



**Sudan University of Science and Technology**  
**College of Graduate Studies**

**Effect of Some Local Plants Extracts on Control of the African  
Bollworm, *Helicoverpa armigera* (Noctuidae) in Tomatos**

**أثر بعض المستخلصات المحلية في مكافحة دودة اللوز الافريقية *Helicoverpa armigera*  
في الطماطم**

**By**

**Saeed Hassan Mohammed Zein Mostamhil**

**B.Sc. Agriculture(Crop Protection)**

**University of Gezira(1999)**

**M.Sc. (Crop Protection)**

**Faculty of Agriculture ,University of Khartoum(2007)**

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**Main Supervisor :Dr .Saif Eldin Mohamed Kheir**

**Co- Supervisor: Prof. Dr. Awad Khalafalla Taha**

**Department of Plant Protection**

**College of Agricultural Studies - Shambat**

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## ***DEDICATION***

*To my wife, family members and friends  
who gave me support and made this work  
possible*

## **A KNOWLEDGEMENTS**

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## ABSTRACT

These studies were conducted in White Nile State on one of the important field pests of tomato, African bollworm *Helicoverpa armigera* Hubner.

The studies included a field survey to determine the presence and distribution of African bollworm in the White Nile State and a laboratory studies to calculate morphometrics measurement s of insect stages , to study some biological activities of the insect and to evaluate

the efficacy of leaves and stem ethanol and aqueous extracts of *Lantana camara* (Verbenaceae), *Ocimum basilcum* (Lamiaceae), *Lupinus termis* (Leguminaceae), *Solenostemma argel* (Asclepiadaceae) and *Nicotiana rustica* (Solanaceae). The evaluated efficacy parameters included mortality, repellency and antifeeding effects on *Helicoverpa armigera* larvae.

The study showed that the pest was present and distributed along the state in all studied area (Elsoufi, Eltawiella and Elmogamaat) with high rate of infestation in December.

The results showed that at an average temperature range of (  $27 \pm 1.1$  °C -  $30 \pm 2.9$  °C) and relative humidity of (  $35.2 \pm 3\%$  -  $40.7 \pm 2.8\%$  ), the morphometrics and the development duration of insect stages were as follows:

The morphometrics study of insect stages revealed that the egg size was (  $0.5 \pm 0.07$  and  $0.44 \pm 0.04$  ) mm in length and width respectively; with yellowish white colour.

The 1<sup>st</sup> , 2<sup>nd</sup> , 3<sup>rd</sup> , 4<sup>th</sup> and 5<sup>th</sup> larval instars size were (  $1.42 \pm 0 \pm$

0.1) , ( $4.2 \pm 0.19$  and  $1.1 \pm 0.2$  ) , ( $8.27 \pm 0.7$  and  $2.3 \pm 0.2$  ) , ( $10.1 \pm 0.9$  and  $2.7 \pm 0.2$  ) and ( $17.1 \pm 3.8$  and  $3.3 \pm 0.2$  ) mm in length and width respectively and they exhibited variations in colour. The pupa was  $18.6 \pm 1.4$  and  $4.9 \pm 0.33$  mm in length and width respectively; the colour started as green then turned brown. The adult male wing span and body length were  $27.6 \pm 2.1$  and  $16 \pm 1$  mm respectively , the female wing span and body length were  $35.8 \pm 2.3$  and  $19.2 \pm 2.4$  mm respectively, the colour ranged from grey to brown with markings on wings.

The incubation period of eggs was  $3.9 \pm 0.9$  days. Where as the larval periods were  $2.4 \pm 0.5$ ,  $2.7 \pm 0.8$ ,  $2.9 \pm 1.1$ ,  $3.2 \pm 0.9$  and  $3.4 \pm 0.5$  days for the 1<sup>st</sup> , 2<sup>ed</sup> , 3<sup>rd</sup> , 4<sup>th</sup> and 5<sup>th</sup> instars respectively. Prepupa and pupa periods were  $3.1 \pm 1.2$  and  $9.6 \pm 1.5$  days respectively.

The percentage of eggs hatching, pupation and adult emergence were  $66.9 \pm 7.1$ ,  $74.6 \pm 4.6$  and  $83.1 \pm 5.8\%$  respectively.

During the period preoviposition (  $2.2 \pm 0.44$ ) days, oviposition ( $10 \pm 0.4$  ) days and postoviposition ( $3.2 \pm 0.45$ ) days, The female laid a total number of  $469.4 \pm 90.9$  eggs.

Generally ethanol extracts were more effective than aqueous extracts. The significant effects on mortality were observed after 72 hours, the effects were dose and time dependent.

The  $LC_{50}$  of *Lantana camara* was 2.76 followed by *solenostemma argel* 6.35 , *Ocimum basilicum* 16.19 , *Lupinus termis* 17.05 and *Nicotiana rustica* 50 .

The different extracts induced significant feeding ratio (FR) except *L.termis* and the effects were dose dependent. The lowest (FR) 0.0054 was caused by *Ocimum basilicum* followed by *Lantana*

*camara* 0.0084, *Solenostemma argel* 0.012 and 0.013 for both *Nicotiana rustica* and *Lupinus termis*, while the Dimethoate and the control showed (FR) of 0.0148 and 0.019 respectively.

The highest percent repellence was shown in the first hour post application and caused by *Ocimum basilicum* 66.7% followed by *Lantana camara* 33.3%, *Nicotiana rustica* 0%, *Lupinus termis* -13.4% while *solenostemma argel* -26.%, and the effects were decreased by increasing the time.

The Preliminary phytochemical screening of *Lantana camara*, *solenostemma argel* and *Ocimum basilicum* indicated the presence of alkaloids, sterols, triterpenes, flavonoids, tannins and saponins compounds in addition to the presence of cyanogenic glycosides in *S, argel*.

## الخلاصة

أجريت هذه التجربة بولاية النيل الأبيض على إحدى أهم الآفات الهامة للطماطم في الحقل وهي دودة اللوز الأفريقية . *Helicoverpa armigera* Hubner . وقد اشتملت الدراسة على مسح حقلي لمعرفة تواجد الآفة وانتشارها في الولاية , وعلى دراسة معملية لحساب حجم وفترة كل طور من أطوار الحشرة مع تسجيل بعض الأنشطة الحيوية للحشرة , ولتقييم الكفاءة الحيوية للمستخلص الكحولي (ايتانول) والمائي لنباتات اللانتانا , الريحان , الترمس , الحرجل و التبغ . وقد اشتملت معايير تقييم الكفاءة على دراسة الاثر القاتل والمانع للتغذية و الطارد على يرقات دودة اللوز الأفريقية .

أظهرت نتائج المسح الحقلية تواجد و انتشار الحشرة على طول الولاية في مناطق الدراسة (الصوفي , الطويلة و المجمعات ) و ان اعلى مستوى للاصابة بالحشرة كان في شهر ديسمبر .

كما أظهرت النتائج انه وتحت ظروف تتراوح فيها متوسط درجات الحرارة بين ( 27  $\pm$  2.9 - 30  $\pm$  1.1  $\pm$  ) و متوسط الرطوبة النسبية بين ( 40  $\pm$  2.8% - 35.2  $\pm$  3 ) ان متوسط حجم و فترة كل طور من اطوار الحشرة كانت كالآتي :  
حجم البيض 0.07  $\pm$  0.5 مم طولاً و 0.04  $\pm$  0.4 مم عرضاً و لونه ابيض مزرق . اما الاطوار اليرقية كان طولها وعرضها على التوالي كالآتي , الطور الاول 0.7  $\pm$  4.1 , 0.2  $\pm$  1.1 مم , الطور الثاني 0.7  $\pm$  4.1 , 0.2  $\pm$  1.1 مم , الطور الثالث 0.7  $\pm$  8.27 , 0.2  $\pm$  2.3 مم , الطور الرابع 0.9  $\pm$  10.1 , 0.2  $\pm$  2.7 مم , والطور الخامس 0.2  $\pm$  3.8 , 0.2  $\pm$  3.3 مم ومختلفة الالوان . العذراء طولها 1.4  $\pm$  18.6 مم وعرضها 0.33  $\pm$  4.9 ولونها يبدأ اخضر ثم يتغير الى البني.

كما اظهرت النتائج ان طول الاجنحة في الذكر 2.1  $\pm$  27.6 مم وفي الانثى 2.1  $\pm$  35.8 مم وطول الجسم كان 1  $\pm$  16 مم في الذكر و 2.9  $\pm$  19.2 مم في الانثى و الحشرة البالغة لونها رمادي الى بني بها علامات على الاجنحة.

متوسط فترة حضانة البيض 0.9  $\pm$  3.9 يوم . ومتوسط فترة الاطوار اليرقية كانت 2.4  $\pm$  0.5 يوم للطور الاول , الطور الثاني 0.8  $\pm$  2.7 يوم , الطور الثالث 1.1  $\pm$  2.9 يوم

، الطور الرابع  $0.9 \pm 0.2$  يوم و الطور الخامس  $0.5 \pm 3.4$  يوم .متوسط فترة ما قبل العذراء  $1.2 \pm 3.1$  يوم ، ومتوسط فترة العذراء (  $1.5 \pm 9.6$ ) يوم .

متوسط نسبة فقس البيض %  $7.1 \pm 66.9$ ، والتعذير  $4.6 \pm 74.6$  اما نسبة خروج الحشرات الحشرات فكانت %  $5.8 \pm 83.1$

متوسط عدد البيض للانثى  $90.9 \pm 469$  بيضة و فترات ما قبل وضع البيض ، وضع البيض ، وما بعد وضع البيض كانت على التوالي  $0.44 \pm 2.2$  ،  $0.4 \pm 10$  ،  $0.42 \pm 3.2$  يوما .

وبصورة عامة المستخلصات الكحولية كانت أكثر تأثيرا في الموت من المستخلصات المائية و أعلا تأثير معنوي كان بعد 72 ساعة من المعاملة كما ان التأثير يزداد بازدياد الجرعة والزمن .

الجرعة النصف قاتلة كانت بتركيز 2.76 للالانتانا يليها ، 6.35 للحرجل ، 16.19 للريحان ، 17.05 للترمس و 50 للتبغ.

كل المستخلصات احدثت تأثيرا معنويا على معدل التغذية ما عدا مستخلص الترمس ويزداد التأثير بازدياد الجرعة حيث كان معدل التغذية خلال 24 ساعة 0.0054 للريحان ، 0.0084 للالانتانا ، 0.012 للحرجل ، 0.013 لكل من التبغ و الترمس و 0.0148 للدايمثويت و 0.019 للشاهد.

اعلا نسبة طرد كانت في الساعة الاولى من المعاملة حيث كانت % 66 للريحان ، 33.3 للالانتانا ، ، صفر للتبغ ، 13.4- للترمس و 26- للحرجل كما تقل نسبة الطرد بازدياد الزمن .

كما دلت نتائج المسح الكيمياء للنباتات الاكثر فعالية الالانتانا والريحان والحرجل على وجود المركبات التالية الكلويد ، فلافونويد ، ترايترين ، صابونين ، استيرول و تانين بالاضافة الى وجود جلايكوسايد في الحرجل.





# CHAPTER ONE

## INTRODUCTION

Solanaceae family is important chiefly for foods, medicines, poisons, oils and ornaments. The major food yielding plants are *Solanum tuberosum* (Potato), *Solanum melongena* (Egg plant), *Lycopersicon esculentum* (Tomato), *Capsicum frutescence* (Pepper).

Several members of this family are found in the Sudan, particularly, tomato which is considered as one most vegetable crop due to its economic and nutritional value (Ahmed, 2009), and is ranking second to onion in vegetables consumption. It is grown almost in every part of the country during the winter season and sometimes during the rainy season .The total cultivated area in the Sudan is about 60.158 ha (Abdallbagi *et al.*, 2010), Ahmed (2009) reported that it occupies about 28% of total area under vegetable production in the Sudan, but Mahadi (2006) mentioned that it occupies nearly about 75% of the total area under vegetables.

Insect pests are known to cause significant damage to crops and affect agricultural return. The monetary loss due to feeding by larvae and adult insects alone contributes to billion dollars USA per annum (Jacobson, 1981).

Tomatoes, wherever grown, are hosts of a wide variety of insect pests and between 100 and 200 species are reported to attack tomatoes worldwide (Lange and Bronson, 1981). .In the Sudan tomato is mostly attacked by, leaf miner *Liriomyza* sp., white fly, and American bollworm *Helicoverpa* (Mahadi, 2006).

*Helicoverpa armigera* is a destructive polyphagous pest occurring on tomato, and many others crops, inflicting substantial loss every year (Reed and Pawar, 1982; Sharma, 2001; Talekar\_ *et al.*, 2006). In the

Sudan African bollworm (ABW) cause reduction in tomato yield by more than 50% (Mahadi, 2006). Bollworms are relatively safe from natural enemies because of the cryptic feeding habits of the larvae within cotton boll. Therefore, large numbers of *Helicoverpa armigera* in cotton and other vegetables survive that may disperse widely, producing progeny that damage high- value crops (Cabanillas and Raulston, 1995; Michael and Donald, 1996).. *Helicoverpa armigera* is also characterized by it's high mobility and fecundity and it has shown great capacity to develop resistance to synthetic insecticides used in its management (Armes *et al.*, 1996; Ramasubramaniam and Regupathy ( 2004)

Repeated application of synthetic organic insecticides resulted into the pest resistance and out break. Most insecticidal compounds fall within four main classes, the organochlorines, organophosphates, the carbamates and pyrethroids. Out of these, the major classes in use today are pyrethroids. organophosphates and carbamates. There are problems of pesticides resistance and negative impacts on non-target organisms including man and the environment (Singh *et al.* 2000; Singh and Saratchandra, 2002; Isman 2006). The environment problems caused by overuse of pesticides have been the matter of concern for both scientists and public in recent year, It has been estimated that about 2.5 million tons of pesticides are used on crops each year and the worldwide damage caused by pesticides reach \$100 billion annually. The reasons for that are two fold: (1) the high toxicity and non biodegradable properties of pesticides and (2) the residues in soil , water resources and crops that affect public health (Koul *et al.*, 2008). Thus , one needs to search the new highly selective and biodegradable pesticides to solve the problem of long term toxicity to mammals and, on the other hand ,one must study the environmental friendly pesticides and develop techniques that can be used to reduce pesticides use while maintaining crop yields. Such

difficulties have caused natural products to gain attention. Among the natural products, plant derived pesticides are more acceptable. This acceptance is due to their abundance, their being nature-friendly, being least toxic to natural enemies, their effect on limited species, species, fast degradation, low phytotoxicity and low toxicity to vertebrates (Kim *et al.* 2003), biodegradable to non-toxic products (Hashem and Youssef, 1991), less persistence in the environment (Isman, 2006). These factors are why natural products are considered suitable for developing new groups of healthy and safe insecticides for insect control .

Plant derived phytochemicals have been widely used in the management of the agricultural pests (Choudhary *et al.*, 2001), also in the Sudan the interest has reversed recently for using natural products for the control of pests and diseases vector ( eg: Siddig ,1991; Abdelgadir, 1993; Elkamali ,2001; Ali, 2004 , Edriss *et al.*,2008) .

Plants are endowed with a potential to produce a range of secondary metabolisms like alkaloids, terpenoids, flavonoids, phenols, glycosides, sitosterol and tannins. These phytochemicals are known to protect the plants from the attack of insect pests (Ahmed, 2007) . A screening of plant extracts from plants could lead to the discovery of new agents for control (Oliverira,1997), active substances extracted from plants may not only act as toxicant, but also as insect growth regulators (Bowers *et al.*, 1972), as repellents or synergists (Su and Harvot,1981) and( Burfield and Reeke, 2005) or phagodeterents (Meisner *et al.*,1988),however, there are many plant families that possess various chemical compounds which act as antifeedants, repellent insecticides or growth inhibitors to many insect species (Srivastava *et al.*,, 2001; Formisano *et al.*, 2008; Odeyemi *et al.*, 2008) .

A number of many plants species has been shown to have pesticidal and antifeedant activity against *Helicoverpa armigera* (Ramy *et al.*,

2008) of which neem has been subjected to extensive investigation by Chopar *et al.*, (1994). Studies have shown that *Acorus calamus*, *Annona squamosa*, *Vitex negundo* are effective in the management of *Helicoverpa armigera* (Murugan *et al.*, 1998).

The undesirability and potential dangers of applying large and uncontrolled quantities of insecticides to tomato plant designed for human consumption must be recognized. The successful use of plant products in the control of certain insect species depends on contained substances that inhibit the developmental process of those insects (Kristensen and Jespersen, 2003). From these points of view, the objectives of this research are to:

- To monitor population density and distribution of *Helicoverpa armigera* in White Nile State.
- To study biology and measurement of different stages of *Helicoverpa armigera* .
- To investigate effects of *Lantana camara*, *Solenostemma argel*, *Ocimum basilicum*, *Lupinus termis* and *Nicotiana rustica* extracts on mortality of *Helicoverpa armigera* 3<sup>rd</sup> instar larvae .
- To investigate effects of *Solenostemma argel*, *Lantana camara*, *Lupinus termis*, *Nicotiana rustica* and *Ocimum basilicum* extracts on repellency of larvae of *Helicoverpa armigera* .
- To investigate antifeedant effects of *Solenostemma argel*, *Lantana camara*, *Lupinus termis*, *Nicotiana rustica* and *Ocimum basilicum* extracts on larvae of *Helicoverpa armigera*.
- To estimate phytochemical screening for *Lantana camara*, *Ocimum basilicum* and *Solenostemma argel*.

# CHAPTER TWO

## LITERATURE REVIEW

### 2.1 Tomato *Lycopersicon esculentum* Mill (Solanaceae)

Tomato, *Lycopersicon esculentum* Mill., is one of the most popular and widely grown vegetables in Sudan. Its grown almost every where around the country in an area estimated to total 65.000.acres. However, it is mostly located in central and eastern Sudan (Omara, 2001).

#### 2.2.1 Taxonomy:

In 1753, Linnaeus placed the tomato in the genus *Solanum*. In 1768, Philip Miller moved it to its own genus, naming it *Lycopersicon esculentum*. Tomato is belonging to the family Solanaceae (Leslie, 1976).

#### 2.1.2 Botanical description:

Tomato plants are dicots, and grow as a series of branching stems, with a terminal bud at the tip that does the actual growing. Tomato vines are typically pubescent. meaning covered with fine short hairs. Most tomato plants have compound leaves The flower yellow, with five pointed lobes on the corolla (Leslie, 1976).

#### 2.1.3: Tomato Pests and diseases:

Common diseases of tomato are leaf curl, verticillum wilt, fusarium wilt, nematodes, tobacco mosaic virus, and alernaria. Some common tomato pests are stink bugs, cutworm, tomato hornworm and tobacco hornworm, aphids, cabbage loopers, white flies, tomato fruitworms, flea beetles, red spider mite (Hahn and Fetzer, 2009)

In Sudan tomato is infected by diseases like tomato yellow leaf curl disease and powdery mildew and attacked by some insects like leaf miner, bollworm and white flies (Omara, 2001). Recently tomatoes attacked were by serious tomato leafminer *Tuta absoluta*.

## **2.2 *Helicoverpa armigera***

### **2.2.1. Taxonomic literature:**

*Helicoverpa armigera* (Hunber,1808) (Lepidoptera: Noctuidae) synonyms (*Heliothis obsotea*) and (*Heliothis armigera*), (Ahmed,1983), because of the number of crops that this pest affects, has many common names, sacra border straw worm, corn worm, African bollworm, American bollworm and tomato bollworm (Begemann and Schoeman, 1999). The American bollworm up to beginning of 1950,s was called African bollworm , and corn earworm (CEW) in oldworld at northern America , respectively, these two old names referred to the same species *Helicoverpa armigera* (Ahmed,1999). Hardwick (1965) classified all the species under the genus *Heliothis* in to two groups:one of them under the old genus *Heliothis* and the other under the name *Helicoverpa*, this involves the following species in the Sudan *Helicoverpa fletcheri* Hardwick, *Helicoverpa peltigera* Schiffermullar and *Helicoverpa armigera* (Hunb) (Ahmed, 1999).

### **2.2.2. Distribution**

The pest is present and widespread in Asia, Africa and Oceania (EPPO, 2006), also occurring through Africa, the middle east the southern Europe, India, central and southern Asia, eastern and northern Australia, Newzialand and many Pacific Island (Fitt, 1989).

In the Sudan it is found wherever cotton is grown (Schumetterer, 1969). Insect surveys covering all major cotton growing area in the Sudan, revealed the presence of ABW in the southern province, Shambat. Nuba mountains , Meridi,Yambio, Gash and Tokear Delta, Gedarif, White Nile and Gezera (Joyce,1951)

### **2.2.3 Biologyof *Helicoverpa armigera*.**

#### **2.2.3.1 Life Cycle.**

In more temperate regions of *H. armigera*'s range, it completes 2½ generations per year with pupae over-wintering in the soil . In more tropical areas, this species may continue to be active throughout the year with multiple overlapping generations, during its lifetime. Fully developed larvae move to the soil where they form an earthen cell 2-10 cm below the surface. The pupal period generally lasts from 8-21 days depending on temperature. Diapausing pupae can over-winter in the soil in more temperate areas for long periods (175 days). In tropical areas, diapauses can be induced by drought. Adults, that are mainly nocturnal in their mating and egg laying activities, can live as long as 2-3 weeks (Jallow and Zalucki, 1998).

*Helicoverpa armigera* is highly migratory, and can fly long distances (Fitt, 1989). It tends to over-winter as pupae in the soil of late-planted summer crops and is therefore considered to remain in the local cropping area. Moth emergence from these over-wintering pupae often begins between September and October and may take several generations to build to high numbers. Crop damage by *H. armigera* is therefore most common during the later parts of summer. The ecology of this species is responsible for it being a predominantly late summer pest. The localized activity of *H. armigera* within cropping regions is also thought to contribute to its ability to readily build resistance to insecticides. The larvae can, however, be difficult to kill with insecticides, even if susceptible. Once they shelter in plant structures or plant tissue, they are difficult with insecticides (Deuter *et al.*, 2000).

In general, *Helicoverpa* species larvae may not always be able to feed on the plant structure on which they hatched, or be able to continue feeding once they have started, if plants respond to feeding by mobilizing secondary metabolites. The preference for fruiting structures and the



tendency to move from one fruit to another, often without consuming each fruit completely.

#### **2.2.3.2 Eggs :**

The freshly laid eggs of *H. armigera* were yellowish-white and glistening at first but changed to dark brown before hatching. The apical area of egg was smooth and the rest of the surface sculptured in the form of longitudinal ribs. The incubation period of eggs was recorded as  $3.37 \pm 0.09$  days, whereas, the size of eggs varied from 4.42-0.60 mm in length and 0.40-0.55 mm in width. The eggs which did not hatch within five days were discarded and termed as infertile eggs. The infertile eggs were yellowish, become increasingly yellow and shriveled after 3 days. The average percentage of eggs hatched was recorded as  $53.33 \pm 0.47\%$ . were yellowish-white to reddish-brown (Ali *et al.*, 2009).

The eggs are spheres approximately 0.5 mm in size, which are initially white in coloration, then darken to grayish brown prior to eclosion. The eggs are sculptured with vertical ridges of alternating length, which surround a smooth apical area that contains the micropyle (King, 1994).

Eggs incubation lasts 3-14 days, depending on temperature (King, 1994; Fowler and Lakin; 2001; CAB, 2003). Eggs hatch in about 3 days at 25c, but at lower temperature, hatching may take up to 11 days (CAB, 2003). Eggs are dome-like with a ribbed surface. The eggs are small (approximately 0.5 mm in diameter; about half the size of a pinhead) and sub-spherical (dome shaped with a slightly flattened bottom) in shape. Eggs are usually laid singly, making detection difficult near buds, flowers, fruits, or on leafy plant parts. They are initially pale green, sometimes with black dots, and they later change to cream and then brown (Zalucki *et al.*, 1986; Deuter *et al.*, 2000; Defra, 2001; CPC, 2002).

### 2.2.3.3 Larvae:

First instars larvae have a black to brown head capsule and a yellowish-white body with a spotted appearance due to sclerotized setae, tubercle bases and spiracle. Larval color darkens with successive molts for the 6 instars stages observed for *H. armigera*, and coloration can vary considerably due to diet content. Larval size in the final instar ranges from 3.5 – 4.2 cm in length (King, 1994). In later instars, stripes, continuous or broken appear on the dorsal and lateral sides.

The larval period of *H. armigera* completed is through six distinct instars. The first and second larval instars are provided with dark brown to black head capsule. However, the length and width of these instars were  $1.40 \pm 0.06$  and  $0.45 \pm 0.01$  mm;  $3.88 \pm 0.11$  and  $0.75 \pm 0.01$  mm,  $7.90 \pm 0.19$  and  $2.28 \pm 0.04$  mm;  $12.83 \pm 0.45$  and  $2.85 \pm 0.04$  mm;  $20.97 \pm 0.61$  and  $3.25 \pm 0.04$  mm,  $32.50 \pm 0.35$  and  $4.03 \pm 0.04$  mm respectively, (Ali *et al.*, 2009). The average duration of these instars were  $2.27 \pm 0.08$  and  $2.42 \pm 0.08$ ,  $2.67 \pm 0.07$ ,  $2.83 \pm 0.07$ ,  $3.40 \pm 0.10$  and  $3.37 \pm 0.11$  days respectively (Ali *et al.*, 2009). During the first and second instars, the color of larvae was more uniform and the movement was very little. The prolegs were developed in third instar stage on 3rd, 4th, 5th, 6th and 10th abdominal segments and remained until last (sixth) larval instar (Bhatt and Patel; 2001). The full grown larva was straw-yellow to green with lateral brown stripes and the head as well as prothoracic legs were dark brown to black in color. Tubercles and spiracles of the larvae were also brown to black giving them a spotted appearance (Cunningham *et al.*, 1999). Larvae may complete up to 7 instars, though generally between 5 and 7 instars (Twine, 1978; King 1994; Fowler and Lakin, 2001). The time required to complete each larval stage varies considerably depending on host plant, temperature and other factors. In laboratory studies, the complete larval period (all instars combined) last between 12-36 days (Kirkpatrick, 1962; Bhatt and Patel, 2001; Fowler and Lakin, 2001).

