Addis Ababa University
School of Graduate studies

Biology, Population Dynamics and Management options of Tomato Leaf Miner, *Tuta absoluta* (Meyrick) (Lepidoptera: Gelechiidae) in West Shewa, Central Ethiopia

By

Tadele Shiberu

A thesis Submitted to the Department of Zoological Sciences,
Addis Ababa University

Presented in partial fulfillment of the requirements for the degree of Doctor of Philosophy in Biology, Insect Sciences

June, 2018

Addis Ababa University
Addis Ababa, Ethiopia

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Declaration

This is to declare that this dissertation is submitted to the School of Graduate Studies of Addis Ababa University for the degree of Doctor of Philosophy (Ph.D.) in Biology, Insect sciences. I would like to corroborate that it is my own work and all other works used in the dissertation are well acknowledged.

Name: Tadele Shiberu

Signature:__________ Date:____________

This Ph.D. Dissertation is submitted for examination with my approval as an advisor.

Name: Emana Getu Degaga (Professor)

Signature:___________ Date ___________

Addis Ababa University

School of Graduate Studies

This is to certify that the thesis prepared by Tadele Shiberu, entitled: **Biology, Population Dynamics and Management options of Tomato Leaf Miner, *Tuta absoluta* (Meyrick) (Lepidoptera: Gelechiidae) in West Shewa, Central Ethiopia** and submitted in fulfillment of the requirements for the Degree of Doctor of Philosophy in Biology (Insect Science) complies with the regulations of the University and meets the accepted standards with respect to originality and quality.

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Chair of Department Zoological Sciences

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DEDICATION

These research efforts are dedicated to all those Oromo people who scarified on Irecha Holiday at Bishoftu Hora-alsadi in 2016

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LIST OF ABBREVIATIONS

A_m	Apparent Mortality
AVRD	The World Vegetable Center
BCR	Benefit cost ratio
CABI	Centre for Agriculture and Biosciences International
CFIA	Canadian Food Inspection Agency
CM	Corrected Mortality
CSA	Central statistical Authority
CV	Coefficient Variation
EARO	Ethiopian Agricultural Research Organization
EC	Emulsifable Concentration
EIA	Ethiopian Investment Agency
EIL	Economic Injury Level
ETL	Economic Threshold Level
EPPO	European Plant Protection Organization
FAO	Food and Agricultural Organization
FERA	Food and Environments Research Agency
I_m	Indispensable Mortality
IPM	Integrated Pest Management
IRAC	Insecticide Resistance Action Committee
LSD	Least Significant Difference
MARC	Melkasa Agricultural Research Center
MoA	Ministry of Agriculture
MSE	Mean Standard Error
MSR	Mortality Survivor Ratio
NAPPO	North American Plant Protection Organization
PPRC	Plant Protection Research Center
RCBD	Randomized Complete Block Design
RH	Relative Humidity
SAS	Statistical Analysis Software

SC	Soluble Concentration
SE	Standard Error
SL	Soluble Liquid
S_x	Survival Fraction
USA	United State of America
USDA	United State Development Agency

Basic Studies and Management Options of Tomato Leaf Miner, *Tuta absoluta* (Meyrick) (Lepidoptera: Gelechiidae) in West Shewa, Central Ethiopia

Tadele Shiberu

ABSTRACT

Tomato leaf miner, *Tuta absoluta* (Meyrick) (Lepidoptera: Gelechiidae) is an important pest infesting Solanaceous plants all over the world. It became a problem on tomato in Ethiopia since 2012. As the pest is new to the country basic studies are paramount importances for designing sound control measure. Such studies includes seasonal abundance and distribution, biology and yield losses, which lay fertile ground for developing sound integrated pest management against the pest. Hence, these studies were intended to monitor the population dynamics, biology, Economic threshold level and management of *T. absoluta* on tomato under laboratory, glasshouse and open field conditions in West Shewa, Central Ethiopia. Different stages of *T. absoluta* were investigated in the glasshouse and open fields during 2015 to 2017 for six and four plantation periods, respectively. The highest populations of *T. absoluta* per plant were recorded in August 2016 under glasshouse conditions whereas under open field conditions, the highest populations of *T. absoluta* per plant were recorded in March and April in 2015-2016. The field studies showed that *T. absoluta* population increased proportionality with growth periods. Accordingly, the period of peak activities of *T. absoluta* was at vegetative, flowering and early fruit setting growth stages of tomato. There were different records on the biology of *T. absoluta* in the world mainly due to variations in temperature and relative humidity. In Ethiopia, the night temperature is very low and the day temperature is very high. Due to this reason, it is expected that the biology of *T. absoluta* is different from what was so far recorded elsewhere. Accordingly, the biology of *T. absoluta* was studied at different temperatures and relative humidities under laboratory and glasshouse conditions for two consecutive seasons. In these studies, developmental stage, adult longevity, fecundity and ovipositon periods were recorded. *T. absoluta* female laid about 60.56% of her eggs on the upper side surface of tomato leaves, while the lowest (0.85%) were on tomato stem. The highest number of eggs, 233.75±14.42 was laid at the temperature of 20.5±2ºC and 55±5% of R.H., whereas the lowest number of eggs, 177.5±9.26 was laid at the temperature of 32.0±2ºC and 40±5% R.H. The life expectancy of adult *T. absoluta* was high at low temperature and low at high temperature. As temperature goes above 20.5°C developmental time of the moth decreased from 9.0±0.3 to 6.8±0.27 days for male and from 18.4±1.45 to 15.2±1.4 days for female *T. absoluta*. Based on the leaf and fruit-infestation data, tomato varieties such as Koshoro, Roman-VF, Galila and Local were found to be susceptible to *T. absoluta*, while potato varieties such as Jalane, Menagesha, Tolcha and Local found to be tolerant to *T. absoluta*. Pepper varieties such as M. Awaze, M. Fana, M. Zala and Local showed high level of resistance to *T. absoluta*. The economic threshold level of *T. absoluta* larvae on tomato plants were conducted from 2015 to 2017 under glasshouse and open field conditions. The economic threshold level of *T. absoluta* was determined under glasshouse and open field conditions. The glasshouse findings showed that control measures should be started at 2.25 larvae per plant while under open field conditions at 2.87 larvae per plant. The yield loss due to *T. absoluta* was found to be 87.50% - 100% and 60.08% - 82.31% under glasshouse and open field conditions, respectively. The studies were conducted to evaluate bio-pesticides against *T. absoluta* under laboratory and glasshouse conditions during 2015-2016. After screening of effective bio-pesticides, further studies were conducted under field conditions during 2016-2017 in three different locations of West Shewa. The extract of *Azadirachta indica*, *Allium sativum*, and *Cymbopogon citrates* were revealed that significant larval mortalities. Similarly, *Beauveria bassiana* and *Metarrhizium anisopliae* at 2.5 x 10^9 concentration were gave good results against *T. absoluta* larvae. The population number of *T. absoluta* was caught on sticky trap colors under glasshouse coditions. The white and blue sticky traps were caught more moths than yellow, green and red traps. Therefore, all these information can be utilized to know when to start monitoring and enhance the use of bio-pesticides and cultural practices as integrated pest management (IPM) strategies to control *T. absoluta* in Ethiopia.

Key words: Biopesticide, Economic threshold, *Entomopathogenic fungi,* Mortality, Oviposition, Sticky trap, Tomato, *Tuta absoluta*

CHAPTER ONE

1. General Introduction

1.1. Background of the study

Tomato (*Lycopersicon esculentum* Mill.) belongs to the family Solanaceae is an important and remunerative vegetable crop grown around the world for fresh consumption and processing. It is widely cultivated in tropical, sub tropical and temperate climates and thus ranks third in terms of world vegetables production. Tomato is a vegetable crop of large importance throughout the world (Abdussamee *et al,* 2014; Mehraj *et al*., 2014; Kaur *et al*., 2014). Global tomato production is currently around 130 million tons of which 88 million tons are intended for the fresh market and the rest 42 million tons are for processing (http://www.hortibiz.com/). In Ethiopia, it is one of the economically important vegetable crops due to a favorable climate conditions of tomato production and the annual production of the crop is 30,700 tons of tomato fruits from about 5,026 ha of land (FAO, 2015; Fact fish, 2016).

Tomato is produced by small and medium scale farmers under open fields and greenhouse conditions for fresh consumption and as a source of income (Bawin *et al*., 2014; Retta and Berhe, 2015). It is an important source of nutrients such as vitamins A, B, C, E and constitutes an important part of the house hold diet and national economy (Baloch, 1994; Bhowmik *et al,* 2012; Kaur *et al*., 2014). It is also a source of basic raw materials required for fresh consumption and local processing industry for the production of processed tomato like tomato paste and tomato juice among others (EIA, 2012; AVRDC, 2014).

The production potential of tomato is 15.9 to 46.3 tons/ha (CSA, 2013), but on an average the production of tomato per hectare estimated to 7.67 tons because of different constraints including insect pests, particularly tomato leafminer, tomato fruit worm, whitefly, leafhopper, aphid, mites and thrips (Daniel and Bajarang, 2017; Tonnang *et al*., 2015; Bawin *et al*., 2015; El-Arnaouty *et al*., 2014; Assaf *et al*., 2013). Tomato leaf miner, *Tuta absoluta* (Meyrick) (Lepidoptera: Gelechiidae) is a invassive pest of tomato and other Solanaceous crops in many areas of the world causing severe damage and yield loss.

In Ethiopia, the occurrence of *T. absoluta* was confirmed under greenhouse and open field in Eastern Ethiopia since 2012 (Retta and Berhe**,** 2015; Muluken *et al*., 2014; Goftishu *et al*., 2014; Gashawbeza and Abiy, 2013). Even though studies of *T. absoluta* distribution and its impact in Ethiopia is in initial phase, it seems that first recorded in potential tomato growing areas of rift valley and central parts of Ethiopia that is a significant economic importance (Bawin *et al*., 2014; Retta and Berhe, 2015; Materu *et al*., 2016). Due to lack of natural barriers, porous borders and lack of quarantine regulation implementation in Ethiopia, the invasive species can move from one ecological community to the other through wind and human activities.

Tuta absoluta is able to cause damage on different genera and species of the solanaceae plants. Larvae may be attacked at any developmental stage from seedlings to mature plants in greenhouses and in open fields reduced tomato yield and fruit quality losses of up to 80-100% by attacking leaves and flowers, burrowing the stalks, apical buds, green and ripe fruits (Harizanova *et al*., 2009; Desneux *et al*., 2010; Garzia *et al*., 2012; Öztemiz, 2012; Yankova, 2012). It has high reproductive rate within a short period of time and capable of producing up to 12 generations per year at favorable temperature (Mollá *et al*., 2011).

The farmers of Ethiopia don’t understand the damage dimension until late, after the development of the Lepidopterous pest by noting amazed their misdeeds on the plants and the fruits which they could not sell. The rapid spread throughout the country, mainly related to the marketing of tomato berries and nursery plants, has allowed the insect to colonize all the areas involved in the production of tomato crop.

1.2. Rationale of the study

The use of chemical pesticides as its control measure is highly sought and the most effective method to reduce *T. absoluta* treat level. However, the need for alternative control measures are encouraged, considering that, the pest has developed resistance to dozens of the pesticides and the negative side effects of pesticides over-use to the environment and beneficial arthropods (Bawin *et al*., 2014).

Tuta absoluta is now one of the major insect pests of tomato in Ethiopia that poses a serious agricultural threat to tomato production areas of the country. The management strategies are extremely difficult and quite challenging due to the following factors:

1. Larvae mines within plant tissue (leaf surface and fruits) and are thus protected from contacting insecticides (Abbes and Chermiti, 2011; Guedes and Picanço, 2012; Guedes and Siqueira, 2013; Sevcan, 2013; Daniel and Bajarang, 2017).
2. They have high reproduction potential, capable of producing 10 to 12 generations per year under favorable environmental conditions. With such high reproduction potential, they are likely to undergo genetic changes (mutation) which in turn causes resistance to pesticides (Arnó and Gabarra, 2010; Muruvanda *et al*., 2012; Daniel and Bajarang, 2017). The pests were reported to be resistant to dozens of insecticides including diamide insecticide chlorantraniliprole, abamectin, methamidophos, permethri cartap (Haddi *et al*., 2012).
3. Megido *et al.* (2012) reported that some female *T. absoluta* are able to reproduce parthenogenically. This showed use of male attractant pheromeone trap is not alone effective.

Consequently, the ongoing invasion of *T. absoluta* has prompted applied research to undertake studies on many aspects of its biology and population dynamics. With the infestation of the entire Ethiopian tomato growing regions by *T. absoluta* now an important threat, there is an urgent need to understand the management options of the pest in its invaded range and develop environmentally sustainable, economically sound and effective Integrated Pest Management (IPM) strategies for this pest. There are efforts to reduce use of insecticides in tomato fields, including cultural control methods such as controlled irrigation, crop rotation, argumentative biological control and destruction of infested plant material the environment (Van Lenteren and Bueno, 2003; Abbes *et al.*, 2012). Thus *T. absoluta* control which relies on synthetic chemical alone is not acceptable. Other control measures and basic studies like cultural practices, botanical insecticides, biological control, biology of the insect, population dynamic studies, use of host resistance varieties, and sticky trap colors also needed. Therefore, experiments were carried out in the laboratory, glasshouse and under the open field conditions with the following objectives:

1.3. General objective:

- To develop knowledge based studies and management options of *Tuta absoluta*

1.3.1 Specific objectives:

- To study the population dynamics and infestation level of *T. absoluta*
- To study the biology and oviposition preferences of *T. absoluta*
- To determine the economic threshold level and yield losses due to *T. absoluta* in tomato plants
- To evaluate different bio-pesticides at different doses and sticky colors traps to control *T. absoluta*

CHAPTER TWO

2. Literature Review

2.1. Importance of Tomato

Tomato (*Lycopersicon esculentum* Mill.) is originated in the Andean region of South America (Ecuador, Peru and Chile). It was first domesticated and cultivated in Central America (Chetelat *et al*., 2009). Tomato has a great importance both as a source of food and health care. Nutritionally tomato is low in calories but high in vitamins A and C, potassium, magnesium, iron, phosphorus, sodium, niacin, riboflavin, thiamine and the valuable antioxidant lycopene and beta-carotene which play an important role in human health and is widely consumed in every household in different forms including raw, as an ingredient in many dishes, sauces, salads, and drinks (Dias, 2012; Fekadu and Dandena, 2006).

It is a seasonal crop of the family *solanaceae* and it is an economically important and widely grown vegetable crop for their fruits by smallholder farmers and commercial state and private farms in Ethiopia (AVRDC, 2014). Fresh tomato and its products are giving as a source of cash income for the households as well as in creating employment opportunity and access to smallholder farmers to participate in the market (EIA, 2012; MoA, 2010). Tomato production faces many problems from several factors which lead to significant yield loss. Recently, due to the collapse of natural barriers to wild species movements mainly in relation to human activities several factors are involved among of these factors, insect pests are the most important (Liebhold and Tobin, 2008). The newly introduced *Tuta absoluta* causes a very high level of damage both in quantity and quality to tomato crops, particularly if no control measures are undertaken (Desneux *et al*., 2011; Guedes and Picanço, 2012; Megido *et al*., 2012).

2.2. Origin and Geographical Distribution of *T. absoluta*

Tuta absoluta was originally described in 1917 by Meyrick as *Phthorimaea absoluta*, based on individuals collected from Huancayo (Peru) but the pest was reported as *Gnorimoschema absoluta*, *Scrobipalpula absoluta* (Povolny), or *Scrobipalpuloides absoluta* (Povolny), but finally described under the genus *Tuta* as *T. absoluta* by Povolny in 1994 (Clarke, 1962; Barrientos *et al.*, 1998; CABI, 2011). It belongs to the family Gelechiidae which includes about 400 genera and 4000 species of small moths represented all over the world, which are the most important insects at this family the pink bollworm (*Platyedra gossypiella* Saund.) in the widest spread and one of the most destructive of cotton and the potato tuber moth (*Phthorimace operculella* zeller) is a widest spread pest of stored potato (Richards and Davies, 1983).

Tuta absoluta is a native to South America (EPPO*,* 2005; Urbaneja *et al*., 2007; Viggiani *et al*., 2009). It was reported since the early 1980s from Argentina, Brazil and Bolivia (NAPPO, 2012). The insect rapidly invaded many European and Mediterranean countries. According to Devaiah *et al*. (2012) continents having problem of *T. absoluta* are Asia, Europe, America, and Africa.

2.3. Life history and Description of *T. absoluta*

The life cycle of *T. absoluta* has four developmental stages such as egg, larva, pupa and adult (Nicolas *et al*., 2010). Adults are nocturnal in habit, usually remain hidden during the day, showing greater morning crepuscular activity and they disappears among crops by flying (Estay, 2000). A single female can lay a total of about 240 to 260 eggs during her lifetime throughout the day on the aerial parts of the plant but peak oviposition occurs at night (Fernandez and Montagne, 1990; Estay, 2000; Riquelme, 2009; Fargalla and Shalaby, 2013). However, *T. absulata* prefers to lay eggs on the leaves. There are four larval stages from first instar to fourth instar (Nicolas *et al*., 2010).

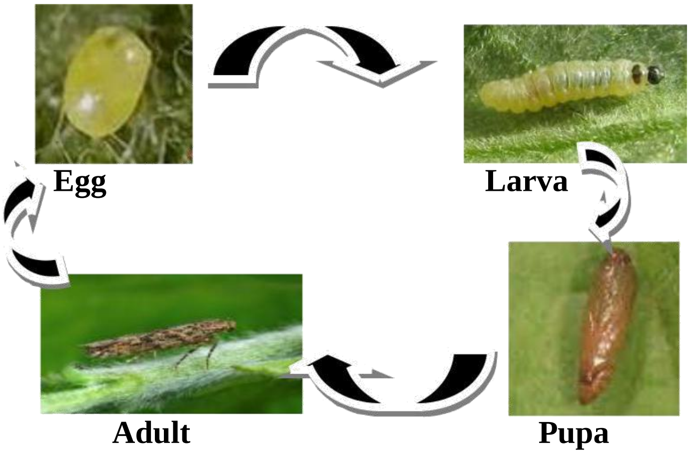


Figure 2.1: Life cycle of *T. absoluta* (**Source:** NAPPO, 2012).

Larvae spin a silken cooon where they transform into pupae. Pupae can be found attached to all plant parts (leaves, main stem, flower, fruits) as well as in the soil (Torres *et al*., 2001). If the environmental condition is conducieve *T. absoluta* may produce 10 to 12 generations per year (Barrientos *et al*., 1998; Gadir *et al*., 2016).

2.4. Economic Importance of *Tuta absoluta*

Tuta absoluta is a key pest in greenhouse and open field conditions for tomato production in South America and in Argentina (Ferrara *et al*., 2001; Botto, 2011). *T. absoluta* is considered as one of the most serious insect pests associated with tomatoes production in different parts of the world specifically in Brazil (Torres *et al*., 2001). The tomato leafminer, *T. absoluta* caused 80 to 100 percent yield losses in tomato crops (Leite *et al.*, 1998; Estay, 2000; EPPO, 2008; Leite *et. al.* 2010; Ramireze *et. al.,* 2010; IRAC, 2014). Both yield and fruit quality can be significantly reduced by direct feeding of the larvae, and their entry-way used by secondary pathogens entering the mines and causing fruit rot (EPPO, 2005).

Tuta absoluta became the major insect pest of tomato in Ethiopia since 2012 causing high infestation level and damage (Gashawbeza and Abiy, 2013). It is considered as a major pest of tomato both in the field and under greenhouse conditions in Ethiopia. It is not only to the intensity of its attack, but also to its occurrence throughout the entire crop cycle (Oliveira *et. al.* 2008).

2.5. Host plants

The hosts were reported from their current distributions, and the host species. If pests are introduced into new areas, they may attack native species that have not previously been identified as host plants. Therefore, host species should be surveyed and survey should be broadened to native species within the host genera. The pest lays eggs in all above ground portions of the plant (leaves, shoots and flowers) including on the fruit. Its main host is tomato, but infestations of *T. absoluta* have also been reported on *Solanum torvum* (egg plant), *Solanum tuberosum* (Potato) and common bean.

2.6. Damage

The female laid her eggs on the aerial parts of the host plants. The larvae of *T. absoluta* infest the leaves, flowers, shoots, buds, stems, calyces, and tomato fruit (Pastrana, 2004; Harizanova *et al*., 2009; Yankova, 2012). Leaf mines are irregular and may later become necrotic. The larvae usually enter the fruit under the calyx and tunnel the flesh, leaving galleries clogged with frass that cause the fruit to drop or to rot on the vine. So it cause negatively effects on plant architecture and can result in a significant reduction of fruit yield (Fernandez and Montagne, 1990; Botto, 2011).

2.7. Population dynamics of tomato leaf miner

Tuta absoluta is oligophagous pest of solanaceous crops (Lietti *et al*., 2005). A closely related Gelechiid species, the tomato pink worm, *Keiferia lycopersicella* (Walshingham), occupies the ecological niche of *T. absoluta* on tomatoes in the United States (CABI, 2011). EPPO (2005) reported that the optimum temperature for *T. absoluta* development ranged from 19-23 ºC. At 19ºC there was 52% survival of *T. absoluta* from egg to adult.

Tuta absoluta is multivoltine and rapid population growth, potential dispersal through environment and expressed resistance to insecticides (Pereyra and Sanchez, 2006; Desneux *et al*., 2010). The biological and ecological characteristics of *T. absoluta* are needed to establish effective management. Thus, various factors must be taken into account such as climate interactions with the biology of the pest and multi-species interactions such as competition and predation (Polat *et al.,* 2016; Tonnang *et al*., 2015). There are several control mrasures were conducted to reduce the infestation level and damage caused by *T. absoluta* which including the use of chemical sprays, mass trapping and releases of natural enemies (Urbaneja *et al*., 2012; Zappalà *et al*., 2013; De Backer *et al*., 2014).

2.8. MANAGEMENT

2.8.1. Cultural Control

Good cultural practices are very important for the control of *T. absoluta* include crop rotations with non-solanaceous crops, ploughing, adequate irrigation and fertilization, removal of infested plants and complete removal of post-harvest plant debris and fruits (Retta and Berhe, 2015). Destruction of infested plant materials is important cultural control practices that would help to tackle this pest in green houses and glasshouse.

The Solanaceous weeds in the vicinity of infestation greenhouse should be removed and destroyed (Kopper, 2009). In the Unite State, a crop rotation with host frees period essential for reducing (tomato pin worm) population in tomato crop (Zalom *et al*., 2008). Daniel and Bajarang (2017) stated that through good agricultural practices to reduce *T. absoluta* population in the field.

To prevent population build up one should not leave infested plant material on the ground, as the larvae will quickly leave them and colonize new plants. After harvesting, crop residues should be destroyed as soon as possible. When the pest infestation is low, it is important to remove any of infested leaves, stems and fruits affected by the presence of larvae or pupae and destroy them. Remove all solanaceous family weeds that may be host to the pest within the area of vicinity.

2.8.2. Botanicals

The botanical pesticides are the important alternative to minimize or replace the use of synthetic pesticides as they possess an array of properties including toxicity to the pest, repellence, anti feeding and insect growth regulatory activities against pests of agricultural importance. The uses of botanical insecticide have many advantages over synthetic pesticides which include: it has low mammalian toxicity thus constitute least or no health hazard environmental pollution, on risk of developing pest resistance to these products, less hazard to non-target organisms, no adverse effect on plant growth, it is less expensive and easily available because of their natural occurrence especially in oriental countries (Prakash and Rao, 1997).

Due to problems and hazards associated to insecticide applications, many studies focused on the use of plant extract to control *T. absoluta* (Moreno *et al*., 2011; Nilahyane *et al*., 2012; Ghanim and Abdel, 2014). Previous studies reported effective larval control of *T. absoluta* with botanical extracts (Trindade *et al*., 2000), for instance, the effects of neem seed and Jatropha seed plant extracts and other seven plants against *T. absoluta* had varying levels of toxici ty on the larvae of *T. absoluta* were effective (Nilahyane *et al*. 2012; Nada *et al*., 2014).

In this study five locally available botanical extracts were tested against larvae of *T. absoluta*. Those botanicals are: *Allium sativum*, *Azadirachta indica, Cymbopogon citrates*, *Nicotiana* sp. and *Phytolacca dodecandra.* Onion and garlic extracts showed significant effects against the tested pests especially *T. absoluta* larvae under laboratory conditions (Nabil and Sherif, 2014). The studies were conducted under laboratory conditions showed that extracts of garlic have been shown effective against larvae of *Culex* and garlic extracts showed the highest effects on *T. absoluta* second instar larvae while basil leaves extract exhibited the least effect (Ghanim and Abdel Ghani, 2014).

The Neem plant contains a number of active metabolites such as alkaloids which can control insect pests. These compounds have been reported to have control efficacy against tomato borer (Goncalves and Vendramis, 2008). A number of references (Stoll, 2000; Hiiesaar *et al*., 2001) indicated that, many of these plant materials show a broad spectrum of activity against insect pests, such as lethal, antifeedant, repellent and growth regulatory effects. Accordingly, many trials were made for its control and suppression of damage through application of chemicals, botanicals and pheromones (Mohamed and Khalid, 2011). Many previous studies reported effective larval control of *T. absoluta* with botanical materials. Trindade *et al*. (2000), Moreno *et al*. (2011) and Hussein *et al. (*2014) reported that Lemon grass extract significantly increased L-ascorbic acid (Vitamin C) contents in tomato fruits and reduced the infestation level of *T. absoluta* under greenhouse conditions.

Phytolacca dodecandra is native to sub-Saharan Africa and Ethiopia (Lemma *et al*., 1972) which is used for different medicinal purposes to treat various ailments in humans and also in animals (Nalule *et al*., 2011). The medicinal values of the plants are well documented in various parts of the World. In Ethiopia, people use leaf juice to treat Malaria (Mesfin *et al*., 2009). The crude berries extracts was reported to have strong toxicity effectiveness against aquatic macro invertebrates such as Baetidae and Hydropsychidide (Karunamoorthi *et al*., 2008). Larvicidal and pupicidal experiment was conducted at botany laboratory University of Gondar, reported that the immature mosquitoes were exposed to selected concentration and high percentage mortality was observed (Nurie *et al*., 2012).

Other plants which are promising in management of *T. absoluta* include Piper whereas compounds from *Acmella oleracea* were revealed to be active against *T. absoluta* (Moreno *et al*., 2011). Though biochemical pesticides have been cited as promising for pest control, their application in the sub-Saharan Africa is limited and none of the compounds have been registered commercially to help farmers. Hence more researches and validation of these natural resources is highly demanded to protect crop damage and loss including those by *T. absoluta* (Cork *et al*., 2009). Plant based pesticides have been documented to be better than synthetic chemical pesticides as they are biodegradable, naturally available and environmentally friend to none targeted organisms (Never *et al*., 2017). Though plant product (botanicals) have been cited as promising for pest control, their application in Ethiopia is limited and none of the compounds have been registered commercially to help farmers and stalk holders. Therefore, more researches and validation of these natural resources is highly demanded to protect crop damage and loss from any pests.

2.8.3. Biological control

2.8.3.1. Predatory insects and Parasitoids

Natural enemies help to maintain a balance among insect pests, by consuming prey, altering prey behavior and prey habitat selection (Smee, 2012). Thus, predators may increase the biodiversity of communities by preventing a single species from becoming dominant (Botkin and Keller, 2010). Although specialized natural enemies are considered as most promising in biological control (Hassell, 1978), the generalist predators may also be of major importance in pest suppression (Rosenheim *et al*., 1993).

In Brazil, anthocorid predator *Xylocoris* sp., *Cycloneda sanguinea* and members of Phlaeothripidae proved to be key predators of both eggs and larvae of *T. absoluta* (Miranda *et al*., 1998). As pointed out by several authors (Nakasu *et al*., 2013), the occurrence of anthicid predator *Anthicus* sp., coccinellid predator *C. sanguinea*, Staphylinidae, *Orius* sp. and *Xylocoris* sp. and Formicidae were observed predating on *T. absoluta*. The predaceous was *protonectarina sylveirae* is considered as one of key mortality factors of *T. absoluta* in the spring-summer (Bacci, 2006; 2008). Oliveira *et al.* (2007) indicated that the mite *Pyemotes* sp. feeds on *T. absoluta* larvae, pupae and adults. They described its potential use in biological control of the pest.

The natural enemies for *T. absoluta* have been reported from their place of origin (South America), which are commercially available and can be used in its control. A studies stated by (Sanchez *et al.*, 2014; Refki *et al.*, 2016) showed that commercially available predators are mentioned. Another done by (Sanchez *et al.*, 2014) in Mediterranean region using *Nesidiocoris tenuis,*, showed highly promising results and effectiveness of predator use when combined with other methods in controlling *T. absoluta*.

The egg parasitoid *Trichogramma achaeae* has been identified as a candidate for biological control of the South American tomato pinworm *T. absoluta* on greenhouse condition a high efficacy of damage reduction was obtained (Cabello *et al*, 2009; Megido *et al*., 2013; Retta and Berhe, 2015).

Mirid *Tuptocort cucurbitaceaus* has been recently evaluated as a potential biological control agent against *T. absoluta* and white flies in Argentina (Lopez, 2010). It is reported that combine application of mass release of *Tichogramma pertiosum* and *Bacillus thuringiensis* resulted fruit damage only 2% in South America (Medeiros *et al*., 2006). Cabello *et al*, (2009) also have been reported the following bio-agents are a potential to control *T. absoluta: Trichogramma pertiosum, Trichogramma achaeae* and *Nabis pseudoferus.*

2.8.3.2. Entomopathogenic fungi

The importance of microorganism as biopesticides for management of pests has increasingly gained popularity in recent years (Mollár *et al*., 2011). The formulations are either by foliar spray or by drenching the roots (Amizadeh *et al*., 2015). One of the best and successful formulations was that of *Metarhizium anisopliae* (fungus) and *Bacillus subtilis* (bacteria) which have been reported to reduce the population of *T. absoluta* on tomato at all developmental stages in America and Europe (Inanli *et al*, 2012). Other formulations reported to be tested against include that of *Metarhizium anisopliae* and *Beauveria bassiana* (Inanli *et al*., 2012; Kaoud, 2014). Most of these reports however were all based on screen house studies (González-Cabrera *et al*., 2011; Sabbour and Nayera, 2014) and only a few have been tested on field conditions and thus they may not be readily available for small-holder farmers.

Currently there are many commercially available bacterial and fungal formulations for controlling pests including *T. absoluta* in America and Europe (Sabbour, 2014). Incontrast, Studies conducted by Youssef and Hassan (2013), Gözel and Kasap (2015) and and Mahmoud (2017) revealed poor documentation on effectiveness of entomopathogens controlling *T. absoluta. Bacillus thuringiensis*, an entomopathogenic bacterium has been used in the control of tomato plant pests and reported by many authors as very effective bio-insecticide. Bio-insecticides like *Bacillus thuringiensis* do not raise any environmental concern as they are environmentally friendly.

Bacillus thuringiensis, an entomopathogenic bacterium has been used in the control of tomato plant pests and reported by many authors as very effective bio-insecticide (Youssef and Hassan, 2013). It has been used extensively to control the pest in crops where IPM programmes based on biological control are applied. Bio-insecticides like *Bacillus thuringiensis* do not raise any environmental concern as they are environmentally friendly. In addition, the entomopathogenic nematodes *Steinernema carpocapsae*, *Steinernema feltiae* and *Heterorhabditis bacteriophora* have proved to be capable of infecting late larval instars of *T. absoluta* and hence be used in its control (Gözel and Kasap, 2015; Mahmoud, 2017).

2.8.3. Pheromone Trap and Sticky trap

There are many types of traps which were used against *T. absoluta* such as Pheromone-baited traps, sex pheromone, mass trapping, rectangular plastic traps, sticky trap, delta traps and mass trapping. The light traps has been used to control *T. absoluta* in the greenhouse tomato production in Italy at a height of 1 meter or less from the ground and at rate of one trapper 50 to 100 m (Laore, 2010). Light traps should be placed near entry doors and used only during sunset and sundown (Bolkman, 2009). Russel IPM (2009a) recently developed a light trap for *T. absoluta* that capable of capturing thousands of male insects.

Studies in Chile revealed that the greatest number of males were captured in pheromone traps during the period of 7 to 11 A.M., suggesting that this is the time when males are searching for calling females (Miranda-Ibarra, 1999). Hickel *et al*. (1991) studied the mating behavior of *T. absoluta* in the laboratory and determined the sequence of male mating behaviors can be divided into two phases that are long-range female location and short-range courtship.

Pheromone-baited with synthetic sex pheromone for monitoring population of *T. absoluta* in open field, greenhouses and packing sites. Studies are being done on the use of synthetic sex pheromones in order to monitor population levels and trigger applications of chemicals (Goettel and Inglis, 1997).

The mass trapping is involving placing a large number Pheromone-Baited traps in the strategic position within a crop which are trapped males resulting in an imbalance to the sex ratio which impacts the mating pattern of *T. absoluta*. It can be used to reduce *T. absoluta* population and is particularly useful in production of greenhouse tomato (Russel IPM, 2009b). The pan traps is easier to maintain and are less sensitive to dust, compared to Delta, Mephail and light traps. It is also have a large trapping capacity than Delta traps. Rectangular plastic traps that hold 6 to 8 liter of water baited with pheromone lure are recommended for it (Info Agro System, 2009b).

The traps are used prior to other control strategies so as to determine the presence and abundance of insects so as to decide on appropriate control measure to apply (Cocco *et al*., 2013; Witzgall and Cork, 2010). Although these traps are designed to control only adult male moth, they have been reported to be effective in managing tomato borer (Cocco *et al*., 2013; Reddy and Guerrero, 2010; Braham, 2014; Cocco et al., 2012; Vacas *et al*., 2011). For effective application in the field, the sex pheromone traps are to be properly hanged at right positions depending on the height of tomato varieties and wind direction (Soliman *et al*., 2013). Another factor reported to be important is the color of the trap which, affects and influences the movement of the pest towards it, thus enhancing trapping efficiency (Megido *et al*., 2013; Mwangi, 2015). Although pheromone traps in combination with active insect killing agent is reported to be used against *T. absoluta*, no study has reported the efficacy of pheromone traps when synergized by active plant compounds. Due to the current *T. absoluta* situation in Ethiopia, it is evident that a pheromone trap baited with active compound could be developed and deployed in fields to improve monitoring and control of *T. absoluta.*

2.8.4. Chemical insecticides

Tuta absoluta has been controlled with chemical, organophosphates and pyrethroids were used during 1970s and 1990s until new products introduced in the 1990s such as abamectin, spinosade, tebufonzide and chlafenpyr) became available (Lietti *et al*., 2005). Since the 1980s, efficacy of organophosphates for *T. absoluta* control has gradually decreased in Bolivia, Brazil and Chile (Siqueira *et al*., 2000, 2001) and rapid development of insecticide resistance. Several treatments are required per growing season and it must be noted that a decrease of the efficacy of products used against *T. absoluta* has been observed since the 1980s in tomato crops (Kaoud, 2014). Resistance to some insecticides has been reported in several countries, for example to abamectin, cartap and permethrin in Brazil (Goettel and Inglis, 1997).

The internal living and feeding habits of the larvae and its ability to produce several generations each year makes it necessary for farmers to apply insecticide every 4–5 days/season with minimum and maximum numbers of sprays 8 to 25 sprays, respectively (Temerak, 2011). In Argentina, *T. absoluta* was reported to be resistance to deltamethrin, and abamectin (Lietti *et al*., 2005). Resistance to cartap, abamectin, permethrin and methamidophos (Siqueira *et. al*., 2000) and acephate and deltamethrin (Branco *et. al.,* 2001) has been reported in Brazil. Intensive use of insecticides results in environmental pollution, the evolution of insecticide resistance and consequent insecticide control failure (Silva *et al*., 2011).

In Ethiopia, recently the pest has been detected throughout the country and it may become a significant problem in greenhouses as well as in open fields. An immediate consequence of the introduction of *T. absoluta* is illustrated by the sudden increase in insecticide use in Ethiopia tomato fields, going from 8-12 applications per cultivation period, at first time requiring every five days in three consecutive spraying times, then after two to three week intervals insecticide applications needed in Dandi district West Shewa of central Ethiopia (Personal communication). Besides the environmental and human safety concerns of such procedures, the tomato production costs more than doubled with the introduction of *T. absoluta* and the prototype of insecticide use required for its control.

Failure of these chemicals in controlling *T. absoluta* opened a new window for development of other methods including biopesticides, pheromone traps, and parasitoids (Regnault-Roger, 2012; Cherif *et al*., 2013; Zappala *et al*., 2013). Though chemical pesticides are economically and environmentally unaffordable, farmers still seek them for their agricultural uses because is the only easily accessible option.

2.8.5. Integrated Pest Maagmet

IPM strategies are being developed in South America to control *T. absoluta*. The current management of *T. absoluta* in the Mediterranean Basin is mainly based on treatments with chemical insecticides. Various active substances are effective and can be used in combination with biological control agents. Nevertheless, few active ingredients are effective against *T. absoluta* and selective to beneficial insects including pollinators. Therefore, integration with other methods becomes imperative, as continued use of chemical insecticides could harm non-target organisms (Landgren *et al*., 2009). Integrated pest management (IPM) strategies have been also adopted to control this insect pest. For implementing environmentally safe strategies, several eco-sustainable control methods and integrated pest management (IPM) programs have been recently evaluated (Batalla-Carrera, 2010; Zappala *et al*., 2012).

