestock manures. *J. Soil Water Conserv.* **40**, 206 – 210.

Szeto, S. Y. and Wan, M. T. (1996). Hydrolysis of azadirachtin in buffered and natural waters. *Journal of Agriculture and Food Chemistry* **44**, 1160-1163.

Taylor, A. L. and Sasser, J. N. (1978). Biology, identification and control of root knot nematode (*Meloidogyne* spp). Coop. Publ. Dept. Plt. Pathol. North Carolina State Univ. and USAID, Raleigh NC 111.

Thonnissen, C., Midmore, D. J., Ladha, J. K., Olk, D. C. and Schmidhalter, U. (2000). Legume decomposition and nitrogen release when applied as green

manures to tropical vegetable production systems. *Agronomy Journal* **92**, 253 – 260.

Tilander, Y. and Bonzi, M. (1997). Water and nutrient conservation through the use of agroforestry mulches and sorghum yield response. *Plant and Soil* **197**, 219 – 232.

Tiquia, S. M. and Tam, N. F. Y. (2000). Fate of nitrogen during composting of chicken litter. *Environmental Pollution* **110**, 535 541.

Tisdale, S. L., Nelson, W. L., Beaton, J. D. and Havlin, J. L. (1993). *Soil fertility and fertilizers*, 5th Edition. Macmillan Pub. Comp. N. Y.

Tiyagi, S. A. and Alam, M. N. (1995). Efficacy of oil seed cakes against plant-parasitic nematodes and soil-inhibiting fungi on mungbean and chickpea. *Bioresource Technology* **51**, 233-239.

Toida, Y., Keereewan, S. and Murakami, T. (1991). Cultural control of java root-knot nematode attacking mungbean in Thailand. *Tropical Agricultural Research Center - Newsletter* **2**, 5.

van Kessel, J. S., Reeves III, J. B. and Meisinger, J. J. (2000). Nitrogen and carbon mineralization of potential manure components. *Journal of Environmental Quality* **29**, 1669 – 1677.

Wan, M. T., Rahe, J. E. and Wong, C. (1997). Persistence of azadirachtin-A in two biological systems used for culturing mycorrhizal fungus *Glomus intraradices*. *Journal of Environmental Science and Health B* **32**, 929-953.

Wani, A. H. (1992). Control of root-knot nematodes on okra with seed soaking in neem leaf extract. *Current Nematology* **3**, 39-40.

West, A. W., Sparling, G. P. and Speir, T. W. (1989). Microbial activity in gradually dried or re-wetted soils as governed by water and substrate availability. *Australian Journal of Soil Research* **27**, 747 – 757.

Widmer, T. L., Ludwig, J. W. and Abawi, G. S. (2001). The Northern root-knot nematode on carrot, lettuce and onion in New York. *New York's Food and Life Science Bulletin, Vegetable MD* on line, 2.

Wieder, R. K. and Lang, G. E. (1982). A critique of the analytical method used in examining decomposition data obtained from litter bags. *Ecology* **63**, 1636 – 1642.

Williams, C. N., Uzo, J. O. and Peregrine, W. T. H. (1991). *Vegetable production in the tropics*. Intermediate Tropical Agric. Series.

Williams, S. (2002). *Fertilizer: Application (organic vs. inorganic)*. University of Saskatchewan, Extension Division and Department of Plant Sciences.

Willmott, S., Gooch, P. S., Siddiqi, M. R. and Franklin, M. (1977). *Commonwealth Institute of Helminthology. Descriptions of Plant Parasitic Nematodes*. Spottiswoode Ballantyne Press by William Clowes & Sons Ltd. London, Colchester and Beccles.

WHO (1990). Global Estimates for Health Situation Assessment and Projections, Geneva.

Wong, M. T. F., Nortcliff, S. and Swift, R. S. (1998). Method for determining the acid ameliorating capacity of plant residue compost, urban waste compost, farmyard manure and peat applied to tropical soils. *Commun Soil Sci Plant Anal.* **29**, 2927 – 2937.

Wong, J. W. C., Ma, K. K., Fang, K. M. and Cheung, C. (1999). Utilization of a manure compost for organic farming in Hong Kong. *Bioresource Technology* **67**, 43 – 46.

WSARE (1995). The basic principles of sustainable agriculture. URL: http://ext.usu.edu/wsare

APPENDIX A

Appendix A1. Effect of soil amendment on populations on *Meloidogyne* spp.

Amendment (per kg of soil)	Nematodes per 100 g of soil							
	Sep. 16	Sep. 30	Oct. 14	Oct. 28	Nov. 11	Nov. 25	Dec. 9	Mean
No amendment (Control)	98 a	120 a	83a	35a	40a	29a	28 a	62 a
10 g C	50 b	36 bc	48b	24b	12c	16b	8 b	28 b
5 g PM	40 c	40 b	30c	16c	24b	12bc	7 bc	24 b
10 g PM	32 d	29 c	22d	11e	19b	13bc	4 cde	19 c
50 g NL	0 e	15 de	17ef	13de	3ef	10c	1 ef	8 de
100 g NL	0 e	15 de	14fgh	0g	0f	2d	0 f	4 de
50 g NL + 10 g C	0 e	18 de	20de	11e	8cde	3d	5 bcd	9 d
100 g NL + 10 g C	0 e	16 de	0i	15cd	9cde	0d	0 f	6 de
50 g NL + 5 g PM	0 e	20 d	9h	6f	0f	4d	3 def	6 de
50 g NL + 10 g PM	0 e	15 de	11gh	8f	10cd	3d	3 def	7 de
100 g NL + 5 g PM	0 e	14 de	15fg	7f	5def	0d	0 f	6 de
100 g NL + 10 gPM	0 e	9 de	12fgh	0g	3ef	0d	0 f	3 e

Means with the same letters within columns are not significantly different ($P \leq 0.05$)
C= Cow dung
PM= Poultry Manure
NL= Neem Leaf

ANOVA Tables – Appendices B9-B16 (pp 160 – 163)

Appendix A2. Effect of soil amendment on populations on *Scutellonema* spp.

Amendment (per kg of soil)	Nematodes per 100 g of soil							
	Sep. 16	Sep. 30	Oct. 14	Oct. 28	Nov. 11	Nov. 25	Dec. 9	Mean
No amendment (Control)	53 a	54 a	34a	18a	20a	16a	14 a	30 a
10 g C	42 b	36 b	21b	12b	17b	7b	9 b	21 b
5 g PM	38 c	24 c	20b	13b	8c	8b	6 c	17 c
10 g PM	27 d	16 d	18b	5c	7c	2c	2 de	11 d
50 g NL	0 e	3 ef	6cde	4cd	4de	1c	1 de	3 ef
100 g NL	0 e	2 f	4def	1de	2fg	0c	0 e	1 fg
50 g NL + 10 g C	0 e	6 e	8c	6c	3ef	0c	2 de	4 e
100 g NL + 10 g C	0 e	2 f	6cde	1de	1fg	2c	0 e	2 efg
50 g NL + 5 g PM	0 e	3 ef	7cd	3cde	1fg	2c	1 de	2 efg
50 g NL + 10 g PM	0 e	0 f	4def	6c	5d	1c	3 d	3 ef
100 g NL + 5 g PM	0 e	0 f	3ef	0e	2fg	0c	0 e	1 fg
100 g NL + 10gPM	0 e	1 f	1f	0e	0g	0c	0 e	0 g

Means with the same letters within columns are not significantly different ($P \leq 0.05$)
C= Cow dung
PM= Poultry Manure
NL= Neem Leaf

ANOVA Tables – Appendices B17-B24 (pp 163 – 164)

Appendix A3. Effect of soil amendment on populations on *Pratylenchus* spp.