During summer larval development is completed in 14-18 days, first instars larvae have high mortality rate, most likely caused by larvae movement or predators (Kyisaki and Zalucki, 1991). The mortality occurs in full sun on leaf, s surfaces (king, 1994). larval developments depend primarily on temperature and secondary on host nutritional quality ( King ,1994; CAB, 2003). Before feeding on their host plant ,newly hatched larvae typically consume all or part of their eggs shells ; larvae may then feed on leaf surface or floral structure ,moving about the plant for short distance before selecting a preferred feeding spot (King,1994).When larval mobility is limited ,development times can vary widely and survival is largely determined by host plant selection of egg-laying females (Jallow and Zalucki,1996; Gu and Walter,1999).Small, young larvae have the ability to feed inside floral structure ,detectable only by a small hole with pun silk at the entrance and visible frass; larger larvae feed with apportion of their body outside the floral or fruiting structure, It is particularly damaging to crop because larvae can move from plant to plant ,particulary when food is scarce (King,1994). Late instars larvae are more damaging to the host plant due to their attraction to full buds (Mabbet *et.al.*,1980)."Antagonism "and "cannibalism" have been observed among older larvae on corn in situations where several eggs were deposited, larvae move between 2.5 to 17.5 cm below the soil surface to pupate depending to soil moisture, organic matter on surface and other factors (King,1994). Early instars are predominantly green, and appear spotted because of dark spiracles and tubercle bases. Larvae pass six instars, and ultimately reach 30 to 40 mm in length, and they usually display striped patterns and may vary in color from light green to brown to black and have distinct hairs when held up to the light. There is a good deal of color variation in the larvae. For example, larvae may have white, instead of black spots. Superimposed on the dorsal bands are

numerous lighter longitudinal lines, which are wrinkled or wavy. There are often dark, raised spots on the back, at the base of fine hairs. In *H. armigera*, there is a dark triangular area on the back of the first abdominal segment of the third, fourth and fifth instars of the larvae. Larvae have a posture when disturbed characteristic of a number of species in this family: it lifts its head and curls it under the front of the body. If even more disturbed, it lets go and drops, rolling into a spiral (Zalucki et al., 1986, Deuter *et al.*, 2000; Defra, 2001; CPC, 2002).

#### **2.2.3.4. Pre-pupae:**

In this stage, the fullgrown larvae become sluggish, wrinkled with suspended feeding and movement. The pre-pupa was noticed as light green yellowish in color but later on it turned to dark brown. This stage lasted for 1-3 days with an average of  $2.15 \pm 0.16$  days . The average length and width of pre-pupa of *H. armigera* varied from 22.50 - 29.00 and 3.90 - 5.00 mm (Ali *et al.*, 2009) .

#### **2.2.3.5 Pupa:**

The pupa was of obtect type with mahogany-brown color. The surface was smooth and it is rounded both anteriorly and posteriorly, with two tapering parallel spines at the posterior tip( Bhatt and Patel,2001). The average length and width of pupa were  $19.00 \pm 0.30$  and  $5.72 \pm 0.08$  mm, respectively. However, this stage took minimum and maximum period of 10 and 14 days, respectively (Ali *et al.*, 2009). Less frequently, pupation occurs with in a spun web on the host plant or on the soil surface depending on temperature, the pupal stage lasts between 6-35 days, unless the insect goes into diapause, in which case pupation may require several months. It over winters as pupae ( Kirkapatric,1962; King,1994; Akashe *et.al.*,1997; Maelzer and Zalucki ,1999; Fowler and Lakin, 2001 ; CAB,2003). Diapause is facultative and occurs during the pupal stage (

King,1994). Diapause induction begins when larvae are exposed to day length 11.5-12.5 hours ,and low temperature 19-23 c , or when larvae are exposed to lengthy periods of extremely hot and dry weather (35c) (King, 1994; Zhou *et al.*,2000; Shimizu and Fujisaki, 2002; CAB, 2003).

#### **2.2.3.6. The adult:**

The adult moth is described as a typical Noctuid. The stout bodied moth has a wing span range of 35-40 mm and the body length range of 18-19 mm. The coloration varies from dull greenish yellow to olive gray or brown and females are darker than males(King, 1994).

The adult moth was stoutbodied with broad thorax. The forewings have a line of seven to eight blackish spots on the margin and a broad, irregular, transverse brown band. Hind wings are pale-straw color with a broad dark-brown border that contains a paler patch; they have yellowish margins and strongly marked veins and a dark, comma-shaped marking in the middle. Antennae are covered with fine hairs (Garcia-Tejero, 1957; Hardwick, 1965; Cayrol, 1972; Delatte, 1973).

#### **2.2.3.7. Fecundity**

Female laid an average  $413.00 \pm 1.89$  eggs over a reproductive /oviposition period of 4.60-5.80 days (Ali *et al.*, 2009). The eggs were laid singly during night time due to nocturnal behavior (Sharma;2001). The percentage hatching/viability of eggs was  $53.33 \pm 0.47\%$ . On the other hand, female required an average  $2.45 \pm 0.08$  days,  $5.33 \pm 0.12$  and  $2.00 \pm 0.05$  days for preoviposition, oviposition and postoviposition, respectively (Butler *et al.*,1996; Jallow *et al.*,1998; Ali *et al.*,2009). A single female may oviposit from 500 to 3,000 eggs, averaging close to 1,000. As many as 1,500 eggs may be laid by a female over a 14 days, with peak laying at 7 day (Jallow and Zalucki, 1998).

Because *H.armigera* exhibits overlapping generations, it can be difficult to determine the number of complete generation, but typically 2-

5 generations are achieved in subtropical and temperate regions, and up to 11 generations can occur under optimal condition, particularly in tropical areas (Tripathi and Singh, 1991; King, 1994; Fowler and Lakin, 2001). In Australia up to 7 generations can be completed in warmer seasons of country (Kirkpatrick, 1962). Larvae do not diapause, approximately 4-5 generations can be completed from late September to early April and 1-2 generations can be completed in winter (Kirkpatrick, 1962; Maelzer and Zalucki, 1999). In China *Helicoverpa armigera* complete 2-3 generations annually (Xiao et al., 2002). In eastern New Zealand Coastal regions, a more temperate climate where the average summer temperature is 23.5°C, 2-3 generations are completed (Cameron et al., 2001). King (1994) reviews several adult longevity studies and report range in adult life span of 5-30 days. Adult longevity depends on several factors including pupal weight, food (nectar) supply, food quality (source contents), temperature, water availability, disease pressure and predator activity (King, 1994). Without adequate food sourcing they die within a few days (King, 1994).

Under adverse conditions, moths can migrate long distance (King, 1994; Zhou et al., 2000; Shimizu and Fujisaki, 2002; CAB, 2003). Adults can disperse within a distance of 10km during "non-migratory flight" and hundreds of kilometers (up to 250km) when making "migratory flights" which probably occur when host quality or availability declines (Saito, 1999; Zhou et al., 2000; Casimero Fujisaki, 2001; Fowler and Lakin, 2001).

#### **2.2.4. Host plants:**

The cotton bollworm *Helicoverpa armigera* (Lepidoptera: Noctuidae) is a major threat to extensive agriculture (Siggaard et al. 2002). Its wide dissemination and pest status has been attributed to its polyphagy, and its ability to undergo both facultative diapauses and seasonal migration (Fitt,

1989). The species is migratory on all continents, and is a key pest on all of them (Feng *et al.*, 2005). This pest feeds on a number of host plants, many of which occur or produced in the Sudan, eg. Cotton (*Gossypium sp.*), okra (*Alemoschus esculentus*), sorghum (*Bicolous sp.*), pear millet (*Pennisctum typhoideum*), maize (*Zea maize*), lubia (*Lablab purpureous*), ground nut (*Arachis hypogaea*), beens (*Vicia faba*), french bean (*Phaseolous vulgaris*), peas (*Pisum sativium*), tomaso, egg plant, sunflower (*Helianthus annus*), cucumber (*Cucumis sativius*), alfa alfa (*Medicago sativa*), chilli pepper (*Capisicum futescens*), pigeon peas (*Cajanus cajanus*), millets sp. and various herbaceous plants, including weeds such as tabar (*Ipomoea cordofana*), hamboko (*Abtilon sp.*) (Schumetterer, 1969 and Balla, 1978).

There are preferences between host-plant species (Roome, 1975; Hillhouse and Pitre 1976). In the Sudan Gezira the crop groundnut is an important alternative host when sorghum and cotton are not available or are not at the attractive growth stage (Topper, 1987). Three consistent groupings were found, sunflower, *Helianthus annus*, tobacco, *Nicotiana tabacum*, and maize, *Zea mays*, being most favoured followed by cotton, *G. hirsutum*, soybean, *Glycine max*, and lucerne, *Medicago sativa*, which were of intermediate preference, and then pigweed, *Trianthema portulacastrum*, cabbage, *Brassica oleraceae*, and linseed, *Linum usitatissimum* which were least favoured. It is notable that cotton was not in the most preferred grouping. It may be chosen in the field because it is more abundant than other host plants which means it is available for a longer time (Wardhaugh *et al.* 1980). Goyal and Rathore (1988) regarding growth and fecundity of followed, in order, by pea, *Pisum sativum*, linseed, *Linum usitatissimum*, tomato, *H. armigera* on different host plants: gram, *Cicer arietinum*, being most favourable *Lycopersicum lycopersicum*, and then cotton, *G. hirsutum*. In a field study of a cotton-

okra intercrop Singh *et al.* (1993) found an oviposition preference for okra, *Abelmoschus esculentus*..

### **2.2.5 Potential Economic Impact.**

*Helicoverpa armigera* is a severe economic pest in most places where it occurs (Agusti *et al.* 1999, CAB 2003). *Helicoverpa armigera* is an important pest of cotton, particularly in Australia and China (King, 1994). All parts of the cotton plant are vulnerable to attack. Cotton yields were reduced by 50-60% by *H. armigera* each year from 1980-1990 in China (Xiao *et al.* 2002). In Queensland Australia, *H. armigera* damage accounted for 7% yield loss in cotton in spite of pest control costs of A\$800/ha in 1998 (Sequeira 2001). In Andhra Pradesh region of India, *H. armigera* reduced yields of seed cotton from 436 kg/ha in 1986-87 to 168 kg/ha in 1987-88 (Sekhar *et al.* 1996, Loganathan *et al.*, 1999). Significant tomato crop loss also occurred in Burkina Faso, India and New Zealand, particularly in unsprayed or late season varieties (Bouchard *et al.* 1992, Cameron *et al.* 2001). In New Zealand, *H. armigera* attacked Monterey pine and “consumed more than 50% [of the] foliage [off] about 60% the trees” (CABI/EPPO 1997). Pigeonpea and chickpea are severely damaged in India, where losses up to 90-100% in the 1992/93 and 1997/98 growing seasons have been reported. Worldwide, annual losses from this pest on chickpea are approximately 10%, equaling \$300 million dollars (Shanower *et al.*, 1997; Mulimani and Sudheendra, 2002; Sidde Gowda *et al.* 2002). The greatest damage is caused to cotton, tomatoes, maize, chick peas, alfalfa and tobacco. The economic threshold of harmfulness in central Asia is three to five larvae per hundred plants of long-staple cotton and eight to twelve larvae per hundred plants on medium-staple cotton (Waring *et al.*; 2003).

Control is most often in the form of chemical sprays, but *H. armigera* has developed resistance to many insecticides (Mabbett *et al.* 1980,



Maelzer and Zalucki, 2000). Overall, the pest affects economies by reducing yields, lowering crop values, and causing market loss from quarantine restrictions (Fowler and Lakin, 2001). The pest is listed by the European and Mediterranean Plant CAPS PRA: *Helicoverpa armigera* Protection Organization as an A2 quarantine pest and is considered a quarantine pest by the Caribbean Plant Protection Commission (CPPC), and the country of Brazil (EPPO, 2000).

#### **2.2.6. Control measures:**

Control measures include the growing of resistant varieties, weeding, inter-row cultivation, removing crop residues, deep autumn ploughing, winter watering to destroy the pupae, the use of insecticides or biological control through the release of monitoring is possible by the use of sex pheromone traps (Waring *et al.*; 2003).

##### **2.2.6.1 Cultural control.**

Several cultural practices have been suggested to control. Cultural control methods include the use of short season cultivars to reduce the time over which the crop is at risk, trap cropping, post-harvest cultivation to destroy pupae, avoiding over-application of fertiliser which makes the crop more attractive to pests, and removal of alternative hosts of *H. armigera* (Sundaramurthy and Chitra, 1992; King, 1994; Russell *et al.*, 1998 ).

##### **2.2.6.2 Biological control.**

The African bollworm is subject to attack by several insects parasites: six are larval parasites belonging to the family Tachinidae: *Hypeuchalcidia soudanensis* Skefl., *Drino imberbis* Wied., *Exorista fallax* Meig ., *Goniophthalmus halli* Mesnil. *Sturmia inconspicus* Meig. And *Isomera cinerascens* Ro-dami. The family Braconidae is represented by two species: *Cardiochiles* sp., *Microbracon kirkpatricki* Wilk. and family Eulophidae represented by the ectoparasite *Euplectrus laphygmae*

Ferr. In addition, the mud wasp, *Eumenus maxilosus*, predate on the larvae (Balla, 1978) and entomophages such as *Trichogramma* spp. and *Habrobracon hebetor* (Waring *et al.*; 2003). According to Sundaramurthy & Chitra (1992) the naturally occurring predators *Menochilus sexmaculatus*, *Coccinella* sp., *Scymus* sp., spiders and birds are important for regulating bollworm populations in the Indian subcontinent and *Chrysopa carnea* was used in an augmentation programme in Gujarat. A commercialised strain of *Helicoverpa* nuclear polyhedrosis virus (NPV) has been found useful and economic in trials in the USA, Mexico and Australia. Control problems when it was promoted in India (Russell *et al.*, 1998). However, it had problems of low persistence (1-2 days) and there were quality network for NPV was more limited than that for conventional insecticides (Shanower, 1999).

### **2.2.6.3 Chemical control.**

The dominant control method that has been used in cotton at least since the 1950s for protection against *H. armigera* has been application of insecticides, mainly directed at the larval stage (Wilson, 1982 and King, 1994). Chemical control is still regarded as the most promising method, and has been adopted as a standard practice for the control of most pests. In the past, several insecticides used were approved by the Pest and Diseases Committee to control the African Boll worm. These comprised Azodrin 55.2% W.S.C., Cidial 50% EC., Malthion 57% E.C., Malthion 96% UL, Sevin 85% W.P., Thimul 35 ULV, Sevamol WP. Several other insecticides are used now to control the African Bollworm on cotton, these include Sumicidin 4 UL, Deltamethrin/Dimethoate UL, Endosulfan /Dimethoate UL, Cypermethrin/Profenophos UL, Fenvalerate/Dimethoate ULV, and the new mixtures such as Sumicidin + Dimethoate E.C., Decis+Ekatin W.E.C, Dursban+Endosulfan E.C., Curacon+Endosulfan E.C., Birlane+ Endosulfan E.C., while some

other insecticides are used to control this pest in vegetable crops, such as Diazinon, Malthion E.C., Kafil10 E.C., Danitol 10%, Danitol-S, and Sumicidin 20% (Balla, 1978). More recently insecticides used Bulldock 125 Sc (Abdalla, 2002).

Application of insecticides against *H. armigera* can disrupt biocontrol of other pest species such as red spider mites and white flies.

#### **2.2.6.4. Alternative Control Methods**

##### **2.2.6.4.1 Host-Plant Resistance**

The development of crop cultivars that resistant to, or tolerant of, feeding damage has great potential in the regional management of *helicoverpa armigera* (Kennedy *et al.*,1987) . Many crops display characters that can be exploited by breeders to reduce attractiveness to ovipositing adults or suitability for larvae to feed (Kennedy,1987).

##### **2.2.6.4.2 Genetically engineered plants**

Two approaches have been used successfully to find genes encoding insect resistance, one to use entomocidal bacterium *Bacillus thuringiensis* as sources of resistance genes, and the other is identify and use resistance genes present in plant source. Inhibitors of proteolytic enzyme, protease inhibitors, Tiol protease inhibitors,  $\alpha$ -amylase inhibitors, trypsin/trypsin inhibitors and CPTI inhibitors which have been effective against *H. armigera* were identified as potential resistance source from plant (Hilder *et al.*,1987).

The most effective and efficient delivery system for Bt toxins is the transgenic plant which effective to control bollworm (Krattiger,1997).

##### **2.2.6.4.3. Semiochemicals**

Mating disruption using sex pheromone did not control *H. armigera*, probably due to its high mobility which increases the chances of mated females immigrating from outside the treated area (King, 1994).

### **2.3 Essential oils constituent and their efficacy.**

Essential oils are defined as any volatile oil (s) that have strong aromatic component and that give distinctive odors, flavors or scent to plant. Essential oils are a complex mixture of natural organic compounds which are predominantly composed of terpenes such as myrcene, pinene, terpinene, limonene, p-cymene,  $\alpha$ - and  $\beta$ -phellandrene etc , and terpenoids such as acyclic monoterpenealcohol (geraniol, linalol), monocyclic alcohol (menthol, 4-carvomenthol , terpineol , carveol , borneol), aliphatic aldehydes (citral, citronella, perillaldehyde) , aromatic phenols (carvacrol, thymol , safrol, eugenol), bicyclic alcohol (verbenol) , monocyclic ketones (menthone, pulegone , carvone), bicyclic monoterpene ketones (thujone, verbenone, fenchone), acids (citronellic acid , cinnamic acid) and esters (linalyl acetate). Some essential oils may also contain oxides (1, 8-cineole), zingiberene, curcumene, farnesol, sequiphellandrene, termerone, nerolidol (Koul *et al.*, (2008).

Insecticides and growth inhibitors essential oil constituents are primarily lipophilic compounds that act as toxins, feeding deterrent, and oviposition deterrent to a wide variety of insect pests. Insecticidal properties of monoterpeneoids to the house fly, red flour beetle, and the southern corn root worm have been reported (Rice and coats, 1994). Eugenol was reported

as toxin Asian army worm, *Spodoptera litura* Fabricius, granary weevil, *Sitophilus granaries* (Linnaeus), western corn root worm , *Diabrotica virgifera* and common house fly, *Mosca domestic* (Obeng-ofori and Rerchumth, 1997; Lee *et al.*, 1997; Hummelbrunner and Isman, 2001). Eugenol is also active against , *Drosophila melanogaster*, yellow fever mosquito *Aedes aegypti* and American cockroach , *Periplanata americana* (Bhatnagar *et al.*, 1993; Ngoh *et al.*, 1998).

Similarly, thymol induces toxicity in, *M. domestica* and *S. litura* (Lee *et al.*, 1997; Hummelbrunner and Isman, 2001) and also toxic to,

*D.melongaster* and northern house mosquito, *Culex pipiens* (Franzios *et al.*, 1997; Traboulsi, *et al.*, 2002). Citronellal toxic to, *M.domestica* and *S.litura* (Franzios *et al.*, 1997; Lee *et al.*, 1997; Chang and Ahn, 2001; Hummellrunner and Isman, 2001; Lee *et al.*, 2001; Chang and Cheng,2002)

Meepagala *et al.*, (2006) found that apoil isolated from *Ligusticum hultenii* exhibited high termiticidal activity of 100% with in 11 days after treatment and similar effect was shown by vulgarone isolated from *Artemisia douglasiana*, where as cncinin isolated from *Centaurea maculosa* showed mortality of 81% with in 15 days after treatment when applied at 1(w/w) concentration to these termites.

Lichtenstien *et al.*,(1974) have reported that carvone isolated from aerial parts of dill plants (*Arethum graveolus* ) was insecticidal to *Drosophila* and *Ades sp.* it also supported larval and adult survival (Ouden *et al.*,1993). Lee *et al.*,1997) evaluated acute toxicity of 34 naturally occurring monoterpenoids against the insect species , they reported that citronellic acid and thymol were the most toxic to house fly , while citronellol and thujone were most effective against western corn root worm. Hierro *et al.*, (2004) has reported that the action of different monoterpenic compounds against *Anisakis simplex* larvae and found that geraniol, citronellol, citral, carvacrol and cuminaldehyde were active at 12.5µg/ml/insect concentration. Menthone, Tran's anethole and cinnamaldehyde are well known anti insect compound that have been studied against variety of insects with wide range of dosages required to kill50% population (65-1735µg/insect) ( Harwood *et al.*, 1990; Lee *et al.*, 1997). Eugenol from cloves, *Eugenia cryophyllus*; 1.8-cineole from *Eucalyptusnella* from lemon grass, *Symbpogon nardus*; pulegone from *Mentha pulegium*, and thymol and carvacrol from *Thymus vulgaris* among the most active constituents against insects. Pulegone is shown

effective against *M.domestica* , *D . virgifera*,*P.saucia* and *S.Litura* in the range of LD50=38-743.9µg/insect (Harwood *et al.*, 1990; Lee *et al.*, 1997) .

## **2.4. Plants used:**

### **2.4.1. *Lantana camara*: commo nname,lantana; family(Verbenaceae)**

#### **2.4.1.1. Botanical description**

Among the innumerous species studied scientifically are the species of the genus *Lantana*, which belongs to the family Verbenaceae. The family Verbenaceae comprises one hundred genera and about 2600 species distributed in tropical and subtropical regions around the world . *Lantana* is mainly native to the tropical and subtropical Americas, but a few taxa are originally from tropical Asia and Africa; currently, they occur in approximately fifty countries with a very large number of species and subspecies. This genus includes herbaceous and shrubby plants, which can reach a height of over 2 m, where they are very often planted for decorative purposes due to the beauty of their flowers (Joly, 1993). *Lantana camara* L., commonly known as wild sage, is the most widespread species of this genus, growing luxuriantly at elevations up to 2000 m in tropical, sub-tropical and temperate regions (The genus *Lantana* was described by Linnaeus in 1753 and contained seven species, six from South America and one from Ethiopia (Munir, 1996; Ghisalberti, 2000).

#### **2.4.1.2.General bioactive activities of lantana genus.**

Species of genus *Lantana* are plants practically immune to attack by herbivores, due to the presence of a great diversity of secondary metabolites (Kohli *et al.*, 2006). They also have anti-rheumatic, stimulant and sudorific properties, and are used to treat broncho-pulmonary disorders, and some species are utilized in biological control as a pesticide

(Ghisalbert, 2000; Dua *et al.*, 2003 and 2010). Organic extracts and essential oils of Lantana have shown a wide variety of biological and pharmacological activities. *L. hispida* Kunth traditionally is used in Mexico to treat tuberculosis, bronchitis, cough, cold, asthma, stomach ailments, kidney pain and diarrhea (Jiménez-Arellanes *et al.*, 2007). In Bolivia, leaves of *L. cujabensis* Schauer are crushed between fingers and sniffed to treat head colds (Okunade and Lewis, 2004). *L. viburnoides* var *kisi* has ethnobotanical importance in Tanzania, where it is used to repel mosquitoes and in traditional medicine for stomach ache relief (Innocent *et al.*, 2008). *L. lilacina* Desf. *L. salvifolia* Jacq is a plant widely used in the Congo as an herbal tea. The decoction of the leaves is used against typhoid fever (Ouamba *et al.*, 2006). *L. indica* Roxb. is used as a sudorific, intestinal antiseptic and diaphoretic, and in treatment of tetanus, rheumatism and malaria in Indian medicine (Ghisalberti, 2000). *L. canescens* Kunth is used mainly for xeriscaping, as it is drought-tolerant. In Northeast Brazil, the local community in Timbauba city (Pernambuco State) uses this species as an analgesic (Sena Filho *et al.*, 2010). The concoction from the leaves of *L. radula* Sw. is used in Brazil as a tea to treat coughs, influenza and bronchitis (Sena Filho *et al.*, 2010). *Lantana trifolia* L. is popularly used to treat colds, flu and sore throat (Silva *et al.*, 2005). *L. fucata* Lindl. is a Brazilian species used in folk medicine for the treatment of respiratory disorders (Julião *et al.*, 2009). *L. camara* has been used for the treatment of various human ailments, such as ulcers, malaria, influenza, tumors, swellings, bilious fever, eczema eruptions, stomach ache, toothache and as antiseptic for wounds (Ghisalberti, 2000). *L. montevidensis* Briq. have been used in folk medicine for the same purposes as *L. camara* (Nagao *et al.*, 2002). The plant is said to have cormitative, anti spasmoidic, and anti rheumatic uses in traditional medicine (Girme *et al.*, 2006).

The plant has antibacterial, antifungal (Kumar *et al.*, 2006), insecticidal (Abdel-Hady *et al.*, 2005), and nematicidal activity (Oamar *et al.*, 2005). Several previous reports have described antifungal, (Tripathi and Shukla 2002, Kumar *et al.* 2006), anti proliferative (Saxena *et al.* 1992, Nagao *et al.*, 2002), and antimicrobial activities of *L. camara* (Saxena *et al.* 1992, Juliani *et al.* 2002, Kasali *et al.* 2002) include termicidal activity reported recently by Verma and Verma (2006). Besides, the plant has been shown to have toxic and repellent effects against certain insect pests of stored grains (Ogendo *et al.*, 2004) and termiticidal effects against adult termite workers (Rajesh and Suman, 2006). The plant has antibacterial, antifungal (Kumar *et al.*, 2006), antioxidant (Basu and *al.*, 2006), insecticidal and nematicidal activity (Oamar *et al.*, 2005).

#### **2.4.1.3 Chemical constituents.**

The toxic effects of the leaves have usually been attributed to a series of pentacyclic triterpenes, in which lantadenes A and B are typical members (Sharma *et al.*, 1988).