CHAPTER THREE

Population dynamics studies of tomato leaf miner, *Tuta absoluta* on Tomato, *Lycopersicum esculentum* (Miller)

3.1 Introduction

Tomato, *Lycopersicum esculentum* (Mill.) is one of the economically important vegetable crops and widely cultivated in the world with a total area and production of 5,227,883 ha and 129,649,883 tons in 2008, respectively (FAO, 2009).

The unintentional introduction of exotic insects has resulted in a high number of established species with considerable negative economic impact like tomato leaf miner. Recently, the production of tomato has been declined due to various factors including insect pest and diseases (Materu *et al*, 2016; Chidege *et al*., 2016). There are several insect species feed on tomato (Assaf *et al*., 2013) including tomato leafminer, whose potential geographic distribution and relative abundance are mostly poorly understood, even after costly and long-standing management programs (Gutierrez and Ponti, 2012).

This invasive pest, *T. absoluta* is considered a serious threat to tomato production worldwide (Chidege *et al*., 2016). Thousands of tomato farmers are suffering from serious production losses due to devastating pest that are destroying their valuable crop (Chidege *et al*., 2016; Materu *et al*, 2015). *Tuta absoluta* can cause losses of 80 to 100% in tomato farms in glasshouse or/and in open fields if control measures are not properly implemented (Öztemiz, 2012). The pest was reported for the first time in Eastern Shewa of Oromia Regional State, Ethiopia since 2012 and then invaded other regions such as Tigiray, Amhara, and Gambella (Gashawbeza and Abiy, 2013; Goftishu *et al*., 2014; Retta and Breh, 2015). *T. absoluta* can devastate an entire tomato farm, if effective control measures are not employed.

Since it was introduced to Ethiopia in 2012, there was no any control mechanisms and action plan being implemented by the government, research institutions, horticultural agencies and other stakeholders to control and mitigate the progress of the spread of *T. absoluta* to new regions of the country. Consequently, the ongoing invasion of *T. absoluta* has prompted applied research to undertake studies on many aspects of its biology, ecology and population dynamics. The infestations of *T. absoluta* in tomato growing regions of Ethiopia are an important threat. There is an urgent need to understand the management options of the pest. It needs to develop environmentally sustainable, economically sound and effective Integrated Pest Management (IPM) strategies. Therefore, the study was carried out to assess the population dynamics, host preferences and infestation level of *T. absoluta* on tomato plants.

3.2 Materials and Methods

3.2.1 Description of the study area

Population dynamics study of *T. absoluta* was conducted during 2015-2017 for greenhouse and open-field studies on tomato plants. The glasshouse studies were carried out at Ambo University plant Sciences laboratory. Ambo is found at geographical coordinate of 8^{O}59’N latitude and 37.85^{O}E longitude with an altitude of 2100 m.a.s.l. (Briggs, 2012). The temperature of the laboratory during the study period was 22 ± 2^{O}C and that of the glasshouse was 32 ± 2^{O}C. The field experiments were carried out on farmers’ field at three different districts (Ambo, Dendi and Toke Kutaye) of West Shewa Zone, Oromia Regional state, Ethiopia on tomato plants. Ambo is at geographical coordinate of 858'59.988"N latitude and 3751'0.000"E longitude with an altitude of 2076 meter above sea level (Briggs, 2012). Toke Kutaye districts is located at 126 km west of Addis Ababa having an altitude of 1990 meter above sea level, latitude of 0859' 01.100" N and longitude of 3746'27.600" E. The average annual rainfall is 1028.7 mm and maximum and minimum temperatures of the area 29.6°C and

11.8°C, respectively. The geographical location of the study area of Dandi district also having an altitude of 2272 meter above sea level, latitude of 4064' 81.00" N and longitude of 9978' 63.00" E.

3.2.2 Leaves sampling for recording eggs, larvae and pupae

Tomato cultivar “Koshoro” was used for the experiment. The experiment was conducted during October 2015 to February 2017 for 24 months. Planting was done on the 1st October and Feburary of the study periods. More one, similar studies was conducted in Ambo University main campus in glasshouse. For glasshouse experiment planting was done every four months starting from 1st October 2015 which ended on 30th September 2017.

The number of eggs, larvae, pupae and mines of *T. absoluta*, on tomato leaves were collected from three different field conditions. Each field with a similar number of plants to ensure that all the area was represented in the samplings. Every 10th day sampling was carried out from October to January, February to May and June to September in glasshouse and open fields. Ten plants were tagged and at each plant five leaves were collected and individually packed in labeled plastic cans then transported to Ambo University Plant Sciences laboratory. On the laboratory, with aid of Binocular microscope, each leaf was examined and the number of eggs, mines and larvae per leaf was recorded (Leite *et. al.*, 2001). The minimum and maximum temperature and relative humidity data were recorded during the study periods.

3.2.3 Data Analysis

The data was subjected to analysis of variance (ANOVA) and the means were compared by least significant different (LSD) test at 0.05 levels, using SAS program version 9.1 (SAS, 2009). Simple linear regression of parameters and the distribution of eggs, larvae and pupae per plant were performed. Coefficient of determination values were also determined to see their role in percentage on the population dynamics of the pest.

3.3 Results

3.3.1 Population Dynamic study

3.3.1.1 Glasshouse Experiment

3.3.1.1.1 Egg, larval and pupal count across months

The results showed that significant ($P < 0.01$) differences were observed among the months. Results of mean monthly counts of different stages of *T. absoluta* are shown in Fig 3.1. *T. absoluta* egg was seen in mid-December from the first cropping cycle of October to January. The number of eggs, larvae and pupae steadily increased towards the end of January. The graph for population dynamics (Fig. 3.2), from the second cropping cycle of Feburary to May all stages of *T. absoluta* increased starting from May through September. During this cycle the flying adults were highly populated in the glasshouse.

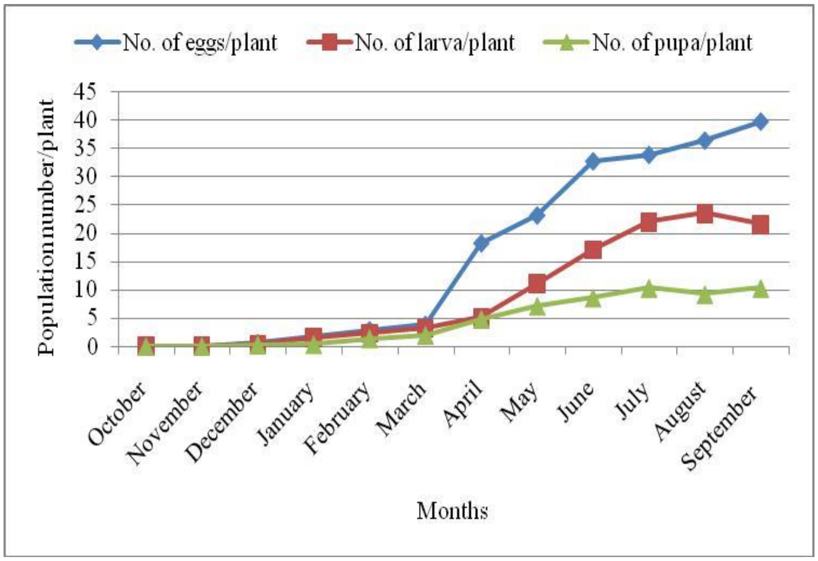


Figure 3.1: Monthly average counts of *T. absoluta* of different stages per plant under glasshouse conditions during 2015-2016.

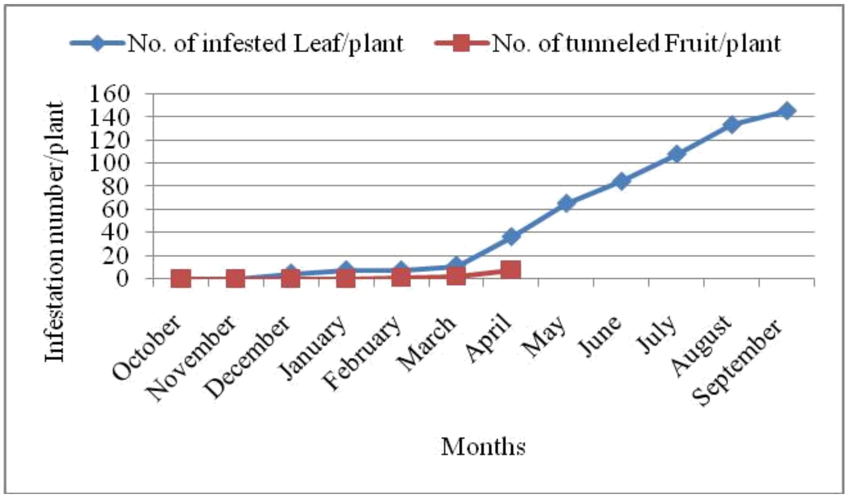


Figure 3.2: Damaged of *T. absoluta* on tomato plant under glasshouse conditions during 2015-2016.

Table 3.1 demonstrated that *T. absoluta* counts and damage are significantly varied across the months. Damaged of leaves and tunneled fruits of tomato plants caused by larval instars occured throughout the months of December 2015 to September 2016.

Table 3.1: Mean *T. absoluta* count and damage to tomato under glasshouse conditions during 2015-2016.

Months	Mean counts of *T. absoluta* per plant			Percent *T. absoluta* Infestation	
	egg	**larva**	**pupa**	**Leaf**	**Fruit**
October	0.00^{d}	0.00^{e}	0.00^{e}	0.00^{f}	0.00
November	0.00^{d}	0.00^{e}	0.00^{e}	0.00^{f}	0.00
December	0.57^{d}	0.37^{e}	0.27^{e}	4.63^{f}	0.00
January	1.70^{d}	1.57^{de}	0.33^{e}	5.56^{f}	0.30
February	2.87^{d}	2.33^{de}	1.37^{e}	8.92^{f}	0.80
March	3.83^{d}	3.17^{de}	2.0^{d}	11.37^{f}	2.15
April	18.27^{c}	5.17^{d}	4.83^{c}	36.17^{e}	7.67
May	23.2^{c}	11.13^{c}	7.13^{b}	65.07^{d}	***

June	32.80^{b}	17.17^{b}	8.60^{ab}	84.17^{c}	***
July	33.93^{b}	22.03^{a}	10.31^{a}	107.73^{b}	***
August	36.47^{ab}	23.60^{a}	9.20^{a}	133.27^{a}	***
September	39.83^{a}	21.60^{a}	10.30^{a}	145.33^{a}	***
LSD at 0.01	5.69	3.63	1.93	14.85	
SE ±	3.34	2.13	1.14	8.72	
CV (%)	20.72	23.66	25.09	17.52	

Note: *** Due to highest damaged of *T. absoluta* the leaf became dried and no fruit setting (100% yield losses)

According to this results, damage on tomato plants were much larger in the third cropping cycle followed by second, apart from damage on tomatoes in the first cropping cycle, with tomato plants from the second cropping cycle, gradual increasing of damages on the leaves. In this cycle, the largest and most evident were damages on the leaves due to the high larval population occurred and also the mean temperature of the glasshouse 20.5-32.5^{o}C during this cropping cycle.

It is important to emphasize that with tomato plants from the first cropping cycle there were damages on the leaves, flowers and fruits. *T. absoluta* larvae were damaged tomatoes in the second cycle, damages on all entire part of the plant were visible. However, the greatest damages were found on the leaves of the plants during the third cropping cycle, because of this damaged no flowering and fruit setting of tomato during the end of second and third cropping cycle shown above in Table 3.1 and Fig. 3.2. As indicated above in Table 3.1 the highest infestation of *T. absoluta* on tomato plants were detected during the 3rd cropping cycle at temperature 20 - 27.5 (84.17 - 145.33 leaves/plant).

Population density and damages were presented in the above Fig. 3.2 for each month of observation. The eggs, larvae and pupal density of *T. absoluta* and months of observations were a strong positive correlation and linear relationship between those two variables. Damaged leaf and tunneled fruit revealed significant ($P < 0.05$) positive correlation in both the years (2015 - 2017). The linear regression co-efficient 'b' value during the same year was significant with higher r^2 value showing more pronounced effect of *T. absoluta* eggs, larvae and pupae on leaves. The highest value of r^2 (0.966) indicated 96.6% variation in yield due to *T. absoluta* larvae damaged of leaves (Fig. 3.3). The regression equations derived were below:

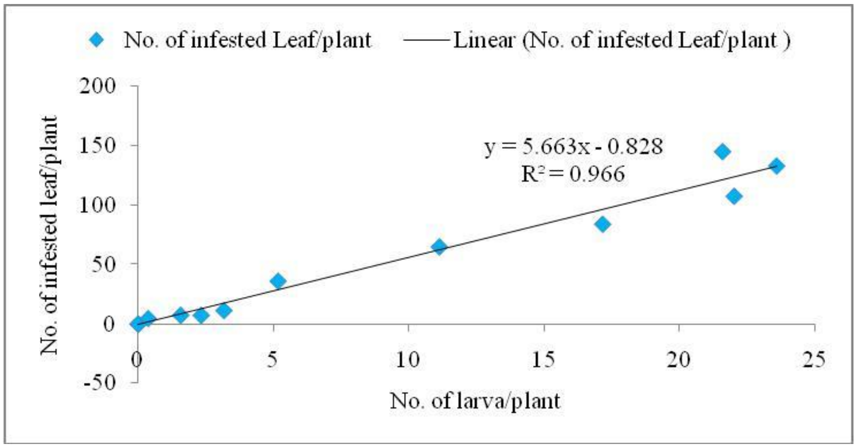


Figure 3.3: Simple linear regression of leaf infestation by number of larvae of *T. absoluta* during 2015 to 2016 under glasshouse conditions

Table 3.2: Mean temperature and relative humidity under glasshouse conditions during 2015-2017

	Temperature (^{0}C)			**Relative humidity (%)**		
Months	**Min**	**Max**	**Mean**	**Min**	**Max**	**Mean**
Oct.	24	30	27.0	57	68	62.5
Nov.	21	28	24.5	52	55	53.5
Dec.	25	33	29.0	44	52	48.0
Jan.	27	35	31.0	39	46	42.5
Feb.	26	39	32.5	37	44	40.5
Mar.	24	37	30.5	42	47	44.5
Apr.	23	35	29.0	41	46	43.5
May	25	37	31.0	40	49	44.5
June	22	33	27.5	47	52	49.5

July	20	28	24.0	60	66	63.0
August	16	25	20.5	55	60	57.5
Sep.	18	27	22.5	56	62	59.0

During second year (2016-2017) study period the population number of all stages of *T. absoluta* were increased during months of June to August 2017 under glasshouse conditions. The infestation level of *T. absoluta* larvae was very high during months of July and August 2017. Due to highly damaged leaves the plant becomes dead at flowering stage and no fruit setting was observed (Fig. 3.5). As compared with first year (2015-2016) the infestation level and population number of the pests somehow similar.

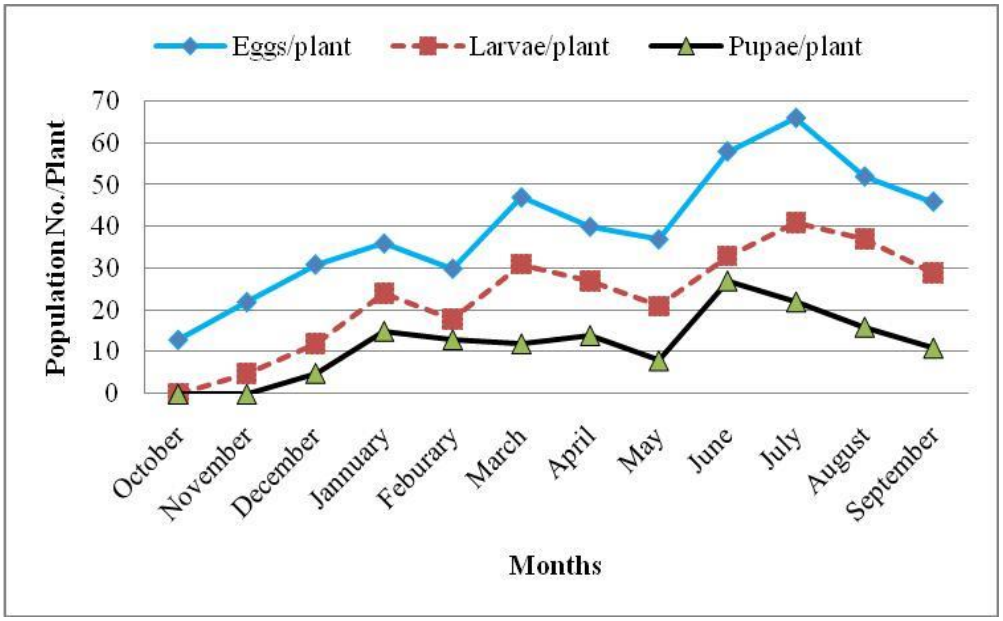


Figure 3.4: Monthly average population dynamics of *T. absoluta* on different stages per plant under glasshouse conditions during 2016-2017.

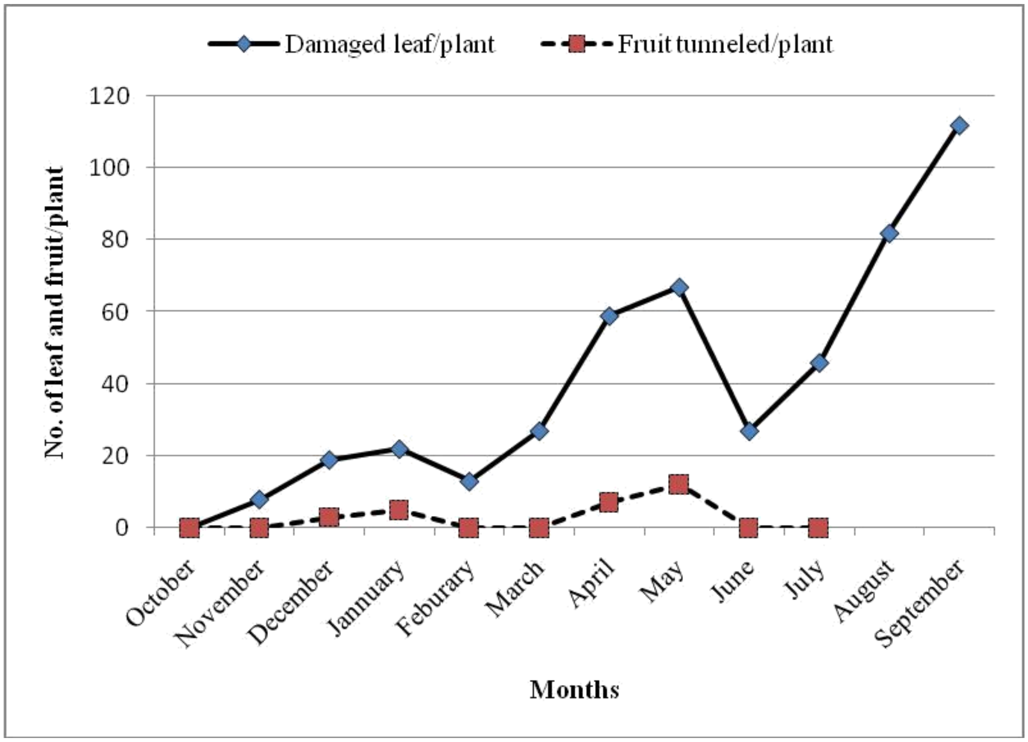


Figure 3.5: Damaged of *T. absoluta* on tomato plant under glasshouse conditions during 2016-2017

3.3.1.2. Field Experiment

3.3.1.2.1. Egg, Larval and pupal-population versus Months

The statistical analysis showed that significant ($P < 0.05$) differences were observed among the treatments. *Tuta absoluta* was observed in these main cropping cycles at Ambo, Dendi and Toke kutaye districts, Western Shewa of Ethiopia, although the maximum densities were found in all the study areas. The results of the three study areas in first and second years (2015-2017) in both cropping cycles are presented in Figures (3.6a-3.8d).

At Ambo district first cropping cycle *T. absoluta* population revealed steady progressive (after crop transplanted) to the field and maximum population recorded in the late of December (5.4 eggs/plant and 2.36 larvae/plant) and declined thereafter. In the second cropping cycle peaked in the late March (24.62 eggs and 11.74 larvae/plant) were recorded in the same district during 2015/2016.

During 2016/2017, at Ambo district first cropping cycle eggs were first recorded at the late of December (Fig. 3.6a) whereas at Dandi district, eggs were first observed at beginning of November (Fig. 3.7a). On the other hand, at Toke kutaye eggs were first found at the mid December (Fig. 3.8a). The first peak was observed at the vegetative to initiation of flowering of the crop at all study areas. The second period of *T. absoluta* activity occurred at the beginning of February at all locations. Similarly, the peak of *T. absoluta* eggs and larvae at Toke kutaye in the first and second cropping cycles were recorded at mid December (14.26 eggs/plant and 9.06 larvae/plant) and the population remained high up to the end of April (Fig. 3.8a & 3.8b), respectively.

The insect infestation appeared in the early November at Dandi compared with Ambo and Toke kutaye (Fig. 3.7a). The early appearance of *T. absoluta* at Dandi in this particular cropping cycle was associated with early planting of the crop in the surrounding farmer’s fields. It was recorded highest population number at mid December (10.48 eggs/plant). In the second cropping cycle the population of eggs, larvae and pupae of *T. absoluta* were high at the end of April (22.62 eggs/plant, 16.30 larvae/plant and 9.36 pupae/plant) compared with Ambo and Toke kutaye districts in all cropping cycles (Fig. 3.8b).

The overall impacts of *T. absoluta* at Ambo and Toke kutaye, the populations were low in the second year in both cropping cycles as compared to the previous year. *T. absoluta* had two peaks per year, the first peak month of December at all study areas whereas, the second peak recorded at the end of March and the beginning of April (Fig. 3.6a, 3.7d & 3.8d). The different stages of *T. absoluta* (egg, larva, and pupa) begun to increase at vegetative stage of the crop before fruit setting and peak numbers attained at flowering stage. During vegetative and flowering stage, most of the product of photosynthesis move upward to developing fruits, at vegetative period the adult *T. absoluta* concentrated to laid eggs on the upper young leaves and developing fruit as it was observed in all occasions. In all study areas, the maximum eggs laid were found at vegetative stages followed by flowering stages. In the first year (2015/16), second cropping cycle the population of *T. absoluta* in all stages (egg to pupa) found at seedling stages and the maximum populations were recorded at March (14.26 - 24.62 eggs/plant) and the minimum eggs were recorded at the mid of May at maturity stages. On Similar manner, at Dandi the maximum eggs were found started mid March to at the end of April (16.14 - 22.62 eggs/plant). In general, the period of peak activities of the *T. absoluta* compromised with vegetative time with flowering and early fruit setting stages of the crop. As a result, large populations, consisting larvae damage the crop leaves at vegetative and then passed to fruit. In 2015/16, patterns of abundances were similar to those of 2016/17. However, in 2016/17 at Ambo and Toke kutaye first and second cropping cycle the population of *T. absoluta* eggs, larvae and pupae were low as compared to Dandi district.

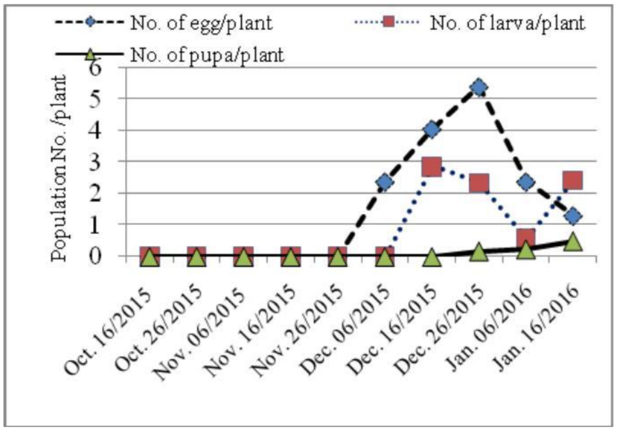


Fig. 3.6a: Mean population number of *T. absoluta* on tomato at Ambo district during 2015/16 1st year 1st Cropping Cycle on open field

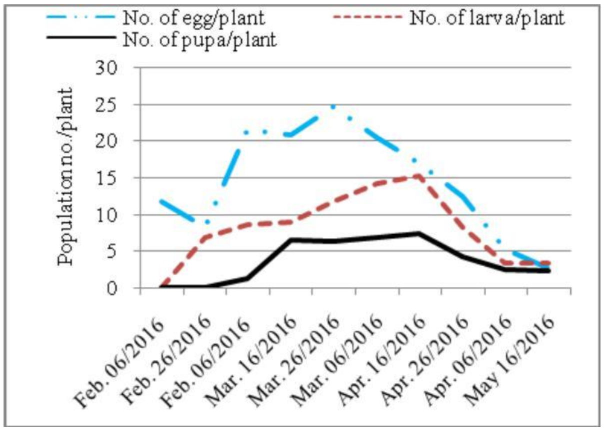


Fig. 3.6b: Mean population number of *T. absoluta* on tomato at Ambo district during 2016 1st year 2nd Cropping Cycle on open field

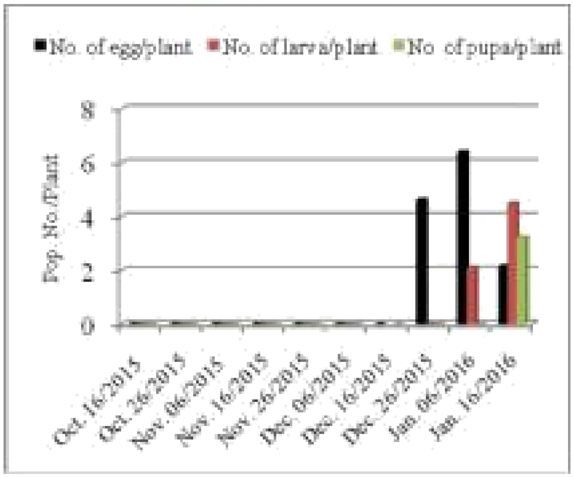


Fig. 3.6c: Mean population number of *T. absoluta* on tomato at Ambo district during 2016/17 2nd year 1st Cropping Cycle on open field

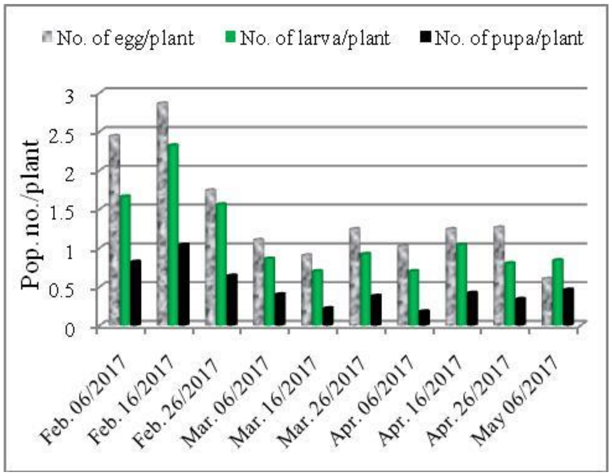


Fig. 3.6d: Mean population number of *T. absoluta* on tomato at Ambo district during 2017 2nd year 2nd Cropping Cycle on open field

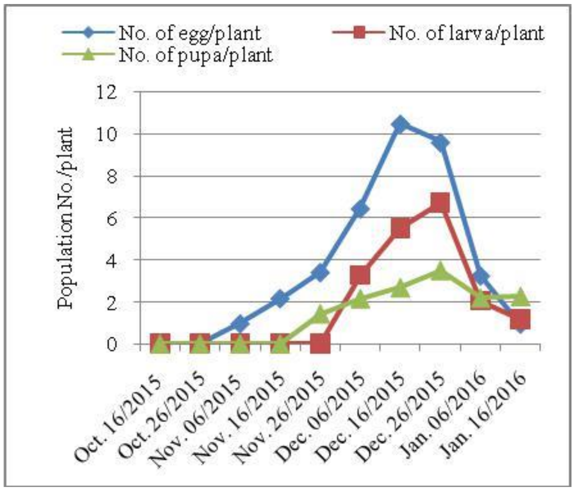


Fig. 3.7a: Mean population number of *T. absoluta* on tomato at Dendi district during 2015/16 1st year 1st Cropping Cycle on open field

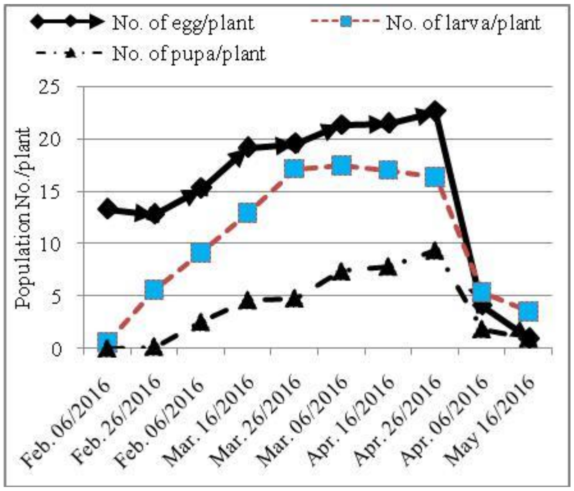
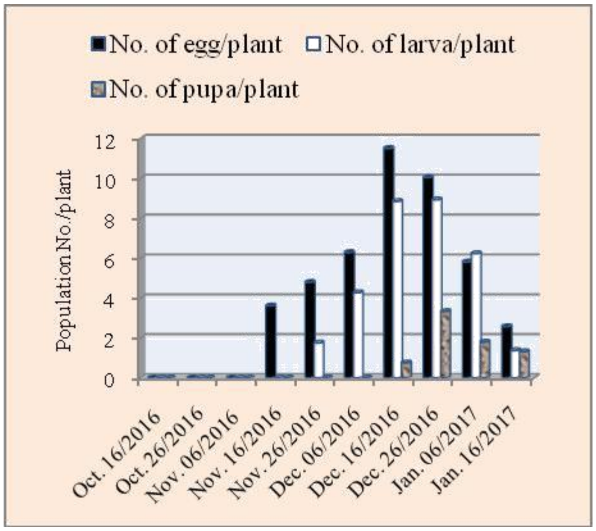


Fig. 3.7b: Mean population number of *T. absoluta* on tomato at Dendi district during 2016 1st year 2nd Cropping Cycle on open field



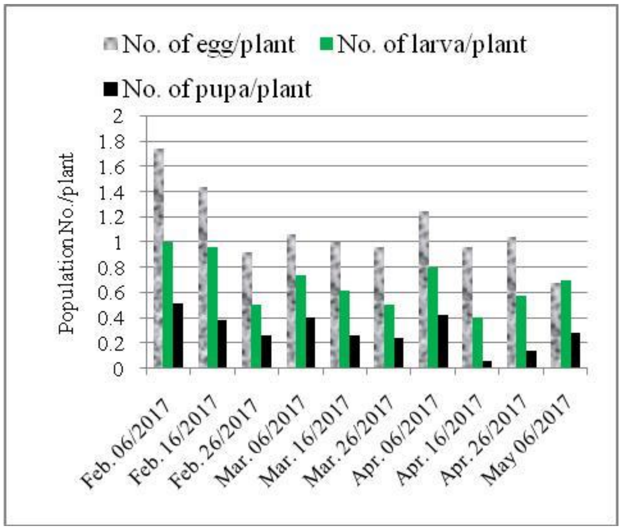
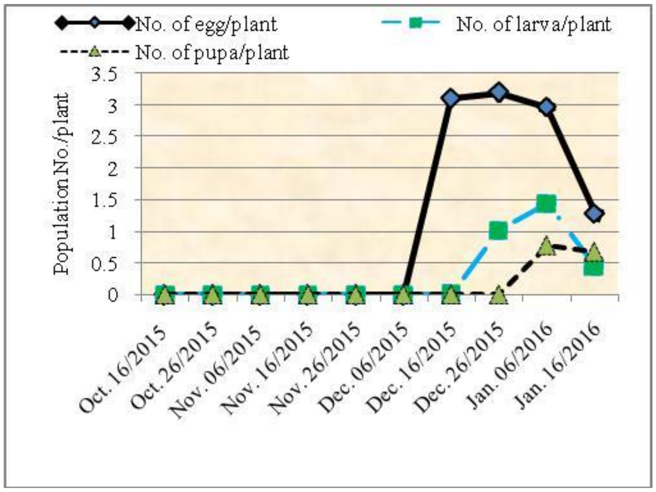


Fig. 3.7c: Mean population number of *T. absoluta* on tomato at Dendi district during 2016/17 1st year 1st Cropping Cycle on open field

Fig. 3.7d: Mean population number of *T. absoluta* on tomato at Dendi district during 2017 1st year 2nd Cropping Cycle on open field



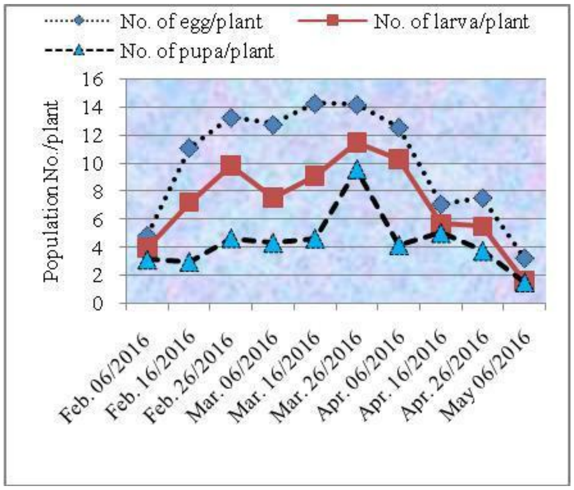


Fig. 3.8a: Mean population number of *T. absoluta* on tomato at Toke kutaye district during 2015/16 (1st year 1st Cropping Cycle) on open field

Fig. 3.8b: Mean population number of *T. absoluta* on tomato at Toke kutaye district during 2016 (1st year 2nd Cropping Cycle) on open field

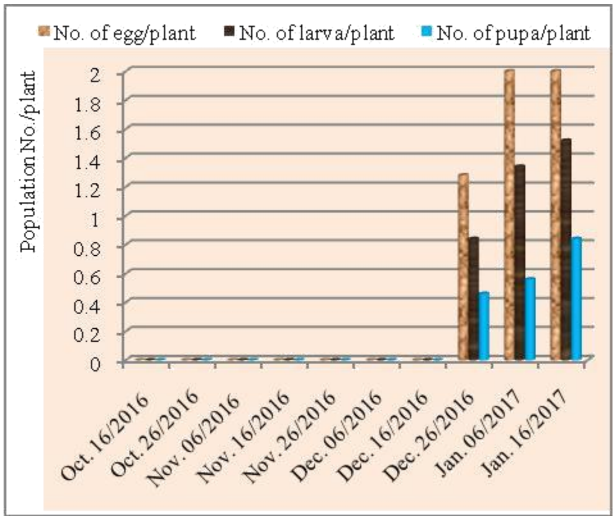


Fig. 3.8c: Mean population number of *T. absoluta* on tomato at Toke kutaye district during 2016/17 (2nd year 1st Cropping Cycle) on open field

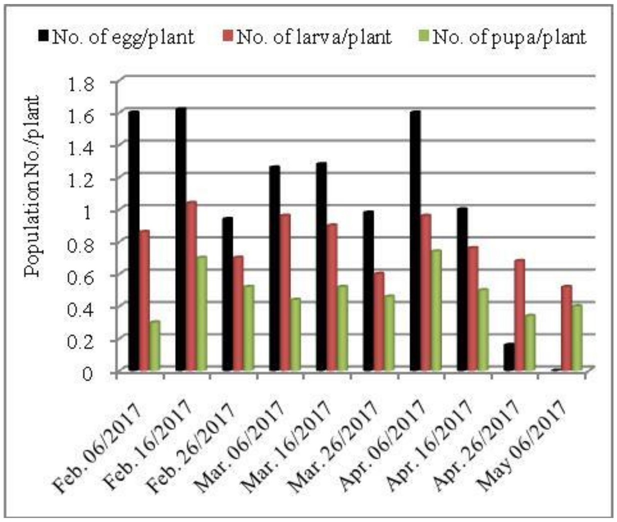


Fig.3. 8d: Mean population number of *T. absoluta* on tomato at Toke kutaye district during 2017 (2nd year 2nd Cropping Cycle) on open

3.4 Discussions

T. absoluta is a key pest of tomato crops in West Shewa of Ethiopia causing high losses. The knowledge of its population dynamics under laboratory, glasshouse and open field conditions is considered as major step to plan effective management strategies. The tomato leafminer, *T. absoluta* eggs number evaluation in the glasshouse study was different from that of field studies. The results showed that *T. absoluta* eggs and larval population number evolving at Ambo glasshouse was larger than that of all field studies. These numbers were influenced perhaps by abiotic factors like temperature, insecticide applications, and distance between glasshouses and by biotic factors i.e. absence of natural enemies. This result confirmed the previous studies of Miranda *et al.* (1998), they reported that during the first phenologic stages of the crop, tomato plants are free from attack of *Tuta absoluta*. The average temperature recorded at this period was approximately 20-25°C. Their number became relatively high, as their attack became intense towards at vegetative stage of crop cycle. These results matched with those found by several authors (Lacordaire and Feuvrier, 2010). These authors underlined the occurrence and increase in *T. absoluta* captures during the crop season.

Allache and Demnati (2014) mentioned that in Algeria, during the first phonologic stages of the crop, tomato plants are free from attack of *T. absoluta*. Harizanova *et al*. (2009) pointed that the leaves were the most heavily damaged plant parts. Leite *et al.* (2004) found that the attack of *T. absoluta* was severe at the end of growing season, these authors suggested removal of crop residues and rotating with crops that were not suitable host for this pest. *Tuta absoluta* females deposited their eggs on all plant parts they prefer laying eggs on leaves (Torres *et al.*, 2001; Faria *et al.*, 2008). Thus, their numbers were high on the upper than lower leaf surface (Leite *et al.*, 2004). In the present study, most of eggs deposited on upper leaves were taken into account. However, minimum eggs laying in October and May were recorded. The nutritional quality of tomato leaves (terpenes) seemed to have a positive effect on the laying behaviour of *T. absoluta* (Leite *et al.*, 2004). The number of eggs increased in the three study areas in the first and second year, second cropping cycles (from mid February to the end of the cropping cycle).

