

**EFFECT OF *Trichoderma harzianum* ON TOMATO (*Solanum lycopersicum* L.) SEED  
GERMINATION, SEEDLING VIGOUR AND ROOT-KNOT INCIDENCE**

**BY**

**MATTHIAS KORLE CONRAD OSSOM (10555217)**

**WEST AFRICA CENTRE FOR CROP IMPROVEMENT COLLEGE OF BASIC AND  
APPLIED SCIENCES UNIVERSITY OF GHANA**

**LEGON**



**JULY, 2017**

**EFFECT OF *Trichoderma harzianum* ON TOMATO (*Solanum lycopersicum* L.) SEED  
GERMINATION, SEEDLING VIGOUR AND ROOT-KNOT INCIDENCE**

**BY**

**MATTHIAS KORLE CONRAD OSSOM (10555217)**

**THIS THESIS IS SUBMITTED TO THE UNIVERSITY OF GHANA, LEGON IN  
PARTIAL FULFILMENT OF THE AWARD OF DEGREE OF MASTER OF  
PHILOSOPHY IN SEED SCIENCE AND TECHNOLOGY**

**WEST AFRICA CENTRE FOR CROP IMPROVEMENT COLLEGE OF BASIC AND  
APPLIED SCIENCES UNIVERSITY OF GHANA  
LEGON**

**JULY, 2017**

**DECLARATION**

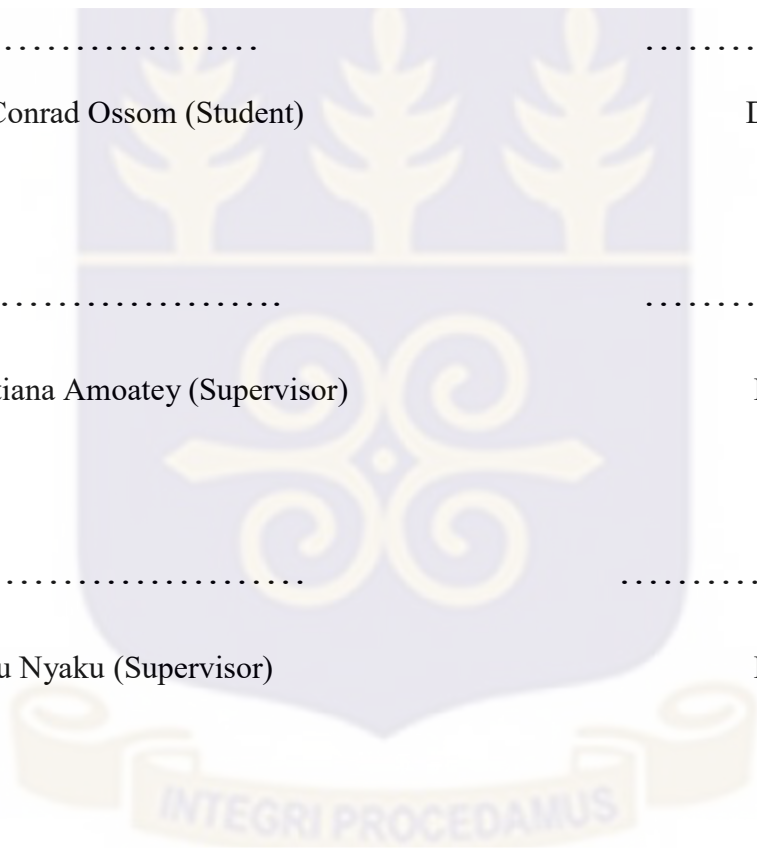
I, Matthias Korle Conrad Ossom, hereby do declare that, except for references to the works of other researchers, which have been duly acknowledged, this thesis is the outcome of my own original study under supervision and has not been submitted elsewhere either in whole or in part for the award of any degree.

.....  
Matthias Korle Conrad Ossom (Student) Date

.....  
Dr. (Mrs.) Christiana Amoatey (Supervisor) Date

.....  
Dr. Seloame Tatu Nyaku (Supervisor) Date

.....  
Dr. Agyemang Danquah (Supervisor) Date



**ABSTRACT**

Tomato is a major crop of high economic importance cultivated worldwide with high yields per unit area of land compared with other tropical vegetables. One of the major challenges with production in the tropics is the infestation of root-knot nematodes. Laboratory and field experiments were carried out to study the effect of different concentrations as well as different durations of exposure of *Trichoderma harzianum* (Eco-T) as seed treatment to manage root knot nematode and to evaluate *T. harzianum* effects on seed germination and seedling performance on tomato (UC 82 variety) conducted in the dry season from February 2017 to May 2017 in the Greater Accra region of Ghana. A Completely Randomised Design (CRD) was used in the laboratory study. It comprised 25 seeds sown in per Petri-dish replicated four times. The effect of the seeds coated with *Trichoderma harzianum* (Eco-T) at concentrations of 0 g, 0.2 g, 0.25 g, 0.3 g, 0.35 g, and 0.4 g/10 g of seed prior to sowing indicated that *T. harzianum* (Eco-T) at the rates of 0.2 g and 0.4 g/10 g of seed produced higher germination percentage, root and shoot lengths of seedlings and seedling vigour. A follow up study arranged in a 3x2 factorial treatment combination laid out in CRD to evaluate the different durations of Eco-T at 30 min, 1hr, 3 hrs, 6 hrs, 8 hrs, 24 hrs, 30 hrs, 48 hrs, and no coating using the recommended rate (0.2 g/10g seeds) showed treatment for 30 minutes and 3 hrs, giving higher seed germination, root and shoot lengths, and seedling vigour respectively. A combination of concentrations (0 g, 0.2 g, and 0.4 g/10 g of seed) and exposure duration (30 min and 3 hrs) resulted in optimum germination, root and shoot lengths as well as seedling vigour for 0.4 g/10 g of seed at 30 minutes exposure. A field study arranged in Randomized Complete Block Design (RCBD) with 4 replications was employed to study the influence of *T. harzianum* (Eco-T) as seed treatment against root-knot nematodes. Velum (Fluopyram 400 SC) at a rate of 3.8 mL/7 L of water was used as a standard reference product. Impact of the treatments on seedling emergence, plant growth, incidence of nematode root knots, nematode populations, egg masses, and yield of UC 82 tomato were recorded. *Trichoderma harzianum* (Eco-T) treatments were effective in reducing nematode populations

and root galling, comparable to the Velum (Fluopyram 400 SC). Plant growth was significantly influenced by both *T. harzianum* and Velum (Fluopyram 400 SC) treatments. Yield however was higher in Velum (Fluopyram 400 SC) although the differences were not significant compared to *T. harzianum*. A partial budget analysis suggested application of *T. harzianum* to as cost-effective option compared to Velum (Fluopyram 400 SC).



## DEDICATION

This work is dedicated to the Lord God Almighty who made me and brought me this far. To Him be all the glory and adorations. Amen.

It is also dedicated to my parents; Mr. Christopher Tettey Ossom and Mrs. Rose Arkor Ossom. Finally, to my siblings; Leo Tetteh Ossom, Elsbeth Korlekie Ossom and Ilona Korlekwor Ossom for their support in prayers, love and encouragement.



## ACKNOWLEDGEMENTS

My help always comes from the Lord God who made the heavens and the earth for His mercies endure forever and they are new every morning. I bless the Lord with all that is in me for He has done exceedingly, abundantly above all that I could ever think of or ask of Him. I thank Him for His protection, favour and blessings He bestowed on me throughout my lifetime and especially throughout this program.

My profound gratitude goes to the Director of the West African Centre for Crop Improvement, Professor Eric Danquah of University of Ghana, Legon for his leadership and Innovative skills that conceived the Plant breeding program at the PhD level and the Seed Science and Technology program at the MPhil level. The latter of which I am pleased to be part of the first cohort. His enormous success has got me a slot to obtain this training and degree. Thank you, Sir and God bless, you.

I am indebted to my supervisors, Dr. (Mrs.) Christiana Amoatey, Dr. Seloame Tatu Nyaku and Dr. Agyemang Danquah for their selfless, relentless and awe-inspiring efforts made through helpful mused suggestions, constructive and cardinal critics and corrections, encouragements and tireless guidance that made this work a success.

Sincere thanks to all the Heads of Departments and lecturers of the College of Basic and Applied Sciences of the University of Ghana for their contributions made to my training one way or another. Big thanks to all.

To the technicians at the Department of Crop Science Pathology laboratory especially Mr. Samuel Osabutey, and those in the Department of Soil Science especially Mr. Adusei Okra for their friendly and fatherly advice given me during the execution of this work and timely duty discharge whenever they were needed.

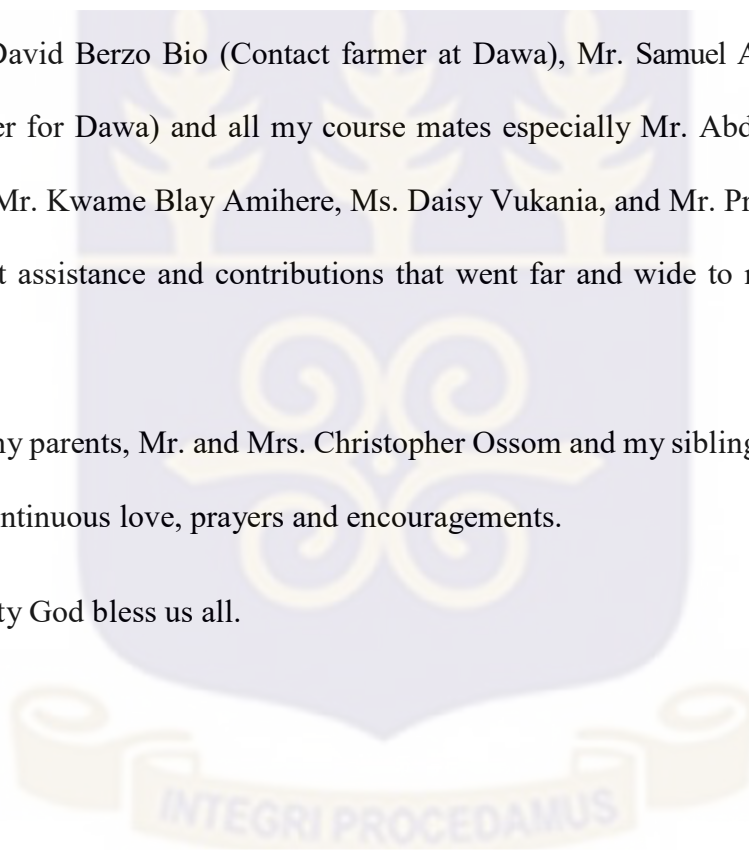
I am also very grateful to the entire staff of Regional Marketing Group (RMG) Ghana Limited for their support and real-world exposure opportunity given me as a seeds expert.

My gratitude also to Mr. Eric Quaye, Director of the Ghana Seed Inspection Division (GSID) of the Plant Protection and Regulatory Service Directorate (PPRSD) at Pokuase for allowing me to use his equipped and internationally accredited laboratory to conduct the first and second parts of my research. To his entire staff, especially Ms. Veronica Nipaah Foster (supervisor) and Mrs. Linda Esther Kortey to mention a few, goes very big thanks for their support, supervision and suggestions.

Thanks to Mr. David Berzo Bio (Contact farmer at Dawa), Mr. Samuel Amoah (Agricultural Extension Officer for Dawa) and all my course mates especially Mr. Abdul Isah Rashid, Mr. Evans Agortey, Mr. Kwame Blay Amihere, Ms. Daisy Vukania, and Mr. Prince Appiah Eburey for their intellect assistance and contributions that went far and wide to make my research a success.

Finally, I bless my parents, Mr. and Mrs. Christopher Ossom and my siblings, Leo, Elsbeth, and Ilona for their continuous love, prayers and encouragements.

May the Almighty God bless us all.



**TABLE OF CONTENTS**

DECLARATION .....	I
ABSTRACT .....	II
DEDICATION .....	IV
ACKNOWLEDGEMENTS .....	V
TABLE OF CONTENTS .....	VII
LIST OF FIGURES .....	XI
LIST OF TABLES .....	XII
LIST OF ABBREVIATIONS .....	XIV
CHAPTER ONE .....	1
1.0 INTRODUCTION .....	1
CHAPTER TWO .....	6
2.0 LITERATURE REVIEW.....	6
2.1 ORIGIN AND DISTRIBUTION OF TOMATO .....	6
2.2 BOTANY OF TOMATO .....	6
2.3 TOMATO GENOTYPE.....	8
2.3.1 TOMATO SIZES .....	8
2.3.2 TOMATO TYPES.....	9
2.4 NUTRITIONAL VALUE AND OTHER HEALTH BENEFITS OF TOMATO.....	9
2.5 PRODUCTION OF TOMATO.....	10
2.5.1 WORLD PRODUCTION OF TOMATO .....	10
2.5.2 TOMATO PRODUCTION IN GHANA.....	11
2.5.3 SOIL REQUIREMENTS .....	12
2.5.4 CLIMATIC REQUIREMENTS .....	13
2.6 CAUSES OF TOMATO PRODUCTION LOSSES.....	13
2.6.1 BIOTIC FACTORS .....	14
2.6.2 ABIOTIC FACTORS.....	15
2.7 <i>TRICHODERMA HARZIANUM</i> .....	16
2.7.1 <i>TRICHODERMA HARZIANUM</i> AS SEED TREATMENT .....	16
2.7.2 FACTORS THAT CAUSE LOSS OF <i>TRICHODERMA HARZIANUM</i> .....	19
2.8 PARTIAL BUDGET ANALYSIS.....	19
CHAPTER THREE .....	21
3.0 MATERIALS AND METHODS .....	21
3.1 STUDY SITES .....	21
3.1.2 CLIMATIC AND EDAPHIC CONDITIONS OF THE FIELD.....	21

<b>3.2 SOURCES OF MATERIALS .....</b>	<b>22</b>
<b>3.3 METHODOLOGY .....</b>	<b>22</b>
<b>3.4 LABORATORY ANALYSES.....</b>	<b>23</b>
<b>3.4.1 EXPERIMENTAL DESIGN.....</b>	<b>23</b>
<b>3.4.2 SEED GERMINATION AND SEEDLING VIGOUR INDEX AS AFFECTED BY DURATION OF EXPOSURE OF TOMATO SEEDS TO <i>TRICHODERMA HARZIANUM</i> (ECO-T)<sup>®</sup>.....</b>	<b>23</b>
<b>3.4.3 APPLICATION OF DIFFERENT CONCENTRATION OF <i>TRICHODERMA HARZIANUM</i> (ECO-T)<sup>®</sup> TO TOMATO SEEDS PRIOR TO SOWING .....</b>	<b>24</b>
<b>3.4.4 SEED GERMINATION AND SEEDLING VIGOUR AS AFFECTED BY DURATION OF ECO-T EXPOSURE TO TOMATO SEEDS AT DIFFERENT CONCENTRATIONS. ....</b>	<b>24</b>
<b>3.4.5 EXPERIMENTAL DESIGN.....</b>	<b>25</b>
<b>3.4.6 DATA COLLECTED FOR LABORATORY EXPERIMENTS 1 – 3. ....</b>	<b>25</b>
<b>3.4.7 DATA ANALYSIS .....</b>	<b>25</b>
<b>3.5 FIELD WORK .....</b>	<b>26</b>
<b>3.5.1 IMPACT OF <i>TRICHODERMA HARZIANUM</i> ON NEMATODE POPULATION AND TOMATO (UC 82) SEEDLING PERFORMANCE IN THE FIELD. ....</b>	<b>26</b>
<b>3.5.2 TREATMENTS: .....</b>	<b>26</b>
<b>3.5.3 LAND PREPARATION AND FIELD LAYOUT.....</b>	<b>26</b>
<b>3.5.4 EXPERIMENTAL DESIGN.....</b>	<b>26</b>
<b>3.5.5 TREATMENT APPLICATION AND PLANTING .....</b>	<b>27</b>
<b>3.5.6 CULTURAL PRACTICES .....</b>	<b>27</b>
<b>3.6 SOIL AND TOMATO PLANT SAMPLING .....</b>	<b>27</b>
<b>3.7 DETERMINATION OF SOIL CHEMICAL AND PHYSICAL PROPERTIES.....</b>	<b>28</b>
<b>3.7.1. TOTAL NITROGEN .....</b>	<b>28</b>
<b>3.7.2 SOIL PH.....</b>	<b>29</b>
<b>3.7.3 AVAILABLE PHOSPHORUS.....</b>	<b>30</b>
<b>3.7.4 ORGANIC CARBON .....</b>	<b>31</b>
<b>3.7.5 EXCHANGEABLE CATIONS: EXTRACTION .....</b>	<b>31</b>
<b>3.7.6 POTASSIUM (K) AND SODIUM (NA) DETERMINATION .....</b>	<b>31</b>
<b>3.7.7 CALCIUM (CA) AND MAGNESIUM (MG) DETERMINATION .....</b>	<b>32</b>
<b>3.7.8 PARTICLE SIZE DISTRIBUTION .....</b>	<b>32</b>
<b>3.7.9 DATA COLLECTED AND REPORTED.....</b>	<b>33</b>
<b>3.8 NEMATODE PARAMETERS .....</b>	<b>33</b>
<b>3.8.1 NEMATODE EXTRACTION AND COUNTING .....</b>	<b>33</b>
<b>3.8.2 NEMATODE IDENTIFICATION .....</b>	<b>34</b>
<b>3.8.3 ROOT-KNOT NEMATODE GALL SCORING.....</b>	<b>35</b>
<b>3.8.4 NEMATODE EGG-MASSSES .....</b>	<b>35</b>

3.8.5 DATA COLLECTED AND REPORTED.....	35
3.9 PLANT PARAMETERS.....	36
3.9.1 SEEDLING HEIGHT (CM) .....	36
3.9.2 NUMBER OF LEAVES.....	36
3.9.3 STEM GIRTH (CM).....	36
3.9.4 ROOT LENGTH (CM).....	36
3.9.5 FRESH WEIGHT OF SHOOTS (G).....	36
3.9.6 DRY WEIGHT OF SHOOTS (G).....	37
3.9.7 FRESH WEIGHT OF ROOTS (G).....	37
3.9.8 DRY WEIGHT OF ROOTS (G).....	37
3.9.9 DATA COLLECTED AND REPORTED.....	37
3.10 DATA ANALYSES .....	38
CHAPTER FOUR.....	39
4.0 RESULTS .....	39
4.1 LABORATORY ANALYSIS .....	39
4.1.1 SEED GERMINATION, SHOOT LENGTH, ROOT LENGTH AND SEEDLING VIGOUR INDEX AS AFFECTED BY DURATION OF EXPOSURE OF TOMATO SEEDS TO <i>TRICHODERMA HARZIANUM</i> AS SEED TREATMENT.....	39
4.1.2 SEED GERMINATION, SHOOT LENGTH, ROOT LENGTH AND SEEDLING VIGOUR INDEX AS AFFECTED BY DIFFERENT CONCENTRATIONS OF <i>TRICHODERMA HARZIANUM</i> USED AS SEED TREATMENT ON UC 82 TOMATO SEEDS .....	40
4.1.3 SEED GERMINATION, SEEDLING SHOOT LENGTH, SEEDLING ROOT LENGTH AND SEEDLING VIGOUR INDEX AS AFFECTED BY DURATION OF EXPOSURE OF UC 82 TOMATO SEED TO <i>TRICHODERMA HARZIANUM</i> AT DIFFERENT CONCENTRATIONS.....	41
4.2.1 PHYSICAL AND CHEMICAL SOIL ANALYSIS OF STUDY AREA. ....	44
4.2.2 IMPACT OF ECO-T ( <i>TRICHODERMA HARZIANUM</i> ) ON UC 82 TOMATO SEEDLING PERFORMANCE.....	45
4.2.2.1 EFFECT OF <i>TRICHODERMA HARZIANUM</i> ON SEEDLING EMERGENCE 2 WEEKS AFTER SOWING.....	45
4.2.2.2 EFFECT OF <i>TRICHODERMA HARZIANUM</i> ON TOMATO PLANT PERFORMANCE .....	45
4.2.2.3 EFFECT OF <i>TRICHODERMA HARZIANUM</i> ON TOMATO ROOT GALLING INDEX 6 WEEKS AFTER.....	53
4.2.2.4 EFFECT OF <i>TRICHODERMA HARZIANUM</i> ON TOMATO ROOT GALLING INDEX AT 12 WEEKS AFTER.....	54
4.3 PARTIAL BUDGET ANALYSIS FOR USING TREATMENTS.....	57
CHAPTER FIVE .....	59
5.0 DISCUSSION .....	59
5.1 IMPACT OF <i>TRICHODERMA HARZIANUM</i> DURATION OF EXPOSURE, CONCENTRATION AND COMBINATION OF EXPOSURE TIME AND CONCENTRATION ON SEED GERMINATION AND SEEDLING VIGOUR .....	59

<b>5.2 EXPERIMENT 2: FIELD STUDY</b> .....	<b>60</b>
<b>5.2.1 PHYSICAL AND CHEMICAL ANALYSES OF FIELD SOIL</b> .....	<b>60</b>
<b>5.2.2 IMPACT OF TRICHODERMA HARZIANUM AND VELUM ON NEMATODE POPULATION, EGG MASSES AND TOMATO ROOT GALLING</b> .....	<b>60</b>
<b>5.2.3 IMPACT OF <i>TRICHODERMA HARZIANUM</i> AND VELUM ON TOMATO VEGETATIVE GROWTH AND YIELD</b> .....	<b>61</b>
<b>5.3 ECONOMIC BENEFITS ANALYSIS OF <i>TRICHODERMA HARZIANUM</i> AND VELUM TREATMENTS WITH A PARTIAL BUDGET</b> .....	<b>63</b>
<b>CHAPTER SIX</b> .....	<b>64</b>
<b>6.0 CONCLUSIONS AND RECOMMENDATIONS</b> .....	<b>64</b>
<b>6.1 CONCLUSIONS</b> .....	<b>64</b>
<b>6.2 RECOMMENDATIONS</b> .....	<b>65</b>
<b>REFERENCES</b> .....	<b>66</b>
<b>APPENDICES</b> .....	<b>76</b>



