BIO-EFFICACY OF SOME INDIGENOUS GHANAIAN BOTANICAL AGAINST THE ALMOND MEAL MOTH: *Ephestia cautella* (WALKER)

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DECLARATION

This is to satisfy that this thesis is the result of research undertaken by PAUL APPIAH towards the award of the Master of philosophy in Crop Science (Post Harvest Technology) in the Department of Crop Science, University of Ghana.

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DEDICATION

I dedicate this work to God Almighty who has seen me through this programme and in particular, this research work.

Secondly, I also dedicate this to my parents, Nana Appiah Hene and Madam Amma Fosua for their unflinching support and encouragement during this period.

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TABLE OF CONTENT

CONTENTS **PAGE**

LIST OF TABLES

LIST OF FIGURES

LIST OF PLATES

ABSTRACT

Ephestia cautella is one of the most important stored product insect pests found in granaries. Its activities tend to reduce germination and nutritive capacity of the infested seed and grains. The increasing rate of pest resistance, high cost and health risk in the use of insecticides to control insect pest have become issue of public concern. Therefore the use of botanical has been found to be very effective against stored product insect pests. Several plant products (leaves, oil, ash, seed extracts, and roots) are processed and mixed with stored products or applied as insecticides to control these pests. These are considered to be cheap, easily biodegradable and readily available for stored product protection. In this research, the bio-efficacies of *Moringa oleifera, Ocimum canum, Securidaca longepedunculata* and *Zanthoxylum xanthoxyloides* as bio-insecticidal agents against *E. cautella* were investigated. The powders of *M. oleifera*, *O.canum*, *S. longepedunculata* and *Z. xanthoxyloides* leaves were screened in the laboratory against the adults *E. Cautella* in the proportion of 50% and 100% wt/wt for all the botanicals. These showed the various bioactivities against the insects with 100% wt/wt treatments of *Zanthoxlum xanthoxyloides* proving to be most effective, as it significantly reduced the survival (90%) of the insects. The effect of petroleum ether extracts of the botanicals at different concentration (0.1g/mL, 0.3g/mL and 0.5g/mL) on the adult insect by grain treatment, effects on immature stages, effects on adult emergence in treated grains repellency, damage to the grains and germination tests were determined in the laboratory. The study showed that, the ability of the petroleum ether extracts to cause mortality was dosage dependent since mortality increased with higher concentration of the extracts. Significantly (p=0.005) lower number of eggs were laid on the treated grains compared with the untreated control. The botanicals had a tremendous effect on the adult emergence of *E. cautella*. There was no adult emergency at the highest concentration (0.5g/ml) for *Moringa oleifera*. There were significant differences in the number of adults

emergence between the various treatments and the control. Overall percentage weight loss was lower (0.0-6.80%) in the botanical treatments than that of the untreated control (9.4-31.51%). Grains treated with botanicals gave an appreciable reduction in weight loss as compared with the control. Studies on the effects of the botanical on seed viability revealed that the botanicals did not hamper seed viability after 60 days storage period. The untreated control had the least percentage germination (25.0-25.3%). There was significant repellence of the insect by the different concentrations of the plant extracts. These results indicate that good protection of maize seed can be offered by indigenous Ghanaians botanicals acting as both repellents and anifeedants against *Ephestia Cautella*.

CHAPTER ONE

1.0 INTRODUCTION

Maize (*Zea mais* L, from Spanish: *maiz* after *Taino mahiz*) known in many countries as corn is a common name for the cereal grass widely grown for food and livestock fodder (FAOSTAT, 2009). It ranks with wheat and rice as one of the world's chief grain crops (Anon, 2012). The global production of maize is estimated to be 817 mmt annually (FAOSTAT, 2009). In sub-Saharan Africa, maize is one of the most important grain staples for Agricultural income and caloric intake accounting for nearly 20% of plant-based food supply (Jones *et al*., 2011).

Maize is the most important cereal crop produced in Ghana and it is also the most widely consumed staple food in Ghana with increasing production since 1965 (FAO, 2008; Morris *et al*., 1999).

About 1.6 million hectares of maize is cultivated annually in Ghana, 80% of which is owned by smallholder farmers (Anankware *et al*., 2012). It is grown throughout Ghana but the leading producing areas are mainly in the middle-Southern part (transitional and forest zones); with an estimated 15% grown in the northern regions of the country (WABS Consulting Ltd., 2008). Average annual production increased from 141,000 metric tons in 1983 to 533,000 metric tons in 1993 (PPMED, 1993) and currently 1,871,700 metric tons (ISSER, 2011). The current value of maize production is approximately US $400,000,000 a year (late 2008 values) (ISSER, 2011).

The Agricultural sector-contributes largely to the economy of most countries in the world today. In Ghana, the Agricultural industry contributed 28.3% to the annual Gross Domestic Product (GDP) in 2011 (ISSER, 2011). The increase in production and exports of Agricultural products as well as creation of jobs for the indigenous people general contribute to the sector's role in the economy of Ghana.

In Ghana, maize is one of the principal crops and is used in the preparation of major meals such as "kenkey, banku, akple" etc. Maize may be roasted with groundnuts and are chewed together as "abro ne nkatie" (meaning: maize and groundnuts). It may also be used in the preparation of local drinks such as "pito" and "meda".

With the increase in the world's population it has become necessary to increase Agricultural production in order to meet the food security demands of the ever increasing world's population. One major constraint to increased and sustainable production of maize is its susceptibility to infestation by insect pests both in the field and during storage (Singh *et al*., 1990).

Insect infestation of maize grains during storage cause considerable loss due to tunneling activity of larvae of maize weevil, *Sitophilus zeamais*, the tropical warehouse moth, *Ephestia cautella* (Walker) the larger grain borer, *Prostephanus trancatus* (Horn) and *Sitotroga cerealla* among others. The larvae of *Ephestia cautella* attack the grain germ, moving from grain to grain and consuming these parts. Damage to the commodity is therefore much greater than the actual weight of grain consumed due to this spoilage factors. Contamination of the infested commodity also occurs from large quantity of webbing which spun over its surface. In heavy moth infestations of stored grains, this webbing looks like a fine white film covering the whole surface of the bagged stack (FAO, 1992).

In the developing countries, food grain production and consumption often fall below demand as a result of postharvest losses, and in Africa where subsistence grain production supports the livelihood of the majority of the population, grains loss caused by storage pests such as *Ephestia cautella* threatens food security. Insect damage in stored grains is a serious problem in developing countries in the tropics due to favourable climatic conditions and poor storage structures (Bekele *et al.*, 1997).

1.1 JUSTIFICATION:

Several methods have been used over the years to protect cowpea and maize grains in storage, but the use of synthetic insecticides have been dominant (Adabie-Gomez *et al*., 2006). Chemical methods of controlling this pest are the easiest and most rapid but come with a lot of adverse effects (Obeng-Ofori, 2008). The pervasive use of these insecticides in granaries of small scale farmers has led to a number of problems such as killing of non-target species, user hazards, food residues, and evolution of resistance to the chemicals, high cost of the chemical and the destruction of the balance of ecosystem. Therefore, the search for alternative insect pest control methods and materials which are relatively cheaper and less harmful to the user and the environment has become essential (Arthur *et al*., 2002). Attention has been focused on the use of indigenous plant sources for cheap and locally available pesticides. Over 200 plant species have insecticidal properties capable of controlling insects (Arnason *et al*., 1989).

Jatropha curcas (Euphorbiaceae) plant is one of the many plant species that have been used to control stored product pest. The efficacy of Jatropha seed oil against cowpea weevils has been reported (Huis, 1991; Adebie-Gomez *et al*., 2006; Henning, 2007). However, other locally prepared powders and extracts from indigenous botanicals have not been fully investigated on their efficacies against stored products and should be evaluated against these weevils so as to provide a wider option for maize seed producers.