The number of eggs mentioned in this work was low compared with results of Pereyra and Sanchez (2006). After Miranda *et al.* (2005), natural enemies played an important role in controlling tomato pests and their preservation by farmers was necessary. *T. absoluta* was attacked at its various stages by several natural enemies, however the rate of parasitism was variable according to species (Marchiori *et al.*, 2004; Faria *et al.*, 2008). *T. absoluta* pupation was frequent in the leaves but the pupae might be found in the ground, mainstem and in fruit (Torres *et al.*, 2001).

In Ethiopia, *T. absoluta* resistance to pesticides was not studied. To manage this problem, using Integrated Pest Management (IPM) and other alternative approaches, reducing pesticides use and preserving natural enemies by growers might be a solution (Lietti *et al.*, 2005; Miranda *et al.*, 2005).

These studies were showed that at third croppinig cycle the populations of *T. absoluta* were very high due to the availability of the host throughout the year under glasshouse conditions. Similarly, in both years on the field studies at second cropping cycle the population number of *T. absoluta* in all stages were increased. Therefore, the populations of these insect pests are very high, because of the same trends. This study highlighted that the three field study areas and one glasshouse study lodged all *T. absoluta* developmental stages. Moreover, it was present during all tomato vegetative cycle and on all plant parts. Hence this finding agreed with the previous study by Oliveira *et al.* (2008), they stated, *T. absoluta* is not only to the intensity of its attack but also to its occurrence during all crop cycles.

3.5 Conclusions

In the second cropping cycle, damaged of tomatoes were high, apart from damage on tomatoes in the first cropping cycle. With tomatoes from the first cropping cycle in both years, the maximum and most evident damages were recorded on the leaves during vegetative stage. It is important to emphasize that with tomatoes from the first cropping cycle there were damages on the leaves and fruits of tomato. It was concluded that the infestation level of *T. absoluta* was exist throughout the year if host is available and mostly depends on phonological stages. From this study, damages of tomatoes from the second cropping cycle, damages on all parts of the plants were visible but specifically the leaf parts totally invaded before fruit setting particularly under the glasshouse conditions.

CHAPTER FOUR

Biology and host preference studies of *Tuta absoluta* on major solanaceous crops

4.1 Introduction

Tomato (*Lycopersicon esculentum* Miller) is important crops in Ethiopia. There are various insect pests are reported to attack tomatoes worldwide (Lange and Bronson, 1981). Some of the pest species are known to be of great economic importance, among of these species the tomato leaf miner, *Tuta absoluta* **(**Meyrick) (Lepidoptera: Gelechiidae) and the tomato fruit borer, *Helicoverpa armigera* (Hubner) (Lepidoptera: Noctuidae) are very serious insect pests of tomato plants.

Among the invasive pests, the tomato leafminer, *Tuta absoluta* (Lepidoptera: Gelechiidae), is a devastating pest that develops principally on solanaceous plants throughout South and Central America and Europe (Thomas *et al*., 2014). It is a very destructive and an oligophagous pest with a strong preference for tomato; it can also attack the aerial parts of potato, eggplant, tobacco and some solanaceous weeds (Notz, 1992). It is a multivoltine species, which rapidly develops in favorable environmental conditions, with overlapping life cycles (Guenaoui *et al*., 2010). The tomato leafminer, *T. absoluta* is one of the most devastating pests of tomato in Ethiopia. This pest was initially reported in Eastern Shewa of Ethiopia in early 2012 (Gashewbaza and Abiy, 2013), and has subsequently spread throughout the country. Since the time of its initial detection, the pest has caused serious damages to tomato in invaded areas, and it is currently considered a key tomato threat to Ethiopia tomato production areas. Tomato leaf miner, *T. absoluta* is a native devastating pest of South America, particularly to the tomato, *L. esculentum* (Desneux *et al*., 2010; Gontijo *et al*., 2013). It may be described as an intercontinental pest. Although *T. absoluta* is an endemic Neotropical pest, it has acquired a wider geographical distribution after its unintended introduction in other tomato production regions. *T. absoluta* is first detection in Europe in late 2006 then distributed to tomato growing areas of the country (Urbaneja *et al*., 2007).

Over the past decade, a substantial amount of research in South America has addressed a wide range of topics related to *T. absoluta* biology, ecology, impacts and management. Since its unintentional introduction from meditation region through Sudan to Ethiopia, this invasive pest has devastated indescribable thousands hectares of tomato crop. Larvae of this insect are known to feed on the leaf mesophyll tissue expanding mines, and fruits of the crop with subsequent reduction of the yield. It has high reproductive rate, where the female lays 260 eggs during its life (EPPO, 2005) on tomato plants. Few authors have studied the life tables of *T. absoluta* on tomato plants (Miranda *et al.* 1998; Pereyra and Sànchez, 2006; Aksu and Çıkman, 2014; Erdogan and Babaroglu, 2014; Berxolli and Shahini, 2017). It is important insect pest in Ethiopia when it was studied on its biology and host preferences. This work is the first in Ethiopia with the Ethiopian strains of *T. absoluta* reared on tomato plants under glasshouse and laboratoty. Hence, the present study was the biology of *T. absoluta* on tomato plants and oviposition preferences on the major solanaceae crops.

4.2. Materials and Methods

4.2.1. Biology of *T. absoluta* and host preferences

4.2.1.1. Biology of *T. absoluta* on tomato plants

The biology of *T. absoluta* on tomato was studied under laboratory and glasshouse temperature; 20.5±2°C and 55±5% R.H. in the laboratory and 32.0±2°C and 40±5% R.H. in the glasshouse. *T. absoluta* larvae were collected from the fields and brought to the laboratory and glasshouse. The tomato leaf miner larvae present on these collected tomato leaves were wrapped with wet cotton kept in plastic box (20 x 15 cm^2) in the laboratory and glasshouse. After the emergence of the adults, cages were prepared under glasshouse and laboratory. Newly emerged adults were collected from the ovipositing female of laboratory culture and placed on the tomato plant in the cages.

Adult *T. absoluta* used in this study were obtained from the culture maintained in the laboratory with temperature and relative humidity, under the natural light phase. Ten (10) pairs of *T. absoluta* from this stock culture were sexed and released into a cage. *T. absoluta* were maintained in the glasshouse. A mixture of sugar, yeast and water was placed in a Petri dish as a food supplement in the cage for adult *T. absoluta*. Temperature and relative humidity were recorded daily starting from egg hatching to adult emergency. Biological parameters of *T. absoluta* such as number of eggs laid, number of larvae and number of puape were recorded daily. Moreover, sex ratio, lonmgevity and oviposition periods were recorded.

4.2.1.1.1. Life table construction

Observations on number of alive and mortality of *T. absoluta* on all stages were recorded daily. The following assumptions were used in the construction of life-table of *T. absoluta*.

$$q_x = [d_x / l_x] \text{ x } 100$$

Where:

x = Age of the insect in days.

l_x = Number surviving at the beginning of each interval

d_x = Number dying during the age interval

q_x = Mortality rate at the age interval x and calculated by using formula

$$e_x = T_x / l_x$$

e_x = Expectation of mean life remaining for individuals of age x

Life expectation was calculated using the equation

$L_x = l_x + 1 (x + 1)/ 2$

To obtain e_x two other parameters L_x and T_x were also computed as below.

L_x = The number of individuals alive between age x and x + 1 and calculated by the equation.

$T_x = l_x + (l_x + 1) + (l_x + 2)$………………..+ l_w.

Where: l_w = The last age interval.

T_x = The total number of individual of x age units beyond the age x and obtained by the equation

K-value = the difference number of $\log l_x$

4.2.1.1.2. Developmental stages of specific life-table

Data on stage specific survival and mortality of eggs, larvae, pupae and adults of *T. absoluta* were recorded from the age specific life-table.

x = Stage of the *T. absoluta*.

l_x= Number surviving at the beginning of the stage x.

d_x = Mortality during the stage indicated in the column x.

The data calculated through above assumptions for computing various life parameters such as Apparent Mortality (A_m), Survival Fraction (S_f), Mortality Survivor Ratio (MSR), Indispensable Mortality (I_M) and K-values were calculated according to Arshad and Parvez (2010).

4.2.2. Host preferences

This study was carried out for two consecutive years using three major Solanaceae crops (tomato, potato and pepper) varieties against *T. absoluta*. Seeds of the crops were obtained from Melkasa Agricultural Research Center. The experiment was laid out in Randomized Complete Block Design (RCBD) with three replications. The plants were transplanted into the experimental glasshouse after 40 days. The experimental pots, 20cm diameter and 25cm height totally 36 pots were prepared and filled with compost, loam soil and sand soil in the ratio of 1:1:2, respectively. All agronomic practices were performed as required and recommended.

4.2.2.1. Data Collection

In this study, different parameters were evaluated on sample plants in each treatment. Data were collected randomly from each variety/line of each replication. Morphological characters such as number of leaves per plant and fruit number per plant were recorded at 30, 50 and 70 days after transplanting (Khanam *et al*., 2003). Total number of infested leaves, healthy leaves and fruits were counted and percent infestations of *T. absoluta* was calculated according to the method of Mukhopadhyay and Mandal (1994).

4.2.2.2. Data analysis

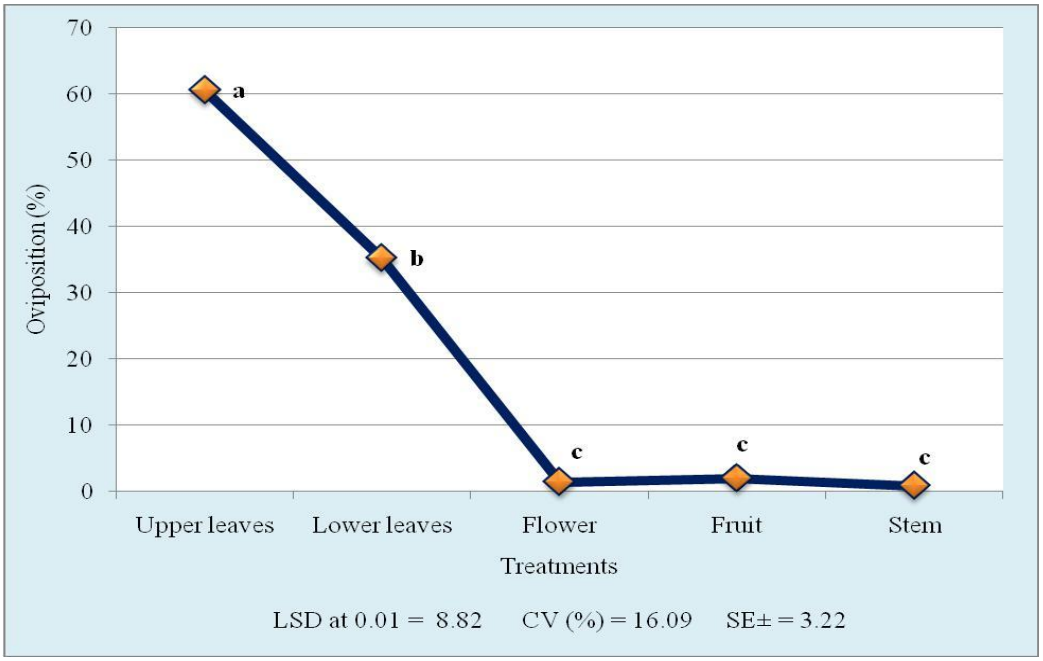
All the necessary data were recorded and analyzed with calculated means of days. Data collected was subjected to Analysis of Variance (ANOVA) in order to determine the significance differences among treatment SAS Programme version 9.1 (SAS, 2009). Life table analyses were calculated according to Jackknife method (Sokal and Rohlf, 1995).

4.3. Results

4.3.1. Biology of *T. absoluta*

4.3.1.1. Eggs laying positions of *T. absoluta* on tomato

The results showed that oviposition preferences were significantly ($P < 0.05$) differences from the other. All adult female *T. absoluta* was laid their eggs on upper side of the leaves (60.56%) followed by lower side of the leaves (35.21%) and lowest egg laying position was recorded on stem (0.85%) followed by flowers (1.41%) and fruit (1.97%) of tomato plants (Fig. 4.1).



Note: Means with the same letter is not significantly different form each other
All treatment effects were highly significant at $P < 0.01$ (DMRT).

Figure 4.1: Oviposition prfereance of *T. absoluta* on tomato plants under glasshouse condition

4.3.1.2 Developmental stage of *T. absoluta*

Significant ($P < 0.05$) differences were observed between laboratory and glasshouse studies on the life cycle of *T. absoluta* in both seasons. The female, *T. absoluta* is lay it their eggs ranged 177.5/female at temperature of 32 ± 2^{o}C (40±5% R.H.) and 233.75 eggs/female at 20.5 ± 2^{o}C (55±5% R.H.) during the life span in 2015. Similarly, in 2016 the female *T. absoluta* was laid their eggs

211.25 and 168.25/female at temperature of 20.5±2^{o}C (55±5% R.H.) and at 32±2^{o}C (40±5% R.H.), respectively. Most of the eggs were embedded on the upper and lower leaves.

The average eggs hatched to the first instar larva at 20.5±2^{o}C (55±5% R.H.) it was taken 13 to 13.5 days while at 32±2^{o}C (40±5% R.H.) it was took 10 to 10.5 days. A total of larval instars were observed in this study was taken 12.5 days at 20.5±2^{o}C (55±5% R.H.) and 11 to 11.51 days in the laboratory at 32±2^{o}C (40±5% R.H.) in the glasshouse, respectively. The developmental period of pupa ranged 8.2–9.8 days at temperature of 20.5±2^{o}C (55±5% R.H.) and 6.5–8.6 days at 32±2^{o}C (40±5% R.H.).

Table 4.1: Some biological parameters of *T. absoluta* studied under labotatory and glasshouse conditions during 2015.

Mean temperature and r.h.	Mean number of egg laid per female	Mean Complete life cycle (days)	Mean Life expectancy (days)	
			Male	Female
20.5±2^{o}C (55±5% r.h.)	233.75±14.42^{a}	30.6±0.59^{a}	9.0±0.30^{a}	18.40 ±1.45^{a}
32±2^{o}C (40±5% r.h.)	177.5±9.26^{b}	27.8±0.57^{a}	6.8±0.27^{b}	15.2±1.40^{b}
LSD at 0.01	26.64	6.25	1.04	2.69
CV (%)	5.76	7.49	7.49	9.12
SE ±	11.84	0.59	0.39	1.53

Note: Means with the same letter(s) in column are not significantly different for each other.
All treatment effects were highly significant at P < 0.01 (DMRT).

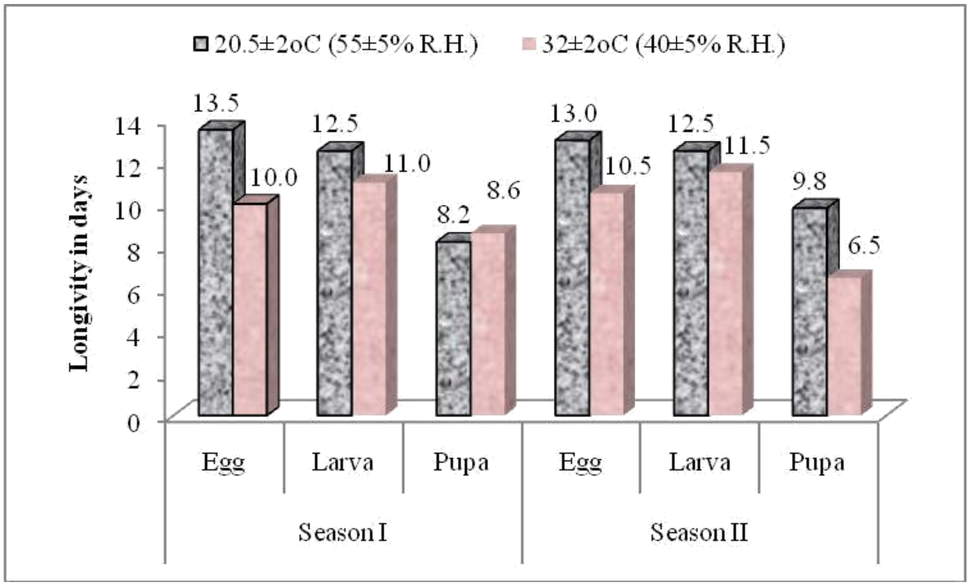


Figure 4.2: Mean developmental stages of *T. absoluta* at different temperature and relative humidity under laboratory and glasshouse condition during 2015-2016.

The mean developmental period for *T. absoluta* from egg to adult was taken 30.6 to 31.2 days at 20.5±2^{O}C (55±5% R.H.) while at 32±2^{O}C (40±5% R.H.) ranged from 26.4 to 27.8 days under glasshouse conditions (Table 4.1 & 4.2).

Table 4.2: Some biological parameters of *T. absoluta* studied under labotatory and glasshouse conditions during 2016.

Mean temperature in ^{O}C)	Mean number of egg laid per female	Mean Complete life cycle	Mean Life span in days	
			Male	Female
20.5±2^{O}C (55±5% r.h.)	211.25 ± 11.37^{a}	31.2 ± 1.87^{a}	8.2 ± 0.91^{a}	16.0 ± 1.04^{a}
32±2^{O}C (40±5% r.h.)	168.25 ± 9.07^{b}	26.4 ± 1.32^{a}	6.2 ± 0.68^{b}	14.4 ± 0.87^{b}
LSD at 0.01	22.54	4.92	1.52	0.68
CV (%)	5.28	9.73	12.60	2.55
SE ±	10.02	2.80	0.87	0.39

Note: Means with the same letter(s) in columns are not significantly different for each other.
All treatment effects were highly significant at $P < 0.01$ (DMRT)

4.3.1.3. Age specific developmental stages

Mortality percentage of eggs was recorded during life cycle 1.40% at 20.5±2^{O}C (55±5% R.H.) and 4.93% at 32±2^{O}C (40±5% R.H.). The highest larval mortalities (26.49%) were recorded at 30.5±2^{O}C

(40±5% R.H.), while the lowest mortalities (15.99%) were at 20.5±2^{O}C (55±5% R.H.) under glasshouse and laboratory, respectively. The different results were recorded the pupal mortality rate during the study periods under laboratory (10.23%) and glasshouse (32.42%) conditions, respectively. This study showed that at high temperature the mortality percentages of all stages were increased during developmental periods of *T. absoluta* under both laboratory and glasshouse conditions.

Significant ($P < 0.05$) differences between treatments of survival percent of eggs were recorded during the life cycle from eggs to larval stage. It was, 143 eggs hatched among of these 137 eggs were emerged to first instar larvae (95.8%) and 6 eggs were dead due to unknown reasons. There were no significant ($P > 0.05$) differences between treatments regarding to larval survival in both conditions. Among 137 larvae hatched, 124 were emerged to pupae (81.01%) was survived and also from hatched pupae only 109 was emerged to adults (89.77) at 20.5±2^{O}C (55±5% R.H.). On the other hand, at the same time 142 eggs were collected and hatched under glasshouse, at 32±2^{O}C (40±5% R.H.), among hatched eggs 119 were emerged to larvae (83.80%). From 119 hatched larvae 92 were emerged to pupae (81.51%). Finally, among hatched pupae 72 adult *T. absoluta* were emerged to Adults (78.26%) during 2015-2016 (Table 4.3).

Table 4.3: Mean survival percent of *T. absoluta* during the life cycle under laboratory and glasshouse temperature and relative humidity during 2015-2016.

Treatments	Mean Survival (%)			Mean mortality (%)		
	Egg	Larva	Pupa	Egg	Larva	Pupa
20.5±2^{O}C (55±5% R.H.)	95.80^{a}	81.01^{a}	89.77^{a}	4.20	19.99	10.23
32±2^{O}C (40±5% R.H.)	83.80^{b}	81.51^{a}	78.26^{b}	16.20	18.49	21.74
LSD	2.74	19.83	3.39			
CV (%)	0.87	6.51	1.16			
SE ±	0.78	5.65	0.96			

Note: Means with the same letter(s) in columns are not significantly different for each other.
All treatment effects were highly significant at $P < 0.01$ (DMRT)

4.3.1.4. Apparent Mortality (A_M)

The maximum apparent mortality was observed in fourth instar larvae (11.68%) and minimum was recorded in second instar larvae (0.72) at 20.5±2°C (55±5% R.H.) while the maximum apparent mortality was observed in last pupal stage (21.74%) and the minimum was recorded in third instar larvae (1.65%) at 32±2°C (40±5% R.H.) during 2015-2016. When a comparison was made between larval instars, the highest mortality (11.68%) was observed at fourth instar, whereas, minimum mortality was recorded at second instar (0.72) at 20.5±2°C. Similarly, the apparent mortality at pre pupa and pupal stages remained minimum 7.03 and 3.23%, at 20.5±2°C, respectively. On similar way, at 32±2°C (40±5% R.H.) maximum was recorded 10.68% and minimum was 2.17%, respectively (Tables 4.4 and 4.5). In this study finding, the early pre pupae were higher than the later pupae and hence, showed higher mortality at pre pupa stage was observed at 20.5±2°C (55±5% R.H.). However, in glasshouse experiment 32±2°C (40±5% R.H.), the highest mortality of *T. absoluta* was observed at last pupal stage.

4.3.1.5. Survival Fraction (S_x)

In tables 8 and 9 indicated that survival fraction was found maximum (0.99), at egg stage to third instar larvae, at 20.5±2°C and minimum (0.88) at 30±2°C. Among larval instars, the S_x remained highest (0.99) and lowest (0.88) at pupal stage at 20.5±2°C (55±5% R.H.). On the other hand, at 32±2°C (40±5% R.H.) the maximum S_x (0.98) was obtained and the minimum was found at pupal stage (0.78).

4.3.1.6. Mortality Survivor Ratio (MSR)

Mortality survival ratio (MSR) was found at egg stage minimum (0.014) at 20.5±2°C (55±5% R.H.) and maximum (0.05) at 32±2°C (40±5% R.H.). Mortality survival ratio of larval instar also found at

20.5±2°C (55±5%) which indicated that (0.007) at second and third instar larvae and (0.117) was found at fourth instar larvae. While at 32±2°C (40±5% R.H.) the minimum larval instar was (0.02) at third instar larvae and (0.13) at fourth instar larvae. Between the pre pupa and pupa, the maximum mortality survival ratio (0.23) was observed at pre pupa and the minimum mortality survival (0.11) at 20.5±2°C (55±5%). On the other hand, the minimum (0.032) ratio was obtained at last pupal stage and (0.07) at pre pupal stage was examined at 32±2°C (40±5% R.H.). In this study, *T. absoluta* showed high natural mortality during its life cycle at larval stage and pupa are known to be the most critical.

4.3.1.7. Indispensable Mortality (I_M)

Indispensable mortality at egg stage was observed maximum (12.75) at fourth larval instar and the minimum results were recorded (0.76) at second and third larval instars at 20.5±2°C (55±5% R.H.). While comparing indispensable mortality between pupal stages, the maximum (7.63) and the minimum (3.49) were observed in similar temperature and relative humidity. At 32±2°C (40±5% R.H.) the lowest value (1.44) was recorded at third instar and the maximum value was observed (16.56) at pupal stage (Tables 4.4 and 4.5).

4.3.1.8. K-values

The k-value was found minimum (0.0061) and maximum (0.0487) in 20.5±2°C (55±5% R.H.) and 32±2°C (40±5% R.H.) at egg stage, respectively. While comparing larval instars, it revealed highest ‘k’ (0.0295) at fourth instar and lowest (0.0030) at third instar larvae at 20.5±2°C (55±5% R.H.). In case of pupal stage in both conditions the highest k-value was recorded at last pupal stage (Table 3 and 4). The total generation mortality k-value was recorded maximum (0.3980) at 32±2°C (40±5% R.H.) and minimum (0.1178) at 20.5±2°C (55±5% R.H.).

4.3.1.9. Adult Longevity of *T. absoluta*

Results from this study revealed that there was a significant ($P < 0.05$) difference in longevity of *T. absoluta* between the treatments. There was no significant ($P > 0.05$) differences in longevity when female *T. absoluta* lived (17.2) at mean temperature and relative humidity of $20.5 \pm 2^{O}C$ (55±5% R.H.) while the male *T. absoluta* lived (8.9) days as an average of two seasons. On the other hand at temperature and relative humidity, $32 \pm 2^{O}C$ (40±5% R.H.) in the glasshouse male and female *T. absoluta* were lived 6.5 and 14.8 days, respectively (Tables 4.1 and 4.2).

The two season results of pupal developmental stages revealed that it was taken 8.2 to 9 days at $20.5 \pm 2^{O}C$ (55±5% R.H.) and 6.5 to 8.6 at $32 \pm 2^{O}C$ (40±5% R.H.). The means of development period from egg to adult was 30.18 days. But our findings showed that at $20.5 \pm 2^{O}C$ (55±5% R.H.) 30.6–31.2 days and at $32 \pm 2^{O}C$ (40±5% R.H.) 26.4 - 27.8 days. The females lived longer than males, allowing them to be sexually mature when the males emerge.

Table 4.4: Stage specific life table of *T. absoluta* on tomato plant at 20.5±2°C (55±5% R.H.) during 2015-2016.

Stage x	No. surviving at the beginning of the stage l_x	No. dying in each stage d_x	Apparent mortality $143q_x$	Survival fraction S_x	Mortality survivor ratio (MSR)	Indispensable mortality I_M	log l_x	K-values
Egg	143	2	1.40	0.99	0.014	1.53	2.1553	0.0061
First instar	141	2	1.42	0.99	0.013	1.42	2.1492	0.0062
Second instar	139	1	0.72	0.99	0.007	0.76	2.1430	0.0033
Third instar	138	1	2.17	0.99	0.007	0.76	2.1397	0.0030
Fourth instar	137	16	11.68	0.93	0.117	12.75	2.1367	0.0295
Pre pupa	128	9	7.03	0.97	0.070	7.63	2.1072	0.0137
Pupa	124	4	3.23	0.88	0.032	3.49	2.0935	0.0560
Adult	109	---	---	---	---	---	2.0375	---
								K=0.1178

Table 4.5: Stage specific life table of *T. absoluta* on tomato plant at 32±2°C (40±5% R.H.) during 2015-2016

Stage x	No. surviving at the beginning of the stage l_x	No. dying in each stage d_x	Apparent mortality $100q_x$	Survival fraction S_x	Mortality survivor ratio (MSR)	Indispensable mortality I_M	log lx	K-values
Egg	142	7	4.93	0.95	0.05	3.60	2.1523	0.0218
First instar	135	9	6.67	0.94	0.07	5.04	2.1305	0.0269
Second instar	127	6	4.72	0.95	0.05	3.60	2.1036	0.0208
Third instar	121	2	1.65	0.98	0.02	1.44	2.0828	0.073
Fourth instar	119	16	13.45	0.87	0.13	9.36	2.0755	0.0629
Pre pupa	103	11	10.68	0.89	0.11	7.92	2.0126	0.0623
Pupa	92	20	21.74	0.78	0.23	16.56	1.9637	0.1062
Adult	72	---	---	---	---	---	1.8575	---
								K= 0.3980

4.3.1.10 Sex ratio

There are 100 eggs were collected and hatched to determined male to female sex ratio under both laboratory and glasshouse conditions. Among hatched eggs 48 male and 32 females were emerged at 20.5±2^{o}C (55±5% R.H.) in the laboratory while the same number of eggs were hatched at 32±2^{o}C (40±5% R.H.) in the glasshouse, among of these 40 male and 36 female adults were emerged from pupae. The total survival of *T. absoluta* adults in the laboratory 80% (3:2) male to female sex ratio and although in the glasshouse the total survival of adults less than in the laboratory it was showed that 76% adults were survived 40 male and 36 female (10:9) male to female sex ratio recorded. In this finding, it was stated that usually males were more than female individuals.

4.3.2 Host preferences and Infestation level study of *T. absoluta*

4.3.2.1 Oviposition preferences of *T. absoluta* on major solanaceous crops

In this study, the tomato was the primary host for *T. absoluta* in both 2015/2016 and 2016/2017 (Table 4.6). On tomato plant, more *T. absoluta* eggs were laid rather than potato and pepper during the study periods. Eggs were almost found to be laid singly on the upper and lower side of the leaf of tomato and potato crops. On tomato varieties percent oviposition on upper leaves ranged from 62.23 to 67.24% and on lower leaves ranged from 32.01 to 36.80% were recorded during 2015/16 while during 2016/17 it was observed that on upper leaves of all varieties of tomato ranged from 63.96 to 68.09% and on lower leaves from 30.82 to 34.50% were recorded. On the other hands, percent ovipostion of *T. absoluta* on potato showed on Table 3, it was recorded that from 23.33 to 35.15% on upper leaves and 63.69 to 75.75% on lower leaves during 2015/16 and ranged from 23.21 to 40.08% on upper leaves and 56.49 to 76.80% on lower leaves of all varieties of potato crops. Very few number of eggs were laid on stem, buds, flowers and calyxes of the fruits of all varieties of tomato.

Larvae were generally found feeding on leaves, creating wide mines giving it a papery appearance and tunneling through the fruits. They were also found boring into the apical buds and the stems. Infested fruits had small hole closer to the calyxes all varieties of tomato. Finally, we observed that *T. absoluta* oviposition preferentially occurs on the upper leaf of tomato and lower leaf of potato plants where highest densities of trichomes are found.

Table 4.6: Mean oviposition of *T. absoluta* on major solanaceous crops during 2015-2017

	Eggs laid by females/plant							
	2015/16				2016/17			
Treatments	Upper leaf	Lower leaf	Stem	Flower	Upper leaf	Lower leaf	Stem	Flower
Koshoro	62.23^{bc}	36.80^{c}	0.48	0.50	67.35^{a}	31.93^{c}	0.33	0.33
VF-Roman	66.39^{ab}	35.97^{c}	0.49	0.23	63.96^{a}	34.35^{c}	0.53	2.17
Galila	60.88^{c}	35.41^{c}	0.27	0.68	68.09^{a}	30.82^{c}	1.09	0.0
Local var.	67.24^{a}	32.01^{c}	0.36	0.36	64.51^{a}	34.50^{c}	1.0	0.0
Jalane	23.33^{e}	73.89^{a}	2.78	0.0	31.75^{d}	56.49^{b}	1.75	0.0
Tolcha	26.84^{e}	71.31^{a}	1.85	0.0	37.11^{c}	57.48^{b}	5.41	0.0
Menagesha	35.15^{d}	63.69^{b}	1.15	0.0	40.08^{c}	59.92^{b}	0.0	0.0
Local var.	23.96^{e}	75.75^{a}	0.0	0.0	23.21^{e}	76.80^{a}	0.0	0.0
M. Awaze	0.0^{f}	0.0^{d}	0.0	0.0	0.0^{f}	0.0^{d}	0.0	0.0
M. Fana	0.0^{f}	0.0^{d}	0.0	0.0	0.0^{f}	0.0^{d}	0.0	0.0
M. Zala	0.0^{f}	0.0^{d}	0.0	0.0	0.0^{f}	0.0^{d}	0.0	0.0
Local var.	0.0^{f}	0.0^{d}	0.0	0.0	0.0^{f}	0.0^{d}	0.0	0.0
LSD	4.90	5.73			6.44	11.91		
CV (%)	9.48	9.56			11.38	21.80		
SE±	2.89	3.39			3.80	7.03		

Note: Means with the same letter(s) in columns are not significantly different for each other.

All treatment effects were highly significant at $P < 0.01$ (DMRT)

4.3.2.2 Host Plant Susceptibility and Infestation Level

The data presented on table 4.7 regarding to larval population of leaf infestation per plant on major solanaceae crop genotypes of tomato, potato and pepper. The data, on the larval population per plant, leaf infestation and fruit damaged were recorded. The analysis of variance revealed that a significant ($P < 0.05$) differences among the crops and non significant ($P > 0.05$) differences were observed among varieties of each crop. The results indicated that the maximum leaf infestation was recorded to be 88.14% per plant during 2015/16 and 92.08% during 2016/17 were observed on tomato varieties. Regarding the fruit damaged also no significance differences were recorded among each variety. They were recorded 81.11 to 97.67% during 2015/17 and 56.06 to 82.22% during 2015/17 (Table 4.7).

On potato varieties, Jalane, Menagesha, Tolcha and Local var. were found to have tolerance response and did not showed significant ($P > 0.05$) differences with one another. The results indicated that the leaf infestation percentage ranging from 16.17% to 24.25% for both years (Table 4.7). The later mentioned on the pepper varieties showed non leaf and fruit infestation and non significant differences with those of pepper varieties.

Table 4.7: Infestation level of *T. absoluta* on different days of observation on major solanaceous crop varieties under glasshouse conditions during 2015-2017

Trts	Year I (2015/16)				Year I (2016/17)			
	Leaf infested (%)			Fruit damaged (%)	Leaf infested (%)			Fruit damaged (%)
	30 days	**50 days**	**70 days**	**90 days**	**30 days**	**50 days**	**70 days**	**90 days**
Koshoro	31.88^{b}	66.62^{b}	88.14^{a}	87.50^{a}	25.98^{d}	33.29^{c}	87.68^{a}	81.48^{a}
Roman-VF	29.53^{b}	76.20^{b}	85.75^{a}	83.81^{a}	47.60^{b}	82.14^{a}	82.96^{a}	56.06^{a}
Galila	29.53^{a}	76.40^{b}	87.78^{a}	81.11^{a}	66.09^{a}	82.55^{a}	92.08^{a}	76.40^{a}
Local var.	36.13^{a}	82.18^{a}	87.15^{a}	97.67^{a}	33.82^{c}	70.74^{b}	90.82^{a}	82.22^{a}
Jalane	9.23^{c}	21.24^{c}	24.19^{b}	---	7.73^{ef}	19.33^{c}	24.25^{b}	---
Tolcha	14.42^{c}	21.94^{c}	23.87^{b}	---	8.89^{ef}	18.62^{d}	23.50^{b}	---
Menagesha	9.87^{c}	17.12^{c}	22.80^{b}	---	6.35^{f}	15.27^{d}	24.43^{b}	---
Local var.	12.14^{c}	16.06^{c}	16.17^{b}	---	12.97^{e}	17.0^{d}	20.12^{b}	---
M. Awaze	0.0^{d}	0.0^{d}	0.0^{c}	---	0.0^{g}	0.0^{e}	0.0^{c}	---
M. Fana	0.0^{d}	0.0^{d}	0.0^{c}	---	0.0^{g}	0.0^{e}	0.0^{c}	---
M. Zala	0.0^{d}	0.0^{d}	0.0^{c}	---	0.0^{g}	0.0^{e}	0.0^{c}	---
Local var.	0.0^{d}	0.0^{d}	0.0^{c}	---	0.0^{g}	0.0^{e}	0.0^{c}	---
LSD	5.34	13.03	9.13	35.86	5.31	9.37	13.86	35.29
SE±	3.15	7.69	5.39	17.95	3.14	5.53	8.18	17.66
CV (%)	21.90	24.43	14.84	20.87	17.97	19.54	22.07	23.85

Note: Means with the same letter(s) in columns are not significantly different for each other.
All treatment effects were highly significant at $P < 0.01$ (DMRT)

4.3.2.3 Mean population trends at various dates of observations

In both years, the mean population tendencies of *T. absoluta* on tomato crops were observed. The egg populations were low during the early months of December 15th and high during the end of February. Then it becomes decline gradually starting from the end of February to early march. Again at mid of March it becomes gradually increased till mid April and then decline slowly to at the end of May it becomes very low. In contrast, the population tendencies of *T. absoluta* on potato varieties increase gradually from mid January till at the end of May 29th. These results indicated that during mid months of December the tomato plants were at vegetative and flowering stages and then matured the ovipositions were declined and the insects were move to potato plants. There was no significance differences were observed among each tomato varieties as well as among each potato varieties. On the other hands, *T. absoluta* did not prefer all pepper varieties.