**LIST OF FIGURES**

**Figure 1: Root galling indices at 6 WAA as influenced by seed treatments .....53**

**Figure 2: Presence of nematode root galls on tomato seedlings in (A) Eco-T (*T. harzianum*) plots; (B) control plot showing heavy galling and (C) velum plot. ....54**

**Figure 3: Root galling indices at 12 WAA as influenced by seed treatments .....55**

**Figure 4: Location of Pokuase.....78**

**Figure 5:Location of Dawa .....78**

**Figure 6: Gall Scoring chat .....79**



LIST OF TABLES

Table 1: Tomato types and their characteristics .....	9
Table 2: World’s leading tomato producers .....	11
Table 3: Soil temperature and days to emergence .....	13
Table 4: Sample Partial Budget with Components .....	38
Table 5: Seed germination, shoot length, root length and seedling vigour index as affected by duration of exposure of tomato seeds to <i>Trichoderma harzianum</i> as seed treatment. ....	40
Table 6: Seed germination, shoot length, root length and seedling vigour index as affected by concentration effect of <i>Trichoderma harzianum</i> used as seed treatment on UC 82 tomato seeds .....	41
Table 7: Seed germination, seedling shoot length, seedling root length and seedling vigour index as affected by duration of exposure of UC 82 tomato Seed to <i>Trichoderma harzianum</i> at different concentrations.....	43
Table 8: Soil physical and chemical analysis .....	44
Table 9: Effect of <i>Trichoderma harzianum</i> on tomato seedling emergence .....	45
Table 10: Effect of <i>Trichoderma harzianum</i> on tomato plant height and plant stem girth at 4 and 8 weeks after application of treatment (WAA).....	46
Table 11: Effect of <i>Trichoderma hazianum</i> on tomato leaves and branches at 4 and 8 weeks after application (WAA).....	47
Table 12: Effect of <i>Trichoderma harzianum</i> on tomato roots at 6 and 12 weeks after treatment application (WAA) .....	48
Table 13: Effect of <i>Trichoderma harzianum</i> on fresh weights at 6 and 12 weeks after application of treatment (WAA) .....	49
Table 14: Effect of <i>Trichoderma harzianum</i> on dry weights at 6 and 12 weeks after treatment application (WAA) .....	50
Table 15: Effect of <i>Trichoderma harzianum</i> on Nematode Population and Egg- mass at 6 and 12 Weeks After Treatment Application (WAA) .....	52
Table 16: Effect of <i>Trichoderma harzianum</i> treatments on tomato yield .....	56
Table 17: Eco-T ( <i>Trichoderma harzianum</i> ) as seed treatment or coating against untreated plots (Control).....	57
Table 18: Partial budget for using velum (fluopyram SC 400) as soil drenching against untreated plot (control).....	58
Table 19: Nutrition value per100g of tomato.....	76

**Table 20: Sample partial budget with components.....77**  
**Table 21: Gall score description .....80**



## LIST OF ABBREVIATIONS

AD	After Death of Christ
CRD	Complete Randomized Design
cm	Centimetres
Cmol	Concentration mol
CV	Coefficient of Variation
EDTA	Ethylene Diamine Tetraacetic Acid
F.A.O.	Food and Agriculture Organization
g	Grams
GSID	Ghana Seed Inspection Division
H <sub>2</sub> O	Water
Hg/ha	Hectogram per Hectare
Kg	Kilogram
L	Litre
LSD	Least Significant Difference
MCF	Moisture Correction Factor
mg	Milligram
mL	Millilitres
nm	Nanometre
NLEA	Nutritional Labelling and Education Act

PPRSD	Plant Protection and Regulatory Service Directorate
PPNs	Plant Parasitic Nematodes
RCBD	Randomized Complete Block Design
RMP	Rounds Per Minute
MDSP	Modified Diagonal Sampling Pattern
USDA	United States Department of Agriculture



## CHAPTER ONE

### 1.0 INTRODUCTION

Tomato (*Solanum lycopersicum*) belongs to the nightshade family, Solanaceae. It is a major horticultural crop that is cultivated on about 5,023,810 hectares of land worldwide for its edible fruits (FAOSTAT, 2014). Its worldwide production is estimated to be 170,750,767 tonnes (FAOSTAT, 2014) and it's one of the world's most consumed vegetable under production after potato (Panthee *et al.*, 2009). Tomato, which is also an herb is usually a sprawling plant but may be pruned and staked to enhance fruit quality. The crop is of high economic importance in many countries because it takes shorter time to harvest and as well gives a higher yield per a small area of cultivation (Obeng-Ofori *et al.*, 2007).

Tomato provides important food nutrients and other healthy and beneficial food supplements for the body when consumed because of its rich source of most vitamins including A, B, C, K, amino acids (tryptophan, threonine, leucine, lysine, valine, glutamic acid etc.), proximates (Protein, fibre, sugars, carbohydrates, energy, water, etc.), minerals (Calcium, iron, phosphorus, magnesium etc.) and some lipids (USDA, 2016). Carotenoids are the most prominent nutrients in tomatoes and specifically with lycopene as the most abundant among beta-carotene, gamma-carotene and other minor carotenoids (USAD, 2016).

In Ghana, tomato is economically attractive and the land area under cultivation by many households is on the rise in all the agro-ecological zones (Diao, 2010).

As at 2012, the production of tomato in Ghana from a total land area of approximately 44,750 hectares stood at approximately 340,218 tonnes with net production value of 128,162.91 dollars (FAOSTAT, 2013). Despite the high production levels of the crop locally, the importation of both

fresh and processed tomato into the country (SEND Ghana, 2008) mostly from neighbouring country Burkina Faso continues to go up especially during the off season.

Due to the high susceptibility to over two hundred diseases including root-knot, anthracnose, early blight, late blight, *Fusarium* wilt, damping off, stem rot, Septoria leaf spot, Verticillium wilt, gray mould, powdery mildew, blossom end rot, fruit cracking etc. caused by several pathogens including bacteria, fungi, nematodes and viruses, tomato production requires a lot more attention than is often given (Lukyanenko, 1991).

Diseases caused by nematodes are often the most difficult to manage because the above ground symptoms of the crop nematodes attack usually result from below ground infection by plant parasitic nematodes whose effects become evident if the nematode population is high and there is great reduction in yield (Mai, 1977).

The level and magnitude of damage caused by nematodes relies on a broad spectrum of factors both biotic and abiotic. The biotic factors include the species or strain's virulence, population density, resistance or tolerance capacity of the host plant and incidence of other pests and diseases. On the other hand, the abiotic factors include climate, soil conditions such as fertility. and the presence or absence of water in the soil (Coyne *et al.*, 2007).

In most experiments, improved plant development and yields are attributed to an alleviation of plant disease. This is achieved using a myriad of control measures including cultural, chemical, biological, and integrated methods. *Trichoderma harzianum* has been utilized as a biological agent to improve crop growth regardless of prevalent crop disease (Baker *et al.*, 1984). It's modes of operation include competition, parasitism, production of pathogen restrictive compounds and enzymes (Lester, 2010). *Trichoderma* spp. have many positive effects on crops which include elevated rate and percentage of seed germination, improved efficiency of plant nutrient uptake from fertilizers and other soil

decompositions. These result in improved systemic resistance of plants to diseases which improves plant growth and yield. In view of this, low yields recorded for tomato production in Ghana could be circumvented if some prevailing pests such as root-knot nematodes causing root-knot disease is managed effectively (Harman *et al.*, 2004).

Plant-parasitic nematodes (PPNs) cause various crop losses throughout the world and are recognized as prime agricultural pathogens. The most destructive plant-parasitic nematode spp. are the root-knot nematodes and they cause the most severe destruction to many crops including tomato (Baker, 1985). Thus, agricultural production worldwide has been greatly affected with decreases of not less than 11% of total yields being attributed to the plant- parasitic nematodes resulting in loss of millions of dollars (Agrios, 2005).

Anastasiadis *et al.* (2008) reported that, “application of chemical nematicide has been found as an effective control of nematodes but with high toxic residual effects on the environment and particularly on non-target organisms”, therefore, the employment of less toxic organic amendments for nematode control appears more sustainable.

Literature on organic amendments employed to suppress plant-parasitic nematodes densities show promising but inconsistent results (Muller and Gooch, 1982; Rodri'guez-Ka' bana,1986; Stirling, 1991; McSorley and Gallaher, 1995; Hassan *et al.*, 2010). However, no single solution is enough for improving seed treatments' ability to establish and perform consistently in the field (O'Callaghan, 2016).

In Ghana, *Meloidogyne incognita* has been reported to be parasitizing tomato in Ashanti, Brong Ahafo, and Upper East regions of the country (Osei *et al.*, 2011). Dawa, a suburb of Greater Accra Region of Ghana is noted for production of tomato and other vegetable crops. However, root-knot nematodes have been identified to be among the major challenges the farmers encounter especially

in tomato production, hampering the growth and development of the crop (Personal Conversation, 2016).

Ghana still imports about 27,000 tonnes of fresh and processed tomato and is second to Germany (Aryeetey 2006; Yeboah, 2011) despite efforts by both commercial and smallholder farmers to increase production to meet local market demands (Osae, 2014).

Several crops in which *T. harzianum* has been used to manage pathogens thereby increasing crop yields have been reported in literature (Agrios,1997, Abdulai *et al.*, 2013). *T. harzianum* significantly increased yields of various crops such as cereals and legumes, from 5% to about 230% (Abdulai *et al.*, 2013). However, not much has been reported on its use in vegetable production. Notwithstanding, increasing demand for pesticide-free vegetables warrants the need for alternative approaches to pest management in vegetable crop production.

Currently, there is insufficient information on the connection between degree of nematode interference and yield for a wide range of crops as well as information on different types of nematodes associated with crops. There is also the need for increased research to address emerging issues from promising but inconsistent studies regarding the capability of organic amendments to manage plant-parasitic nematodes (Muller and Gooch, 1982; Rodri' guez-Ka' bana, 1986; Stirling, 1991; McSorley and Gallaher, 1995; Hassan *et al.*, 2010).

To overcome challenges posed by various nematode species in tomato production, research using different control methods with high adoption potential and hence enhance tomato production especially in the Dawa suburb in the Greater Accra Region are needed.

The object of this research is to evaluate the effect of *T. harzianum* on nematode infestation growth and yield of tomato compared to chemical control.

The specific objectives of the study were therefore to:

- ❖ assess the effect of *T. harzianum* levels on tomato seed germination and seedling vigour

- ❖ evaluate effect of *T. harzianum* levels on nematode population and root galling
- ❖ evaluate effect of *T. harzianum* levels on tomato yield
- ❖ estimate the benefit associated with using *T. harzianum* and Velum (Fluopyram 400 SC G) in tomato production using the partial budget analysis tool.



## CHAPTER TWO

### 2.0 LITERATURE REVIEW

#### 2.1 Origin and distribution of tomato

Among the sprawling plants of the nightshade family, Tomato species (*Solanum lycopersicum*) is cultivated for its edible fruits worldwide (Peralta and Spooner 2001).

Tomato originated from the Western South along the coast and high Andes which extended from Central Ecuador through Peru, Colombia, Venezuela, Bolivia to Northern Chile and Argentina where they occurred as wild types and were first cultivated by the Incas and Aztecs as early as 700 A.D (Oncken, *et al.*, 2006). The crop was introduced for cultivation in Africa via Southern Africa by the Portuguese in 1850 (Blanca *et al.*, 2013)

Early cultivars introduced were seemingly yellow instead of red in colour and in most places, the tomatoes were remarkably slow in gaining acceptance, except as an ornamental curiosity (Basset, 1986).

Its history as a commercially processed commodity began at the Lafayette College at Easton, Pennsylvania, in 1847 where the commercial ‘canning’ possibilities of the “love apple” was first demonstrated and has become the leading processed vegetable crop in the U.S. (Basset, 1986).

#### 2.2 Botany of tomato

Tomato, also known as the “edible wolf-peach” (Cutler, 1998), belongs to the family, Solanaceae and Linnaeus placed it in the genus *Solanum* as *Solanum lycopersicum* L. in 1753. The ‘*lycopersicum*’ came about from the derivatives: ‘lyco’ which means wolf, and ‘persicum’ which means peach. Although the scientific name of tomato underwent some variations, the name *Solanum lycopersicum* has been maintained.

The typical height of tomato plant is around 1–3 meters tall. The plant is identified with a sprawling weak stem that sprawls with pubescent vines. The hairs associated with the tomato plants facilitate

vining process. In situations where original roots are damaged, new roots form or sprout wherever the plant gets in contact with the soil (Cox, 2000).

In spite of the fact that it is regularly cultivated outside in mild climates as an annual crop, tomato is a perennial in its natural habitat. Tomato plants are either determinate or indeterminate. While the determinate plants have their terminal buds becoming flowers with their axillary shoots suppressed, indeterminate plants have their terminal buds continuing into vegetative growth without flowers (Smith *et al.*, 1994).

Indeterminate plants are long-vined plants having a blossom bunch that occurs at each third internode along the fundamental hub which bears fruits continuously and grow indefinitely as long as that environmental condition stays favourable. Determinates on the other hand are short-vined plants and sometimes referred to as “self-pruning.” These have bloom bunches that occur at practically every node on which blossoms and fruits develop. Growth proceeds along the axis until the point when terminal bunch forms then development of that specific branch ends (McCormack, 2004). Due to a greater number of blossom groups per length of vine, determinate plants tend to bear and grow into a crop within a shorter timeframe. The amount of blooms per bunch are regularly 4 -5. In little-fruited cherry tomatoes for example, quantity of flowers in a bunch can vary from 8- 12. Some varieties like ‘Riesentraube’ have piles of flowers per cluster but not necessarily having all of them bearing fruits (McCormack 2004).

The style is encompassed by anthers in the tomato flowers that fuse together to form tubes. Flowers in local cultivars have a tendency to be self-fertilizing and these blooms are 1 – 2 cm, yellow, with five pointed projections on the corolla and are borne together in a cyme of 3 - 12 (Acquaah, 2005).

Most tomato plants are referred to as regular leaf plants simply because they have compound leaves while others are referred to as potato leaf plants due to the simple nature of the leaves. Irregular-pinnate tomato leaves are usually 10 – 25 cm long. The leaflets on the petioles are normally 5 - 9 in

number with each notch margined leaflet about 8 cm long. A typical feature about the leaves and stem of tomato plants are that they are thickly hairy (Acquaah, 2005).

The root system of tomato plant is mostly vigorously extensive and is located primarily in the first 30 cm of the soil. These plants have a solid taproot that can develop as long as about 185 cm. Most of the time, tomatoes root systems are fibrous. This is a result of the breakage suffered by the taproots during transplanting. Majority of the horizontal roots develop 5 to 25 cm deep subsurface of the soil (McCormack, 2004). A dense lateral root system develops some few days after transplanting (Norman, 1992).

### **2.3 Tomato genotype**

Tomatoes are valued for their bright red colour but that notwithstanding they can also be black, green, pink, purple, white, yellow and sometimes multicoloured. There are so many genotypes of tomatoes marketed today unlike before the last 15 years where the types of tomato for cultivation were basically only the processor and fresh market types. Mostly, open-pollinated tomato varieties that are cultivated by local farmers are imported from other countries. Cherry, Cun Dayri, Eva T-89, Feston, Roma, Money maker, Nemadina, Pakit, and Peelo are some varieties of tomato that are produced globally

#### **2.3.1 Tomato sizes**

Tomato fruits differ in shapes and sizes. An example is the 'currant' tomato which is approximately 0.6 – 0.9 cm thick produces both yellow and red fruits (McCormack 2004); tom berries are about 5 cm thick, cherry tomatoes are about 1- 2 cm in diameter similar in size to the wild tomato, and finally the beefsteak tomatoes which are approximately  $\geq 10$  cm (Freeman *et al.*, 2016).

There are about four (4) main sizes of tomato under cultivation in Florida, USA. They are the large fruited and beefsteak types such as the Amelia, Florida 47 and Red morning; the Cherry types such

as Sweet Treats, Camelia and Sakura; the Plum and Roma types such as Mariana, Moniticello and Supremo; finally, the grape types such as Brixmore, Cupid and Jolly Girl (Freeman *et al.*, 2016).

### 2.3.2 Tomato types

Table 1 below shows the groupings of tomato varieties as used in South Africa. According to Adu-Dapaah *et al.* (2002), "Laurano 70", "Racci", "Zaussi", "Power Laurano", and "Power rasta" were mostly grown in Ghana. Currently in Ghana, "Roma", "UC 82", "Pectomech", "Tropimech", "Jaguar", "Heinz", "Ada lorry tyre" among others are those mostly cultivated on a large scale for both local and foreign markets. These cultivated varieties fit mainly into the South African classification of determinate varieties.

**Table 1: Tomato types and their characteristics**

DETERMINATE TYPES	INDETERMINATE TYPES
Fresh large and medium sizes with long shelf life	Fresh large, medium and small sizes with long shelf life
Fresh round and oval red cherries	Fresh round and oval red cherries
Fresh cylindrical and blocky saladette	Fresh oval yellow cherries
Processors	Fresh round and oval yellow cocktail
Fresh tomatoes suitable for non-trellising	Fresh round and oval red cocktail
Fresh cylindrical and blocky saladette	
Fresh truss tomatoes	

*Source: <http://www.Starkeayeres.co.za>, tomato pdf*

### 2.4 Nutritional value and other health benefits of tomato

Although low in cholesterol, saturated fat and sodium, tomatoes are major sources of multivitamins such as vitamins A, B and C and also contain some amounts of Fe, K and P elements. Tomato's medicinal properties were endorsed in Continental Europe in the 16th century. Its ingesting was believed to profit the heart and other mammalian organs due to its lycopene content. Heart ailment, cancer and untimely aging were proven to have been kept under control in the presence of Lycopene, which is a standout amongst the most effective antioxidants and a dietary carotenoid in tomatoes and other vegetables (Wener, 2000). Processing and/or cooking tomato is actually beneficial to health because it increases the bioavailability of lycopene as heating up tomato breaks down its cell walls and release more lycopene which helps prevent colorectal, oesophageal, lung, pancreatic, prostate, stomach, oral, breast and cervical cancers ([renatovicario.com/pdf/tomato.pdf](http://renatovicario.com/pdf/tomato.pdf)).

## **2.5 Production of tomato**

### **2.5.1 World production of tomato**

Tomato is one of the major horticultural crops cultivated worldwide with a global production estimated at 170,750,767 tonnes and a total land area of 5,023,810 hectares (FAOSTAT, 2014).

The proportions of tomato production by region according to FAOSTAT (2014), Asia had the largest (59.5 %) contribution to tomato production and availability worldwide while Africa contributed 11.3 %.

Less than 20 countries produced above one (1) million metric tonnes (FAOSTAT, 2014). The world's leading producers of tomato are China, India, USA (Table 2). The various volumes of production are presented in Table 2.

**Table 2: World's leading tomato producers**

<b>Rank</b>	<b>Country</b>	<b>Production (tonnes)</b>
1	China	52,722,967
2	India	18,735,910
3	USA	14,516,060
4	Turkey	11,850,000
5	Egypt	8,288,043
6	Islamic Republic of Iran	5,973,275
7	Italy	5,624,245
8	Spain	4,888,880
9	Brazil	4,302,777
10	Mexico	3,536,305
11	Russian Federation	2,819,193
12	Uzbekistan	2,285,801
13	Ukraine	2,147,880
14	Nigeria	2,143,500
15	Portugal	1,399,535
16	Tunisia	1,250,000
17	Morocco	1,230,954
18	Algeria	1,065,609
19	Greece	1,032,220
20	Indonesia	916,001

*Source: FAOSTAT, (2014)*

### **2.5.2 Tomato production in Ghana**

In Ghana, the efforts of tomato producers to attain same production levels as countries like China and the United States have been unsuccessful (Robinson and Kolavalli, 2010). Even commercial producers are unable to match up the needed supplies for sustainable processing. Ghana had cultivated tomato on a total land area of 47,000 hectares, yielded 78,037 hg/ha which in turn

indicated that the total tonnage of tomato production in Ghana stood at 366,772 tonnes (FAOSTAT, 2014).

### 2.5.3 Soil requirements

Kelley and Boyhan (2017) soil requirements for tomato production is as follows: Fertile organic matter-rich sandy loam soils that are deep, well drained, quite rough and have slightly acidic to neutral pH (6 - 7). Plants depend intensively on the soil for root growth and anchorage; water and nutrient uptake, nutrients necessary for plants growth must be available and accessible in sufficient and balanced volumes in order to attain optimal growth of plants (Chen, 2006). But this can only be possible if the topography, soil type and soil structure are just appropriate and soil management practices such as tillage are perfectly executed (Kelly and Boyhan 2017).

Low soil fertility is the most occurring limitation hindering various crop yields in developing countries and especially among smallholder farmers (Mokwunye *et al.*, 1996) but this can partially be circumvented if proper land preparation is done to significantly reduce soil compaction and hard pans. This also aids plant roots to develop extensively to their full potential (Kelly and Boyhan, 2017).