*Lantana camara* whole plant and plant parts viz). leaves, flowers, and essential oils have been thoroughly studied for their chemical compositions, previously and currently (Saleh, 1974; Hart *et al.*, 1976; Sharma and Sharma, 1989; Siddiqui *et al.*, 1995; Ghisalberti 2000). All these studies have revealed the presence of terpenoids, steroids, and alkaloids as major chemical constituents in *L. camara* (Saleh, 1974; Hart *et al.*, 1976; Sharma and Sharma, 1989; Siddiqui *et al.* 1995.). However, sesquiterpenes with mainly  $\beta$ -caryophyllene, zingiberene, -humulene, arcurcumene gemacrene-D and major leaf and flower essential oil constituents bisabolene were reported as (Singh *et al.* 1991; Nagassoum *et al.*, 1999; Khan *et al.*, 2002; Andersson and Dobson, 2003).



Compounds isolated from *Lantana camara* include triterpenoids, flavonoid and phenyllethanoid glucosides, iridoidglycosides, furanonaphthoquinones. Some of which could be responsible for the observed insecticidal properties (Morton, 1994; Siddiqui *et al.*, 1995, Sharma *et al.*, 2000; Yadav and Tripathi, 2000). The active ingredient, lantadene A and lantadene B and high flavonoid content, which associated with deterrent against insects, are present in lantana (Ghisalberti, 2000). The chemical composition of the whole plant and parts and essential oils were reported to be influenced by geographical, and seasonal factors as well as developmental stages of the concerned plant, previously, Randrianalijaona *et al.*, (2005) have reported the several changes in the chemical composition of essential oils in more than seventy *lantana camara* from different parts of the world. Very recently, we reported ontogenic variation in secondary metabolites such as venolic, anthocyanins, and proanthocyanidins in *lantana camara* (Bhakta and Cianjewala, 2009). Lantadenes present in all *L. camara* is believed to be responsible for almost all the biological activities (Barre *et al.*, 1997)

In addition, other secondary metabolites such as alkaloids, terpenoids and phenolics could be held partially responsible for some of these biological activities (Barre *et al.*, 1997). However constituents like 1,8-cineol, subinone and caryophyllene and other major constituents viz. , E-nerolidol, Bicycloger mocene- me and pinene identified in leaf essential oils were also found to be responsible for the biological activities of essential oils (Chowdhury *et al.*, 2007; Sonibare and Effiong, 2008). The phytochemical present in *L.camara* are saponine, flavonoids, terpenoids and cardiacglycosides (Kumar, 2008). Previously Randrianalijaona *et al.*, (2005) have reported composition of essential oils in more than the seasonal changes in the chemical seventy *L. camara* from different parts of the world. Very recently, we reported ontogenic variation in secondary

metabolites such as phenolics, anthocyanins, and proanthocyanidins in *L. camara* (Bhakta and Ganjewala, 2009). However, in *L. camara* very few studies have so far been focused on the influence of seasonal, genetic, ontogenic, and developmental factors of the chemical composition. Lantanin by Louw (1943), lantadene B (Barton *et al.*, 1954), lantanolic acid (Barua *et al.*, 1969). Various steroids, terpenoids, and flavonoids have isolated by different groups from the different parts of the plant (Johns *et al.*, 1983; Begum *et al.*, 2000 and Pullaiah, 2006). lantanoic acid and camaranoic acid, lantic acid (Begum *et al.*, 2008), camarinic acid (Siddiqui *et al.*, 1995), camangeloyl acid, camarinin (Begum *et al.*, 2003, 2006), oleanonic acid, and ursonic acid (Siddiqui *et al.*, 2000) lantanolic acid (Begum *et al.*, 2008, lantanilic acid (Barua *et al.*, 1976, 1985),  $\alpha$ -amyrin,  $\beta$ -sitosterol and lantadene B (Ahmed *et al.*, 1972), lantoic acid (Roy and Barua, 1985), lantadene D (Sharma *et al.*, 1990), lantadenes. (Sastry and Mahadevan, 1963; Sharma *et al.*, 2000), lantanolic acid, oleanolic acid, 22/-O-angeloyl-oleanolic acid, 22  $\beta$ -O-seneciroyl-oleanolic acid, 22 $\beta$  hydroxyl-oleanonic acid, 19 $\alpha$ -hydroxy ursolic acid and a new triterpenoid 3 $\beta$ - isovaleroyl-19 $\alpha$ -hydroxy-ursolic acid (lantaiursolic acid) (Pandey *et al.*, 1993), camarinic acid, camaric acid, camarilic acid and camaracinic acid (Siddiqui *et al.*, 1995; Begum *et al.*, 1995), 25-hydroxy-3-oxolean-12-en-28-oic acid, hederagenin and 19-hydroxyursolic acid (Singh, *et al.*, 1996), novel trans lactone containing euphane triterpenes A, B and C (O'Neill *et al.*, 1998), phenylpropanoid glycosides verbascoside, isoverbascoside isonuomioside A, calceolarioside E and derhamnosyl verbascoside (Taoubi *et al.*, 1997), martynoside and verbascoside (Syah *et al.*, 1998), theveside (Ford and Bcndall, 1980).

#### **2.4.1.4. Isecticidal activities.**

A wide variety of extracts/essential oils of *Lantana* and their constituents possess varying degrees of pest-controlling properties. A

recent study investigated the insecticidal activity of essential oil from the leaves of *L. camara* L. against mosquito vectors (Dua *et al.*, 2010. Dharmagadda *et al.* (2005) showed that 200 ppm oil of *L. camara* L. produced 100% mortality in *C.quinquefasciatus* larvae in 15 minutes.

Dried leaf powder was reported to show antifeedant activity when tested against rice weevil *Cytophilus oryzae* (Morallo-Rajessus and Decena, 1982) , where its wood extracts showed toxicity to weevil *Sitophilus grenarius* (Morallo-Rajessus and Decena 1984), its leaf and flower extracts showed antifeedancy (Pandey *et al.*,1986) and toxicity (Saxena,1992) when tested against *Callosobruchus Chinensis*. Biological activity field pests, it's leaf water extract was reported to be toxic to black bear aphid *Aphis rumicis* and repellency to the diamond black moth, *Plutella xylostella* (Jacobson, 1975).

However Pandey *et al.*, (1982) found its extract in ether to show antifeedant activity to 3<sup>rd</sup> instars larvae of the mustard sarfly *Athalia proxima*. It's oil was reported to be repellent to honey bee s (Singh; 1977). Further, it's dried leaf powder showed insecticidal, repellent and antifeedant activity against the Asian corn borer *Ostrin furnacalis* (Morallo-Rajessus and Decena,1982) and against turnip aphid *Hyadaphis erysimic*. However was found to be toxic to the rice brown plant hopper *Nilaparvata lugens* (Morallo-Rajessus and Decena, 1984). Lal *et al.*, (1987) reported it's dried leaf powder over the stored potato to show significant protection to the potato against infestation of potato tuber moth *Phthormiaea operculella*.

Reddy *et al.*, (1990) reported it's petroleum ether extract to reduce the population of the brinjal spotted leaf beetle *Henosepilachna vigintioctopunctata* significantly when sprayed on the potted plants,similary it's leaf extract showed toxicity to 2<sup>nd</sup> instars larvae of the hairy caterpillars *Amsactamoovei*. Sharma *et al.*, (1990) . Also reported

it's crude leaf extract in water to show significant antifeedancy to the 3<sup>rd</sup> instars larvae of jule semi-hopper *Anomis sabulif* and 4<sup>th</sup> instars larvae of Bihar hairy caterpillar, *Spilosoma* Saxena *et al.*, (1992 ) has studied the insecticidal action of aerial parts of *lantana camara* against *Callosobruchus chinensis* (Coleoptera: Bruchidae) and found that 10-43% morality complete feeding deterrent and loss of fecundity was also noticed .Ogendo *et al.*, (2003) has studied the Efficiency of Lantana against *Sitophilus zeamaze*(Coleolptera: Curculionidae) in stored maize grains and it was found that 82.7-90% mortality was caused by *lantana camar*. Suryakala *et al.*, (2007) did topical application for insect growth regulating activity on *Dysdercus koengii*,a pest of cotton plant, it was observed that the freshly moulted 5<sup>th</sup> instars resulted in various morphological abnormalities. Mesbah *et al.*, (2006) reported that, all the efficiency tested essential and or volatile oils acted principally as insect growth inhibitors (IGI) other than antifeedant causing disruption of insect development, abnormal larvae,pupae,adults that were lead finally to death.Akhtar *et al.*, (2007), stated that most extracts and botanicals tested proved to be strong growth inhibitors, contact toxins and significant feeding deterrent to lepidopteron species.

Arti *et al.*,. (2009) stated that morphological changes were observed on 4<sup>th</sup> insrars treated with different formulated bases of lantana leaves extracts such as general sluggishness and cessation of feeding , the legs were all black and paralysed and significant mortality rate.

The morphological changes in 4<sup>th</sup> instars larvae of *Helicoverpa armigera* on application of leaf extract of *Lantana camara* (L.) were evaluated .The powder was extracted on ethanol at different dosages were applied on different developmental stages of the larvae, abnormal morphological changes with mortality was observed . The larval pupal intermediates and abnormal pupae were also observed, when 4<sup>th</sup> instars

were treated with different formulations the morphological changes were observed. General sluggishness and a cessation of feeding were observed after 2 days of treatment that increase significantly at the time enhances. Gradually the body became black particularly towards the posterior position. The body got paralyzed with dark brown to black skin and green color was observed in leg region of the mid gut segments. The whole body turned brown with to black leaving a slight faded portion in posterior part through the legs were all black and paralyzed. During the later phases, dry appearance of body, crumpled skin, over all shrinkage of body segments and reduction due to the shorting of body segments can be seen (Arti *et al.*, 2009).

Kumar and Maneemegalai (2008) investigated the methanol and ethanol extracts of leaves and flowers of *L. camara* L. and showed mosquito larvicidal activity against 3<sup>rd</sup> and 4<sup>th</sup> instars larvae of the mosquito species *A. aegypti* and *C. quinquefasciatus*. Extracts at 1.0 mg/mL caused maximal mortality in *A. aegypti* exposed for 24 h. In the case of *C. quinquefasciatus*, maximal mortality was seen when the concentration was increased to 3.0 mg/mL. Repellent properties of different fractions obtained from *L. camara* L. flowers have been evaluated against *Aedes* mosquitoes (*Ae. Albopictus*, *Ae. Vittatus* and *Ae. Aegypti*) (Dua *et al.*, 2003). The results showed that one application of the chloroform fraction gave 100% protection for 2 h and up to 75.8% protection at 7 h against *Ae. Mosquito* bites. Significant reduction in aphid establishment A methanolic extract of *L. camara* L. was tested on larval weight, pupation and adult emergence of cabbage butterfly (Sharma & Mehta, 2009 b). *L. camara* resulted in significantly lower effect on reduction in weight (1.25%). Pupal formation increased significantly (0.0-43.1%) with a decrease in concentration from 5.0 to 1.25%. A similar trend was observed with respect to adult emergence of *Plasmodiophora brassicae*

(Sharma & Mehta, 2009 a). On the other hand, the methanolic extract of *L. camara* caused a (less than 50%) at 5.0%. *L. viburnoides* var kisi was tested for insecticidal activity against late 3<sup>rd</sup> or early 4<sup>th</sup> instars larvae of *Anopheles gambiae* s.s., which were exposed to various concentrations of extracts, fractions, blends and pure compounds (Innocent *et al.*, 2008). The crude extract (LC50 7.70 ppm at 72 h) and fractions exhibited different levels of mosquito larvicidal activity with subtraction of some fractions resulting in activity enhancement. The active fractions contained furanonaphthaquinones regioisomers (LC50 5.48-5.70 ppm at 72 h) and camaric acid (29) (LC50 6.19 ppm at 72 h) as active principles, while betulinic acid (65) (LC50 <10 ppm in 72 h) was obtained from the least active fraction.

Extracts of *L. camara* leaves were studied for their termiticidal effects against adult termite workers (Verma & Verma, 2006). Only 5% chloroform extract exhibited excellent termite mortality. Iannacone & Lamas (2003) studied the effects of extracts of *L. camara* L. on eggs, first instars larvae and adults of *Phthorimaea operculella* in bioassays of insecticidal effectiveness. The results showed that hatched eggs were affected by the hexane extract, and that first instars larva mortality was affected by hexane, acetone and water extracts at 10% concentrations.

The petroleum ether and methanol extracts of the aerial part of *L. camara* L. have been reported to be toxic to *Callosobruchus chinensis* (Dixit *et al.*, 1992). In other studies, the essential oils of leaves and flowers of *L. camara* L. revealed insecticidal activity against 3<sup>rd</sup> instars larvae of *Musca domestica* (Abdel-Hady *et al.*, 2005). Essential oil of *L. camara* L. leaves also showed insecticidal properties against 3<sup>rd</sup> instars larvae of *Helicoverpa armigera*, causing 56% inhibition (Kathuria and Kaushik, 2006), and activity against fresh 5<sup>th</sup> instars nymphs of *Dysdercus similis* (Singh & Upadhyay, 1993). Research studies on plant essential oils and

their constituents as fumigants against stored product insects have been reviewed (Rajendran and Sriranjini, 2008). *Lantana camara* L. showing fumigant and topical toxicity extracts. (Remia and Logaswamy, 2010).

#### **2.4.2. *Solenostemma argel*: Common name, Argel; family(Asclepiadaceae)**

##### **2.4.2 .1. Botanical Description.**

Erect perennial under shrubs, reaching up to 60 cm high. Leaves opposite, lanceolate to oblong-ovate, with acute to subacute apex, cuneate base. The inflorescence is cymose. Bracts broad, linear-lanceolate, acute. Flowers white. Fruit a follicle, 5 cm long, 17-18 mm broad, ovoid lanceolate and acuminate at the apex. It is very hard and dark purple. Seeds are turgid, ovoid, channelled down one face, minutely tuberculated, bearing one apical tuft of hairs (Osborn, 1968 and El-Fishawy, 1976). Is a member of family Asclepiadaceae. Hargal grows naturally in North parts of the Sudan and extend from Barbar to Abu Hamad. It was also widely distributed through North Africa (Egypt, Libya and Algeria and Saudi Arabia (Ahmed, 2004).

##### **2.4.2 .2. General uses:**

*Solenostemma argel* is a medicinal African herb that has long been used by Africans for its antispasmodic, purgative, loss of appetite and carminative effects. It has also been known for its anti-famontory, immunostimulatory as well as antimicrobial and fungicidal properties (Hahn *et al.*, 1998).

##### **2.4.2 .3. Chemical constituents**

The family Asclepiadaceae is a rich source of indoline, alkaloids, steroidal, perogen -ine and their glucosides (Si-Qi *et al.*, 1993; Deepak *et al.*, 1989; Srivastava *et al.*, 1993). Its other chemical constituents are cyanogenetic (glucosides, saponin, tannins, coumarins, flavonoids, phenolic acids and triterpenoids Aeri, 2007). Murwan *et al.*, (2010) found that its leaves contained phytic acid and tannin, Inno-centi *et al.*,

(2005) was found that its aerial parts contained two monoterpene, glucosides, apregnane glucoside, benzyl alcohol O- $\beta$  apiofuranosyle (1-6)  $\beta$ glucopyrano -side, 2- phenyl-ethyl O- $\alpha$  arbinopyranosyl (1-6) $\beta$  glucopyranoside, astragalin and kaempferol 3-O-neohesperidose. Shafec *et al.*, (2012) isolated two new natural kaempferol glucosides namely kaempferol 3-O- $\beta$ -D-glucopyranosyl (1-2)  $\beta$ -D-xylopyranoside (S2) and kaempferol 3-O- $\alpha$ -L-arabinopyranosyl (1-2)  $\beta$ -D-galactopyranoside (S3). (Osborn,1968 and El-Fishawy,1976) Sterosterols, choline, flavonoids, glycosides, namely argelin and argelosid and a triterpinoid saponin. Previous studies have reported the presence of monoterpenes (Kamel *et al.*, 2000) pregnane glycosides (Hassan *et al.*, 2001; Hamed, 2001). Palaza *et al.*, (2003) reported the occurrence of novel pregnane glycosides namely argeloside from *S. argel*. Also monoterpene, glycosides, pregnane glycosides, flavonoids and tannins as well as other steroids and alkaloids were isolated and identified from different parts of the herb (Hamed, 2001 ; Kamel *et al.*, 2000) (Fig:2).

#### **2.4.2 .4. Insecticidal activities**

Elkamali (2001) and Al-Dogharri *et al.*, (2002) reported its mosquitocidal effect. Sidahmed *et al.*, (2009) demonstrated its termiticidal effects on cotton soil termite (*Microtermis thoracalis* Sjost.) Incorporated in to rearing media of *Culex pipiens*.1, showed effect on oviposition, egg hatchability and larval viability. The ovicidal effect of *Solenostemma argel* was relatively less pronounced, however the 0.1% concentration reduce egg hatchability by 33.7% ,complete suppression of oviposition with the first 2 days was observed, *Solenostemma argel* was studied against *Spodoptera littoralis* by Abd El-Aziz and Ezz El-Din (2007), and the study showed that 45.25% mortality and the residual affect remained for three days after the treatment , Abd El-Aziz and Ezz El-Din also found that methanolic extract of Hargel when tested against



24h , 48h and 72 hours egg age showed reduction in eggs hatchability by 1.4, 19.22, 66.67% respectively, this result indicate that, in general the higher susceptibility of the older eggs. A moderate knock down effect of *S. argel* was reported with initial time sample, larvae fed on cotton leaves for one day and followed by untreated ones , showed percentage of mortality after three days,20%. however its action was gradually lost (Al-Dogharri *et al.* 2007) . Hargal extract showed best LD 50=0.006 ml/l against *Culex quinque fasciatus* as compared with Usher extract which show LD50 = 0.108 ( Hag El-Tayeb *et al.*, 2009).

Biocide effects were mainly attributed to the presence of a variety of bioactive organic substances mainly terpenes, perogenine, glycosides, alkaloids and sterols (Al-Dogharri *et al.*, 2004). For instance flavonoides were reported toxic to some insects (Salama *et al.*1997), the effect of tannins in growth inhibition of lepidopterous larvae was reported by Klock and Chan (1982).

Kogar (1986) mentioned the alkaloids had repellent, toxic and antifeedant inhibition affect on some insect species. Sterols were also reported by Nayer and Fraenkel (1962) to inhibit the feeding of some insects.

#### **2.4.3.Ocimum basilicum: Common name, Rehan; Family (Lamiaceae)**

Basil belonging to genus *Ocimum* (Lamiaceae) contain up to 150 species of herb and shrub in tropical regions of Asia, Africa, and central and South America (Simon *et al.*, 1990).

##### **2.4.3.1 Botanical Description**

basil is an aromatic plant reaching a height of 60 to 70 cm. It is a glabrous or slightly pubescent herb with petiolate leaves and white or slightly purplish flowers. Sweet basil (*Ocimum basilicum*) will grow to a size of 1-2 feet in height. Basil will prolifically produce large green leaves

measuring around 2 inches in length, throughout the summer. Basil flowers are white, and are commonly removed to increase yield of leaves.

*Ocimum basilicum* is indigenous to France, Italy, Spain, Germany, Haiti, Indonesia, Samoa, Africa, India, Pakistan, and the Philippines.

#### **2.4.3.2. General uses:**

Essential oil distilled from the fresh flowers or the entire basil plant is employed extensively as a flavor in confectionery, baked goods, condiments, and spiced meats and as an aroma in certain perfume compounds. It has antipyretic, antiseptic, diaphoretic, diuretic, and stimulant properties and, therefore, has been recommended for gastric Disorders, malarial fevers and skin diseases.

Basil oil, extracted via steam distillation from the leaves and other parts, is used to flavor foods dental and oral products, and fragrance in traditional rituals and medicines (Simon *et al.*, 1990) ,however that different parts of this plant have been used in medicine practices, the leave of ocimum were used as anti. helminthics,( Javanmardi *et al.*, 2002) and to treat ulcer ,rheumatism, diarrhoe, dysentery,malaria and high blood pressure (Ngassun *et al.*,2003; Nakamura *et al.*,2004).There are reports that extracts of *Ocimum sp.* are effective against HIV1 and HIV2 infection.Basil is well known source of flavoring principles (Javanmardi.,2003).Thai basil oil derived from the aerial part of *Ocimum basilicum* and *Ocimum amricana* has been used science ancient time as traditional for various tropical applications. Such as poultice or slave for insecticides and ring worm (Viyoch *et al.*, 2006). Basil contain phytochemical with significant antioxidant capacities and health benefits (Exarchou *et al.*, 2002).

Active compounds:

Although not all of the repellent compounds in the essential oil have

been identified, cineole, linalool, and methyl chavicol-which account for 3 percent, >50 percent, and 33 percent of the oil, respectively-are implicated (Guenther,1949). The fruit fly attractant has been identified as methyl eugenol. Two compounds, designated as “juvocimene I“ and “juvocimene I I,” are responsible for the juvenilizing effects

#### **2.4.3.3 Chemical constituents**

The compounds of basil essential oil linalool, methyl chavicol, eugenol, estragol, thymol and *p*-cymen were found (Akgul 1989; Khatri *et al.* 1995; Pino *et al.* 1996; Martins *et al.* 1999; Keita *et al.* 2000). It was previously reported that the oil of *O. basilicum* contained linalool , eugenol , (E)- $\alpha$ -bergamotene and thymol . Linalool , eugenol, methyl eugenol and fenchyl alcohol(Akgul 1989). Khatri *et al.* (1995) found methyl chavicol, linalool, methyl eugenol,  $\beta$ -caryophyllene,  $\alpha$ -pinene,  $\beta$ -pinene, limonene and camphene. Marotti *et al.* (1996) reported the presence of linalool, methyl chavicol and eugenol as main components of *O. basilicum*. Özcan *et al.*,(2002) represented that the chemical composition of *O. minimum* oil linalool, eugenol and bornyl acetate .

Basil oil contain bioactive constituents that are insecticidal (Chogo and Cranis, 1981; Charan and Nikam, 1982; Deshpande and Tipinis, 1997; Keita *et al.*, 2000; Salvatore *et al.*, 2004), repellent (Maganga *et al.*, 1996; Tawatsin *et al.*, 2001; Paula *et al.*,2004;Popovic *et al.*,2006), nematocidal (Chatterje *et al.*,1982) , fungi static (. Reuveni *et al.*,1984).These properties can frequently be attribute to predominant essential oil constituents such as methyl charvicol (estragol), methyl eugenol, linalool, champhor, and methyl cinamate (Baritiaux *et al.*,1992) . Aftab (1996) reported constituents of *Ocimum basilicum* are mostly phenolic compounds (volatile oil) e.g. linalool, chavicol, methyl ether (estragol), eugenol, caffeic acid derivative and flavonoids.

Recently Chang (2009) demonstrated that 12 volatile compounds were identified on GC-MS by comparison of MS spectra with these NIST mass spectra database. These chemicals include  $\alpha$  pinene, linalool, trans-anethole, 4-(cymethoxypropyl)-benzene, trans-5-octyl, 3-caryophyllene, methyl eugenol, 3,11-trimethyl-(E)-1,6-dodecatriene-3,2,3-dihydro-1H-indole-[3-ido-2-(iodine thyl)-2-methyl]-1,2,4,5-dimethyl benzene, 1-(1,1-dimethyl)-2-methoxy-4-methyl-3,5-dimethylbenzene. Eugenol is the major constituent of basil and was shown to be effective against *Sitophilus granarius* (Obeg-ofori and Reichumth, 1997).

According to the study of Regnault-Roger (1997), *O. basilicum* L. has linalool, limonene, eugenol and estragole. Bunrathep *et al.*, (2007) found Monoterpenes such as ( $\beta$ -myrcene, limonene, (E)-( $\beta$ -ocimene), oxygenated monoterpenes (1,8-cineole, fenchone, linalool, champhor), sesquiterpenes (trans- $\alpha$ -bergamotene- $\alpha$ -bulnesene,  $\gamma$ -cadinene).

#### **2.4.3.4. Insecticidal activities**

Basil seed extracts were found to show toxicity to the 5<sup>th</sup> instar larvae of potato tuber moth *Gnorimoschema operculella* (Pandey *et al.*, 1982), its stem and leaf extracts and oil were reported to be toxic to cotton aphid *Aphis gossypii* (Jacobson, 1975) and showed growth-inhibiting activity to cotton stainer, *Dysdercus cingulatus*. (Mohiuddin *et al.*, (1987) found its oil shows repellency to the red flour beetle *T. castaneum* under laboratory test, also Shaaya *et al.*, (1997) found its essential oil shows fumigant toxicity to saw-toothed grain beetle, *Oryzaephilus surinamensis*. Jacobson (1989) reported that its whole plant extract contained Juvocinine-I and Juvocinine-II, which were found to show juvenile hormone analogue mimic toxicity against milkweed bug, *Oncopeltus fasciatus*.

Methanol extract from *Ocimum basilicum* plant tested on 3<sup>rd</sup> instar larvae of *Spodoptera lituralis* was highly toxic with LC50 1.7  $\mu$ g/ml and the

(RGR) relative growth rate value was 3.75 mg/day which 80% lower than the control group (17.75mg /day), where the consumption diet treated with extracts from 0-6 decreased by 60% in comparison with the control (Pavela *et al.*, 2004), and the efficiency of *Ocimum sp.* Against garden pest has been recently reviewed (Quarles, 1999). Ibrahim (2003), showed that powder and water extracts of Rehan *Ocimum basilicum* have repellent and insecticidal activity against *B.incarnatus*, also *Ocimum* leaf extracts were evaluated by (Oladimeji and Kannike, 2010) against *Podagrica sp.* On okra and they found that at 5,10, and 20ml/l gave 12, 41, and 43% reduction in leaf damage. In addition, volatile oils of *O. basilicum* were found effective as repellent against cowpea beetle, *Callosobruchus aculates* Fab. (Boeke *et al.*, 2004).

The essential oil of *Ocimum basilicum* has been found to be effective as carbofuran (synthetic insecticide) in inhibiting the survival of *Paratylenchus brachycerus* in vitro and under green house, it has been suggested that essential oils could disrupt or change the permeability of the cell membrane of the nematode (Oka *et al.*, 2000).