Table 4.8: Mean population tendency on major solanaceous crops at different months of observations under glasshouse conditions during 2015-2017

Mean number of *T. absoluta* population tendency on different stages at different Months of Observation on major solanaceae crops												
Months	Stages	Koshoro	VF-Roman	Galila	Local var.	Jalane	Tolcha	Mena gesha	Local var.	M. Awaze	M. Fana	M. Zala	Local var.
December 15th	E	1.75	3.25	8.75	5.25	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
	L	1.25	3.0	6.25	4.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
	P	1.0	2.50	5.75	2.75	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
December 30th	E	8.25	10.75	19.25	13.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
	L	6.75	9.25	15.25	9.25	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
	P	5.25	7.25	11.25	5.75	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
January 14th	E	19.50	26.50	33.75	29.25	3.75	2.50	5.25	2.50	0.0	0.0	0.0	0.0
	L	17.25	20.25	22.50	20.50	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
	P	14.25	13.25	12.75	16.25	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
January 29th	E	22.50	25.25	19.75	32.0	2.25	6.0	4.25	8.50	0.0	0.0	0.0	0.0
	L	14.25	17.75	13.50	26.25	2.0	5.0	3.25	5.0	0.0	0.0	0.0	0.0
	P	10.25	9.75	11.0	19.75	0.0	3.75	2.75	2.25	0.0	0.0	0.0	0.0
February 13th	E	20.67	28.50	23.25	24.25	8.25	12.50	13.25	10.75	0.0	0.0	0.0	0.0
	L	13.0	18.25	14.75	20.25	3.75	5.75	7.25	6.75	0.0	0.0	0.0	0.0
	P	10.75	10.0	7.25	13.75	1.25	2.50	3.75	3.25	0.0	0.0	0.0	0.0
February 28th	E	24.25	32.25	35.0	27.25	10.75	15.25	8.50	16.25	0.0	0.0	0.0	0.0
	L	14.75	18.25	18.25	22.0	7.0	9.50	3.25	9.25	0.0	0.0	0.0	0.0

	P	6.50	8.75	7.50	14.75	3.25	4.75	1.25	4.75	0.0	0.0	0.0	0.0
March 15th	E	8.25	9.75	13.25	12.75	7.25	19.50	14.25	10.75	0.0	0.0	0.0	0.0
	L	11.75	18.25	22.25	18.25	12.50	8.25	9.50	6.00	0.0	0.0	0.0	0.0
	P	6.50	11.50	14.50	12.0	5.75	4.25	6.25	4.75	0.0	0.0	0.0	0.0
March 30th	E	16.25	22.75	18.25	19.0	26.25	32.25	29.50	25.75	0.0	0.0	0.0	0.0
	L	9.25	14.50	10.0	13.50	14.25	17.25	20.25	21.0	0.0	0.0	0.0	0.0
	P	5.50	4.75	5.25	9.75	9.0	8.50	10.0	14.25	0.0	0.0	0.0	0.0
April 14th	E	26.75	28.25	21.50	27.25	32.50	40.50	37.25	29.0	0.0	0.0	0.0	0.0
	L	14.0	15.75	17.0	22.50	14.25	24.25	18.25	25.75	0.0	0.0	0.0	0.0
	P	6.0	9.0	12.25	10.0	12.25	12.25	8.50	18.75	0.0	0.0	0.0	0.0
April 29th	E	17.25	19.75	13.25	16.25	38.0	37.75	42.50	35.50	0.0	0.0	0.0	0.0
	L	14.75	13.75	8.25	11.75	16.75	21.50	18.25	23.0	0.0	0.0	0.0	0.0
	P	8.75	6.0	5.75	6.25	6.0	15.0	8.25	14.0	0.0	0.0	0.0	0.0
May 14th	E	3.75	6.25	5.50	7.25	47.25	31.50	29.50	40.25	0.0	0.0	0.0	0.0
	L	0.0	2.50	3.0	3.25	21.75	15.50	17.25	33.25	0.0	0.0	0.0	0.0
	P	0.0	0.0	1.25	2.75	14.25	8.25	11.50	20.0	0.0	0.0	0.0	0.0
May 29th	E	2.25	4.50	0.0	3.0	40.75	36.25	31.75	48.0	0.0	0.0	0.0	0.0
	L	1.75	0.0	2. 50	2.25	28.25	19.75	22.25	22.25	0.0	0.0	0.0	0.0
	P	1.0	0.0	1.25	0.0	14.75	11.25	16.25	14.75	0.0	0.0	0.0	0.0

Note: E = No. of eggs/plant L = No. of larvae/plant P = No. of pupae/plant

4.4 Discussion

T. absoluta is a major insect pest of tomato crops in Ethiopia causing heavy infestation and yield losses. The knowledge of its biology and host preferences is considered as major steps to plan effective control strategies. This study results disagree with the previous work of Torres *et al*. (2001), they were reported that before flowering, *T. absoluta* females chose on the underside of the leaves for oviposition in the apical part of the plant canopy but we agreed with Leite *et al*. (2004) demonstrated that there was a preferential deposition of *T. absoluta* eggs on the apical upper side of the leaves of the tomato plant canopy. Furthermore, Leite *et al*. (1999) found that *T. absoluta* oviposited more on leaves of the apical and medium portions than in the basal parts of the tomato plant. According to different authors report, the preferential oviposition in the upper leaves are probably due to the fact that these leaves are more tender, as they have lower calcium content and percentage of leaf fibre insoluble in acid detergent as compared to the middle and lower leaves (Marschner, 1995; Silva *et al*., 1998; Leite *et al*., 1999a; Wei *et al*., 2000).

This finding was disagreed with the previous work of Erdogan and Babaroglu (2014), EPPO (2005) and Torrest *et al*. (2001) previous studies they found that the period of eggs hatching was between 4-5 days. Most of the eggs were embedded on the upper and lower side of the leaves. Similarly this study results supported by Uchoa-Fernandes *et al*. (1995) they stated that the oviposition period was 7 days after first mating with a maximum life time fecundity of 260 eggs per female. This present studies were confirmed with the work of Erdogan and Babaroglu (2014). They found that the total period of larvae instar was 10.97 days, Pereyra and Sanches (2006) also reported that the period of larvae instar of *T. absoluta* was 12.14 days at 25±1ºC. It was also determined that the period of larvae instar of *T. absoluta* was 13-15 days (EPPO, 2005). Torrest *et al.* (2001) who stated the period of larvae instar of *T. absoluta* was taken 12 and 16 days at 27ºC.

The developmental time of pupa ranged 8.2–9.8 days at temperature of 20.5±2^{O}C (55±5% R.H.) and 6.5–8.6 days at 32±2^{O}C (40±5% R.H.). Gadir *et al*. (2016) stated that the pupal development time of the tomato leafminer varied from 7.07 to 8.62 days on two tomato cultivars. It was found that the period of pupae instar of *T. absoluta* was 9.53 days Erdogan and Babaroglu (2014). Torres *et al.* (2001) also found the pupal developmental time of *T. absoluta* as 7–9 days, those studies are similar to our findings.

These results agreed with the findings of Erdogan and Babaroglu (2014) they found that means of developmental period from eggs to adults are 30.18 days. EPPO (2005) was reported under optimal conditions *T. absoluta* developed in about 30 days. Barriontes *et al.* (1998) was also found that average development time of *T absoluta* was 23.8 days at 27.1ºC. Cuthbertson (2011) reported that the development from egg to adult period taken 35 days at 25ºC. He also mentioned in the same year in England under greenhouse conditions, the mean total development time of *T. absoluta* was 39.8 days at 19.7 ^{O}C. Erdoghan and Babaroglu (2014) showed that the mean total development time of *T. absoluta* was 30.18 days on tomato at 25–26 ^{O}C. *T. absoluta* developed slightly faster in our studies compared to the results obtained by the other researchers which might be in part attributed to the tomato cultivar differences and possible differences in the population of *T. absoluta* in these results. According to Du *et al*. (2004) the developmental period of the herbivorous insects were strongly affected by the nutritional qualities of the host plant, which in turn influences its population growth. On the other hand, the chemical components of host plants can also affect survival, growth and reproduction of herbivorous insects (Wilson and Huffaker, 1976; Bernays and Chapman, 1994; Adebayo and Omoloyo, 2007).

The study was showed that the 'k' increased from pupa to pre-pupal stage, this indicated that during this stage the mortality of *T. absoluta* was high. Similarly, Erdogan and Babaroglu (2014) found that the survival rate of pupa was 63.10% i.e 36.9% mortality; it was very high as compared with these results. Mortality percentage of eggs were recorded during the life cycle 1.40% at 20.5±2^{O}C (55±5% R.H.) and 4.93% at 32±2^{O}C (40±5% R.H.). But Cuthbertson (2011) stated that the developmental stage, the survival of the egg stage was 100%, no mortality rate was recorded. But in this study mortalities were observed in all the developmental stages. In another experiment much lower survival of *T. absoluta* was recorded at 20-23°C (Miranda *et al*., 1998). In this study, *T. absoluta* showed high natural mortality during its life cycle at larval stage and pupa are known to be the most critical. Therefore, this study on the mortality performance of *T. absoluta* showed corroboration with Miranda *et al*. (1998).

This finding was closest to the previous studies of Estay (2000), who was observed that adult *T. absoluta* lifespan was ranged between 10 - 15 days for females and 6 -7 days for males. It was found that adult longevity for male and female individuals were 15.8 days and 18.16 days, respectively (Erdogan and Babaroglu, 2014). This study results agreed that the findings of Pereyra and Sanches (2006) who reported that the period of larvae instar of *T. absoluta* was 12.14 days at 25±1ºC. It was resolute that the developmental period of larvae of *T. absoluta* was 13–15 days, and found that the period of pupae of *T. absoluta* was as an average it was taken 9.53 days (EPPO, 2005). This result was similar to those reported by Torrest *et al.* (2001) who reported the developmental period of larval instar of *T. absoluta* was 12–16 days at 27ºC. This finding at 20.5±2^{O}C (55±5% R.H.) showed that in both season 13 to 13.5 days while at 32±2^{O}C (40±5% R.H.) shorter than as an average 11 to 11.5 days. EPPO (2005) also reported under optimal conditions the *T. absoluta* developed in about 30 days. We also agreed with Barriontes *et al.* (1998) who reported that average development time of *T. absoluta* was 23.8 days at 27.1ºC. Similar results were found by Fernandez and Montagne (1990) reported that the females lived longer than males, allowing them to be sexually mature when the males emerge. The present study results showed that usually males were more than female individuals. These results confirmed the work of Cuthbertson (2011), who was reported males were more than females in ratio.

This finding showed that the oviposition site choice by female *T. absoluta* was likely to be dependent on the highest densities of trichomes and infestation level. A good assumption could be that tomato leafminer infestation could be resulted in a high favorable and susceptible hosts of tomato varieties rather than potato and pepper and compared to the non-infested pepper genotypes sites are unlikely to support growth and development of a newly laid egg because of very less trichomes are observed. This study results were disagreed with the previous work of Muniappan (2013), he was stated that *Capsicum annum* (Pepper) was host preference by *T. absoluta*. However, high larvae density on tomato plants could be effectively associated by females *T. absoluta* for egg laying. Indeed, the number of eggs laid on the potato plant infested by *T. absoluta* larvae was lower than on other plants with lower infestation levels in tomato plants. Therefore, female eggs laying preferences were restricted to the least infested plant and oviposition seems to be stimulated at a certain density of trichomes, as the number of eggs was found to be statistically similar between each plant varieties.

Under similar conditions, tomato leafminer females may have develop a system of host plant assessment manifested by moderate oviposition level on varieties of potato, increased infestation level during maturity of tomato varieties and also, gradually increased infestation level in absence of all tomato varieties. But *T. absoluta* was non-preferentially oviposited of any pepper varieties during glasshouse conditions even no host chooses. However, many insect species mark the host patch during egg laid to regulate the oviposition site selection (Renwick, 1989). This study findings are confirmed with the laboratory studies of Thomas *et al.* (2014), they suggested that the female oviposition response could be more sophisticated than could be expected from a simple intraspecific competition model based on the detection of previous infestation. It is generally assumed that larval survivability depends on the female's oviposition choice because most lepidopteran larvae are unable to move to an alternative food source (Awmack and Leather, 2002; Gripenberg *et al*., 2010). The present study found differences in terms of total number of eggs laid on tomato varieties, but no difference in terms each crop varieties. It is also true that similar trends were observed among potato varieties. Considering *T. absoluta* antagonists may provide more explanations about this behavior according to Thomas *et al*. (2014). In their natural environment, Zappala *et al.* (2013) reported that the solanaceous leafminers were significantly affected by many factors that keep them at endemic levels. In contrast, tomato leafminer are not easily affected by many factors because of high generation and host specificity.

4.5 Conclusions

Based on these findings, it can be concluded that the different parts of tomato plant in their respective preference ranges *T. absoluta* should preferentially laid more eggs on leaves of tomato. On the other hand, *T. absoluta* at 32±2^{O}C (40±5% R.H.) was less conducive to maintaining a colony in a glasshouse conditions than rearing them at 20.5±2^{o}C (55±5% R.H.) in laboratory conditions. It has been proved as a most suitable for superior development, maximum survival and minimum mortality on *T. absoluta*. The current study paves the way to provide awareness to the farmers. Therefore, the management strategies of *T. absoluta* need high attention in Ethiopian climatic conditions.

CHAPTER FIVE

Determination of Economic threshold level and yield losses due to *Tuta absoluta*

5.1 Introduction

Tomato, *Lycopersicon esculentum* (Miller) is a vegetable crop of large importance throughout the world (Abdussamee *et al,* 2014; Mehraj *et al*., 2014). It is an important source of nutrients such as vitamins A, C, E and constitutes an important part of the house hold diet and national economy (Bhowmik *et al,* 2012; Kaur and Rishi, 2014). Tomato is devastating by an array of pests; however, the major damage is caused by the Tomato fruit borer, *Helicoverpa armigera* (Hubner) (Lepidoptera: Noctuidae) and tomato leafminer, *Tuta absoluta* Meyrick (Lepidoptera: Gelechiidae). The tomato leaf miner, *T. absoluta* attacks a wide range of host plants, and also has a wide geographical distribution throughout the world (Desneux *et al*. 2010). Nevertheless, its larvae feed mainly on the leaf and fruits of tomato and it are regarded as the most severe pest in tomato plant in Mediterranean Basin, but also in Africa, where it has become much more abundant over recent years particularly in Ethiopia (Gashawbeza and Abiy, 2013).

The economic injury level and economic threshold level (ETL) of *T. absoluta* is depends on population number and infestation level. However, currently the decision making management action of this devastating insect pest is not known in Ethiopia. The basic components of decision making in pest management; these are the economic injury level and economic threshold level (Stern *et al*., 1959; Pedigo *et al*., 1986). Integrated Pest Management poses some added considerations to the estimation of the economic injury level (ElL). It involves substitution of information for pesticide applications, recognizes costs of pest control beyond the direct chemical and application costs, and requires consideration of management options to reduce the necessity for chemical treatment. The challenge is to incorporate more host and pest dynamics into the economic injury level, which can help to verify pest management decisions and to recognize more efficient economic thresholds. The present study is conducted to the applications of economic injury and economic threshold level analysis to the calculation of cost/benefit (C/B) ratios for control of tomato leafminer, *T. absoluta* demonstrate the definition of the ElL advanced by Pedigo and Higley (1992) to estimate the cost-benefit ratios.

To manage *T. absoluta* larvae, pest management programmes employ intensive curative and/or preventive treatments that are economically unjustified. However, to control *T. absoluta* a number of chemical treatments are applied arbitrarily during the infestation of this insect pest with no regard to economic benefit. The current work was initiated to quantify the marketable yield loss from larval feeding of the tomato plants and to estimate the EIL in order to determine economic threshold level per plant when, and how many times, control measures against this devastating insect pest is justified economically. Regular monitoring is of paramount importance in IPM to ensure timely intervention before economic damage occurs, thereby saving farmers unnecessary cost. The functional relationship between pest infestation levels and benefit from control measures can be visualized as a benefit plane that shows increasing benefit with increasing infestation levels of either pest (Blackshaw, 1986). It is thus important to determine economic injury level for effective management of pests to prevent avoidable yield losses.

The rapid spread throughout the country, mainly related to the marketing of tomato berries and nursery plants, which allowed the insect to colonize all the areas involved in the production of tomato crop. It causes significant economic loss to tomato cropping in many countries in South America (Siqueira *et al.*, 2000). This pest has become one of the most severe pests of Solanaceae with estimated, crop losses of up to 100% in South America (Garzia *et al*., 2012; Öztemiz, 2012). The yield loss due to *T. absoluta* on tomato production in Ethiopia yet not studied. With this background, the objectives of these studies were to determine economic threshold level of *T. absoluta* and to assesse yield losses due to this insect pest.

5.2 Materials and Methods

5.2.1 Economic threshold level (ETL) and yield losses

5.2.1.1 Economic threshold level

The experiment was carried out at West Shewa of central Ethiopia under open farmer's field and glasshouse conditions during 2015/16 and 2016/17 using natural infestation. There were different levels of larval population to establish the economic injury levels of *T. absoluta.* The economic threshold level (ETL) was determined based on economic injury level and benefit cost ratio as suggested by Farrington (1977). The experiment was laid out in randomized complete block design (RCBD) with three replications. There were seven treatments consisted of six different larval densities i.e., 0, 1, 2, 3, 4, 5 and 6 larvae per plant. When the number of larvae reaches each treatment spray with standard chemical was sprayed accordingly. If the number of larvae at 1st instar exceeds larval density/ies removed unnecessary larval number from the plant before application. The damaged leaves and fruits were counted and fruit yields weighed from each treatment. Relationship between the larval densities and the number of leaf and fruit damaged was worked out by simple regression equations. Yield losses due to different treatments were derived by deducting the yield of the respective treatment from the yield of control (where no egg and larva protected treatment). The value of yield losses was determined according to the wholesale market price of tomato at current market situations.

Benefit cost ratio (BCR) was worked out as the ratio of the value of yield saved to the cost of insecticidal application. Standard chemical Chlorantraniliprole (Coragen 200 SC) was considered for calculating the cost of insecticidal application.

5.2.1.1.1 Data Collection

Regular periodical counts number of damaged (leaves and fruits) were taken from each pot, where the leaves and fruits had checked for eggs and larvae. However, when the sprayable level was attained, the pots were sprayed one day later with the appropriate insecticides, according to the pest situation. On Similar trends, on field conditions, counts of the *T. absoluta* (eggs and Larvae) and number of damaged (leaves and fruits) were taken from 10 plants in each plot, where the leaves and fruits had checked for eggs and larvae. However, when the sprayable level was attained, the plots were sprayed one day later with the appropriate insecticides, according to the pest situation. Spraying was carried out using a knapsack sprayer. Finally yield data was taken to compare the treatments.

5.2.1.2 Yield losses

5.2.1.2.1 Glasshouse Experiment

In glasshouse, sixteen pots were prepared and filled with compost, loam soil and sand soil in the ratio of 1:1:2, respectively. A Pot having a height of 25cm and a diameter of 20cm was used. Eight pots were protected from any insect infestation put in four different cages and eight pots were open for insect infestation (unprotected) treatment. Purposefully the door and windows of the glasshouse was kept open for 24 hours for the entrance of moths. Unprotected treatment was kept for *T. absoluta* infestation.

5.2.1.2.2 Field Experiment

T. absoluta control was done using fortnightly foliar application of recommended insecticide using a knapsack sprayer. The plots were separated by a 3m buffer zone of bare ground. Whenever insects were observed to make the plot free of insect infestation as much as possible. Treated tomato plants were sprayed with insecticides every five days with Chlorantraniliprole (Coragen 200 SC) at 200ml/hectare. Since it is not possible to completely protect and obtain pest free check plots in the experiment, percent yield losses were calculated based on estimated yield for pest free.

5.2.1.3 Yield Assessment

Yield data in treated and untreated plots in the tomato harvest seasons (2015 to 2017), under glasshouse and field conditions represented by weight in tons per hectare was estimated. Yield loss was estimated according to the following formula:

$$\text{Yield loss (\%)} = \frac{\text{Potential yield} - \text{Actual yield}}{\text{Potential yield}} \times 100$$

Potential yield is protected treatment was considered the standard for comparison with the other ones unprotected from *T. absoluta* (Actual yield).

5.2.1.4 Data recorded

Number of leaf infested, marketable and non-marketable fruits, bored/tunneled fruits were recorded during maturity stage before harvesting and finally yield data were weighed.

5.2.1.5 Data analysis

The collected data for each treatment was subjected to analysis of statistical software package (SAS, 2009) for analysis of variance of the experiment. The economic threshold level for leaf miner larvae were calculated by fitting regression equation Y= a + bx, between larval population levels and Benefit Cost Ratio. The larval density corresponding to unit benefit cost ratio was the economic injury level and economic threshold levels were set at 75% of EIL (Pedigo, 1991). Benefit-cost ratio was calculated using the following formula**:**

$$\text{BCR} = \frac{\text{Value of yield loss saved}}{\text{Total Cost}} \text{ x } 100$$

The correlation between the observed and predicted values of marketable fruit weight was calculated for each data set to assess the fit of the model. However, the economic injury level (EIL) is often expressed mathematically by the following formula:

$$\text{EIL} = \frac{\text{C x N}}{\text{V x I}}$$

Where:

"C" is the unit cost of controlling the pest (Birr/plant)

"N" is the number of pests injuring the commodity unit (number of pest/plant)

"V" is the unit value of the commodity (Birr/plant)

"I" is the percentage of the commodity unit injured (% loss)/plant

5.3 Results

5.3.1 Determine Economic Threshold Level

5.3.1.1 Glasshouse Experiment

5.3.1.1.1 Infestation of tomato plant

In both years, the differences among the various treatments were found significant ($P < 0.05$) on effect of leaf infestation and fruit yield damaged. Total number of damaged leaves per plant ranged from 0 to 87.33 during 2015 and 0 to 94.0 during 2016/2017. The percent fruit tunneled due to larval damage of *T. absoluta* increased significantly with rise in larval density per plant during the two seasons ranged from 0 to 11.67 during 2015 and 0 to 15.67 during 2016/2017 (Table 5.1).

Table 5.1: Infestation level of *T. absoluta* on tomato plants and marketable yield per plant in kg under glasshouse conditions during 2015 – 2017.

Trts	Year I (2015/16)			Year II (2016/17)		
	No. of leaves infested/plant	No. of fruit tunneled/plant	Marketable yield/plant in (kg)/plant	No. of leaves infested/plant	No. of fruit tunneled/plant	Marketable yield/plant (kg)/plant
T$_1$	0.0^{e}	0.0^{d}	0.92^{a}	0.0^{f}	0.0^{e}	0.93^{a}
T$_2$	11.0^{d}	2.67^{c}	0.82^{a}	9.67^{e}	2.0^{de}	0.83^{a}
T$_3$	23.0^{c}	3.33^{c}	0.43^{b}	19.0^{d}	3.0^{cd}	0.40^{b}
T$_4$	29.0^{ab}	4.0^{c}	0.44^{b}	26.0^{cd}	5.0^{c}	0.43^{b}
T$_5$	31.0b^{a}	7.33^{b}	0.40^{bc}	29.33^{c}	5.67^{c}	0.38^{b}
T$_6$	34.67^{b}	8.33^{b}	0.33^{bc}	40.33^{b}	10.0^{b}	0.30^{bc}
T$_7$	87.33^{a}	11.67^{a}	0.23^{c}	94.0^{a}	15.67^{a}	0.18^{c}
LSD	7.32	2.54	0.20	7.74	2.75	0.15
MSE ±	4.08	1.41	0.11	13.80	1.53	0.08
CV (%)	13.21	21.345	21.73	4.30	20.88	16.98

Note: Means with the same letter(s) in columns are not significantly different for each other.
All treatment effects were highly significant at $p<0.01$ (DMRT)
T$_1$= 0 larva density, T$_2$= 1 larval densities T$_3$= 2 larval densities T$_4$= 3 larval densities
T$_5$= 4 larval densities T$_6$= 5 larval densities T$_7$ = Control

Under glasshouse experiment, the marketable fresh fruit yield per hectare varied from 8,571 kg to 34,787 kg during 2015/2017 and from 6,932 kg to 35,039 kg during 2016/2017 (Table 14), corresponding to larval densities above 5 to 0 per plant, respectively. Marketable fresh fruit yield results were indicated that significant variations among the treatments (Table 5.1 and 5.2). 'T_1' recorded maximum marketable fresh fruit yield per hectare (34,787 kg) followed by 'T_2' (31,132 kg), 'T_3' (25,912.84 kg), and 'T_4' (21,376.61 kg) and minimum fresh fruit yield (8,571 kg) was recorded by 'T_7' during 2015 study period. On similar manner, the study was conducted during 2016 the maximum fresh fruit yields were recorded by 'T_1' (35,039 kg) followed by 'T_2'' (30,880 kg) per hectare, the minimum marketable fresh fruit yields also recorded by 'T_7' (6,932 kg) followed by 'T_6' (11,218 kg) may be attributed to the maximum net profit per hectare (Table 5.2). A density of one larva per plant caused about 10.51% marketable yield loss was recorded during 2015 which represent 3,655kg minimum yield loss per hectare and maximum yield loss 26,307 kg which represents 75.62% yield loss was recorded. In other year 2016 a density of one larva per plant indicated 11.87% marketable yield loss was observed, that represent 4,159.0 kg minimum yield reduction per hectare and maximum yield reduction was observed 28,107.0 kg/ha which showed 80.22% yield losses.

The data on *T. absoluta* infestation at different larval densities and fruit yield loss showed significant ($P < 0.05$) positive correlation in both years. The linear regression co-efficient 'b' value during 2015/2017 was highly significant with higher r^2 value (0.965) showing more pronounced effect of *T. absoluta* infestation on yield. The highest value of ($r^2 = 0.965$) indicated 96.5 percent variation in yield due to *T. absoluta* infestation. Similarly during 2016 also the linear regression co-efficient 'b' value was highly significant with high value of r^2 (0.873) which explained 87.3 percent variation in total yield.

Table 5.2: Marketable yield in kg/hectare and interms of money under glasshouse conditions during 2015-2017.

	Year I (2015/16)				**Year II (2016/17)**			
Treatments	Marketable yield (kg)	Marketable Yield loses			Marketable yield (kg)	Marketable yield losses		
		(kg/ha)	(%)	U.S ($/ha)		(Kg/ha)	(%)	U.S. ($/ha)
T_1	34,787^{a}	---	---	---	35,039.00^{a}	---	---	---
T_2	31,132^{ab}	3,655.0	10.51	1,589.13	30,880.00^{a}	4,159.0	11.87	1,808.26
T_3	25,912.84^{b}	8,874.16	25.51	3472.50	26,902.94^{b}	8,136.06	23.22	3183.67
T_4	21,376.61^{bc}	13,410.39	38.55	5,247.54	22,260.28^{b}	12,778.7	36.47	5,000.36
T_5	14,999^{d}	19,788.0	56.88	8,603.48	14,243.00^{c}	20,796.0	59.35	9,041.74
T_6	12,478^{d}	22,309.0	64.13	9,699.57	11,218.00^{c}	23,821.0	67.98	10,356.96
T_7	8,571^{d}	26,307.0	75.62	11,437.83	6,932.00^{cd}	28,107.0	80.22	12,220.44
LSD	8362.60				5663.70			
MSE ±	5209.83				3151.56			
CV (%)	25.63				16.98			

Note: Means with the same letter(s) in columns are not significantly different for each other.
All treatment effects were highly significant at $p<0.01$ (DMRT)
Cost of insecticides 365.22 U.S.$/liter
Average price of tomato 0.35 U.S.$/kg
Labor charge 3.04 U.S.$/day
1.0 U.S $=23.00 Eth. Birr

Table 5.3: Economic analysis of *T. absoluta* on management of tomato crop under glasshouse conditions during 2015-2016.

				Cost of production (U.S.$/ha)							
		Mean yield loss		Value of	Mean	Mean	Other production			Net in	
Trts	Marketable yield (kg/ha)	(kg/ha)	(%)	Yield loss (U.S.$/ha)	Cost of insecticide (U.S.$/ha)	Labor charges U.S.$/ha	cost (E.g. Fertilizer, cost of planting pot, cost of marketing and distribution) (U.S.$/ha)	Total cost U.S.$/ha)	Total income U.S.$/ha)	come U.S.$/ha)	**CBR**
T_1	34,913.0	---	---	---	1,552.17	2,482.83	3,166.96	7,201.96	13,661.61	6,459.65	---
T_2	31,006.0	3,907.0	11.19	1,698.670	1,004.35	2,482.83	3,166.96	6,654.13	12,132.78	5,478.65	1.82
T_3	18,843.0	8,505.03	24.37	3,328.09	821.74	2,482.83	3,166.96	6,471.52	7,373.35	901.83	1.14
T_4	17,141.5	13,094.55	37.51	5,123.95	547.83	2,482.83	2,881.74	5,912.40	5,962.26	49.86	1.00
T_5	14,621.0	20,292.0	58.12	8,822.61	547.83	2,482.83	2,881.74	5,912.40	5,721.26	-191.14	0.97
T_6	11,848.0	23,065.0	66.04	10,028.26	456.52	2,482.83	2,553.35	5,492.70	4,636.17	-856.53	0.84
T_7	7,751.5	27,161.5	77.80	11,809.35	0.0	2,482.83	1,814.35	4,297.18	3,033.20	-1263.98	0.71

Note: Cost of insecticides 365.22 U.S.$/liter
Average price of tomato 0.35 U.S.$/kg
Labor charge 3.04 U.S.$/day
1.00 U.S $=23.00 Eth. Birr

A positive correlation was found between number of leaf infestation and marketable yield loss (r^2 =0.720) and (r^2 =0.637) during 2015 and 2016, respectively. However, increase in larval population per plant did show proportionate increase in marketable yield loses. The relationship between *T. absoluta* number of leaf infestation and marketable yield loss percent was expressed by the regression equation Y= 0.854x + 15.12 and Y = 0.771x + 22.96 for years 2015 and 2016, respectively.

A density of two larvae per plant caused about 6.33% leaves damaged during 2015/2017 which indicated 10.51% yield losses. In 2016/2017, 5.83% leaves damaged was observed which showed 11.87% yield losses (Table 5.2). The control (unsprayed) treatments was caused 56.97 leaf damaged during 2015/2016 and 59.44% during 2016/2017. The control treatments in both years revealed that 75.62% and 80.22% yield losses during 2015/2016 and 2016/2017, respectively.

5.3.1.1.2 Benefit-cost ratio, Economic injury level and Economic threshold

The cost of *T. absoluta* control (cost index in EIL formula) was estimated by adding the cost of insecticide per plant, number of pest/plant, total cost of plant production and yield loss percent/plant. Actuality, the EIL value can be complicated to calculate exact number because of the dynamic nature of pest infestation level, damage system and crop value.

The ratio of the value of yield saved to the cost of insecticide application at three larvae per plant was 1.08 during 2015 - 2017. EIL lies at the pest population density where BCR would be 1.08. In order to calculate the exact larval density at which BCR would be 2.25 larvae, the correlation of larval density (X) with the BCR (Y) was calculated. There was a strong positive correlation and linear relationship between those two variables. The regression equations derived were y= 0.157x + 0.19 during 2015- 2017. It should be noted that estimate of damage caused by pests rather than pest density is especially important for tomato leafminer, *T. absoluta*. Where: x =Larval density per plant and y= BCR.

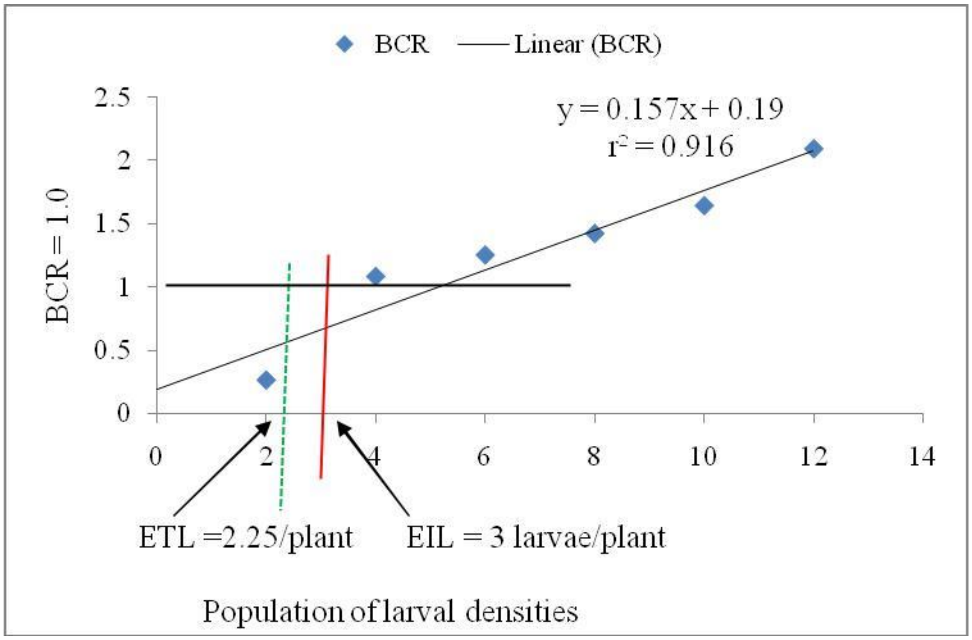


Figure 5.1: Relationship between larval densities and BCR under glasshouse conditions during 2015-2017.

From the above equations the EILs of *T. absoluta* larvae determined as three larvae during 2015-2016/2017 (Fig. 5.1). On the basis of means of two years, the EIL value was 3 larvae per plant under glasshouse. Therefore, the economic threshold level was determined as 2.25 larvae per plant.

5.3.1.1.3 Yield loss-larval population relationship

Larval density and yield loss percent are presented in (Table 5.4) for each year of the experiment. Larval density and yield loss percentage was a strong positive correlation and linear relationship between those two variables. The regression equations derived were y=7.013x + 1.418 during 2015/16 under glasshouse during 2015 and y = 6.410x + 8.550 during 2016/2017. The values of the parameters for calculation of yield loss and EIL over the two years of the experiment are presented in (Fig. 5.2 & 5.3).

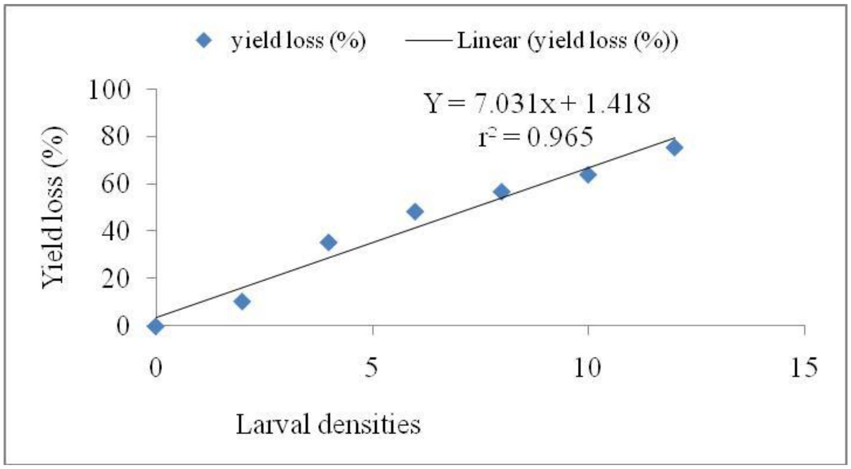


Figure 5.2: Relationship between larval densities of *T. absoluta*/plant and percent yield loss correlation coefficient under glasshouse conditions during 2015/2017.

Number of larvae was indicates that initiated control prevented pests from causing economic injury while number of larval population rate above the limit indicates that the economic threshold was inadequate to prevent an increasing infestation from causing economic damage.

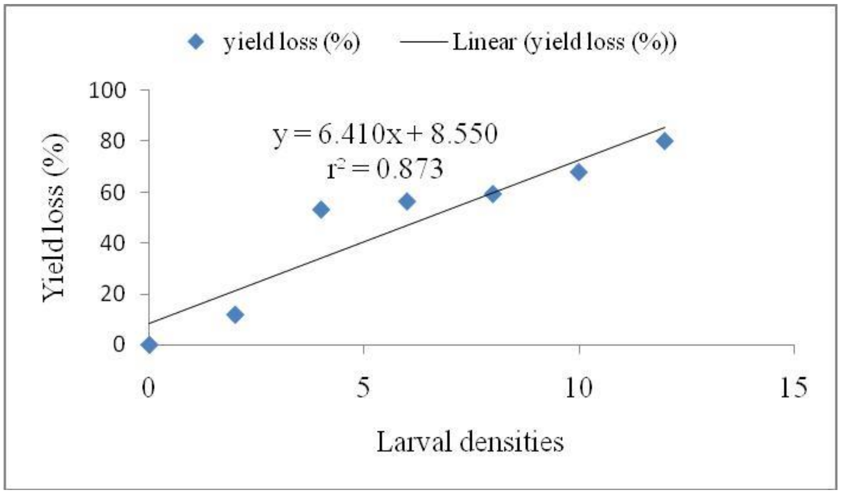


Figure 5.3: Relationship between larval densities of *T. absoluta*/plant and percet yield loss correlation coefficient under glasshouse condition during 2016/2017.

The economic threshold is the population level where insect control should be initiated to avoid exceeding the economic injury level. The ElL is the most persistent and influential element in pest management.

5.3.1.1.4 Yield-leaf infestation relationship

The data on *T. absoluta* infestation at different exposure periods and marketable fruit yield losses showed significant ($p < 0.05$) positive correlation in the years i.e. 2015/2016 and 2016/2017. The linear regression co-efficient 'b' value during 2015/2016 was significant with higher r^2 value showing more pronounced effect of *T. absoluta* infestation on yield. The highest value of r^2 (0.720) indicated 72% variation in yield due to *T. absolut* infestation. Similarly during 2016/2017 also the linear regression co-efficient 'b' value was significant with high value of r^2 (0.637) which explained 63.7 percent variation in total yield (Fig. 5.4 & 5.5). The relationship between *T. absoluta* infestation and marketable yield loss was expressed by the regression equation $Y = 0.854x + 15.22$ and $Y= 0.771x + 22.96$ for years 2015/2016 and 2016/2017, respectively.

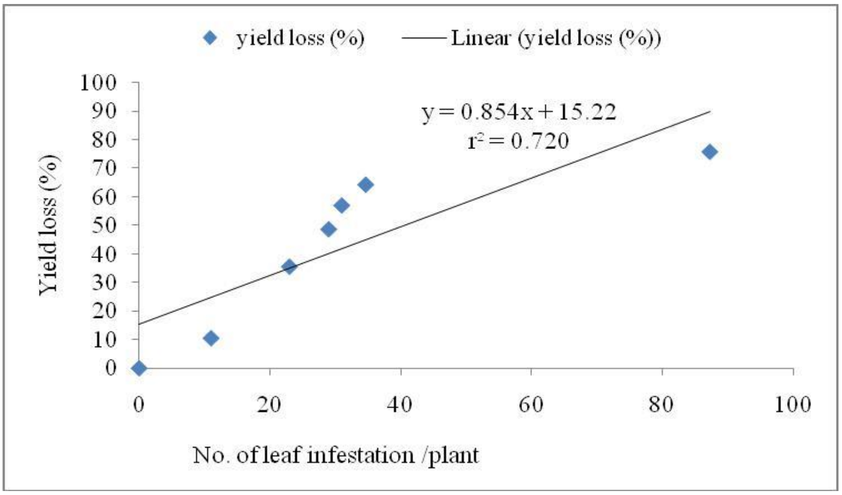


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Figure 5.4: Relationship between no.of leaf infestation/plant and percent yield loss correlation coefficient under glasshouse condition during 2015.