1.2 OBJECTIVES:

The study aims at evaluating the efficacies of *Ocimum canum*, *Moringa oleifera Gaertn*, *Securidaca longependunculata* Fress and *Zanthoxylum xanthoxyloides* Lam extracts against *E. cautella*. The specific objectives were to:

- determine the insecticidal efficacies of the powdered form of *O. canum*, *M. oleifera*, *S. longependunculata* and *Z. xanthoxyloides* against the adult *E. Cautella*
- Assess the insecticidal efficacies of petroleum ether extracts of the plants against adult *E. Cautella*
- Determine the effect of the,

a. extracts on the emergence of adult *E. cautella* in treated grains.

b. extracts on grain damage by adult *E. cautella*.

c. plants extracts on maize seed viability.

- Determine the protective potential of the best extract under semi-field conditions against *E. cautella*.

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Maize Production and Importance in Ghana.

Maize (*Zea mais* L.), the third most important cereal crop after rice and wheat in the world (FAO, 1992) is Ghana's most important cereal. It is grown by the vast majority of rural households in all parts of the country except the Sudan Savannah zone of the far north (Morris *et al* 1999). It was introduced into Ghana in the 16th century by the Portuguese traders. Maize is cultivated in all the agro ecological zones in Ghana, however cropping systems and production technologies vary in these ecological zones (Morris *et al*., 1999). Depending on the agro ecological location, there may be one or two cropping seasons. In Ghana, maize production is done largely by small scale farmers whose farm sizes are usually less than five hectares (CIMMYT, 1990). However, large scale production is done in the maize belt of Ejura-Techiman-Wenchi-Odumasi area in the forest savannah transition zone (Boateng, 1996).

Maize is also an important non-traditional agricultural export commodity which together with rice contributes tremendously to the export volume of non-traditional cereal crops compared to other cereals crops in Ghana.

2.2. Maize Storage Practices in Africa

Storage of farm produce during off-season is an anti-famine and long-lasting food security strategy which ensures that there is enough food to meet the needs of consumers across years in case there is famine; particularly when drought, locust invasion, and other problems such as wars prevent farmers from producing food crops in one or more seasons (Gari, 2004).

Food and seed insecurity in Africa is recurrent problem which is more acute among the poorest farmers (Gari, 2004). Among the causes of food and seed insecurity in Africa is inadequate storage facilities and inappropriate methods for food, and seed storage especially among rural farmers. Successful seed storage is paramount to farmers' seed security and may also enable communities to generate income through collecting, storage and selling seeds.

Specific storage practices vary widely according to climate zone, cultural traditions and production scale or socio-economic condition of farmers. Since most of the grains produced in Africa are destined for human consumption, storage in family granaries predominates. The common storage techniques utilized by small-holder producers in West Africa vary greatly, but include on-field, open storage, jute bags, polyethylene or polypropylene bags, raised platforms, conical structures with thatched roofs, clay structures and giant woven baskets (Addo *et al*., 2002, Jones *et al*., 2011). Farmers may also store bags in their personal rooms, on cobs above fireplaces, or simply heaped on the floor (Hell *et al.*, 2000). These are generally considered ‘’traditional’’ storage method, while improved covered structures or ‘’cribs’’ may be termed ‘’semi-modern’’ and formal silos and warehouses termed ‘’modern’’ storage systems (Jones *et al*., 2011). Though shelling of grain and insecticide application is officially promoted by many Ministries of Agriculture, storage of maize on cobs (husked and de-husked) is almost universal (Jones *et al*., 2011). If maize is transported during the storage season, however, it is very commonly threshed and bagged (Addo *et al*., 2002).

Producers in East and Southern Africa store in small bags with cow dung ash, wood and wire cribs, pits, metal bins, wooden open-air or roofed cribs, raised platforms and roofed iron drums enclosed with mud, or may hang cobs over a fireplace. In Zambia, many farmers prefer to store maize in a temporary house structures after harvest until fully dry, then shell and transfer the grain to polypropylene bags inside the house before sale, consumption, or planting. Insecticide, when used, is applied directly on husked stores as well as admixing with shelled grain in sacks (Jones *et al*., 2011).

Unfortunately, the management and effectiveness of family granaries and other storage structures are seriously wanting. These predispose the grains to serious attacks from biotic constraints such as insects, rodents, birds, and micro-organisms (Nukenine, 2010). The rate of insect proliferation in these storage structures could be alarmingly high, especially with the warm climate in tropical Africa (Nukenine, 2010). Annual grain losses of up to 50% in cereals and 100% in pulses have been reported (Obeng-Ofori, 2011), although average losses stand at roughly 20% (Obeng-Ofori, 2011), thus, the need for better storage systems to protect grains and other food crops.

2.2.1 **Post Harvest Losses in Stored Maize**

Post production practices involved in the handling of maize grains are harvesting, transporting, drying, shelling, grain cleaning, storage and processing (FAO, 1992). Qualitative and quantitative losses are the main types of post harvest losses in maize. Quantity losses can occur as a result of inappropriate harvesting methods, spillage during transportation, or damage by pest organisms causing reductions in weight or volume. Quality losses occur as changes in colour, smell or taste, contamination with toxins, pathogens, insects or rodent excreta, reduction in nutritional value or loss of viability if the harvest is meant for seed (CIMMTY, 2008).

In developing countries, post harvest losses in durable stored grains are estimated to be about 30% (cited Obeng-Ofori, 2007). Generally, insects are the most serious pests of stored maize (CIMMTY, 2008) and in developing countries like Ghana, the greatest losses to cereals and grain legumes during storage are caused by insect pests (Obeng-Ofori, 2007).

2.3 Storage Insect Pests of Maize

In Ghana, stored maize is susceptible to attack by several insect pests including the maize weevil *Sitophilus zeamais*, the larger grain borer, *Prostephanus truncatus* Horn (Coleoptera: Bostrichidae), the Angoumois grain moth, *Sitotroga cereallela* Oliver (Lepidoptera: Gelechiidae) and *Ephestia cautella*. *Ephestia cautella* is one of the commom pests of stored maize (CABI, 2006).

2.4. The Tropical Warehouse Moth, *Ephestia Cautella*

2.4.1. Description

The eggs of the tropical warehouse moth are translucent yellow with a distinctly sculptured surface pattern. Its larvae range from 1.2-1.5 cm in length and are pale gray or light brown in colour with many setae and small dark brown spots on the cuticle. The head capsule is dark and they have a sparse covering of hair. In the males the testes can be seen through the cuticle as a dark patch in the posterior region. The pupae are dark-brown and found within a relatively light pupal case. The adult of the tropical warehouse moth has reddish brown forewings with faintly coloured white cross lines and the hind wings are pale gray. The wing span is 1.4-2.2 cm and wing fringes are short. Its labial palpi curve upwards in front of the head and are rather blunt at the tip (Sedlacek *et al*., 1995; CABI, 2006).

2.4.2 Biology and ecology

Pairing takes place shortly after emergence and egg laying starts 24 hours later (Navarro and Noyes, 2000). The eggs of *E. cautella* are laid by the female directly in the food commodity at the end of copulation. They are globular and white when laid and gradually turn orange as they develop. About 300 eggs are laid in the first 3 to 4 days and few in the fourth or fifth day. The eggs hatch in about 4 days between 10 °C and 38 °C with optimum conditions of 25 °C and 80% to 90% relative humidity (Navarro and Noyes, 2000).

Newly hatched larvae wander in search of food and are able to penetrate very small cracks in packages. There are five larval development of stages ranges from 28 to 35 days, 22 to 50 days in copra, 49 days in cocoa and 64 days in raisins (Navarro and Noyes, 2000). The first instar larvae are visible to the naked eye and begin to feed immediately upon hatching forming small galleries of silk and frass within the food. Fully grown larvae leave the food layer if they are numerous and enter a wandering stage searching for cracks and crevices in which they pupate

(Takahashi, 1955; Navarro and Noyes, 2000). They spun large quantities of silk during this period and this covers breeding containers, walls and ceilings of warehouses and flour mills. The last instar larva manufactures a cocoon before pupation. The pupal stage is completed in about 7 days at 30°C and 70% relative humidity (cited in Benson, 1973). Duration of the eggs and pupa stage is not influenced by relative humidity but by temperature. Lower humidity retards larval growth and very low humidity results in death.