Larvicidal and antifeedant activity of acetone, chloroform, ethyl acetate, hexane, and methanol extract of peel, leaf, and flower of *ocimum canum* and *ocimum sanctum* were used against the 4<sup>th</sup> instar larvae of *Helicoverpa armigera* at 1,000 ppm concentration for 24h of exposure. All extracts showed moderate larvicidal effects (Kathuria *et al.*, (2006). Inyang and Emosairue (2005) found good repulsion and antifeedant activity of aqueous solution of *O. gratissimum* L. leaves against banana weevil *Cosmopolites sordidus* Germar in Nigeria. Oladimeji and Kannik (2010) found that *O. basilicum* L. leaf extract was not phytotoxic.

Sathyaseelan and Bhaskaran (2010) found the highest repellency by *O. basilicum* L. (90.1%) leaf extract after 48 h of release of against *Maconellicoccus hirsutus* Green which was a major pest of mulberry

crop. Singh *et al.*, (2012) found that *O.bassilicum* have repellency with 91% against *Aphis gossypii*.

#### **2.4.4. *Lupinus termis*; Common name, termis; Family (Leguminaceae)**

##### **2.4.4.1. Botanical description:**

A plant of about 2 feet high, with leaves cut palmately into five or seven divisions, 1 to 2 inches long, smooth above, and white, hairy, beneath. The flowers are in terminal racemes, on short footstalks, white and rather large, the pod 3 to 4 inches long, flattish, containing three to six white, circular, flattened seeds, which have a bitter taste( Grieve, 2001).

##### **2.4.4.2. General uses:**

The bruised seeds of White Lupine, after soaking in water, are sometimes used as an external application to ulcers, etc., and internally are said to be anthelmintic, diuretic and emmenagogue.

##### **2.4.4.3. Chemical constituents:**

The bitter principle Lupinin is a glucoside occurring in yellowish needles. On boiling with dilute acids, it is decomposed into Lupigenin and a fermentable glucose ( Grieve, 2001).

##### **2.4.4.4. Insecticidal activities:**

In addition to its good nutritional properties, it has important functional characteristic related to its hypocholesterolemic (Sirtori *et.al.*,2004 , antioxidant (Tsaliki *et.al.*,1999) and antimicrobial properties (Lampartszczapa *et.al.*, 2003) . Mogahed (1997) studied the influence of petroleum ether extract of *Lupinus termis* on some biological aspects of *Spodoptera littoralis* under laboratory conditions the study revealed that the concentration of 1, 2, 3 and 4 petroleum ether extract of *Lupinus termis* had the following effects on the different stages of *Spodoptera littoralis* :

Ovicidal efficiency: all concentrations affect the egg viability and percent of hatchability was negatively correlated with the extract concentration. The percentages of hatchability were 53.6, 30.6, and 22.3

and 15.4% for 1, 2, 3 and 4% respectively compared with control 95.6%. LC50 value were 1.11. A significant differences in egg hatchability were noticed between control and other treatments, these result maybe due to the lipophilic properties of lupine extract which might facilitate its permeability through egg chorion inhibiting the embryonic development. Barakat *et al.*, (1984) found that acetone extract of lupine *Lupinus termis* is effective against *D. melanogaster* adults.

Mogahed results of 1<sup>st</sup>, 2<sup>nd</sup> and 3<sup>rd</sup> instars which are treated with lupine *Lupinus termis* extract had also the ability to decrease the mean period of larval duration. On the contrary, low concentration caused high decrease, where as this period decreased from 17.1 days in case of untreated larvae to 8.7, 8.8, 10 and 13.1 days for these treated with 1, 2, 3 and 4% of lupine extract respectively. It maybe due to the chemical constituents of *Lupinus termis*, the phloem of family legumiosae contains calcium oxalate in rhomboidal crystals, rosette crystals have been also observed in some members (Mohran, 1967).

#### **2.4.5. *Nicotiana rustica*: Common name, Tobacco; Family (Solanaceae)**

Alkaloids are characteristic of plants in the genus of solanaceae (Shmuck, *et al.*, 1941; Bush and Growe, 1989; Sisson and Severson, 1990). Nicotine is a most prevalent of these compounds, and it typically makes up > 95% of the total alkaloids in commercial flue-cured tobacco (Sisson, 1990). The insecticidal properties of nicotine have been known since at least 1690 (Schmeltz, 1971). Through the 18<sup>th</sup> century. Crude aqueous extracts or dust from tobacco was recommended for control of insect pests (Metcalf *et. Al.*, 1962).

##### **2.4.5.1 Botanical description:**

*Nicotiana rustica* is a wild plant in the solanaceae family, the plant is of about 2 feet high, with leaves cut palmately into five or seven divisions, 1 to 2 inches long, smooth above, and white, hairy, beneath. The flowers

are in terminal racemes, on short footstalks, white and rather large, the pod 3 to 4 inches long, flattish, containing three to six white, circular, flattened seeds, which have a bitter taste (Garner, 1946).

#### **2.4.5.2 General uses:**

Leaves are sedative, narcotic, emetic, antispasmodic and fish poison; used for the treatment of rheumatic swellings, skin diseases and scorpion-sting. A decoction of the leaf is used as a fomentation to the spine in tetanus; induce muscular relaxation, reduce strangulated hernia and dislocations. An ointment made by simmering the leaves in lard is used in curing old ulcers and painful tumours. The dried leaf is used as a styptic. Nicotine has been recommended in tetanus and as an antidote to strychnine. Since harmful effects of tobacco outweigh its therapeutic actions, its use as a drug is now discouraged (Ussuf *et al.*, 2001).

#### **2.4.5.3 Chemical constituents:**

Leaves contain alkaloid- nicotine, choline, anabasine and nor-nicotine. Leaves produce 6.6-8.8% nicotine when grown in greenhouse; while leaves of the same variety grown in the field yield 1.54-2.64%. Stem and root contains nicotine and anabasine (Chopra *et. al.*, 1992).

The primary active ingredient, nicotine, was isolated and named in 1928 (Schmeltz, 1971). Other pyridine alkaloids found in *Nicotiana sp.*, such as anabasine, anatabasine and nornicotine, also have insecticidal properties (metacalf *et al.*, 1962). Self *et al.*, (1964) reported that tobacco bollworm Larvae *Heliothis virescens* do not metabolize tobacco alkaloids but they are able to excrete compounds to some extent.

Plants protect themselves against insect attack by the use of different constitutive and induced defense metabolisms (Howe and Jander, 2008). Constitutive defense provides the plant with a basal defense level necessary to withstand first encounter during infestations. In addition, plants alter their gene expression profile during insect attack, resulting in



accumulation of newly synthesized biochemical compounds with insecticidal properties (Lawrence *et al.*,2006). The inducible defense response is mostly a combination of secondary metabolites, volatile and defense potent that can act directly on pest insects or make the plant less preferable to herbivores. Defense proteins such as proteinase inhibitors or amylase inhibitors distribute the normal enzyme activity in the insect mid gut after ingestion (Ussuf *et al.*, 2001; Franco *et al.*, 2002; Zavala and Baldwin, 2004). Other defense proteins are plant enzymes that reduce the nutritional quality of plants such as polyphenol oxidase (ppo) or enzymes that are detrimental towards pest insects, e.g. specific propeinase or threonine deaminase (Kang *et al.*2006, Mohan *et al.*, 2006; Thipyapong *et al.*;2007; Mahanil *et al.*, 2008). A third category of plant proteins involved in the complex mechanisms of plant defense are carbohydrate-binding proteins of lectins (Peumans and Van Damme, 1995; Vandenborre *et al.*, 2009) (Fig:5).

#### **2.4.5.4 Insecticidal activities**

Plant lectins are the insecticidal properties of many plants (Sharma *et al.*, 2004; Van Damme, 2008). Lectins have been shown to reduce the performance of several insect species belonging to the orders Lepidoptera, Coleoptera, Diptera and Hemiptera (Vandenborre *et al.* 2009).

Aqueous tobacco (*Nicotiana tabacum*, *N. glauca* or *N. rustica*) extracts containing the alkaloid nicotine have long been used to control crop insect pests (Schmeltz, 1971). Nicotine exerts its insecticidal effect by mimicking acetylcholine and interacting with nicotinic acetylcholine receptors (nAChRs), a major excitatory neurotransmitter in the insect CNS.

Unfortunately, nicotine is highly toxic to mammals and extreme care must be used since it is readily absorbed through the skin.

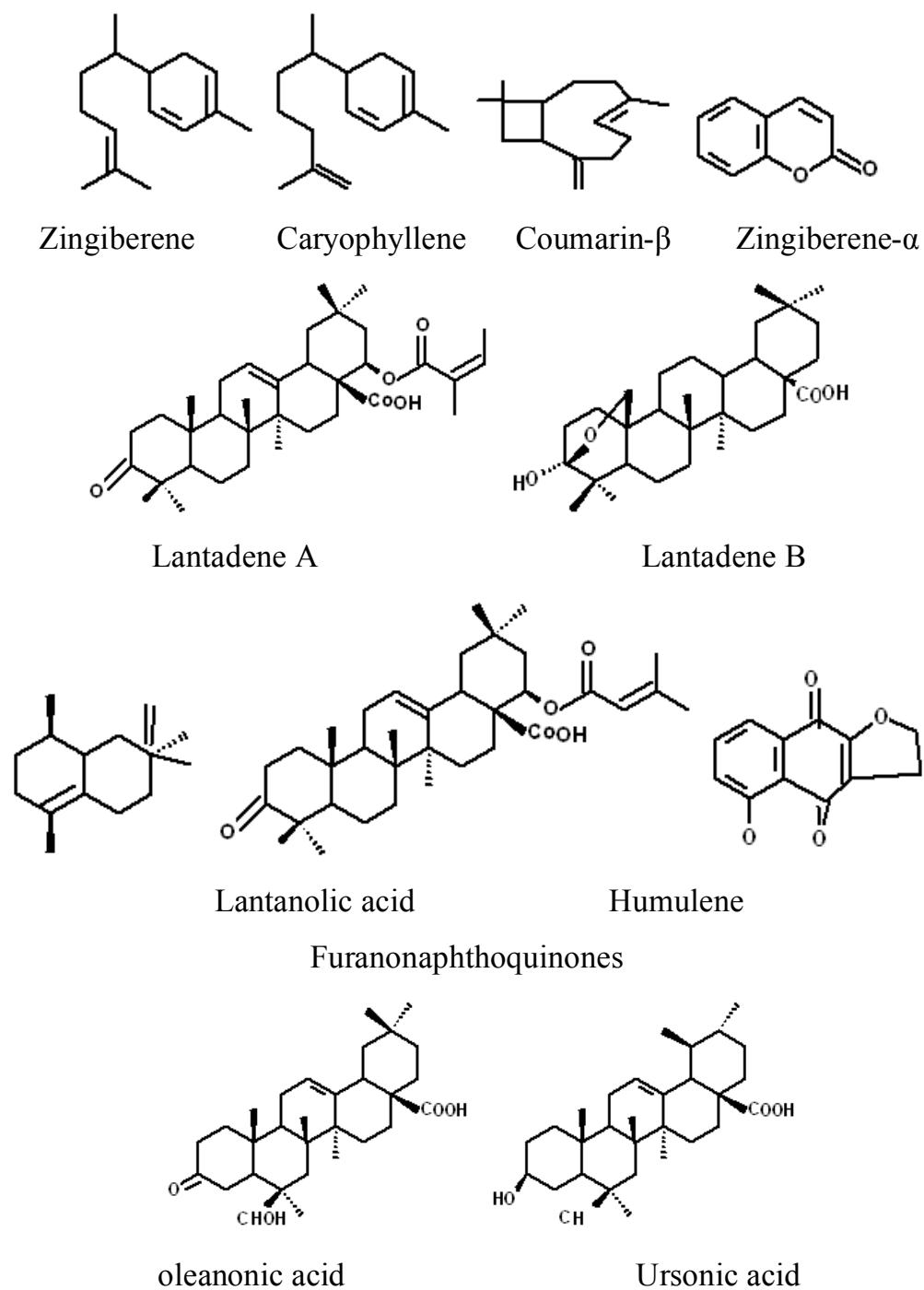
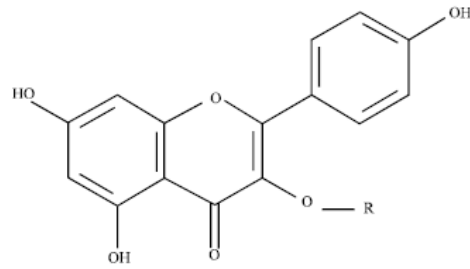
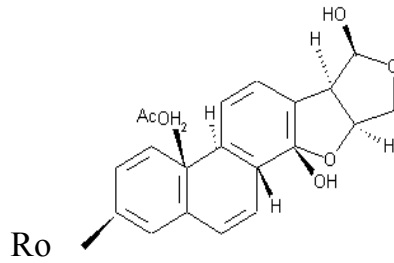


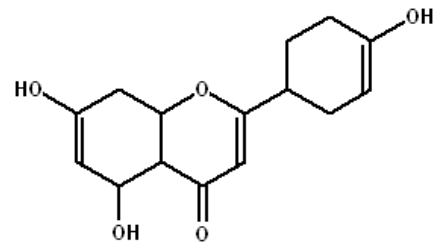
Fig (1) Chemical constituents of *lantana camara* (source: Ghisalberti, 2000)



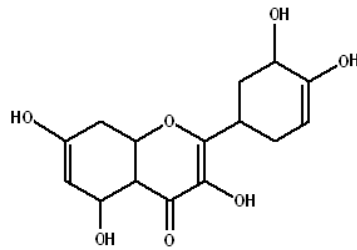
(S<sub>1</sub>) R = β-D-glucopyranosyl (1→2) β-D-xylopyranoside, (S<sub>3</sub>) R = α-L-arabinopyranosyl (1→2) β-D-galactopyranoside,



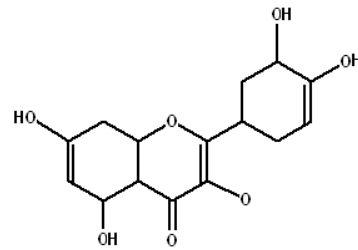
Argeloside



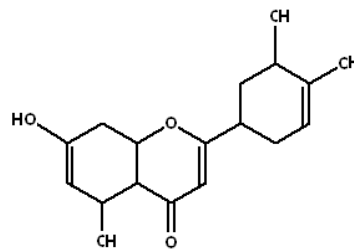
Kaempferol



Quercetin



Rutin



Luteolin

Fig (2) Chemical constituents of *Solenostemma argel* (source:Shafek and Michael, 2012)

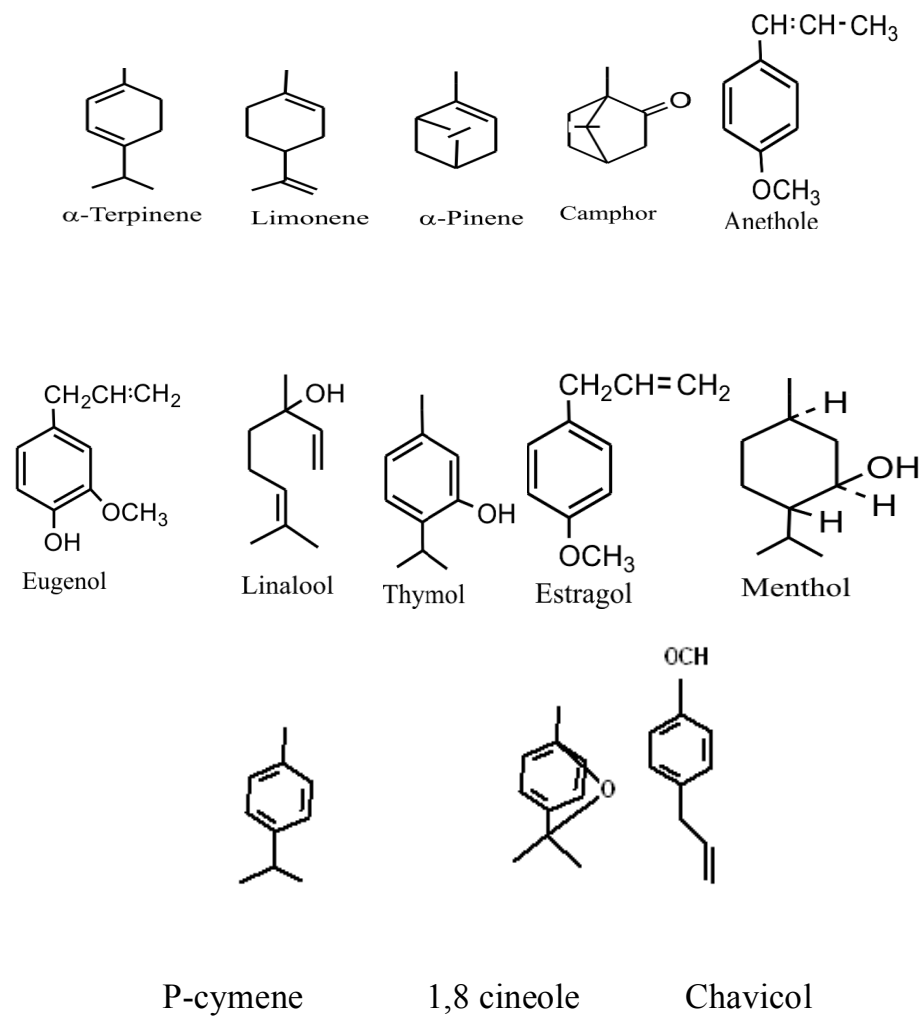


Fig (3) Chemical constituents of *Ocimum basilicum* (source: Koul *et al.*, 2008))

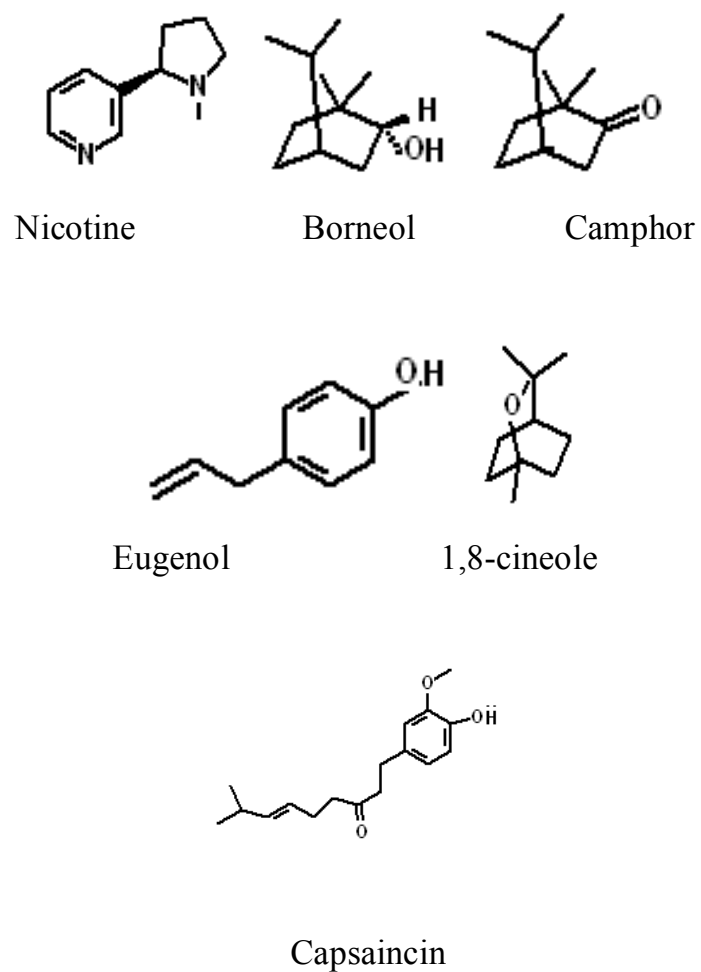


Fig (4) Chemical constituents of *Nicotiana rustica* (Source: John C., 2201)

## 2.5. Dimethoate:

Dimethoate recognized in the 1950s by American Cyanamid, is a widely used insecticide used to kill mites and insects systemically and on contact. It is used against a wide range of insects, including aphids, thrips, planthoppers and whiteflies on ornamental plants, alfalfa, apples, corn, cotton, grapefruit, grapes, lemons, melons, oranges, pears, pecans, safflower, sorghum, soybeans, tangerines, tobacco, tomatoes, watermelons, wheat and other vegetables. It is also used as a residual wall spray in farm buildings for house flies. Dimethoate has been administered to livestock for control of botflies. Dimethoate is available in aerosol spray, dust, emulsifiable concentrate, and ULV concentrate. Dimethoate is highly toxic to fish and to aquatic invertebrates (Hayes, 1990). Dimethoate is a colorless crystalline solid with a camphor-like (mercaptan) odor (Worthing, 1987). It will decompose rapidly when heated to temperatures above 80 degrees C, creating the possibility of explosion. It should never be heated above 35 degrees C. Thermal decomposition may release toxic and hazardous fumes of dimethylsulfide, methyl mercaptane, carbon monoxide, carbon dioxide, phosphorus pentoxide, nitrogen oxides (Meister, 1992).

Dimethoate structure (Fig: 5)

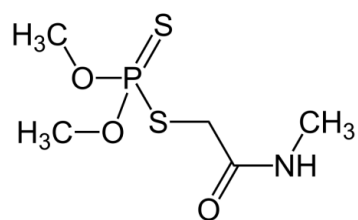


Fig ( 5) Dimethoate structure source (Sigma.2014)

# CHAPTER THREE

## MATERIALS AND METHODS

### 3.1. Study area

The study was carried out at the White Nile State ( Fig, 1 ). The study areas are demarcated by longitude 20-32E , latitude 13-39N and altitude 380 m . It has a saline cracking clay soil. The climate of the region is hot dry zone with rainfall during summer ( July-October). The mean annual rainfall ranges between 75 to 300mm, resulting from few moderate and heavy showers during July and August (Adam,2002) (Appendix:1)

### 3.2. Field studies:

During the course of study a number of regular surveys and general observations were conducted weekly to investigate the abundance of the pest African bollworm (*Helicoverpa armigera*) in the State. Larvae were randomly collected from host plants using hand picking (Plate, 6 ). The specimens were identified at insect collection units, at Gazira Research Station (Wad madani).

Three areas in the White Nile State namely Elsoufi, Eltawiela and Elmogmaat were selected for field study (Fig, 6). Four farms were selected randomly from each area.

From each area 4 farms were selected randomly. From each farm 25 plants were selected for counts of eggs and larvae on leaves, flowers and fruits were done weekly during period from first September to 20 January season (2012 – 2013) (Appendix, 2).

### 3.3.Laboratory studies:

Laboratory studies were carried out in Entomology Laboratory, Department of Crop Protection, Faculty of Agriculture and Natural Resources, University of Bakht – eruda .

#### 3.3.1 Insect rearing



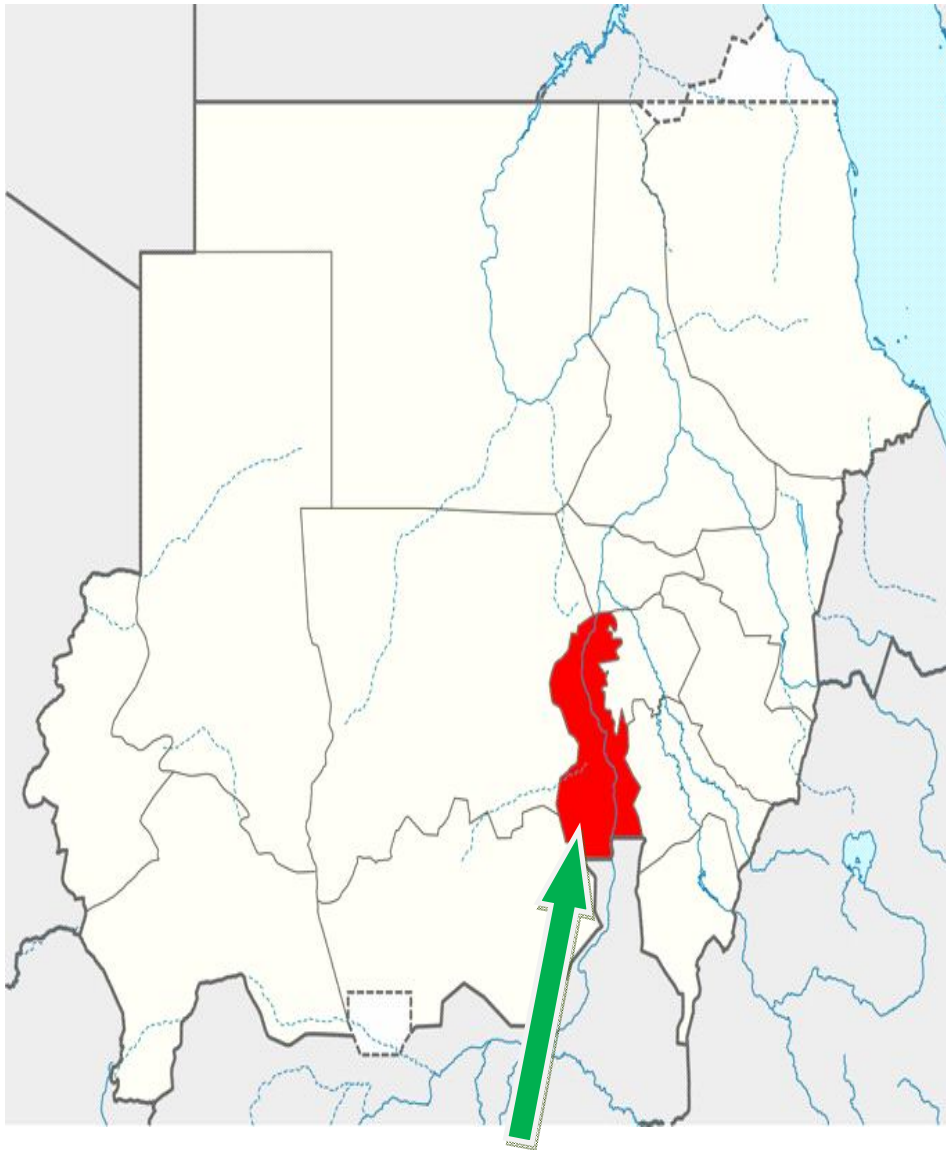
To maintain the culture of *H armigera*, the larvae were collected from tomato plants which were maintained in micro-plot (3 x 4 m) at the experimental field of Bakht er-uda University. Each larva was placed in a Petri dish (9 cm in diameter ) individually (to avoid cannibalism ) and was fed with tomato leaves . Before entering into pre-pupal and pupal stage, fully grown larvae were transferred into vials with small hole on their covers . One third of each vial was filled with moist sand, to provide optimal condition for pupation . When larvae were transformed into pupae, they were collected from sand and transferred individually to other glass vails for adult emergence . Emerged adults were paired according to the method suggested by Bhatt and Patel (2001) in glass jars covered with a fine muslin cloth . A 10% honey solution on a cotton swab was placed in each jar for moth feeding, then reared in jar till sufficient for conducting the experiment.