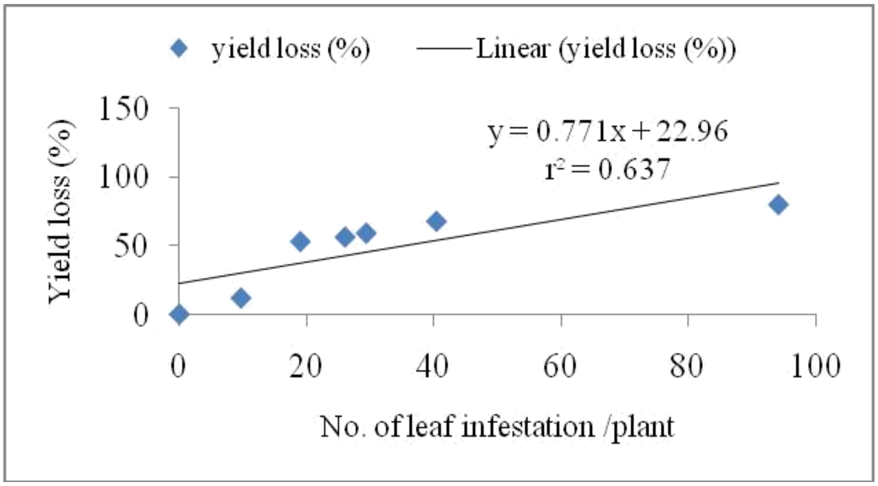


Figure 5.5: Relationship between no.of leaf infestation/plant and percent yield loss under glasshouse condition correlation coefficient during 2016.

5.3.1.1.5 Frequency of application

The frequency of application made sixteen times within 5 days intervals at the recommended application rates during 2015/2016 and eighteen times within 5 days intervals in 2016/2017 for protected (insect free) treatment. On the other hand, insecticide application times were ranged between 4 to 12 times during both years in others treatments (Table 5.4). The management option of *T. absoluta* by insecticide was made when the economic threshold level was determined 8 to 10 times application per season during 2015-2017.

Table 5.4: Frequency of insecticide application on management of *T. absoluta* on tomato crop under glasshouse during 2015-2017.

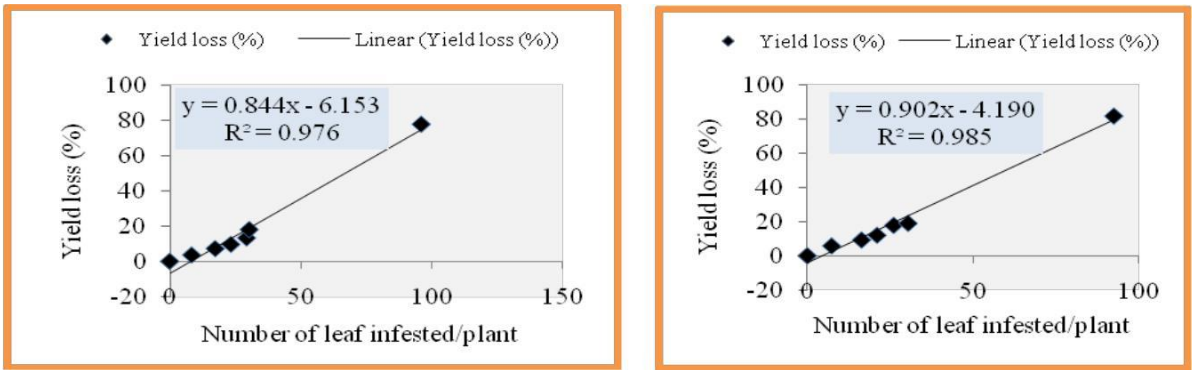
Treatments	Frequency of application	
	(2015)	(2016)
T_1	16	18
T_2	10	12
T_3	8**	10**
T_4	5*	7*
T_5	5	7
T_6	4	6
T_7	0	0

Note: ** ETL * EIL

5.3.1.2 Field conditions

A population density of one larva per plant caused about 6.33% leaf damage during 2015/2016 which indicated 3.61% yield losses. In 2016/17, 8.03% leaf damage was observed which showed 5.75% yield losses. The control (unsprayed) treatments were caused 50.76 leaf damaged during 2015/16 and 56.97% in 2016/2017. The control treatments in both years revealed that 77.91% and 81.61% yield losses in 2015/2016 and 2016/2017, respectively. On the other hand, 5 larvae of *T. absoluta* per plant were showed 18.07% marketable yield losses were observed during 2015/2016, in similar manner 5 larvae of *T. absoluta* per plant revealed that 18.61% marketable yield reduction during 2016/2017.

The relationship between *T. absoluta* number of leaf infestation and marketable yield loss percent was expressed by the regression equation y = 0.844x - 6.153 and y = 0.902x - 4.190 for years 2015/2016 and 2016/2017, respectively. A strong positive correlation was found between number of leaf infestation and marketable yield loss (r^2 =0.976) and (r^2=0.985) during 2015/2016 and 2016/2017, respectively (Fig. 5.6). However, increase in larval population per plant did show proportionate increase in marketable yield loses (Table 5.6 and 5.6).



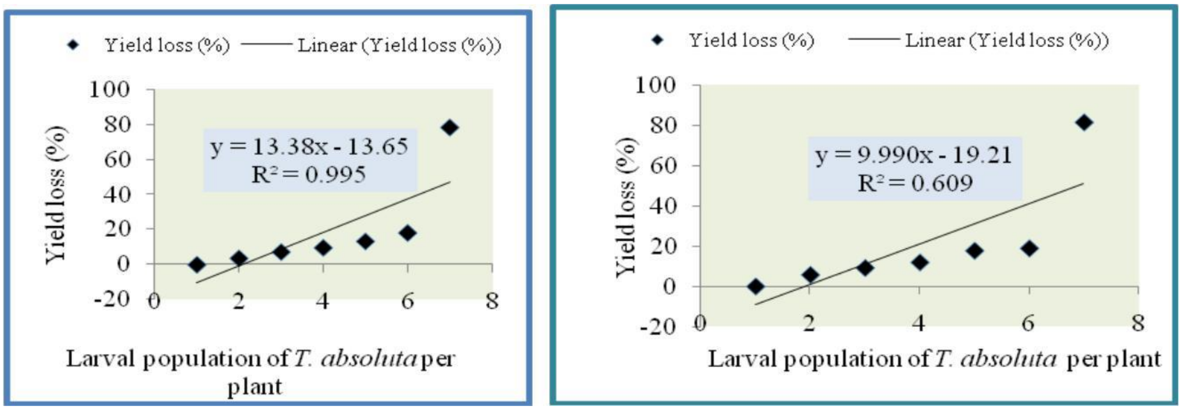
(a) 2015/2016 (b) 2016/2017

Figure 5.6: Relationship between number of leaf infested/plant and percent yield loss correlation coefficient on field conditions during 2015- 2017.

5.3.1.2.1 Economic threshold and Economic injury level

A linear relationship between tomato yield and larvae of *T. absoluta* per plant was detected on field during 2015-2017. There was a significant linear relationship between larval infestation and marketable yield loss when tomato fruit and leaf were infested in larvae (Fig. 5.8 & 5.9). Data presented on Table 5.5 indicated that marketable yields per hectare were statistically significant $P < 0.05$) different, a population density of one larva per plant caused 394.57 U.S dollar during 2015/2016 which indicated 1,134.4 kg/ha yield losses. In 2016/2017, one larva caused 569.45 U.S dollar which showed 1,637.16 kg/ha yield losses. The control (unsprayed) treatments were caused 8,548.81 U.S dollar during 2015/2016 which showed 24,577.84 kg/ha yield losses and 9,249.77 U.S dollar lost in 2016/2017 which showed that 26,593.08 kg/ha yield losses were observed. The criterion of selection of slope for economic injury level calculation was based on the worst-case scenario of yield loss per insect, i.e., the control unsprayed treatment (Table 5.6).

The economic threshold levels were developed from EILs, expressed as the percentage of infested plants at least three larvae/plant (Table 5.7). Larval survival was incorporated in the ETL calculation considering the range of larval survival observed during 2015-2017.



(a) 2015/16

(b) 2016/17

Figure 5.7: Relationship between larval densities of *T. absoluta*/plant and percent yield loss correlation coefficient on field condition during 2015 - 2017.

Table 5.5: Infestation level of *T. absoluta* on tomato plants on field conditions during 2015-2017

Treatments	Year I (2015/2016)			Year II (2016/2017)		
	No. of leaves infested/plant	No. of fruit tunneled/plant	Marketable yield/plant in (kg)	No. of leaves infested/plant	No. of fruit tunneled/plant	Marketable yield/plant (kg)
T_1	0.0^{e}	0.0^{e}	0.83^{a}	0.0^{e}	0.0^{e}	0.87^{a}
T_2	8.33^{d}	2.33^{de}	0.80^{ab}	13.33^{d}	1.33^{de}	0.82^{ab}
T_3	17.33^{c}	3.33^{d}	0.77^{abc}	16.33^{d}	3.33^{cd}	0.79^{abc}
T_4	23.33^{bc}	3.67^{d}	0.75^{bc}	21.0^{cd}	4.67^{c}	0.76^{bc}
T_5	29.33^{b}	9.0^{c}	0.72^{c}	26.0^{bc}	6.33^{bc}	0.71^{c}
T_6	30.33^{b}	12.33^{b}	0.68^{c}	30.37^{b}	9.33^{b}	0.70^{c}
T_7	96.0^{a}	23.67^{a}	0.18^{d}	92.33^{a}	26.67^{a}	0.16^{d}
LSD	7.77	3.16	0.10	6.89	3.17	0.07
MSE ±	4.33	1.76	0.06	3.83	1.77	0.04
CV (%)	14.80	22.66	8.11	13.86	23.92	5.5

Note: Means with the same letter(s) in columns are not significantly different for each other.
All treatment effects were highly significant at $p<0.01$ (DMRT)
T_1= 0 larva density, T_2= 1 larval densities T_3= 2 larval densities T_4= 3 larval densities
T_5= 4 larval densities T_6= 5 larval densities T_7= Control

Table 5.6: Marketable yield in ton per hectare on field conditions during 2015-2017.

Treatments	**Year I (2015/2016)**				**Year II (2016/2017)**			
	Marketable yield (kg/ha)	Marketable Yield loses (kg/ha)	Marketable Yield loses (%)	Marketable Yield loses (U.S. $/ha)	Marketable yield (kg/ha)	Marketable yield losses (Kg/ha)	Marketable yield losses (%)	Marketable yield losses (U.S. $/ha)
T_1	31,384.0^{a}	---	---	---	32,643.0^{a}	---	---	---
T_2	30,249.6^{ab}	1,134.4	3.61	394.57	31,005.84^{ab}	1,637.16	5.75	569.45
T_3	29,115.24^{abc}	2,268.76	7.23	789.13	29,871.48^{abc}	2,771.52	9.20	964.01
T_4	28,359.0^{bc}	3,025.0	9.64	1,052.17	28,737.12^{bc}	3,905.88	11.97	1,358.57
T_5	27,224.64^{c}	4,159.36	13.25	1,446.73	26,846.52^{c}	5,796.48	17.76	2,016.17
T_6	25,712.16^{c}	10,587.0	18.07	3,682.43	26,468.4^{c}	6,174.6	18.92	2,147.69
T_7	6,806.16^{d}	24,577.84	77.91	8,548.81	6,049.92^{d}	26,593.08	81.61	9,249.77
LSD	2348.70				3785.80			
MSE ±	1306.97				2106.65			
CV (%)	5.05				8.11			

Note: Means with the same letter(s) in columns are not significantly different for each other.

All treatment effects were highly significant at $P < 0.01$ (DMRT)

T_1= 0 larval density, T_2= 1 larval densities T_3= 2 larval densities T_4= 3 larval densities
T_5= 4 larval densities T_6= 5 larval densities T_7= Control

Table 5.7: Mean economic analysis of *T. absoluta* on management of tomato crop at Western Shewa of central Ethiopia on field during 2015-2017

Cost of production (U.S.$/ha)

Trts	Mean Marketable yield	Mean yield loss (kg/ha)	(%)	Value of Yield loss (U.S.$/ha)	Mean Cost of insecticide (U.S.$)	Mean Labor charges (U.S.$)	Other production cost (E.g. Fertilizer, Land rent, transport, cost of marketing and distribution) (U.S.$)	Total production cost (U.S.$)	Total income (U.S.$)	Net income (U.S.$)	**BCR**
T_1	32,013.5	---	---	---	1,552.17	1,865.65	6,803.91	10,221.74	11,135.13	913.39	---
T_2	30,627.72	1,385.78	4.68	481.23	1,004.35	1,865.65	6,803.91	9,673.91	10,653.12	979.21	0.91
T_3	29,493.36	2,520.14	8.22	876.57	821.74	1,865.65	6,803.91	9,491.30	10,258.56	767.26	0.93
T_4	28,548.06	3,465.44	10.81	1,205.37	821.74	1,865.65	6,803.91	9,491.30	9,929.76	438.46	0.96
T_5	27,035.44	4,978.06	15.51	1,731.50	730.43	1,865.18	6,803.91	9,400.00	9,403.63	3.63	1.00
T_6	26,090.28	5,923.22	18.50	2,060.25	639.13	1,865.65	6,803.91	9,308.70	9,074.88	-233.82	1.03
T_7	6,428.04	25,585.46	79.76	8,899.29	0.00	1,865.65	6,803.91	8,669.57	2,235.84	-6,433.73	3.88

Note: Cost of insecticides 365.22 U.S.$/liter
Average price of tomato 0.35 U.S.$/kg
Labor charge 3.04 U.S.$/day
1.00 U.S $=23.00 Eth. Birr

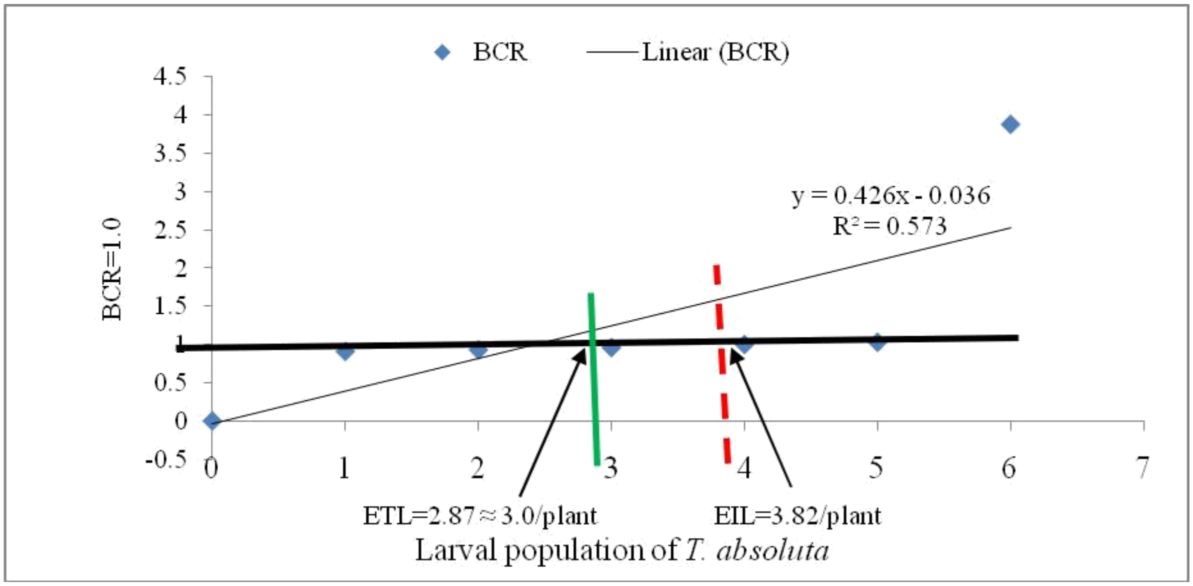


Figure 5.8: Relationship between larval densities and BCR under open field during 2015-2017

From the above equations the EILs of *T. absoluta* larvae determined as three larvae during 2015-2017 (Fig. 5.8). On the basis of means of two years, the EIL value was 3.82 larvae per plant under field conditions. Therefore, the economic threshold level was determined as 2.87 larvae per plant.

5.3.1.2.2 Cost-benefit ratio

Cost of management reflects a range of actual price scenarios in current market price. Crop value considers a range of crop market price for the period of 2015-2017, including low, average, and high prices of tomato. The crop values also are converted in U.S. dollars per Ethiopia Birr. Based on 0.03 kg of yield loss per plant with one tomato leafminer larva, conversion was done to kilograms per hectare considering 37,812 kg/ha and resulted in a yield loss mean of 1,134.36 kg/ha and untreated control was observed 0.65 kg/plant which was converted to kg per hectare 24,577.8 kg during 2015/2016. In the second year (2016/2017), also yield loss per plant with one larva and untreated control were 0.05 and 0.71 kg which was converted per hectare was 1,890.6 kg/ha and 26,846.52 kg/ha, respectively.

Table 5.8: Economic injury level of *T. absoluta* on management of tomato crop at Western Shewa of central Ethiopia on field during 2015-2017.

Trt	Pest control Birr/plant "C"	No. of *Tuta absoluta*/plant "N"	Total cost of tomato production/plant "V"	Percent of yield loss/plant "I"	EIL	BCR
T_1	0.94	0	3.49	---	---	---
T_2	0.61	1	3.16	4.68	4.07	0.91
T_3	0.50	2	3.04	8.22	4.00	0.93
T_4	0.50	3	3.04	11.38	4.29	0.96
T_5	0.44	4	2.99	15.51	3.82	1.00
T_6	0.39	5	2.93	18.50	3.61	1.03
T_7	0.0	6 and above	2.54	79.76	---	3.88

Note: EIL=3.82 larvae/plant   ETL=2.87 ≈ 3.0 larvae/plant

5.3.1.2.3 Frequency of application

The frequency of application made sixteen times within 5 days intervals at the recommended application rates during 2015/2016 and eighteen times within 5 days intervals in 2016/2017 for protected (insect free) treatment. On the other hand, application times were ranged between 5-13 times during both years (Table 5.9).

Table 5.9: Frequency of insecticide application on management of *T. absoluta* on tomato crop at Western Shewa of central Ethiopia on field during 2015-2017.

Treatments	Frequency of application	
	(2015/2016)	(2016/2017)
T_1	16	18
T_2	10	13
T_3	8	10
T_4	8**	8**
T_5	7*	7*
T_6	5	7
T_7	0	0

Note: ** ETL * EIL

5.3.2 Study on yield loss assessments due to *T. absoluta*

5.3.2.1 Glasshouse Experiment

Results on (Table 5.10) showed that highly significant ($P < 0.01$) different was observed between protected and unprotected treatments. The infestation number on protected leaves (2.25/plant) was very low as compared with unprotected leaves (43.75/plant) during November 2015/2016 to February 2016/2017. Similarly, fruit bored per plant under protected treatment completely free while on unprotected treatment it was recorded (11.5/plant). It also highly significant ($P < 0.01$) different was observed regarding to marketable yield per plant between protected and unprotected treatments. It was recorded 86.95 and 108.5 gm/plant in protected and unprotected treatments, respectively. Weight of single fruit of unprotected treatment due to *T. absoluta* was considerably less as compared to the protected treatment. The result has also shown that number of infested leaves is lowest in the treatment sprayed with Coragen 200 SC. The protected treatment sprayed with Coragen 200 SC has the highest fruit marketable weight (867.95gm/plant) as shown in table below.

Table 5.10: Impact of tomato leafminer, *T. absoluta* on yield and yield components of tomato under glasshouse condition during November 2015/2016 to Feburary 2016/2017.

Treatments	No. of Infested leaf/plant	No. of tunneled fruit/plant	Average single fruit weight in gm	Marketable fruit wt/plant in gm	Total fruit yield/plant in gm
Protected	2.25^{b}	0.00^{b}	57.32^{a}	867.95^{a}	1003.58^{a}
Unprotected	43.75^{a}	11.50^{a}	41.00^{b}	108.5^{b}	382.50^{b}
LSD at 0.01	10.80	5.59	6.78	225.22	477.40
SE±	2.61	1.35	1.64	54.53	115.61
CV (%)	11.37	23.55	3.34	11.17	16.68

Note: Means with the same letter(s) in columns are not significantly different for each other.
All treatment effects were highly significant at $P < 0.01$ (DMRT)

In the second season (March to June 2016) the infestation level of *T. absoluta* was very high and completly damaged the leaves of tomato under glasshouse condition. A significant ($P < 0.01$) different was also noted on the marketable fruit yield during this season. Hence, due to all leaves dried rate of photosynthesis reduced and finally flowering and fruit setting were stopped and finally dead. The unprotected control had 100% yield loss recorded.

Table 5.11: Impact of tomato leafminer, *T. absoluta* on tomato leaf under glasshouse condition during March to June 2016.

Treatments	**No. of Infested leaf/plant**
Treated	2.25^{b}
Untreated	170.25^{a}
LSD at 0.01	38.08
SE±	9.22
CV (%)	10.69

Note: Means with the same letter(s) in column are not significantly different for each other.
All treatment effects were highly significant at $P < 0.01$ (DMRT)

5.3.2.2 Field Experiment

The first year experiments were conducted during November 2015 to March 2016. In other season the second year experiments were conducted to November 2016 to March 2017 at three locations of West Shewa of central Ethiopia. Results on yield loss assessment with insecticides spray (treated) and untreated were highly significant ($P < 0.01$) different presented in (table 5.10). The treated was imposed irrespective of *T. absoluta* occurrence from 10 days after transplanting till harvesting. It is evident that, significantly lower leaf infested, low fruit bored and higher marketable and total yield were recorded from all locations compared to the untreated control.

Chlorantraniliprole (Coragen 200 SC) spray was quite effective in reducing population density of *T. absoluta*. Leaf infested and fruit tunneled were least in all three locations, that is twelve times sprays of Chlorantraniliprole (Coragen 200 SC) at 200 ml per hectare and the *T. absoluta* was completely minimized with treated treatments. Maximum marketable yield (30.8, 28.18 and 27.48 t/ha) was obtained with treated treatments in Dendi, Ambo and Toke kutaye districts, respectively. While minimum marketable yields in two years were recorded at Dandi (5.45 t/ha) followed by Toke kutaye (7.95 t/ha) and Ambo (11.25 t/ha) Districts. Two years mean data presented on Table 5.12 showed that the minimum total yield loss was recorded 43.83% and marketable yield was 60.08% in Ambo district whereas, the maximum total yield and marketable yield losses were observed in Dendi district 62.89% and 82.31%, respectively. These results indicated that *T. absoluta* can cause 60.08% to 82.31% marketable tomato fruit yield loss under field condition at Western Shawa of central Ethiopia.

Table 5.12: Impact of tomato leafminer, *T. absoluta* on yield and yield components of tomato in open farmers field during 2015- 2017

Treatments	No. of Infested leaf/plant	No. of tunneled fruit/plant	Marketable yield in ton/ha	Total yield in ton/ha	Percent yield losses
Ambo district					
Protected	3.75^b	0.25^b	28.18^a	35.43^a	
Unprotected	24.25^a	13.5^a	11.25^b	19.9^b	43.83-60.08
LSD at 0.01	5.06	5.0	7.82	11.85	
CV (%)	8.75	17.57	9.60	10.38	
SE±	1.22	1.21	1.89	2.87	
Dendi district					
Protected	1.0^b	0.5^b	30.8^a	35.38^a	
Unprotected	35.75^b	15.75^b	5.45^b	13.13^b	62.89-82.31
LSD at 0.01	9.65	6.48	7.96	9.04	
CV (%)	12.71	19.30	10.63	9.02	
SE±	2.34	1.57	1.93	2.19	
Toke kutaye district					
Protected	3.5^b	0.5^b	27.48^a	34.2^a	
Unprotected	33.5^a	18.0^a	7.95^b	16.2^b	52.63-71.67
LSD at 0.01	6.31	5.0	10.63	16.68	
CV (%)	8.26	13.24	14.53	16.02	
SE±	1.53	1.24	1.57	4.04	

Note: Means with the same letter(s) in columns are not significantly different for each other.
All treatment effects were highly significant at $P < 0.01$ (DMRT)

5.4 Discussions

The experiment was conducted to uses control costs, crop value and plant-monitoring, marketable fruit loss to calculate an injury level for leaf infestation and fruit damaged. Number of larvae was indicates that initiated to prevente pests from causing economic injury while number of larval population rate above the limit indicates that the economic threshold was inadequate to prevent an increasing infestation from causing economic damage. This result consistent with the work of Pedigo and Higley (1992) to recapitulate the primary variables to define the ElL is not a discrete pest density, but is variable depending on control costs, the value of the production protected, production losses per pest, and the efficacy of the control.

The ElL is the most persistent and influential element in pest management. Indeed, ElL and ETL continue to function as the primary mechanisms for making pest management decision (Sarmahl *et al*., 2011). Various workers (Stern *et al.,* 1959; Stone and Pedigo, 1972; Smith and Vanden Bosch, 1967; Ram and Patil, 1986) highlighted the importance of ElL and ETL in the pest management of different crop plants. It should be noted that estimate of damage caused by pests rather than pest density is especially important for tomato leafminer, *T. absoluta*.

Currently, no any report on economic threshold level of *T. absoluta* globally. There are different damage indices and developmental stages have been used by different authors to estimate the economic injury and economic threshold level on different crops and different insect pests. For instance, Bahrami *et al.* (2003) and Naranjo *et al*. (1996) studied the number of insects per bush for the sunn pest, *Eurygaster integriceps* (Hem: Scutelleridae) and the whitefly, *Bemisia tabaci* (Hem: Aleyrodidae), respectively, while Jemsi (2007) used the density of pest on surface unit to estimation the economic injury level in the grain leaf miner *Syringopais temperatella* (Lepdoptera: Elachistidae). Reddy *et al.* (2001) found that EIL of other lepidopteran insect, *Helicoverpa armigera* in pigeon pea was 0.78 to 0.80 larvae per plant and also Nath and Rai (1995) reported EIL of gram pod borer under natural condition to be 1.77 to 2.00 larvae per m row length was determined. Furthermore, Prabhakar *et al.* (1998) found EILs of chickpea pod borer were 0.9 and 1.23 larvae per m for unirrigated and irrigated crops, respectively. However, our method is different and easily applicable for tomato leafminer, *T.absoluta* management. This study used the number of larvae per plant to simplify the observation and exact estimation of the economic injury level. This index can easily be used by farmers to estimate the intensity of damages on their tomato fruits and develop management options using their available materials.

T. absoluta larval feeding not only reduces the marketability of plants, but it also reduces the photosynthetic capacity of plants, which reduces plant vigor, growth and yield (Al-Khateeb and Al-Jabr, 2006). In this study, *T. absoluta* feeding on plants result in reduced rate of photosynthesis, facilitate for pathogen, and an increase in leaf transpiration. Maximum leaf and fruit damages were recorded in untreated control treatment whereas, minimum infestation of leaves and fruit damage were recorded on treated treatment. High marketable yield was obtained from the treated treatment. The results of the present study can be corroborated with the findings of earlier researchers **(**Lopez, 1991) and (CFIA, 2010). These findings are also confirmed the work of Mohamed and Khalid (2011) observations in Sudan, *T. absoluta* is recorded as a serious problem to tomato and potato crops after the official report in the country in 2011 with infestation levels from 50% and up to 80%.

This study findings also consistent with the findings of Öztemiz (2012), who was mentioned larvae reduce tomato yield and fruit quality losses range from 80 to 100% by attacking leaves and flowers, burrowing stalks, apical buds, green and ripe fruits. Tomato plant in the world suffered about 100% yield loss due to *T. absoluta* (Derbalah *et al*., 2012). The larvae can destroy up to 100% of the leaf surface and damage 50-100% of fruits in severely attacked on fields (EPPO, 2005). Öztemiz (2012) reported 100 percent yield loss due to *T. absoluta* infestation. Similarly, Garzia *et al*. (2012) have reported 100% yield loss in unprotected tomato crops by *T. absoluta,* this finding also in concord with the present study. The present study was indicated that the yield loss depends on the level of leaves infestation and fruits bored. Therefore, appropriate timely spray schedule is an important component which is quite effective in mitigating the losses caused by the *T. absoluta*. This suggests the need for avoiding timely yield losses due *T. absoluta* in tomato.

5.5 Conclusions

Results of the study revealed that the control measures should be initiated when the *T. absoluta* larval population reaches 2.25 larvae per plant in glasshouse and 2-3 larvae per plant on field in order to prevent the population in reaching economic injury levels. The frequency of application time also depends on infestation level of *T. absoluta* and effectiveness of insecticides.

The larvae of *T. absoluta* is a serious pest of tomato in West Shewa, central Ethiopia and causes extensive feeding on the leaves due to which leaves dry up, become defoliated and even ceased flower and fruit setting and finally yield of fruits are significantly reduced.

CHAPTER SIX

Evaluation of bio-pesticides and sticky colors trap on management of *Tuta absoluta* on tomato in West Shewa, Central Ethiopia

6.1. Introduction

Tomato (*Lycopersicon esculentum* Mill.) belongs to the family Solanaceae is an important and remunerative vegetable crop grown around the world for fresh market and processing. It is widely cultivated in tropical, sub tropical and temperate climates and thus ranks third in terms of world vegetables production. Global tomato production is currently around 130 million tones, of which 88 million are destined for the fresh market and 42 million are processed (FAO, 2015). The leading tomato producing countries are China, India, Nigeria, Turkey, Egypt and United State. They account for 70% of global production (FAO, 2015; Fact fish, 2016). It is one of the economically important vegetable crops with a total area and production of 5,023,810 ha and 170,750,767 tons in 2014 (FAO, 2015).

In Ethiopia, due to a favorable climate conditions tomato production and the annual production of the crop is 30,700 tones of tomato fruits from about 5,026 ha of lands (FAO, 2015; Fact fish, 2016). Tomato is a seasonal plant which is one of the most economically important and widely grown vegetable crops in Ethiopia both in the rainy and dry seasons for their fruits by smallholder farmers and commercial state and private farms (AVRDC, 2014). It is also a source of basic raw materials required for fresh consumption and local processing industry for the production of processed tomato like tomato paste and tomato juice among others (EIA, 2012).

Tomato production faces many problems from several factors which lead to significant yield loss. Among the newly introduced insect species, some can become invasive, with subsequent significant economic impacts. Tomato leaf miner, *Tuta absoluta* (Meyrick) (Lepidoptera: Gelechiidae) is an oligophagous notorious pest of a number of economic crops including tomato.

It is a serious pest of tomato and other Solanaceous crops in many areas of the world causing severe damage and yield loss (El-Arnaouty *et al*., 2014; Tonnang *et al*., 2015; Bawin *et al*., 2015). Damage is caused by the larvae that mine leaves and bore into fruits which eventually facilitate plant pathogen invasion (EPPO, 2005). *T. absoluta* has recently invaded Europe, Africa and Asia (Desneux *et al*., 2011), being presently found in 63 countries of the world. It can affect tomato from seedling to fruit maturity stages. Feeding is caused by all larval stages throughout the plant growth period. On leaves, the larvae feed inside, forming irregular leaf mines which may later become dead tissue and serve as avenue for the entry of pathogens. The larvae can form extensive mines in the stems and tunnel into fruits, forming galleries which alter the normal growth of the plants and qualities of the fruits.

To overcome the problem of this pest, insecticides play a significant role globally. Tomato is a perishable commodity with a relatively short shelves life after harvest. This pest was initially reported in the Central Rift Valley region of Ethiopia in 2012 (Gashawbeza and Abyi, 2013). Since the time of its initial detection, the pest has caused serious damages to tomato in invaded areas (Gashawbeza, 2015) and it is currently considered as a key threat to Ethiopian tomato production. If no control measures are taken, the pest can cause up to 80% - 100% yield losses by attacking leaves, flowers, stems and fruits (Öztemiz, 2012).

Currently, chemical insecticides are heavily used by tomato growers against *T. absoluta*. However, the chemicals which are under use have negative impacts as the other chemical have. Hence, combination with other control methods like use of botanicals and entomopathogen fungi becomes imperative, as the continued use of chemical insecticides could harm non-target organisms (Landgren *et al.*, 2009) and the environment among others. The recommended waiting period which is required between application of conventional organophosphate pesticides group and consumption can hardly be afforded. On the other hands, Insecticide control against *T. absoluta* has been developed and widely applied in different countries such as several South American, Asia and Africa including Ethiopia, using the new active ingredient which is likely to be the core of the IPM program is Chlorantraniliprole (Coragen 200 SC). At present there are no other alternative insecticides to control *T. absoluta* in Ethiopia.

Different authors stated that pesticides is available in the market to control *Tuta absoluta* but most of these pesticides have not effective in the field as the pest have develop resistance to dozens of applied pesticides (Megido *et al.*, 2013). *T. absoluta,* with a high reproductive capacity and very short generations has an increased risk of developing resistance (Konus, 2014). For instance, resistance to abamectin, cartap, methamidophos and permethrin has been reported from Brazil (Siqueira *et al*., 2000). However, the need for alternative control methods is encouraged, that the newly introduced pesticides may be replaced the failure of insecticides against *T. absoluta* in the field. Even though, the use of new insecticide against *T. absoluta* is highly sought and effective control methods to reduce the population of this insect pest.

Pest control tactics are frequently based on different sticky trap catches as a cultural control measure. Insects are differentially attracted to colored surfaces, particularly yellow as a general insect attractant; a feature exploited among entomologists for collection of Coleoptera, Hemiptera, Hymenoptera and Thysanoptera (Kersting and Baspinar, 1995). But few studies were conducted in Diptera and Lepidoptera insects. Successful population monitoring is important for effective implementing insect control strategies, for properly timing control applications and for assessing their effects (Gillespie and Quiring, 1987). Sticky traps have been widely used to sample harmful and beneficial insects in wild and cultivated plants worldwide. Traps based on the response of insects to color have been widely used in integrated pest management program in diverse cultivated crops (Meyerdirk and Oldfield, 1985).

There is a real need to improve crop protection against *T. absoluta* and in the meanwhile reducing the use of synthetic insecticidal compounds (De Backer, 2014). Therefore, the current studies were initiated to evaluate the efficacy of botanicals, entomopathogenic fungi, insecticides and sticky trap colors, which were relatively safer to human and environment used against *T. absoluta* in the laboratory, glasshouse and open field conditions.

6.2. Materials and Methods

6.2.1. Evaluation of botanicals at different concentrations

6.2.1.1. Botanical collection and preparation

Fresh fruits and leaves of *Phytolacca dodecandra*, leaves of *Nicotiana* sp., and *Cymbopogon citratus* were collected and simultaneously separated from any infestation of disease and insect pest and then washed and cut into small pieces. *Allium sativum* was obtained from a local market in Ambo and used as fresh extract. *A. sativum* extraction was prepared according to the method described by Stoll (2000) using the following items: 250 gm of garlic fresh bulbs were chopped and strained in grinder, then the shopped bulbs were soaked in one liter of distilled water for one hour. *C. citratus* extraction was also prepared according to Stoll (2000) as follows: dried leaves of lemon grass were powdered and strained. Fifty grams of powdered dried leaves were soaked in two liters of distilled water for six hours. All the botanicals were ground, mixed, strained and filtered through cheese cloth and made stock solution. The stalk solution mixed with water at 5, 7.5 and 10% concentration level (v/v) in 100 ml of water.

Table 6.1: List of some crude extracts of medicinal plants used against Tomato leafminer, *Tuta absoluta* under laboratory and glasshouse conditions

Common name	Amharic name	Scientific name	Family	Used part(s)
Soap berry	Endod	*Phytolacca dodecandra*	Phytolacaceae	Leaf and seed
Tobacco	Timbaho	*Nicotiana* sp.	Solanaceae	Leaf and stalk
Lemon grass	Key sar	*Cymbopogon citrates*	Gramineae	Leaf
Garlic	Nich shinkurti	*Allium sativum*	Lilliaceae	Cloves
Neem	Yekinin zaf	*Azadirachta indica*	Meliaceae	Seed

6.2.1.2 Laboratory Experiment

The experiment was conducted under laboratory conditions at the room temperature of 22±2 ^{0}C.

Larvae were inserted in a Petri dish having 9cm diameter within the leaves and provided with coated cotton moist that is kept as fresh leaves of tomato that was collected from the glasshouse. All medicinal plants were prepared in three doses (50, 75, and 100 ml/L) from prepared stalk solution as follows: Endod fruit crude water extract (*P. dodecandra*), Endod leaf (*P. dodecandra*), Garlic clove (*A. sativum*), Tobacco leaves (*Nicotiana* sp.) local variety, Lemon grass leaf (*C. citrates*) and Neem seed (*A. indica*). Medicinal plant crude extracts were sprayed on *T. absoluta* larvae in the Petri dish using micro pipette. For the control treatment, larvae were treated with distilled water. After 24, 48, 72 and 120 hrs of exposure, the mortality rates at different concentrations were obtained. Dead larvae were removed as soon as possible in order to prevent decomposition which may cause rapid death of the remaining larvae.