Amendment (per kg of soil)	Nematodes per 100 g of soil							
	Sep. 16	Sep. 30	Oct. 14	Oct. 28	Nov. 11	Nov. 25	Dec. 9	Mean
No amendment (Control)	16 a	12 a	13a	8a	4a	2a	2 a	8 a
10 g C	3 b	8 b	7b	3b	2b	1b	1 b	4 b
5 g PM	2 b	5 c	4c	1cd	2b	2a	0 c	2 c
10 g PM	2 b	4 cd	2de	3b	1bc	1b	1 b	2 c
50 g NL	0 c	4 cd	0f	2bc	2b	0c	0 c	1 c
100 g NL	0 c	3 cd	1ef	1cd	0c	0c	0 c	1 c
50 g NL + 10 g C	0 c	2 d	2de	2bc	0c	1b	0 c	1 c
100 g NL + 10 g C	0 c	2 d	3cd	0d	1bc	0c	0 c	1 c
50 g NL + 5 g PM	0 c	5 c	2de	1cd	0c	1b	0 c	1 c
50 g NL + 10 g PM	0 c	4 cd	3cd	0d	1bc	0c	0 c	1 c
100 g NL + 5 g PM	0 c	3 cd	1ef	1cd	2b	0c	0 c	1 c
100 g NL + 10 gPM	0 c	2 d	1ef	0d	0c	0c	0 c	1 c

Means with the same letters within columns are not significantly different ($P \leq 0.05$)
C= Cow dung
PM= Poultry Manure
NL= Neem Leaf

ANOVA Tables – Appendices B25-B32 (pp 166 –168)

Appendix A4. Effect of soil amendment on populations on *Paratricodorus* spp.

Amendment (per kg of soil)	Nematodes per 100 g of soil							
	Sep. 16	Sep. 30	Oct. 14	Oct. 28	Nov. 11	Nov. 25	Dec. 9	Mean
No amendment (Control)	20 a	14 a	15a	12a	9a	8a	8 a	12 a
10 g C	17 a	12 ab	10c	7c	5b	8a	6 b	9 b
5 g PM	10 b	10 b	8d	10b	4b	3c	1 c	7 b
10 g PM	13 b	11 ab	12b	4d	1c	5b	1 c	7 b
50 g NL	0 c	2 c	2ef	1e	0c	0d	1 c	1 c
100 g NL	0 c	0 c	1fg	0e	0c	1d	0 c	0 c
50 g NL + 10 g C	0 c	1 c	3e	1e	0c	0d	0 c	1 c
100 g NL + 10 g C	0 c	2 c	1fg	0e	0c	0d	1 c	1 c
50 g NL + 5 g PM	0 c	1 c	3e	1e	0c	0d	0 c	1 c
50 g NL + 10 g PM	0 c	1 c	2ef	0e	1c	0d	0 c	1 c
100 g NL + 5 g PM	0 c	2 c	0g	1e	0c	0d	0 c	0 c
100 g NL + 10 gPM	0 c	0 c	1fg	1e	0c	0d	0 c	0 c

Means with the same letters within columns are not significantly different ($P \leq 0.05$)
C= Cow dung
PM= Poultry Manure
NL= Neem Leaf

ANOVA Tables – Appendices B33-B40 (pp 168 – 171)

Appendix A5. Effect of soil amendment on populations on *Tricodorus* spp.

Amendment (per kg of soil)	Nematodes per 100 g of soil							
	Sep. 16	Sep. 30	Oct. 14	Oct. 28	Nov. 11	Nov. 25	Dec. 9	Mean
No amendment (Control)	14 a	16 a	8b	11a	9a	5a	4 a	10 a
10 g C	11 b	8 b	12a	3bc	4b	0d	3 a	6 b
5 g PM	9 c	6 c	6c	4b	2cd	2bc	1 b	4 b
10 g PM	10 bc	5 cd	6c	1de	3bc	2bc	1 b	4 b
50 g NL	0 d	3 def	3d	0e	1de	0d	1 b	1 c
100 g NL	0 d	0 g	1ef	2cd	0e	1cd	0 b	1 c
50 g NL + 10 g C	0 d	2 efg	3d	1de	0e	0d	1 b	1 c
100 g NL + 10 g C	0 d	0 g	2de	0e	1de	3b	0 b	1 c
50 g NL + 5 g PM	0 d	1 fg	3d	2cd	0e	1cd	0 b	1 c
50 g NL + 10 g PM	0 d	4 cde	1ef	0e	1de	0d	0 b	1 c
100 g NL + 5 g PM	0 d	0 g	2de	2cd	0e	0d	0 b	1 c
100 g NL + 10 gPM	0 d	0 g	0f	0e	1de	2bc	0 b	0 c

Means with the same letters within columns are not significantly different ($P \leq 0.05$)
C= Cow dung
PM= Poultry Manure
NL= Neem Leaf

ANOVA Tables – Appendices B41-B48 (pp 171 – 173)

Appendix A6. Effect of soil amendment on populations on *Helicotylenchus* spp.

Amendment (per kg of soil)	Nematodes per 100 g of soil							
	Sep. 16	Sep. 30	Oct. 14	Oct. 28	Nov. 11	Nov. 25	Dec. 9	Mean
No amendment (Control)	10 a	3 a	5a	2a	3a	1a	3 a	4 a
10 g C	7 b	2 ab	0c	2a	0b	0b	0 b	2 b
5 g PM	5 b	1 bc	0c	0b	0b	0b	0 b	1 bc
10 g PM	6 b	1 bc	2b	0b	0b	0b	0 b	1 bc
50 g NL	0 c	0 c	0c	0b	0b	0b	0 b	0 c
100 g NL	0 c	0 c	0c	0b	0b	0b	0 b	0 c
50 g NL + 10 g C	0 c	0 c	0c	0b	0b	0b	0 b	0 c
100 g NL + 10 g C	0 c	0 c	0c	0b	0b	0b	0 b	0 c
50 g NL + 5 g PM	0 c	0 c	0c	0b	0b	0b	0 b	0 c
50g NL + 10 g PM	0 c	0 c	0c	0b	0b	0b	0 b	0 c
100 g NL + 5 g PM	0 c	0 c	0c	0b	0b	0b	0 b	0 c
100 g NL + 10 gPM	0 c	0 c	0c	0b	0b	0b	0 b	0 c

Means with the same letters within columns are not significantly different ($P \leq 0.05$)
C= Cow dung
PM= Poultry Manure
NL= Neem Leaf

ANOVA Tables – Appendices B49-B56 (pp 174 – 176)

Appendix A7. Effect of soil amendment on populations on *Criconemella* spp.