Table 3 shows the germination temperatures that tomato seeds need to rapture and emerge. The optimum temperature for tomato seedling to emerge is 29.4°C (Boyhan and Kelly (2017). Thus, a temperature range of 20°C to 30°C would be most appropriate for rapid germination and early overall establishment of tomato. Too low temperatures (below 20°C) or too high temperatures (above 30°C) are however detrimental to tomato production.

**Table 3: Soil temperature and days to emergence**

<b>Soil Temperature</b>	15.6°C	20°C	25°C	29.4°C	35°C
<b>Days to Emergence</b>	14	8	6	5	9

*Source: (Boyhan and Kelly 2017)*

#### **2.5.4 Climatic requirements**

For tomato to develop and yield quality fruits, the crop needs a moderately cool, dry climate but it can however be cultivated in both temperate and hot climates (Srinivasan, 2010).

Data gathered by the South African Agricultural Department, Forestry and Fisheries (2017) however noted that, although tomato is a warm season crop, the average temperature required for seed germination, plant growth, fruit set and fruit quality lie between 20 – 24°C. Temperatures below 12°C and above 35°C, are unsuitable for tomato development and fruit quality. Unnecessary blossom abortion is mostly triggered by hot dry breezes. Foliar disease incidence and outbreak is facilitated by persistent sodden, rainy climate. Growing tomatoes in dry areas under irrigation is therefore highly recommended.

#### **2.6 Causes of tomato production losses**

Estimation of production losses in developing countries are very hard to evaluate although the trend is easily noticeable through the yield losses of the crop (Adeoye *et al.*, 2009). Losses in tomato production have been factored down to diverse causal agents ranging from biotic to abiotic factors even though these may appear very similar (Flynn, 2003).

Biotic effects are more restricted to certain species while abiotic destruction frequently occurs in a considerable number of species. Biotic effects can cause diseases to spread from one plant to neighboring plants of identical species mostly facilitated by wind-blown rain while abiotic effects are not transmitted from one crop to another even over time (Flynn, 2003).

When plants are affected by abiotic diseases, symptoms may not show until crops are almost damaged whereas for biotic diseases symptoms of the pathogen infection are expressed.

Losses in production relatively mean waste of scarce resources such as naturally occurring soil nutrients, formulated soil amendments like fertilizers and growth enhancers and water and human efforts (Flynn (2003).

### **2.6.1 Biotic factors**

These include various living organisms such as insect pests and pathogens. Pathogenic bacteria, nematodes, fungi and viruses are causal agents of the numerous tomato diseases. Tomato is susceptible to so many diseases caused by the aforementioned pathogens. Most of these pathogenic organisms are usually associated with the seed thus causing damage right from the rupture of the radicle and plumule (i.e. onset of germination) (Lukyanenko, 1991)

Fungal diseases of tomato include: Early blight, late blight, *Fusarium* wilt, *Septoria* leaf spot, Target spot and Southern stem blight. Bacterial diseases of tomato also include bacterial spot, bacterial speck and bacterial wilt (David and Langston, 2017). Viral diseases include: tomato spotted wilt virus (TSWV), cucumber mosaic virus (CMV) and tomato yellow leaf curl virus (TYLCV).

Insect pests cause different magnitudes of damage per the location and time of year of tomato production. Some of these insects includes foliage feeders: aphids, flea beetles, Colorado potato beetle, hornworm, cabbage looper, whiteflies, leaf miners, spider mites; fruit feeders: stink bugs and leaf footed bugs, beet armyworms, tomato fruit worm (corn earworm), tarnished plant bugs and tomato pinworm; seedling pests: cutworms. (Alton and Sparks, 2017). Some plants can endure some amount of insect defoliation without adverse effect on yield (McCormack, 2004; Watt, 2000).

Another major constraint to tomato production is infection by plant parasitic nematodes. Nematodes are worm-like organisms which are extremely widespread and practically found everywhere, both as free-living organisms and as parasites (Coyne *et al.*, 2007).

Generally, nematodes although very minute and virtually impossible to see with the naked eye, some species are reported to reach considerable length (Coyne *et al.*, 2007).

Root-knot nematodes (*Meloidogyne* spp.) are destructive agricultural pests, attacking a lot of crops under cultivation (Baker *et al.*, 1985). Development of the root-knot disease is initiated when nematode second stage juveniles penetrate the roots of tomato plants. (Baker *et al.*, 1985). In as much as they are very microscopic, nematodes are very destructive and they exhibit non-specific symptoms. They perpetrate damages that are often related to other reasonable and visible factors (Coyne *et al.*, 2007).

Nematodes cause physical damage that affects the uptake of water and nutrients, they also allow the manifestation of other diseases. Growth of diseased plants are also suppressed with development of pale green to light yellow foliage (David and Langston, 2017).

### **2.6.2 Abiotic factors**

These on the contrary are non-living. They comprise harsh and inclement soil and weather conditions (chemical drift, drought stress, freeze injury, nutrient deficiency, sunscald, wind injury and too low and/or too high soil pH) and improper cultural practices.

Adverse environmental conditions affect tomato production in that they cause various unappealing physiological problems. These problems include blossom drop which results in reduced yields; blossom-end rot resulting from lack of calcium; radial and concentric fruit cracking caused by higher temperatures and prolonged rains; puffiness as a result of too cool temperatures; rain check caused by healed tiny cracks; sunscald caused by fruit exposure to intense sunlight and heat; cat

facing occurring when fruits are formed during humid or cooler weather which happens to favour corolla adhering to developing fruits (Kelly and Boyhan, 2017).

Warm and/or wet climates facilitate significantly high prevalence of tomato diseases. Since tomato is highly susceptible to most of these diseases, significant loss of the crop occurs throughout its growth cycle and attacks could be extremely damaging in warm and wet climates (Parker *et al.*, 2000).

Tomatoes are warm season vegetables and they need relatively moderate temperatures to set fruit. In the night, when temperatures go above  $-6^{\circ}\text{C}$ , blossom drops may be induced which consequently will result in reduced yields if not zero yields (Boyhan and Kelly, 2007).

## **2.7 *Trichoderma harzianum***

### **2.7.1 *Trichoderma harzianum* as seed treatment**

Tomato seeds germinate in a temperature range of  $10^{\circ}\text{C}$  to  $35^{\circ}\text{C}$ . with optimum germination occurring  $29^{\circ}\text{C}$  (McCormack, 2004). According to *T. harzianum* is known to increase not only growth and yield of plants, increase nutrient uptake, increase fertilizer utilization efficiency but also seed germination rate, seed germination percentage, and instigated systemic resistivity to plant diseases Harman *et al.*, 2004).

Tomato seeds can be dressed with *Trichoderma* and directly sown into the soil or any media for cultivation. Seed treatment before sowing/planting warrants that the fresh emerging roots are colonized at an opportune time for defense against root decays caused by water mould. Bare roots of plants can be dipped into pre-activated *T. harzianum* solutions. Here, newly emerged roots might be predisposed to parasitic pathogens before the dip (Lester, 2010).

*Trichoderma* contends with other microorganisms in that, it competes for important exudates from seeds that invigorate the germination of propagules of soil-borne plant pathogenic fungi as well as competes with other microorganisms for space and/or available soil nutrients (Howell, 2002).

The most emphatic rhizosphere competent strains of *Trichoderma* spp. can be added to soil or seeds. They germinate and colonize the root surface once into contact with plant roots and some moisture, preventing intrusion of other parasitic organisms (Ghahfarokhy *et al.*, 2011).

The use of *T. harzianum* as biological control agent is because of its functional characteristics: parasitism, competition, production of pathogen restrictive compounds and enzymes. They deactivate pathogen enzyme systems rendering them ineffective (Lester, 2010). Competitively, *Trichoderma* actively invades root rhizosphere making it tough for other pathogenic microbes to contend for limited root surface space and also available plant nutrients. Regarding parasitism, *Trichoderma* spirally covers the hyphae of pathogenic organisms and then produces enzymes such as chitinase and cellulase to dissolve the cell walls of soil pathogens like *Pythium*, *Fusarium* and *Rhizoctonia* among others. The cellulase produced by *Trichoderma* breaks down cellulose that is readily available in plant fibers, crop residues or those in other parasitic organisms living in the soil around the root zone. The enzyme chitinase decomposes chitin – a structural constituent in fungal cell walls and a key component in insect exoskeletons (Lester, 2010).

*Trichoderma* spp. produces a wide range of antibiotic substances to parasitize other soil pathogens. They are free-living, ubiquitous but mostly found in root, soil and foliar environments (Sivasithamparam *et al.*, 1998 and Ghahfarokhy 2011) but no record of *Trichoderma* attacking insects has been documented (Lester, 2010).

*T. harzianum* is one of the most effective biocontrol agents that is commercially available for application to control pathogens as well as prevent development of several soil borne pathogens (Ghahfarokhy, 2011).

Sivan *et al.* (1984) also reported 85 % disease reduction in tomatoes using *Trichoderma* species. A research by Ojha and Chatterjee (2011) evaluated the mycoparasitic potential of three (3) taxonomic groups of *Trichoderma*: *T. harzianum*, *Trichoderma viride* and *T. hamatum* in relation to minimizing the consequences of *Fusarium oxysporum*, the causal agent of *Fusarium* wilt in tomato. It was reported that *T. harzianum* was superior over the others in that, it was more efficient in producing chitinase,  $\beta$ -1,3 glucanase and lytic enzymes which enhanced its capability to diminish the frequency of the wilt disease.

Lee *et al.* (2006) also provided evidence that *Botrytis cinerea* was successfully controlled by *Trichoderma* species. Windham *et al.* (1989) reported significant reduction in egg production in *Meloidogyne arenaria* in *T. harzianum* and *T. koningii* treated soils.

Even though *Trichoderma* fungus is accepted as an essential biocontrol agent, Baker (1986) mentioned that when *Pisium sativum* seeds dressed with conidia of *T. harzianum* were planted at temperatures that were harsh to the growth of the beneficial fungus, the incidence of *Rhizoctonia* damping-off was similar to the untreated plots.

The fungus, *T. harzianum* became pathogenic when its population density was excessively increased which resulted in callused seedlings. That notwithstanding, *Trichoderma* can perfectly perform its “traditional role” as a biocontrol agent (Baker, 1986).

### **2.7.2 Factors that cause loss of *Trichoderma harzianum***

*Trichoderma* fungi are lost in nature because of soil degradations such as erosion, mining, chemical treatments, excessive heat, drought and flooding (Lester, 2010).

Temperature is also another abiotic factor that causes loss of *Trichoderma*. Too low temperatures are unfavourable for *Trichoderma* growth. The safest temperature for *T. harzianum* lies within the range 30 to 37°C (Lester, 2010). *T. harzianum* was most effective at a relative humidity between 80% and 97% under temperatures above 20°C (Elad *et al.*, 1993). *T. harzianum* took 96 hours long to germinate at a lower temperature of 12°C (Hjeljord, 2000).

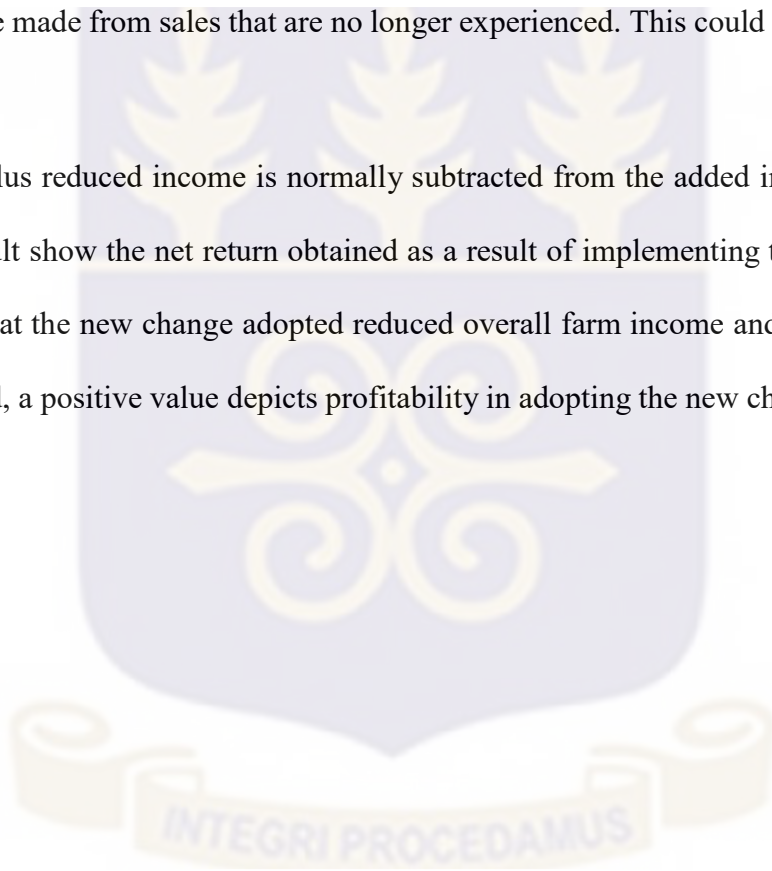
### **2.8 Partial budget analysis**

Partial budget (Appendix 2) is a tool used in comparing positive and negative likely effects that come along when new implementations and or changes that are introduced in a business (Robert, 2006). This analysis is essential in assessing only a small portion of the business where a change is likely to affect, not necessarily the whole business. It only allows for items that are particularly affected due to the alternative changes introduced. Partial budgets also help in determining revenue and expense items that are affected by the decision of implementing a change by further showing the difference in net income over relevant expenses. Robert (2006) mentioned that in view of employing partial budgets in businesses, four basic principles are inquired. Firstly, whether there will be an increase in income, secondly, whether the new implementation will reduce or eliminate unnecessary costs, thirdly, if there will be increase in costs of production and finally, whether there will be reduction or elimination of income.

The partial budget has four major components in two columns that need to balance out. The components include:

- a. the added income component: This illustrates what new product or item is introduced and what income estimate could be accrued.
- b. added costs component: This component contains list of all increased expenses that resulted from the additional input or implementation.
- c. reduced costs component: This portion accounts for the previous expenses that were being incurred that are no longer going to be experienced.
- d. reduced income: This section of the budget includes possible reduction in progress or farm incomes that were made from sales that are no longer experienced. This could be reduced yields for example.

Increased costs plus reduced income is normally subtracted from the added incomes plus reduced costs and the result show the net return obtained as a result of implementing the change. Negative values indicate that the new change adopted reduced overall farm income and hence insignificant. On the other hand, a positive value depicts profitability in adopting the new change.



## CHAPTER THREE

### 3.0 MATERIALS AND METHODS

#### 3.1 Study sites

The study was conducted from June 2016 to June 2017. Laboratory experiments were carried out at the Ghana Seed Inspection Division's (GSID) Seed Testing Laboratory of the Plant Protection and Regulatory Service Directorate (PPRSD) Head Office and the Soil Science and the Plant Pathology Laboratories at the University of Ghana, Legon.

GSID is located at Pokuase a suburb of Accra, off the Accra-Kumasi highway at a Latitude  $5^{\circ} 42' 0''$  North and Longitude  $0^{\circ} 18' 7.2''$  West. The University of Ghana also located in Greater Accra region is at a latitude  $5^{\circ} 39' 1.79''$  North and Longitude  $0^{\circ} 11' 7.80''$  East. The field work was conducted in Dawa, situated at the outskirts of Ada in the Greater Accra Region of Ghana at Latitude  $5^{\circ} 52' 0''$  North, Longitude  $0^{\circ} 18' 0''$  East

#### 3.1.2 Climatic and edaphic conditions of the field

Rainfall pattern in Dawa is bimodal with mean annual rainfall of 500 - 1,100 mm fairly distributed from April to October (Kouadio, 2011). During the dry season, monthly temperatures are very high in the day to a maximum value of about  $34^{\circ}\text{C}$  and minimum of  $28^{\circ}\text{C}$  and in the night, temperatures are very low to about  $24^{\circ}\text{C}$ . During the wet season, monthly temperatures are as high as  $29^{\circ}\text{C}$  and as low as  $22^{\circ}\text{C}$  and at night, temperatures are as low as  $20^{\circ}\text{C}$ . The area has an average relative humidity of 76% with an average cloud cover of 75% and dew point of about  $26^{\circ}\text{C}$  (AccuWeather, 2017).

The soils in Ada are mostly sandy clay loams and classified as Costal Sodium Vleisols (Obeng, 2000).

### 3.2 Sources of materials

- Tomato Variety (UC 82) was obtained from RMG (Regional Marketing Group) Ghana Limited, Accra.
- Velum (Floupyram SC 400 G) which is a Nematicide was acquired from Bayer West and Central Africa Limited (Bayer WCAFA).
- Eco-T was used as the source of the *Trichoderma harzianum* and was provided by Bioteq Company.
- Petri dishes were obtained from GSID.

### 3.3 Methodology

The study was divided into Two (2): Experiment one (1) consisted of three (3) different studies conducted in the laboratory to investigate the effect of *T. harzianum* on tomato seed germination and seedling vigour and experiment two (2) had only one (1) study conducted on the field to find out the effect of *T. harzianum* on incidence of root-knot and yield of tomato.

Experiment one (laboratory work) was conducted in the Ghana Seed Inspection Division (GSID) laboratory of the Plant Protection and Regulatory Service Directorate (PPRSD) at Pokuase.

Experiment two (field work) was conducted between February 2017 to May 2017, after a preliminary field study during the internship period revealed a high prevalence of nematodes which the farmers in the area reported to be responsible for hindering successful tomato production in the Dawa locality.

### 3.4 Laboratory analyses

#### 3.4.1 Experimental design

All laboratory experimental setups were arranged in a Completely Randomized Design with four (4) replications on a Jacobson Table for the germination test.

#### 3.4.2 Seed germination and seedling vigour index as affected by duration of exposure of tomato seeds to *Trichoderma harzianum* (Eco-T)<sup>®</sup>.

Seeds were treated with Eco-T (*T. harzianum*) at different time intervals prior to sowing using the recommended dose of 0.2 g of Eco-T for every 10 g of tomato seeds. Twenty-five (25) mL of distilled water was mixed with Eco-T to activate the *T. harzianum* spores before applying to the seeds. Ten (10)g of tomato seeds were poured into the slurry after they had been washed thoroughly with distilled water, well mixed and allowed to dry for various durations after which 25 seeds were plated per Petri-dish on a wet blotter paper.

There were nine treatments designated as follows;

- No coating of tomato seeds with Eco-T (control 1)
- Coating of tomato seeds for 30 mins (Check)
- Coating of tomato seeds with Eco-T for 1 hr
- Coating of tomato seeds with Eco-T for 3 hrs
- Coating of tomato seeds with Eco-T for 6 hrs
- Coating of tomato seeds with Eco-T for 8 hrs
- Coating of tomato seeds with Eco-T for 24 hrs
- Coating of tomato seeds with Eco-T for 30 hrs
- Coating of tomato seeds with Eco-T for 48 hr

### **3.4.3 Application of different concentration of *Trichoderma harzianum* (Eco-T)<sup>®</sup> to tomato seeds prior to sowing**

Seeds were coated with Eco-T (*T. harzianum*) at different concentrations prior to sowing using the recommended dose of 0.2 g of Eco-T for every 10 g of tomato seeds as baseline. For each treatment, 25 mL of distilled water was mixed with Eco-T to activate the *T. harzianum* spores before applying to the seeds. Ten (10)g of UC 82 tomato seeds were poured into the slurry after they had been washed thoroughly with distilled water, well mixed and allowed to dry after which 25 seeds were plated per Petri-dish on a wet blotter paper.

There were six treatments designated as follows;

- No coating of tomato seeds with Eco-T (control)
- Coating 10g of tomato seeds with 0.2g Eco-T (check)
- Coating 10g of tomato seeds with 0.25g Eco-T (25% above recommended)
- Coating 10g of tomato seeds with 0.3g Eco-T (50% above recommended)
- Coating 10g of tomato seeds with 0.35g Eco-T (75% above recommended)
- Coating 10g of tomato seeds with 0.4g Eco-T (100% above recommended)

### **3.4.4 Seed germination and seedling vigour as affected by duration of Eco-T exposure to tomato seeds at different concentrations.**

A set up to evaluate the interaction between duration of exposure and concentration was carried out. The best two (2) treatments and a control from the duration of exposure of Eco-T experiment, and also, the best two (2) application concentrations from the application of different concentration of Eco-T experiment were analyzed to determine the best application concentration to use *T. harzianum* (Eco-T) and at what duration of exposure for optimum results. Twenty-five (25) seeds were plated per petri-dish on a wet blotter paper.

The *T. harzianum* (Eco-T) treatments employed were two application concentrations with a control with two durations of exposure.

- No Eco-T x 30 mins
- No Eco-T x 3 hrs
- Coating 10g of tomato seeds with 0.2g Eco-T x 30 mins
- Coating 10g of tomato seeds with 0.2g Eco-T x 3 hrs
- Coating 10g of tomato seeds with 0.4g Eco-T x 30 mins
- Coating 10g of tomato seeds with 0.4g Eco-T x 30 hrs

### **3.4.5 Experimental design**

A 3x2 factorial treatment combinations laid out in CRD with four (4) replicates was used in this setup which was arranged on a Jacobson Germination Table.