6.2.1.3. Glasshouse Experiment

Seeds of tomato cultivar ‘Koshoro’ were planted on November 1st 2015 in the nursery site. The plants were transplanted into the experimental glasshouse after 40 days. The experimental pots were 20cm diameters and 25cm height totally 21 pots were prepared and filled with compost, loam soil and sand soil in the ratio of 1:1:2, respectively. The experiment was laid in a completely randomized block design (RCBD) with three replications. Purposefully the door and windows of the glasshouse was kept open for 24 hours for the entrance of moths. Hence, plants were infested under natural conditions. Planted tomato plants were sprayed with medicinal plant insecticides at recommended doses from laboratory studies. Each potted plant was covered with nylon cloth to avoid escape of larvae. Mortality data was recorded after treatment application of 1st, 3rd, 5th and 7th days. The best promising medicinal plant extracts were selected for further field experiment.

6.2.2. Evaluation of *Beauveria bassiana* and *Metarrhizium anisopliae*

6.2.2.1. Experimental design and materials used

The laboratory and glasshouse experiments were laid out in a Randomized Complete Block Design (RCBD) with three replications. Eight treatments were considered such treatments were *Beauveria bassiana* (PPRC-56) isolate at three different concentrations (2.5×10^7, 2.5×10^8 and 2.5×10^9 conidia ml^{-1}), similar concentrations were performed in *Metarhizium anisopliae* (PPRC-2) isolate. Chlorantraniliprole (Coragen 200 SC) as a standard check and untreated control was also considered for comparision.

Tomato cultivar “Koshoro” seeds were obtained from Melkasa Agricultural Research Center. The seeds were planted on the field for natural infestation of *T. absoluta*. Harboring *T. absoluta* larvae were collected from the fields of tomato and brought to the laboratory and glasshouse. The tomato leaf miner present on these collected tomato leaves were wrapped with wet cotton kept in plastic box (20 x 15 cm^2) in laboratory and glasshouse. After the emergency of adults rearing cages were prepared under glasshouse.

The insect was reared and maintained on tomato plants in the glasshouse until use. Leaves were examined under binocular microscope and *T. absoluta* larvae were counted. Spore suspensions were sprayed using a hand sprayer (1 liter of capacity). After treatment applications, the percent mortalities of the agents were observed at: 3, 5 and 7 days in the laboratory and 3, 5, 7 and 10 days under glasshouse conditions.

6.2.2.2. Fungus Culture and Viability Test

Isolates of *Beauveria bassiana* (PPRC-56) and *Metarhizium anisopliae* (PPRC-2) were obtained from Ambo Plant Protection Research Center. These entomopathogenic fungi were cultured on Potato Dextrose Agar (PDA) medium containing 20g glucose, 20g starch, 20g agar, and 1000 ml of distilled water in test tubes. The test tubes containing PDA medium was autoclaved at 121°C for 15-20 minutes and incubated at 27±1°C, 80±5% relative humidity and photophase of 12 hours for 15 days. The relative humidity was measured using Huger Hygrometer. The conidia were harvested by scraping the surface of 14-15 days old culture gently with inoculation needle. The mixture was stirred with a magnetic shaker for ten minutes. The hyphal debris was removed by filtering the mixture through fine mesh sieve. The conidial concentration of final suspension was determined by direct count using Haemocytometer. A serial dilution was prepared in distilled water containing 0.1% Tween-80 and preserved at 5°C until used.

Conidial viability was assessed according to Goettel and Inglis (1997). Three different concentrations were evaluated. The droplet of a diluted suspension was placed on a thin film of potato dextrose agar medium incubated at 27±1°C and 80±5% relative humidity in the dark for 24 hours. The conidia were stained with lacto-phenol cotton blue and germination was observed under microscope.

6.2.2.3. Mortality of *T. absoluta* under laboratory

The concentration of the stock suspension was adjusted to 2.5 x 10^7. 2.5 x 10^8 and 2.5 × 10^9 conidia ml^{-1} using an improved neubaour heamocytometer. To evaluate the efficiency of each of the fungal isolates on *T. absoluta*, 20 larvae were placed on a filter paper in 9 cm diameter petri-dish and 100 µl of the suspension was then spread. On similar trend the suspension was spread in glasshouse using hand sprayed was performed and after 3rd days of observation all counted larvae were collected from plants to brought in the laboratory to determined how many *T. absoluta* larvae were dead without being infested with fungal isolates. A control treatment was sprayed with only sterile distilled water as negative control.

6.2.3. Evaluation of Insecticides

6.2.3.1. Laboratory and Glasshouse Experiments

Laboratory and glasshouse studies were under taken to evaluate the newly introduced Prosuler Oxymatrin (Levo 2.4SL^{TM}) and Emamectin Benzoate (Prove 1.9 E.C^{TM}) insecticides in three different concentrations (1ml, 2ml and 3ml/liter). The experiments were conducted at a temperature of 22 ± 2^0C and 50 ±5% RH for a period of 72 hrs in the laboratory. Larvae were inserted in a Petri dish within the leaves and provided with coated cotton moist that was kept as fresh leaves of tomato that had collected from the glasshouse. Seventy two hours latter larval percent mortalities were counted. The best performing, insecticides of each concentration were selected for subsequent glasshouse experiments.

In glasshouse 24 pots were prepared and filled with compost, loam soil and sand soil in the ratio of 1:1:2, respectively. A Pot having a height of 25cm and a diameter of 20cm was used. The experiment was arranged in a randomized completely block design (RCBD) in three replications.

Purposefully the door and windows of the glasshouse was kept open for 24 hours for the entrance of moths. Hence, plants were infested under natural conditions. After treatment application of 24 hours the counted larvae in the leaves were collected and put under laboratory till five days to observe the mortality rate. Mortality data were recorded after application at 1st, 3rd and 5th days.

Table 6.2: List of experimental treatments at different Concentration

S. N.	Common name	Trade name	Concentration (%)
1	Prosuler Oxymatrin	Levo 2.4SL™	1, 2 and 3ml/ liter.
2	Emamectin Benzoate	Prove 1.9 E.C™	1, 2 and 3ml/liter.
3	Prosuler Oxymatrin + Emamectin Benzoate	Levo 2.4SL + Prove 1.9 E.C	2:2 ml/liter
4	Tetraniliprole	Vayego 200 SC	2ml/liter
5	Chlorantraniliprole	Coragen 200 SC	1 ml/ liter
6	Control		

6.2.4. Open Fields Experiment

6.2.4.1. Description of the study area

The field experiment was carried out on farmers' field at three different districts of West Shewa Zone, Oromia Regional state, Ethiopia, such as Ambo, Dendi and Toke Kutaye districts. Ambo is at geographical coordinate of 858'59.988"N latitude and 3751'0.000"E longitude with an altitude of 2076 meter above sea level (Briggs, 2012). Toke Kutaye districts is located at 126 km west of Addis Ababa having an altitude of 1990 meter above sea level, latitude of 0859' 01.100" N and longitude of 3746'27.600"E. The average annual rainfall is 1028.7 mm and maximum and minimum temperatures of the area 29.6°C and 11.8°C, respectively. The geographical location of the study area of Dendi district also having an altitude of 2272 meter above sea level, latitude of 4064' 81.00"N and longitude of 9978' 63.00"E.

6.2.4.2. Experimental materials and layout

Tomato cultivar “Koshoro” seeds were drilled in flat seed bed having 2m length and 1.2m width ($2.4m^2$) for raising seedlings for the experiment. Nine selected treatments were considered for the open field experiments such as *A. sativum, Nicotiana* sp., *C. citrates, A. indica, B. bassiana, M. anisopliae*, Tetraniliprole (Vayego 200 SC), Chlorantraniliprole (Coragen 200 SC) as a standard check and untreated control was also considered for comparision. The field study was laid out in a Randomized Complete Block Design with three replications. Plant to plant and row-to-row distances were kept at 40 cm and 80 cm, respectively. The experimental plot was consisted of an area 4m long and 3m wide, with a total area of $12m^2$ and a working area of 9.6 m^2/plot comprising the central row. All agronomic practices were conducted as recommended.

6.2.4.3. Data collection

The field experiment was scouted every week for the signs and symptoms of tomato damage and occurrence until leaves infested by *T. absoluta*. Before treatment applications pre-spray counts were made and then treatment application follows. The treatments were applied at vegetative, flowering and fruit setting stages of the crop. After treatment applications, five plants were randomly selected in each treatment and data on *T. absoluta* larvae both live and dead (per plant) were observed under microscope at 3, 5, 7 and 10 days intervals. After three days of post spray, all leaves were counted during pre spray would collected and brought to laboratory. The dead larvae were removed and live larvae were observed under microscope until 10 days of post spray. Larvae were considered as dead when they were not able to move back to the ventral position after being placed on their dorsum. Finally, the fruit yield per hectare was recorded.

6.2.4.4 Data analysis

Obtained data were transformed using Arcsine transformed and analysis of variance was computed using SAS program version 9.1 (SAS, 2009). The mean percent mortality was corrected using Abbott's formula (Abbott, 1925) and efficacy analysis was done based on data transformation to Arcsine $\sqrt{x+0.5}$ when necessary according to Gomez and Gomez (1984).

$$\text{CM (\%)} = \frac{[T\,(\%) - C(\%)]}{[100 - C(\%)]} X100$$

Where: CM (%) - Corrected mortality

T- Mortality in treated insects

C- Mortality in untreated insects

6.2.5. Evaluation of Sticky Trap Colors

The present study was carried out under glasshouse conditions during 2016-2017. The experiment was arranged in Randomized Complete Block Design (RCBD) with four replications. The following five treatments were used in different sticky trap colors namely: blue, green, red, yellow and white were hanged in glasshouse at different phonological stages (vegetative, flowering and fruit setting). The traps were distributed between tomato plants at constant height (60 cm above the ground). The captured *T. absoluta* moths were collected weekly and counted until the end of the crop harvest.

6.2.5.1. Data Analysis

The effect of different colors and phonological stages of the plants were evaluated for caught of *T. absoluta*. The data was subjected to analysis of variance (ANOVA) and the means were compared by Least Significant Different (LSD) test at 0.05 levels, using SAS program version 9.1 (SAS, 2009).

6.3. Results

6.3.1. Effect of crude extracts of medicinal plants against *T. absoluta* on tomato

6.3.1.1. Effect of medicinal plants on larvae of *T. absolute* under Laboratory Conditions

Efficacy of medicinal plant extracts against *T. absoluta* in the laboratory is shown in Table 6.3. All treatments significantly (P < 0.01) different on plant treated with various treatments and reduced the total number of *T. absoluta* larvae per treatment. Effect of medicinal plant crude extracts was evaluated against *T. absoluta* at three different concentrations (5, 7.5 and 10%) after four exposure time (24, 48, 72 and 120 hours). Table 25 below revealed that medicinal plant extract at 10% concentration *A. indica* seed had maximum toxicity (98.33%) against *T. absoluta* after which *C. citratus* (96.67%) while *P. dodecandra* leaf showed the minimum mortality (55.0%) after 120 hours of application. On the other hand, at 7.5% concentration biocidal potential of *C. citratus* was high (91.67%) following by *A. indica* seed (90.0%) and *A. sativum* (86.67%). Low toxicological effect was observed of some extracts at 5% concentration especially *P. dodecandra* leaf and seed showed least mortality (36.67%). Similarly, among time interval for exposure of plant extracts was significantly recoded at all concentrations as shown in Table 6.3. Medicinal plant extract did not showed quick response after 24 hours but effect was significantly high after 72 and 120 hours as killed more larvae of *T, absoluta*.

Table 6.3: Mean efficacy of some medicinal plant extracts against *T. absoluta* larvae at different rates and time of exposure in the laboratory

			Mean Efficacy percentage (%)			
S.N	**Treatments**	**Conc.**	**After 24 hrs**	**After 48 hrs**	**After 72 hrs**	**After 120 hrs**
		5%	21.67^{gh}	33.33^{ef}	35.00^{hi}	40.0^{h}
1	Endod seed	7.5%	26.67 f^{g}	36.67^{def}	38.33^{h}	43.33^{h}
	(*P. dodecandra*)	10%	31.67^{ef}	36.67^{def}	53.33^{ef}	56.67^{fg}
		5%	23.33^{h}	26.67^{g}	28.33^{j}	36.67^{h}
2	Endod leaf	7.5%	23.33^{gh}	26.67^{g}	33.33^{ij}	36.67^{h}
	(*P. dodecandra*)	10%	38.33^{fg}	31.67^{fg}	51.67^{fg}	55.0^{g}
		5%	38.33^{d}	38.33^{de}	46.67^{g}	66.67^{ef}
3	Garlic cloves	7.5%	38.33^{d}	53.33^{b}	63.33^{bc}	86.67^{bc}
	(*A. sativum*)	10%	56.67^{b}	66.67^{a}	73.33^{a}	95.0^{ab}
		5%	38.33^{d}	56.67^{a}	56.67^{def}	61.67^{efg}
4	Tobacco	7.5%	46.67^{c}	58.33^{a}	61.67^{cde}	70.0^{de}
	(*Nicotiana* sp.)	10%	66.67^{a}	71.67^{a}	71.67^{a}	80.0^{cd}
		5%	33.33^{de}	41.67^{cd}	53.33^{ef}	88.33^{abc}
5	Lemongrass	7.5%	36.67^{de}	46.67^{c}	56.67^{def}	91.67^{ab}
	(*C. citratus*)	10%	38.33^{d}	56.67^{b}	63.33^{bc}	96.67^{ab}
		5%	46.67^{c}	46.67^{c}	58.33^{cde}	90.0^{abc}
6	Neem seed	7.5%	66.67^{a}	68.33^{a}	68.33^{ab}	90.0^{abc}
	(*A. indica*)	10%	66.67^{a}	68.33^{a}	71.67^{a}	98.33^{a}
7	Control		0.00^{i}	0.00^{h}	0.00^{k}	6.67^{i}
	LSD		3.73	3.81	3.74	10.81
	CV (%)		4.54	4.14	3.73	7.17
	MSE ±		1.68	1.72	1.69	4.87

Note: Means with the same letter(s) in columns are not significantly different for each other.
All treatment effects were highly significant at $p<0.01$ (DMRT)

6.3.1.2. Effect of medicinal plant crude extract on larvae of *T. absolute* under glasshouse conditions

Under glasshouse conditions, toxicity of medicinal plant crude extracts were evaluated against *T. absoluta* at 10% concentrations after five exposure dates (1, 3, 5 and 7 days). The tested medicinal plant crude extracts at 10% concentration illustrated in (Table 6.4) showed that significant (P < 0.01) differences were observed among the treatments from the untreated control. The presented data are pertaining to mean percent reduction of *T. absoluta* larvae reveals that *A. indica* seed had maximum toxicity 66.54% against *T. absoluta* after which *Nicotiana* sp. 62.10% while *P. dodecandra* seed showed significantly lowest effect mortality 36.51% for entire days. In general, the results indicated that each medicinal plant extract caused significant mortality rate against larvae of *T. absoluta* after 120 hours exposure in the laboratory and after 7 days in the glasshouse. Therefore, these findings suggested that medicinal plant insecticides are a good alternative management option of *T. absoluta*.

Table 6.4: Mean efficacy of medicinal plant crude extracts against *T. absoluta* larvae at different rates and time of exposure under glasshouse

S.N	Treatments	Mean Efficacy Percentage			
		After 1 day	After 3 day	After 5 day	After 7 day
001	*P. dodecandra* seed	18.65^{d}	24.03^{c}	36.51^{bc}	36.51^{b}
002	*P. dodecandra* leaf	23.33^{cd}	26.11^{c}	32.72^{c}	36.94^{b}
003	*A. sativum* clove	43.45^{a}	58.59^{a}	58.93^{a}	59.92^{a}
004	*Nicotiana* sp.	33.61^{ab}	45.71^{ab}	45.83^{abc}	62.10^{a}
005	*C. citratus*	30.63^{bc}	33.97^{abc}	47.62^{ab}	57.9^{a}
006	*A. indica* seed	24.52^{bcd}	46.79^{a}	50.95^{a}	66.54^{a}
007	Control	0.00^{e}	0.00^{d}	0.00^{d}	0.00^{c}
	LSD	6.17	12.0	7.98	11.64
	CV (%)	8.91	14.98	8.83	11.59
	SE±	2.47	4.81	3.20	4.67

Note: Means with the same letter(s) in columns are not significantly different for each other.
All treatment effects were highly significant at P<0.01 (DMRT)

T. absoluta are capable of reducing the total yield of *tomato* plants as well as facilitate it to secondary attack by pathogens. The application of botanicals on T. *absoluta* larvae resulted good performance. The results revealed that *A. sativum, Nicotiana* sp. and *C. citrates* were gave promising results to minimize the impact of *T. absoluta* larvae on leaves and fruit's, similarly to the standard treatment *A. indica* seeds.

6.3.2. Effects Entomopathogenic fungi in the management of *T. absoluta*

6.3.2.1. Laboratory Experiment

The laboratory results showed that the percent mortality of *T. absoluta* larvae due to entomopatogenic fungi significant ($P < 0.01$) differences among the concentrations of *B. bassiana* and *M. anisopliae* (Table 6.5). All concentrations of *B. bassiana* caused mortality of *T. absoluta* above 75% after treatment application of 7 days, indicating that 2.5×10^9 conidia ml^{-1} caused the highest mortality. For *M. anisopliae*, at the concentration of 2.5×10^9 conidia ml^{-1}, mortalities obtained with all concentrations were higher than 50%; however, the concentrations did differ statistically from each other after treatment application, and the highest mortality of *T. absoluta* larvae were observed with concentration 2.5×10^9 (87.5%) under laboratory conditions (Table 6.5).

After 7th day of treatment application *B. bassiana* raveled that 79.17%, 83.33% and 95.83% mortality at 2.5×10^7, 2.5×10^8 and 2.5×10^9 concentrations, respectively. Similarly, *M. anisopliae* concentrations showed that 66.67%, 79.17% and 87.50% mortality at 2.5×10^7, 2.5×10^8 and 2.5×10^9 concentrations, respectively. There was a highly significant variation among the concentrations in causing mortality of *T. absoluta* larvae. The lowest mean percent mortality was caused by the *B. bassiana* at 3rd days of observation 37.50% which was not significantly different from *M. anisopliae* at 3^{rd} days of 58.33 %. The highest mortality of *T. absoluta* was caused by *B. bassiana* 95.83% which did not significantly differ from the *M. anisopliae* which was 87.50% mortality. Based on the results of the virulence assays of *B. bassiana* and *M. anisopliae* had time taken by the three concentrations to caused percent mortality of *T. absoluta*. The effects of the concentrations varied significantly ($P < 0.01$) with the lowest (3 days) recorded from concentration 2.5×10^7 in *B. bassiana* followed by *M. anisopliae* (5 days) which recorded 58.33%. In the 7^{th} day of the three concentrations the highest was recorded due to *B. bassiana* which was significantly ($P < 0.01$) different from *M. anisopliae* concentrations.

The comparison among the different concentrations and treatments against *T. absoluta* the results indicated good performance and gradually increased from 3 to 7 days treatment applications. The percent mortality according to Abbott formula (1925), both agents at 2.5×10^9 conidial/ml gave statistically no significant ($P < 0.01$) differences from the standard check (Coragen 200 SC) while highly significant different from untreated check after 3 days of treatment application (Table 6.5).

Concentration-response test

Percent mortality of *T. absoluta* larvae at different concentrations of *B. bassiana* and *M. anisopliae* shown in Table 6.5. There were no significant differences in mortality rates within each concentration except for the concentration of 2.5×10^9 conidia/ml in which the *B. bassiana* showed significantly higher mortality than *M. anisopliae*. The results of all concentrations except the concentration 2.5×10^7 in *B. bassiana* revealed the lowest at 3^{rd} days of application but also highly significantly ($P < 0.01$) among the concentrations requiring higher concentration (2.5 $10^9 \times 10^9$conidia ml^{-1}).

Table 6.5: Mean percent mortality of *T. absoluta* treated with fungal isolates at different concentration over time under laboratory conditions

Treatments		**Mean percent mortality after treatment application**		
	Conc.	**3 days**	**5 days**	**7 days**
Beauveria bassiana (PPRC-56)	2.5×10^{7}	37.50^{c}	58.33^{c}	79.17^{b}
	2.5×10^{8}	70.83^{bc}	70.83^{bc}	83.33^{ab}
	2.5×10^{9}	79.17^{ab}	91.67^{ab}	95.83^{a}
Metarhizium anisopliae (PPRC-2)	2.5×10^{7}	58.33^{bc}	58.33^{bc}	66.67^{b}
	2.5×10^{8}	58.33^{bc}	79.17^{bc}	79.17^{ab}
	2.5×10^{9}	66.67^{bc}	83.33^{abc}	87.50^{ab}
Chlorantraniliprole (Coragen 200 SC)		95.83^{a}	95.83^{a}	95.83^{a}
Control (water)		0.0^{d}	0.0^{d}	0.0^{c}
LSD at 0.01		20.15	19.95	21.82
CV (%)		16.47	14.19	14.61
SE±		8.29	8.21	8.96

Note: Means with the same letter(s) in columns are not significantly different for each other.
All treatment effects were highly significant at p<0.01 (DMRT)

B. bassiana, strain presented the highest pathogenicity on *T. absoluta* larvae with 95.83% an average mortality, $LC_{50} = 2.5\times10^{9}$ conidia ml^{-1} and LT_{50}=5.01 days (Table 6.8). *M. anisopliae* strain was the most virulent on *T. absoluta* larvae presenting 87.50% mortality, $LC_{50} = 2.5\times10^{9}$ conidia ml^{-1} and LT_{50}=4.82 days. The LT_{90} values to *B. bassiana* strains on *T. absoluta* larvae ranged from 8.06 to 9.32 days, and for *M. anisopliae* strains on *T. absoluta* larvae ranged from 8.14 to 9.04 days (Table 6.6). The *M. anisopliae* strain presenting the lowest LC_{90} on *T. absoluta* larvae was 2.5×10^{9} conidia ml^{-1}) and the highest LC_{90} was presented by *B. bassiana* 2.5×10^{7} conidia ml^{-1}). Finally, for *T. absoluta* larvae the LC_{90} of both *B. bassiana and M. anisopliae* varied from 2.5×10^{7} to 2.5×10^{9} conidia ml^{-1} concentration (Table 6.7).

Table 6.6: Median lethal time (LT_{50}) and (LT_{90}) of *B. bassiana* and *M. anisopliae* against *T. absoluta*

Treatments		95% Confidence Limit		95% Confidence Limit	
	(Conidia ml^{-1})	LT_{50} (days)	Slope ± SE	LT_{90} (days)	Slope ± SE
B. bassiana	2.5 x 10^{7}	5.45	3.17 ± 0.52	9.32	2.28 ± 0.48
(PPRC-56)	2.5 x 10^{8}	5.21	3.80 ± 0.61	8.87	2.66 ± 0.37
	2.5 x 10^{9}	5.01	4.29 ± 0.82	8.06	3.06 ± 0.68
M. anisopliae	2.5 x 10^{7}	5.14	3.64 ± 0.56	9.04	2.86 ± 0.46
(PPRC-2)	2.5 x 10^{8}	5.02	3.63 ± 0.48	8.56	3.04 ± 0.58
	2.5 x 10^{9}	4.82	3.31 ± 0.64	8.14	3.31 ± 0.72

Table 6.7: Mean concentration (LC_{50}) and (LC_{90}) of *B. bassiana* and *M. anisopliae* (100µ /larva) of *T. absoluta*

Treatments				95% Confidence Limit	
	(Conidia ml^{-1})	LC_{50}	Slope ± SE	LC_{90}	Slope ± SE
B. bassiana	2.5 x 10^{7}	4.23 ± 0.52	4.29 ± 0.82	9.68 ± 0.82	3.36 ± 0.41
(PPRC-56)	2.5 x 10^{8}	3.93 ± 0.61	3.80 ± 0.61	9.22 ± 0.61	2.64 ± 0.38
	2.5 x 10^{9}	3.50 ± 0.82	3.17 ± 0.52	8.46 ± 0.52	2.45 ± 0.28
M. anisopliae	2.5 x 10^{7}	3.59 ± 0.56	3.31 ± 0.64	8.71 ± 0.64	2.47 ± 0.77
(PPRC-2)	2.5 x 10^{8}	3.26 ± 0.48	3.63 ± 0.48	8.13 ± 0.48	2.63 ± 0.58
	2.5 x 10^{9}	2.91 ± 0.64	3.64 ± 0.56	7.52 ± 0.56	3.54 ± 0.72

6.3.2.2. Glasshouse Experiment

The entomopathogenic fungal isolates were tested at three different concentrations for their percent mortality against *T. absoluta* in glasshouse to explore their potential to manage the pest population. Percent mortality of *T. absoluta* larvae were calculated for the different concentrations of the two isolates and showed increasing mortality with increasing spore concentration. Cumulative mortality of *T. absoluta* larvae over exposure period (3, 5, 7 and 10 days) was significantly ($P < 0.01$) different for fungi isolates (Table 6.8). On the 3rd days of exposure maximum mortality 91.84 recorded from standard check, while the untreated control had 2.78% mortality. These were significantly different from all concentrations of the fungal isolates. Among the concentrations of entomopathogenic fungi maximum percent mortality was recorded at 2.5 x 10^9 conidial ml^{-1} of *B. bassiana* (84.04%) followed by *M. anisopliae* (76.31%) on 10th day after treatment application. At the highest concentration of conidial ml^{-1}, all *B. bcassiana* concentration gave the highest percent mortality (Table 6.8). The results indicated for pathogencity of all the concentrations revealed that all of them are virulent, even three days after application causing significant mortality up to 64.05% when compared with untreated control.

Table 6.8: Mean percent mortality of Entomopathogenic fungi at different concentration on larvae *T. absoluta* under glasshouse conditions

Treatments	Conc.	Mean percent mortality after treatment application			
		3 days	**5 days**	**7 days**	**10 days**
	2.5 x 10^7	43.85^c	57.57^{bc}	75.17^{ab}	81.64^{ab}
Beauveria bassiana	2.5 x 10^8	56.27^{bc}	56.27^{bc}	73.0^{ab}	76.62^{abc}
(PPRC-56)	2.5 x 10^9	63.84^b	67.05^b	67.05^{bc}	84.04^{ab}
	2.5 x 10^7	38.76^c	42.93^c	53.37^c	53.37^d
Metarhizium anisopliae	2.5 x 10^8	44.07^c	51.98^{bc}	61.49^{bc}	64.65^{cd}
(PPRC-2)	2.5 x 10^9	64.05^b	68.21^{bc}	71.98^{abc}	76.31^{abc}
Chlorantraniliprole (Coragen 200 SC)		91.84^a	91.84^a	91.84^a	91.84^a
Control		2.78^d	4.76^d	4.76^d	7.14^e
LSD at 0.01		18.61	21.12	21.73	14.85
CV (%)		15.10	15.65	14.21	9.09
SE±		7.66	8.69	8.94	6.11

Note: Means with the same letter(s) in columns are not significantly different for each other.
All treatment effects were highly significant at $p<0.01$ (DMRT)

A positive relationship was recorded between mortality percentages and concentrations among the *B. bassiana* and *M. anisopliae* concentrations. Concurrently, with the increase in conidia concentration, a reduction in LT_{50} was observed. Concentrations of 2.5 x 10^9 from *B. bassiana*, at the concentrations 2.5 x 10^8 and 2.5 x 10^9 conidia ml^{-1}, presented the shortest lethal time (Table 6.8).

The effect of entomopathogenic fungi were evaluated to determined the concentrations with high efficacy against larvae *T. absoluta* under laboratory and glasshouse conditions. Both fungal isolates were found to be pathogenic to *T. absoluta*. Though, there was a variation in their virulence against *T. absoluta*. The percent mortality for all the concentrations gradually increased. The spore formation appeared on the larvae of *T. absoluta* took place after treatment exposure of the concentrations of the two isolates starting from the day three after treatment exposure, and thereby no hatched larvae were appeared in the concentrations of both isolates comparing the control treatment. The *M. anisopliae* in all concentrations were significantly less effective when compared with that of *B. bassiana* in terms of virulence. Virulence due to *B. bassiana* on 10th day was not significantly different from each other. This indicated that all *B. bassiana* concentrations were the best management option of *T. absoluta*.

6.3.3. Effects of synthetic insecticides on the management of *T. absoluta*

6.3.3.1. Effect of insecticides against *T. absoluta* under laboratory

The data on the effectiveness of insecticides against *T. absoluta* revealed that highly significant ($P < 0.01$) differences among treatments in laboratory after treatment exposure of 72 hours (Table 6.11). The results showed that the insecticide Coragen 200 SC and the mixture of Prove 1.9 E.C + Levo 2.4 SL were found to be highly significant ($P < 0.01$) superior over the control 95.55% mortality followed by the mixture of Prove 1.9 E.C and Levo 2.4 SL 2% concentration 80.0% mortality. The lowest mortality percent recorded at 1% concentration of Levo 2.4 SL, 22.22% followed by Prove 1.9 E.C, 31.11% were observed after 24 hours treatment application while they were found to be highly significant ($P < 0.01$) and superior over the control (Table 6.9).

The mortality of *T. absoluta* was higher after 48 and 72 hours of insecticidal application as compared with 24 hours because of the lethal influence gradually the insect physiology which culminated in the highest control. The best performance by Chlorantraniliprole (Coragen 200 SC) among the insecticides could be due to their desirable mode of action that effectively blocked a physiological mechanism in the target insect right 48 hours of application and after 72 hours mortality reached the highest percent mortality followed by the mixture of Emamectin benzoate (Levo 2.4 SL) and Prosuler oxymatrin (Prove 1.9 E.C). These findings indicated that increase in mortality rate in all the treatments in concentration and time dependent manner.

Table 6.9: Mean percent mortality of *T. absoluta* caused by different concentrations of insecticides after treatment exposure at 24, 48 and 72 hours intervals under laboratory conditions

S.N	Treatments	Conc.	Mean Efficacy percent		
			After 24 hrs	**After 48 hrs**	**After 72 hrs**
1	Emamectin benzoate (Prove 1.9 E.C)	1%	31.11^{cd}	42.22^{de}	51.11^{cd}
		2%	37.78^{c}	44.45^{cd}	57.78^{c}
		3%	37.78^{c}	46.67^{c}	60.0^{c}
2	Prosuler oxymatrin (Levo 2.4 SL)	1%	22.22^{d}	33.33^{e}	37.78^{e}
		2%	35.55^{c}	35.55^{de}	42.22^{de}
		3%	35.55^{c}	37.78^{cde}	42.22^{de}
3	Prove 1.9 E.C + Levo 2.4 SL	2:2%	62.22^{b}	77.78^{b}	80.0^{b}
4	Chlorantraniliprole (Coragen 200 SC)	1%	93.33^{a}	95.55^{a}	95.55^{a}
5	Control		0.0^{e}	0.0^{f}	0.0^{f}
	LSD at 0.01		12.79	12.68	12.27
	CV (%)		13.96	12.26	11.07
	SE ±		5.36	5.31	5.15

Note: Means with the same letter(s) in columns are not significantly different for each other.
All treatment effects were highly significant at $p<0.01$ (DMRT)

6.3.3.2. Effect of insecticides against *T. absoluta* under Glasshouse

The data on the effectiveness of insecticides sprayed to overcome the *T. absoluta* revealed a highly significant ($P < 0.01$) difference among treatments in glasshouse (Table 6.10). The presented data are pertaining to mean percent reduction of *T. absoluta* showed that, among all the treatments the Coragen 200 SC gave the highest mortality 89.68% percent mortality followed by the mixture of Prove 1.9 E.C + Levo 2.4 SL insecticides 78.94% percent mortality within 5 days of application. Prove 1.9 E.C and Levo 2.4 were recorded to be 39.81% and 25.28% significantly inferior in efficacy against *T. absoluta* within five days of application, respectively. All insecticides gave the mean percent reduction of *T. absoluta* and significantly higher than control treatments.

Management of resistance to prevent or delay the development of resistance to an insecticide and cross resistance to additional insecticides is necessary for increasing the chance of insecticide control of *T. absoluta*. This glasshouse and laboratory studies suggested that the good performance of the tested compounds, Chlorantraniliprole (Coragen 200 SC) followed by (a mixture of Emamectin Benzonate and Prosuler Oxymatrin were important for the managment of tomato leafminer, *T. absoluta.*

Table 6.10: Mean efficacy of two commercial insecticides in different concentrations against tomato leafminer, *T. absoluta* under glasshouse conditions

S.N	Treatments	Conc (%)	Mean Mortality percent		
			1 day	3 day	5 day
1	Emamectin benzoate (Prove 1.9 E.C)	2%	30.55^{c}	34.26^{c}	39.81^{b}
2	Prosuler oxymatrin (Levo 2.4 SL)	2%	17.49^{d}	22.25^{c}	25.28^{c}
3	Prove 1.9 E.C + Levo 2.4 SL	2:2%	51.52^{b}	74.77^{ab}	78.94^{a}
4	Chlorantraniliprole (Coragen 200 SC)	1%	76.59^{a}	89.68^{a}	89.68^{a}
5	Control		3.33^{d}	7.50^{d}	7.50^{d}
	LSD at 0.01		12.42	24.70	17.83
	CV (%)		12.63	19.73	13.49
	SE±		4.53	9.02	6.51

Note: Means with the same letter(s) in columns are not significantly different for each other.
All treatment effects were highly significant at $p<0.01$ (DMRT)

6.3.4. Effects of Bio-pesticides in the management of *T. absoluta* on field conditions

6.3.4.1. Effect of bio-pesticides on larvae of *T. absoluta*

Effect of bio-pesticides on larvae of *T. absoluta* showed that significant ($P < 0.01$) differences were found among the treatments in all districts (Table 6.11). After 3 days of treatment application, about 0 – 96.19% mean larval percent mortalities were observed due to application of different bio-pesticides when compared to untreated control and standard check (2.09%) and (94.74%), respectively. The highest percent mortality was recorded in Vayego 200 SC 96.19% and the lowest percent mortality was observed in *Nicotiana* sp. 39.35% but both entomopathogenic fungi (*B. bassiana* and *M. anisopliae)* were no effects on *T. absoluta* within 3 days of treatment application, because of the establishment of fungi on the larvae of insect pests take some days.

Data in (Table 6.10 and 6.11) showed that at the 5th day treatment application of the experiment, mortality percentage of *T. absoluta* on tomato fruit were better than the 3rd day in all treatment plots and in all locations except the standard check (Coragen 200 SC) it gave high percent mortality in all districts. The other treatments percent mortality gradually, increased from 0% to 74.26% after 10 days from the third day application of the experiment. Meanwhile, in the plots receiving bio or chemical insecticides experienced a marked gradual increased in percent mortality and decreased the infestation level of *T. absoluta*. This effect was most evident post spraying by *A. sativum* (76.40) and *C. citrates* (77.30) both in Dandi and Ambo districts, whereas *A. indica* (76.07) in Toke kutaye district. The least toxic effect was exhibited by *Nicotiana* sp. 46.29, 49.90 and 51.85 in Ambo, Dandi and Toke kutaye districts, respectively.

The chemical insecticide Tetraniliprole (Vayego 200 SC) showed very high toxic effect on *T. absoluta* following 10 days treatment applications, and percent mortality was low in all locations as compared with other treatments but highly significant ($P < 0.01$) different from untreated control. In general, after 5th, 7th and 10th day of treatment application the results of all treatments proved better percent mortality on larvae of *T. absoluta* as compared with 3rd day of application (Table 6.11).

Table 6.11: Mean efficacy percentage of bio-pesticides against *T. absoluta* after 3 days of treatment application on field conditions.

	Locations			
Treatments	**Ambo**	**Dendi**	**Toke kutaye**	**Mean**
Allium sativum	50.86^{bc}	57.57^{bcd}	43.10^{c}	50.51^{b}
Nicotiana sp.,	33.33^{bc}	43.60^{d}	41.11^{c}	39.35^{b}
Cymbopogon citratus	59.26^{b}	66.67^{b}	38.91	54.55^{b}
Azadirachta indica	27.61^{c}	61.24^{bc}	42.26^{c}	43.70^{b}
Beauveria bassiana	0.00^{d}	0.00^{e}	0.00^{d}	0.00^{c}
Metarhizium anisopliae	0.00^{d}	0.00^{e}	0.00^{d}	0.00^{c}

Vayego 200 SC	96.78^{a}	93.38^{a}	98.42^{a}	96.19^{a}
Coragen 200 SC	91.41^{a}	96.97^{a}	95.83^{a}	94.74^{a}
Control	6.27^{d}	0.00^{e}	0.00^{d}	2.09^{c}
LSD at 0.01	16.11	8.95	12.77	13.53
CV (%)	20.47	20.29	16.57	19.11
SE ±	6.76	3.75	5.35	5.67

Note: Means with the same letter(s) in columns are not significantly different for each other.
All treatment effects were highly significant at $p<0.01$ (DMRT)

Table 6.12: Mean efficacy percentage of bio-pesticides against *T. absoluta* after 5 days of treatment application on field conditions.

	Locations			
Treatments	**Ambo**	**Dendi**	**Toke kutaye**	**Mean**
Allium sativum	62.48^{d}	69.80^{b}	48.90^{bc}	60.39^{b}
Nicotiana sp.,	41.67^{b}	60.12^{b}	41.10^{bc}	47.63^{bc}
Cymbopogon citratus	65.74^{b}	60.32^{b}	49.14^{bc}	58.40^{b}
Azadirachta indica	52.31^{b}	59.02^{bc}	52.80^{b}	54.71^{b}
Beauveria bassiana	37.29^{b}	13.35^{de}	29.17^{cd}	26.60^{d}
Metarhizium anisopliae	45.19^{b}	30.16^{cd}	16.31^{d}	29.45^{cd}
Vayego 200 SC	96.78^{a}	93.38^{a}	98.42^{a}	96.19^{a}
Coragen 200 SC	91.41^{a}	96.97^{a}	95.83^{a}	94.74^{a}
Control	6.27^{c}	6.27^{e}	0.00^{d}	4.18^{e}
LSD at 0.01	18.66	16.36	12.77	13.59
CV (%)	18.66	20.65	13.32	13.19
SE ±	8.37	6.86	5.35	5.70

Note: Means with the same letter(s) in columns are not significantly different for each other.
All treatment effects were highly significant at $p<0.01$ (DMRT)

Table 6.13: Mean efficacy percentage of bio-pesticides against *T. absoluta* after 7 days of treatment application on field conditions.