### **3.3.2 Morphometrics**

A total of ten eggs, ten larvae, ten pupae and ten adults (5 males + 5 females) were measured. The size of eggs was obtained with help of a binocular microscope, other life stages (larvae, pupa and adult) were measured via vernier caliper as well as simple scale.

### **3.3.3 Biology and lifecycle**

The developmental period of *H. armigera* was calculated by recording means of developmental time from ten eggs, ten larvae, ten pupae each in a separate petri dish and ten adults each in a separate glass vail covered by a muslin. Larvae were fed daily by fresh tomato slides while adults were provided with 10% honey solution. On the other hand, preoviposition, oviposition, post oviposition period , fecundity of female and hatchbilty of eggs were also recorded. The data collected on morphometrics, development and fecundity of *H. armigera* were subjected to analysis of standard error of means .

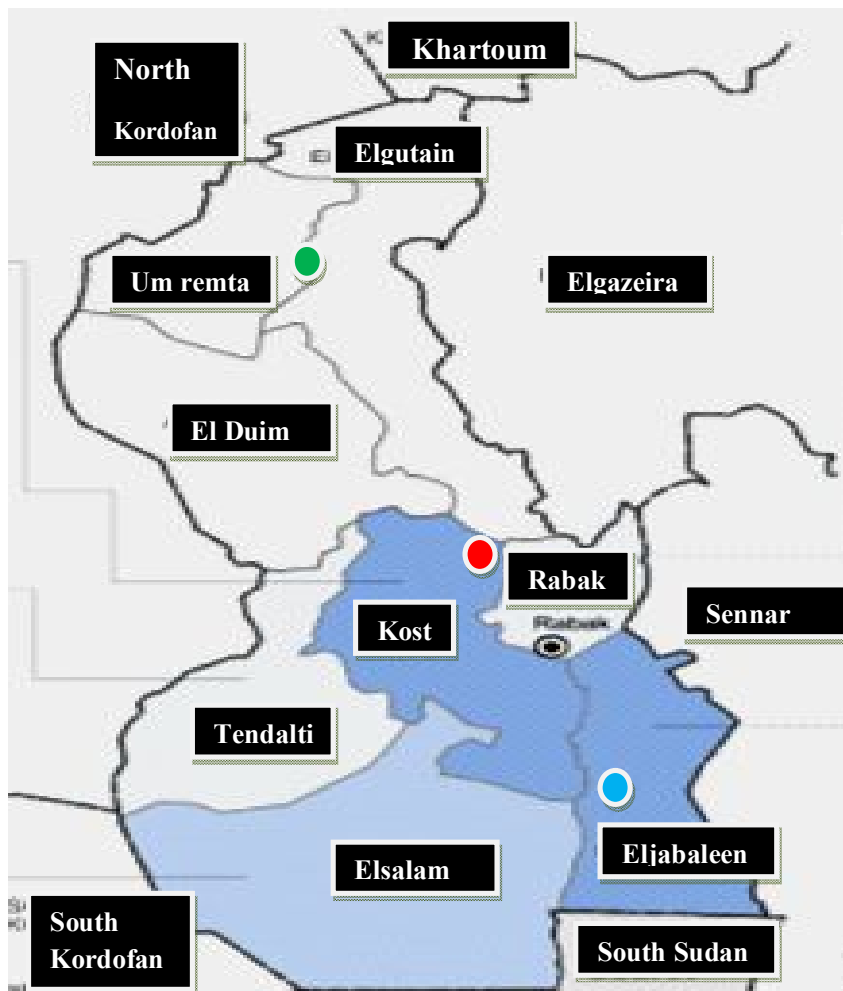


White Nile State

Fig (6) White Nile State (source) Google Map.



Plate (1) Larvae collected from experiment farm



Key: ● Elsuofi area  
 ● Eltawilla area  
 ● Elmogamaat area

Fig (7) Studied areas (source: Plant Protection Directorate, Kosti).

### **3.4 Plants extract preparation:**

Five plants (leaves and stems) namely Lantana ( *Lantana camara* : Verbenaceae) Plate 2, Hargal (*Solenostemma argel*: Asclepiadaceae) Plate 3 , Rehan (*Ocimum basilicum* : Lamiaceae) Plate 4 , Termis (*Lupinus termis* : Leguminaceae) plate5 , Tobacco ( *Nicotana rustica*: Solanaceae) Plate 6, were collected from the loacal area and the local market and then dried in the shade.



Plate (2) *Lantana camara* (Verbenaceae)



Plate (3) *Solenostemma argel* (Asclepiadaceae)



Plate (4) *Ocimum basilicum* (Lamiaceae)



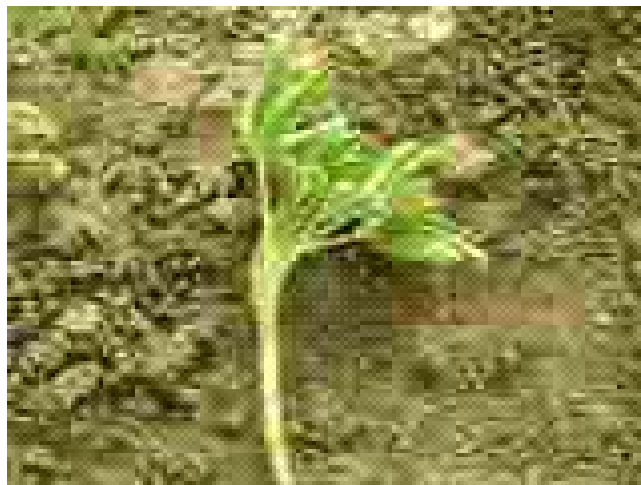


Plate (5) *Lupinus termis* (Leguminaceae)



Plate (6) *Nicotiana rustica* (Solanaceae)

### **3.4.1 Ethanolic extraction:**

Whole plant parts were collected from different areas and the local market, the collected parts were shade dried and then powdered by using electric blender. Extract was prepared according to the method of Visetson Milne (2004) by Soxhlet extractor in the ratio 1:10 (powder: solvent (Ethanol 96%) for 8 hours of extraction process. The plants extract were evaporated under room condition for 8 hrs to remove solvent then the crude extracts were kept in the refrigerator as stock solution (100%) until experiment commenced .

### **3.4.2 Aqueous extraction**

The required plant aqueous extracts were prepared according to the method suggested by Ascher (1981). Hence, an aqueous solution was prepared by adding 20 mg of the plant powder to 80 ml distilled water. The mixture was thoroughly shaken every 8hrs, for 10 min and was left for 24hrs . The mixture was then strained by a fine cloth and filtered by filter paper. The stock solution (20%w/v) was kept in the refrigerator until used for bioassay.

### **3.5 Mortality test**

The contact toxicities of aqueous and ethanolic extracts were evaluated according to the method of Brem *et al.* (2002) . Twelve (12) third instar larvae of *H. armigera* were introduced individually in separate petri dish ( to avoid cannibalism) used as a unit. The treatments were graded concentrations of ehtanolic extracts of selected plants stated above by using dipping method. Separate crude aqueous extract was applied as one rate 10% while ethanolic extracts were applied at four rates (2.5, 5, 10 and15%) . Ordinary water and Dimethoate (Rogor E40) ® at 2%v/v were included as negative and positive controls, respectively. Treatments were arranged in a complete randomize design CRD with three replicates per treatment. Fresh

tomato leaves were supplied every 24 h. The number of dead larvae (ND) was recorded 24, 48 and 72 h post-treatment. The actual percent larvae mortality was corrected for natural mortality using Abott (1925) formula.

$$\% \text{ corrected mortality} = \frac{\% \text{ treatment mortality} - \% \text{ control mortality} \times 100}{100 - \% \text{ control mortality}}$$

### 3.6 Probit analysis

The statistical analysis was done by MStat program (ANOVA) and probit analysis was carried out according to Finney (1964).

### 3.7 Antifeedant Test

In order to determine the amount of food consumed by larvae, starved larvae for 6 hours were used, leaves of the same weight (1gm) were immersed in test aqueous extracts 10% and ethanolic extracts at three rates (2.5, 5 and 10%) and Dimethoate (Rogor E40) ® at 2%v/v and control treatments for 30 minutes. Separately, treated leaves were removed using forceps, placed inside plastic Petri dishes (15 cm in diameter) lined with filter paper (Whatman No. 1) at the base according to Erturk (2006). Ten 2nd and 3rd instar larvae were introduced into each of the treated diets. Treatments were arranged in a complete randomized design with three replicates per treatment for 24hrs. Data on the amount of food consumed by larvae were recorded. The feeding ratio (FR) was calculated according to Owusu *et al.* (2000) formula:  $FR = FW/50$

Where FW remains food weight.

### 3.8. The absolute deterrence coefficient (DC)

The absolute deterrence coefficient (DC) was calculated by using

Kielczewski and Nawrot (1979) formula as follows:

$$\text{Deterrence Coefficient (DC)} = \frac{(C - T)}{(c + t)} \times (1000)$$

Where T represent the weight of food consumed by larvae in the experimental unit and C represent the weight of food consumed in the control unit.

### **3.9. Repellency test.**

Choice bioassay tests using 10 of 2<sup>nd</sup> and 3<sup>rd</sup> instar larvae were conducted in a circular flat bottomed plastic basin (45cm in diameter by 30 cm high) whose base was divided into four equal portions according the method described by Ogendo *et al.* (2004) . Separate aqueous and ethanolic crude extracts obtained from selected plants were evaluated against *H. armigera* larvae at one rate (10 %) and Dimethoate(Rogor E40) ® at 2%v/v was included as positive controls, treatments were arranged in a CRD with three replicates per treatment.

Alternate treated and untreated 10 tomato leaves were placed equidistant from the centre of the circular base. The top of the basin was covered with a nylon mesh to prevent the larvae from escaping. In each treatment, ten 2<sup>ed</sup> or 3<sup>rd</sup> instar larvae were released at the centre of the basin. The number of larvae that settled on the treated tomato leaves was recorded after 1 , 6 and 12 hrs of exposure . Percent repulsion (PR) was calculated according to the method that described by Talukder and Howse (1993) as follows

$$PR = 2 \times (C - 50)$$

C is the percent of larvae that settled on the untreated tomato leaves.

Trails showing a positive (+) PR value demonstrate repellency.

### **3.10. Phytochemical screening**

The phytochemical screening tests were carried out at Biochemistry Laboratory of Medicinal and Aromatic Plant Research (National Research Center). Screening was done for *Lantana camara*, *Ocimum basilicum* and *Solenostemma argel* according to the methods described by

Wall *et al.*, (1952), Harborne (1984), Sofowora (1993) and Martinez *et al.*,(1999).

### **3.10.1. Identification of tannins:**

0.2 g of each fraction of all plants extracts were dissolved in 10 ml of hot saline solution and divided in two test tubes. To one tube 2-3 drops of ferric chloride were added and to the other one 2-3 drops of gelatin salts reagent were added. The occurrence of blackish blue color in the first test tube and turbidity in the second one denotes the presence of tannins.

### **3.10.2. Test of sterols and triterpenes:**

0.2 g of each fraction was dissolved in 10 ml of chloroform. To 0.5ml of th solution, 0.5 ml acetic anhydride was added and then 3 drops of sulphuric acid at the bottom of the test tube. At the contact zone of the two layers, gradual appearance of green blue, pink to purple color was taken as an evidence of the presence of sterol (green to blue) and or triterpenes (pink to purple) in the sample.

### **3.10.3. Test for Alkaloids:**

0.5g of each fraction of each plant extract was dissolved in 2 ml of 2N Hcl in at water path and stirred while heating for 10 minutes. Then cooled and filtered . The filtered was divided in 2 test tubes. To one test tube few drops of Mayer's reagent was added while to the other tube few drops of Valsers reagent was added. A slight turbidity or heavy precipitate in either of the two test tubes was considered as presumptive evidence for the presence of alkaloids.

### **3.10.4. Test for Flavonoids:**

0.5 g of each fraction was dissolved in 30 ml of 80% ethanol and filtered. The filtered was tested for the presence of falvonoids as follows:  
A) 1ml of 1% aluminum chloride methanol solution was added to 3ml of filtrate in test tube. Formation of a yellow color indicated the presence of flavonoids.

B) 1ml of 1% of potassium hydroxide solution was added to 3 ml of of the filtrate in test tube. A dark yellow colour indicated the presence of flavonoids compounds (flavones or flavonenes) chalcone and flavonols.

C) 0.5 g of magnesium turnings were added to 2ml of the filtrate and 1ml of HCl . Apperance of defiant of color pink or red was taken as presumptive

evidence that flavonenes were present in the plant sample.

#### **3.10.5. Test for saponins:**

0.3 g each fraction was placed in a clean test tube. 10 ml of distilled water was added. The tube stoppered and vigorously shaken for about 30 seconds. The tube was then allowed to stand and observed for the formation of foam, which persisted for least an hour. Was taken as evidence for presence of saponins.

#### **3.10.6. Test for anthraquinone glycoside:**

0.2 g of each fraction was boiled with a solution of 9 ml of 0.5 N Koll and 1 ml 3% hydrogen peroxide. The mixture was extracted by shaking it with 10 ml of benzene . 5 ml of benzene solution was shaken with 3 ml of 10% ammonium hydroxide solution and the two layers were allowed to seprate. The presence of anthraquinones was indicated if the alkaline layer was found to have assumed pink or red color.

#### **3.10.7. Test for cyanogenic glycoside:**

0.2 g of each fraction was placed in Erlenmeyer flask and sufficient amount of water was added to moisten the sample, followed by 1 ml of chloroform ( to enhance every activity). A piece of freshly prepared sodium picrate paper was carefully inserted between a spilt crock which was used as stopper to the flask, a change in color of the sodium picrate paper from yellow to various shades of red was taken as an indication of the presence of cyanogenic glycoside.

# CHAPTER FOUR

## RESULTS

### **4.1 Field study results:**

The results of the field survey are shown in Table 1, Figs 8 and 9 and Appendix 2. These results showed that the African bollworm *Helicoverpa armigera* abundance varied with the season, time and place. The highest infestation during the season was recorded in December at Eltawiella and Elsoufi.

The early appearance of the pest was noticed in September at Elmogamaat area (southern of State) and the later appearance was noticed in November at Elsoufi area (northern of state).



Table:( 1) Number of eggs and larvae of *H.armigera*/montht

Month	Area					
	Elmogmaat		Eltawiella		Elsoufii	
	eggs	larvae	eggs	larvae	eggs	larvae
September	100	200	-	-	-	-
October	500	750	700	500	-	-
November	3700	1270	800	1000	200	100
December	100	400	700	200	600	1130
January	-	-	100	100	200	200

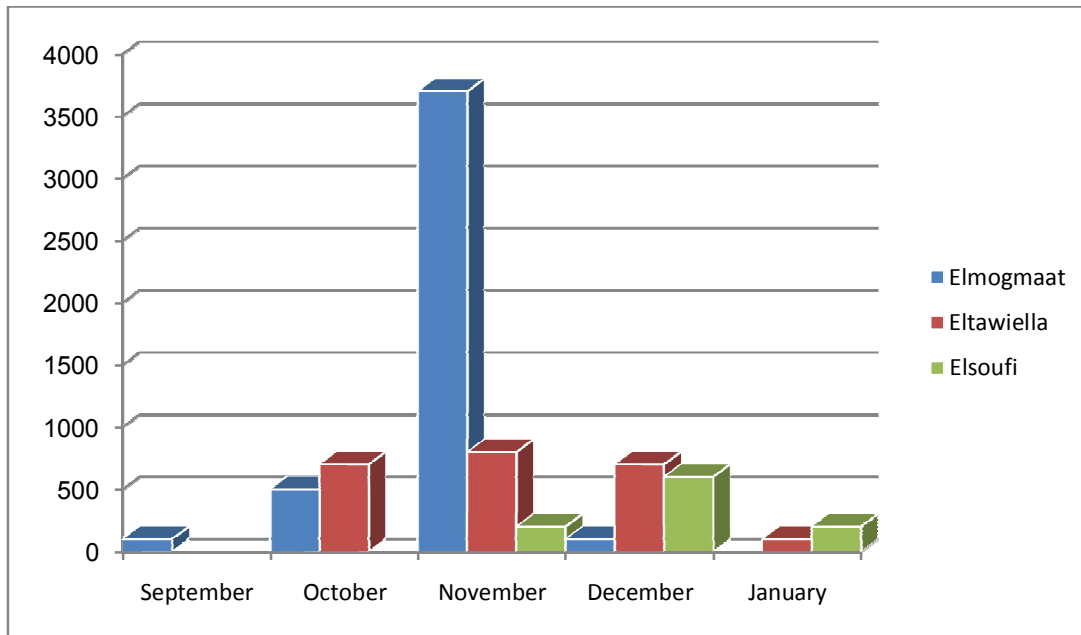


Fig: (9) Number of eggs of *H. armigera* /month

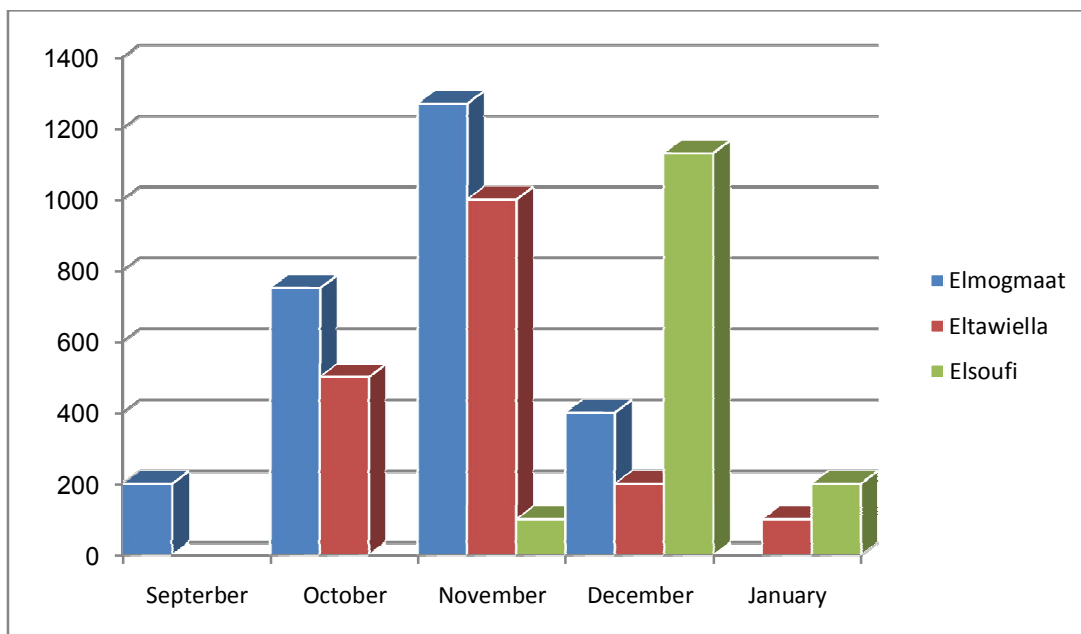


Fig: (8) Number of larvae *H. armigera* /month

## **4.2. Laboratory studies :**

### **4.2.1. Morphometrics measurements**

Results of these measurements are shown in Table 2 and in Plates 7 and 8 and Appendix 3. These results showed that, with an average range of temperature ( $27 \pm 1.1$  °C -  $30 \pm 2.9$  °C) and relative humidity ( $35.2 \pm 3\%$  -  $40.7 \pm 2.8\%$ ) morphometrics of insect stages are as follows:

The morphometrics measurements of length and width (mm) showed that the egg size was  $0.5 \pm 0.07$  and  $0.44 \pm 0.04$  mm yellowish white in colour.

The first instar was  $4.2 \pm 0.19$  and  $1.1 \pm 0.2$ , second instar was  $4.1 \pm 0.7$  and  $1.1 \pm 0.2$ , third instar was  $8.27 \pm 0.71$ , fourth instar was  $10.1 \pm 0.9$  and  $2.7 \pm 0.2$ , fifth instar was  $17.1 \pm 3.8$  and  $3.3 \pm 0.2$  mm in length and width respectively and the larvae exhibited variation in colours.

The pupa was  $18.6 \pm 1.4$  and  $4.9 \pm 0.33$  mm in length and width respectively with colour ranged from green to brown.

The adult male with wing span  $27.6 \pm 2.1$  mm and body length  $16 \pm 1$  mm, the female wing span  $35.8 \pm 2.3$  and body length mm  $19.2 \pm 2.4$ , adults colour ranged from grey to brown.

Table (2): Morphometrics measurements means of different stages of *H.armigera*

Stage	Width (mm)	Length (mm)
Egg	0.44±0.04	0.5±0.07
First instar	0.5±0.07	1.42±0.19
Second instar	1.1±0.2	4.1±0.7
Third instar	2.3±0.2	8.27±0.7
Fourth instar	2.7±0.2	10.1±0.9
Fifth instar	33.±0.2	17.1±3.8
Pupa	4.9±0.33	18.6±1.4
Male body	-	16±1
Female body	-	19.2±2.4
Male wing span	-	27.6±2.1
Female wing span	-	35.8±2.3

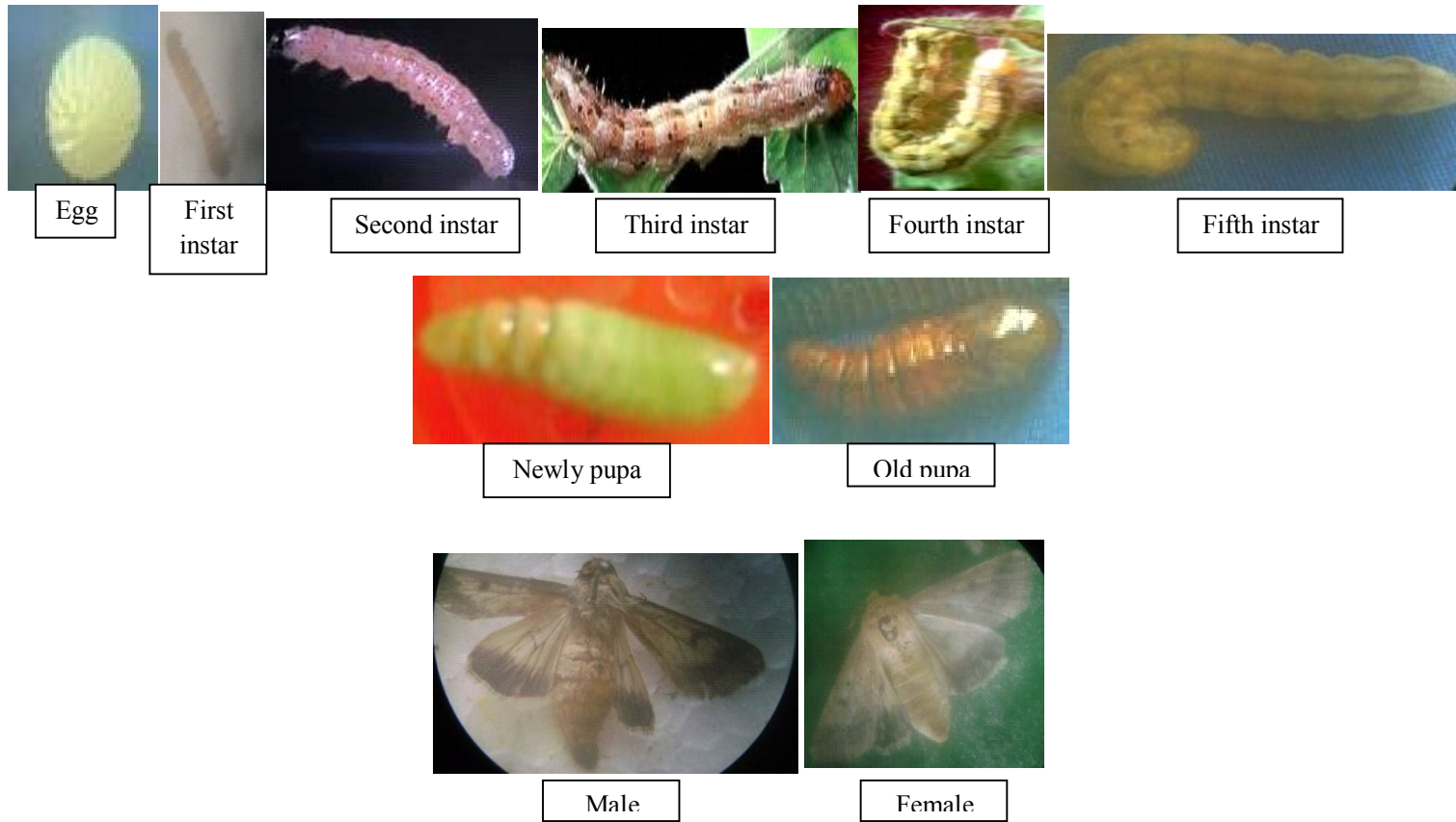


Plate (7) Life cycle of *Helicoverpa armigera* .



Pate (8) Different colours of *H.armigera* larvae

#### **4.2.2. Biology and lifecycle of African bollworm *Helicoverpa armigera*.**

Results are shown in Tables 3, 4, 5 and in appendices 4, 5, 6. These results revealed that the eggs are laid singly. The incubation period of eggs was  $3.9 \pm 0.9$  days. The larval periods were  $2.4 \pm 0.5$ ,  $2.7 \pm 0.8$ ,  $2.9 \pm 1.1$ ,  $3.2 \pm 0.9$  and  $3.4 \pm 0.5$  days for first, second, third, fourth and fifth instar respectively, prepupa and pupa periods were  $3.1 \pm 1.2$ ,  $9.6 \pm 1.5$  days respectively.