### **3.4.6 Data collected for laboratory experiments 1 – 3.**

- Percentage (%) germination =  $(\text{[Number of seeds germinated / Total number of seeds]} \times 100)$
- Shoot length (cm) measured with a meter rule
- Root length (cm) measured with a meter rule
- Seedling vigour index =  $(\text{[Mean root length + Mean shoot length]} \times \text{percentage of seed germination})$

### **3.4.7 Data analysis**

Analysis of variance was done with Genstat statistical package

### **3.5 Field work**

#### **3.5.1 Impact of *Trichoderma harzianum* on nematode population and tomato (UC 82) seedling performance in the field.**

#### **3.5.2 Treatments:**

There were 6 treatments. Based on the outcome of the Laboratory studies carried out prior to the field work, the following treatments were administered:

T0 = Zero Eco-T (Control)

T1 = Velum (Floupyram SC 400) drenching 4 weeks after planting at 3.8 mL/7 L of water

T2 = Seed coating with 0.2 g of Eco-T for 30 minutes prior to sowing

T3 = Seed coating with 0.4 g of Eco-T for 30 minutes prior to sowing

T4 = Seedling drenching with 0.2 g of Eco-T four weeks after planting

T5 = Seedling drenching with 0.4 g of Eco-T four weeks after sowing

#### **3.5.3 Land preparation and field layout**

The field was shallowly ploughed to loosen up the soil and not to disturb the nematode community in the soil since the original population of nematodes was needed for the experiment. No inoculants were used.

A plot size of 3.5 x 3 m<sup>2</sup> was allocated for each treatment. A (1)m alley was left between plots hence a total land area of 26 x 15 m<sup>2</sup> was used for the study.

#### **3.5.4 Experimental design**

A Randomized Complete Block Design (RCBD) with four (4) replicates was used in this setup.

### **3.5.5 Treatment application and planting**

As a seed treatment, 0.2 g and 0.4 g of Eco-T (*T. harzianum*) was applied to 10 g of UC82 tomato seeds respectively by dissolving various concentrations in 25 mL of water before adding the seeds to the slurry and then left to sit for 30 minutes to get seeds quite dried. Seeds were picked and sown with a pair of forceps after drying. Four (4) seeds per hole were sown hence, ninety-six (96) seeds were sown in 16 lines. giving a total of 2,304 seeds on the entire field.

Drenching with 0.2 g and 0.4 g of Eco-T was done at 4 weeks after sowing likewise application of Velum (Fluopyram SC 400 G) treatment at 3.8 mL/7 L of water.

### **3.5.6 Cultural practices**

Irrigation was carried out every single day using the dam at the location as the source of water due to drought conditions during the first two months of cultivation.

Weeds were controlled mostly by handpicking and where needed, hoe was used. No weedicides were applied.

N.P.K 15:15:15 was applied on entire field at 4 weeks after planting using 20 g/plant. Insects such as the lady bird, grasshopper, canker worm, etc. found on the field were controlled by spraying Decis forte (Deltamethrine) at a rate of 4 mL/16L of water.

### **3.6 Soil and tomato plant sampling**

The (MDSP) Modified Diagonal Sampling Pattern was adopted for soil sampling on various plots. Using the auger, eight (8) cores were collected from 0-30 cm after which they were composited to obtain homogenous samples representing each block. Auger, cutlass and wellington boots were cleaned of soil post sampling of each plot before moving onto another plot. This was to ensure avoidance of introduction of nematodes from one plot contaminating the initial populations of other plots. Soils were placed into white rubber bags pre-labelled with each plot's name, and date written

with permanent black marker before final placement in black polythene bags. GPS coordinates were recorded immediately.

For the physico-chemical analysis, half a kilogram of submitted samples was removed and air dried. This helped to reduce the build-up of nitrate in the soils. Soils were thinly spread on clean piece of plastic slates in the open and allowed to air dry over three (3) days after which they were sieved with a 2 mm mesh sieve to eliminate broken sticks and debris prior to analysis. At the end of the study, that is, after harvesting, soil samples were collected again for final analysis of physico- chemical properties.

For the nematode analysis, sample soils were individually sealed and placed on ice cubes in an ice chest. Samples were later transferred into the deep freezer in the laboratory. This was done to prevent the death of nematodes in the sampled soils prior to analysis.

At 100 % flowering, the auger was used to sample soils from root rhizosphere of the tomato plants on each plot. Three tomato plants were sampled from various plots and placed on ice to prevent dehydration of plants in order to preserve nematodes and nematode eggs.

### **3.7 Determination of soil chemical and physical properties**

#### **3.7.1. Total nitrogen**

The Kjeldahl procedure (usually the wet oxidation method) involved two steps which were – the digestion of the samples to convert the N to ammonium ( $\text{NH}_4$ ) and then the determination of the ammonium in the digest.

Reagents used were concentrated sulphuric acid, digestion accelerators (10g  $\text{K}_2\text{SO}_4$ , 1g  $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$  and 1g selenium) mixed together, 40% sodium hydroxide (NaOH), mixed indicator (0.13g of methyl red plus 0.066g of methylene blue dissolved in 100mL of 95% ethanol), hydrochloric acid 0.01M and 2% boric acid (2g of  $\text{H}_3\text{BO}_3$  in 100mL of distilled  $\text{H}_2\text{O}$ ).

Two (2)g of < 2 mm air-dried soil sample was weighed into the 500mL Kjeldahl digestion flask and 4mL of distilled H<sub>2</sub>O was added to it so as to wet the soil thoroughly for about 30 minutes. After which a scoop (0.5g) of the digestion catalyst mixture as well as 5mL of concentrated H<sub>2</sub>SO<sub>4</sub> were added. The mixture altogether was heated gently on a digesting rack in a fume cupboard until vigorous effervescence subsided. This was done to digest the solution. Boiling was continued until the digest became clear. The digest was then allowed to cool and settle after adding 20mL of distilled H<sub>2</sub>O. The supernatant solution was decanted into a 50mL volumetric flask. A blank consisting of acid and catalyst was also digested. Five (5) mL of the aliquot was taken and transferred into a Markham distillation apparatus. Ten (10) mL of the 40% NaOH was added to the aliquot and the mixture distilled. NH<sub>4</sub> was trapped in 5mL of 2% boric acid. The distillate (NH<sub>4</sub>) was then titrated with 0.1 N HCl until a green to reddish end point was observed and the value recorded. The total nitrogen content was then determined as described by Bremner and Malvaney (1982).

### 3.7.2 Soil pH

The soil pH in water method was employed to determine the soil pH. Twenty (20) g of air dried soil sample was weighed into a 50mL beaker after which 20 mL of H<sub>2</sub>O was added. The suspension was stirred severally for the next 30 minutes. The suspension was then allowed to stand for about 30 minutes so as to ensure that most of the suspended clay particles settled out from the suspension. Prior to dipping it into the suspension, the pH meter was standardized using the standard buffer solutions of pH 4.0 and 7.0. The glass electrode was then inserted into the supernatant of the suspension and then the result read for the pH was recorded as soil pH<sub>w</sub> 1:1

The pH of the sediment of that same sample was also taken and the first step in paragraph one was repeated. The glass electrode was inserted into the supernatant of the suspension and the pH was

measured and the results reported as soil pHw 1:2. The results (pHw 1:1 and pHw 1:2) were then tabulated.

### 3.7.3 Available phosphorus

The method proposed by Bray and Kurtz (1945) was followed. Bray 1 solution (which consists of a mixture of 0.03 M  $\text{NH}_4\text{F}$  and 0.025 M  $\text{HCl}$ ) was used to remove easily acid-soluble phosphorus forms, largely calcium phosphates and a portion of iron phosphates and aluminium. The reagents included Ammonium fluoride ( $\text{NH}_4\text{F}$ ) 1M of which 37 g was dissolved in distilled water and diluted to 1 litre stored in a polyethylene bottle, hydrochloric acid ( $\text{HCl}$ ) 0.5 M of which 20.2 mL was diluted to a volume of 500 mL with distilled water and the extract which was prepared by adding 15 mL of 1.0 M ammonium fluoride ( $\text{NH}_4\text{F}$ ) and 25 mL of 0.5 M hydrochloric acid ( $\text{HCl}$ ) to 460 mL of distilled water. This gave a solution of 0.03 M in  $\text{NH}_4\text{F}$  and 0.025 M  $\text{HCl}$ .

Thirty (30) g of air-dried soil sample was weighed into a shaking bottle of volume 100 mL. Sixty (60) mL of the extractant (Bray 1) was added to the shaking bottle content and was shaken for 2 minutes on a mechanical reciprocal shaker. The content was then passed via a Whatman No. 42 filter paper. Twenty (20) mL aliquots of the filtrate were taken in duplicates into 50 mL flask and 10 mL colouring reagent (ammonium paramolybdate) was added and then a small drop of ascorbic acid followed. The content was well mixed and left to settle for 20 minutes for a blue colour to be formed. The colour was measured at a wavelength of 660 nm with the spectrophotometer. Standard series of 0, 1.2, 2.4, 3.6, 4.8, and 6.0 mg P/l was prepared by pipetting 0, 10, 20, 30, 40 and 50 mL of 12.0 mg P/L respectively into a 100 mL volumetric flask and distilled water was added to the mark. The available phosphorus was then deduced from the standard curve.

### **3.7.4 Organic carbon**

Nelson and Sommers (1982) description of a modification of the wet oxidation procedure based on the reduction of the  $\text{Cr}_2\text{O}_7^{2-}$  ion by organic matter which is known as the Walkley and Black procedure was followed. Ten (10) mL of 1N  $\text{KCr}_2\text{O}_7^{2-}$  solution and 20mL  $\text{H}_2\text{SO}_4$  were added to soil samples. One (1) g of the soil sample was weighed into a conical flask after screening with 0.5 mm sieve. A blank was also included in separate conical flask. Addition of 10mL of potassium dichromate solution followed by 20mL of concentrated  $\text{H}_2\text{SO}_4$  to the soil and blank flasks was done. The flasks were carefully swirled for maximum contact with all the particles of the soil. After some few minutes, the content in the flask was allowed to settle for 30 minutes. Two hundred and fifty (250) mL of distilled water was added and 10mL orthophosphoric acid was also added. Finally, 2mL of barium diphenylamine sulphate indicator was added to the flask content and titrated with 0.1M ferrous ammonium sulphate solution until the colour changed to bright green end point.

### **3.7.5 Exchangeable cations: Extraction**

Cation exchange is a critical factor in soil fertility from two important standpoints. Firstly, it affects the total amount of nutrient availability to plants as exchangeable cations. Secondly, it influences the degree to which the exchange complex is saturated with basic ions as opposed to hydrogen ions. Exchangeable (metallic) cations most frequently found in the soils include Ca, Mg, K and Na, all of which are easily extracted and determined in 1 M Ammonium ethanoate ( $\text{NH}_4\text{OAc}$ ) extract of soil (Black, 1965). Two hundred and fifty (250) mL of 1.0 M ammonium acetate ( $\text{NH}_4\text{OAc}$ ) buffer solution at pH 7 was used to leach 10 g soil sample in leaching tube.

### **3.7.6 Potassium (K) and sodium (Na) determination**

In determining the exchangeable K and Na, the flame photometry method was employed as described by Helmke and Sparks (1996). One hundred (100) mg/L of K and Na standard series

solutions were prepared. The 100 mg/l standard solution was subdivided into portion of 0, 5, 10, 15 and 20 mL into 200 mL volumetric flasks each. One hundred (100) mL of 1.0 M ammonium acetate solution was added to the contents of each flask and distilled water added to mark up the volume. 0, 2.5, 5.0, 7.5, 10.0 mg/l standard series were obtained for potassium and sodium. Finally, Na and K were sprayed directly into the flame as directed at wavelengths of 589.0 nm and 766.5 nm respectively and the photometer readings were obtained.

### **3.7.7 Calcium (Ca) and magnesium (Mg) determination**

One (1) M of ammonium acetate ( $\text{NH}_4\text{OAc}$ ) extract was used as described by Black (1965). Soil sample was weighed into a shaking bottle and 100 mL of 1 M Ammonium ethanoate solution was added and then shook thoroughly in a mechanical shaker for an hour. The supernatant solution was passed via No. 42 Whatman filter paper into clean empty plastic bottles. An aliquot of the extract was used for the determination of Ca, Mg. A 25 mL portion of the extract was taken into a conical flask and distilled  $\text{H}_2\text{O}$  was added to make the volume to the 50 mL mark. Three drops (2 %) of potassium ferrocyanide (1 mL), potassium cyanide (1 mL) from a burette, hydroxylamine hydrochloride (1 mL), ethanolamine buffer (10 mL) and Enriochrome Black T solution (0.2 mL) were added. The mixture was then titrated with 0.01 M EDTA solution until an end point from red to blue green colour (or turquoise blue colour) was obtained. To attain a standard blue colour from the titration, an addition of 20 mL of 0.01 M EDTA and 25 mL of 1.0 M ammonium acetate solution was made and the titre value was documented. To calculate for the titre value of magnesium, the titre value for Ca was deducted from this value.

### **3.7.8 Particle size distribution**

Hundred (100) mL of distilled water was added to previously weighed 50 g of air dried soil sample placed into a 1 litre shaking bottle. The content was swirled thoroughly for about 2 minutes and then

20 mL of 30 % H<sub>2</sub>O<sub>2</sub> was added, followed by 50 mL of 5 % sodium hexametaphosphate after which drops of amyl alcohol was added and gently swirled. The mixture was placed on a mechanical shaker and was shaken for 120 minutes after which the mixture was poured out into a 1 litre sedimentation cylinder. The first hydrometer reading (specific gravity value) as well as the first thermometer reading (temperature value) were recorded after 40 seconds. The 1 litre sedimentation cylinder together with its content was allowed to settle for 3 hours after which the second set of readings were taken.

### **3.7.9 Data collected and reported**

- % Total Nitrogen
- Soil pH
- Available Phosphorus (mg/kg)
- % Organic Carbon
- Exchangeable Potassium (Cmol/kg)
- Exchangeable Sodium (Cmol/kg)
- Exchangeable Calcium (Cmol/kg)
- Exchangeable Magnesium (Cmol/kg)
- Particle size distribution (% sand, % silt, % clay)

### **3.8 Nematode parameters**

#### **3.8.1 Nematode extraction and counting**

Nematode extraction and counting were performed in the Plant-Pathology laboratory of the Department of Crop Science, University of Ghana, Legon. Nematode populations were determined before sowing, 6 weeks after sowing, and after harvesting at 12 weeks. Extraction of nematodes was done using the water and sucrose extraction methods. Sample soil lumps were mashed into fine

texture before mixing with water. Soil samples were mashed in water and left to settle for about 5 minutes. Clayey samples were left to settle for about 7 minutes. After every round of mashing, hands were washed prior to another round of mashing to prevent contamination and introduction of foreign nematodes. The initial step was repeated once again. Water was poured on residue and allowed to settle for 5 minutes.

Three laboratory test sieves with different apertures were used; 90  $\mu\text{m}$ , 71  $\mu\text{m}$  and 36  $\mu\text{m}$ . The 90 and 71  $\mu\text{m}$  sieves trapped the soil particles while the 36  $\mu\text{m}$  trapped both nematodes and nematode eggs. Residue on the 36  $\mu\text{m}$  sieve after decantation was sent to the biotechnology laboratory for centrifugation at 1700 rpm for five minutes. The centrifuge only works on a principle of balancing so water was added to the plastic vials containing the residue collected from the 36  $\mu\text{m}$  sieve to equal volumes prior to arrangement in opposite but balanced positions in the centrifuge. The supernatant was discarded and sucrose was added to the settled particle in the plastic vials. Second centrifugation at 1000 rpm for 1 minute was then done after addition of the sucrose which aided in separating the nematodes from the particles causing the nematodes to be in the suspension. The suspension was then sieved out via a 36  $\mu\text{m}$  sieve trap the nematodes and then transferred into the plastic vials. Water was added to the content until the 10 mL mark using the fine spray water bottle after which they were taken to the pathology laboratory for nematode identification and count. Hand tally counter was used to keep track of each nematode counted in the suspension under the compound microscope. Counting was repeated three times and the average nematode numbers determined in 200 cc of soil, and 10 g of tomato roots.

### **3.8.2 Nematode identification**

Nematode suspension in plastic vials were separately emptied gently into 50 mm round plastic counting petri dishes calibrated on the inside with 2 mm by 2 mm blue grid lines. These lines served

as a guide for systematic observation of nematodes across the liquid. Nematodes were sucked using a small syringe and transferred to another counting dish where selected nematodes for identification were mounted on a drop of water on a microscopic slide and placed on a 60 °C hot plate to allow nematode to straighten for easy identification (Hooper, 2005). The compound light microscope with magnification 400x, manufactured by Optec optical technology, was used for nematode identification to the genera level. They were identified based on their morphological features as described by Siddiqi, (2000), Luc, *et al.* (1990), and the website of the University of Nebraska - Lincoln for nematode identification:

(<http://nematode.unl.edu/konzlistbutt.htm>). Identification of nematodes was mainly based on the length of stylet, stylet knob, head region, intestine thickness and shape of tail of adult nematodes

### **3.8.3 Root-knot nematode gall scoring**

The stem to the crown of each tomato plant sampled was separated from the roots after which they were washed thoroughly under running tap water and allowed to dry up over natural air. Each plant root sample was assessed for galls and were scored according to the Bridge and Page (1980) chart for gall rating. (See Appendix 4)

### **3.8.4 Nematode egg-masses**

Nematode egg-masses on roots were determined by dipping seedlings in 0.015% Phloxine B solution for 20 minutes as described by Daykin and Hussey (1985).

### **3.8.5 Data collected and reported**

- Root-knot nematode gall scores
- Nematode egg-mass
- Reproduction factor (Final population divided by initial population)

### **3.9 Plant parameters**

#### **3.9.1 Seedling height (cm)**

Seedling height was taken from the ground level to the apical bud with a meter rule for shorter plants and tape measure for taller plants at 4 and 8 weeks after application of treatment.

#### **3.9.2 Number of leaves**

Number of leaves were counted at 2, 5 and 8 weeks after emergence. Leaves of 12 sampled plants were counted on each plot and the mean number of leaves per plot was then determined for each treatment.

#### **3.9.3 Stem girth (cm)**

Stem girth of seedling was measured with the digital micrometre screw gauge at 4 and 8 weeks after application of treatments. The digital micrometre screw gauge was clamped around the stem of 12 sampled plants (one at a time) and the diameter read from the screen and recorded.

#### **3.9.4 Root length (cm)**

Root length was measured with a meter rule at 6 weeks after sowing and then 12 weeks after sowing. Root lengths of sampled plants for gall scoring were measured and recorded appropriately.

#### **3.9.5 Fresh weight of shoots (g)**

Fresh weight of shoots was taken at harvesting. The plants were uprooted, shoots separated from the roots, washed off the attached soil, allowed to air dry and then weighed on the weighing balance.

The fresh weights of the shoots were then recorded.

### **3.9.6 Dry weight of shoots (g)**

Dry weight of shoots was taken after harvesting. The shoots of the 12 plants sampled were separated from the roots. They were dried in the oven at 70 °C for 72 hours after which the dry weights of the shoots were taken with the weighing balance and then recorded.

### **3.9.7 Fresh weight of roots (g)**

Fresh weight of roots was also taken after harvesting. After the shoots were separated from the roots of the 12 sampled plants, the roots were taken and washed thoroughly off all attached soils and then allowed to air dry. They were then weighed on the weighing balance and values recorded accordingly.

### **3.9.8 Dry weight of roots (g)**

Dry weight of roots was taken after harvesting. After the fresh weight of the 12 sampled plants were taken, they were dried in the oven at 70 °C for 72 hours after which the dry weights of the roots were taken with the weighing balance and then recorded accordingly.

### **3.9.9 Data collected and reported**

- Seedling height (cm)
- Number of leaves
- Stem girth (cm)
- Root length (cm)
- Shoot fresh weight (g)
- Shoot dry weight (g)
- Root fresh weight (g)
- Root dry weight (g)



## CHAPTER FOUR

### 4.0 RESULTS

#### 4.1 Laboratory analysis

##### 4.1.1 Seed germination, shoot length, root length and seedling vigour index as affected by duration of exposure of tomato seeds to *Trichoderma harzianum* as seed treatment

When ten (10) g of tomato seeds were treated with recommended dose of 0.2 g of *Trichoderma harzianum* (Eco-T) and left for different time intervals before plating, percentage germination values indicated that there was no significant difference in seed germination among the various treatments (Table 5).

Shoot length was also not significantly ( $p \leq 0.05$ ) affected by *T. harzianum* although seeds coated and exposed for 3 hours before plating recorded greater length of shoots at 7 days after germination as compared to the other treatments. Seeds exposed for 30 hours recorded the lowest shoot lengths and differed only from *T. harzianum* (Eco-T) exposed 3 hrs before planting.

Root lengths were significantly affected ( $p \leq 0.05$ ) by *T. harzianum* (Table 5). Seeds coated for 30 hrs and 48 hrs recorded lower root lengths which were not significantly different from each other. However, seeds coated for a period of 30 minutes recorded the highest mean root length and this differed significantly from 30 hrs and 48 hrs exposure treatments.

Vigour index of seedling was also evaluated (Table 5). Treatments did not significantly affect seedling vigour however seeds coated for a period of 30 minutes recorded the highest seedling vigour index of 868.20 but differed significantly only from the 30 hrs and 48 hrs exposure treatments.