Amendment (per kg of soil)	Nematodes per 100 g of soil							
	Sep. 16	Sep. 30	Oct. 14	Oct. 28	Nov. 11	Nov. 25	Dec. 9	Mean
No amendment (Control)	3 a	5 a	1a	1b	0	2a	1 a	2 a
10 g C	0 c	0 d	1a	2a	0	1b	1 a	1 b
5 g PM	2 b	2 b	0b	0c	0	2a	1 a	1 b
10 g PM	0 c	1 c	0b	1b	0	1b	0 b	0 c
50 g NL	0 c	0 d	0b	0c	0	0c	0 b	0 c
100 g NL	0 c	0 d	0b	0c	0	0c	0 b	0 c
50 g NL + 10 g C	0 c	1 c	0b	0c	0	0c	0 b	0 c
100 g NL + 10 g C	0 c	0 d	0b	0c	0	0c	0 b	0 c
50 g NL + 5 g PM	0 c	0 d	0b	0c	0	0c	0 b	0 c
50 g NL + 10 g PM	0 c	0 d	0b	0c	0	0c	0 b	0 c
100 g NL + 5 g PM	0 c	0 d	1a	0c	0	0c	0 b	0 c
100 g NL + 10 gPM	0 c	0 d	0b	0c	0	0c	0 b	0 c

Means with the same letters within columns are not significantly different (P≤ 0.05)
C= Cow dung
PM= Poultry Manure
NL= Neem Leaf

ANOVA Tables – Appendices B57-B63 (pp 176 – 178)

Appendix A8. Effect of soil amendment on populations on *Rotylenchus* spp.

Amendment (per kg of soil)	Nematodes per 100 g of soil							
	Sep. 16	Sep. 30	Oct. 14	Oct. 28	Nov. 11	Nov. 25	Dec. 9	Mean
No amendment(Control)	5 a	2 a	0	0	0	0	0	1 a
10 g C	2 b	0 c	0	0	0	0	0	0 b
5 g PM	1 c	0 c	0	0	0	0	0	0 b
10 g PM	0 c	1 b	0	0	0	0	0	0 b
50 g NL	0 c	0 c	0	0	0	0	0	0 b
100 g NL	0 c	0 c	0	0	0	0	0	0 b
50 g NL + 10 g C	0 c	0 c	0	0	0	0	0	0 b
100 g NL + 10 g C	0 c	0 c	0	0	0	0	0	0 b
50 g NL + 5 g PM	0 c	0 c	0	0	0	0	0	0 b
50 g NL + 10 g PM	0 c	0 c	0	0	0	0	0	0 b
100 g NL + 5 g PM	0 c	0 c	0	0	0	0	0	0 b
100 g NL + 10 gPM	0 c	0 c	0	0	0	0	0	0 b

Means with the same letters within columns are not significantly different ($P \leq 0.05$)
C= Cow dung
PM= Poultry Manure
NL= Neem Leaf

ANOVA Tables – Appendices B64-B66 (pp 179)

Appendix A9. *Nutrient content, pH and moisture content of amended soil at the 12th week of incubation*

Treatment	Organic Carbon (%)	CO_2 – C (mg/g soil/day)	NH^+_4 – N (mgN/kg soil)	NO^-_3 – N (mgN/kg soil)	Total N (%)	Avail. P (mg/kg soil)	Exch. K (cmol/kg soil)	Exch. Ca (cmol/kg soil)	Exch. Mg (cmol/kg soil)	pH	Moisture Content (%)
Control (No amendment)	0.46 e	0.042 f	3.37 g	13.13 h	0.04 e	6.75 f	0.18 e	1.04 f	0.90 de	5.00 b	12 11 f
5 gPM/kg	0.53 e	0.057 f	4.86 f	24.23 g	0.05 de	39.73 e	0.20 de	1.01 f	0.63 f	4.90 b	13.55 e
10 gNL/kg soil	0.74 d	0.095 e	5.13 e	32.51 f	0.06 cde	7.59 f	0.23 de	1.29 e	0.93 d	5.20 b	15.40 d
10 gNL + 5 gPM/kg soil	0.74 d	0.129 d	5.60 d	34.38 e	0.07 cd	45.58 d	0.30 cd	1.44 e	0.82 e	5.30 b	16.84 c
20 gNL + 5 gPM/kg soil	0.90 c	0.141 d	6.92 c	71.09 a	0.07 cd	48.84 c	0.40 c	1.68 d	1.50 c	5.30b	18.80 b
30 gNL + 5 gPM/kg soil	1.03 b	0.232 c	7.03 c	40.69 d	0.08 bc	49.41 bc	0.51 b	2.09 c	2.06 b	5.90 a	18.90 b
40 gNL + 5 gPM/kg soil	1.30 a	0.279 b	7.87 b	50.04 c	0.10 ab	51.15 b	0.64 a	2.65 ab	2.30 a	6.20 a	19.30 b
50 gNL + 5 gPM/kg soil	1.36 a	0.351 a	8.98 a	67.50 b	0.11 a	59.26 a	0.68 a	2.80 a	2.39 a	6.30 a	21.50 a

Figures with the same letter within columns are not significantly ($P \leq 0.05$) different from each other.
ANOVA Tables – Appendices B67-B78 (pp 180 – 183)

APPENDIX B

ANOVA TABLES

Appendix B 1. ANOVA for Table 4.2 – Sep. 16

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	88.167	44.083	4.2197	0.0281
2	Factor A	11	179726.000	16338.727	1563.9681	0.0000
-3	Error	22	229.833	10.447		
	Total	35	180044.000			

Coefficient of Variation: 7.08%

Appendix B 2. ANOVA for Table 4.2 – Sep. 30

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	337.500	168.750	43.9349	0.0011
2	Factor A	11	121448.000	11040.727	2874.5089	0.0000
-3	Error	22	84.500	3.841		
	Total	35	121870.000			

Coefficient of Variation: 3.46%

Appendix B 3. ANOVA for Table 4.2 – Oct. 14

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	308.167	154.083	20.9464	0.0010
2	Factor A	11	63492.750	5772.068	784.6684	0.0000
-3	Error	22	161.833	7.356		
	Total	35	63962.750			

Coefficient of Variation: 5.74%

Appendix B 4. ANOVA for Table 4.2 – Oct. 28

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	145.167	72.583	14.9470	0.0001
2	Factor A	11	20765.000	1887.727	388.7363	0.0000
-3	Error	22	106.833	4.856		
	Total	35	21017.000			

Coefficient of Variation: 8.76%

Appendix B 5. ANOVA for Table 4.2 – Nov. 11

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	92.056	46.028	38.0522	0.0012
2	Factor A	11	19328.306	1757.119	1452.6493	0.0000
-3	Error	22	26.611	1.210		
	Total	35	19446.972			

Coefficient of Variation: 4.99%

Appendix B 6. ANOVA for Table 4.2 – Nov. 25

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	121.500	60.750	9.5125	0.0011
2	Factor A	11	11468.750	1042.614	163.2562	0.0000
-3	Error	22	140.500	6.386		
	Total	35	11730.750			

Coefficient of Variation: 16.22%

Appendix B 7. ANOVA for Table 4.2 – Sec. 9

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	37.500	18.750	25.0000	0.0013
2	Factor A	11	10772.000	979.273	1305.6970	0.0000
-3	Error	22	16.500	0.750		
	Total	35	10826.000			

Coefficient of Variation: 7.42%

Appendix B 8. ANOVA for Table 4.2 – Mean

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	150.000	75.000	13.9831	0.0001
2	Factor A	11	45990.000	4180.909	779.4915	0.0000
-3	Error	22	118.000	5.364		
	Total	35	46258.000			

Coefficient of Variation: 7.24%

Appendix B 9. ANOVA for Appendix A1 – Sep. 16

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	66.667	33.333	4.3307	0.0259
2	Factor A	11	32084.000	2916.727	378.9449	0.0000
-3	Error	22	169.333	7.697		
	Total	35	32320.000			