The adults are mainly nocturnal and many emerge in the late afternoon and evening and usually live for 1-3 weeks (cited in Benson, 1973). The development from eggs to adult depends on temperature, moisture content and the type of food (cited in Benson, 1973). Optimal development occurs at 30-32°C and 70-80% relative humidity.

The complete life cycle takes 1 to 3 months under field conditions according to climate and food substrate and in the laboratory, under constant temperature and humidity conditions it takes 6 weeks at 24°C and 75% relative humidity (Navarro *et al*., 1991).

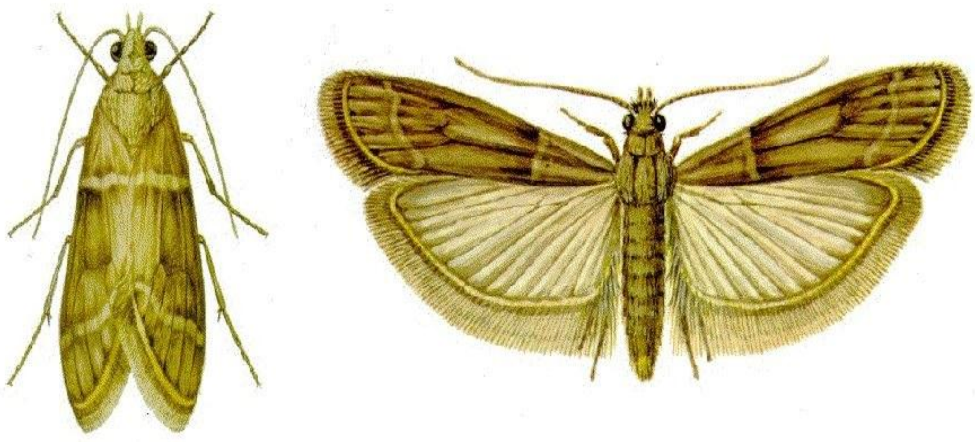


Plate 2.1: The adult *E. cautella*

2.4.3 Distribution

Ephestia cautella is a predominant pest in the tropics and sub tropics but less common in the arid areas (CABI, 2006). It has been reported to be present in Europe, Asia, Africa, North, South and Central America and Caribbean (CABI, 2006). In Africa, it is found in countries such as Ghana, Nigeria, Togo, and Ethiopia (Haines, 1991).

2.4.4 Economic Importance

The larva of tropical warehouse moth is the damaging phase and a general feeder. It attacks plant products by causing damage to the fruits, pods, and seeds. Larvae are found as primary pest in a wide range of commodities. The major host includes groundnut (*Arachis hypogeal*), cocoa (*Theobroma cacao*), maize (*Zea mays*), soyabean *(Glycerine max*), cassava (*Minihot esculenta*) and rice (*Oryza sativa*) (CABI, 2006). The preferred food includes stored vegetable matter such as flour, grains, dates, cocoa beans, nut and seeds (Sedlacek *et a*l., 1995). It causes damage by feeding on the germ and spins silk webbing which they attach to the food substrate forming a loose tunnel in which the develop. Webbing and frass produced are nuisance factors (cited in Navarro and Noyes, 2000). Contamination caused by webbing is often more serious than the actual damage by consumption (Navarro and Noyes, 2000). Older larvae may leave the food and wander to find a pupation site in cracks, nooks, and crevices. The adults are short lived and do not feed (CABI, 2006).

2.5 Management of stored product pests

Stored product insect pest population under favourable environmental conditions and availability of food tend to increase exponentially and this eventually leads to substantial losses in the stored produce (Talukder, F.A., 1995). Throughout history, man has employed a variety of preventive and curative measures against pests of food, designed either to prevent infestation or to inhibit pests in their development by repelling or destroying them (Zehrer, 1994).

The basic requirement or effective pest management strategy is a good knowledge of the pest (example biology, ecology and population dynamics), its host and environment in which the pest lives. These can be manipulated to make conditions unfavourable for the development of the pest (Talukder, F.A., 1995). Protection of stored produce by judicious application of pesticides, in conjunction with the use of improved warehouse sanitation, and other physical methods, host –

plant resistance and biological control methods invariably remain a vital factor in reducing losses during storage (Owusu *et al*., 2008).

2.5.1 Physical Control

This involves the manipulation of the physical environment of the pest to make it inimical for its growth and survival (Fields and Muir, 1995). By so doing, the insect population does not increase, it is reduced and eliminated. This physical attributes relates to temperature, relative humidity, and moisture content of grains, storage structures, packaging, forces in commodity (Compression and impaction), irradiation and the use of inert dusts.

Stored product insects have been controlled using physical means for thousands of years. In the Neolithic times in Africa, this method was employed in the Nile Delta by placing seeds to be stored in glass jars underground to keep them cool and dry (Fields and Muir, 1995,).

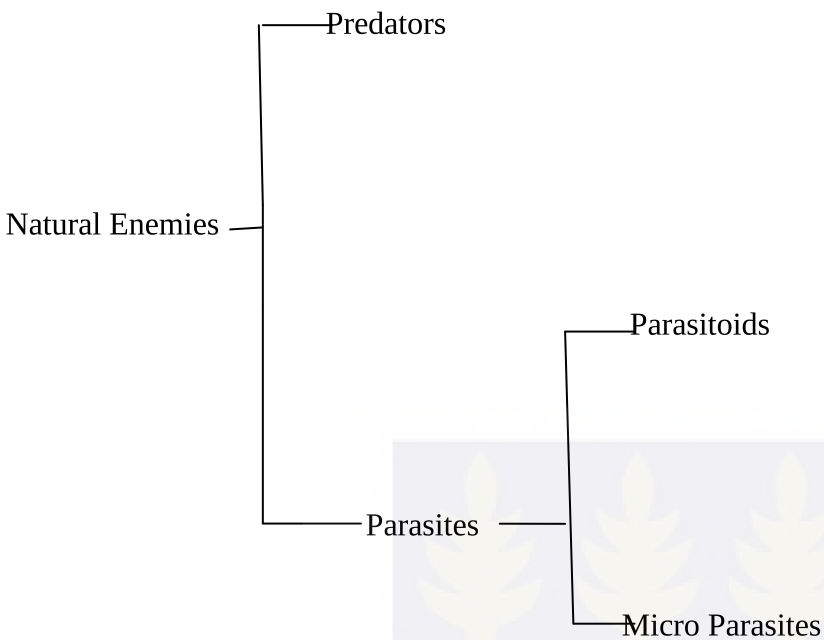
One aspect of physical control mostly used in stored product protection is temperature regulation. Some insects are susceptible to high temperatures while others to low temperatures. Some are also susceptible to extreme high and low temperatures. For instance in general *Ephestia cautella* is most susceptible to very low and very high temperatures.

One other aspect of physical control deals with controlled or modified atmospheres. Examples are the underground storage in Egypt and grains stored in sealed containers in most African countries example Senegal. The term usually refers to the process of changing the atmosphere of a facility by introducing carbon dioxide or nitrogen and it is expected that an environment will be created which will not support the growth and development of the insect. The Hermetic storage of grians is one form of modified atmospheres (Obeng-Ofori, 2008).

2.5.2. Biological Control

All insect populations tend to increase exponentially as long as there is adequate food and suitable environment, and no predators or parasites (Fields and Muir, 1995). Biological control employs the use of natural enemies such as parasites, predators or pathogens to suppress pest populations. Natural enemies can be classified into types based on their history, ecology and population dynamics (Brower *et al*., 1995).