**Table 5: Seed germination, shoot length, root length and seedling vigour index as affected by duration of exposure of tomato seeds to *Trichoderma harzianum* as seed treatment.**

Treatments	% Germination	Mean Shoot Length(cm)	Mean Root Length(cm)	Vigour Index
Control (No coating with Eco-T)	88.00	2.66	5.97	757.40
Seed coated with Eco-T for 30 min	95.00	3.08	6.05	868.20
Seed coated with Eco-T for 1 hr	93.00	2.96	5.55	791.10
Seed coated with Eco-T for 3 hrs	94.00	2.76	5.92	817.90
Seed coated with Eco-T for 6 hrs	93.00	3.52	5.13	804.80
Seed coated with Eco-T for 8 hrs	95.00	2.72	5.82	813.00
Seed coated with Eco-T for 24 hrs	93.00	2.95	5.46	787.90
Seed coated with Eco-T for 30 hrs	93.00	2.29	4.58	644.70
Seed coated with Eco-T for 48 hrs	94.00	2.29	4.71	660.9
LSD (0.05)	8.68	6.40	0.96	168.5
Grand Mean	93.11	3.53	5.47	772.00
CV (%)	6.40	24.9	12.2	15.00

#### **4.1.2 Seed germination, shoot length, root length and seedling vigour index as affected by different concentrations of *Trichoderma harzianum* used as seed treatment on UC 82 tomato seeds**

Treating seed with *T. harzianum* (Eco-T) at different concentrations from (0.2 to 0.4 g/10 g seeds) did not significantly differ ( $p \leq 0.05$ ) for all the seedling parameters measured (Table 6).

**Table 6: Seed germination, shoot length, root length and seedling vigour index as affected by concentration effect of *Trichoderma harzianum* used as seed treatment on UC 82 tomato seeds**

Treatments	% Germination	Mean shoot length (cm)	Mean root length (cm)	Vigour index
Control (No coating)	92.00	3.11	4.14	671.20
Seed coated with Eco-T 0.2 g/10 g seeds	89.00	3.06	4.47	671.90
Seed coated with Eco-T 0.25 g/10 g seeds	90.00	3.15	4.20	659.80
Seed coated with Eco-T 0.3 g/10 g seeds	90.00	2.95	3.87	613.00
Seed coated with Eco-T 0.35 g/10 g seeds	93.00	3.22	3.83	654.40
Seed coated with Eco-T 0.4 g/10 g seeds	93.00	9.55	4.19	673.00
LSD (0.05)	5.28	7.88	2.35	240.9
Grand Mean	91.17	4.20	4.12	657.00
CV (%)	3.90	26.9	38.50	24.70

#### **4.1.3 Seed germination, seedling shoot length, seedling root length and seedling vigour index as affected by duration of exposure of UC 82 tomato Seed to *Trichoderma harzianum* at different concentrations**

From the analysis (Table 7), it is evident that the durations of exposure had no significant ( $P \leq 0.05$ ) effect on tomato seed germination. The observed highest germination percentages recorded in *T. harzianum* used for seed treatment at 0.4g/10g seeds for both 30 minutes and 3 hours were not significantly different from the other treatments respectively.

The effect of concentration and duration of exposure on the shoot lengths of seedlings of seeds pre-coated with different levels of *T. harzianum* (Eco-T) concentrations for different durations prior to

sowing were also reported in Table 7. The treatments had no significant ( $p \leq 0.05$ ) effect on shoot length of seedlings but 0.4 g of *T. harzianum* (Eco-T) used as seed treatment for a period of 30 minutes and 0.4 g of *T. harzianum* (Eco-T) for a period of 3 hours recorded highest values. Coating tomato seeds with 0.4g of *T. harzianum* (Eco-T) for a period of 30 minutes recorded highest mean values of shoot length as compared to 0.4g of Eco-T for a period of 3 hours.

Seedling root length values (Table 7) indicate that the treatments were not significantly different.

Same could be said about seedling vigour index values (Table 7) that also indicate there was no statistical difference ( $P \leq 0.05$ ) among the treatments



**Table 7: Seed germination, seedling shoot length, seedling root length and seedling vigour index as affected by duration of exposure of UC 82 tomato Seed to *Trichoderma harzianum* at different concentrations**

Treatments	%Germination		Mean seedling shoot length		Mean seedling root length		Seedling vigour index (I)	
	Duration							
Concentration	30 min	3 hr	30 min	3 hr	30 min	3 hr	30 min	3 hr
Control	60.00	60.00	4.42	4.06	1.42	1.84	18.59	18.79
Seed coated with Eco-T 0.2 g/10 g seeds	63.00	63.00	4.44	4.48	1.65	1.93	19.45	20.01
Seed coated with Eco-T 0.4 g/10 g seeds	67.00	65.00	4.68	3.75	2.25	1.83	21.47	18.81
LSD (0.05)	9.26	16.03	0.93	0.54	0.79	0.46	4.06	2.34
Grand Mean	63.30	62.70	4.51	4.10	1.77	1.86	19.84	19.20
CV (%)	17.10		14.70		29.50		14.00	

#### 4.2.1 Physical and chemical soil analysis of study area.

Table 8 shows the details of soil analysis conducted on the study field. In the analysis, pH was observed to be slightly acidic to neutral.

**Table 8: Soil physical and chemical analysis**

Soil Nutrient	Value
pH 1:1	6.18
Available phosphorus	2.90 (mg/kg)
Total nitrogen	0.11 (%)
Organic carbon	0.24 (%)
Exchangeable calcium	1.17 (Cmol/kg)
Exchangeable magnesium	0.21 (Cmol/kg)
Exchangeable potassium	0.26 (Cmol/kg)
Exchangeable sodium	0.32 (Cmol/kg)
Sand	61.24 (%)
Silt	10.64 (%)
Clay	28.13 (%)
Textural Class	Sand Clay Loam

NB: Soil sample was taken from a depth of zero to thirty (0-30) cm

#### 4.2.2 Impact of Eco-T (*Trichoderma harzianum*) on UC 82 tomato seedling performance.

##### 4.2.2.1 Effect of *Trichoderma harzianum* on seedling emergence 2 weeks after sowing

Treated seeds with 0.4 g and 0.2 g *T. harzianum* (Eco-T) had significant number of the seedling emerged as compared to the control (Table 9). The 0.4 g *T. harzianum* (Eco-T) treated seed was not significantly different from the control and the 0.2 g *T. harzianum* (Eco-T) treated seeds but the latter was significantly ( $p \leq 0.05$ ) different from the control.

**Table 9: Effect of *Trichoderma harzianum* on tomato seedling emergence**

Treatment	% Number of Emergence (Plant Stand) 2 WAS
Control (No Treatment)	46.10
Seed coated with Eco-T 0.2 g/10 g of seeds	60.68
Seed coated with Eco-T 0.4 g/10 g of seeds	52.87
LSD ( $_{0.05}$ )	8.37
Grand Mean	53.2
CV (%)	9.10

##### 4.2.2.2 Effect of *Trichoderma harzianum* on tomato plant performance

There were significant differences ( $p \leq 0.05$ ) in the treatments' effect on plant height and girth (Table 10). Seeds coated with 0.4 g and 0.2 g Eco-T recorded the highest and second highest plant heights respectively.

On the other hand, plant stem girth was not significantly ( $p \leq 0.05$ ) affected by any of the treatments in the first 4 weeks however, the situation changed by the 8th week. Stem girths varied highly significantly ( $p \leq 0.05$ ) across the treatments, with seeds treated with 0.4 g and 0.2 g *T. harzianum* (Eco-T) recording the greatest tomato plant girths.

**Table 10: Effect of *Trichoderma harzianum* on tomato plant height and plant stem girth at 4 and 8 weeks after application of treatment (WAA)**

Treatments	Mean plant height(cm)		Mean plant stem girth (cm)	
	4WAA	8WAA	4WAA	8WAA
Control	9.43	23.00	1.35	2.15
Velum at 3.8 mL/7 L of water	10.50	30.65	1.46	2.81
Seed coated with Eco-T 0.2 g/10 g seeds	10.96	33.96	1.51	3.11
Seed coated with Eco-T 0.4 g/10 g seeds	12.56	34.22	1.73	3.07
Drenching with Eco-T 0.2 g/25 mL water	10.10	23.29	1.81	2.39
Drenching with Eco-T 0.4 g/25 mL water	9.71	27.63	1.37	2.73
LSD (0.05)	1.92	5.56	0.49	0.41
Grand Mean	10.54	28.79	1.54	2.71
CV (%)	12.10	12.80	21.20	10.10

In week 4, seeds treated with 0.4 g and 0.2 g *T. harzianum* (Eco-T) as well as seedlings drenched with 0.2 g *T. harzianum* were observed to have highly significant ( $p \leq 0.001$ ) increase in leaf development over the other treatments (Table 11). The control, Velum and drenching with 0.4 g treatments were however not significantly ( $p \leq 0.05$ ) different from each other. However, there were no differences in treatments at 8 weeks after application.

Velum drenched treatment, 0.2g and 0.4g *T. harzianum* (Eco-T) treatments recorded the highest observed values of plant leaves: 4.60, 5.08 and 5.35 respectively. These treatments were not however significantly different from the other treatments in week 4. By week 8, 0.2 g *T. harzianum*

(Eco-T) was significantly different producing 23.35% more leaves than the control but not different from the rest of the other treatments.

**Table 11: Effect of *Trichoderma hazianum* on tomato leaves and branches at 4 and 8 weeks after application (WAA)**

Treatments	Mean Leaf length (cm)		Mean Number of leaves (cm)	
	4WAA	8WAA	4WAA	8WAA
Control	3.69	5.36	4.12	9.75
Velum at 3.8 mL/7 L of water	3.92	6.46	4.60	11.75
Seed coated with Eco-T 0.2 g/10 g seeds	4.30	8.36	5.08	12.72
Seed coated with Eco-T 0.4 g/10 g seeds	4.98	7.15	5.35	12.27
Drenching with Eco-T 0.2 g/25 mL water	4.11	6.97	4.43	12.00
Drenching with Eco-T 0.4 g/25 mL water	3.77	5.96	4.52	12.35
LSD (0.05)	0.45	2.49	4.68	1.49
Grand Mean	4.12	6.71	0.79	11.81
CV (%)	7.30	20.40	5.60	8.30

Mean root length 6 weeks after treatment application showed no significant ( $p \leq 0.05$ ) differences among the treatments. By the 12<sup>th</sup> week, velum treated plants recorded the longest root length of 62.82cm which was 40.30% longer than the control treatment root. The root length of velum treated plants was however not different from the other treatments (Table 12).

**Table 12: Effect of *Trichoderma harzianum* on tomato roots at 6 and 12 weeks after treatment application (WAA)**

Treatments	Mean Root Length (cm)	
	6WAA	12 WAA
No treatment	18.75	37.50
Velum at 3.8 mL/7 L of water	20.12	62.82
Seed coated with Eco-T 0.2 g/10 g seeds	20.95	59.34
Seed coated with Eco-T 0.4 g/10 g seeds	21.70	61.52
Drenching with Eco-T 0.2 g/25 mL water	18.57	58.92
Drenching with Eco-T 0.4 g/25 mL water	18.30	52.19
LSD (0.05)	6.33	12.47
Grand Mean	19.73	55.4
CV (%)	21.30	14.90

Mean shoot and root fresh weights of tomato plants 6 and 12 weeks after treatment application (Table 13) showed no significant ( $p \leq 0.05$ ) differences among the treatments in week 6 but there in week 12, there was significant difference between the controls and the rest of the treatments.

**Table 13: Effect of *Trichoderma harzianum* on fresh weights at 6 and 12 weeks after application of treatment (WAA)**

Treatments	Mean shoot fresh weight (g)		Mean root fresh weight (g)	
	6WAA	12 WAA	6WAA	12 WAA
Control	68.24	137.00	11.12	41.78
Velum at 3.8 mL/7 L of water	96.52	229.8	17.19	76.27
Seed coated with Eco-T 0.2 g/10 g seeds	89.79	218.4	15.91	76.39
Seed coated with Eco-T 0.4 g/10 g seeds	90.69	260.4	17.43	91.65
Drenching with Eco-T 0.2 g/25 mL water	90.99	249.0	15.30	83.68
Drenching with Eco-T 0.4 g/25 mL water	93.73	275.5	16.36	76.78
LSD (0.05)	12.24	68.35	4.40	28.06
Grand Mean	88.30	228.4	15.55	74.40
CV (%)	9.20	19.90	18.80	25.00

In week 6, the treatment that recorded the highest shoot dry weight, ie. drenching with 0.2 g *T. harzianum* (Eco-T) per 25 mL of water was significantly different from the control but not different from the rest of the treatments. By week 12 seedlings drenching with 0.4 g of *T. harzianum* (Eco-T) and 0.2 g of *T. harzianum* (Eco-T) per 25 mL of water were observed to have significantly ( $p \leq 0.05$ ) increased shoot dry weight compared to the other treatments (Table 14).

There was no significant ( $p \leq 0.05$ ) differences in treatments' effect on root dry weight in week 6 likewise week 12.

**Table 14: Effect of *Trichoderma harzianum* on dry weights at 6 and 12 weeks after treatment application (WAA)**

Treatments	Shoot dry weight (g)		Root dry weight (g)	
	6 WAA	12 WAA	6 WAA	12 WAA
Control	31.31	85.10	0.93	18.28
Velum at 3.8 mL/7 L of water	47.20	112.40	1.68	21.40
Seed coated with Eco-T 0.2 g/10g seeds	45.25	107.40	1.75	19.77
Seed coated with Eco-T 0.4 g/10g seeds	48.72	121.8	2.12	21.57
Drenching with Eco-T 0.2 g/25 mL water	53.63	124.4	1.60	15.13
Drenching with Eco-T 0.4 g/25 mL water	49.98	132.0	2.06	21.29
LSD (0.05)	12.44	29.06	0.92	11.10
Grand Mean	46.00	113.80	1.69	19.60
CV (%)	17.90	16.90	36.30	37.60

Naturally occurring nematode populations on the field in each plot were sampled and counted before treatment application. No significant ( $p \leq 0.05$ ) differences were observed for initial numbers for each treatment (Table 15).

In weeks 6 and 12, although there was no significant difference in nematode initial populations across treatments, the control was significantly ( $p \leq 0.05$ ) higher than the other treatments.

The reproductive factor indicates the efficacy of the treatments in controlling the nematodes' attack on the tomato plants.

Treatments with RF values less than one are considered effective against the incidence of the tomato root knot nematode (Table 15). It is clear that the Velum treatment had the least RF values at 6 and 8 WAA indicating the efficiency of the treatment.

Egg masses were extracted and counted in weeks 6 and 12 respectively. The control treatment again recorded significantly ( $p \leq 0.05$ ) highest egg masses compared to the other treatments.



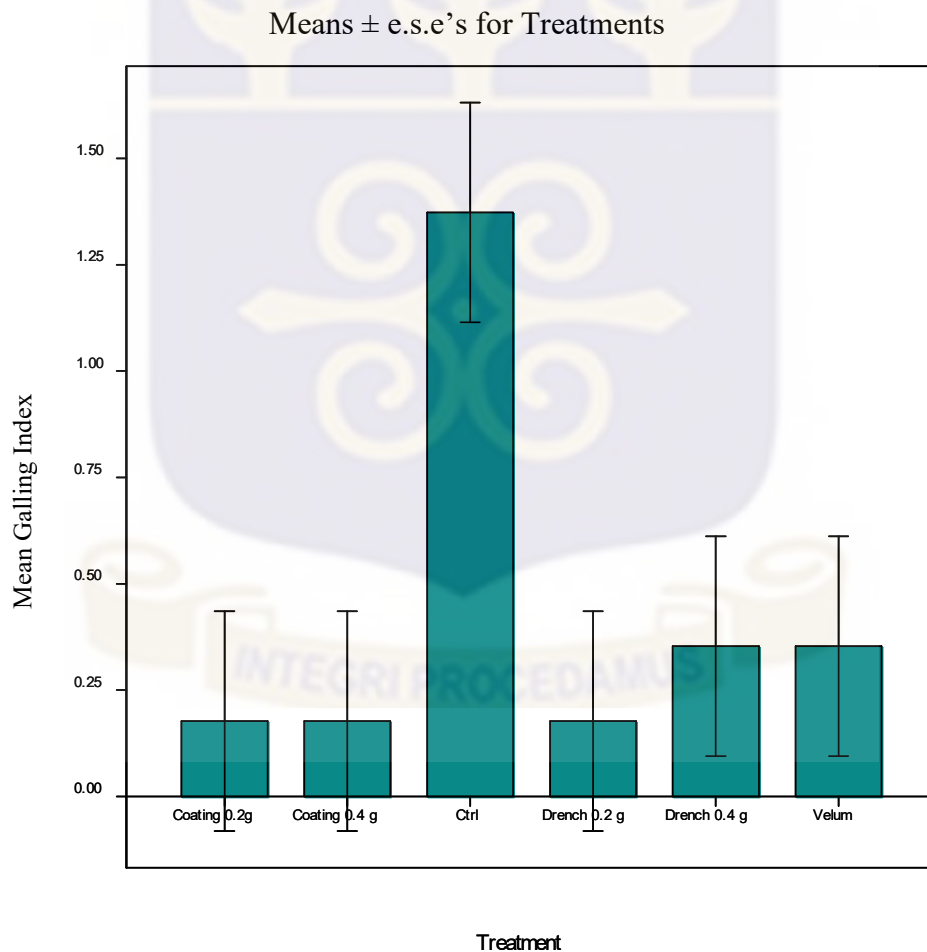
**Table 15: Effect of *Trichoderma harzianum* on Nematode Population and Egg- mass at 6 and 12 Weeks After Treatment Application (WAA)**

Treatments	Nematode population			Reproductive factor		Egg masses	
	Initial	6 WAA	12 WAA	6 WAA	12 WAA	6 WAA	12 WAA
Control	390.50 (19.62)	408.20 (20.06)	475.00 (21.59)	1.02	1.08	222.80 (14.60)	363.80 (17.76)
Velum at 3.8 mL/7 L of water	319.50 (17.40)	199.50 (13.50)	77.80 (8.65)	0.78	0.64	35.00 (5.48)	16.50 (4.01)
Seed coated with Eco-T 0.2 g/10 g seeds	284.50 (16.36)	189.50 (12.99)	279.00 (16.02)	0.79	1.23	55.50 (7.35)	98.20 (9.450)
Seed coated with Eco-T 0.4 g/10 g seeds	210.00 (14.07)	123.00 (11.05)	129.00 (11.30)	0.79	1.02	48.20 (6.64)	76.00 (8.65)
Drenching with Eco-T 0.2 g/25 mL water	205.00 (13.31)	145.80 (11.79)	137.50 (11.43)	0.89	0.97	144.00 (11.44)	109.00 (9.45)
Drenching with Eco-T 0.4 g/25 mL water	228.50 (14.86)	197.20 (13.95)	151.00 (11.86)	0.94	0.85	124.20 (10.66)	115.80 (9.95)
LSD (0.05)	7.13	5.01	5.43			4.99	7.03
Grand Mean	15.94	13.89	13.47			9.37	9.88
CV (%)	29.7	23.9	26.70			35.40	47.2

Nematode populations and Egg mass values in parenthesis were transformed with the square root transformation too

#### 4.2.2.3 Effect of *Trichoderma harzianum* on tomato root galling index 6 weeks after treatment application (WAA)

Galling on roots of the plants was evaluated at 6 WAA. The gall scoring scale (Appendix 4) was used to determine the magnitude of gall formation on the roots (Figure 1). Three treatments: Coating with *T. harzianum* (Eco-T) at 0.2 g concentration per 10 g of tomato seeds, Coating with 0.4 g and Drenching with *T. harzianum* (Eco-T) at a rate of 0.2 g/25 mL of water recorded the least gall index on tomato roots. All the treatment effects did not differ significantly ( $p \leq 0.05$ ) for gall indices but for the control which recorded the highest galling index which was statistically significantly ( $p \leq 0.05$ ) different from the others.



**Figure 1: Root galling indices at 6 WAA as influenced by seed treatments**

(NB: Original means values were transformed with the square root transformation tool).

#### 4.2.2.4 Effect of *Trichoderma harzianum* on tomato root galling index at 12 weeks after treatment application

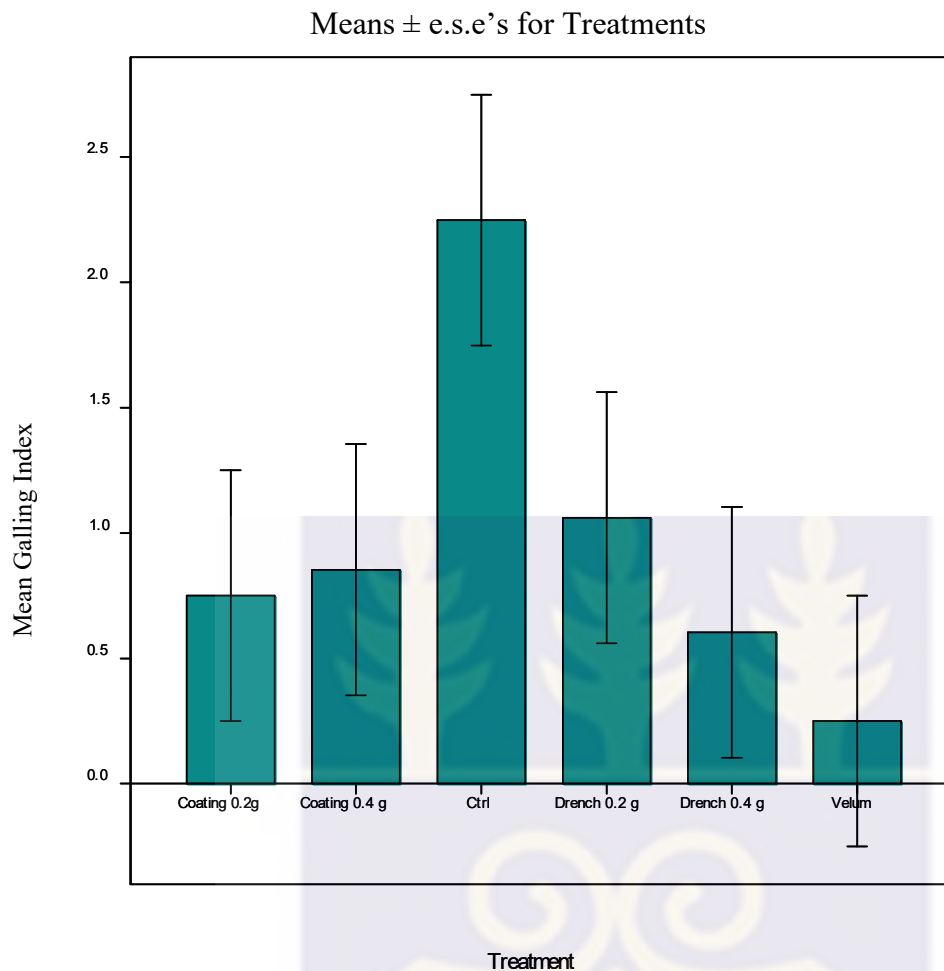
Pictures of the various degrees of root gall formation on the tomato roots are shown as Figure 2. Severity of gall formation is intense in the control compared to the *T. harzianum* and velum treatments.