The adult male longevity was  $9 \pm 2.5$  days, while the adult female longevity was  $16.2 \pm 1.5$  days.

The percentage of eggs hatching, pupation and adult emergence were 66.9, 74.72 and 83.6% respectively.

Female have preoviposition, oviposition and postoviposition periods of  $2.2 \pm 0.44$ ,  $10.4 \pm 0.9$ ,  $3.2 \pm 0.44$  days respectively. The female insect laid about  $469.4 \pm 90.9$  eggs.

Table (3) Means of duration period of different stages of *H.armigera*.

Stage	Period (days)
Egg	3.9±0.9
First instar	2.4±0.5
Second instar	2.7±0.8
Third instar	2.9±1.1
Fourth instar	3.2±0.9
Fifth instar	3.4±0.5
Prepupa	3.1±1.2
Pupa	9.6±1.5
Male longefity	9±2.5
Female longefity	16.2±1.5
Lifespan	32.1

Table (4): Means of hatching, pupation and adult emergence of *Helicoverpa armiera*.

Number of eggs / mass	Number of eggs hatched	Haching %	Number of pupa formed	Pupation %	Number of adults emerged	Emergence %
24±6.9	16.4±6.2	66.9±7.1	12.4±5	74.6±4.6	10.4±4.6	83.1±5.8

Table (5): Means of fecundity of *Helicoverpa armiera*.

Preovipositon period (days)	Oivipositon period (days)	Eggs number	Postoivipositon period (days)
2.2±0.44	10.5±1	469.4±90.9	3.2±0.45



### **4.2.3 Effects of different botanical extracts on mortality of *H.armiera* 3<sup>rd</sup> instar larvae**

Various concentrations of ethanol and aqueous extracts of *Lantana camara*, *Solenostemma argel*, *Ocimum basilicum*, *Lupinus termis*, *Nicotian rustica* and Dimethoate (standard chemical) were tested as potential source of control agent for *H.armigera* larvae, the results of the effects are in Appendies 7, 8, 9, 10, 11, 12. These results can be discussed as follows:-

#### **4.2.3.1. Effects of different botanical extracts on mortality of *H.armiera* 3<sup>rd</sup> instar larvae (After 24 hours).**

All tested concentrations of *L.camar* and *S.argel* gave significantly high mortality% than the control.

All tested concentrations of *N.rustica* gave 0% mortality and were not different from the control.

Only the two highest concentrations of *L.termis* and *O.basilicum* were significantly high than the control, and effects were dose dependent.

All water extract of tested plants gave 0% mortality and were not different from the control (Table 6, Fig 10).

Table: (6) Effects of of different botanical extracts on mortality of 3<sup>rd</sup> larval instar of *H.armigera* (after 24 hours).

Ethanol extract con.	<i>L.camara</i>	<i>S.argel</i>	<i>O.basilicum</i>	<i>L.termis</i>	<i>N.rustica</i>
15	50 <sup>b</sup>	41.7 <sup>b</sup>	33.3 <sup>b</sup>	33.3 <sup>b</sup>	0 <sup>c</sup>
10	41.7 <sup>c</sup>	33.3 <sup>c</sup>	25 <sup>c</sup>	25 <sup>c</sup>	0 <sup>c</sup>
5	25 <sup>d</sup>	16.7 <sup>d</sup>	0 <sup>d</sup>	0 <sup>d</sup>	0 <sup>c</sup>
2.5	8.3 <sup>e</sup>	8.3 <sup>e</sup>	0 <sup>d</sup>	0 <sup>d</sup>	0 <sup>c</sup>
Water 10%	0 <sup>f</sup>	0 <sup>f</sup>	0 <sup>d</sup>	0 <sup>d</sup>	0 <sup>c</sup>
Dimethoate (1mm/L)	100 <sup>a</sup>	100 <sup>a</sup>	100 <sup>a</sup>	100 <sup>a</sup>	100 <sup>a</sup>
Control	0 <sup>f</sup>	0 <sup>f</sup>	0 <sup>d</sup>	0 <sup>d</sup>	0 <sup>c</sup>
C.V	12.73	19.14	10.75	21.4	.12.34
Lsd	18.16	18.35	18.69	18.74	19.9
SE	7,42	7.5	7.64	7.66	7.8

- Means having the same letters within the same column are not significantly different at P=0.05.

LSD = Least significant difference

C.V = Coefficient of variation

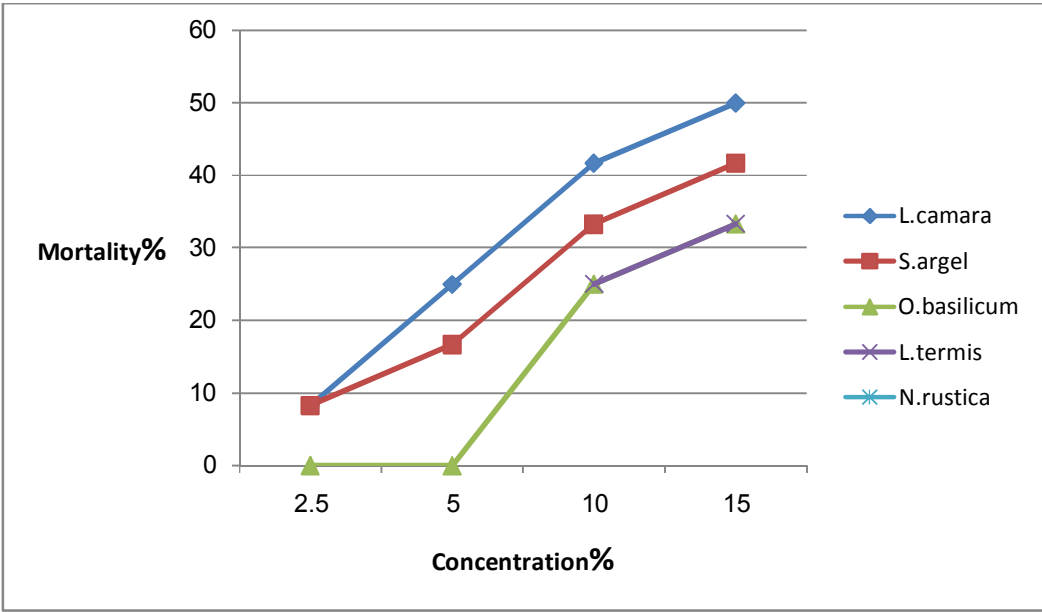


Fig (10) ) Effects of of different botanical extracts on mortality of 3<sup>rd</sup> larval instars of *H.armigera* (after 24 hours).

#### **4.2.3.2. Effects of different botanical extracts on mortality of *H.armiera* 3<sup>rd</sup> instar larvae (after 48 hours).**

All tested concentrations of *L.camar* gave significantly high mortality% than the control.

The two highest concentrations of *S.argel*, *O.basilicum* and *L.termis* were significantly high than the control.

Only the highest concentration of *N.rustica* gave significantly mortality% than the control, and effects were dose dependent.

All water extract of tested plants gave 0% mortality and were not different from the control (Table 7, Fig 11).

Table: (7) Effects of of different botanical extracts on mortality of 3<sup>rd</sup> larval instar of *H.armigera* (after 48 hours).

Ethanol extract con.	<i>L.camara</i>	<i>S.argel</i>	<i>O.basilicum</i>	<i>L.termis</i>	<i>N.rustica</i>
15	66.6 <sup>b</sup>	41.7 <sup>b</sup>	41.7 <sup>b</sup>	41.7 <sup>b</sup>	16.7 <sup>b</sup>
10	58.3 <sup>bc</sup>	41.7 <sup>b</sup>	33.3 <sup>b</sup>	25 <sup>c</sup>	0 <sup>c</sup>
5	50 <sup>c</sup>	33.3 <sup>c</sup>	8.3 <sup>c</sup>	0 <sup>d</sup>	0 <sup>c</sup>
2.5	25 <sup>d</sup>	16.7 <sup>d</sup>	0 <sup>c</sup>	0 <sup>d</sup>	0 <sup>c</sup>
Water 10%	0 <sup>e</sup>	0 <sup>e</sup>	0 <sup>c</sup>	0 <sup>d</sup>	0 <sup>c</sup>
Dimethoate	100 <sup>a</sup>	100 <sup>a</sup>	100 <sup>a</sup>	100 <sup>a</sup>	100 <sup>a</sup>
Water control	0 <sup>e</sup>	0 <sup>e</sup>	0 <sup>c</sup>	0 <sup>d</sup>	0 <sup>c</sup>
C.V	12.96	17.72	17.33	10.5	12.01
SE	7.16	7.38	7.58	7.63	7.77
Lsd	17.52	18.06	18.55	18.67	19.01

- Means having the same letters within the same column are not significantly different at P=0.05.

LSD = Least significant difference

C.V = Coefficient of variation

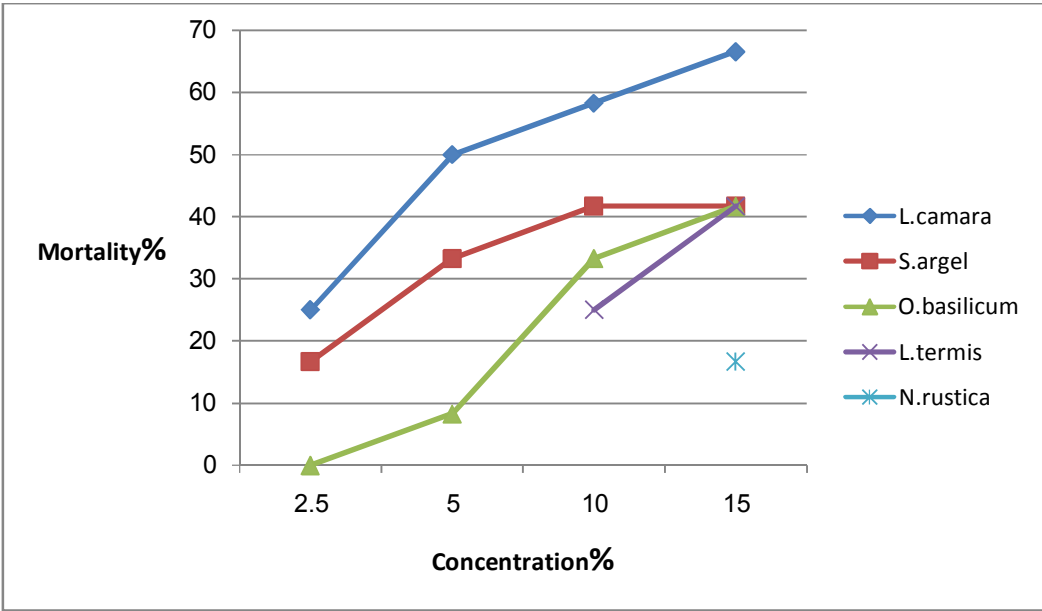


Fig (11) Effects of of different botanical extracts on mortality of 3<sup>rd</sup> larval instars of *H.armigera* (after 48 hours).

#### **4.2.3.3. Effects of different botanical extracts on mortality of *H.armiera* 3<sup>rd</sup> instar larvae(After 72 hours).**

All tested concentrations of *L.camar*, *S.argel* and *O.basilicum* gave significantly high mortality% than the control.

The three highest concentrations of *L.termis* were significantly high than the control.

The two highest concentration of *N.rustica* gave significantly mortality% than the control, and effects were dose dependent.

Only water extract of *L.camar* gave significantly mortality% than the control (Table 8, Fig 12).

Generally all extracts caused significant mortality compared to the control, and effects were dose and time dependent. The highest effects were noticed after 72 hours of exposure. The results indicated that *Lantana camara* caused distinguished symptoms of toxicity like shrinkage of body segments and green colour on prolegs (Plate: 9).

Table: (8) Effects of of different botanical extracts on mortality of 3<sup>rd</sup> larval instar of *H.armigera* (after 72 hours).

Ethanol extract con.	<i>S.argel</i>	<i>L.camara</i>	<i>O.basilicum</i>	<i>L.termis</i>	<i>N.rustica</i>
15	66.7 <sup>b</sup>	83.3 <sup>b</sup>	50 <sup>b</sup>	50 <sup>b</sup>	16.7 <sup>b</sup>
10	50 <sup>c</sup>	66.7 <sup>c</sup>	41.7 <sup>b</sup>	25 <sup>c</sup>	8.3 <sup>c</sup>
5	50 <sup>c</sup>	58.3 <sup>d</sup>	18.37 <sup>c</sup>	8.3 <sup>d</sup>	0 <sup>d</sup>
2.5	25 <sup>d</sup>	50 <sup>e</sup>	8.3 <sup>d</sup>	0 <sup>d</sup>	0 <sup>d</sup>
Water 10%	0 <sup>e</sup>	8.3 <sup>f</sup>	0 <sup>d</sup>	0 <sup>d</sup>	0 <sup>d</sup>
Dimethoate	100 <sup>a</sup>	100 <sup>a</sup>	100 <sup>a</sup>	100 <sup>a</sup>	100 <sup>a</sup>
water control	0 <sup>e</sup>	0 <sup>f</sup>	0 <sup>d</sup>	0 <sup>d</sup>	0 <sup>d</sup>
C.V	17.87	13.42	15.87	9.2	16.59
SE	7.23	7.02	7.46	7.58	7.74
Lsd	17.69	17.18	18.25	18.55	18.94

- Means having the same letters within the same column are not significantly different at P=0.05.

LSD = Least significant difference

C.V = Coefficient of variation



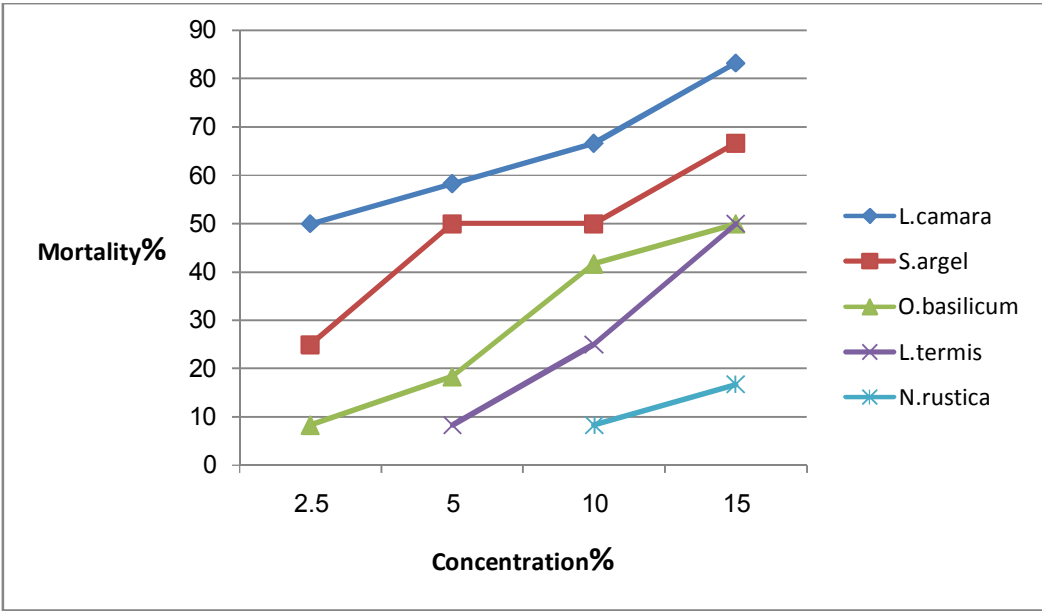


Fig (12) Effect of of different botanical extracts on mortality of 3<sup>rd</sup> larval instars of *H.armigera* (after 72 hours).



Plate (9) symptoms of *Lantana camara* toxicity

#### **4.2.4 Calculation of the lethal dose (LD<sub>50</sub>).**

As seen in table () *L.camara* was the most potent as showed by its low LC<sub>50</sub> 2.76 followed by *solenostemma argel* 6.35 , *Ocimum basilicum* 16.19 , *Lupinus termis* 17.05 and *Nicotiana rustica* 50 (Table 9, Fig 13).

Table: (9) LC<sub>50</sub> of of different botanical ethanol extracts on mortality of 3<sup>rd</sup> larval instar of *H.armigera* (after 72 hours).

	<i>L.camara</i>	<i>S.argel</i>	<i>O.basilicum</i>	<i>L.termis</i>	<i>N.rustica</i>
Slope	1.4	2.91	7.25	9.47	2.73
Chi-square	0.189	1.82	0.26	0.45	0.52
LC <sub>50</sub>	2.76	6.35	16.19	17.05	50.9
DF	4	4	4	4	4

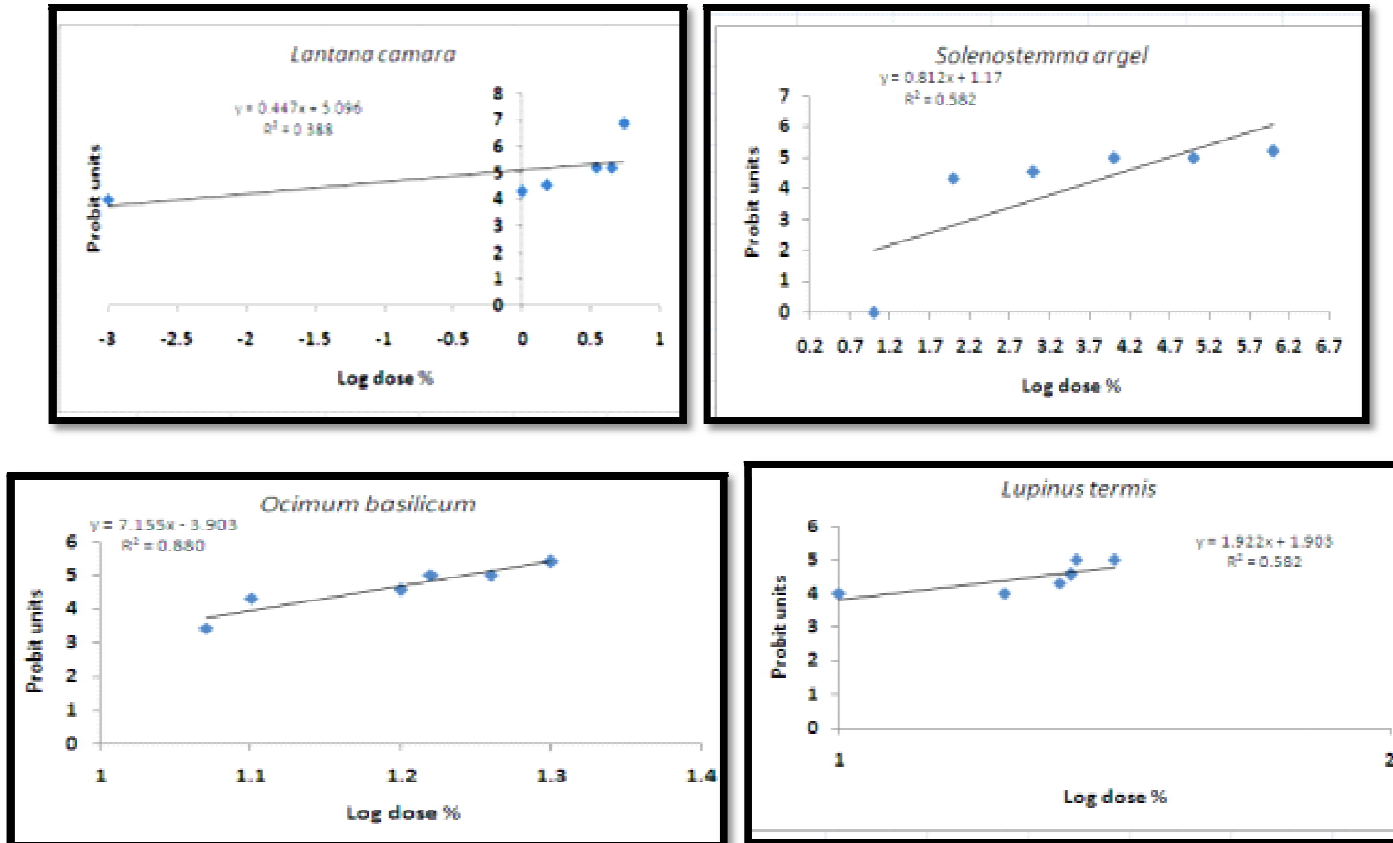
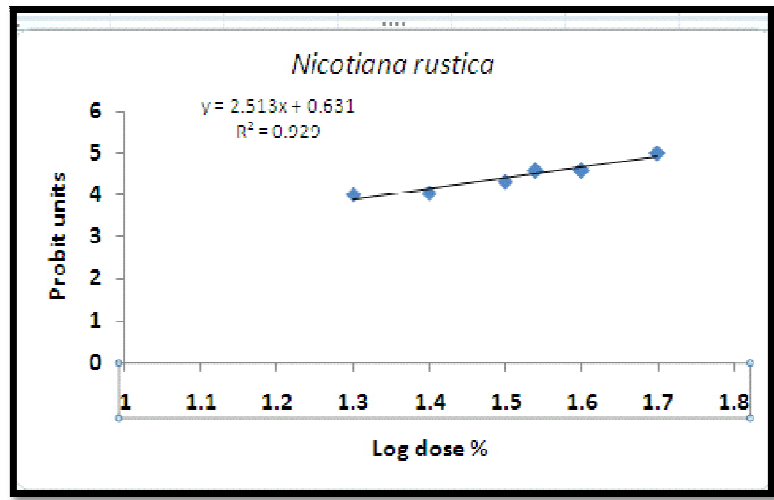


Fig (13) Log dose/ probit regression line of different botanical extracts on 3<sup>rd</sup> instar of *H. armigera* larvae



Fig(13) (Continued)

#### **4.2.5 Antifeedant effects of different botanical extracts on *H.armigera* 3<sup>rd</sup> instar larvae.**

Ethanol and aqueous extracts of *Lantana camara*, *Solenostemma argel*, *Ocimum baslicum*, *Lupinus termis* *Nicotian rustica* were tested as antifeedant agent for *Helicoverpa armigera* larvae. The results of these tests are shown in Table 10, 11, Figs 14, 15 and Appendix 8.

All extracts showed significant differences in Feeding Ratio except *L.termis* which showed no significant differences and the effects were dose dependent compared to the control. Generally *O.basilicum* was most potent as showed by its low feeding ratio 0.0054 and by its high food Deterrence Coefficient 557.3. The feeding ratio decreased with increase of concentration and the food Deterrence Coefficient increased with increase of concentration .

Table ; (10) Effects of different botanical extracts on feeding ratio(Fr) of 3<sup>rd</sup> larval instar of *H.armigera* (after 24 hours).

Feeding ratio for 24 hours

Feeding ratio (Fr)					
Con (%)	Lantana	Hargel	Rehan	Termis	Tobacco
Ethanol					
10	0.0084 <sup>c</sup>	0.0120 <sup>b</sup>	0.0054 <sup>c</sup>	0.0130 <sup>a</sup>	0.0130 <sup>b</sup>
5	0.0090 <sup>bc</sup>	0.0150 <sup>ab</sup>	0.0066 <sup>c</sup>	0.0142 <sup>a</sup>	0.0150 <sup>a</sup>
2.5	0.0110 <sup>bc</sup>	0.0176 <sup>ab</sup>	0.0070 <sup>c</sup>	0.0145 <sup>a</sup>	0.0172 <sup>a</sup>
Water 10%	0.0090 <sup>bc</sup>	0.012 <sup>b</sup>	0.0054 <sup>c</sup>	0.0160 <sup>a</sup>	0.0180 <sup>a</sup>
water	0.0190 <sup>a</sup>	0.0190 <sup>a</sup>	0.0190 <sup>a</sup>	0.0190 <sup>a</sup>	0.0190 <sup>a</sup>
control					
Dimethoate	0.0148 <sup>b</sup>	0.0148 <sup>ab</sup>	0.0148 <sup>b</sup>	0.0148 <sup>a</sup>	0.0148 <sup>a</sup>
LSD	0.042	0.07	0.039	0.013	0.026
C.V	14.6	19.54	15.78	3.52	18.2

- Means having the same letters within the same column are not significantly different at P=0.05.

LSD = Least significant difference

C.V = Coefficient of variation



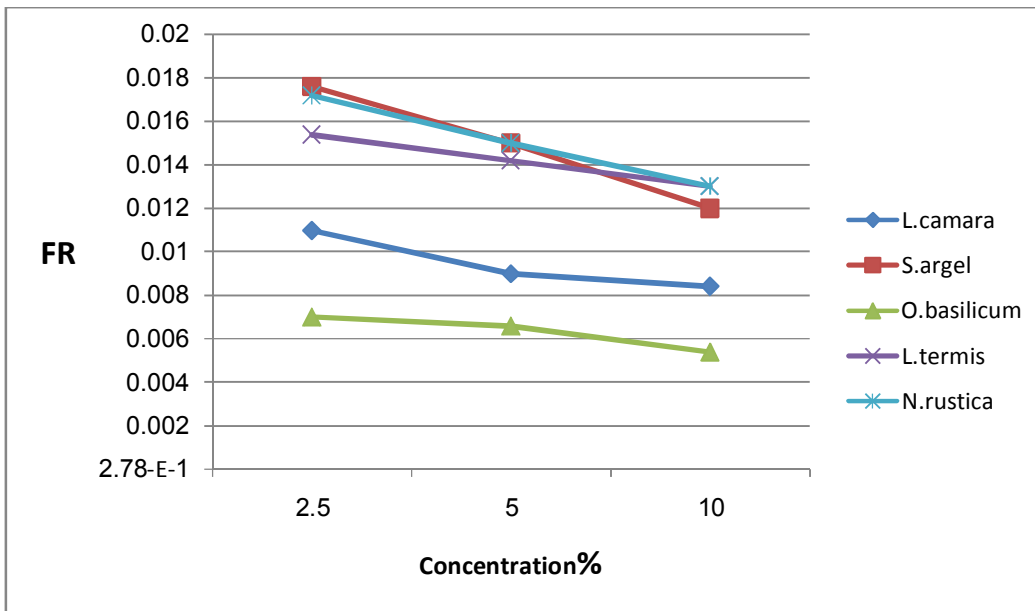


Fig (14) Effects of of different botanical extracts on feeding ratio of 3<sup>rd</sup> larval instar of *H.armigera* (after 24hours).