Predators generally are organisms that capture another and feed on the captured organism during their life time.



Parasites are generally smaller than their hosts and can be classified into parasitoids and microparasites. Parasitoids are insects whose immature stages develop as a parasite on or in another insect. Microparasites are microbial pathogens such as viruses, bacteria, fungi, protozoa which cause contagious diseases in target pest (Brower *et al*., 1995).

Examples of organism used as biological control agents include: *Teretrus nigriscenes* Lewis has been effectively used to control *P. tuncatus* in most countries. Females of the parasitoid wasp Bracon hebetor Say seek out and sting wandering-stage larvae of Pyralid moth pests such as the Indianmeal moth and the Mediterranean flour moth (Brower *et al*., 1995).

Females of the Pteromalids *Anisopteromalus calandrae* (Howard) and *Choetospila elegans* Westwood have been observed to forage through stored grains, select a kernel that contains a larva or pupa of a grain beetles (Brower *et al*., 1995). Pirate bugs such as the warehouse pirate bug, *Xylocoris flavipes* (Reuter), and the larger pirate bug, *Lyctocoris campestris* (Fabricus) are generalist predators whose adults and nymphs feed on any life stage of pests that can be subdued (Billups, 1980). *Trichrogramma* species lay their eggs in the caterpillar of moths e.g. *C. cephalonica* and *Ephestia spp*. (Obeng-Ofori, 2008).

Insect pathogens (microbial agents) are facultative pathogens that kill by means of insecticidal proteins (toxins). Infections are acute and cause rapid mortality. For example, formulations containing *Bacillus thuringiensis* (Bt) kill the caterpillar stage of a wide array of moths such as *P. interpunctella, Ephestia spp., C. cephalonica* as well as the lesser grain borer *R. dominica* (Taura *et al*., 2004)

2.5.3 Chemical control

Insecticide use under many circumstances is the easiest, most rapid and most economical method of controlling insects. However, according to Obeng-Ofori (2006), all insecticides used on or near stored food must meet certain criteria and intense regulatory review to ensure human safety.

An ideal insecticide must rapidly kill the insect without killing non target organisms, must be easily degradable with low residual activity, must be easily handled and prepared with low mammalian toxicity, must be cheap and readily available for the farmer to use (Boateng and Obeng-Ofori, 2008).

Currently, the classes of contact insecticides used on or near stored products include organophosphates, carbamates and synergized pyrethroids. These synthetic insecticides are available in a wide variety of compounds and formulations and are applied using different application methods including spraying, fumigation, dusting, smoking, fogging, sparkling and evaporation (Billups, 1980). These insecticides may cause neurological poisoning, desiccation, suffocation or other complex physiological abnormalities in the target pests (Obeng-Ofori, 2008).

The organophosphates (OPs) act by inhibiting the enzyame cholinesterase, thus preventing new impulse transmission resulting in eventual death of the insect (Walker, 1994). An OP insecticide Pirimiphosmethyl (Actelic) with effective dose as 10g ai/tone of grain is a broad spectrum insecticide with a remarkable knockdown effect, persistent for several months and is used in the control of stored product moths and beetles. Other OPs used include Etrimfos, Chloropyrifosmethyl (Reldan), Methacrifos (Damfin) (Obeng-Ofori, 2010).

The synthetic pyrethroids including Bioresmethrin have low toxicity and their effect can be reinforced by the addition of a synergist piperonyl butoxide. It is applied at the rate of 1.5cm^3

ai/tone of grain and it acts by disrupting transmission of nerve impulse, by contact, respiratory effect as well as poison.

Other potential grain protectants include permethrin, diazinon, etrimphos, fenvalerate, iodofenphos, methacrifos, phenothrim, phoxin and tetrachlorvinphos (Obeng-Ofori, 2010). Sometimes two or more insecticides are combined to control certain stored product pests. Although chemical control is the fastest method of pest control, problems of insecticide residues which may be detrimental to the consumer, build up insecticide resistance by the pest, as well as the destruction of non target pests are a major draw backs to its use. However, if appropriate application techniques are employed these problems may be reduced (Koomson, 2003).

2.5.4. Hocklicombi as a grain protectant

Hocklicombi, one of the insecticides under evaluation is a binary insecticide and its active ingredients are fenitrothion and fenvalerate. Fenitrothion is a contact insecticide and selective acaricide of low ovividal properties (Spencer, 1981). It belongs to the organophosphate family of insecticides (Meister, 1994) and considered as a cholinesterase inhibitor. It is effective against a wide range of pests including penetrating, chewing and sucking insects on cereals, cotton, orchard fruits, rice, vegetables and forests. It is also used as an insecticide in public health for the control of household insects (Tomlin, 2005). It is formulated as dust, emulsifiable concentrate, flowable, fogging concentrate, granules, ULV, oil based liquid spray and wettable powder (Meister, 1994). Fernitrothion is available as 95% concentrate, 50% emulsifiable concentrate, 40% and 50% wettable powder and 2%, 2.5%, 3%, and 5% dusts (Hayes and Law 1991). Fenitrothion can be applied as disinfectant of stores and for treatment of stored products. It has a broad-spectrum effect against several insects. It penetrates the grain minimally so that the deposits are mostly removed either with the husk or bran.

Fenvalerate is a pyrethriod insecticide of moderate mammalian toxicity. It acts on the nervous system of insects and disrupts the function of neurons by interaction with the sodium channel (WHO, 1996). It is a non-systemic insecticide and acaricide with contact and stomach action. Fenvalerate is used to control a wide range of pests including those resistant to organochlorines and organophosphorus and carbamates insecticides (Tomlin, 2005). Species that can be controlled include chewing, sucking and boring insects in fruits, vines, olives, nuts, vegetables, cereals and forestry. It is formulated as emulsifiable concentrates, flowable, UVL, wettable powders, slow release formulations, insecticidal fogs, granules and dusts (WHO, 1996)

2.6 The use of botanicals in stored product protection.

Different plant products (leaves, ash, seeds, essential oils, plant oils etc) are mixed with different food stuffs to protect them against insect pest damage (Schmutterer, 1985); Hassanali *et al*., 1990; Obeng-Ofori *et al*., 1997).

The most widely studied plant for this purpose is the Neem, *Azadirachta indica* A. Juss which is potent against several species of insect and mite pests (Schmutterer, 1985). Other plants that have shown promise for the control of storage pests include the siam weed, *Ocimum* plant species, Mahoghany tree, Candlewood, Jathropha, etc. (Bekele *et al*., 1997; Obeng-Ofori and Akuamoah,2000; Udo, 2000).

Since these naturally occurring botanicals are usually biodegradable and not toxic to plants and animals, they offer the potential for safe and effective control of stored product pests (Rembold, 1994). More than 30,000 secondary metabolites have been reported from plants possessing major group of compounds with insecticidal activity being alkaloids, amines, non-protein amino acid, cynogenic glycosides, glucosinolates lectins, protease inhibitors (Wink, 1988). Other allelochemicals are monoterpenes,sesquiterpenes, diterpenes, triterpenes/steroids,tetraterpenes, polyketides, polycetylenes, flavonoids and phenylpropanoids (Wink, 1993).

Boeke *et al*. (2004) repoted that in a survey conducted in Benin West Africa, out of 33 plants collected and tested the powders of *Nicotiana tabacum* L, *Tephrosia vogeliii* Hook F and *S. longepedunculata* significantly reduced progeny production of *C. maculatus* in stored cowpea while *Clausena anisata* (Willd) Hook, *Dracaena arborea* L., *T. vogelii*, *Momordica charantia* L. and *Blumea aurita* Linn f. were repellent to beetles.

In a similar survey, plants used as traditional insecticides in 12 districts in forest areas of Ashante Region of Ghana involving 500 farmers, it was found that 26 different plant species were found to be used as grain storage protectants (Cobbinah *et al*., 1999). The most common were *Chromoleana odorata* L., A. *indica* and *Capsicum annum* L.