*Source: Researcher's field*

**Figure 2: Presence of nematode root galls on tomato seedlings in (A) Eco-T (*T. harzianum*) plots; (B) control plot showing heavy galling and (C) velum plot.**

Figure 3 displays the mean gall incidence for the various treatments. The control treatment recorded the highest root gall index compared to the other treatments. Plants that were drenched with Velum recorded the least root gall index. Velum treatment significantly ( $p \leq 0.05$ ) reduced the gall index compared to the control but the effect was not significantly ( $p \leq 0.05$ ) different from the *T. harzianum* treatments.



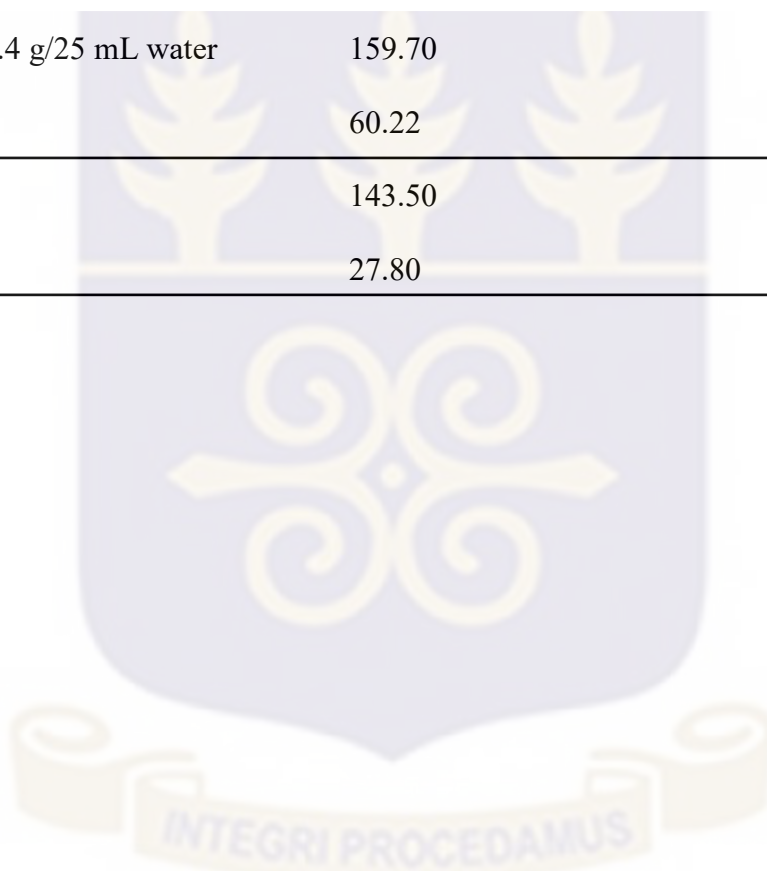
**Figure 3: Root gallling indices at 12 WAA as influenced by seed treatments**

(NB: Original mean values were transformed with the square root transformation tool).

The effects of the treatments on average tomato weight and tomato yield were statistically significant (Table 16). All the treatments effects did not show any significant differences for tomato weight and yield among themselves but these all differed significantly from the control. Velum (at 3.8 mL/7 L of water) treatment recorded the highest yield and weight of fruits with control plot recording the least.

**Table 16: Effect of *Trichoderma harzianum* treatments on tomato yield**

Treatments	Average tomato weight (g)/plot	Tomato yield (t)/ha
Control	49.50	6.43
Velum at 3.8 mL/7 L of water	170.90	22.22
Seed coated with 0.2 g/10 g seeds	166.40	21.62
Seed coated with 0.4 g/10 g seeds	169.40	22.00
Drenching with 0.2 g/25 mL water	145.30	18.89
Drenching with 0.4 g/25 mL water	159.70	20.76
LSD (0.05)	60.22	7.83
Grand Mean	143.50	18.7
CV (%)	27.80	27.9



### 4.3 Partial budget analysis for using treatments.

Positive change in net income (+ 478.94) shows that the use of *T. harzianum* (Eco-T) on tomato fields to control nematodes is beneficial (Table 17).

**Table 17: Eco-T (*Trichoderma harzianum*) as seed treatment or coating against untreated plots (Control)**

Increase in Net Income		Reduction in Net Income	
a. Added income because of the change		c. Added costs because of the change	
Yield Sales	GH¢ 511.94	<u>Eco-T (<i>T. harzianum</i>)</u>	GH¢ <u>8.00</u>
<hr/>	<hr/>		
<b>Total</b>	GH¢ <u>511.94</u>	<b>Total</b>	GH¢ <u>8.00</u>
b. Decreased costs because of the change		d. Decreased income because of the change	
<u>Banned Furadan</u>	GH¢ <u>30.00</u>	<u>Yield Sales</u>	GH¢ <u>150.00</u>
<u>Extra seed for replacement</u>	GH¢ <u>45.00</u>	<hr/>	<hr/>
<u>Fungicide</u>	GH¢ <u>35.00</u>		
<u>Extra NPK fertigation</u>	GH¢ <u>15.00</u>		
<b>Total</b>	GH¢ <u>125.00</u>	<b>Total</b>	GH¢ <u>150.00</u>
<b>Sub Total</b>	GH¢ <u><u>636.94</u></u>	<b>Subtotal</b>	GH¢ <u><u>158.00</u></u>
<b>Change in net income =</b>	<b>+ (478.94)</b>		

Positive change in net income (+ 407.50) shows that the use of Velum at 3.8 mL/7 L of water on tomato fields to control nematodes is beneficial (Table 18).

**Table 18: Partial budget for using velum (fluopyram SC 400) as soil drenching against untreated plot (control)**

<b>Increase in net income</b>	<b>Reduction in net income</b>
a. Added income because of the change	c. Added costs because of the change
<u>Yield sales</u> GH¢ <u>532.5</u>	<u>Velum (Fluopyram SC 400)</u> GH¢ <u>100.00</u>
<b>Total</b> GH¢ <u>532.5</u>	<b>Total</b> GH¢ <u>100.00</u>
c. Decreased costs because of the change	d. Decreased income because of the change
<u>Banded Furadan</u> GH¢ <u>30.00</u>	<u>Yield Sales</u> GH¢ <u>150.00</u>
<u>Extra seed for replacement</u> GH¢ <u>45.00</u>	
<u>Fungicide</u> GH¢ <u>35.00</u>	
<u>Extra fertigation</u> GH¢ <u>15.00</u>	
<b>Total</b> GH¢ <u>125.00</u>	<b>Total</b> GH¢ <u>150.00</u>
<b>Sub Total</b> GH¢ <u><u>657.5</u></u>	<b>Subtotal</b> GH¢ <u><u>250.00</u></u>
<b>Change in net income =</b> + (407.50)	

## CHAPTER FIVE

### 5.0 DISCUSSION

#### 5.1 Impact of *Trichoderma harzianum* duration of exposure, concentration and combination of exposure time and concentration on seed germination and seedling vigour

The effect of Eco-T (*T. harzianum*) on percentage germination of tomato seeds in the laboratory indicated that, *T. harzianum* used as a slurry at varying concentrations ranging from 0.2 g/10 g of seed (recommended) to 0.4 g/10 g marginally increased tomato seed germination but was not different from the control. There was no difference between the other treatments and control, therefore, results are different from that of Asaduzzaman *et al.* (2010) that five *Trichoderma* strains (*T. virens* IMI-392430, *T. pseudokoningii* IMI-392431, *T. harzianum* IMI-392432, *T. harzianum* IMI-392433 and *T. harzianum* IMI-392434) when used for seed treatment before planting gave early and higher germination percentages of chilli.

Seedling vigour was not significantly impacted by some of the treatments compared to the control. However, the recommended rate (0.2 g/10 g seed) at different durations of exposure (30 mins to 48 hrs) significantly influenced the seedling vigour. The highest seedling vigour was observed with *T. harzianum* at a duration of exposure of 30 minutes. Similar finding was reported by Asaduzzaman *et al.* (2010) that chili seeds treated with *Trichoderma* strains had better seedling vigour index compared to the control. Poor seedling vigour observed in the other duration of exposure treatments in the laboratory could be attributed to extended periods of soaking in slurry which might have influenced available oxygen for gas exchange during the germination period.

Similar result was reported by Fausey and McDonald (1985) that retarded germination of corn was observed when seeds were exposed to excess moisture or flooding. Optimum moisture levels are necessary for seeds imbibition in order to soften hard seed coats, activate enzyme systems and

oxidative phosphorylation to facilitate germination processes in plant. Extended exposure was also observed to have favoured increased contamination with fungi especially *Aspergillus flavus* during incubation period probably due to the high seed moisture contents prior to plating. The fact that tomato seed germination percentage was not significantly affected by any of the treatments indicated that tomato seed germination is influenced by several other factors aside seed treatments.

## **5.2 Experiment 2: Field Study**

### **5.2.1 Physical and chemical analyses of field soil**

The initial soil physical and chemical analyses showed low soil fertility as the most limiting factor in the study area. Although soil pH was adequate for tomato production, all other available nutrients were below sufficient proportions necessary to boost crop growth, development and yield. Poor soil fertility has been reported as a major challenge to crop production in developing countries and especially among resource-poor farmers (Mokwunye *et al.*, 1996). NPK fertilizer 15:15:15 at a rate of 20 g/plant was applied across all the plots in order to bolster plant growth and development as well as yield. Chen (2006) stated that nutrients necessary for plants growth must be available in sufficient and right proportions in order to attain optimal growth of plants.

### **5.2.2 Impact of *Trichoderma harzianum* and velum on nematode population, egg masses and tomato root galling**

Velum (Fluopyrum SC 400) at a rate of 3.8 mL/7 L of water was effective in controlling nematode populations and subsequently egg production and root galling and hence increased the chance of nutrient and water uptake through the roots. This is in line with the report by Abdulai (2014) stating that plots that received Velum treatments (VSC at 3.8 mL/7 L of water) had the least reproductive factor (RF) values compared to control plots.

Nematode populations on control plots were about twice as much as nematode populations on Velum and *T. harzianum* plots. Also, root galling was observed to be very severe on roots of tomato plants on control plots but on roots of tomato plants on Velum at 3.8 mL/7 L and *T. harzianum* plots, galling was drastically reduced. Nematode egg mass was relative to nematode populations in that, the higher the nematode population, the higher the egg mass and vice versa. Higher reproductive factor (RF) of nematodes in control plots as compared to the other treatment plots was reported indicating the faster rate at which nematodes in control plots multiplied. These findings are in agreement with Chindo *et al.* (2006) and Nadary *et al.* (2006) who reported that high reproduction rates of nematode inoculums of *Meloidogyne incognita* induced greater root galling and egg masses due to the larger populations of the nematode. Windham *et al.* (1989) reported a decrease in egg production by the root-knot nematode *Meloidogyne arenaria* after *T. harzianum* and *T. koningii* preparations were used as soil treatments. However, Baker (1986) reported that extremely high concentrations of *T. harzianum* make the fungus pathogenic causing hardening of seedlings (Callus formation).

### **5.2.3 Impact of *Trichoderma harzianum* and Velum on Tomato Vegetative Growth and Yield**

Most emphatic rhizosphere competent strains of *Trichoderma* spp. can be added to soil or seeds because they germinate and colonize root surfaces once into contact with plant roots and some moisture, preventing intrusion of other parasitic organisms that could hinder root development (Ghahfarokhy *et al.* 2011)

Seedling emergence was significantly influenced by *T. harzianum* treatments. High emergence (plant stand) levels were observed on *T. harzianum* treated plots compared to the control by 6 to 14%.

Leaf development, plant height and stem girth of tomato plants on the field were significantly influenced by both *T. harzianum* and Velum (Fluopyram 400 SC) treatments. Plant height of tomato on *Trichoderma* and *Velum* treated plots were also taller than the control. Increased vegetative growth can be attributed to *T. harzianum*'s potential to immediately take over the young radicle of germinating seeds (Sivasithamparam *et al.*,1998) by forming nests around the root zone promoting rapid root growth for nutrients absorption. *T. harzianum* prevents invasion of other soil organisms that may compete for resources by parasitizing these organisms and breaking down chitin and available cellulose. Leaf formation as well as rapid plant growth was observed in *T. harzianum* and *Velum* (Fluopyram) treatments.

Harman *et al.* (2004) reported that, *T. harzianum* is known to increase fertilizer and nutrient uptake and utilization that boost rapid plant growth and increase in the number of leaves which results in increase in surface area occupied by the plant hence more sunlight trapping by these leaves for photosynthesis which will result in healthy start and increased yield per plant.

*T. harzianum* treated plots at 0.4 g/10 g seeds were observed to have significantly increased leaf length at 4 weeks after application. Leaf lengths among the other treatments and the control were however not significant. For the number of leaves however, difference was observed between the control and the other treatments. Application of *Velum* did not affect both the leaf length and leaf number compared to the *Trichoderma* treatments. These results explain the fact that *Velum*, unlike *T. harzianum* only controls nematode populations and prevents galling but does not produce enzymes that break down soil organic matter to release nutrients for plant uptake and development.

Tomato plots that were drenched with *T. harzianum* (0.4 g/25 mL of water) recorded highest fresh shoot and dry weights followed by plants that were drenched with *T. harzianum* 0.2 g/25 mL of water. Plants on *Velum* plots recorded fresh and dry weights a little higher than plants on *T.*

*harzianum* treated seeds plots (0.4 g/10 g seeds and 0.2 g/10 g seeds) although the differences were very slight. The increase in plant height, number and length of leaves, fresh and dry weights of plants could be due to a reduction in nematode population resulting in lower gall formation that enhances plant nutrient uptake (Pattison, 2007).

A similar trend was observed for the length, fresh and dry weights of the roots of tomato plants. Sharon *et al.* (2001) reported similar observations in their study with tomato.

Yield data showed that Velum treatment was effective in giving higher yields. This is because Velum was highly effective in keeping nematode populations lower than *T. harzianum* treated plots, totally reducing root galling close to zero and allowing free uptake of nutrients supplied to the tomato plants. Similar result was reported in maize production by Windham *et al.* (1989). Yield on *T. harzianum* (Eco-T) plots were comparably lower probably because *T. harzianum* was effective to only a stage in the plant development cycle. *T. harzianum* is known to have a short life span of 21 to about 35 days as reported by (Lester, 2010).

### **5.3 Economic benefits analysis of *Trichoderma harzianum* and velum treatments with a partial budget**

A positive difference between the net incomes of using *T. harzianum* and Velum in tomato production shown by partial budgeting suggested that using *T. harzianum* resulted in an additional income of GH¢ 71.44/ha and was economically beneficial compared to Velum treatment. This is in line with similar findings from analytical research on “Economic feasibility of using the parasitic wasp *Pediobius foveolatus* to manage the mexican bean beetle in soyabean” (Reichelderfer, 1979). It showed a lower cost incurred for the use of biological control which resulted in higher net return per hectare as compared with the use of chemical insecticides.

## CHAPTER SIX

### 6.0 CONCLUSIONS AND RECOMMENDATIONS

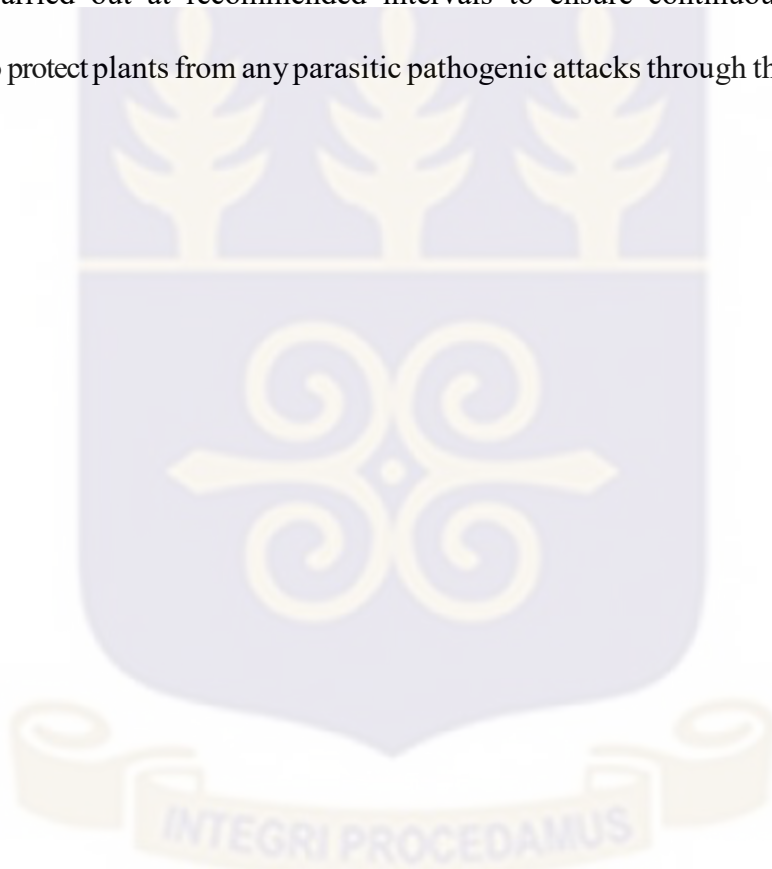
#### 6.1 Conclusions

- Tomato seed germination, seedling vigour and plant development were improved when *Trichoderma harzianum* was used as seed treatment compared to the non-treated controls. However, the different concentrations of *T. harzianum* employed for seed treatment (0.2 g/10g seeds and 0.4g/10g seeds) and for seedling drenching (0.2 g/25 mL of water and 0.4g/25 mL of water) showed no significant differences among treatments.
- Velum (Fluopyram 400 SC) 3.8 mL/7L of water and *T. harzianum* seed treatments prior to sowing at 0.2 g/10 g seeds; 0.4 g/10 seeds for 30 mins; and drenching at 0.2 g/25 mL of water; 0.4 g/25 mL of water 6 weeks after emergence were effective in reducing nematode populations and the incidence of root galling.
- Tomato fruit yield on the field for Velum and *T. harzianum* treated plots were not statistically different.
- *T. harzianum* was more cost effective with a change in net income of (+ 478.94) compared to Velum (Fluopyram 400 SC) with a change in net income of (+ 407.50) after tomato yield sales.

## 6.2 Recommendations

Generally, low yields were recorded in the study due to the execution of the experiment under extreme climatic conditions prevailing on the experimental site at the time, therefore, a repeat of the study is recommended in the major season to ascertain the effect of *T. harzianum* either as a seed treatment or for drenching in the control of root knot nematodes.

Agronomic integration of soil/seedling drenching as well as foliar application of *T. harzianum* slurry must be carried out at recommended intervals to ensure continuous availability of the beneficial fungi to protect plants from any parasitic pathogenic attacks through the plant growth cycle.



## REFERENCES

- Abdulai, M., Moss, R., Nboyine, J. A., Opore-Atakora, D. and Seidu, A. (2013).** The effects of the microbial amendment, Eco-T (*Trichoderma harzianum*) and Enrich (*Herbaspirillum seropedicae* and *Bacillus subtilis*) on growth and yield of cotton in Ghana. *Advanced Crop Science*. 3(11): 729–735.
- Abdulai S. (2014).** Efficacy of selected nematicide for control of nematodes on tomato (*Solanum lycopersicon*).
- AccuWeather, (2017).** Inc <https://www.accuweather.com/en/gh/ada/467/weather-forecast/467>
- Acquaah, G. (2005).** *Horticulture, Principles and Practices* (3<sup>rd</sup> Edition). Pearson Education Inc., Upper Saddle River, New Jersey.
- Adeoye, I. B., Odeleye, O. M. O., Babalola, S. O. and Afolayan, S. O. (2009).** Economic Analysis of Tomato Losses in Ibadan Metropolis, Oyo State, Nigeria. *National Horticultural Research Institute, African Journal of Basic & Applied Sciences* 1(5-6): 87-92
- Adu-Dapaah, H. K. and Oppong-Konadu, E. Y. (2002).** Tomato production in four major tomato-growing districts in Ghana. Farming practices and production constraints in Ghana. *Ghana Journal of Agricultural Science*. 35, 11-22.
- Agrios, G. N. (2005).** Plant diseases caused by nematodes. In: *Plant Pathology*. Agrios, G. N.(eds). Burlington, MA 01803, San Diego, California, USA. Elsevier Academic Press. (pp. 826-874).
- Agrios, G. N. (1997).** *Plant pathology*. New York, USA, Academic Press. (pp. 803).
- Alton N. and Sparks, Jr. (2017).** Insect Management. In: *Commercial Tomato Production Handbook*. UGA extension. (pp 31-35)
- Anastasiadis, I. A., Giannakou I. O., Athanasiadou, D. A. P. and Gowen, S. R., (2008).** The combined effect of the application of a biocontrol agent *Paecilomyces lilacinus*, with the control of root-knot nematodes. *Crop Protection*, 27, 352-361.