Table : (11) Effect of different botanical extracts on food deterrence (Absolute coefficient deterrence) on 3<sup>rd</sup> larval instar of *H.armigera* (after 24hours).

Tobacco	Termis	Rehan	Hargel	Lantana	Con (%) Ethanol
200	187.5	557..3	225.1	386.86	10
117.65	144.58	484.38	117.65	357.14	5
49.72	104.75	461.54	38,25	266.7	2.5
38.46	85.71	418.03	55.56	357.14	Water 10%
124.26	124.26	124.26	124.26	124.26	Dimethoate
0	0	0	0	0	control

- Means having the same letters within the same column are not significantly different at P=0.05.

LSD = Least significant difference

C.V = Coefficient of variation

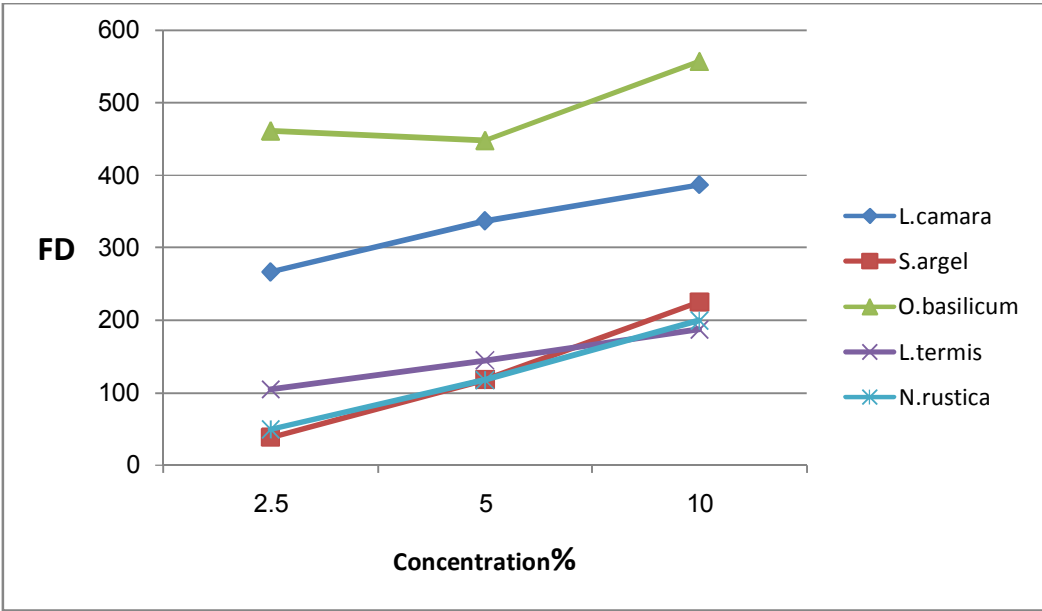


Fig (15) Effects of different botanical extracts on food deterrent of 3<sup>rd</sup> larval instar of *H.armigera* (after 24hours).

#### **4.2.6. Repellency effect of different botanical extracts on *H.armigera* 3<sup>rd</sup> instar larvae.**

In all extracts the highest percent repellence occurred in the first hour of exposure and the effects were time dependent. Generally *O.basilicum* was most effective showed by its high percent replusion 66.7 followed by *L.camara*. *Nicotiana rustica*, *L.termis* and *S.argel* were showed negative percent repellence results as repellent agents. Generally ethanol extracts were more effective (Table 12, Fig 16) than aqueous extracts (Table 13, Fig 17 and appendix 9).

Table (12) Effects of different botanical extracts (Ethanol 10%) on percent repellency of 3<sup>rd</sup> larval instar of *H.armigera*/hours.

Plant	Percent replusion /hours		
	1 hrs	6 hrs	12 hrs
<i>Ocimum basilicum</i>	66.7	40	- 6.6
<i>Solenostemma argel</i>	-26.6	- 33.4	- 33.4
<i>Lantana camara</i>	33.3	13.4	- 33.4
<i>Lupinus termis</i>	-13.4	-13.4	- 33.4
<i>Nicotiana rustica</i>	0	-40	- 46.7
Dimethoate	100	46.6	-

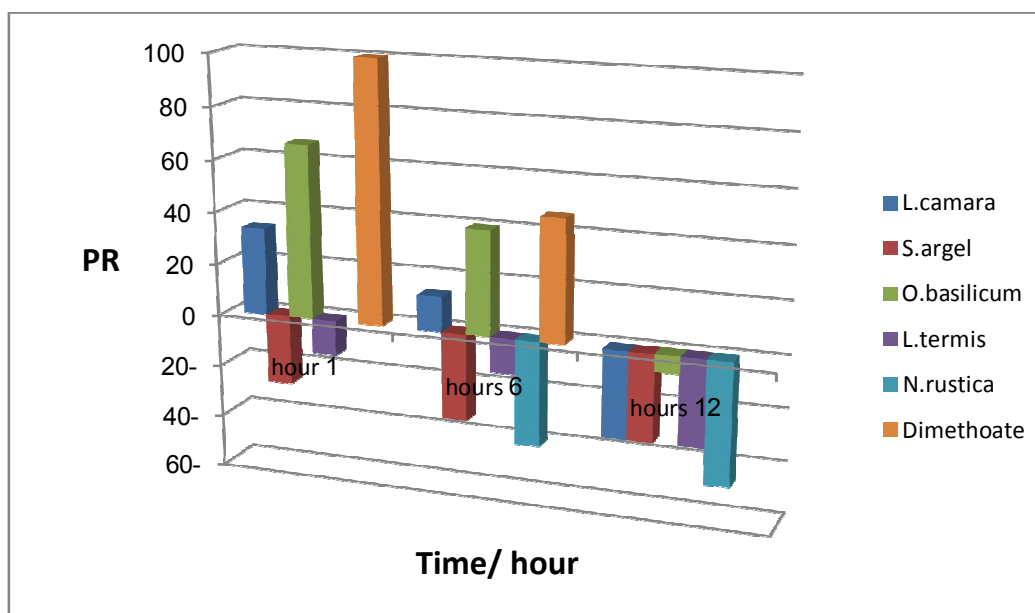


Fig (16): Effect of different botanical extracts (Ethanol 10%) on repellency of 3<sup>rd</sup> larval instar of *H.armigera* / hours.

Table (13) Effect of different botanical extracts (water 10%) on repellency of 3<sup>rd</sup> larval instar of *H.armigera*/hours.

plant	Percent replusion/hours		
	1 hrs	6 hrs	12 hrs
<i>Ocimum basilicum</i>	33.4	0	- 20
<i>Solenostemma argel</i>	-20	-26	- 33.4
<i>Lantana camara</i>	6.6	6.6	- 40
<i>Lupinus termis</i>	6.6	0	- 40
<i>Nicotiana rustica</i>	-6.6	-6	- 73.4

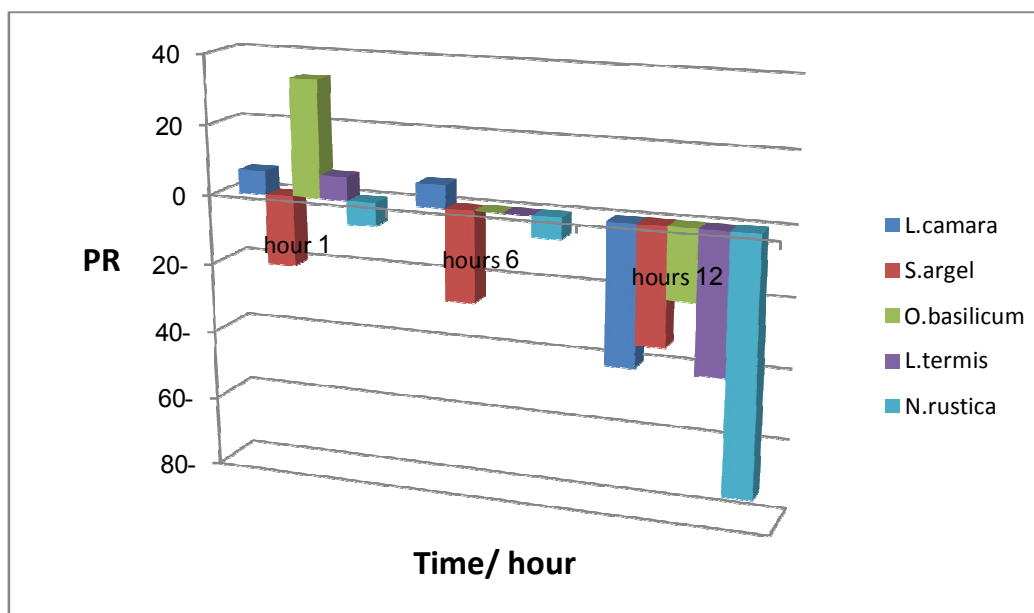


Fig (17) : Effect of different botanical extracts (water 10%) on repellency of 3<sup>rd</sup> larval instar of *H.armigera* / hours

#### **4.7. Phytochemical screening of the most potent plant extracts**

Table (14) exhibited phytochemical screening of the most potent plants *L.camara*, *O.basilicum* and *S.argel*. Tests showed the presence of alkaloids (Plate: 10-A) sterols and triterpenes (Plate: 10-B) , flavonoids (Plate: 10-C), tannins (Plate: 10-D), saponins (Plate: 10-E) and in addition to presence of cyanogenic glycosides in *S.argel* (Plate: 10-F).

Table: ( 14 ) **Phytochemical screening of the most potent plants.**

observations	Result			Test
	Hargel	Lantana	Rehan	
turbidity	+	+	+	Alkaloids
green to blue color	+	+	+	Sterols
Pink to purple color	+	+	+	Triterpenes
Yellow color	+	+	+	Flavonoids
-	-	-	-	Athraquenones
Foam formation	+	+	+	Saponins
ULV adsorbtion	+	+	+	Cumarins
Blackish green color	+	+	+	Tannins
Chage of yellow color of sodium picrate paper to red	+	-	-	Cyanogenic glycosides

**Key:**

+ Present

- not present





Plate (10-A ): Appearance of slight turbidity , which indicates the presences of Alkaloids.

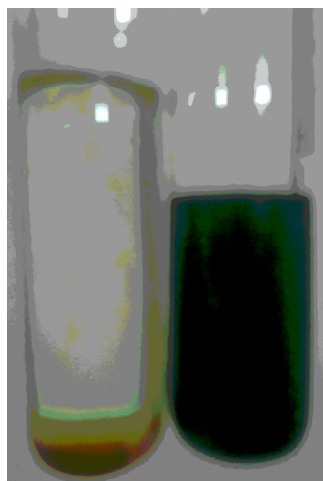


Plate (10-B ): Appearance of purple and green colour, which indicates the presences of Sterols and triterpens.

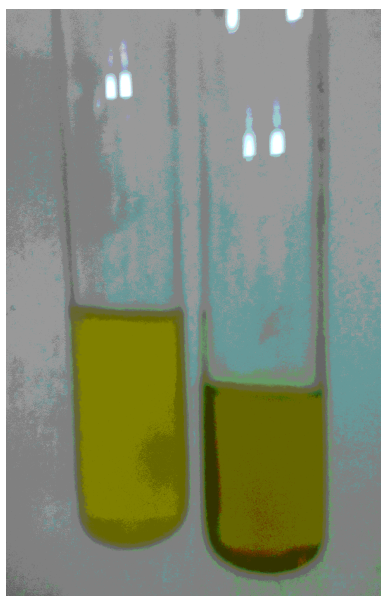


Plate ( 10-C ): Appearance of yellow colour, which indicates the presences of flavonoids.

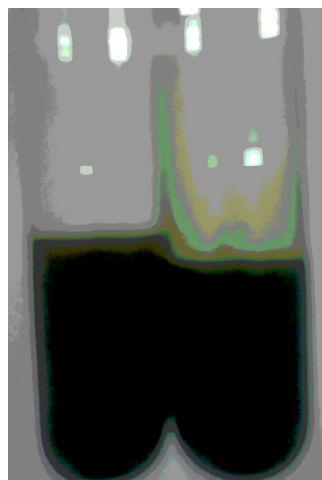


Plate ( 10-D ): Appearance of Black green colour, which indicates the presences of Taninns.



Plate ( 10-E ): Appearance of foam, which indicates the presences of saponins

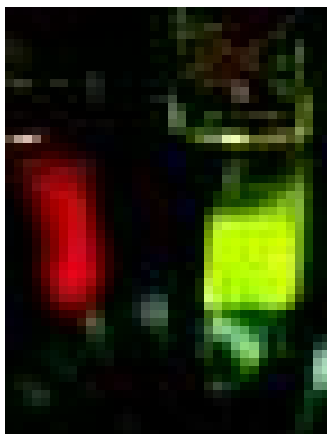


Plate (10 -F ): Appearance of red color on sodium picrate paper, which indicates the presences of Cyanogenic glycosides

# CHAPTER FIVE

## DISCUSSION

The control of African bollworm *H.armiera* mostly depends on chemical sprays, but *H.armiera* has developed resistance to many insecticides. In addition, the use of chemical pesticides have many problems such as negative impacts on nontarget organisms including man and the environment (Isman, 2006) high toxicity, non biodegradable residues in soil, water resources and crops that affect human health (Koul, 2008). These problems have been the matter of concern for both scientists and public.

Thus. one needs to search the new highly selective, biodegradable and environmental friendly pesticides to solve these problems by developing natural products pesticides.

Among the natural products, plants which derived pesticides are more acceptable . This acceptance is due to their abundance, nature friendly, least toxic to natural enemies, their effect on limited species, fast degradation and low phytotoxicity and low toxicity to vertebrates (Krim *et al.*, 2003), biodegradable to non toxic products (Hashem and Youssef, 1991) and less persistence in the environment (Isman, 2006).

Plants which produce secondary metabolism such as alkaloids, terpenoids, flavonoids, phenols, glycosides and tannins , which were known to protect plants from the attack of insect pests (Ahmed, 2007). These compounds act as toxicant, growth regulators (Bower *et al.*, 1972), repellent anifeedant and growth inhibitors (Odeyemi *et al.*, 2008).

Based on the above there is a necessary need for active and safe alternatives to use for the control of *H.armigera* . This active search was focused on the evaluation of efficacy of natural products especially botanical extracts against *H.armigera*.

Many reports about the toxicity of several extracts from different plant species belonging to various families are used for controlling *H.armiera*, of which neem (chopar *et al.*, 1994), *Acorus calamus*, *Annona squanosa* and *Vitex negunado* (Murugan *et al.*, 1998).

Control of any pest needs to some ideas about the morphology characteristics and biology activities of the different stages of target pest and general survey to the study area.

In this present study, general survey was done at three different sites of the White Nile State to count the number of eggs and larvae of *H.armigera*. This is followed by some morphological description and some biological activities. Five local plants namely Lantana, Hargal,Rehan, Termis and Tobacco were used to investigate their insecticidal activities against *H.armigera* a polyphagous pest of several crop plants under laboratory conditions.

General survey, some morphological description, some biological activities, mortality of 3<sup>rd</sup> instars caused by topical method application, antifeedant and repellency were applied under the following sub-titles.

### **Field study**

The field studies carried out in the study areas showed that the African bollworm *Helicoverpa armigera* was present along the whole State. This result agree with both Joyce (1951) and Schumetterer (1969) who confirmed the presence of *H.armigera* in Shambat, Nuba mountains, Toker Delta, Gedarif, White Nile and Gezera. The time of insect incidence varied gradually from south of the state (Elmogamaat area) in September, Eltwiella (middle of the state) in October and to the north area ( Elsoufi) in November. This may be referred to the variations in sowing date or to the raining time. The high infestation occurred during the winter season in December. This may be due to the optimum temperature, abundance of

host plants and early application of insecticides for other pests which kill natural enemies of the insect.

### **Morphometrics measurements**

Morphometrics data showed that the length and width of eggs, larval instar and the adult male and female body length and wing span are similar to that reported by Ali *et al.*, (2009) who found that the size of eggs varied from 0.42 – 0.6 mm in length and 0.4 – 0.55 mm in width. First, second, third, fourth and fifth instars length and width are (1.40 ± 0.06 and 0.45 ± 0.01 mm; 3.88 ± 0.11 and 0.75 ± 0.01 mm, 7.90 ± 0.19 and 2.28 ± 0.04 mm; 12.83 ± 0.45 and 2.85 ± 0.04 mm; 20.97 ± 0.61 and 3.25 ± 0.04 mm) respectively. The adult ( male and female) body length and wingspan were 17.65 ± 0.18 and 34.73 ± 0.59 mm; 20.08 ± 0.38 and 40.93 ± 0.55 mm, respectively.

### **Biology and lifecycle**

The biological study revealed that the insect larval development passed through 5 larval instars. This result agrees with Twine (1978) and Fowler and Lakin, (2001) who reported that the larvae take between 5 to 7 instars depending on host plant, temperature and other factors. The larval instars exhibited variation in colours. This result is similar to that reported by King (1994) who wrote larvae may vary in colour from light green to brown to black. The duration periods of insect stages (eggs, larvae, prepupae and pupae) are in line of periods represented by Ali *et al.*, (2009) who mentioned that the incubation period of eggs was 3.37 ± 0.09 days, the average duration of first, second, third, fourth and fifth instars were 2.27 ± 0.08, 2.42 ± 0.08, 2.67 ± 0.07, 2.83 ± 0.07 and 3.40 ± 0.10 respectively, prepupae lasted 2.15 ± 0.15 days and pupae took time from 10 -14 days. The life span of male and female result indicated that female

lived longer than male, this finding agrees with King (1994) and Akashe *et al.*, (1997) who reported females live for 2 -3 days longer than males .

The preoviposition and postoviposition periods are similar to that stated by Butler *et al.*, (1996), Jallow *et al.*, (1998) and Ali *et al.*, (2009) who stated that preoviposition was  $2.45 \pm 0.08$  days and postoviposition was  $2 \pm 0.05$  days. Oviposition period, eggs hatching%, pupation% and adult emergence% differed from previous studies. This may be due to the variations in time, food and place of experiments.

### **Effects of different botanical extracts**

The results of the mortality data indicated that *Lantana camara* ethanol extract was most potent against *Helicoverpa armigera* 3<sup>rd</sup> larval instars as showed by its low LC<sub>50</sub> 2.76 and its mortality percent of 83.3%. These findings agree with that reported by Dharmagadda *et al.*, (2005) who found that 200 pp oil of lantana produced 100% mortality in larvae of *C.quinquefasciatus* in 15 minutes, Kathuria (2006) who reported that lantana extract caused 56% inhibition on 3<sup>rd</sup> instars of *H.armigera* and Arti *et al.*, (2009) who reported that Lantana extracts caused significant mortality rate on 4<sup>th</sup> instars of *H.armigera*. Lantana also caused morphological changes in larval instars and pupae. These results agree with Arti and Prasad and Purohit (2009) who observed that sluggishness, cessation of feeding, green colour in leg region and overall shrinkage occurred . The high effects may be due to the presence of Lantadene A and B in the chemical constituents of Lantana which were believed to be responsible for most biotic activities of *lantana camara* as reported by Barre *etal.*, (1997). Morphological changes may be caused by the presence of tannins which were reported by Alock and Chan (1982) to cause growth inhibition of lepidopterous spp.

Generally the mortality was dose and time dependent, the significant mortality was shown at all concentrations of ethanol extract. A high

mortality rate (83.3%) was observed in 15% concentration after 72 hours of exposure, while (2.5%) concentration showed the least mortality rate (8.3%) after 24 hours of exposure .

The published data reported that Lantana have been used against different insect species, Ogendo *et al.*, (2003) used it against *Sitophilus zeamaze* (Coleoptera) and found that it caused 82.7-90% mortality. In other study Sharma *et al.*, (1990) reported that it was toxic to *Aphis rumicis* (Homoptera), and showed insecticidal properties against *Dysdercus similis* (Hetroptera). Also lantana showed growth regulating activity on *Dysdercus koengii* a pest of cotton (Suryakala et al., (2007) . Abdel-Hady *et al.*, (2005) reported that *lantana camara* revealed insecticidal activity against 3<sup>rd</sup> instars larveae of *Musca domestica* (Diptera).

*Solenostemma argel* mortality effects results obtained in this study are in agreement with that reported by Abd El-Aziz and Ezz El-Din (2007) who found that *S.argel* caused 45.25% mortality on *Spodoptera littoralis*. The effects were attributed to the presence of a variety of bioactive substances mainly terpens, pergenine, glycosides, alkaloids and sterols as indicated in the present results of phytochemical screening. These results are in line with that reported by Al-Dgharri *et al.*, (2004) who stated that the bioactive effects of *S.argel* is due to the presence of terpens, pergenine, glycosides, alkaloids and sterols . For instance flavonoides were reported to be toxic to some insects by (Salama *et al.*, 1997) .

In other studies Dgharri *et al.*, (2007) found that Hargal caused 20% mortality percent on *S.littoralis* . El-Tayeb et al., (2009) represented that Hargal extract LD 50=0.006 ml/l against *Culex quinque fasciatus*. Sidahmed *et al.*, reported its termicidal effects on cotton soil termite (*Microtermis thoracalis*).



Generally the mortality was dose and time dependent, the significant mortality was shown at all concentrations of ethanol extract. A high mortality rate (66.7%) was observed in 15% concentration after 72 hours of exposure, while (2.5%) concentration showed the least mortality rate (8.3%) after 24 hours of exposure .

*Ocimum basilicum* caused moderate mortality effects, these results agree with that reported by Kathuria *et al.*, (2006) who found that acetone, chlorophorm, hexane and methane extracts of *O.canum* and *O.sanactum* induced moderate larvicidal effects on 4<sup>th</sup> instars larvae of *H.armigera* . These results can be explained by the fact that *O.basilicum* contains compounds reported to possess insecticidal activities such as eugenol (Ngho *et al.*, 1998), thymol (Hummelbrunner and Isman (2001), anethole and 1.8 – cineole (Lee *et al.*, 1997).

Generally the mortality was dose and time dependent, the significant mortality was shown at all concentrations of ethanol extract. A high mortality rate (50%) was observed in 15% concentration after 72 hours of exposure, while (2.5% and 5%) concentration showed no significant mortality.

In other studies *O.basilicum* was found to show toxicity to 5<sup>th</sup> instar larvae of potato tuber moth (Pandey *et al.*,1982) and to *Aphis gossyii* (Jacobson, 1975) and showed inhibition activity to *Dysdercus cingulatus*(Jacobson, 1975).

The mortality effects caused by *L.termis* agree with that stated by Barakat *et al.*, (1984) who reported *L.termis* extracts is effective on *D.melanogastra* adults. This may be related to its hypocholestrolemic characteristic and antioxidant ( *Tsaliki etal.*, 1999). In other studies Termis was found to cause reduction in eggs hatchability of *S.littoralis* and decrease the mean period of larval duration (Mogahed,1997).

Generally the mortality was dose and time dependent, the significant mortality was shown at 10% and 15% concentrations of ethanol extract. A high mortality rate (50%) was observed in 15% concentration after 72 hours of exposure, while 2.5% and 5% concentration showed no significant mortality.

*Nicotiana rustica* mortality effects agree with Metacalf *et al.*, (1962) who reported crude extracts from Tobacco recommended for control of insect pests. This may be due to the presence of alkaloids such as nicotine, anabasine, anatabasine and nornicotine which have insecticidal properties.

Generally the mortality was dose and time dependent, the significant mortality was shown at 10% and 15% concentrations of ethanol extract. A high mortality rate (16.7) was observed at 15% concentration after 72 hours of exposure, while 2.5% and 5% concentration showed no significant mortality.

### **Antifeedant effects**

The antifeedant results showed that *Ocimum basilicum* caused high effects (least FR) and this agrees with Pavella and Skaya (2004) who found *O.basilicum* decreased the consumption diet of 3<sup>rd</sup> instars of *H.armiera* by 60% in comparison with control. This high effects may be due to *O.basilicum* contain eugenol as major compound

The feeding ratio was dose dependent. The significant FR was shown at all treatments (Ethanol and water extracts). The least FR (0.0054) was observed at 10% ethanol extract, while 2.5% showed the highest FR (0.007). In other study Oladimeji and Kannike evaluated Rehan leaf extracts against *Podagrica* sp. on okra and they found that at 5, 10 and 20ml/L gave 12, 41 and 45% reduction in leaf damage.

*Lantana camara* antifeedant results agree with that reported by Ahmed (1983) who reported that lantana show antifeedant activity to *Athalica proxima* 3<sup>rd</sup> instars larvae. This may be attributed to the presence of lantadenes A and B and high flavonoids content which associate with deterrence against insects (Ghisalberti, 2000).

The published data mentioned that lantana caused significant feeding deterrence to lepidopera species (Akhtar et al., 2007), significant protection to the potato against potato tuber moth *Phthormiaea operculella* as powder fumigant (Lal et al., 1987) and show significant antifeedancy to the 3<sup>rd</sup> larvae of julesemi-hoppers *Anomissabulif* (Sharma et al., 1990).

The feeding ratio was dose dependent. The significant FR was shown at all treatments (Ethanol and water extracts). The least FR (0.0084) was observed at 10% ethanol extract, while 2.5% showed the highest FR (0.011).

Antifeedant results of *S.argel* agree with that reported by Kogar (1982) who mentioned that alkaloids had antifeedant inhibition effect on some insect species. The antifeedant action of *S.argel* due to the presence of compounds mentioned to have antifeedant inhibiton effect on some insects like alkaloids (Kogar, 1986) and sterols (Nayer, 1962) . The same active compounds were found in the present Phytochemical screening results.

The feeding ratio was dose dependent. The significant FR was shown at all treatments (Ethanol and water extracts). The least FR (0.012) was observed at 10% (ethanol and water) extract, while 2.5% showed the highest FR (0.0176).

*Lupinus termis* and *N.rustica* showed no significant effects as antifeedant to *H.armigera* larvae. These results ensured that Termis and Tobacco are host plants of *H.armigera* .