Another plant found to be commonly used by subsistence farmers as dry powder and admixed with grains in the Northern region of Ghana to protect stored cowpea, Bambara groundnut, millet, sorghum and maize was *Cassia* species (Belmain *et al*., 1999; 2001). Niber (1994) also investigated the bioactivity of 10 indigenous plant species reputed to have both medicinal and insecticidal properties by local herbalists in Ghana against *P. truncatus* and *S. oryzea.*

Based on the above research in Ghana, Belmain *et al*. (2001) reported 16 different plant species the Ministry of Food and Agriculture of Ghana has adopted to be used by farmers for stored product protection in Ghana. (Table 1.1)

Table 2.1: plant species adopted by the Ministry of Food and Agriculture of Ghana to be used by farmers for stored product protection in Ghana (Belmain *et al*., 20001).

Azadirachta indica (Neem tree)	9. *Ocimum Americana* (American Basil tree)
Capsicum annum (Chilli pepper)	10. *Pleiocapa mutica*
Cassia sophera (Coffee pod)	11.*Pterocarpus erinaceus* (Barwood)
Chamaecrista nigricens (Partrige pea)	12. *Securidaca longependunculata* (African Violet tree)
Citrus sinensis, (Sweet orange)	13. *Synedrella nodiflora* (Cenderella weed)
Cymbopogon schoenanthus (Lemon grass)	14. *Chromolaena odorata* (Siam weed)
Khaya senegalesis (Mahogany tree)	15. *Vitellaria paradoxa* (shea butter tree)
Lippia multiflora (Bush tea)	16. *Mitragyna inermis.* (False abura)

2.6.1 **The use of plant parts**

Different parts of several plants show various levels of bioactivities against stored product insect pests. Many part of the neem tree including the leaves, bark and seed are used for plant protection purposes in many parts of the tropical world (Belmain *et al*., 2001). The seed kernels are rich in oil which even at extremely low concentration as a water emulsion, deter insect larvae from feeding and interfere with reproduction, growth and development (Addae-Mensah, 1998).

In some cases groundnut and coconut oils are mixed with grains as protectants before storage, thus protection is offered by inhibiting and damaging the eggs of the pests (Zehrer, 1994). Jotwani and Sircar (1994) found that powdered neem seed kernels when mixed with wheat seeds at the rate of 1-2 parts per 100 parts protected stored wheat against *S. oryzea*, *R. dominica* and *Trogoderma granarium* Everts for at least 379, 321 and 269 days, respectively. In another study it was found that coarse powder of crushed neem seed kernels mixed at the rate of 1-2% with different pulses gave protection against the bruchid, *C. maculatus* for 8-11 months (Jotwani and Sircar, 1997).

2.6.2 The use of solvent extract of plant parts

Crude extracts from these plants have also been used against stored product pests. The use of jute bags impregnated with 10% aquous extract from *Chenopodium ambrosiodes* L. were found to be effective in reducing infestatation by *C. maculatus* for more than months (Zehrer, 1994).

Owusu *et al*., (2007) reported that out of four solvent extracts investigated from the roots of *Z. xanthoxyloides* the methanol extracts cause 100% mortality of *S. zeamais* and *C. maculatus* in the laboratory. Yadava (1973) found that 2 and 4 % emulsion of essential oil of *Acorus calamus*

L. in kerosene oil, water and absolute alcohol were effective against *Callosobruchus chinensis* (L).

Shehu (2009) reported that 3 ml/L, 7 ml/L of Calneem oil (derived from the neem tree *A. indica*) reduced the number of grains that were damaged by *C. maculatus* and *E. cautella* (Walker) infestation and caused repellency of 50-70% against them. Shay and Ikan (1980) found that the fractionated cotton seed oil possessed insecticidal activity against *C. chinensis* when applied at a concentration of 400 g/ton of stored chick peas. The fractions were also active against *S. oryzae* on stored wheat.

2.6.3 Classification of Botanicals

On the basis of physiological activities on insects, Jacobson (1982) conventionally classified the plant components into six groups, namely; repellents, feeding deterrents/anti feedants, toxicants, growth retardant, chemisterilants and attractants. However, the bio-potential plant products were also classified by Talukder (2006) as follows (Table 2.2).

Table 2.2: A list of some botanicals and their physiological effect on insects (Talukder, 2006).

Common Name/Local name	**Scientific Name**	**Physiological Effect**	**Form in which it used**
Neem	*Azadirachta indica*	Tox, Antf, Rep.	Extracts of all parts
AfricanVoilet tree/Palaga	*Securidaca longependuculata*	Tox, Antf, Rep.	Water from soaked roots, powdered root or bark.
African Basil/ 'Akoko besa'	*Ocimum canum*	Tox	Powdered form of leaves, methanol extract.
'kulenka'	*Cybopogon schoenanthus*	Rep	Whole/powdered flower heads or entire plant.
Lodel	*Chamaecrista nigricens*	Tox, Antf	Powdered leaves
Lidikonja	*Lippa multiflora*	Antf	Powdered whole leaves and/or flowers
Kimkim	*Synedrella nidiflora*	Antf	Water from boiled leaves/whole plant powdered leavs
Tikublaakum	*Cassia sophera*	Tox, Antf	Powdered leaves
Garlic	*Allium sativum*	Rep, Tox	Oil and powdered form of leaves
Candle wood	*Xanthoxylum xanthoxyloides*	Tox, Rep, Antf	Powdered form of bark

NB: Tox: Toxicant, Antf: Antifeedant, Rep: Repellent

2.6.4. Repellents

According to Dethier *et al*. (1960) an insect repellent is a chemical stimulus, which causes the insect to make oriented movements away from the source of stimulus. These are desirable chemicals as they offer protection with minimal impact on the ecosystem, as they give the insect-pest from the treated materials by stimulating olfactory or other receptors of insects (Talukder, 2006). Repellents from plant origins are considered safe in pest control operations as they minimize pesticide residues; ensure safety of the people, food, environment and wild life (Khan, 1982; Talukder and Howse, 1995; Talukder *et al*., 2004). The essential oil of *Artemisia annua* L. was found as repellent against *T. castemneum* and *C.maculatus* (Tripathi *et al*., 2000).

2.6.5. Feeding deterents/Antifeedants

Antifeedants are defined as chemicals that inhibit feeding, although they do not kill the insect directly (Munakata, 1997). Saxena *et al.*, (1988) defined antifeedants as chemicals, which retard or disrupt insect feeding by rendering the treated material unattractive or unpalatable. Antifeedants are of great value in protecting stored commodities from insect. Insects remain on treated food indefinitely and eventually starve to death without eating (Munakata, 1997; Talukder and Howse 1994; Talukder and Howse, 1995; Talukder., *et al*., 2004),

Some naturally occurring antifeedants, which have been characterized, include glycosides of steroidal alkaloids, aromatic steroids, hydroxylated steroid meliantriol, triterpene hemizectal etc.

(Nowrot *et al*., 1986; Jacobon 1990; Klocke and Kubo, 1991; Kubo and Nakanishi, 1997; Talukder and Howse 2000).

The screening of several medicinal herbs showed that the root bark of *Dictamnus dasycarpus* Turcz possessed significant feeding deterrence against two stored product insects, (Liu *et al*., 2002).

2.6.6. Toxicant

These are specific type of chemicals, which directly kill insects. Worldwide reports on the toxicity of different plant derivatives showed that many plant products are toxic to stored product insects (Weaver et al., 1994; Xie *et al*., 1995; Obeng-Ofori and Reichmuth, 1997). Talukder (1995) listed the use of 43 plant species expressing toxicant effect on different species of stored product insects. Pascual and Robledo (1998) carried out screening of plant extracts from 50 different wild plants species of South-eastern Spain for insecticidal activity towards. *T. castaneum* and reported that four species namely, *Anabasis hispaica* Pau, *Senecio lopezii* Boiss, *Bellardia trixago* L. and *Asphodelus fistulosus* L. were found most bioactive.