<http://doi.org/10.1016/j.cropro.2007.06.008>

**Aryeetey, E. (2006).** Ghana – Second largest importer of tin tomato ISSER – *Merchant Bank Development Seminar Series*. Retrieved 25th May, 2017 from:

<http://www.ghanaweb.com.GhanaHomePage/election2008/artikel.php?ID=101256>.

**Asaduzzaman, M., Alam, M. J., and Islam, M. M. (2010).** Effect of Trichoderma on seed germination and seedling parameters of chili. In *Journal of National Science Foundation*, 8(1&2):141-150

**Baker, R., Elad, Y. and Chet, I. (1984).** The controlled experiment in the scientific method with special emphasis on biological control. *Phytopathology*, 74:1019-1021.

**Baker, K. R., (1985).** The application of microplot techniques in nematological research. In Baker, K.R., Carter, C.C., Sasser, J.N. (Eds.), In: *An Advanced Treatise on Meloidogyne. Methodology, Raleigh*. North Carolina State University Graphics, Vol. 2,127–134.

**Baker, K. R. (1986).** Biological Control. An Overview. *Canadian Journal of Plant Pathology*. 8:218–221.

**Basset, M. J. (1986).** Breeding Vegetable Crops. *A.V.I. Publishing Corporation*, 2:137-157.

**Black, C. A. (1965).** Methods of soil analysis. Part 2. Chemical and microbiological properties. First edition. *American Society of Agronomy and Soil Science Society of America*. Madison, Wisconsin, USA. (pp. 914 – 925).

**Blanca, J., Cañizares, J, Cordero, L, Pascual, L., Diez, M. J, Nuez, F. (2013).** Variation revealed by SNP genotyping and morphology provides insight into the origin of the tomato. *PLoS ONE* 7 (10): e48198.

**Bray, R. H. and Kurtz, L. T. (1945).** Determination of Total Organic and Available Forms of Phosphorus in Soils. *Soil Science*. 59, 39-54.

**Bremner, J. M. and Mulvaney, C. S. (1982).** Total nitrogen. In: Page, A. L., Miller, R. H. and Keeney, D. R. (Eds.). *Methods of soil analysis Part 2. Chemical and microbiological*

*properties*. American Society of Agronomy and Soil Science Society of America, Madison Wisconsin Inc. pp. 593-624

**Bridge, J. and Page, S. I. J. (1980).** Estimation of root-knot nematode infestation levels on roots using a rating chart. *Topical Pest Management* 26, 296-298.

**Boyhan, G. E. and Kelley, W. T. (2017).** Transplant Production. In *Commercial Tomato Production Handbook*. UGA extension. (pp. 9)

**Boyhan, G. E. and Kelley, W. T. (2017).** Physiological Problems. In *Commercial Tomato Production Handbook*. UGA extension. (pp. 15-16).

**Chen, J. (2006).** The combined use of chemical and organic fertilizer and or biofertilizer for crop growth and soil fertility. *International Workshop on Sustained Management of the Soil-Rhizosphere System for Efficient Crop Production and Fertilizer Use*. Thailand. (pp. 16-20).

**Chindo, P. S., Emechebe, A. M. and Marley, P. S. (2006).** Effect of initial population density of *Pratylenchus brachyurus* on maize. *Department of Crop Protection, Faculty of Agriculture/Institute for Agricultural Research, Ahmadu Bello University, Zaria, Nigeria Archives of Phytopathology and Plant Protection*. 39, 205 – 208

**Coyne, D. L., Nicol, J. M. and Claudius-Cole, B. (2007).** *Practical plant nematology: a field and laboratory guide*. SP-IPM Secretariat, International Institute of Tropical Agriculture (IITA). Cotonou, Benin. pp. V

**Cox, S. E. (2000).** *History of the Tomato. I Say Tomato, You Say Tomato*. Retrieved from <http://www.landscapeimagery.com/articles.html>. Accessed: 10<sup>th</sup> June, 2017.

**Cutler, K. D. 1998.** *From Wolf Peach to Outer Space*.

[www.bbg.org/gardening/kitchen/tomatoes/cutler.html](http://www.bbg.org/gardening/kitchen/tomatoes/cutler.html)

**Daykin, M. E., Hussey, R. S., (1985).** Staining and histopathological techniques in nematology. In: Baker, K.R., Carter, C.C., Sasser, J.N. (Eds.). In: *An Advanced Treatise on Meloidogyne*.

*Methodology, Raleigh, North Carolina State University Graphics. Vol. II (pp. 39-48).*

**David, B. and Langston, Jr (2017).** Diseases. In *Commercial Tomato Production Handbook*. UGA extension. (pp. 28-30).

**Diao, X. (2010).** Economic importance of agriculture for sustainable development and poverty reduction: findings from a case study of Ghana global forum on agriculture. In *Policies for agricultural development, poverty reduction and food security*. OECD headquarters, Paris. (pp. 5-79).

**Elad, Y., Zimand, G., Zaqs, Y., Zuriel, S., Chet, I. (1993).** Use of *Trichoderma harzianum* in combination or alternation with fungicides to control cucumber grey mould (*Botrytis cinerea*) under commercial greenhouse conditions. *Plant Pathology*. 42, 324–332.

**FAOSTAT (2013).** Commodities by country rankings. <http://www.faostat.fao.org>. Last accessed September 29, 2016.

**FAOSTAT (2014).** Commodities by country rankings <http://www.fao.org>. Last accessed 1<sup>st</sup> June 2017.

**Fausey, N. R., and McDonald, M. B. (1985).** Emergence of Inbred and Hybrid Corn Following Flooding. *Agronomy Journal*. 77:51-56.

**Flynn Paula (2003).** Biotic vs. Abiotic - Distinguishing Disease Problems from Environmental Stresses. Department of Plant Pathology. Iowa State University Extension and Outreach. *Horticulture and Home Pest news article*. Last accessed: 12<sup>th</sup> June, 2017.

**Freeman, H. J., Eugene, J. M., Nathan, S. B., Peter, J. D., Ozores-Hampton, M., Smith, H. A., Vallad, G. E., and Webb, S. E. (2016).** *Vegetable Production Handbook for Florida, Chapter18. Tomato Production*. University of Florida. IFAS Extension. (pp. 315-217).

**Ghahfarokhy, M. R., Goltapeh, E.M., Purjam, E., Pakdaman, B.S., Modarres, S.A.M. and Varma, A. (2011).** Potential of mycorrhiza-like fungi and *Trichoderma* species in biocontrol

of Take-all Disease of wheat under greenhouse condition. *Journal of Agricultural Technology* 7(1): 185-195.

**Harman, G. E, Howell, C. R, Viterbo, A., Chet, I., Lorito, M. (2004).** *Trichoderma* spp. opportunistic avirulent plant symbiosis. *Nat Rev Microbiology* 2: 43-56.

**Hassan, M. A., Chindo, P. S., Marley, P. S., Alegbejo, M. D., (2010).** Management of root-knot nematodes (*Meloidogyne spp.*) on tomato (*Lycopersicon lycopersicum*) using organic wastes in Zaria, Nigeria. *Plant Protect. Sci.* 46 (1), pp. 34-38.

**Helmke, P. A. and Sparks, D. L. (1996).** Methods of Soil Analysis. Part 3. Chemical Methods - Soil Science Society of America. Book series no. 5. Madison, Wisconsin, USA.

**Hjeljord, L. G., Stensvand, A., Tronsmo, A. (2000).** Effect of temperature and nutrient stress on the capacity of commercial *Trichoderma* product to control *Botrytis cinerea* and *Mucor piriformis* in greenhouse strawberries. *Biological Control*, 19: 149–160.

**Hooper, D. J., Hallmann, J. and Subbotin, S. (2005).** Methods for extraction, processing and detection of plant and soil nematodes. In: Luc, M., Sikora, R.A. and Bridge (Eds.). *Plant Parasitic Nematodes in Subtropical and Tropical Agriculture, 2nd Edition*. Wallingford, UK: CAB International. (pp. 53-86)

**Howell, C. R. (2002).** Cotton seedling pre-emergence damping-off incited by *Rhizopus oryzae* and *Pythium* spp. and its biological control with *Trichoderma* spp. *Phytopathology* 92: 177–180.

**Kelley, W. T. and Boyhan, G. (2017).** Culture and Varieties. In *Commercial Tomato Production Handbook* (pp. 4). UGA extension.

**Kouadio, K. Y., Aman, A., Ochou, A. D., Ali, K. E. and Assamoi, P.A. (2011),** Rainfall Variability Patterns in West Africa: Case of Cote d'Ivoire and Ghana. *Journal of Environmental Science and Engineering*, 5 (2011) 1229-1238.

- Lee, S. K., Sohn, H. B., Kim, G. G. and Chung, Y. R. (2006).** Enhancement of biological control of *Botrytis cinerea* on Cucumber by foliar sprays and bed potting mixes of *Trichoderma harzianum* YC459 and its application on Tomato in the greenhouse. *Plant Pathology Journal*. 22 (3): 283-288.
- Lester, D. (2010).** Understanding and using *Trichoderma* fungi. In *Maximum yield Australia*. pp. 48-52.
- Luc, M., Sikora, R. A. and Bridge, J. (1990).** Reflection on nematology in subtropical and tropical agriculture, In: Luc, M., Sikora, R. A. and Bridge, J. (Eds.) *Plant parasitic nematodes in subtropical and tropical agriculture*. CAB International Wallingford, U.K. (pp. 319-392)
- Lukyanenko, A. N. (1991).** Disease resistance in tomato. In Genetic improvement of tomato (ed. G., Kalloo). *Monographs on Theoretical and Applied Genetics*. Springer Verlag, Berlin, Heidelberg. pp. 24, 99 – 119.
- Mai, W. F. (1977).** Worldwide distribution of potato cyst nematodes and their importance in crop production. *Journal of Nematology*. 9 pp. 30-34.
- McCormack, J. H. (2004).** Tomato seed production. *An organic seed production manual for seed growers in the Mid-Atlantic and Southern U.S.* (Version 2.6: pp 3)
- McSorley, R., Gallaher, R. N., (1995).** Effect of yard waste compost on plant-parasitic nematode densities in vegetable crops. *Supplementary to Journal of Nematology*. 27, pp. 545–549.
- Mokwunye, A. U., De Jager, A. and Smaling, E. M. A. (1996).** *Restoring and maintaining the productivity of West Africa Soils: Key to sustainable development*. International Fertilizer Development Center (IFDC), Muscle Shoals, Alabama. pp. 94.
- Muller, R., and Gooch, P.S., 1982.** Organic amendments in nematode control. An examination of the literature. *Nematropica* 12, 319–326.

- Nadary, S. N., Al-Hazmi, A. S., Dawabah, A. A. M. and Al-Yahya, F. A. (2006).** Relationship between the initial inoculum density of *Meloidogyne incognita*, infection and reproduction on green beans. 9th Arab congress of plant protection Damascus, Syria NB- E. 112.
- Nelson, D. W., and Sommers, L. W. (1982).** Total carbon, organic carbon and organic matter. In: Page, A. L., Miller, R. H and Keeney, D. R. (eds.). *Methods of soil analysis. Part 2. Second edition. Chemical and microbiological properties.* American Society of Agronomy and Soil Science Society of America. Madison, Wisconsin USA. (pp. 301-312).
- Norman, J. C. (1992).** *Tropical vegetable crops.* Arthur, H. Stockwell Ltd. Devon, pp. 47
- Obeng-Ofori, D., Danquah, Y. E and Ofori Anim, J. (2007).** *Vegetable and spice crop production in West Africa.* Smart line publishers, pp.53-59.
- Obeng, H. (2000).** Soil Classification in Ghana. Centre for policy Analysis (CEPA). Selected Economic Issues No. 3. pp 23
- O’Callaghan, M. (2016).** Microbial inoculation of seed for improved crop performance: issues and opportunities. *Applied Microbiology and Biotechnology*, 100, 5729–5746.
- Ojha, S., Chatterjee, N. C. (2011).** Mycoparasitism of *Trichoderma* spp. in biocontrol of *Fusarium wilt* of tomato. *Archives of Phytopathology and Plant Protection*, 44(8): 771-782,
- Osae Richard (2014).** Assessment of locally produced waxing materials on the shelf life and fruit quality of two tomato varieties (*Solanum lycopersicum*). pp. 4
- Osei, K., Moss, R., Nafeo, A., Addico, R., Agyemang, A., Danso, Y. and Asante, J.S., (2011).** Management of plant parasitic nematodes with antagonistic plants in the forest-savanna transitional zone of Ghana. *Journal of Applied Biosciences*, 37, 2491-2495.
- Parker, S.K., Nutter Jr., F.W. and Gleason, M.L. (2001).** Directional Spread of *Septoria* Leaf Spot in Tomato Rows. *Plant Disease*, 81. 272-276.
- Panthee, D. R. and Chen, F. (2009).** Genomics of Fungal Resistance in Tomato. *Current Genomics*. 11, 30-39.

- Pattison, T. (2007).** *Tomato root knot nematode: Biology and control*. Department of Primary Industry and Fisheries. Queensland, Australia.
- Peralta, I. E. & Spooner, D. M. (2001).** GBSSI gene phylogeny of wild tomatoes (*Solanum* L. section *Lycopersicon* [Mill], Wettst. Subsection *Lycopersicon*). *American Journal Bot* 88:1888-1902.
- Robert, T. (2006).** *Partial Budgeting: A Tool to Analyze Farm Business Changes*. University Extension. Iowa State University. FM 18877 Revised May 2006. File C1-50.
- Robinson, J. Z. E. and Kolavalli, S. L. (2010).** *The Case of Tomato in Ghana – Marketing*. (International Food Policy Research Institute - IFPRI).  
<http://www.ifpri.org/publication/case-tomato-ghana-marketing>.
- Rodri'guez-Ka' bana, R., (1986).** Organic and inorganic nitrogen amendments to soil as nematode suppressants. *Journal of Nematology*. 18 (2), 129–135.
- Reichelderfer, K. H. (1979).** Economic feasibility of a biological control technology: using a parasitic wasp, *Pediobius foveolatus*, to manage Mexican bean beetle on soybean. U. S. Department of Agriculture. ESCS, Agricultural Economics. Rept. No. 430.
- SEND Ghana (2008).** *Bacterial canker of Tomato. Plant pathology fact sheet vegetable production guide for commercial growers*, ID- 36 (University of Kentucky). Retrieved on 5th January 2013 from <http://www.ca.uky.edu/agc/pubs/id/id36/id36.htm>.
- Sharon, E., Bar-Eyal, M., Chet, I., Herrera-Estrella, A., Kleifeld, O., and Spiegel, Y. 2001.** Biological control of the root-knot nematode *Meloidogyne javanica* by *Trichoderma harzianum*. *Phytopathology* 91, 687-693.
- Starkeyres (2014).** **Tomato production guideline**. Retrieved from <http://www.Starkeyres.co.za>.

- Siddiqi, M. R. (2000).** *Tylenchida parasites of plants and insect (2nd edition)*. Wallingford: CAB International.
- Sivan A., Elad Y., Chet I. (1984).** Biological control effects of a new isolate of *Trichoderma harzianum* on *Pythium aphanidermatum*. *Phytopathology*, 74, 498–501.
- Sivasithamparam, K. and Ghisalberti, E. L. (1998).** In *Trichoderma and Gliocladium* Vol. 1 (eds) Kubicek, C. P. and Harman, G. E. Taylor and Francis, London. (pp. 139–191).
- Smith, M. A. L. and Spomer, L.A. (1994).** Vessels, gels, liquid media, and support systems. In: *Automation and Environmental Control in Plant Tissue Culture*. Aitken-Christie *et al.* (eds.), *Kluwer Academic Publishers*. 371-404.
- South African Department of Agriculture Forestry and Fisheries (2017).** Production guidelines Production guidelines for Tomato. *Published by the Directorate Plant Production*. Last visited: 19<sup>th</sup> June 2017.
- Srinivasan, R. (Ed.) (2010).** Safer tomato production methods: A field guide for soil fertility and pest management. AVRDC – *The World Vegetable Centre, Shanhua, Taiwan*. *AVDRC 10 (740)*: 2-97.
- Stirling, G.R., (1991).** *Biological Control of Plant Parasitic Nematodes: Progress, Problems and Prospects*. (pp. 282). CAB International, Wallingford, UK.
- Slow food Upstate. Tomato.** Retrieved from [Renatovicario.com/pdf/tomato.pdf](http://Renatovicario.com/pdf/tomato.pdf)
- USDA (2016).** *National Nutrient Database for Standard Reference Release 28*. (revised May, 2016). Retrieved from <https://ndb.nal.usda.gov>. Last accessed 9<sup>th</sup> June, 2017.
- Watt, B.A. (2000).** *Septoria Leaf Spot of Tomato*. *University of Maine-Fact Sheet*. Retrieved from <http://www.pmo.umext.maine.edu/factsht/septum.htm>
- Wener, J. N. (2000).** <http://www.agrisupportonline.com>.

**Windham, G. L., Windham, M. T., and Williams, W. P. (1989).** Effects of *Trichoderma* spp. on maize growth and *Meloidogyne arenaria* reproduction. *Plant Disease*. 73: 493-494.

**Yeboah A., (2011).** *A survey on postharvest handling, preservation and processing methods of tomato (Solanum lycopersicum) in the Dormaa and Tano south districts of the Brong Ahafo region of Ghana.* A thesis submitted to the school of graduate studies, Kwame Nkrumah University of Science and Technology, Kumasi. (pp. 4-29).



## APPENDICES

## APPENDIX 1

Table 19: Nutrition value per 100g of tomato.

NUTRIENT	NUTRIENT VALUE	NLEA SERVING 148 (g)
<b>Proximates</b>		
Energy	18 Kcal	27
Water	94.52 g	139.89
Carbohydrates	3.9 g	5.76
Protein	0.9 g	1.30
Fibre, Total dietary	1.2 g	1.8
Sugars, total	2.63 g	
<b>Lipid</b>		
Cholesterol	0.00 g	0.00g
Fatty acids, total saturated	0.028 g	0.041
Fatty acids, total monounsaturated	0.031 g	0.046
Fatty acids, total polyunsaturated	0.083 g	0.123
<b>Vitamins</b>		
Folate, total	15 µg	22
Niacin	0.594 mg	0.879
Pyridoxine (Vitamin B-6)	0.080 mg	0.118
Thiamin	0.037 mg	0.055
Vitamin A	833 IU	1233
Vitamin A, RAE	42 µg	62
Vitamin C (Total ascorbic acid)	13.7 mg	20.3
Vitamine D	0 IU	0
Vitamin E (Alpha-tocopherol)	0.54 mg	0.08
Vitamin K (Phylloquinone)	7.9 µg	11.7
Riboflavin	0.019 mg	0.028
Vitamine B-12 (cobalamin)	0.0 µg	0.0
Carotene, beta	449 µg	665
Carotene, alpha	101 µg	149
Lutein + zeaxanthin	123 µg	182
Lycopene	2573 µg	3808
<b>Minerals</b>		
Calcium (Ca)	10 mg	15
Iron (Fe)	0.27 mg	0.40
Magnesium (Mg)	11 mg	16
Potassium (K)	237 mg	351
Sodium, (Na)	5 mg	7

<b>Phosphorus</b>	24 mg	36
<b>Zinc</b>	0.17 mg	0.25
<b>Amino acids</b>		
<b>Tryptophan</b>	0.006 g	0.009
<b>Threonine</b>	0.027 g	0.040
<b>Isoleucine</b>	0.018 g	0.027
<b>Leucine</b>	0.025 g	0.037
<b>Lysine</b>	0.027 g	0.040
<b>Methionine</b>	0.006 g	0.009
<b>Cystine</b>	0.009 g	0.013
<b>Phenylalanine</b>	0.027 g	0.040
<b>Tyrosine</b>	0.014 g	0.021
<b>Valine</b>	0.018 g	0.027
<b>Arginine</b>	0.021 g	0.031
<b>Histidine</b>	0.014 g	0.021
<b>Alanine</b>	0.027 g	0.040
<b>Aspartic acid</b>	0.135 g	0.200
<b>Glutamic acid</b>	0.431 g	0.638
<b>Glycine</b>	0.019 g	0.028
<b>Proline</b>	0.015 g	0.022
<b>Serine</b>	0.026 g	0.038

(Source: USDA National Nutrient database 2016)

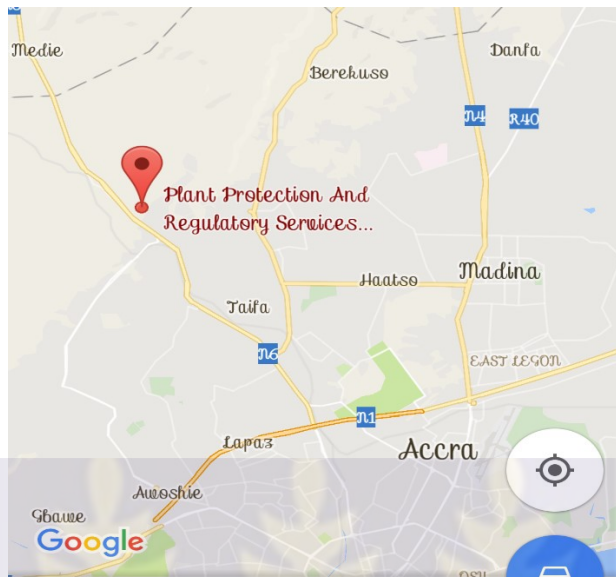
## APPENDIX 2

**Table 20:sample partial budget with components.**

<p>a. Added income because of the change</p> <p>_____ GH¢ _____</p> <p>_____ GH¢ _____</p> <p>b. Decreased costs because of the change</p> <p>_____ GH¢ _____</p> <p>_____ GH¢ _____</p> <p><b>Subtotal</b> <b>GH¢</b> _____</p> <p><i>The net change</i> = (a+b) – (c+d)</p>	<p>c. Added costs because of the change</p> <p>_____ GH¢ _____</p> <p>_____ GH¢ _____</p> <p>d. Decreased income because of the change</p> <p>_____ GH¢ _____</p> <p>_____ GH¢ _____</p> <p><b>Subtotal</b> <b>GH¢</b> _____</p>
---	--

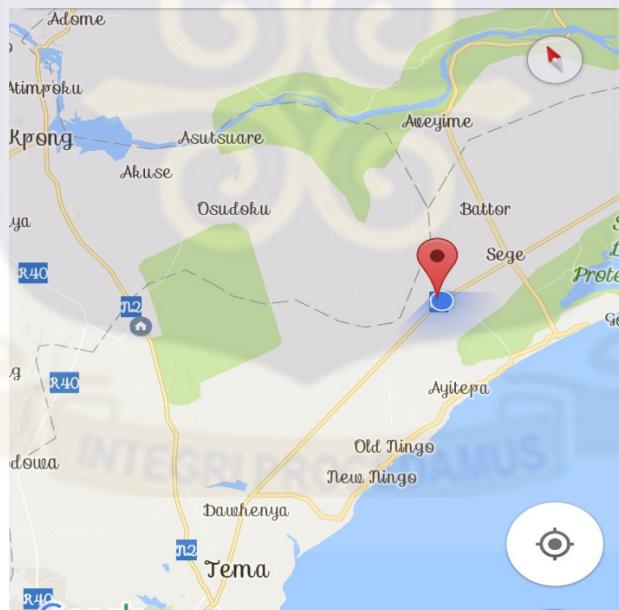
Source: Robert (2006).