Coefficient of Variation: 15.13%

Appendix B 10. ANOVA for Appendix A1 – Sep. 30

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	450.667	225.333	43.7412	0.0021
2	Factor A	11	30104.750	2736.795	531.2603	0.0000
-3	Error	22	113.333	5.152		
	Total	35	30668.750			

Coefficient of Variation: 7.85%

Appendix B 11. ANOVA for Appendix A1 – Oct.14

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	308.167	154.083	19.0619	0.0001
2	Factor A	11	16358.750	1487.159	183.9784	0.0000
-3	Error	22	177.833	8.083		
	Total	35	16844.750			

Coefficient of Variation: 12.14%

Appendix B 12. ANOVA for Appendix A1 – Oct.28

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	112.667	56.333	24.1429	0.0042
2	Factor A	11	3197.000	290.636	124.5584	0.0000
-3	Error	22	51.333	2.333		
	Total	35	3361.000			

Coefficient of Variation: 12.56%

Appendix B 13. ANOVA for Appendix A1 – Nov. 11

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	20.667	10.333	0.8024	
2	Factor A	11	4484.750	407.705	31.6571	0.0000
-3	Error	22	283.333	12.879		
	Total	35	4788.750			

Coefficient of Variation: 32.38%

Appendix B 14. ANOVA for Appendix A1 – Nov. 25

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	301.167	150.583	14.4771	0.0001
2	Factor A	11	2528.000	229.818	22.0947	0.0000
-3	Error	22	228.833	10.402		
	Total	35	3058.000			

Coefficient of Variation: 38.07%

Appendix B 15. ANOVA for Appendix A1 – Dec. 9

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	15.167	7.583	2.0639	0.1508
2	Factor A	11	2000.750	181.886	49.5031	0.0000
-3	Error	22	80.833	3.674		
	Total	35	2096.750			

Coefficient of Variation: 38.99%

Appendix B 16. ANOVA for Appendix A1 – Mean

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	105.167	52.583	7.0182	0.0044
2	Factor A	11	9395.000	854.091	113.9939	0.0000
-3	Error	22	164.833	7.492		
	Total	35	9665.000			

Coefficient of Variation: 18.05%

Appendix B 17. ANOVA for Appendix A2 – Sept. 16

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	48.167	24.083	5.0063	0.0161
2	Factor A	11	13838.000	1258.000	261.5055	0.0000
-3	Error	22	105.833	4.811		
	Total	35	13992.000			

Coefficient of Variation: 16.45%

Appendix B 18. ANOVA for Appendix A2 – Sept. 30

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	100.167	50.083	11.7425	0.0003
2	Factor A	11	9918.750	901.705	211.4121	0.0000
-3	Error	22	93.833	4.265		
	Total	35	10112.750			

Coefficient of Variation: 16.86%

Appendix B 19. ANOVA for Appendix A2 – Oct. 14

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	70.167	35.083	8.2256	0.0022
2	Factor A	11	3288.000	298.909	70.0817	0.0000
-3	Error	22	93.833	4.265		
	Total	35	3452.000			

Coefficient of Variation: 18.77%

Appendix B 20. ANOVA for Appendix A2 – Oct. 28

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	28.667	14.333	4.4206	0.0243
2	Factor A	11	1092.750	99.341	30.6379	0.0000
-3	Error	22	71.333	3.242		
	Total	35	1192.750			

Coefficient of Variation: 31.32%

Appendix B 21. ANOVA for Appendix A2 – Nov. 11

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	57.167	28.583	21.8092	0.0000
2	Factor A	11	1361.000	123.727	94.4046	0.0000
-3	Error	22	28.833	1.311		
	Total	35	1447.000			

Coefficient of Variation: 19.63%

Appendix B 22. ANOVA for Appendix A2 – Nov. 25

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	15.167	7.583	3.1577	0.0623
2	Factor A	11	768.750	69.886	29.1009	0.0000
-3	Error	22	52.833	2.402		
	Total	35	836.750			

Coefficient of Variation: 37.68%

Appendix B 23. ANOVA for Appendix A2 – Dec. 9

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	20.667	10.333	8.3171	0.0020
2	Factor A	11	635.000	57.727	46.4634	0.0000
-3	Error	22	27.333	1.242		
	Total	35	683.000			

Coefficient of Variation: 35.20%

Appendix B 24. ANOVA for Appendix A2 – Mean

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	40.167	20.083	9.6400	0.0010
2	Factor A	11	3128.750	284.432	136.5273	0.0000
-3	Error	22	45.833	2.083		
	Total	35	3214.750			

Coefficient of Variation: 18.23%

Appendix B 25. ANOVA for Appendix A3 – Sept. 16

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	1.500	0.750	1.3200	0.2875
2	Factor A	11	686.750	62.432	109.8800	0.0000
-3	Error	22	12.500	0.568		
	Total	35	700.750			

Coefficient of Variation: 39.33%

Appendix B 26. ANOVA for Appendix A3 – Sept. 30

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	100.167	50.083	27.6611	0.0000
2	Factor A	11	279.000	25.364	14.0084	0.0000
-3	Error	22	39.833	1.811		
	Total	35	419.000			

Coefficient of Variation: 29.90%

Appendix B 27. ANOVA for Appendix A3 – Oct. 14

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	57.167	28.583	27.5401	0.0000
2	Factor A	11	420.750	38.250	36.8540	0.0000
-3	Error	22	22.833	1.038		
	Total	35	500.750			

Coefficient of Variation: 31.35%

Appendix B 28. ANOVA for Appendix A3 – Oct. 28

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	26.167	13.083	20.8072	0.0000
2	Factor A	11	161.000	14.636	23.2771	0.0000
-3	Error	22	13.833	0.629		
	Total	35	201.000			

Coefficient of Variation: 23.25%

Appendix B 29. ANOVA for Appendix A3 – Nov. 11

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	16.667	8.333	11.9565	0.0003
2	Factor A	11	48.750	4.432	6.3587	0.0001
-3	Error	22	15.333	0.697		
	Total	35	80.750			

Coefficient of Variation: 16.79%

Appendix B 30. ANOVA for Appendix A3 – Nov. 25

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	4.167	2.083	2.8947	0.0066
2	Factor A	11	20.000	1.818	2.5263	0.0309
-3	Error	22	15.833	0.720		
	Total	35	40.000			

Coefficient of Variation: 27.25%

Appendix B 34. ANOVA for Appendix A4 – Sept. 30

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	57.167	28.583	25.3221	0.0000
2	Factor A	11	944.000	85.818	76.0268	0.0000
-3	Error	22	24.833	1.129		
	Total	35	1026.000			

Coefficient of Variation: 22.77%

Appendix B 35. ANOVA for Appendix A4 – Oct. 14

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	45.500	22.750	30.3333	0.0000
2	Factor A	11	845.000	76.818	102.4242	0.0000
-3	Error	22	16.500	0.750		
	Total	35	907.000			

Coefficient of Variation: 17.92%

Appendix B 36. ANOVA for Appendix A4 – Oct. 28

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	20.167	10.083	22.5593	0.0000
2	Factor A	11	581.000	52.818	118.1695	0.0000
-3	Error	22	9.833	0.447		
	Total	35	611.000			

Coefficient of Variation: 21.11%

Appendix B 37. ANOVA for Appendix A4 – Nov. 11

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	8.167	4.083	6.4940	0.0061
2	Factor A	11	272.000	24.727	39.3253	0.0000
-3	Error	22	13.833	0.629		
	Total	35	294.000			