Two major constituents of the essential oil of *Allium sativum*, methyl allyl disulfide and dially trisulfide were found as potent contact toxicant, fumigant and feeding deterrent against *S. zeamais* and *T. castaneum* (Huang *et el*., 2000). The essential oil vapours distilled from eucalyptus, oreganus and rosemay was reported to have caused 100% mortality of the eggs of *T. confusum* and *Ephestia kuehniella* (Tunc *et al*., 2000).

2.6.7. Grain Protectants

From the very early time, plant materials have been used as a kind of natural protectants to stored grain. Neem plant parts such as the leaves, crushed seeds, powdered fruits, oil etc. are most traditional examples in this regard (Jotwani and Sircar, 1967; Talukder *et al*., 2004). In parts of Eastern Africa, leaves of some plants have traditionally been mixed as grain protectants (Hasanali *et al*., 1990).

Worldwide reports show that when mixed with stored grains, leaves, barks, seed powder or oil extracts of plants reduced oviposition rate and suppressed adult emergence of stored product insects, and also reduced seed damage (Talukder and Howse, 1994; Onu and Aliyu, 1995; Keita *et al*., 2001; Talukder *et al*., 2004).

2.6.8. Reproduction inhibitors

Ground plant parts, extracts, oil and vapour also suppressed fecundity and fertility of many insects (Tunc *et al*., 2000). Many researchers reported that plant parts, oils or extracts mixed with grains reduced insect oviposition, egg hatchability, post embryonic development and progeny production (Ivbijaro, 1983; Saxena and Yadav, 1984; Schmidt *et al*., 1991). Reports also indicated that plant derivatives including the essential oils caused mortality of insect eggs (Tunc *et al*., 2000)

2.6.9. Insect growth and development inhibitors

Plant extracts showed deleterious effects on the growth and development of insects and reduced larval, pupal and adult weight significantly, lengthened the larval and pupal recovery and adult eclosion (Khanam *et al*., 1990). The crude extracts also retarded development and caused mortality of larvae, cuiticle melanisation and high mortality in adults (Jamil *et al*., 1984). It was reported that grains coated with plant extracts completely inhibited the development of *S. oryzae* (Rajasekaran and Kumaraswami, 1985). Plant derivatives have also been reported to reduce the survival rates of larvae and pupae and adult emergence (Tripathi *et al*., 2000). Development of eggs and immature stages inside grain kernels were also found to be inhibited by plant derivatives (Obeng-Ofori *et al*., 1997).

2.7. Control of *Ephestia cautella*

Infestation in store is minimized by cultural control and sanitary methods, biological and chemical control (CABI, 2006). Infestation can be reduced by storage of good quality grains such as whole cereals with fewer broken grains and foreign matter and by ensuring good hygienic measures.

The use of protectant insecticides has been the predominant control strategy (Arthur, 1989). In Ghana, stored product protectants used include pirimiphos-methyl, permethrin, fenitrothion and fenvalerate. However, the use of residual insecticdes is now limited because of pest resistance to insecticides and other adverse environmental and health hazards (Obeng-Ofori, 2007). Other measures such as fumigation with phosphine (CABI, 2006) have been used to control *E. cautella*. Also, the use of fumigants is becoming more restricted because of problems with environmental contamination. Other control strategies which have been tried but have not been widely used commercially include modified atmospheres, irradiation, low temperatures, high temperatures, biological control, mating disruption with pheromones and host plant or commodity resistance (cited in Throne *et al*., 1998).

Currently, Tutuncu *et al*. (2007) has recommended the use of modified atmospheres for the control of *E. cautella* in stored products such as dried figs. In their studies, carbon dioxide (CO2) application was very effective against the almond moth within short exposure times. Finkelman *et al*. (2004) also evaluated the effect of low pressure on the survival of *E. cautella*, *Plodia interpunctella* Hubner (Lepidoptera:Pyralidae) and *T. castanum*. Their results showed that the use of low pressure allows the control of the insect pests at shorter exposure intervals without the need for toxic chemicals.

Irradiation has also been recommended as a safe method for food preservation and as suitable alternative to chemical fumigants (Ozyadimici *et al.,* 2006; Faruki *et al.,* 2007). Ozyadimici *et al.*

(2006) investigated the inhibition of egg hatcing and development stages of the Indian meal moth and the almond moth by irradiation in hazelnuts. Irradiation doses of 450 and 300 Grays were found to inhibit the development of eggs in both species, respectively. Also, no adult of either species emerged from the treated samples. Faruki *et al.* (2007) evaluated the effect of ultra violet (254 nm) irradiation on egg hatching and adult emergence of the flour beetles, *T. castaneum*. *Tribolium confusum* Jacquelin du Val (Coleoptera: Tenebrionidae) and *E. cautella*. The eggs of *E. cautella* were found to be less sensitive to UV rays than *T. castaneum* and *T. confusum*. However, all the exposure periods significantly reduced the emergence of adults in all the experimental insects.

A variety of parasitoid species, including egg parasitoids in the genus *Trichogramma* as well as the larval parasitoid *Habrobracon hebetor* Say (Hymenoptera: Braconidae) have been studied as potential natural enemies for a variety of stored product moth in bulk peanunt storage, bulk wheat storage and bakeris (cited in Grieshop *et al*., 2006).

2.7.1. Insecticides used as grain protectants in Ghana.

In recent times, stored product insecticides have been formulated as binary insecticides with active ingredients consisting of an organophosphorus and synthetic pyrethriod to increase their spectrum of activity (Hoffman *et al.*, 2008). This has come about largely as result of development of resistance by the insects with ensuring reduced efficacy. In Ghana, several of these binary insecticides have been evaluated and used for the protection of stored grains. These include Betallic Super EC, Hocklicombi, super guard, guard, Super dust and Sumbicombi (Boateng *et al.*, 2006).

2.7.2. Hocklicombi as a grain protectant

Hocklicombi, one of the insecticides under evaluation is a binary insecticide and its active ingredients are fenitrothion and fenvalerate. Fenitrothion is a contact insecticide and selective acaricide of low ovicidal properties (Spencer, 1981). It belongs to the organophosphate family of insecticide (Meister, 1994) and considered as a cholinesterase inhibitor. It is effective against a wide range of pests including penetrating, chewing and sucking insects on cereals, cotton, orchard fruits, rice, vegetables and forests. It is also used as an insecticide in public health for the control of household insects (Tomlin, 2005). It is formulated as dust, emulsifiable concentrate, flowable, fogging concentrate, granules, ULV, oil based liquid spray and wettable powder (Meister, 1994). Fernitrothion is available as 95% concentrate, 50% emulsifiable concentrate, 40% and 50% wettable powder and 2%, 2.5%, 3% and 5% dust (Hayes and Laws, 1991). Fenitrothion can be applied as a disinfectant of stores and for treatment of stored products. It has a broad-spectrum effect against several insects. It penetrates the grain minimally so that the deposits are mostly removed either with the husk or bran.

Fenvalerate is a pyrethroid insecticide of moderate mammalian toxicity. It acts on the nervous system of insects and disrupts the function of neurons by interaction with the sodium channel (WHO, 1996). It is a non-systemic insecticide and acaricide with contact and stomach action.

Fenvalerate is used to control a wide range of pests including those resistant to organochlorines and organophosphorus and carbamates insecticides (Tomlin, 2005). Species that fenvalerate can control include chewing, sucking and boring insects in fruits, vines, olives, nuts, vegetables, cereals and forestry. It is formulated as emulsifiable concentrates, flowable, ULV, wettable powders, slow release formulations, insecticidal fogs, granules and dusts (WHO, 1996).