Treatments	Locations			
	Ambo	**Dendi**	**Toke kutaye**	**Mean**
Allium sativum	68.29^{b}	69.10^{bc}	70.45^{b}	69.28^{b}
Nicotiana sp.,	46.22^{bc}	50.80^{cd}	51.85^{b}	49.65^{c}
Cymbopogon citrates	65.66^{bc}	77.3^{b}	64.71^{b}	69.22^{b}
Azadirachta indica	69.69^{b}	60.10^{bcd}	71.29^{b}	67.03^{b}
Beauveria bassiana	47.75^{bc}	54.13^{cd}	67.04^{b}	56.31^{bc}
Metarhizium anisopliae	55.71^{bc}	39.68^{d}	58.40^{b}	51.26^{c}
Vayego 200 SC	96.78^{a}	93.38^{a}	98.42^{a}	96.19^{a}
Coragen 200 SC	94.19^{a}	93.64^{a}	95.83^{a}	94.55^{a}
Control	9.97^{d}	6.23^{e}	0.00^{c}	5.40^{d}
LSD at 0.01	21.44	13.35	11.67	12.22
CV (%)	18.69	20.65	19.78	10.44
SE ±	8.99	5.60	4.89	5.13

Note: Means with the same letter(s) in columns are not significantly different for each other.
All treatment effects were highly significant at P<0.01 (DMRT)

Table 6.14: Mean efficacy percentage of bio-pesticides against *T. absoluta* after 10 days of treatment application on field conditions.

	Locations			
Treatments	**Ambo**	**Dandi**	**Toke kutaye**	**Mean**
Allium sativum	68.28^{b}	76.40^{b}	72.78^{bcd}	72.49^{b}
Nicotiana sp.,	46.29^{bc}	49.90^{de}	51.85^{e}	49.35^{c}
Cymbopogon citrates	65.57^{bc}	77.30^{b}	64.71^{bcde}	69.19^{b}
Azadirachta indica	72.39^{b}	74.03^{bc}	76.07^{bc}	74.26^{b}
Beauveria bassiana	78.37^{b}	65.66^{bcd}	78.40^{b}	74.14^{b}
Metarhizium anisopliae	64.76^{bc}	54.76^{cde}	58.40^{cde}	59.31^{bc}
Vayego 200 SC	96.78^{a}	93.38^{a}	98.42^{a}	96.19^{a}
Coragen 200 SC	94.19^{a}	96.97^{a}	95.83^{a}	95.66^{a}
Control	9.97^{d}	6.23^{f}	0.00^{d}	5.40^{d}
LSD at 0.01	20.04	12.32	12.02	10.87
CV (%)	18.83	19.94	19.81	18.81
SE ±	8.31	5.16	5.04	4.56

Note: Means with the same letter(s) in columns are not significantly different for each other.
All treatment effects were highly significant at P<0.01 (DMRT)

6.3.4.2 Effect of bio-pesticides on leaf and fruit of tomato infestation

In Ambo district, data recorded on number of leave infested and fruits bored per plant were presented in Table (6.15). All treatments were significantly reduced *T. absoluta* leaf infestation compared to untreated control. Plants treated with *A. sativum*, *C. citrates, A. indica*, *B. bassiana* and Coragen 200 SC were recorded the lowest infestation of leaves by *T. absoluta* followed by *M. anisopliae*. On the other hand, the effect of *A. indica* (1.33), *C. citrates* (2.33), *B. bassiana* (2.33), Vayego 200 SC (1.33) and Coragen 200 SC (1.33) on tunneld fruits per plant at the same district and not significantly differed to each other but highly significant ($P < 0.01$) different from untreated control (12.33)/plant. The highest leave infestation was recorded in untreated control followed by *Nicotiana* sp. (8.0)/plant was recorded.

In Dandi district, infestation of leaves were observed on standard check (Coragen 200 SC) (4.33)/plant the lowest infestation and followed by *C. citrates* (10.67), *B. bassiana* (11.0), *A. indica* (11.67) and A. *sativum* (13.67)/plant were recorded. The standard check was significant (P < 0.05) different from other treatments. Similarly, the untreated control also highly significant (P < 0.01) different from all treatments and recorded the highest leave infestation (42.67)/plant. The effects of bio-pesticides on fruit of tomato against *T. absoluta* were evaluated. It was showed that most of the treatments gave low fruit tunneled, among of these Coragen 200 SC (0.0), *B. bassiana* (0.33) *A. sativum* (1.33), *C. citrates* (1.67), and *A. indica* (1.67) per plant. Most of the treatments reduced tunneled tomato fruits significantly compared to untreated control. Similar results were observed in Toke kutaye district (Table 6.15).

In all districts, treatments were found significantly superior over the control. Leaf infestation and fruits bored by of tomato leaf miner, *T. absoluta* was reduced after treatment application. Results proved a significant P < 0.01) difference in infestation level of *T. absoluta* as affected by the different control treatments under field conditions. The differences can be attributed to different modes of action of the products and also the number of days after treatment application. The results showed that Vayego 200 SC, *A. sativum, C. citratus, A. indica,* and *B. bassiana* were reduced number of leaf infested and fruit tunneled per plants in all districts (Table 6.15).

Table 6.15: Mean infestation of leaf and fruit per plant on different districts of West Shewa, Central Ethiopia

Treatments	**Locations**					
	Ambo		**Dendi**		**Toke kutaye**	
	Leaf infested	Fruit bored/plant	Leaf infested	Fruit bored/plant	Leaf infested	Fruit bored/plant
Allium sativum	9.33^{d}	1.67^{b}	13.67^{cd}	1.33^{d}	11.0^{de}	2.33^{c}
Nicotiana sp.,	23.67^{b}	8.0^{b}	22.67^{b}	7.33^{b}	18.67^{bc}	6.33^{b}
Cymbopogon citratus	8.0^{d}	2.33^{c}	10.67^{d}	1.67^{d}	15.0^{cd}	2.33^{c}
Azadirachta indica	8.0^{d}	1.33^{c}	11.67^{d}	1.67^{d}	11.33^{d}	1.33^{c}
Beauveria bassiana	8.0^{d}	2.33^{c}	11.0^{d}	0.33^{d}	10.33^{de}	0.67^{c}
Metarhizium anisopliae	15.0^{c}	6.0^{b}	17.33^{c}	4.67^{c}	15.33^{bcd}	5.67^{b}
Vayego SC 200	3.0^{d}	1.33^{c}	3.67^{e}	1.0^{d}	2.67^{e}	1.0^{c}
Coragen 200 SC	6.33^{d}	1.33^{c}	4.33^{e}	0.0^{d}	4.67^{e}	0.67^{c}
Control	49.0^{a}	12.33^{a}	42.67^{a}	18.0^{a}	41.33^{a}	9.0^{a}
LSD at 0.01	4.84	2.66	4.25	2.56	6.38	2.22
CV (%)	12.32	21.52	10.58	22.47	16.13	24.46
SE ±	2.03	1.12	1.78	1.07	2.68	0.93

Note: Means with the same letter(s) in columns are not significantly different for each other.
All treatment effects were highly significant at P<0.01 (DMRT)

6.3.4.3 Effect of bio-pesticides on yield of tomato

Evaluation of different bio-pesticides against *T. absoluta* under three different districts of West Shawa on effect tomato fruit yields. No significant (P > 0.05) differences were recorded among the treatments of Vayego 200 SC, *B. bassiana*, *A. indica* and the standard check (Coragen 200 SC) depicted on (Table 6.16). The highest mean yield at all districts obtained in treatment of Vayego 200 SC, the standard check (Coragen 200 SC) followed by *B. bassiana* and *A. indica*. The minimum yields were recorded in untreated control followed by *Nicotiana* sp. Considering the mean marketable fruit yield production, the fruits that were highly damaged by *T. absoluta*, for each treatment (Table 6.16), the maximum and minimum production were obtained from plants treated with *A. sativum* (23.63 ton/ha) and *Nicotiana* sp. (12.83 ton/ha) in Ambo district, *B. bassiana* (27.53 ton/ha) and *Nicotiana* sp. (15.33 ton/ha) in Dandi district and similar results were observed in Toke kutaye districts, respectively.

Table 6.16: Mean marketable yield per hectare in tons on different locations of West Shewa, Central Ethiopia

Treatments	**Locations**			Mean Marketable yield/ha in tons
	Ambo	**Dendi**	**Toke kutaye**	
	Marketable yield/ha	Marketable yield/ha	Marketable yield/ha	
Allium sativum	23.63^{b}	26.8^{ab}	19.30^{bc}	22.30
Nicotiana sp.,	12.83^{d}	15.33^{e}	11.61^{e}	13.26
Cymbopogon citratus	19.72^{c}	24.19^{abc}	17.83^{cd}	20.58
Azadirachta indica	25.37^{ab}	25.00^{abc}	20.37^{abc}	23.58
Beauveria bassiana	25.17^{ab}	27.53^{ab}	21.62^{ab}	24.77
Metarhizium anisopliae	19.25^{c}	21.17^{cd}	15.70^{d}	18.71
Vayego 200 SC	29.69^{a}	28.05^{a}	25.33^{a}	26.69
Coragen 200 SC	28.01^{a}	29.03^{a}	23.53^{a}	26.86
Control	8.43^{e}	5.60^{f}	4.62^{f}	6.22
LSD at 0.01	3.37	4.38	3.27	
CV (%)	7.16	8.53	8.41	
SE ±	1.41	1.84	1.37	

Note: Means with the same letter(s) in columns are not significantly different for each other.
All treatment effects were highly significant at P<0.01 (DMRT)

6.3.4.3.1 Yield loss Estimation

Highly significant ($P < 0.01$) differences were observed from untreated check in all Districts in terms of yield losses. Low yield losses recorded in the plots treated with *B. bassiana* 6.87% to 10.08% followed by *A. indica* 7.33 to 13.13% at harvesting time. The maximum yield losses were recorded from plots treated with *Nicotiana* sp (44.01, 42.32 and 37.88%) in Ambo, Dandi and Toke kutaye Districts, respectively. In the control plots the yield losses of the untreated plots were between 59.16 to 70.12% in all three Districts of West Shewa of Central Ethiopia (Table 6.17). Therefore, yield losses depend on infestation level of *T. absoluta*.

Table 6.17: Yield losses of tomato fruits after treatment application against *T. absoluta* during 2015/2016 in three different locations of West Shewa, Centeral Ethiopia

Treatments	**Locations**					
	Ambo		**Dendi**		**Toke kutaye**	
	Wt of tomatoes (tons/ha)	Yield Loss in (%)	Wt of tomatoes (tons/ha)	Yield Loss in (%)	Wt of tomatoes (tons/ha)	Yield Loss in (%)
Allium sativum	27.29^{be}	21.40	31.11^{d}	8.26	24.95^{b}	12.09
Nicotiana sp.,	19.44^{de}	44.01	19.56^{d}	42.32	17.63^{e}	37.88
Cymbopogon citratus	23.92^{cd}	31.11	28.36^{bc}	16.37	23.36^{bc}	17.69
Azadirachta indica	30.16^{abc}	13.13	30.46^{ab}	10.17	26.23^{ab}	7.33
Beauveria bassiana	31.22^{ab}	10.08	31.01^{ab}	8.55	26.43^{ab}	6.87
Metarhizium anisopliae	26.64^{bc}	23.28	27.22^{bc}	19.73	21.47^{cd}	24.35
Vayego 200 SC	35.31^{cd}	---	36.22^{c}	---	28.62^{de}	---
Coragen 200 SC	34.72^{a}	---	33.91^{a}	---	28.38^{a}	---
Control	14.18^{e}	59.16	11.64^{e}	65.67	8.48^{f}	70.12
LSD at 0.01	6.12		4.22		2.92	
CV (%)	10.77		6.64		5.58	
SE ±	2.78		1.77		1.23	

Note: Means with the same letter(s) in columns are not significantly different for each other.
All treatment effects were highly significant at $p<0.01$ (DMRT)

6.3.5. Evaluation of sticky trap colors on capture of *T. absoluta* Moths

The results (Table 6.18 and 6.19) showed that significant ($P < 0.01$) differences were observed among the treatments. The results revealed that the different sticky trap colors were captured Tomato leaf miner during 2016 to 2017. This study showed *T. absoluta* attraction has differential attraction to various colors in the sequence of white > blue > yellow > green > red during both study years 2016 and 2017. The maximum of moths (*T. absoluta*) were caught in white sticky color traps followed by blue sticky traps. The minimum number of moths of *T. absoluta* was caught in red and green sticky colored traps, respectively. The study of colored sticky traps on capturing of *T. absoluta* moths showed effective control measures against the insect pest of *T. absoluta*. The mean number of caught in stick trap with different colors and phonological developmental stages of tomato plants. It was observed that the maximum number of *T. absoluta* caught in white sticky trap followed by blue colored at all developmental stages. In both years, maximum sticky trap catch of *T. absoluta* was significantly to higher in white traps (Table 6.18 and 6.19) and relatively more trap catch was observed during 2016 might be due to more incidence of *T. absoluta* moths. The captured percent of *T. absoluta* also showed on (Fig. 6.1) during 2016 and on (Fig. 6.2) during 2017. The highest captured percent was observed in white stick trap during first year 47%, 39% and 33%, at vegetative, flowering and fruit setting stages, respectively. Although the lowest captured percent was indicated on red sticky color. In 2017, the maximum captured percents of *T. absoluta* moths were recorded 32%, 39%, and 38% at vegetative, flowering and fruit setting stages, respectively. Similar to the first year the minimum captured percent recorded on red sticky colors.

Table 6.18: Cumulative Mean of *T. absoluta* caught in sticky traps with different colors during 2016 cropping season

Treatments	Number of adult captured at different phonological stages		
	At vegetative	At flowering	At fruit setting
Blue	1.85^a	3.85^a	5.08^a
Green	0.63^b	1.13^c	1.63^b
Red	0.35^b	0.48^c	1.00^b
Yellow	0.83^b	2.58^b	2.70^b
White	3.15^a	4.33^a	5.03^a
LSD at 0.01	2.12	1.17	1.75
SE±	0.98	0.54	0.81

Note: Means with the same letter(s) in columns are not significantly different for each other.
All treatment effects were highly significant at P<0.01 (DMRT)

Table 6.19: Cumulative Mean of *T. absoluta* caught in sticky traps with different colours during 2017 cropping season

Treatments	Number of adult captured at different phonological stages		
	at vegetative	at flowering	at fruit setting
Blue	2.50^a	2.10^b	2.50^{ab}
Green	0.28^c	0.58^d	1.63^{bc}
Red	0.00^c	0.78^{cd}	0.85^c
Yellow	0.90^c	1.80^{bc}	1.55^{bc}
White	2.28^{ab}	3.45^a	3.05^a
LSD at 0.01	1.38	1.11	1.36
SE±	0.64	0.51	0.63

Note: Means with the same letter(s) in columns are not significantly different for each other.
All treatment effects were highly significant at P<0.01 (DMRT)

It was recorded that the minimum numbers of *T. absoluta* moths were caught at vegetative stage in all colored sticky traps indicating to starting the infestation and appearance of *T. absoluta* moths while the maximum numbers were caught at fruit setting in both years. Also, white sticky trap caught higher number of moths than other traps during the whole tested phonological stages of the plant, white and blue sticky traps showed insignificant and yellow sticky traps came in the second order while the green and red are approximately equal in their efficiency for attraction *T. absoluta* moths and less efficient in comparison to white, blue and yellow sticky traps.

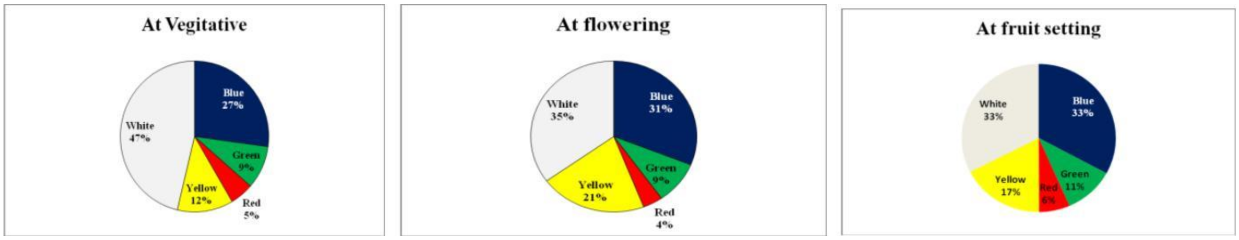


Figure 6.1: Effect of different color sticky traps for capturing of *T. absoluta* moths under glasshouse conditions during 2016

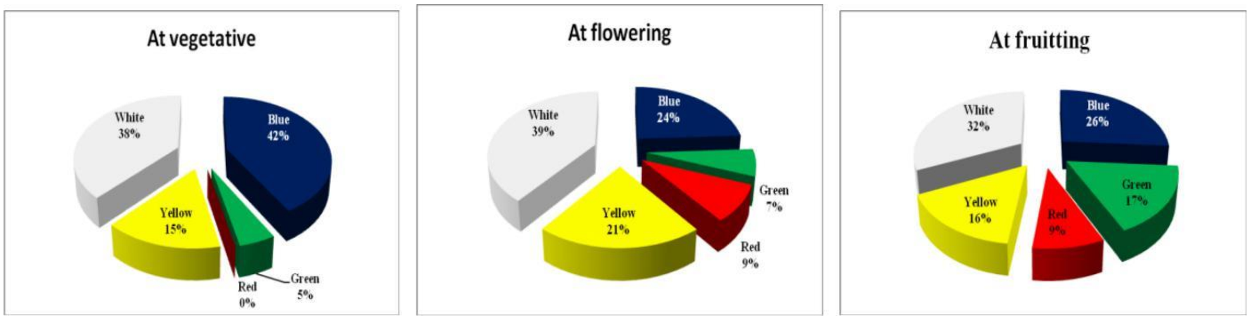


Figure 6.2: Effect of different color sticky traps for capturing of *T. absoluta* moths under glasshouse conditions during 2017.

6.4 Discussion

Tuta absoluta are capable of reducing the total yield of *tomato* plants as well as facilitate it to secondary attack by pathogens. The application of botanicals on *T. absoluta* larvae resulted good performance. These results were confirmed with those reported by Ghanim and Abdel Ghani (2014) they stated that the highest reduction of *T. absoluta* population was obtained after tomato plants treated with *A. sativum and C. citrates* extract. They showed that highest effects of *A. sativum* on *T. absoluta* second instar larvae under laboratory conditions. *C. citrates* extract reduced the population of *T. absoluta* on plant moderately as compared with *A. sativum, A. indica* and *Nicotiana* sp. The results obtained were in agreement with the work of Hussein *et al.*

(2014) they found that the aqua extract of *C. citrates* and *A. sativum* showed good insecticidal activity and can be used to control *T. absoluta* in the laboratory and glasshouse. This finding is also concordant with the report of Gonçalves-Gervásio and Vendramim (2008) they found that *A. indica* seeds extracts made high larval mortality of *T. absoluta* under laboratory condition. Oparaeke (2007) verified that *A. indica* and *A. sativum* extracts contain insecticidal properties that are lethal to a wide range of insects. The results revealed that *A. sativum* and *C. citrates* were gave promising results to minimize the impact of *T. absoluta* larvae on leaves and fruits.

A positive relationship was recorded between mortality percentages and concentrations among the *B. bassiana* and *M. anisopliae* concentrations. Concurrently, with the increase in conidia concentration, a reduction in LT_{50} was observed. Concentrations of 2.5×10^9 from *B. bassiana*, at the concentrations 2.5×10^8 and 2.5×10^9 conidia ml^{-1}, presented the shortest lethal time. These low values are probably associated to the presence of enzymes that aid in the process of penetration of the fungi (St. Leger *et al*., 1988).

The effect of entomopathogenic fungi were evaluated to determined the concentrations with high efficacy against larvae *T. absoluta* under laboratory and glasshouse conditions. Both fungal isolates were found to be pathogenic to *T. absoluta*. Though, there was a variation in their virulence against *T. absoluta*. The percent mortality for all the concentrations gradually increased. The spore formation appeared on the larvae of *T. absoluta* took place after treatment exposure of the concentrations of the two isolates starting from the day three after treatment exposure, and thereby no hatched larvae were appeared in the concentrations of both isolates comparing the control treatment. The *M. anisopliae* in all concentrations were significantly less effective when compared with that of *B. bassiana* in terms of virulence. Virulence due to *B.*

bassiana on 10th day was not significantly different from each other. This indicated that all *B. bassiana* concentrations were the best management option of *T. absoluta*. This finding confirms with earlier reports (Shalaby *et al*., 2013) who obtained high percent mortality during the evaluation time for *B. bassiana* and *M. anisopliae*.

The amount of conidia used should to attain a certain concentration and thus, achieving an efficacious penetration of the fungus on the insect cuticle and causing host death. Similar findings by Garcia *et al.* (2011) were obtained, evaluating the insecticidal activity of *B. bassiana* strains and *M. anisopliae* on *Spodoptera frugiperda* and *Epilachna varivestis* larvae at six concentrations (10^4 to 10^9); *B. bassiana* strain was more virulent for *E. varivestis* larvae with a 93.3% mortality, LC_{50}= $1.20×10^6$ conidia ml^{-1} and LT_{50}= 5.1 days. *B. bassiana* strain presented the highest mortality on *S. frugiperda* larvae (96.6%, LC_{50}= $5.92×10^3$ conidia ml^{-1} and LT_{50}= 3.6 days). These results are disagreed with Khalid *et al*. (2012), evaluating the virulence of various strains of *B. bassiana* and *M. anisopliae* on *G. mellonella* larvae using 10^2, 10^3, 10^4, 10^5 and 10^6 conidia ml^{-1} concentration.

Thus, laboratory and glasshouse experiments suggested that *B. bassiana* and *M. anisopliae* have good effect on both eggs and larvae of *T. absoluta.* Sabbour (2014) also confirmed the effectiveness of both *B. bassiana* and *M. anisopliae* against larvae of *T. absoluta* under laboratory and greenhouse conditions. The same results were obtained by Sabbour and Singer (2014); Sabbour and Abdel-Raheem (2014). These results agree with these findings and Cabello *et al*. (2009**)** where stated that; the higher mortality of larvae under laboratory studies indicated *B. bassiana* could cause good larval mortality. At present, the knowhow of entomopathogenic fungi on *T. absoluta* was very limited because of very few studies that are available to indicate that the isolates causes the high mortality on other lepidopteran insects (Kannan *et al*., 2008). In this study it has been shown that all the fungal concentrations are effective against *T. absoluta*.

These results confirmed that, the previous study of Shalaby *et al*. (2013), they stated that when the second instar larvae fed on *M. anisopliae* the pathogen effect was evident by the 3rd day of evaluation after exposure in the concentration (10^7 and 10^8 conidia/ml). Dahliz *et al*. (2014) have reported similar results with *Metarhizium*. This result was confirmed the work of Inanl and Oldargc (2012), they reported the studies conducted in Turkey, researchers compared the efficacy of *B. bassiana* and *M. anisopliae* on *T. absoluta* eggs and larvae; these two agents provided highly effective to control of *T. absoluta* larvae. Our results also indicated the potential of *B. bassiana* and *M. anisopliae* to control the larvae of *T. absoluta* in an integrated pest management programs. Neves and Alves (2000) also noted, as more conidia penetrating, more toxins or enzymes are released, increasing the insect mortality. Though, the fungus action speed depends, besides the concentration, of the host species involved (Sosa-Gomez and Moscardi, 1992). According to Kleespies and Zimmermann (1998), variation in virulence of entomopathogenic strains was a result of differences in the enzymes and toxins production in conidia germination speed, mechanical activity in the cuticle penetration, colonization capacity and cuticle chemical composition.

Throughout the experiments, the mixture of Emamectin Benzoate and Prosuler Oxymatrin products proved more efficacy suppressed *T. absoluta* larval populations followed by Emamectin Benzoate alone. Indeed, several authors reported the performance of Emamectin Benzoate product against several insects. Seal (2005), reported the efficacy of emamectin benzoate at various rates in reducing the densities of the melon thrips, *Thrips palmi* adults and larvae.

Stanley *et al.,* (2005) reported the high acute toxicity of emamectin benzoate to *Helicoverpa armigera* under laboratory conditions.

It was reported by several authors that the active ingredient of Emamectin benzoate has a high potency against a broad spectrum of Lepidopterous pests with an efficacy potent against certain armyworm species (Jansson *et al.,* 1996). Cook *et al*., (2004) conducted field and laboratory trials on cotton and soybean for the control of the beet armyworm *Spodoptera exigua* (Hübner) and the fall armyworm *Spodoptera frugiperda* using emamectin benzoate demonstrated the good efficacy of tested products compared with the control. But we obtained low percent mortality of *T. absoluta* larvae under laboratory and glasshouse conditions.

The efficacies of spraying using mixtures of natural products and synthetic chemicals for the control of the pests are crucial. Indeed, insecticides that work in synergy when mixed together are an avenue to explore in *T. absoluta* control. We agreed that the work of Bielza *et al*. (2009), they have been proposed that pesticides mixtures with different modes of action may delay the onset of resistance developing in pest populations. However, some problems need to be considered when two or more insecticides are mixed together especially Phytotoxicity (Mohamed and Lobna, 2012).

Management of resistance to prevent or delay the development of resistance to an insecticide and cross resistance to additional insecticides is necessary for increasing the chance of insecticide control of *T. absoluta*. This glasshouse and laboratory studies suggested the good performance of the tested compounds Chlorantraniliprole (Coragen 200 SC^{TM}) followed by (a mixture of Emamectin Benzonate and Prosuler Oxymatrin were important for the managment of tomato leafminer, *T. absoluta.*

Limited information is available regarding the efficacy of bio-pesticides against *T. absoluta*. However, the use of traditional botanical pesticides during the past decades and their efficacy against different pests has led to their wide acceptance throughout the world. Researchers have focused on the use of botanical extracts, oils and plant powders, which are cheap, of short persistence and of low mammalian toxicity. A number of authores indicated that, many of these plant materials show a broad spectrum of activity against insect pests, such as lethal, antifeedant, repellent and growth regulatory effects (Stoll, 2000; Hiiesaar *et al*., 2001).

These studies also revealed that after 3 days of treatment application *B. bassiana* and *M. anisopliae* indicated no effect on the larvae of *T. absoluta*. It can be supposed that, the establishment of fungi on the larvae of insect pests take some days. The results of the present experiments were quite similar to that of (Trindade *et al*., 2000), who applied Neem seed extract against larvae of *T. absoluta*. The result of the experiments showed high percent mortalities of entomopathogenic fungi against *T. absoluta* larvae up to 78.40% obtained after 10 days of treatment application on different locations. Many findings indicated effective larval control of *T. absoluta* with different botanical extracts were recorded in different countries. It was contrast with the work of Trindade *et al.* (2000), he was reported that application of Neem seed extract against larvae of *T. absoluta* resulted 84-100% mortality after 4 days while our findings proved that Neem seed extracts at 10% concentration showed that 52.31 to 59.02 percent mortality after 5 days of application. However, it was increased after 7 to 10 days of treatment application 65.66 to 78.40% percent mortality on different locations. Coragen 200 SC exhibit the good mode of action, this explains their similar mean reduction in *T. absoluta* infestation (Hamdy and Walaa, 2013). Our findings were agree with the work of Shalaby *et al.* (2013), they were suggested that *B. bassiana* and *M. anisopliae* has a potential effect on larvae of *T. absoluta.*

These studies were agreed with the work of Sabbour (2014) control of *Tuta absoluta* by three microbial control agents including *B. bassiana* which increase the yield of tomato fruit. Solufeed Ltd. (2015) reported that garlic extracts and other naturally derived plant chemicals are becoming increasingly subject to official recognition in regard to their pesticidal activity. Our findings also confirmed plant extracts including garlic were prepared and tomato plants infested with leaf miner were sprayed three times at vegetative, flowering and fruit setting, all treatments reduced population density of tomato leaf miner significantly. The highest reduction was recorded by *A. sativum* extract followed by *C. citratus* extract and also, garlic extract increased the yield of tomato significantly.

Effective management option in conjunction with integrated pest management (IPM) is vital to global crop protection, sustainable agriculture and improved public health (IRAC, 2014). Both yield and fruit quality can be significantly reduced by the direct feeding of *T. absoluta* and secondary pathogens that may enter through the wounds made by the insect. Similar results obtained by Cabello *et al*. (2009) who controlled the pinworm by bio-insecticides. Several Researchers reported, if no control measures are taken, then the pest can reduce tomato yield and fruit quality up to 80-100% yield losses by attacking leaves, flowers, stems and especially fruits **(**Öztemiz, 2012; CFIA, 2010; Lopez, 1991**)**. But, in these field experiments at three different districts the results showed that 59.16 - 70.12% yield losses were recorded.

These results inconsistence with white pheromone traps caught more moths than yellow, blue, green and red traps. Previous findings by various authors differed regarding attraction of various colors in attracting tomato leaf miner (Knight and Miliczy, 2003; Mahmoud *et al*., 2014). White and blue have been considered as the preferred or the most preferred colors for *T. absoluta*.

White traps caught significantly more *T. absoluta* than the yellow ones in this study. Similarly, this result was assured the previous work of Mahmoud *et al.* (2014) where the means of weekly catches of both white and yellow traps are slightly different from one another while significant differences between means of sticky trap colors catches of white and blue traps from one point and other three traps from another point were recorded.

On the other hands, green and yellow traps caught more Grab Root Borer (GRB) moths than other traps (white and blue) and the males prefer green and yellow pheromone-baited traps (Craig and Oscar, 2008), while red traps were most effective in trapping moths of *Helicoverpa armigra*, *Earis insulana* and *Plutella xylostella*, while yellow pheromone traps attracted maximum number of *Spodoptera littoralis* moths (Kumar, 2009). On the other insect pest study conducted by Christos *et al.* (2004) on *Palpita unionalis* are closed with our results where they found that among the five colored sticky traps tested, white and yellow traps were most effective where white and yellow colors revealed. Although the previous study of Mahmoud *et al*., (2014), they demonstrate the impotence of considering the visual stimuli of Lepidopterous moths in the design of pheromone traps and further study is required however, to answer the question as to why *T. absoluta* moths are more attracted to white and yellow traps than to the other traps. This study showed that plant phonological stages are strongly affects the response of *T. absoluta* moths to sticky traps colour. Therefore, the plant phonology is one of the most important aspects for capturing of *T. absoluta*.

6.5 Conclusions

Considering, the high risks of chemical insecticides on human being, mammals and birds as well as in the environment bio-pesticides are a cheap, valuable, safe and environmentally friendly alternative insect pest management. There are indications that producers consider available information on management of *T. absoluta* when choosing different control strategies. The newly tested bio-pesticides and report of effectiveness efforts, in addition to the synthetic chemical insecticides. It might be concluded that *A. indica*, *A. sativum* and *C. citratus* were potential effect on larvae of *T. absoluta* after 7th day of application. Moreover, the newly introduced insecticide Tetraniliprole (Vayego 200 SC) gave effectively control *T. absoluta* after application of 24 hours. Consequently, from the obtained results, it can be concluded that foliar application of the mentioned insecticide, medicinal plants extract and entomopathogenic fungi on tomato plants reduced *T. absoluta* population and improved the quality and quantity of tomato fruit yield. The sticky trap colors and tomato developmental stages were strongly affect the response of *T. absoluta* moths. Furthermore, the infestation level of *T. absoluta* was exist throughout the year if host is available and mostly depends on phonological stages. Hence, more researches with new materials in the future will constitute an asset in the field of botanicals, entomopathogenic fungi and different sticky trap colors on different climatic conditions.

CHAPTER SEVEN

7. General Conclusions and Recommendations

7.1 Conclusions

The population number of all stages of *T. absoluta* and the infestation level were increased during months of June to August, the period of peak activities of the *T. absoluta* compromise with vegetative time with flowering and early fruit setting stages of the crop. As a result, large populations, consisting larvae damage the crop leaves at vegetative and then passed to fruit under glasshouse conditions. The infestation level of *T. absoluta* was exist throughout the year if host is available and mostly depends on phonological stages.

The moths of *T. absoluta* preferred tomato plants more than potato plants both in glasshouse and open field studies. *T. absoluta* should preferentially lay more eggs on upper surface leaves of tomato plants where as on lower surface leaves of potato plants. The densities of trichomes may have played an important role in the *T. absoluta* oviposition.

Tuta absoluta at 32 ± 2^{O}C is less conducive to maintaining a colony in a glasshouse than rearing them at 20.5 ± 2^{O}C in laboratory. It has been proved as a most suitable for superior development, maximum survival and minimum mortality.

Tuta absoluta larval population reaches 2.25 larvae per plant in glasshouse and 2.87 larvae per plant on field in order to prevent the population in reaching economic injury levels. The infestation of *T. absoluta* can cause 87.50 to 100% marketable yield loss under glasshouse and 60.08% to 82.31% on open field conditions.

Foliar applications of *B. bassiana* and *M. anisopliae* at 2.5×10^{9} concetration have potential effect on arvae of *T. absoluta*. Moreover, foliar applications of medicinal plants crude extract such as *A. indica*, *A. sativum* and *C. citratus* on tomato plants reduce the larval population density of *T. absoluta*. The frequency of bio-pesticide application period depends on infestation level of *T. absoluta* and effectiveness of bio-pesticides.

In glasshouse study, the moths of *T. absoluta* preferred white sticky trap color more than blue and yellow.

7.2 Recommendations

The economic injury and economic threshold level was determined under glasshouse and open field conditions. So, based on the results the control measures should be initiated when the *T. absoluta* larval population reaches 2.25 larvae per plant in glasshouse and 2.82 larvae per plant on open field in order to prevent the population in reaching economic injury levels.

Under glasshouse and open field conditions high estimation of yield losses were recorded during March to June 2016 and low during November, 2015 to February, 2016. This indicated that during March to June the population of *T. absoluta* was increased and infestation level also increased. Therefore, tomato planting is more prefer from November to February rather than from March to June to escape from highly devastating of *T. absoluta.*

Concerning the effectiveness of treatments, it was found that the lower infestation level the higher marketable yield and less yield percent loss. Thus laboratory, glasshouse and field studies suggested that *A. indica*, *A. sativum* and *C. citratus* at 10% concentrations have a potential effect on larvae of *T. absoluta* under glasshouse and open field conditions. The entomopathogenic fungi, *B. bassiana* and *M. anisopliae* present different capacity cause mortality of the insects, with the 2.5×10^9 conidian ml^{-1} *B. bassiana* strains as the most pathogenic for *T. absoluta*, as well as 2.5 x 10^9 conidian ml^{-1} *M. anisopliae* strains was also good virulence for *T. absoluta* and presenting the lowest LC_{50} and LT_{50} values.

The use of sticky colors trap at this period is an effective and easy to use method of early pest population control. It was found that, white and blue sticky traps were more effective in capturing moths of *T. absoluta* population in glasshouse conditions.

To control *T. absoluta* effectively it is critical to combine all the control measures available and not to rely only on insecticide sprays. Various active substances are effective and can be used in combination with cultural, biological and bio-pesticide control measures. Hence, the insecticidal substances those have potential for use as alternative management options of *T. absoluta* based on different tactics and environmental friendly is an important component of an integrated pest management strategies. Therefore, these informations could be utilized to know when and how to begin monitoring and control measures to be implemented.

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LIST OF APPENDICES

Annex 1: Summary table for analysis of variance (ANOVA) on monthly average population dynamics of *T. absoluta* Eggs per plant under glasshouse conditions during 2015-2016.

Source of Variation	DF	SS	MS	F-value	Pr > F
Model	15	14733.33	982.22	88.00	< 0.0001
Error	44	491.13	11.16		
Corrected total	59	15224.47			
Replication	4	120.79	30.19	2.71	0.04
Treatment	11	14612.52	1328.41	119.01	< 0.0001

R-square	LSD	CV (%)	SE±	Mean	Critical value of t
0.97	5.69	15.72	3.34	16.12	2.69

Annex 2: Summary table for analysis of variance (ANOVA) on monthly average population dynamics of *T. absoluta* Larva per plant under glasshouse conditions during 2015-2016.

Source of Variation	DF	SS	MS	F-value	Pr > F
Model	15	4999.14	338.28	73.33	< 0.0001
Error	44	198.98	4.56		
Corrected total	59	5199.12			
Replication	4	10.09	2.52	0.55	0.69
Treatment	11	4989.07	453.85	99.79	< 0.0001

R-square	LSD	CV (%)	SE±	Mean	Critical value of t
0.96	3.63	13.66	2.13	9.01	2.69

Annex 3: Summary table for analysis of variance on monthly average population dynamics of *T. absoluta* Pupa per plant under glasshouse conditions during 2015-2016.