## Repellency effects

Generally the repellency results indicated that only *O.basilicum* and *L . camara* demonstrated repellency effects on *H.armigera* larvae as shown by their positive percent repulsion. While *S.argel*, *L.termis* and *N.rustica* showed negative percent repulsion. The latest result may be due to absence of phenolic compounds in their constituents . The PR and time are in opposite relation.

The repellency results showed that *O.basilicum* caused high effects as indicated by its high percent repulsion 66.7 . These results are in line of that reported by Sathyaseelan and Bhaskaran (2010) who found highest repellency by *O .basilicum* 90.1% against *Maconellicoccus hirsutus* and Singh *et al.*, (2012) who found that *O.basilicum* have repellency with 91% against *Aphis gossypii*. This high effect may be due to the fact that *O.basilicum* contains phenolic compounds in its constituents like linalool, charvicol, eugenol and esragol.

In other studies , its oil was reported to be repellent to garden pest (Quarles, 1999), *B.incarnatus* (Ibrahim,2003) and to cowpea beetle *Callosobruchus aculates* (Boeke *et al.*,2004).

The percent repulsion was time and extract type dependent. In ethanol extract the highest PR (66.7) was shown at the first hour of exposure and the least PR (40) was shown after 6 hrs of exposure. In water extract the highest PR (33.3) was shown at the first hour of exposure and the least PR (0) was shown after 6 hrs of exposure. As mentioned above the ethanol extract was most potent than water extract as repellency agent.

Results of the repellency effects of *L.camara* agree with that stated by Jacobson (1975) who mentioned repellency of Lantana to the diamond black moth, *Plutella xylostella* and Lal *et al.*, (1987) who reported that

leaf powder of Lantana show significant protection to the potato against potato tuber moth *Phthormiaea operculella*. This may be due to the phenolic compounds.

In other studies, lantana showed repellency to honey bees (Singh, 1977), Asian corn borer *Ostrin furnacalis* and aphid *Hyadaphis erysimic* (Morallo, 1984) and to stored product insects as fumigant (Rajendran and Sriranjini, 2008).

The percent repulsion was time and extract type dependent. In ethanol extract the highest PR (33.3) was shown at the first hour of exposure and the least PR (13.4) was shown after 6 hrs of exposure. In water extract the PR was (6.6) for both 1 and 6 hours of exposure. As mentioned above the ethanol extract was most potent than water extract as repellency agent.

### **Phytochemical screening**

Phytochemical screening results showed that *L.camara* compounds (alkaloids, sterols, triterpenes, flavonoids, tannins and saponins) are in line

with that reported by Begum *et al.*, (2000).

In other studies, lantana was found to have, triterpenoids, flavonoid and phenyllethanoid glucosides, iridoid glycosides, furanonaphthoquinone (Morton, 1994; Siddiqui *et al.*, 1995, Sharma *et al.*, 2000; Yadav and Tripathi, 2000), lantadene A and lantadene B and high flavonoid content are present in lantana (Ghisalberti, 2000), lantanoic acid and camaranoic acid, lantic acid (Begum *et al.*, 2008), camarinic acid (Siddiqui *et al.*, 1995), camangeloyl acid, camarinin (Begum *et al.*, 2003, 2006), oleanonic acid, and ursonic acid (Siddiqui *et al.*, 2000) lantanolic acid (Begum *et al.*, 2008), lantanilic acid (Barua *et al.*, 1976),  $\alpha$ -amyrin,  $\beta$ -sitosterol and lantadene B (Ahmed *et al.*, 1972), lantoic acid (Roy and

Barua, 1985), lantadene D (Sharma *et al.*, 1990), 22/-O-angeloyl-oleanolic acid, 22  $\beta$ -O-senecioid-oleanolic acid, 22 $\beta$  hydroxyl-oleanolic acid, 19 $\alpha$ -hydroxy ursolic acid and a new triterpenoid 3 $\beta$ - isovaleroyl-19 $\alpha$ -hydroxy-ursolic acid (lantaiursolic acid) (Pandey *et al.*, 1993), camarinic acid, camaric acid, camarilic acid and camaracinic acid (Siddiqui *et al.*, 1995; Begum *et al.*, 1995), 25-hydroxy-3-oxolean-12-en-28-oic acid, hederagenin and 19-hydroxyursolic acid (Singh, *et al.*, 1996), novel trans lactone containing euphane triterpenes A, B and C (O'Neill *et al.*, 1998), phenylpropanoid glycosides verbascoside, isoverbascoside isonuomioside A, calceolarioside E and derhamnosylverbascoside (Taoubi *et al.*, 1997), martynoside and verbascoside (Syah *et al.*, 1998), theveside (Ford and Bendall, 1980) in its chemical constituents.

*Ocimum basilicum* compounds (alkaloids, sterols, triterpenes, flavonoids, tannins and saponins). This agrees with that reported by Akgul (1989). *Solenostemma argel* compounds (alkaloids, sterols, triterpenes, flavonoids, tannins, saponins and cyanogenic glycosides). This result agrees with that reported by Innocenti *et al.*, (2005)

In other studies, the compounds of basil essential oil linalool, methyl chavicol, eugenol, estragol, thymol and *p*-cymen were found (Akgul 1989; Khatri *et al.* 1995; Pino *et al.* 1996; Martins *et al.* 1999; Keita *et al.* 2000), linalool, eugenol, (E)- $\alpha$ -bergamotene and thymol. Linalool, eugenol, methyl eugenol and fenchyl alcohol (Akgul 1989), methyl chavicol, linalool, methyl eugenol,  $\beta$ -caryophyllene,  $\alpha$ -pinene,  $\beta$ -pinene, limonene and camphene Khatri *et al.* (1995), linalool, methyl chavicol and eugenol Marotti *et al.* (1996), linalool, eugenol and bornyl acetate Özcan *et al.*, (2002), Recently Chang (2009) demonstrated that 12 volatile compounds were identified on GC-MS by comparison of MS spectra with these NIST mass spectra database these chemical include  $\alpha$

pinene, linalool, trans anethole, 4-(cimethoxy propyl)-benzene, trans-5-ol, 3 coryophyllene, methyl eugenol, 3, 11-trimethyl-(E), 1,6 doscatriene-3, 2, 3-dihydro-1H idone-[3-ido-2-(Iodinethyl)-2- methyl]-1,2-4,5 Idranethyl benzene, 1-(1,1-dimethyl)-2-methoxy-4-methyl-3, 5dini-triobenzene.

*Solenostemma argel* compounds (alkaloids, sterols, triterpenes, flavonoids, tannins, saponins and cyanogenic glycosides). This result agree with that reported by Innocenti *et al.*, (2005).

In other studies it was found that *S. argel* have the following compounds: indoline, alkaloids, steroidal, perogenine and their glucosides (Si-Qi *et al.*, 1993; Deepak *et al.*, 1989; Srivastava *et al.*, 1993). It's other cyanogenetic (glucosides, saponin, tannins, cumarins, flavonoids, phenolic acids and triterpenoides (Aeri, 2007). Murwan *et al.*, (2010) found that it's leaves contained phytic acid and tannin, Inno -centi *et al.*, (2005) was found that its aerial parts contained two monoterpene, glucosides, apregnane glucoside, benzyl alcohol O- $\beta$  apiofuranosyle (1-6)  $\beta$ glucopyrano -side, 2- phenyl-ethyl O- $\alpha$  arbinopyranosyl (1-6) $\beta$  glucopyranoside, astragaline and kaempferol 3-O-neohesperidose. Shafec *et al.*, (2012) isolated two new natural kaempferol glucosides namely kaempferol 3-O- $\beta$ -D-glucopyranosyl (1-2)  $\beta$ -D-xylopyranoside (S2) and kaempferol 3-O- $\alpha$ -L-arabinopyranosyl (1-2)  $\beta$ -D-galactopyranoside (S3), (Osborn, 1968 and El-Fishawy, 1976) Sitosterols, choline, flavonoids, glycosides, namely argelin and argelosid and a triterpinoid saponin. Previous studies have reported the presence of monoterpenes (Kamel *et al.*, 2000) pregnane glycosides (Hassan *et al.*, 2001; Hamed, 2001). Palaza *et al.*, (2003) reported the occurrence of novel pregnane glycosides namely argeloside from *S. argel*. Also monoterpene, glycosides, pregnane glycosides, flavonoids and tannins as well as other seroids and alkaloids were isolated and identified from different parts of the herb (Kamel *et al.*, 2000; Hamed, 2001). This variations of chemical

constituents may be to that plant chemical constituents reported to be influenced by genetic, geographical seasonal factors , plant stage and the processing method.

### **Probit analysis**

Results in probit analysis showed that lantana was most effective against *H.armigea* as indicated by their low values of LD50. Chi-square was small indicated good excusion of experiment. The slope were steep indicated homogenous population. Increase in concentrations increase the slope of regression line and caued progressive improvement of homogeneity.

This study reveals the insecticidal properties , antifeedant and repellency effects of Lantana and Rehan and the potential of these products for developing commercial formulation for managing *H. armigera*.

### **Conclusion**

In acordance with the present study one can conclude that :

The present work is an attempt to add something new and useful to efforts through the use of lantana, Rehan and Hargal plant in ABW.control.

The study concludes that lantana was the most effective in killing African bollworm in comparison to other plant extracts tested.

The study demonstrates that lantana was most effective as contact toxicity agent.

The study demonstrates that Rehan was the most effective as antifeedant and repellent agent.

These plants may be used as population controlling agents for *H.armigera* as they are cheaper and biodegradable, producing minimal polloution and can be prepared esasily by farmers.

Recommendations:



- In spite of good results of lantana on *H.armigera* there is no information about its side effects so extensive research is needed before it finds its place in *H.armigera* control.
- Future lines of research should include the efficacy of natural products against other tomato insect pests.
- More future studies of mode of action and toxicity for the above mentioned plants are needed .
- Improvement of farmers' knowledge about biopesticide are needed.
- An IPM approach to control a complex insect pests of tomatoes.
- More management for insecticides used in tomatoes.

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## APPENDICES

### Appendix (1)

Metrological data for the dry climatic zone in the Sudan.

Evap. mm/annum	Rainfall mm/annum	Temp. range	Min. temp.c°	Max. temp.c°	Solar radiation mj/m <sup>2</sup> /day	Sunlight Duration hr/year
2300	100-250	30	12 (Jan)	42 (May)	22.3	3550

Source;Adam (2002)



Appendix : (2)

Number of eggs and larvae of *H.armigera*/100 plants

Date	scheme					
	Elmogmaat		Eltawiella		Elsoufii	
	eggs	larvae	eggs	larvae	eggs	larvae
25/9	70	20	-	-	-	-
30/9	30	180	-	-	-	-
8/10	400	200	300	-	-	-
14/10	50	250	300	1	-	-
22/10	50	30	600	4	-	-
4/11	100	-	100	700	-	-
11/11	3100	270	100	-	-	-
18/11	300	800	200	300	200	-
29/11	200	200	300	-	-	100
8/12	100	300	200	100	300	600
17/12	-	100	100	1600	200	330
27/12	-	-	-	300	100	200
4/1	-	-	-	100	100	100
15/1	-	-	-	-	-	100
25/1	-	-	-	-	-	-
<b>Total</b>	<b>4400</b>	<b>2350</b>	<b>2200</b>	<b>3105</b>	<b>900</b>	<b>1430</b>
<b>Mean</b>	<b>440</b>	<b>231.6</b>	<b>244.4</b>	<b>310.5</b>	<b>150</b>	<b>238.3</b>
<b>Sd±</b>	<b>942.5</b>	<b>220.7</b>	<b>159</b>	<b>504</b>	<b>104.9</b>	<b>199</b>

### Appendix (3)

#### Morphometrics of *Helicoverpa armigera* development stages

Stage	Eggs		Larval instar										Pupa		Adult			
			1st		2d		3d		4th		5th				male		Female	
Rep.	length	width	length	width	length	width	length	width	length	width	length	width	length	width	Wing span	Body length	Wing span	Body length
1	0.6	0.5	1	0.40	3.2	0.8	8	2.2	9.2	2.6	15	3.3	20	5.5	28	17	38	21
2	0.4	0.4	1.3	0.42	3	0.8	8.5	2.4	10	2.7	14	3	18.8	5.1	30	17	34	18
3	0.5	0.45	1.7	0.55	4.5	1	7.8	2	9.7	2.6	18	3.4	19	5	26	15	38	22
4	0.4	0.43	1.4	0.46	4.7	1.3	8.5	2	11	2.8	23	3.5	17.8	5	25	16	36	19
5	0.6	0.48	1.6	0.52	4.5	1.2	6.7	2	10	2.7	24	3.7	16	4.5	29	15	32	16
6	0.45	0.4	1.4	0.53	3.2	1	7.4	2.3	9	2.5	13	3	20	4.8				
7	0.55	0.43	1.5	0.6	4	1.3	9	2.4	12	3	18	3.4	18	4.42				
8	0.5	0.47	1.45	0.55	5	.9	8.3	2	10.5	2.7	17	3.2	17	4.65				
9	0.47	0.4	1.35	0.45	4.5	1	8	2.6	9.5	2.4	13	3.3	16	5.2				
10	0.56	0.45	1.5	0.43	4.7	1.2	8.5	2.5	10	2.6	16	3	18	4.9				
Mean	0.5	0.44	1.42	0.5	4.1	1.1	8.27	2.3	10.1	2.7	17.1	3.3	18.6	4.9	27.6	16	35.8	19.2
± Sd	0.07	0.04	0.19	0.07	0.7	0.2	0.7	0.2	0.9	0.2	3.8	0.2	1.4	0.33	2.1	1	2.3	2.4

Appendix (4)

Stages development periods of *H.armigera* reared on tomato leaves/days

Stage Rep.	Egg	Larval instars					Prepupa	Pupa	M
		1 <sup>st</sup>	2 <sup>d</sup>	3 <sup>d</sup>	4 <sup>th</sup>	5 <sup>th</sup>			
1	3	2	2	2	2	4	3	12	9
2	3	2	2	3	3	3	1	9	7
3	4	3	3	2	4	3	4	11	11
4	5	2	2	2	3	4	2	9	6
5	3	3	3	4	3	3	3	10	11
6	4	3	2	2	5	4	4	8	11
7	4	2	4	3	2	4	4	7	11
8	5	3	3	2	4	3	5	9	11
9	5	2	2	4	3	3	2	11	11
10	3	2	4	5	3	3	3	10	11
mean	3.9	2.4	2.7	2.9	3.2	3.4	3.1	9.6	9
±Sd	0.9	0.5	0.8	1.1	0.9	0.5	1.2	1.5	2

### Appendix (5)

Eggs hatching ,pupation and adult emergence percent of *Helicoverpa armiger* reared on tomato

Stage Rep.	Number of eggs/mass	Number hatched	Hatching %	Number of pupae formed	pupation %	Adult emergence
1	16	9	56.5	6	66.7	5
2	26	17	65.4	13	76.5	10
3	18	12	66.7	9	75	7
4	33	25	75.8	19	76	16
5	27	19	70.4	15	78.9	14
total	120	82	334.8	62	373.1	52
mean	24	16.4	66.95	12.4	74.62	10.4
±Sd	6.9	6.2	7.1	5	4.6	4.6

## Appendix (6)

### Fecundity duration of *Helicoverpa armigera* reared on tomato leaves

Day Rep.	Eggs number/female/day																		total
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	
1	-	-	16	27	35	41	87	65	30	22	12	11	6	-	-	-	D	-	351
2	-	-	13	32	64	71	99	70	35	14	9	3	-	-	-	-	D	-	410
3	-	-	-	9	25	51	78	107	86	65	36	18	9	6	-	-	-	D	490
4	-	-	20	46	57	77	77	98	63	42	38	40	19	-	-	-	D	-	585
5	-	-	22	31	49	69	86	115	82	57	-	-	-		D	-	-	-	511
Mean	-	-	15.8	29	46	61.8	85.4	91	60.8	40	19	14.4	6.6	1.2	-	-	-	-	469.4
±Sd			8.6	13	28.8	15.1	9	22.3	25.9	21.9	25.9	10	7.9	2.7					90.9

Appendix (7)

Effects of *Lantana camara* extracts on *Helicoverpa armigera* 3<sup>rd</sup> instar larvae

Extract		Ethanol											
Concentration%		2.5			5			10			15		
Rep.	N. treated	Number death/hr			Number death/hr			Number death/hr			Number death/hr		
		24	48	72	24	48	72	24	48	72	24	48	72
1	12	1	3	6	3	6	8	6	7	8	4	7	9
2	12	1	3	6	3	6	7	4	8	9	5	8	9
3	12	1	3	6	3	6	6	5	6	7	3	9	12
total	36	3	9	18	3	18	21	15	21	23	12	24	30
mean		1	3	6	1	6	7	3	7	8	4	8	10
Sd		1.7	0	0	0	0	1	1	1	1	1	1	1.7
Mortality%		8.3	25	50	25	50	58.3	41.6	58.3	66.6	50	66.6	83.3

Appendix (8)

Effects of *solenostemma argel* extracts on *Helicoverpa armigera* 3<sup>rd</sup> instar larvae

Extract		Ethanol											
Concentration%		2.5			5			10			15		
Rep.	N. treated	Number death/hr			Number death/hr			Number death/hr			Number death/hr		
		24	48	72	24	48	72	24	48	72	24	48	72
1	12	1	3	3	4	6	7	2	2	2	2	2	2
2	12	1	1	3	1	3	5	1	2	2	2	2	2
3	12	1	2	3	1	3	6	1	1	2	1	1	3
total	36	3	6	9	6	12	18	4	5	6	5	5	7
mean		1	2	3	2	4	6	1.33	1.66	2	1.66	1.66	2.33
Sd		0	0.58	0	0.58	0.58	0	0.58	0.58	0.0	0.58	0.58	0.58
Mortality%		8.3	16.7	25	16.7	33.3	50	33.3	41.7	50	41.7	41.7	58.3

Appendix (9)

Effects of *Ocimum basilicum* extracts on *Helicoverpa armigera* 3<sup>rd</sup> instar larvae

Extract		Ethanol											
Concentration%		2.5			5			10			15		
Rep.	N. treated	Number death/hr			Number death/hr			Number death/hr			Number death/hr		
		24	48	72	24	48	72	24	48	72	24	48	72
1	12	0	0	1	0	0	1	3	5	5	5	5	6
2	12	0	0	1	0	0	1	2	4	6	5	5	6
3	12	0	0	1	0	0	1	4	3	4	3	4	6
<b>total</b>	<b>36</b>	<b>0</b>	<b>0</b>	<b>3</b>	<b>0</b>	<b>0</b>	<b>3</b>	<b>9</b>	<b>12</b>	<b>15</b>	<b>13</b>	<b>13</b>	<b>18</b>
<b>mean</b>		<b>0</b>	<b>0</b>	<b>1</b>	<b>0</b>	<b>0</b>	<b>1</b>	<b>3</b>	<b>4</b>	<b>5</b>	<b>4.3</b>	<b>4.3</b>	<b>6</b>
<b>Sd</b>		<b>0</b>	<b>0</b>	<b>1.7</b>	<b>0</b>	<b>0</b>	<b>1.7</b>	<b>1</b>	<b>1</b>	<b>1</b>	<b>1.2</b>	<b>1.2</b>	<b>0</b>
<b>Mortality%</b>		<b>0</b>	<b>0</b>	<b>8.3</b>	<b>0</b>	<b>0</b>	<b>8.3</b>	<b>25</b>	<b>33.3</b>	<b>41.7</b>	<b>33.3</b>	<b>33.3</b>	<b>50</b>



Appendix (10)

Effects of *Lupinus termis* extracts on *Helicoverpa armigera* 3<sup>rd</sup> instar larvae

Extract		Ethanol											
Concentration%		2.5			5			10			15		
Rep.	N. treated	Number death/hr			Number death/hr			Number death/hr			Number death/hr		
		24	48	72	24	48	72	24	48	72	24	48	72
1	12	0	0	0	0	0	1	3	3	3	4	6	6
2	12	0	0	0	0	0	1	4	5	3	4	5	6
3	12	0	0	0	0	0	1	2	1	3	3	4	6
total	36	0	0	0	0	0	3	9	9	9	11	15	18
mean		0	0	0	0	0	1	3	3	3	3.7	5	6
Sd		0	0	0	0	0	0	1	1	1	0.58	1	0
Mortality%		0	0	0	0	0	8.33	25	25	25	33.3	41.7	50

Appendix (11)

Effects of *Nicotiana rustica* extracts on mortality of *Helicoverpa armigera* 3<sup>rd</sup> larvae

Extract		Ethanol											
Concentration%		2.5			5			10			15		
Rep.	N. treated	Number death/hr			Number death/hr			Number death/hr			Number death/hr		
		24	48	72	24	48	72	24	48	72	24	48	72
1	12	0	0	0	0	0	0	0	0	1	0	1	1
2	12	0	0	0	0	0	0	0	0	1	0	4	4
3	12	0	0	0	0	0	0	0	0	1	0	1	1
<b>total</b>	<b>36</b>	<b>0</b>	<b>0</b>	<b>0</b>	<b>0</b>	<b>0</b>	<b>0</b>	<b>0</b>	<b>0</b>	<b>3</b>	<b>0</b>	<b>6</b>	<b>6</b>
<b>mean</b>		<b>0</b>	<b>0</b>	<b>0</b>	<b>0</b>	<b>0</b>	<b>0</b>	<b>0</b>	<b>0</b>	<b>1</b>	<b>0</b>	<b>2</b>	<b>2</b>
<b>Sd</b>		<b>0</b>	<b>0</b>	<b>0</b>	<b>0</b>	<b>0</b>	<b>0</b>	<b>0</b>	<b>0</b>	<b>0</b>	<b>0</b>	<b>1.7</b>	<b>1.7</b>
<b>Mortality%</b>		<b>0</b>	<b>0</b>	<b>0</b>	<b>0</b>	<b>0</b>	<b>0</b>	<b>0</b>	<b>0</b>	<b>8.3</b>	<b>0</b>	<b>16.7</b>	<b>16.7</b>

. Appendix (12)

Effects of Dimethoate and control

Treatment		Dimethoate			
Concentration%		1ml/L			
Replication	N. treated	Number death/hr			
		24	48	72	24
1	12	12	—	—	0
2	12	12	—	—	0
3	12	12	—	—	0
total	36	36	—	—	0
mean	12	12	—	—	0
Sd		0	—	—	0
Mortality%		100	—	—	0

Appendix (13)  
Food deterrent and feeding ratio of botanical extracts /24 hours

plant	No. treated	Extract	Concentration	Mean of consumed food (g)/24 hrs
<i>Ocimum basilicum</i>	3	Ethanol	2.5	35.07
	3		5	0.33± 0.06
	3		10	0.27 ±0.07
	3	water	10	0.27± 0.05
<i>Lantana camara</i>	3	Ethanol	2.5	0.55± 0.2
	3		5	0.45± 0.1
	3		10	0.42± 0.1
	3	water	10	0.45± 0.16
<i>Lupinus termis</i>	3	Ethanol	2.5	0.77± 0.1
	3		5	0.71± 0.1
	3		10	0.65± 0.17
	3	water	10	0.8± 0.07
<i>Nicotiana rustica</i>	3	Ethanol	2.5	.086± 0.1
	3		5	0.75± 0.2
	3		10	.065± 0.13
	3	water	10	0.90± 0.22
<i>Solenostemma argel</i>	3	Ethanol	2.5	.088± 0.08
	3		5	0.75± 0.1
	3		10	0.6± 0.22
	3	water	10	0.85± 0.1
Dimetoate	3		1 ml/l	0.74± 0.03
control	3	water	0	0.95± 0.1

D.C: deternt coefficient      FR: Feeding ratio

Appendix (14)  
 Repellency Effects of different botanical extracts on *H.armigera* 3<sup>rd</sup> instar  
 larvae applied at 10% /hour

Plant	Replication	Number treated	Number settled on untreated leaf/hrs					
			Water extract			Ethanol extract		
			1hrs	6hrs	12hrs	1hrs	6hrs	12hrs
<i>L.camara</i>	1	10	8	6	4	8	8	3
	2	10	7	7	3	7	7	5
	3	10	5	3	5	9	6	6
	Total	30	20	16	12	24	21	14
	Number settled on untreated leaf %		66.7%	53.3%	40%	80%	70%	47.7%
	Percent replusion		33.4	6.6	-20	60	40	-6.6
<i>N.rustica</i>	1	10	3	2	3	6	3	2
	2	10	4	1	0	4	3	3
	3	10	7	3	1	5	3	3
	Total	30	14	6	4	15	9	8
	Number settled on untreated leaf %		46.7%	20%	13.3%	50%	30%	26.7
	Percent replusion		- 6.6	-60	- 73.4	0	-40	-46.7
<i>O.basilicum</i>	1	10	5	5	3	8	6	4
	2	10	5	5	4	6	6	3
	3	10	6	5	2	6	5	3
	Total	30	16	15	9	20	17	10
	Number settled on untreated leaf %		53.3	50%	30%	66.7	56.7	33.3
	Percent replusion		6.6	0	-40	33.3	13.4	-33.4
<i>L.termis</i>	1	10	5	5	3	4	3	4
	2	10	5	5	4	6	5	3
	3	10	6	5	2	3	5	3
	Total	30	16	15	9	13	13	10
	Number settled on untreated leaf %		53.3	50%	30%	43.3	34.3	33.3
	Percent replusion		6.6	0	-40	-13.4	-13.4	-33.4
<i>S.argel</i>	1	10	4	4	3	3	3	2
	2	10	3	4	2	3	4	5
	3	10	5	3	5	5	3	3
	Total	30	12	11	10	11	10	10
	Number settled on untreated leaf %		40	36.7	33.3	36.7	33.3	33.3
	Percent replusion		-20	-26.6	-33.3	-26.6	-33.4	-33.4
Dimethoate 1ml/L	1	10	5	5	-			
	2	10	5	4	-			
	3	10	5	5	-			
	Total	30	30	14	-			
	Number settled on untreated leaf %		100%	46.7	-			
Percent replusion		0	-6.8	-				