2.7.3 Stored Products

Food products harvested and stored for future use can be classified broadly into perishable and durable stored products (Boateng and Obeng-Ofori, 2008). The perishable stored products cannot be stored for long without deterioration unless they are processed. They have natural storage life of a few days to months depending on the produce. Examples include fruits, vegetables, and root crops.

The durables on the other hand have the inherent ability for long term storage without deterioration and storage life may range from months to several years. Plant products first stored and transported by man were all durables. This includes the cereals, and legumes (Obeng-Ofori, 2008).

2.8 The African violet Tree: *Securidaca longependunculata* Fress

2.8.1. Taxonomy, distribution and ecology

This plant belongs to the family Polygalaceae. It is found in a wide range of climates, from sub-tropical, hot and arid climate to rainfall and equatorial humid regions (Orwa *et al.*, 2009). It also occurs in a broad range of vegetation, from semi-arid scrub to dense forest, including many woodland and bush habitats and gallery forests (Burkill, 1997). In Ghana it is found in the Northern and middle belts of the country. The various local names given to this plant include *'Aforo' or 'Kyirito*' in Twi, *'Kpaliga'* or *'Palaga'* in Dagbani and *'Warramangungumi'* in Hausa.

It is a semi-decidous shrub or small tree that grows to 12 m tall, with an often flattened or slightly fluted bole. It is spiny and much branched, with an open, rather straggly looking crown (plate 2.2). Leaves are either alternative or clustered with small flowers of about 10 mm long and pink to lilac or purple in colour. They are sweetly scented, on long slender stalks and produced in beautiful profusion in terminal axillary sprays 3-5 cm long. Appearing with the very young leaves are 5 unequal bisexual sepals. The fruit is more or less a round nut. Its hatchet like appearance is referred to in the generic name, while the specific name *'longepedunculata'*, refers to the long, slender stalks of the flower (Orwa *et al.*, 2009).


Plate 2.2: The leaves and flowers of the African violet tree *Securidaca longependunculata* Fress

2.8.2. Uses

S. longepedunculata is a very useful plant. It is used for fire wood and production of charcoal in Africa (Bein, 1996). Fine quality fibre is obtained from inner bark of the straight, annual shoots, which when retted could possibly be useful for flax- like textiles (Cmelik & Ley, 1984). The solid portion of the root is said to be the most lethal. In Zambia the crushed and powdered roots are used as intravaginal or intrarectal poison, and in Gambia as fish poison (Storrs, 1995). The bark, roots and seeds are used in arrow poison, and roots can be used as a snake repellant. Roots at 350 ppm are 100% effective as molluscicide (Burkill, 1997). Powder from the burnt roots is rubbed into small incisions made on the temple and forehead to relief headaches (Coates-Palgrave, 1983). The leaves are also used for the treatment of snakebites, veneral diseases and cough; bark is also used against stomach problems and as an arrow poison antidote (Leeuwenberg, 1987).

Research shows that the bark contains methyl salicylate. The presence of saponins also in the bark and crushed seeds give a soapy solution in water and are used as soap for washing or bleaching clothes (Orwa *et al.*, 2009). Seck and Lognay (1998) reported on the efficacy of *S. longepedunculata* at concentrations of 5% and 10% against *C. maculatus* on 'niebe' bean in Senegal.

2.9. African Basil *Ocimum canum* Sims

2.9.1. Taxanomy, distribution and ecology

Ocimum canum belongs to the family Lamiaceae and is an annual plant native to Africa. Two important relatives with these similar properties are the *Ocimum gratissum* (basil), and *Ocimum sanctum* (holy basil).

There are various vernacular names in Ghana for this plant. It known as *'eme'* amongst the Akans, *'Kowe'* or *'danue'* among the Gas and *'ahame'* or *'defetsui'* amongst the Ewes. *O. canum* is a bushy herb which grows to a height of 0.6 m (plate 2.3) and it is common to tropical Africa and other areas of the tropics and was later introduced into the Americas (Guenther, 1952).

It has a characteristic mint flavor with hairy leaves and scented flowers. It is a weed of waste places and villages and usually occurs in both grassland and forest in Ghana (Dokosi, 1998). The plant grows quickly and the seeds sprout easily when placed in warm soils, especially indoors and takes about eight weeks to mature (Thomas *et al.*, 2000).

Plate 2.3: The African Basil *Ocimum canum* Sims

2.9.2. Uses

O. canum is grown for its medical and culinary value. It is highly useful in treating various types of diseases and also in lowering blood glucose especially in type 2 diabetes levels (Dokosi, 1998). It is used as an insect repellent during storage. It also works as a mosquito repellent to prevent malaria and dengue fever (Dokosi, 1998).

The essential oils and leaves are used in flavouring foods, chewing gums, sweets, teas, soft drinks, energy drinks, milk products, cosmetics, shampoos, soaps, shower gels, body lotion and tooth pastes (Dokosi, 1998).

2.10. Candle Wood *Zanthoxylum xanthoxylodes* Lam.

2.10.1. Taxonomy, distribution and ecology

Z. xanthoxyloides belong to the family Rutaceae which contains 1700 species placed in 161genera. In Ghana several vernacular names are used for this plant. The Gas call it 'haatso', Fantes call it 'kanfu', the Ewes call it 'xetsi' and the Akans call it 'okonto'. It is a shrub that can grow to small tree of up to 1.25 m high and 0.13 m girth (Irvine,1961) (plate 2.4). It thrives well in dry and well drained soils (Neumann and Muller-Haude, 1999). The tree is widely grown closed and savanna forest habitats and sometimes in coastal thickets (Nakanishi and Suzuki, 1998).

Plate 2.4: The leaves of Candle wood, *Zanthoxylum xanthoxyloides* Lam.

2.10.2 Uses

This is an important plant with all its parts having many uses. The seed are used for making necklaces in Senegal (Irvine, 1961). The leaves are fed to sheep in certain Ga villages in Ghana and pulverized leaves are sometimes used to flavour food in Cote d'Ivoire (Dalziel, 1937). The pounded roots are sometimes mixed with guinea grass or capsicum peppers and are used to control stomach aches. The bark can be made into poultice and applied against rheumatism and swellings. Udo (2000) reported that the bark and root of *Z. xanthoxyloides* provided up to 100% protection of grains against infestation by *S. zeamais, T. casteneum* and *C. maculatus*. Tablet formulations of the various parts induced about 50% mortality, and offered 95% protection of stored maize and cowpaes against *S. zeamais* and *C. maculatus*, respectively (Koomson, 2003). Application rates ranging from 0.125 to 3 g per 20 g seed, root bark powder of *Z. xanthoxyloides* was used as an effective oviposition suppressant against the cowpea seed bruchid *C. maculatus* (Nakanishi and Suzuki, 1998).

2.11. Horseradish Tree *Moringa oleifera* Gaetn

2.11.1 Taxonomy, distribution and ecology

Moringa oleifera, Lam (*M. oleifera),* also known as *Moringa pterygosperma* Gaertn, is a member of the Moringaceae family of perennial angiosperm plants, which includes 12 other species (Ramachandran *et al.,* 1980). Native of the sub-Himalayan northern parts of India, it is cultivated throuhout tropical and sub-tropical areas of the world, where it is known by various vernacular names with drumstick tree, horseradish tree, and malunggay being the most commonly found in the literature (Ramachandran *et al*.,1980).

2.11.2. Uses

Moringa oleifera is an edible plant (plate 2.5). A wide variety of nutritional and medicinal virtues have been attributed to its roots, bark, leaves, flowers, fruits and seeds. Phytochemical analyses have shown that its leaves are particularly rich in potassium, calcium, phosphorous, iron, vitamins A and D, essential amino acids, as well as such known antionxidants such as β-carotene, vitamin C, and flovonoids (Ramachandran *et al*.,1980).