Coefficient of Variation: 27.58%

Appendix B 38. ANOVA for Appendix A4 – Nov. 25

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	8.167	4.083	6.4940	0.0061
2	Factor A	11	332.750	30.250	48.1084	0.0000
-3	Error	22	13.833	0.629		
	Total	35	354.750			

Coefficient of Variation: 38.06%

Appendix B 39. ANOVA for Appendix A4 – Dec. 9

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	10.667	5.333	8.8000	0.0016
2	Factor A	11	231.000	21.000	34.6500	0.0000
-3	Error	22	13.333	0.606		
	Total	35	255.000			

Coefficient of Variation: 31.90%

Appendix B 40. ANOVA for Appendix A4 – Mean

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	16.667	8.333	9.4828	0.0011
2	Factor A	11	584.000	53.091	60.4138	0.0000
-3	Error	22	19.333	0.879		
	Total	35	620.000			

Coefficient of Variation: 28.12%

Appendix B 41. ANOVA for Appendix A5 – Sept. 16

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	8.167	4.083	5.0374	0.0158
2	Factor A	11	1010.000	91.818	113.2710	0.0000
-3	Error	22	17.833	0.811		
	Total	35	1036.000			

Coefficient of Variation: 24.55%

Appendix B 42. ANOVA for Appendix A5 – Sept. 30

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	28.167	14.083	14.1908	0.0001
2	Factor A	11	726.750	66.068	66.5725	0.0000
-3	Error	22	21.833	0.992		
	Total	35	776.750			

Coefficient of Variation: 26.57%

Appendix B 43. ANOVA for Appendix A5 – Oct. 14

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	42.667	21.333	50.2857	0.0000
2	Factor A	11	398.750	36.250	85.4464	0.0000
-3	Error	22	9.333	0.424		
	Total	35	450.750			

Coefficient of Variation: 16.63%

Appendix B 44. ANOVA for Appendix A5 – Oct. 28

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	20.167	10.083	16.0361	0.0001
2	Factor A	11	311.000	28.273	44.9639	0.0000
-3	Error	22	13.833	0.629		
	Total	35	345.000			

Coefficient of Variation: 36.60%

Appendix B 45. ANOVA for Appendix A5 – Nov. 11

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	13.500	6.750	17.4706	0.0000
2	Factor A	11	221.000	20.091	52.0000	0.0000
-3	Error	22	8.500	0.386		
	Total	35	243.000			

Coefficient of Variation: 33.90%

Appendix B 46. ANOVA for Appendix A5 – Nov. 25

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	13.500	6.750	11.8800	0.0003
2	Factor A	11	80.000	7.273	12.8000	0.0000
-3	Error	22	12.500	0.568		
	Total	35	106.000			

Coefficient of Variation: 16.53%

Appendix B 47. ANOVA for Appendix A5 – Dec. 9

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	10.667	5.333	8.8000	0.0016
2	Factor A	11	56.750	5.159	8.5125	0.0000
-3	Error	22	13.333	0.606		
	Total	35	80.750			

Coefficient of Variation: 24.93%

Appendix B 48. ANOVA for Appendix A5 – Mean

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	51.056	25.528	28.6374	0.0000
2	Factor A	11	287.889	26.172	29.3598	0.0000
-3	Error	22	19.611	0.891		
	Total	35	358.556			

Coefficient of Variation: 36.16%

Appendix B 49. ANOVA for Appendix A6 – Sep 16

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob-
1	Replication	2	16.667	8.333	4.2308	0.0279
2	Factor A	11	434.000	39.455	20.0308	0.0000
-3	Error	22	43.333	1.970		
	Total	35	494.000			

Coefficient of Variation: 30.15%

Appendix B 50. ANOVA for Appendix A6 – Sep 30

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	2.667	1.333	5.5000	0.0116
2	Factor A	11	32.750	2.977	12.2812	0.0000
-3	Error	22	5.333	0.242		
	Total	35	40.750			

Coefficient of Variation: 34.41%

Appendix B 51. ANOVA for Appendix A6 – Oct. 14

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	1.167	0.583	1.8780	0.0166
2	Factor A	11	74.750	6.795	21.8780	0.0000
-3	Error	22	6.833	0.311		
	Total	35	82.750			

Coefficient of Variation: 25.54%

Appendix B 52. ANOVA for Appendix A6 – Oct. 28

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	2.667	1.333	2.2000	0.0346
2	Factor A	11	20.000	1.818	3.0000	0.0136
-3	Error	22	13.333	0.606		
	Total	35	36.000			

Coefficient of Variation: 33.55%

Appendix B 53. ANOVA for Appendix A6 – Nov. 11

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	0.667	0.333	1.0000	0.0412
2	Factor A	11	24.750	2.250	6.7500	0.0001
-3	Error	22	7.333	0.333		
	Total	35	32.750			

Coefficient of Variation: 30.94%

Appendix B 54. ANOVA for Appendix A6 – Nov. 25

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	0.167	0.083	1.0000	0.0421
2	Factor A	11	2.750	0.250	3.0000	0.0136
-3	Error	22	1.833	0.083		
	Total	35	4.750			

Coefficient of Variation: 34.41%

Appendix B 55. ANOVA for Appendix A6 – Dec. 9

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	0.167	0.083	1.0000	0.0331
2	Factor A	11	24.750	2.250	27.0000	0.0000
-3	Error	22	1.833	0.083		
	Total	35	26.750			

Coefficient of Variation: 25.47%

Appendix B 56. ANOVA for Appendix A6 – Mean

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	2.667	1.333	5.5000	0.0116
2	Factor A	11	50.000	4.545	18.7500	0.0000
-3	Error	22	5.333	0.242		
	Total	35	58.000			

Coefficient of Variation: 33.85%

Appendix B 57. ANOVA for Appendix A7 – Sept. 16

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	1.500	0.750	1.9412	0.0473
2	Factor A	11	32.750	2.977	7.7059	0.0000
-3	Error	22	8.500	0.386		
	Total	35	42.750			

Coefficient of Variation: 34.18%

Appendix B 58. ANOVA for Appendix A7 – Sept. 30

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	6.000	3.000	4.7143	0.0198
2	Factor A	11	72.750	6.614	10.3929	0.0000
-3	Error	22	14.000	0.636		
	Total	35	92.750			

Coefficient of Variation: 26.36%

Appendix B 59. ANOVA for Appendix A7 – Oct. 14

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	1.500	0.750	3.6667	0.0422
2	Factor A	11	6.750	0.614	3.0000	0.0136
-3	Error	22	4.500	0.205		
	Total	35	12.750			

Coefficient of Variation: 18.91%

Appendix B 60. ANOVA for Appendix A7 – Oct. 28

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	2.667	1.333	3.1429	0.0130
2	Factor A	11	14.000	1.273	3.0000	0.0136
-3	Error	22	9.333	0.424		
	Total	35	26.000			

Coefficient of Variation: 21.40%

Appendix B 61. ANOVA for Appendix A7 – Nov. 25

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	6.000	3.000	4.7143	0.0198
2	Factor A	11	21.000	1.909	3.0000	0.0136
-3	Error	22	14.000	0.636		
	Total	35	41.000			

Coefficient of Variation: 19.54%

Appendix B 62. ANOVA for Appendix A7 – Dec. 9

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	1.500	0.750	3.6667	0.0222
2	Factor A	11	6.750	0.614	3.0000	0.0136
-3	Error	22	4.500	0.205		
	Total	35	12.750			