**APPENDIX 3.1: Study Site for Laboratory analysis**



*Location source: Google maps*  
**Figure 4: Location of Pokuase**

**APPENDIX 3.2: Study Site for Field work**

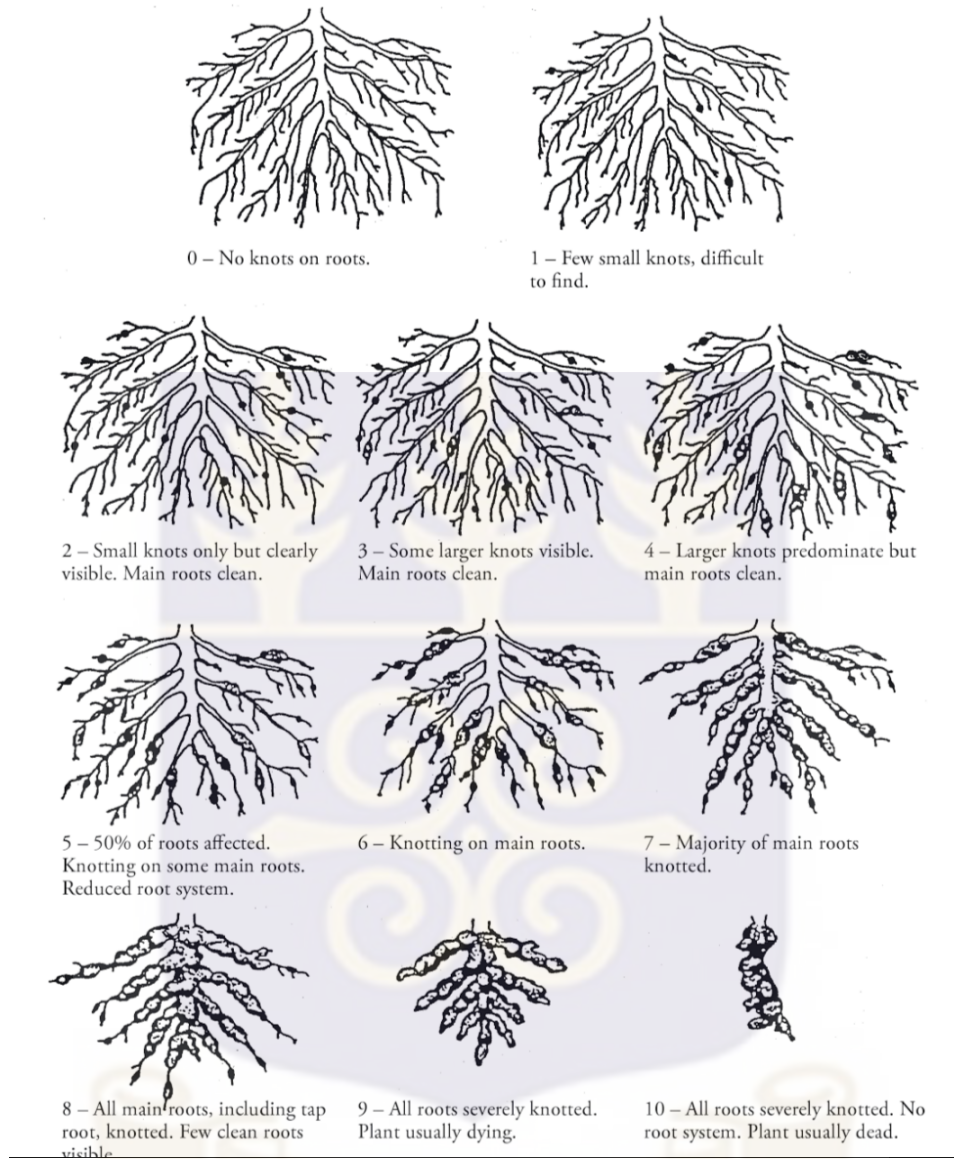


*Location source: Google maps*  
**Figure 5: Location of Dawu**

**APPENDIX 4: Root Gall Scoring (Bridge and Page, 1980)**

**Diagrammatic root-knot scoring chart**

Courtesy of John Bridge and Sam Page (1980).



**Figure 6: Gall Scoring chart**

*Image source: (Coyne et al., 2007)*

**Table 21: Gall score description**

Score	Galling observation
0	No knots on roots
1	Few small knots difficult to find
2	Small knots only but clearly visible; main roots clean
3	Some larger knots visible, but main roots clean
4	Larger knots predominate but main roots clean
5	50 % of roots knotted; knotting on parts of main root system
6	Knotting on some of main roots
7	Majority of main roots knotted
8	All main roots knotted; few clean roots visible
9	All roots severely knotted; plant usually dying
10	All roots severely knotted; no root

**APPENDIX 5: pH Meter Scale**

Acidity pH – H<sub>2</sub>O

< 4.0	extremely acid
4.0 – 5.0	strongly acid
5.0 – 5.5	acid
5.5 – 6.0	slightly acid
6.0 – 7.5	neutral
7.5 – 8.0	slightly alkaline
8.0 – 9.0	alkaline
> 9.0	strongly alkaline

Inference drawn from pHw 1:1 soil: water of mineral soils.

1. pH < 3.5: associated with significant pH drop after wetting – drying cycles: acid sulphates present.
2. Ph < 4.5: a significant amount of exchangeable hydrogen is probably present in addition to exchangeable Al.
3. pH 4.5 to 5.8 in mineral soils: sufficient exchangeable Al is present to affect plant growth significantly, and % base saturation is low.
4. pH 5.8 to 6.5: acidity is present (apparently from hydroxyl- Al and organic functional groups) ordinary hydroxonium, in amounts sufficient to affect acidity – sensitive crops such as Alfalfa.
5. pH 6.5 to 8: the soil is essentially base saturated and free calcium carbonate is present in the system.
6. pH 8 to 8.5: the soil is fully base saturated and free calcium carbonate is present in the system.
7. pH 8.5 to 10: the soil contains large amounts of soluble salts and conductivity is high.
8. pH > 10: the soil is highly Na saturated – “alkali” soil.

**APPENDIX 6:** Chart for organic carbon rating (% C)

< 0.3	Very low
0.3 – 1.0	low
1.0 – 2.0	medium
2.0 – 5.0	high
> 5.0	very high

**APPENDIX 7: ANALYSIS OF VARIANCE TABLE FOR PARAMETERS**

## LABORATORY ANALYSIS

(Concentration Experiment)

## Analysis of Variance for % Germination

Variate: %\_Germination

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
TRT_Lab_Conc	5	59.33	11.87	0.94	0.481
Residual	18	228.00	12.67		
Total	23	287.33			

## Analysis of Variance for seedling Shoot Length

Variate: Seedlg\_shoot\_length\_conc

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
TRT_Lab_Conc	5	139.00	27.80	0.99	0.452
Residual	18	505.92	28.11		
Total	23	644.92			

## Analysis of Variance for Seedling Root Length

Variate: Seedlg\_root\_lgth\_conc

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
TRT_Lab_Conc	5	1.108	0.222	0.09	0.993
Residual	18	45.341	2.519		
Total	23	46.449			

## Analysis of Variance for Seedling Vigour Index

Variate: Seedling\_vigour\_conc

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
TRT_Lab_Conc	5	10503.	2101.	0.08	0.995
Residual	18	473411.	26301.		
Total	23	483913.			

(Duration Experiment)

Analysis of Variance for % Germination

Variate: %\_Germination

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
TRT_Lab_Dura	8	139.56	17.44	0.49	0.855
Residual	27	968.00	35.85		
Total	35	1107.56			

Analysis of Variance for Seedling Shoot Length

Variate: Seedlg\_shoot\_lgth\_dura

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
TRT_Lab_Dura	8	153.88	19.23	0.99	0.467
Residual	27	525.53	19.46		
Total	35	679.41			

Analysis of Variance for Seedling Root Length

Variate: Seedling\_root\_length

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
TRT_Lab_Dura	8	9.6574	1.2072	2.73	0.024
Residual	27	11.9177	0.4414		
Total	35	21.5751			

Analysis of Variance for Seedling Vigour

Variate: Seedlg\_vigour\_dura

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
TRT_Lab_Dura	8	173983.	21748.	1.61	0.168
Residual	27	364112.	13486.		
Total	35	538095.			

(Interaction Experiment)

Analysis of Variance for % Germination

Variate: %\_Germination

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Concentration_inter	2	144.0	72.0	0.62	0.550
Duration	1	2.7	2.7	0.02	0.881
Concentration_inter.Duration	2	5.3	2.7	0.02	0.977
Residual	18	2096.0	116.4		
Total	23	2248.0			

Analysis of Variance for Seedling Shoot Length

Variate: Seedling\_shoot\_length

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Concentration_inter	2	0.2837	0.1419	0.36	0.706
Duration	1	1.0168	1.0168	2.55	0.128
Concentration_inter.Duration	2	0.9505	0.4753	1.19	0.327
Residual	18	7.1881	0.3993		
Total	23	9.4392			

Analysis of Variance for Seedling Root Length

Variate: Seedling\_root\_length

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Concentration_inter	2	0.6744	0.3372	1.17	0.332
Duration	1	0.0504	0.0504	0.18	0.681
Concentration_inter.Duration	2	0.8204	0.4102	1.43	0.266
Residual	18	5.1815	0.2879		
Total	23	6.7268			

Analysis of variance

Variate: Seedlg\_vigour\_inter

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Concentration_inter	2	16420.	8210.	0.76	0.481
Duration	1	4081.	4081.	0.38	0.546
Concentration_inter.Duration	2	15138.	7569.	0.70	0.509
Residual	18	194043.	10780.		
Total	23	229681.			

FIELD EXPERIMENT

Analysis of Variance for Seedling Emergence on Field

Variate: Emergence

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_field_Emer stratum	3	1281.91	427.30	18.25	
REP_field_Emer.*Units* stratum					
TRT_field_Emerg	2	426.02	213.01	9.10	0.015
Residual	6	140.47	23.41		
Total	11	1848.40			

Analysis of Variance for Number of Leaves

Variate: No\_of\_Leaves\_4wks

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	0.48691	0.16230	2.39	
REP_Field.*Units* stratum					
TRT_Field	5	4.05807	0.81161	11.96	<.001
Residual	15	1.01821	0.06788		
Total	23	5.56320			

Analysis of Variance for Number of Leaves

Variate: No\_of\_leaves\_8wks

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	0.9813	0.3271	0.34	
REP_Field.*Units* stratum					
TRT_Field	5	22.5095	4.5019	4.72	0.009
Residual	15	14.2993	0.9533		
Total	23	37.7901			

Analysis of Variance for Leaf Length 1

Variate: Leaf\_Length\_1

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	0.78008	0.26003	2.83	
REP_Field.*Units* stratum					
TRT_Field	5	4.48982	0.89796	9.78	<.001
Residual	15	1.37730	0.09182		
Total	23	6.64720			

Analysis of Variance for Leaf Length 2

Variate: Leaf\_Length\_2

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	3.828	1.276	0.68	
REP_Field.*Units* stratum					
TRT_Field	5	21.738	4.348	2.32	0.095
Residual	15	28.125	1.875		
Total	23	53.691			

Analysis of Variance for Plant Height 1

Variate: Plant\_Height\_1

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	15.516	5.172	3.19	
REP_Field.*Units* stratum					
TRT_Field	5	25.590	5.118	3.15	0.038
Residual	15	24.357	1.624		
Total	23	65.464			

Analysis of Variance for Plant Height 2

Variate: Plant\_Height\_2

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	32.75	10.92	0.80	
REP_Field.*Units* stratum					
TRT_Field	5	498.94	99.79	7.32	0.001
Residual	15	204.58	13.64		
Total	23	736.28			

Analysis of Variance of Stem Girth 1

Variate: Plant\_Girth\_1

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	0.3382	0.1127	1.05	
REP_Field.*Units* stratum					
TRT_Field	5	0.7313	0.1463	1.36	0.292
Residual	15	1.6092	0.1073		
Total	23	2.6787			

Analysis of Variance for Stem Girth 2

Variate: Plant\_Girth\_2

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	0.00438	0.00146	0.02	
REP_Field.*Units* stratum					
TRT_Field	5	2.86177	0.57235	7.62	<.001
Residual	15	1.12655	0.07510		
Total	23	3.99270			

Analysis of Variance of Shoot Dry Weight 1

Variate: Shoot\_dry\_weight\_1

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	181.34	60.45	0.89	
REP_Field.*Units* stratum					
TRT_Field	5	1197.12	239.42	3.52	0.027
Residual	15	1021.43	68.10		
Total	23	2399.88			

Analysis of Variance for Shoot Dry Weight 2

Variate: Shoot\_dry\_weight\_2

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	4205.6	1401.9	3.77	
REP_Field.*Units* stratum					
TRT_Field	5	5500.8	1100.2	2.96	0.047
Residual	15	5578.0	371.9		
Total	23	15284.4			

Analysis of Variance for Root Dry Weight 1

Variate: Root\_dry\_weight\_1

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	0.3263	0.1088	0.29	
REP_Field.*Units* stratum					
TRT_Field	5	3.6314	0.7263	1.92	0.150
Residual	15	5.6619	0.3775		
Total	23	9.6197			

Analysis of Variance for Root Dry Weight 2

Variate: Root\_dry\_weight\_2

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	354.69	118.23	2.18	
REP_Field.*Units* stratum					
TRT_Field	5	126.96	25.39	0.47	0.794
Residual	15	812.92	54.19		
Total	23	1294.57			

Analysis of Variance for Shoot Fresh Weight1

Variate: Shoot\_fresh\_weight\_1

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	417.57	139.19	2.11	
REP_Field.*Units* stratum					
TRT_Field	5	2059.01	411.80	6.25	0.003
Residual	15	988.90	65.93		
Total	23	3465.49			

Analysis of Variance for Shoot Fresh Weight 2

Variate: Shoot\_fresh\_weight\_2

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	4778.	1593.	0.77	
REP_Field.*Units* stratum					
TRT_Field	5	48450.	9690.	4.71	0.009
Residual	15	30848.	2057.		
Total	23	84077.			

Analysis of Variance for Root Fresh Weight 1

Variate: Root\_fresh\_weight\_1

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	18.340	6.113	0.72	
REP_Field.*Units* stratum					
TRT_Field	5	106.770	21.354	2.50	0.077
Residual	15	128.054	8.537		
Total	23	253.164			

Analysis of Variance for Fresh Weight 2

Variate: Root\_fresh\_weight\_2

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	1780.3	593.4	1.71	
REP_Field.*Units* stratum					
TRT_Field	5	5842.2	1168.4	3.37	0.031
Residual	15	5199.8	346.7		
Total	23	12822.3			

Analysis of Variance of Gall Score 1

Variate: Transformed\_gall1

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	1.0852	0.3617	1.36	
REP_Field.*Units* stratum					
TRT_Field	5	4.3716	0.8743	3.28	0.034
Residual	15	4.0007	0.2667		
Total	23	9.4575			

Analysis of Variance for Gall Score 2

Variate: Transformed\_gall2

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	9.400	3.133	3.13	
REP_Field.*Units* stratum					
TRT_Field	5	9.419	1.884	1.88	0.158
Residual	15	15.021	1.001		
Total	23	33.840			

Analysis of Variance for Root Length 1

Variate: Root\_length\_1

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	51.61	17.20	0.97	
REP_Field.*Units* stratum					
TRT_Field	5	39.46	7.89	0.45	0.809
Residual	15	264.80	17.65		
Total	23	355.87			

Analysis of Variance for Root Length 2

Variate: Root\_length\_2

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	38.10	12.70	0.19	
REP_Field.*Units* stratum					
TRT_Field	5	1804.26	360.85	5.27	0.005
Residual	15	1026.16	68.41		
Total	23	2868.51			

Analysis of Variance for Egg Mass

Variate: Egg\_mass\_1

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	6847.	2282.	0.42	
REP_Field.*Units* stratum					
TRT_Field	5	105310.	21062.	3.84	0.019
Residual	15	82266.	5484.		
Total	23	194423.			

Analysis of Variance for Transformed Egg Mass 1

Variate: Transformed\_egg\_mass1

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	8.92	2.97	0.27	
REP_Field.*Units* stratum					
TRT_Field	5	239.90	47.98	4.36	0.012
Residual	15	164.97	11.00		
Total	23	413.79			

Analysis of Variance for Egg Mass 2

Variate: Egg\_mass\_2

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	54192.	18064.	0.91	
REP_Field.*Units* stratum					
TRT_Field	5	288357.	57671.	2.90	0.050
Residual	15	298679.	19912.		
Total	23	641229.			

Analysis of Variance for Transformed Egg Mass 2

Variate: Transformed\_Egg\_Mass2

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	52.24	17.41	0.80	
REP_Field.*Units* stratum					
TRT_Field	5	394.20	78.84	3.62	0.024
Residual	15	326.45	21.76		
Total	23	772.89			

Analysis of Variance for Nematode Count 1

Variate: Nematode\_count\_1

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	10730.	3577.	0.15	
REP_Field.*Units* stratum					
TRT_Field	5	106696.	21339.	0.89	0.514
Residual	15	360964.	24064.		
Total	23	478390.			

Analysis of Variance for Transformed Nematode Count 1

Variate: Transformed\_nematode\_count1

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	10.47	3.49	0.16	
REP_Field.*Units* stratum					
TRT_Field	5	109.54	21.91	0.98	0.463
Residual	15	336.48	22.43		
Total	23	456.50			

Analysis of Variance for Nematode Count 2

Variate: Nematode\_count\_2

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	39468.	13156.	1.38	
REP_Field.*Units* stratum					
TRT_Field	5	206766.	41353.	4.35	0.012
Residual	15	142634.	9509.		
Total	23	388868.			

Analysis of Variance for Transformed Nematode Count 2

Variate: Transformed\_nematode\_count2

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	49.34	16.45	1.49	
REP_Field.*Units* stratum					
TRT_Field	5	205.82	41.16	3.72	0.022
Residual	15	166.05	11.07		
Total	23	421.21			

Analysis of Variance for Nematode Count 3

Variate: Nematode\_count\_3

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	18532.	6177.	0.51	
REP_Field.*Units* stratum					
TRT_Field	5	431020.	86204.	7.14	0.001
Residual	15	181063.	12071.		
Total	23	630616.			

Analysis of Variance for Transformed Nematode Count 3

Variate: Transformed\_nematode\_count3

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	16.95	5.65	0.44	
REP_Field.*Units* stratum					
TRT_Field	5	428.40	85.68	6.60	0.002
Residual	15	194.72	12.98		
Total	23	640.07			

Analysis of Variance for Weight of Tomato Fruits

Variate: Average\_weight\_of\_tomato

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	1014.	338.	0.21	
REP_Field.*Units* stratum					
TRT_Field	5	44179.	8836.	5.54	0.004
Residual	15	23944.	1596.		
Total	23	69138.			

Analysis of Variance of Yield parameter

Variate: Yield\_t/ha

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
REP_Field stratum	3	17.15	5.72	0.21	
REP_Field.*Units* stratum					
TRT_Field	5	746.68	149.34	5.53	0.004
Residual	15	405.14	27.01		
Total	23	1168.97			

- END -