Source of Variation	**DF**	**SS**	**MS**	**F-value**	**Pr > F**
Model	15	1030.60	68.69	53.23	< 0.0001
Error	44	56.79	1.29		
Corrected total	59	1087.19			
Replication	4	4.69	1.17	0.91	0.47
Treatment	11	1025.70	93.25	72.25	< 0.0001

R-square	**LSD**	**CV (%)**	**SE±**	**Mean**	**Critical value of t**
0.95	1.93	17.09	1.14	4.53	2.69

Annex 4: Summary table for analysis of variance on leaf damaged by *T. absoluta* on tomato plant under glasshouse conditions during 2016-2016

Source of Variation	**DF**	**SS**	**MS**	**F-value**	**Pr > F**
Model	15	168613.91	11240.93	147.72	< 0.0001
Error	44	3348.34	76.09		
Corrected total	59	171962.26			
Replication	4	673.82	168.45	2.21	0.083
Treatment	11	167940.09	15267.28	200.62	< 0.0001

R-square	**LSD**	**CV (%)**	**SE±**	**Mean**	**Critical value of t**
0.98	14.85	14.52	4.72	49.79	2.69

Annex 5: Summary table for analysis of variance on egg laying position of *T. absoluta* on tomato plant under glasshouse condition

Source of Variation	DF	SS	MS	F-value	Pr > F
Model	6	8746.63	1457.77	140.72	< 0.0001
Error	8	82.88	10.36		
Corrected total	14	8829.51			
Replication	2	5.03	2.54	0.25	0.7881
Treatment	4	8741.55	2185.39	210.95	< 0.0001

R-square	LSD	CV (%)	SE±	Mean	Critical value of t
0.99	8.82	16.09	3.22	20.00	3.36

Annex 6: Summary table for analysis of variance on mean egg survival percent of *T. absoluta* during their life cycle under laboratory and glasshouse conditions.

Source of Variation	DF	SS	MS	F-value	Pr > F
Model	6	384.68	128.23	211.36	0.0047
Error	5	1.21	0.61		
Corrected total	11	385.89			
Replication	5	149.05	74.53	122.85	0.0081
Treatment	1	235.63	235.63	235.63	0.0066

R-square	LSD	CV (%)	SE±	Critical value of t
0.99	2.74	0.87	0.78	4.30

Annex 7: Summary table for analysis of variance on mean larva survival percent of *T. absoluta* during their life cycle under laboratory and glasshouse conditions

Source of Variation	**DF**	**SS**	**MS**	**F-value**	**Pr > F**
Model	6	161.60	53.87	1.69	0.3967
Error	5	63.74	31.87		
Corrected total	11	225.35			
Replication	5	1.44	0.72	0.02	0.9780
Treatment	1	160.17	160.17	5.03	0.1542

R-square	**LSD**	**CV (%)**	**SE±**	**Critical value of t**
0.91	19.83	6.51	5.65	4.30

Annex 8: Summary table for analysis of variance on mean pupa survival percent of *T. absoluta* during their life cycle under laboratory and glasshouse conditions

Source of Variation	**DF**	**SS**	**MS**	**F-value**	**Pr > F**
Model	6	357.25	119.08	128.77	0.0077
Error	5	1.85	0.92		
Corrected total	11	359.10			
Replication	5	217.86	108.92	117.79	0.0084
Treatment	1	139.39	139.39	150.73	0.0066

R-square	**LSD**	**CV (%)**	**SE±**	**Critical value of t**
0.98	3.39	1.16	0.98	4.30

Annex 9: Summary table for analysis of variance on mean oviposition of *T. absoluta* on upper leaf of major Solanaceae crops during 2015/2016

Source of Variation	DF	SS	MS	F-value	Pr > F
Model	13	25262.23	1943.25	232.55	< 0.0001
Error	22	183.87	8.36		
Corrected total	35	25446.06			
Replication	2	11.78	5.90	0.71	0.5045
Treatment	11	25250.43	2295.49	274.70	< 0.0001

R-square	LSD	CV (%)	SE±	Critical value of t
0.99	4.89	9.48	2.89	2.07

Annex 10: Summary table for analysis of variance on mean oviposition of *T. absoluta* on lower leaf of major Solanaceae crops during 2015/2016

Source of Variation	DF	SS	MS	F-value	Pr > F
Model	13	30703.97	2361.84	206.06	< 0.0001
Error	22	252.17	11.47		
Corrected total	35	30956.13			
Replication	2	25.06	12.53	1.09	0.3527
Treatment	11	30678.90	2788.99	243.32	< 0.0001

R-square	LSD	CV (%)	SE±	Critical value of t
0.99	5.73	9.56	3.89	2.07

Annex 11: Summary table for analysis of variance on mean oviposition of *T. absoluta* on upper leaf of major Solanaceae crops during 2016/2017

Source of Variation	DF	SS	MS	F-value	Pr > F
Model	13	26267.48	2020.58	139.60	< 0.0001
Error	22	38.44	14.47		
Corrected total	35	26585.92			
Replication	2	13.36	6.68	0.46	0.6364
Treatment	11	26254.14	2386.74	164.89	< 0.0001

R-square	LSD	CV (%)	SE±	Critical value of t
0.98	6.44	11.38	3.80	2.07

Annex 12: Summary table for analysis of variance on mean oviposition of *T. absoluta* on lower leaf of major Solanaceae crops during 2016/2017

Source of Variation	DF	SS	MS	F-value	Pr > F
Model	13	24515.14	1885.78	38.10	< 0.0001
Error	22	1088.78	49.49		
Corrected total	35	25603.92			
Replication	2	11.05	5.53	0.11	8949
Treatment	11	24504.09	2227.64	45.01	< 0.0001

R-square	LSD	CV (%)	SE±	Critical value of t
0.95	11.91	21.80	7.03	2.07

Annex 13: Summary table for analysis of variance on infestation level of *T. absoluta* on tomato leaf under glasshouse conditions during 2015/16

Source of Variation	**DF**	**SS**	**MS**	**F-value**	**Pr > F**
Model	9	16067.88	1563.10	94.12	< 0.0001
Error	11	182.69	16.61		
Corrected total	20	14250.57			
Replication	3	1606.64	73.55	4.43	0.0284
Treatment	6	12761.30	2126.88	128.06	< 0.0001

R-square	**LSD**	**CV (%)**	**SE±**	**Critical value of t**
0.98	7.32	13.21	4.08	2.20

Annex 14: Summary table for analysis of variance on effect of *T. abnsoluta* on tomato fruit tunneled/plant under glasshouse during 2015/16

Source of Variation	**DF**	**SS**	**MS**	**F-value**	**Pr > F**
Model	9	768.98	85.44	42.78	< 0.0001
Error	11	21.97	2.0		
Corrected total	20	790.95			
Replication	3	135.57	45.19	22.63	< 0.0001
Treatment	6	633.41	105.57	52.86	< 0.0001

R-square	**LSD**	**CV (%)**	**SE±**	**Critical value of t**
097	2.54	21.35	1.41	2.20

Annex 15: Summary table for analysis of variance on infestation level of *T. absoluta* on tomato leaf under glasshouse conditions during 2016/17

Source of Variation	DF	SS	MS	F-value	Pr > F
Model	9	16943.44	1882.60	101.61	< 0.0001
Error	11	203.80	18.53		
Corrected total	20	17147.24			
Replication	3	2681.95	893.98	48.25	< 0.0001
Treatment	6	14261.48	2376.91	128.49	< 0.0001

R-square	LSD	CV (%)	SE±	Critical value of t
0.98	7.74	13.80	4.30	2.20

Annex 16: Summary table for analysis of variance on effect of *T. abnsoluta* on tomato fruit tunneled/plant under glasshouse during 2016/17

Source of Variation	DF	SS	MS	F-value	Pr > F
Model	9	1360.87	151.21	64.46	< 0.0001
Error	11	25.80	2.35		
Corrected total	20	1386.67			
Replication	3	3.53	1.18	0.50	0.6887
Treatment	6	1137.15	189.53	80.80	< 0.0001

R-square	LSD	CV (%)	SE±	Critical value of t
0.98	2.75	20.88	1.53	2.20

Annex 17: Summary table for analysis of variance on marketable yield in kg/ha under glasshouse during 2015/16

Source of Variation	DF	SS	MS	F-value	Pr > F
Model	9	1,783.691,795.0	198,187,977.0	7.30	0.0016
Error	11	298,565,003.0	27,142,273.0		
Corrected total	20	2,082,256,799.0			
Replication	3	605,205,242.0	201,735,081.0	7.43	0.0054
Treatment	6	1,178,486,553.0	1,964,144,225.0	7.24	0.0025

R-square	LSD	CV (%)	SE±	Critical value of t
0.86	9,362.60	25.63	5,209.83	2.20

Annex 18: Summary table for analysis of variance on marketable yield in kg/ha under glasshouse during 2016/17

Source of Variation	DF	SS	MS	F-value	Pr > F
Model	9	1,959,847,731.0	217,760,859.0	21.92	< 0.0001
Error	11	109,255,391.0	9,932,308.0		
Corrected total	20	2,069,103,123.0			
Replication	3	304,474,909.0	101,491,636.0	10.22	0.0016
Treatment	6	1,655,372,822.0	275,895,470.0	27.78	< 0.0001

R-square	LSD	CV (%)	SE±	Critical value of t
0.95	5663.70	16.98	3151.56	2.20

Annex 19: Summary table for analysis of variance on mean efficacy of some medicinal plant extracts against Larvae of *T. absoluta* at different rates and time of exposure after 24 hours day of application under laboratory conditions

Source of variation	DF	SS	MS	F-value	Pr > F
Model	20	8611.24	430.56	152.30	< 0.0001
Error	36	101.78	2.83		
Corrected Total	56	8713.02			
Replication	2	15.81	7.91	2.80	0.0743
Treatment	18	8595.43	477.52	168.91	< 0.0001

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
0.99	3.73	4.54	1.68	37.01	2.72

Annex 20: Summary table for analysis of variance on mean efficacy of some medicinal plant extracts against Larvae of *T. absoluta* at different rates and time of exposure after 48 hours day of application under laboratory conditions

Source of variation	DF	SS	MS	F-value	Pr > F
Model	20	9342.22	467.11	158.33	< 0.0001
Error	36	106.21	2.95		
Corrected Total	56	9448.43			
Replication	2	2.03	1.02	0.34	0.7108
Treatment	18	9340.18	518.90	175.88	< 0.0001

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
0.99	3.81	4.14	1.72	41.49	2.72

Annex 21: Summary table for analysis of variance on mean efficacy of some medicinal plant extracts against Larvae of *T. absoluta* at different rates and time of exposure after 72 hrs day of application under laboratory conditions

Source of variation	DF	SS	MS	F-value	Pr > F
Model	20	9980.55	499.03	175.58	< 0.0001
Error	36	102.32	2.84		
Corrected Total	56	10082.87			
Replication	2	13.13	6.57	2.31	0.1138
Treatment	18	9967.41	553.75	194.83	< 0.0001

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
0.99	3.74	3.73	1.69	45.15	2.72

Annex 22: Summary table for analysis of variance on mean efficacy of some medicinal plant extracts against Larvae of *T. absoluta* at different rates and time of exposure after 120 hrs day of application under laboratory conditions

Source of variation	DF	SS	MS	F-value	Pr > F
Model	20	37194.74	1857.74	78.52	< 0.0001
Error	36	852.63	23.68		
Corrected Total	56	38047.37			
Replication	2	97.37	48.68	2.06	0.1428
Treatment	18	37097.37	2060.96	86.02	< 0.0001

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
0.98	10.81	7.17	4.87	67.89	2.72

Annex 23: Summary table for analysis of variance on mean efficacy of some medicinal plant extracts against Larvae of *T. absoluta* at different rates and time of exposure after 1 day of application under glasshouse conditions

Source of variation	DF	SS	MS	F-value	Pr > F
Model	8	3190.26	398.78	65.25	< 0.0001
Error	12	73.34	6.11		
Corrected Total	20	3263.60			
Replication	2	26.12	13.06	2.14	0.1608
Treatment	6	3164.14	527.36	86.28	< 0.0001

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
0.98	6.17	8.91	2.47	27.75	3.05

Annex 24: Summary table for analysis of variance on mean efficacy of some medicinal plant extracts against Larvae of *T. absoluta* at different rates and time of exposure after 3 day of application under glasshouse conditions

Source of variation	DF	SS	MS	F-value	Pr > F
Model	8	4313.88	539.23	23.31	< 0.0001
Error	12	277.60	23.13		
Corrected Total	20	4591.49			
Replication	2	52.94	26.47	1.14	0.3509
Treatment	6	4260.94	710.16	30.73	< 0.0001

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
0.94	14.98	12.0	4.81	32.11	3.05

Annex 25: Summary table for analysis of variance on mean efficacy of some medicinal plant extracts against Larvae of *T. absoluta* at different rates and time of exposure after 5 day of application under glasshouse conditions

Source of variation	DF	SS	MS	F-value	Pr > F
Model	8	5116.55	639.57	62.42	< 0.0001
Error	12	122.95	10.25		
Corrected Total	20	5239.50			
Replication	2	43.10	21.55	2.10	0.1648
Treatment	6	5073.45	845.57	82.53	< 0.0001

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
0.98	7.98	8.83	3.20	36.27	3.05

Annex 26: Summary table for analysis of variance on mean efficacy of some medicinal plant extracts against Larvae of *T. absoluta* at different rates and time of exposure after 7 day of application under glasshouse condition

Source of variation	DF	SS	MS	F-value	Pr > F
Model	8	66.22.86	827.86	30.04	< 0.0001
Error	12	261.15	21.76		
Corrected Total	20	6884.01			
Replication	2	14.96	7.48	0.34	0.7160
Treatment	6	6607.90	1101.32	50.61	< 0.0001

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
0.96	11.64	11.59	4.67	40.26	3.05

Annex 27: Summary table for analysis of variance on mean percent mortality of *T. absoluta* caused by different concentrations of insecticides after treatment exposure at 24 hours intervals under laboratory conditions

Source of variation	DF	SS	MS	F-value	Pr > F
Model	10	15670.83	1567.08	67.38	< 0.0001
Error	16	372.14	23.26		
Corrected Total	26	16042.97			
Replication	2	42.87	21.43	0.92	0.4180
Treatment	8	15627.96	1953.49	83.99	< 0.0001

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
	12.79	13.96	5.36	39.75	2.92

Annex 28: Summary table for analysis of variance on mean percent mortality of *T. absoluta* caused by different concentrations of insecticides after treatment exposure at 48 hours intervals under laboratory conditions

Source of variation	DF	SS	MS	F-value	Pr > F
Model	10	16215.83	1621.58	82.06	< 0.0001
Error	16	316.17	19.76		
Corrected Total	26	16352.00			
Replication	2	128.45	64.22	3.25	0.0654
Treatment	8	16087.38	2010.92	101.76	< 0.0001

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
0.98	12.68	12.26	5.31	46.67	2.92

Annex 29: Summary table for analysis of variance on mean percent mortality of *T. absoluta* caused by different concentrations of insecticides after treatment exposure at 72 hours intervals under laboratory conditions

Source of variation	DF	SS	MS	F-value	Pr > F
Model	10	16341.14	1634.41	76.30	< 0.0001
Error	16	342.65	21.43		
Corrected Total	26	16683.79			
Replication	2	42.83	21.42	1.0	0.3897
Treatment	8	16298.31	2037.29	95.13	< 0.0001

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
0.98	12.27	11.07	5.15	52.34	2.92

Annex 30: Summary table for analysis of variance on mean percent mortality of *T. absoluta* caused by different concentrations of insecticides after treatment exposure at 1 day intervals under glasshouse conditions

Source of variation	DF	SS	MS	F-value	Pr > F
Model	6	10836.04	167.67	81.37	< 0.0001
Error	8	164.45	20.56		
Corrected Total	14	10200.49			
Replication	2	53.88	26.94	1.31	0.3219
Treatment	4	9982.16	2495.54	121.4	< 0.0001

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
0.98	12.42	12.63	4.54	35.89	3.36

Annex 31: Summary table for analysis of variance on mean percent mortality of *T. absoluta* caused by different concentrations of insecticides after treatment exposure at 3 day intervals under glasshouse conditions

Source of variation	DF	SS	MS	F-value	Pr > F
Model	6	14837.92	2472.99	30.42	< 0.0001
Error	8	650.36	81.30		
Corrected Total	14	15488.28			
Replication	2	79.14	39.57	0.49	0.6317
Treatment	4	14758.77	3609.69	45.39	< 0.0001

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
0.95	24.70	19.73	9.02	45.69	3.36

Annex 32: Summary table for analysis of variance on mean percent mortality of *T. absoluta* caused by different concentrations of insecticides after treatment exposure at 5 day intervals under glasshouse conditions

Source of variation	DF	SS	MS	F-value	Pr > F
Model	6	14798.89	2466.48	58.25	< 0.0001
Error	8	338.72	42.34		
Corrected Total	14	15137.62			
Replication	2	45.86	22.93	0.54	0.6018
Treatment	4	14753.84	3688.26	87.11	0.0041

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
0.98	17.83	13.49	6.51	48.24	3.56

Annex 33: Summary table for analysis of variance on mean percent mortality of *T. absoluta* treated with fungal isolates at different concentration after treatment application at 3 day intervals under laboratory conditions

Source of variation	DF	SS	MS	F-value	Pr > F
Model	9	10217.03	1135.22	16.22	< 0.0001
Error	14	962.58	68.76		
Corrected Total	23	11179.61			
Replication	2	149.17	74.59	1.08	0.3648
Treatment	7	10067.86	1438.27	20.92	< 0.0001

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
0.91	20.15	16.47	8.29	50.34	2.98

Annex 34: Summary table for analysis of variance on mean percent mortality of *T. absoluta* treated with fungal isolates at different concentration after treatment application at 5 day intervals under laboratory conditions

Source of variation	DF	SS	MS	F-value	Pr > F
Model	9	8956.32	995.15	14.78	< 0.0001
Error	14	942.85	67.35		
Corrected Total	23	9899.17			
Replication	2	325.79	162.89	2.42	0.1252
Treatment	7	8630.53	1232.93	18.31	< 0.0001

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
0.90	19.95	14.19	8.21	57.82	2.98

Annex 35: Summary table for **a**nalysis of variance on mean percent mortality of *T. absoluta* treated with fungal isolates at different concentration after treatment application at 7 day intervals under laboratory conditions

Source of variation	DF	SS	MS	F-value	Pr > F
Model	9	9399.82	1044.42	12.96	< 0.0001
Error	14	1127.86	80.56		
Corrected Total	23	10527.68			
Replication	2	176.49	88.24	1.10	0.3614
Treatment	7	9223.34	1317.62	16.36	< 0.0001

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
0.89	21.82	14.61	8.96	61.41	2.98

Annex 36: Summary table for analysis of variance on mean percent mortality of *T. absoluta* treated with fungal isolates at different concentration after treatment application at 3 day intervals under glasshouse conditions

Source of variation	DF	SS	MS	F-value	Pr > F
Model	9	13955.73	1550.64	26.46	< 0.0001
Error	14	820.42	58.60		
Corrected Total	23	14776.14			
Replication	2	142.51	71.26	1.22	0.33
Treatment	7	13813.21	1973.32	33.67	< 0.0001

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
0.94	18.61	15.10	7.66	50.68	2.78

Annex 37: Summary table for analysis of variance on mean percent mortality of *T. absoluta* treated with fungal isolates at different concentration after treatment application at 5 day intervals under glasshouse conditions

Source of variation	DF	SS	MS	F-value	Pr > F
Model	9	16512.05	1501.34	19.88	< 0.0001
Error	14	1057.38	75.53		
Corrected Total	23	14569.43			
Replication	2	133.92	66.96	0.89	0.4340
Treatment	7	13378.13	1911.16	25.30	< 0.0001

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
0.93	21.12	15.65	8.69	55.52	2.98

Annex 38: Summary table for analysis of variance on mean percent mortality of *T. absoluta* treated with fungal isolates at different concentration after treatment application at 7 day intervals under glasshouse conditions

Source of variation	DF	SS	MS	F-value	Pr > F
Model	9	14496.23	1610.69	20.15	< 0.0001
Error	14	1118.88	79.92		
Corrected Total	23	15615.12			
Replication	2	191.17	95.58	1.20	0.3315
Treatment	7	14305.07	2043.58	25.27	< 0.0001

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
0.93	21.73	14.21	8.94	62.91	2.98

Annex 39: Summary table for analysis of variance on mean percent mortality of *T. absoluta* treated with fungal isolates at different concentration after treatment application at 10 day intervals under glasshouse conditions

Source of variation	DF	SS	MS	F-value	Pr > F
Model	9	15445.79	1716.20	45.96	< 0.0001
Error	14	522.81	37.34		
Corrected Total	23	15968.59			
Replication	2	66.49	33.24	0.89	0.4326
Treatment	7	15379.31	2197.04	58.83	< 0.0001

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
0.97	14.85	9.09	6.11	67.25	2.98

Annex 40: Summary table for analysis of variance of transformed data after 3rd day treatment application under Field condition at Ambo District, West Shewa, Oromia Regional State, Ethiopia

Source of variation	DF	SS	MS	F-value	Pr > F
Model	10	15396.68	1539.67	33.72	< 0.0001
Error	16	730.51	45.66		
Corrected Total	26	16127.18			
Replication	3	12.74	6.37	0.14	0.8708
Treatment	8	15384	1923.0	42.12	< 0.0001

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
0.95	16.11	20.47	6.76	33.0	2.98

Annex 41: Summary table for analysis of variance of transformed data after 5th day treatment application under Field condition at Ambo District, West Shewa, Oromia Regional State, Ethiopia

Source of variation	DF	SS	MS	F-value	Pr > F
Model	10	7033.73	703.37	10.03	< 0.0001
Error	16	1121.76	70.11		
Corrected Total	26	8155.49			
Replication	3	18.49	9.24	0.13	0.8774
Treatment	8	7015.25	876.91	12.51	< 0.0001

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
0.86	19.97	18.66	8.37	44.87	2.98

Annex 42: Summary table for analysis of variance of transformed data after 7th day treatment application under Field condition at Ambo District, West Shewa, Oromia Regional State, Ethiopia

Source of variation	DF	SS	MS	F-value	Pr > F
Model	10	7158.58	715.86	8.86	< 0.0001
Error	16	1292.62	621933	80.79	
Corrected Total	26	8451.21			
Replication	3	133.66	66.83	0.83	0.4551
Treatment	8	7024.92	878.11	10.87	< 0.0001

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
0.85	21.44	18.69	6.29	48.08	2.98

Annex 43: Summary table for analysis of variance of transformed data after 10th day treatment application under Field condition at Ambo District, West Shewa, Oromia Regional State, Ethiopia

Source of variation	DF	SS	MS	F-value	Pr > F
Model	10	6340.48	634.05	29.04	< 0.0001
Error	16	349.32	21.83		
Corrected Total	26	6689.81			
Replication	3	45.60	22.80	1.04	0.3747
Treatment	8	6294.89	786.86	36.04	< 0.0001

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
0.95	20.04	18.83	8.99		

Annex 44: Summary table for analysis of variance of transformed data after 3rd day treatment application under Field condition at Dandi District, West Shewa, Oromia Regional State, Ethiopia

Source of variation	DF	SS	MS	F-value	Pr > F
Model	10	21499.90	2150.0	152.67	< 0.0001
Error	16	225.32	14.08		
Corrected Total	26	21725.22			
Replication	3	68.88	34.44	2.45	0.1184
Treatment	8	21431.02	2678.88	190.23	< 0.0001

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
0.92	8.95	20.29	3.75	36.48	2.98

Annex 45: Summary table for analysis of variance of transformed data after 5th day treatment application under Field condition at Dandi District, West Shewa, Oromia Regional State, Ethiopia

Source of variation	DF	SS	MS	F-value	Pr > F
Model	10	10819.34	1081.93	23.00	< 0.0001
Error	16	752.71	47.04		
Corrected Total	26	11572.05			
Replication	3	33.35	16.67	0.35	0.7069
Treatment	8	10785.99	1348.25	28.66	< 0.0001

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
0.93	16.36	20.65	6.86	44.54	2.98

Annex 46: Summary table for analysis of variance of transformed data after 7th day treatment application under Field condition at Dandi District, West Shewa, Oromia Regional State, Ethiopia

Source of variation	DF	SS	MS	F-value	Pr > F
Model	10	7472.69	747.27	23.83	< 0.0001
Error	16	501.72	31.36		
Corrected Total	26	7974.41			
Replication	3	58.80	29.40	0.94	0.4120
Treatment	8	7413.88	926.74	29.55	< 0.0001

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
0.94	13.35	20.65	5.60	48.05	2.98

Annex 47: Summary table for analysis of variance of transformed data after 10th day treatment Application under Field condition at Dandi District, West Shewa, Oromia Regional State,Ethiopia

Source of variation	DF	SS	MS	F-value	Pr > F
Model	10	8846.55	884.65	33.17	< 0.0001
Error	16	426.71	26.67		
Corrected Total	26	9273.26			
Replication	3	130.95	65.47	2.46	0.1175
Treatment	8	8715.60	1089.45	40.85	< 0.0001

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
0.95	12.32	19.94	5.16	51.97	2.98

Annex 48: Summary table for analysis of variance of transformed data after 3rd day treatment application under Field condition at Toke kutaye District, West Shewa, Oromia Regional State, Ethiopia

Source of variation	DF	SS	MS	F-value	Pr > F
Model	10	20502.80	2050.28	437.15	< 0.0001
Error	16	75.04	4.69		
Corrected Total	26	20577.84			
Replication	3	11.13	5.57	1.19	0.3306
Treatment	8	20491.66	2561.46	546.14	< 0.0001

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
0.93	12.77	16.57	5.35	32.95	2.98

Annex 49: Summary table for analysis of variance of transformed data after 5th day treatment application under Field condition at Toke kutaye District, West Shewa, Oromia Regional State, Ethiopia

Source of variation	DF	SS	MS	F-value	Pr > F
Model	10	11751.10	1175.11	40.98	< 0.0001
Error	16	458.81	28.68		
Corrected Total	26	12209.91			
Replication	3	8.23	4.12	0.14	0.8673
Treatment	8	11742.86	1467.86	51.19	< 0.0001

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
0.96	12.77	13.32	6.38	40.20	2.98

Annex 50: Summary table for analysis of variance of transformed data after 7th day treatment application under Field condition at Toke kutaye District, West Shewa, Oromia Regional State, Ethiopia

Source of variation	DF	SS	MS	F-value	Pr > F
Model	10	11373.14	1137.31	47.48	< 0.0001
Error	16	383.28	23.95		
Corrected Total					
Replication	3	82.74	41.37	1.73	0.2094
Treatment	8	11290.40	1411.30	58.92	< 0.0001

R-square	LSD	CV (%)	SE±	an Efficacy (%)	Critical value of t
0.97	11.67	19.78	4.89	50.01	2.98

Annex 51: Summary table for analysis of variance of transformed data after 10th day treatment application under Field condition at Toke kutaye District, West Shewa, Oromia Regional State, Ethiopia

Source of variation	DF	SS	MS	F-value	Pr > F
Model	10	11994.24	1199.42	47.24	< 0.0001
Error	16	406.24	23524	25.39	
Corrected Total	26	12400.47			
Replication	3	107.77	53.89	2.12	0.1522
Treatment	8	11886.46	1485.81	58.52	< 0.0001

R-square	LSD	CV (%)	SE±	Mean Efficacy (%)	Critical value of t
0.97	12.02	19.81	5.04	51.37	2.98

Annex 52: Summary table for analysis of variance on cumulative Mean of *Tuta absoluta* caught in sticky traps with different colours at vegetative stage during 2016 under glasshouse conditions

Source of Variation	DF	SS	MS	F-value	Pr > F
Model	7	23.88	3.41	3.56	0.0262
Error	12	11.51	0.96		
Corrected total	19	35.39			
Replication	3	2.72	0.90	0.94	0.45
Treatment	4	21.16	5.29	5.52	0.0093

R-square	LSD	CV (%)	SE±	Mean	Critical value of t
0.87	2.12	26.94	0.98	1.36	3.05

Annex 53: Summary table for analysis of variance on cumulative Mean of *Tuta absoluta* caught in sticky traps with different colours at flowering stage during 2016 under glasshouse conditions

Source of Variation	DF	SS	MS	F-value	Pr > F
Model	7	45.43	6.49	21.94	< 0.0001
Error	12	3.55	0.29		
Corrected total	19	48.98			
Replication	3	0.85	0.28	0.96	0.4440
Treatment	4	44.58	11.14	37.67	< 0.0001

R-square	LSD	CV (%)	SE±	Mean	Critical value of t
0.92	1.17	22.02	0.54	2.47	3.05

Annex 54: Summary table for analysis of variance on cumulative mean of *Tuta absoluta* caught in sticky traps with different colours at fruit setting stage during 2016 under glasshouse conditions

Source of Variation	DF	SS	MS	F-value	Pr > F
Model	7	57.68	8.24	12.61	0.0001
Error	12	7.84	0.65		
Corrected total	19	65.23			
Replication	3	0.28	0.09	0.14	0.93
Treatment	4	57.40	14.35	21.96	< 0.0001

R-square	LSD	CV (%)	SE±	Mean	Critical value of t
0.88	1.75	26.20	0.81	3.09	3.05

Annex 55: Summary table for **a**nalysis of variance on cumulative mean of *Tuta absoluta* caught in sticky traps with different colours at vegetative stage during 2017 under glasshouse conditions

Source of Variation	DF	SS	MS	F-value	Pr > F
Model	7	12.05	1.78	4.47	0.115
Error	12	4.78	0.40		
Corrected total	19	12.22			
Replication	3	0.52	0.17	0.44	0.7307
Treatment	4	11.93	2.98	7.49	0.0029

R-square	LSD	CV (%)	SE±	Mean	Critical value of t
0.82	1.36	28.14	0.63	1.92	3.05

Annex 56: Summary table for analysis of variance on cumulative mean of *Tuta absoluta* caught in sticky traps with different colours at flowering stage during 2017 under glasshouse conditions

Source of Variation	DF	SS	MS	F-value	Pr > F
Model	7	22.97	3.28	12.46	0.0001
Error	12	3.16	0.26		
Corrected total	19	26.13			
Replication	3	1.58	0.53	2.00	0.1672
Treatment	4	21.38	5.35	20.29	< 0.0001

R-square	LSD	CV (%)	SE±	Mean	Critical value of t
0.88	1.11	22.29	0.51	1.74	3.05

Annex 57: Summary table for **a**nalysis of variance on cumulative mean of *Tuta absoluta* caught in sticky traps with different colours at fruit setting stage during 2017 under glasshouse conditions

Source of Variation	**DF**	**SS**	**MS**	**F-value**	**Pr > F**
Model	7	21.63	3.09	7.55	0.0013
Error	12	4.91	0.41		
Corrected total	19	26.54			
Replication	3	0.71	0.24	0.58	0.6421
Treatment	4	20.92	5.23	12.79	0.0003

R-square	**LSD**	**CV (%)**	**SE±**	**Mean**	**Critical value of t**
0.92	1.36	23.75	0.63	1.19	3.05

LIST OF PLATES

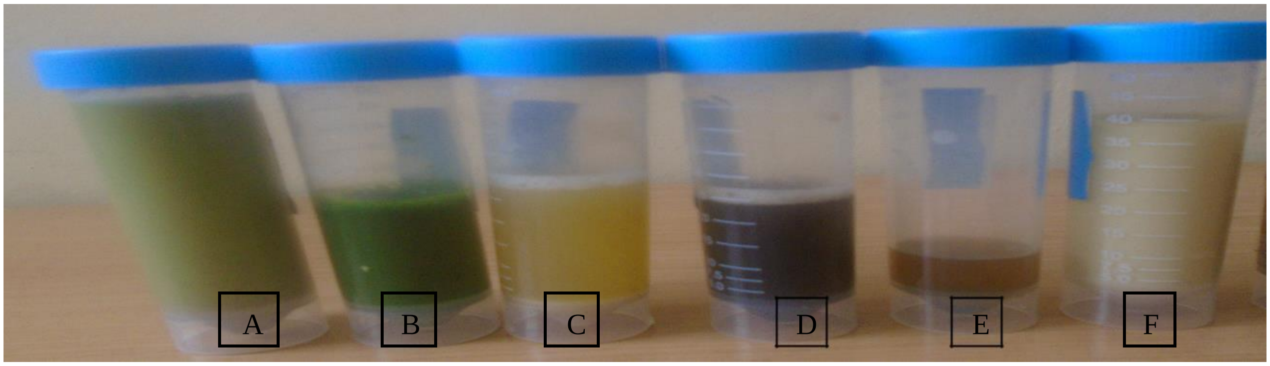

Plate 1: Crude water extract of botanicals

A = *Phytolacca dodecandra* seed B= *Phytolacca dodecandra* leaf C= *Allium sativum* D = *Nicotiana* sp. E= *Cymbopogon citrates* F= *Azadirachta indica*

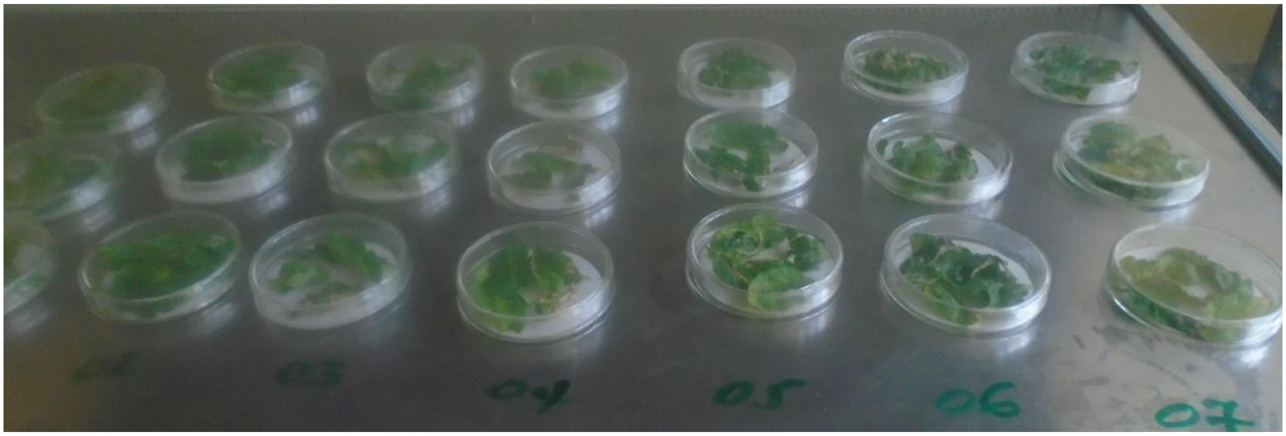

Plate 2: Effect of botanicals on larvae of *T. absoluta* under laboratory conditions

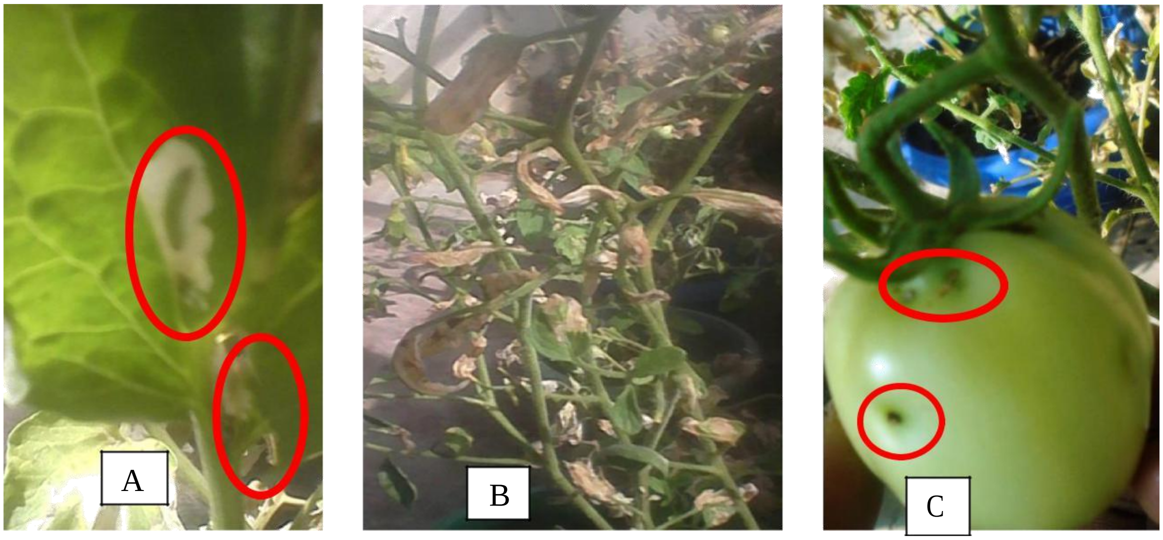

Plate 3: Damage of tomato varieties by *T. absoluta* on leaves and fruits under glasshouse conditions (A) Larva in the leaf surface of tomato, (B) Leaf damaged, and (C) Fruit tunneled

Plate 4: Effect of tomato leafminer, *T. absoluta* on tomato crop during 2015-2016 under glasshouse conditions: (A) Damaged tomato leaf at different phonological stages, (B) Tunneled tomato fruit, and (C) Desiccated leaf

Plate 5: Infested tomato plant by *T. absoluta* during November 2015 to February 2016 under glasshouse condition A: leaves; B: Unripe fruits C: Ripe fruits

Plate 6: Infestation of potato varaities by *T. absoluta* under glasshouse conditions

Plate 7: Non-infestation of pepper varieties by *T. absoluta* under glasshouse conditions during 2015-2017

Plate 8: Effects of *T. absoluta* on Protected and unprotected tomato plants during March to June 2016 under glasshouse. A & C = Protected tomato plants from any pest infestation, B & D = Un protected tomato plants

Plate 9: Glassehouse experiment on management of *T. absoluta*

Plate 10: Field layout of bio-pesticide treatments on *T. absoluta* at different locations of Western Shewa, Centeral Ethiopia: A=Ambo district, B= Danadi district, and C= Toke kutaye district