Coefficient of Variation: 18.91%

Appendix B 63. ANOVA for Appendix A7 – Mean

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	2.667	1.333	3.1429	0.0130
2	Factor A	11	14.000	1.273	3.0000	0.0136
-3	Error	22	9.333	0.424		
	Total	35	26.000			

Coefficient of Variation: 15.40%

Appendix B 64. ANOVA for Appendix A8 – Sept. 16

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	2.667	1.333	3.1429	0.0430
2	Factor A	11	74.000	6.727	15.8571	0.0000
-3	Error	22	9.333	0.424		
	Total	35	86.000			

Coefficient of Variation: 37.70%

Appendix B 65. ANOVA for Appendix A8 – Sept. 30

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	1.500	0.750	1.9412	0.0373
2	Factor A	11	12.750	1.159	3.0000	0.0136
-3	Error	22	8.500	0.386		
	Total	35	22.750			

Coefficient of Variation: 28.63%

Appendix B 66. ANOVA for Appendix A8 – Mean

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	0.167	0.083	1.0000	0.0500
2	Factor A	11	2.750	0.250	3.0000	0.0136
-3	Error	22	1.833	0.083		
	Total	35	4.750			

Coefficient of Variation: 36.41%

Appendix B 67 ANOVA for Appendix A9 – Organic Carbon

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	0.017	0.008	62.2632	0.0000
2	Factor A	7	2.303	0.329	2424.4749	0.0000
-3	Error	14	0.002	0.000		
	Total	23	2.322			

Coefficient of Variation: 1.32%

Appendix B 68. ANOVA for Appendix A9 – $CO_2 - C$

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	0.002	0.001	443.7722	0.0000
2	Factor A	7	0.257	0.037	20345.5479	0.0000
-3	Error	14	0.000	0.000		
	Total	23	0.258			

Coefficient of Variation: 0.81%

Appendix B 69. ANOVA for Appendix A9 – $NH^{+}_{4} - N$

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	0.065	0.033	16.2707	0.0002
2	Factor A	7	69.092	9.870	4939.5701	0.0000
-3	Error	14	0.028	0.002		
	Total	23	69.185			

Coefficient of Variation: 0.72%

Appendix B 70. ANOVA for Appendix A9 – Org. N

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	0.681	0.340	0.8639	0.0002
2	Factor A	7	8578.416	1225.488	3110.8411	0.0000
-3	Error	14	5.515	0.394		
	Total	23	8584.612			

Coefficient of Variation: 1.51%

Appendix B 71. ANOVA for Appendix A9 – Total N

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	0.002	0.001	2017612.5000	0.0000
2	Factor A	7	0.012	0.002	4269412.1000	0.0000
-3	Error	14	0.000	0.000		
	Total	23	0.013			

Coefficient of Variation: 2.00%

Appendix B 72. ANOVA for Appendix A9 – Avail. P

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	9.000	4.500	9.0000	0.0031
2	Factor A	7	8496.185	1213.741	2427.4814	0.0000
-3	Error	14	7.000	0.500		
	Total	23	8512.185			

Coefficient of Variation: 1.83%

Appendix B 73. ANOVA for Appendix A9 – Exch. K

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	0.018	0.009	4.9115	0.0242
2	Factor A	7	0.825	0.118	63.5111	0.0000
-3	Error	14	0.026	0.002		
	Total	23	0.869			

Coefficient of Variation: 10.97%

Appendix B 74. ANOVA for Appendix A9 – Exch. Ca

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	0.027	0.014	21.2340	0.0001
2	Factor A	7	10.177	1.454	2267.8973	0.0000
-3	Error	14	0.009	0.001		
	Total	23	10.213			

Coefficient of Variation: 1.45%

Appendix B 75. ANOVA for Appendix A9 – Exch. Mg

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	0.137	0.068	8.3548	0.0041
2	Factor A	7	10.867	1.552	189.4833	0.0000
-3	Error	14	0.115	0.008		
	Total	23	11.118			

Coefficient of Variation: 6.28%

Appendix B 76. ANOVA for Appendix A9 – Exch. Na

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	0.013	0.007	20.2350	0.0001
2	Factor A	7	0.062	0.009	27.1312	0.0000
-3	Error	14	0.005	0.000		
	Total	23	0.080			

Coefficient of Variation: 4.29%

Appendix B 77. ANOVA for Appendix A9 – pH

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	1.322	0.661	6.5309	0.0099
2	Factor A	7	6.206	0.887	8.7566	0.0003
-3	Error	14	1.417	0.101		
	Total	23	8.946			

Coefficient of Variation: 5.77%

Appendix B 78. ANOVA for Appendix A9 – Moisture

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	0.689	0.344	12.0467	0.0009
2	Factor A	7	212.311	30.330	1060.7577	0.0000
-3	Error	14	0.400	0.029		
	Total	23	213.400			

Coefficient of Variation: 0.99%

Appendix B 79. ANOVA for Table 4.3 – Sept. 16

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	308.167	154.083	9.3685	0.0011
2	Factor A	11	78203.000	7109.364	432.2598	0.0000
-3	Error	22	361.833	16.447		
	Total	35	78873.000			

Coefficient of Variation: 11.53%

Appendix B 80. ANOVA for Table 4.3 – Sept. 30

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	23940.167	11970.083	8.0666	0.0024
2	Factor A	11	4155219.000	377747.182	254.5635	0.0000
-3	Error	22	32645.833	1483.902		
	Total	35	4211805.000			

Coefficient of Variation: 5.11%

Appendix B 81. ANOVA for Table 4.3 – Oct. 14

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	677.056	338.528	0.2138	0.0001
2	Factor A	11	22425360.222	2038669.111	1287.4963	0.0000
-3	Error	22	34835.611	1583.437		
	Total	35	22460872.889			

Coefficient of Variation: 4.09%

Appendix B 82. ANOVA for Table 4.3 – Oct. 28

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	2016.667	1008.333	7.9132	0.0026
2	Factor A	11	27874292.750	2534026.614	19886.5347	0.0000
-3	Error	22	2803.333	127.424		
	Total	35	27879112.750			

Coefficient of Variation: 1.13%

Appendix B 83. ANOVA for Table 4.3 – Nov. 11

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	816.667	408.333	53.6853	0.0000
2	Factor A	11	17845226.750	1622293.341	213289.5628	0.0000
-3	Error	22	167.333	7.606		
	Total	35	17846210.750			

Coefficient of Variation: 17.33%

Appendix B 84. ANOVA for Table 4.3 – Nov. 25

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	6208.167	3104.083	7.0071	0.0044
2	Factor A	11	28163738.750	2560339.886	5779.6471	0.0000
-3	Error	22	9745.833	442.992		
	Total	35	28179692.750			

Coefficient of Variation: 11.71%

Appendix B 85. ANOVA for Table 4.3 – Dec. 9

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	12973.500	6486.750	8.7678	0.0016
2	Factor A	11	26338482.750	2394407.523	3236.3816	0.0000
-3	Error	22	16276.500	739.841		
	Total	35	26367732.750			

Coefficient of Variation: 22.36%

Appendix B 86. ANOVA for Table 4.3 – Mean

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	12421.500	6210.750	8.9116	0.0015
2	Factor A	11	10553334.750	959394.068	1376.5967	0.0000
-3	Error	22	15332.500	696.932		
	Total	35	10581088.750			

Coefficient of Variation: 13.08%

Appendix B 87. ANOVA for Table 4.5 – Half life of azadirachtin A

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	169.000	84.500	20.0508	0.0001
2	Factor A	7	2296.921	328.132	77.8617	0.0000
-3	Error	14	59.000	4.214		
	Total	23	2524.921			

Coefficient of Variation: 8.10%

Appendix B 88. ANOVA for Table 4.6 – Moisture content

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	6.250	3.125	4.2752	0.0355
2	Factor A	7	139.491	19.927	27.2617	0.0000
-3	Error	14	10.233	0.731		
	Total	23	155.974			

Coefficient of Variation: 6.25%

Appendix B 89. ANOVA for Table 4.6 – pH

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	1.690	0.845	4.8683	0.0248
2	Factor A	7	0.356	0.051	0.2932	
-3	Error	14	2.430	0.174		
	Total	23	4.476			

Coefficient of Variation: 6.45%

Appendix B 90. ANOVA for Figure 4.2 (a)

x	-0.016378	0.003905	-4.19	0.009

S = 0.2893 R-Sq = 77.9% R-Sq(adj) = 73.4%

Source	DF	SS	MS	F	P
Regression	1	1.4720	1.4720	17.59	0.009
Error	5	0.4184	0.0837		
Total	6	1.8904			

Appendix B 91. ANOVA for Figure 4.2 (b)

x	-0.020730	0.005172	-4.01	0.010

S = 0.3831 R-Sq = 76.3% R-Sq(adj) = 71.5%

Source	DF	SS	MS	F	P
Regression	1	2.3583	2.3583	16.06	0.010
Error	5	0.7340	0.1468		
Total	6	3.0923			

Appendix B 92. ANOVA for Figure 4.2 (c)

x	-0.021811	0.005556	-3.93	0.011

S = 0.4116 R-Sq = 75.5% R-Sq(adj) = 70.6%

Source	DF	SS	MS	F	P
Regression	1	2.6108	2.6108	15.41	0.011
Error	5	0.8469	0.1694		
Total	6	3.4577			

Appendix B 93. ANOVA for Figure 4.2 (d)

x	-0.024821	0.005403	-4.59	0.006

S = 0.4003 R-Sq = 80.8% R-Sq(adj) = 77.0%

Source	DF	SS	MS	F	P
Regression	1	3.3812	3.3812	21.10	0.006
Error	5	0.8012	0.1602		
Total	6	4.1823			

Appendix B 94. ANOVA for Figure 4.3 (a)

x	-0.031071	0.005633	-5.52	0.003

S = 0.4173 R-Sq = 85.9% R-Sq(adj) = 83.1%

Source	DF	SS	MS	F	P
Regression	1	5.2983	5.2983	30.43	0.003
Error	5	0.8707	0.1741		
Total	6	6.1690			

Appendix B 95. ANOVA for Figure 4.3 (b)

x	-0.039033	0.008378	-4.66	0.006

S = 0.6206 R-Sq = 81.3% R-Sq(adj) = 77.5%

Source	DF	SS	MS	F	P
Regression	1	8.3615	8.3615	21.71	0.006
Error	5	1.9260	0.3852		
Total	6	10.2875			

Appendix B 96. ANOVA for Figure 4.3 (c)

x	-0.046378	0.006766	-6.85	0.001

S = 0.5012 R-Sq = 90.4% R-Sq(adj) = 88.5%

Source	DF	SS	MS	F	P
Regression	1	11.804	11.804	46.99	0.001
Error	5	1.256	0.251		
Total	6	13.060			

Appendix B 97. ANOVA for Figure 4.3 (d)

x	-0.05697	0.01094	-5.21	0.003

S = 0.8106 R-Sq = 84.4% R-Sq(adj) = 81.3%

Source	DF	SS	MS	F	P
Regression	1	17.811	17.811	27.11	0.003
Error	5	3.285	0.657		
Total	6	21.096			

Appendix B 98. ANOVA for Figure 5.3 – Cumulative CO_2 – C mineralized

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	0.096	0.048	0.6850	0.0000
2	Factor A	6	29.891	4.982	71.0087	0.0000
-3	Error	12	0.842	0.070		
	Total	20	30.829			

Coefficient of Variation: 7.48%

Appendix B 99. ANOVA for Figure 5.6 – Cumulative N mineralized

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	4483.591	2241.796	28.0647	0.0000
2	Factor A	6	1137030.273	189505.046	2372.3857	0.0000
-3	Error	12	958.554	79.880		
	Total	20	1142472.419			

Coefficient of Variation: 1.35%

Appendix B 100. ANOVA for Table 5.5 – C:N Ratio (0 Week of incubation)

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	1	1.103	1.103	0.6586	0.0000
2	Factor A	7	40.725	5.818	3.4755	0.0612
-3	Error	7	11.717	1.674		
	Total	15	53.545			

Coefficient of Variation: 7.54%

Appendix B 101. ANOVA for Table 5.5 – C:N Ratio (2 Week of incubation)

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	1	2.103	2.103	1.2349	0.3032
2	Factor A	7	34.451	4.922	2.8908	0.0924
-3	Error	7	11.918	1.703		
	Total	15	48.471			

Coefficient of Variation: 8.99%

Appendix B 102. ANOVA for Table 5.5 – C:N Ratio (4 Week of incubation)

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	1	0.137	0.137	0.0949	0.0000
2	Factor A	7	33.043	4.720	3.2707	0.0703
-3	Error	7	10.103	1.443		
	Total	15	43.282			

Coefficient of Variation: 8.80%

Appendix B 103. ANOVA for Table 5.5 – C:N Ratio (6 Week of incubation)

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	1	6.503	6.503	6.0549	0.0434
2	Factor A	7	24.299	3.471	3.2323	0.0722
-3	Error	7	7.517	1.074		
	Total	15	38.319			

Coefficient of Variation: 7.89%

Appendix B 104. ANOVA for Table 5.5 – C:N Ratio (8 Week of incubation)

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	1	0.087	0.087	0.0584	0.0000
2	Factor A	7	19.470	2.781	1.8662	0.2146
-3	Error	7	10.433	1.490		
	Total	15	29.990			

Coefficient of Variation: 9.62%

Appendix B 105. ANOVA for Table 5.5 – C:N Ratio (10 Week of incubation)

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	1	4.000	4.000	2.3333	0.1705
2	Factor A	7	19.328	2.761	1.6106	0.2723
-3	Error	7	12.000	1.714		
	Total	15	35.328			

Coefficient of Variation: 10.42%

Appendix B 106. ANOVA for Table 5.5 - C:N Ratio (12 Week of incubation)

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	1	3.629	3.629	2.8597	0.1347
2	Factor A	7	22.407	3.201	2.5225	0.1227
-3	Error	7	8.883	1.269		
	Total	15	34.920			

Coefficient of Variation: 9.10%

Appendix B 107. ANOVA for Table 6.2 - %N

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	0.002	0.001	13.5714	0.0014
2	Factor A	5	0.014	0.003	41.1429	0.0000
-3	Error	10	0.001	0.0001		
	Total	17	0.017			

Coefficient of Variation: 9.30%

Appendix B 108. ANOVA for Table 6.2 - %K

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	0.000	0.000	0.4545	
2	Factor A	5	0.001	0.000	1.5455	0.2606
-3	Error	10	0.001	0.000		
	Total	17	0.002			

Coefficient of Variation: 13.39%

Appendix B 109. ANOVA for Table 6.2 - %P

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	0.001	0.001	2.5000	0.0317
2	Factor A	5	0.003	0.001	2.3333	0.1191
-3	Error	10	0.002	0.0001		
	Total	17	0.006			

Coefficient of Variation: 29.05%

Appendix B 110. ANOVA for Table 6.2 – $NH_4 - N$

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	3.307	1.654	9.0990	0.0056
2	Factor A	5	53.361	10.672	58.7194	0.0000
-3	Error	10	1.817	0.182		
	Total	17	58.486			

Coefficient of Variation: 6.76%

Appendix B 111. ANOVA for Table 6.2 – $NO_3 - N$

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	0.128	0.064	0.2669	0.0000
2	Factor A	5	5971.816	1194.363	4975.1313	0.0000
-3	Error	10	2.401	0.240		
	Total	17	5974.345			

Coefficient of Variation: 1.71%

Appendix B 112. ANOVA for Table 6.2 – Avail. P

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	5.333	2.667	4.0000	0.0529
2	Factor A	5	5066.680	1013.336	1520.0041	0.0000
-3	Error	10	6.667	0.667		
	Total	17	5078.680			

Coefficient of Variation: 2.42%

Appendix B 113. ANOVA for Table 6.2 – Exch. K

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	0.003	0.001	9.0000	0.0058
2	Factor A	5	0.053	0.011	71.2000	0.0000
-3	Error	10	0.001	0.0001		
	Total	17	0.058			

Coefficient of Variation: 6.12%

Appendix B 114. ANOVA for Table 6.2 – pH

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	0.998	0.499	14.0052	0.0013
2	Factor A	5	0.696	0.139	3.9083	0.0318
-3	Error	10	0.356	0.036		
	Total	17	2.050			

Coefficient of Variation: 3.61%

Appendix B 115. ANOVA for Table 6.3 – % N in carrot leaf

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	3.413	1.707	13.9130	0.0013
2	Factor A	5	4.897	0.979	7.9850	0.0029
-3	Error	10	1.227	0.123		
	Total	17	9.537			

Coefficient of Variation: 16.51%

Appendix B 116. ANOVA for Table 6.3 – % K in carrot leaf

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	0.572	0.286	26.8392	0.0001
2	Factor A	5	0.700	0.140	13.1411	0.0004
-3	Error	10	0.107	0.011		
	Total	17	1.379			

Coefficient of Variation: 7.48%

Appendix B 117. ANOVA for Table 6.3 – % P in carrot leaf

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	0.012	0.006	7.3673	0.0108
2	Factor A	5	0.081	0.016	19.8367	0.0001
-3	Error	10	0.008	0.001		
	Total	17	0.101			

Coefficient of Variation: 9.85%

Appendix B 121. ANOVA for Table 6.3 – % P in carrot root

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	0.041	0.020	54.2035	0.0000
2	Factor A	5	0.037	0.007	19.5664	0.0001
-3	Error	10	0.004	0.0001		
	Total	17	0.081			

Coefficient of Variation: 8.14%

Appendix B 122. ANOVA for Table 6.3 – % Ash in carrot root

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	0.555	0.277	13.9022	0.0013
2	Factor A	5	11.467	2.293	114.9558	0.0000
-3	Error	10	0.200	0.020		
	Total	17	12.221			

Coefficient of Variation: 2.73%

Appendix B 123. ANOVA for Table 6.4 – Root knot Index of carrot

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	0.963	0.482	11.5600	0.0025
2	Factor A	5	44.425	8.885	213.2400	0.0000
-3	Error	10	0.417	0.042		
	Total	17	45.805			

Coefficient of Variation: 7.80%

Appendix B 124. ANOVA for Table 6.5 – Mean root length of carrot

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	0.464	0.232	10.7108	0.0033
2	Factor A	5	21.023	4.205	194.0603	0.0000
-3	Error	10	0.217	0.022		
	Total	17	21.704			

Coefficient of Variation: 1.56%

Appendix B 125. ANOVA for Table 6.5 – Mean circumference of carrot root

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	0.120	0.060	0.4380	0.0000
2	Factor A	5	41.595	8.319	60.7226	0.0000
-3	Error	10	1.370	0.137		
	Total	17	43.085			

Coefficient of Variation: 3.86%

Appendix B 126. ANOVA for Table 6.5 – Mean root weight of carrot

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	3.203	1.602	17.0997	0.0006
2	Factor A	5	1301.205	260.241	2778.3753	0.0000
-3	Error	10	0.937	0.094		
	Total	17	1305.345			

Coefficient of Variation: 1.17%

Appendix B 127. ANOVA for Table 6.5 – Mean shoot weight of carrot

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	2.083	1.042	11.1210	0.0029
2	Factor A	5	273.666	54.733	584.3409	0.0000
-3	Error	10	0.937	0.094		
	Total	17	276.686			

Coefficient of Variation: 3.07%

Appendix B 128. ANOVA for Figure 6.1 – %N in Carrot leaves

Source	DF	SS	MS	F	P
Regression	1	0.0033468	0.0033468	98.09	0.001
Error	4	0.0001365	0.0000341		
Total	5	0.0034833			

Appendix B 129. ANOVA for Figure 6.1 – %N in Carrot roots

Source	DF	SS	MS	F	P
Regression	1	0.23291	0.23291	41.27	0.003
Error	4	0.02257	0.00564		
Total	5	0.25548			

Appendix B 130. ANOVA for Figure 6.2 – %K in Carrot leaves

Source	DF	SS	MS	F	P
Regression	1	0.15123	0.15123	7.36	0.053
Error	4	0.08217	0.02054		
Total	5	0.23340			

Appendix B 131. ANOVA for Figure 6.2 – %K in Carrot roots

Source	DF	SS	MS	F	P
Regression	1	0.053763	0.053763	8.36	0.044
Error	4	0.025720	0.006430		
Total	5	0.079483			

Appendix B 132. ANOVA for Figure 6.3 – %P in Carrot leaves

Source	DF	SS	MS	F	P
Regression	1	0.024300	0.024300	36.00	0.004
Error	4	0.002700	0.000675		
Total	5	0.027000			

Appendix B 133. ANOVA for Figure 6.3 – %P in Carrot roots

Source	DF	SS	MS	F	P
Regression	1	0.011408	0.011408	52.15	0.002
Error	4	0.000875	0.000219		
Total	5	0.012283			

Appendix B 134. ANOVA for Table 5.4 - % N mineralized

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	7.143	3.571	6.2500	0.0138
2	Factor A	6	1489.079	248.180	434.3148	0.0000
-3	Error	12	6.857	0.571		
	Total	20	1503.079			

Coefficient of Variation: 1.54%

Appendix B 135. ANOVA for Table 5.4 – Rate of N mineralized

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	65.146	32.573	7.3925	0.0081
2	Factor A	6	26514.962	4419.160	1002.9439	0.0000
-3	Error	12	52.874	4.406		
	Total	20	26632.982			

Coefficient of Variation: 2.09%

Appendix B 136. ANOVA for Table 5.4 – $\%CO_2$ mineralized

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	68.667	34.333	34.3333	0.0000
2	Factor A	6	2821.469	470.245	470.2449	0.0000
-3	Error	12	12.000	1.000		
	Total	20	2902.136			

Coefficient of Variation: 1.67%

Appendix B 137. ANOVA for Table 5.4 – Rate of CO_2 mineralized

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
1	Replication	2	0.003	0.001	23.0769	0.0001
2	Factor A	6	0.684	0.114	1842.2303	0.0000
-3	Error	12	0.001	0.0001		
	Total	20	0.688			

Coefficient of Variation: 2.82%