Plate 2.5: Horseradish Tree *Moringa oleifera.*

CHAPTER THREE

3.0 MATERIALS AND METHOD

3.1 Location of experiments

The experiments were conducted at the Entomology Laboratory of the Crop Science Department, School of Agriculture, University of Ghana, under laboratory conditions of 27±2°C and 55-60% relative humidity, and in the storage Crib of the University Farm, Legon.

3.2 Culturing of *Ephestia cautella*.

The initial stock was obtained from a culture at Ghana Cocoa Board (Quality Control - Tema). The adult of *E.cautella* were cultured on mixed substrate (Plate 3.1) constituting wheat powder, maize flour and glycerol (Rogers, 1970).

Plate 3.1: Culture of *E. cautella*

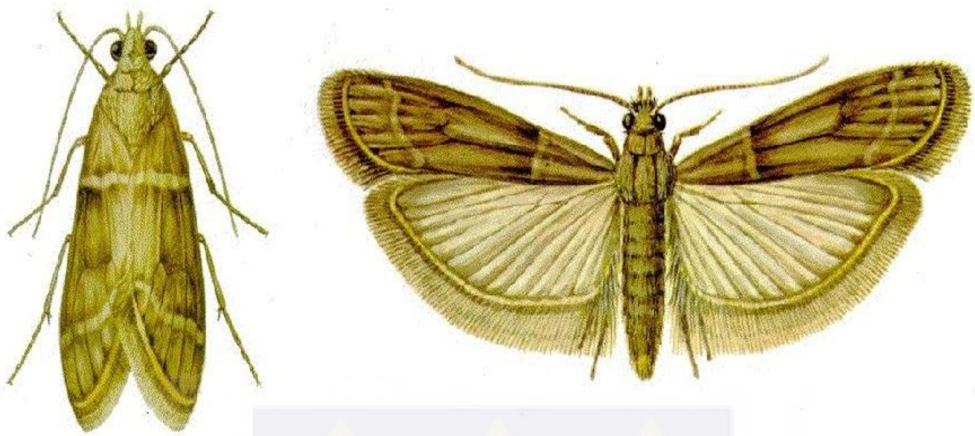


Plate 3.2a: Adult *E. cautella*

Plate 3.2b: 5th instar larva of *E. cautella*

The mixed diet consisted of 8 parts of maize flour to 8 parts of wheat to 1 part of glycerol and 1 part of honey (Allotey and Azalekor, 1999). The maize grains and 1 L kilner jars were sterilized in an oven at 60°C for 3 hours. Maize grains (100 g) were weighed into the kilner jars. Fifty adults of the *E. cautella* were introduced into each jar and left under laboratory condition of 27 ± 2°C and 55-60% relative humidity for 30 days to allow for egg laying and the development of the 5th instar larvae of *E. cautella* (Plate 3.2b). The set up was placed in trays containing industrial oil to prevent the crawling of other insects into the culture.

3.4 Collection of the plant samples

The plant species that were used were obtained from the following places:

Securidaca longepedunculata: Northern Ghana

Zanthoxylum xanthoxyloides: University farms, Legon

Moringa oleifera : Ashalley Botwe, Accra

Ocimum canum: Dome, Accra

3.5 Preparation of plant powders

The leaves were air-dried under shade for 14 days to ensure that they were well dried (Plate 3.2). They were pounded, blended and sieved to obtain fine powders.

3.6 Preparation of plant extracts

Petroleum ether (100%) was used as the solvent for the crude extracts. Four sets of 500 g of the powders of leaves were soaked in 750 mL petroleum ether (100%) and left to stand for two days, filtered and concentrated using the Rotary evaporator. Concentrations of 0.1 g/mL, 0.5 g/mL and 0.3 g/mL of each extract was prepared in acetone and used for the bioassays.

3.7 Repellent effect of the extracts

The repellent effect of the extracts was determined by using the method adopted by Sheehu (2009). Concentrations (0.1 g/mL, 0.5 g/mL, and 0.3 g/mL) of the extracts were applied at 0.2 mL to half filter paper disc; the other half was treated with 100% petroleum ether only as a control. Hocklicombi 25 EC applied at 2ml/L of water was used as reference insecticide. The treated filter was air dried for one hour after which the two halves were joined together and placed in a petri dish.

Ten of the larvae were then placed at the middle of the joined filter papers and covered. There were three replications of each treatment. Larvae on the treated and untreated portions of the filter paper disc were counted after 24, 48 and 72 hours and the repellency determined using the formula adopted by Obeng-Ofori *et al*. (1997).

PR = [(Nc –Nt)/ Nc + Nt)] x 100%;

Where Nc: Number of insect pests on control.

Nt: Number of insects on the extract sides.

All negative PR were treated as zero.

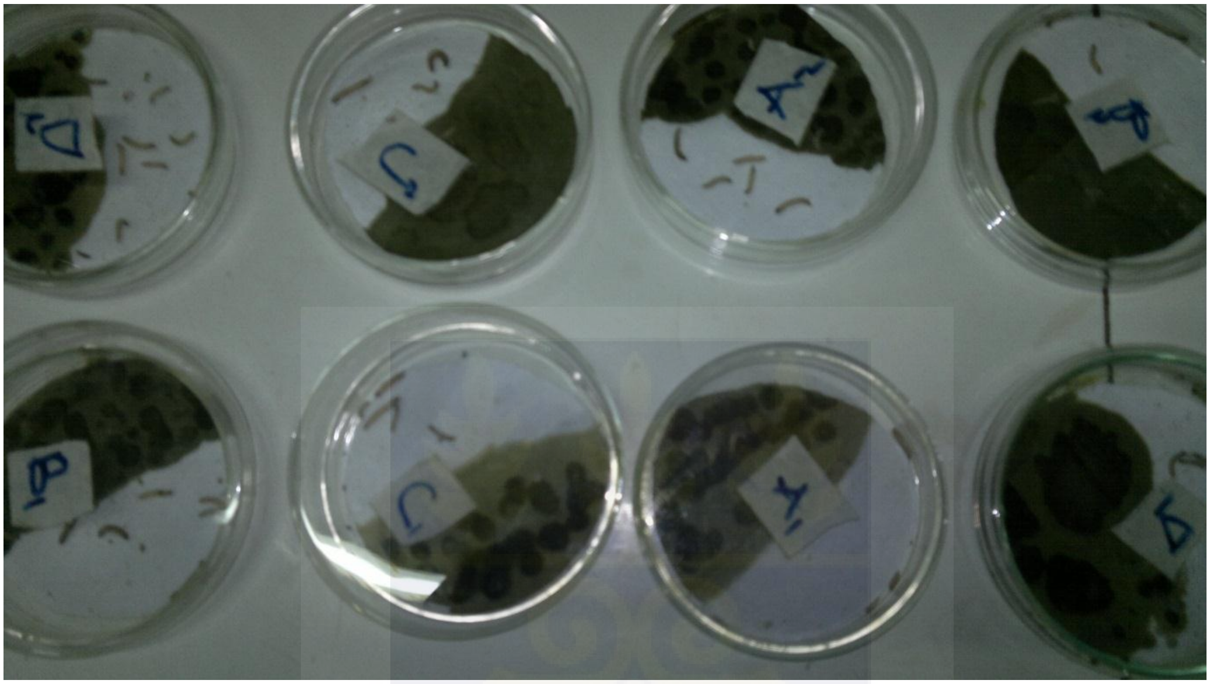


Plate 3.4: Insects being tested for repellence against plant extracts and Hocklicombi

3.8 Bioassays

3.8.1 Effect of the plant powders on adult *Ephestia cautella*

Uninfested, whole maize (2 kg) was sterilized in an oven and 100 g was put into glass jars. The plant powders of *M. oleifera*, *O.canum*, *S. longepedunculata* and *Z. xanthoxyloides* were admixed to it in the proportion, 50% and 100% wt/wt. The negative control however had no plant powders added.

After one hour, a cohort 20 of the adult insects were introduced into the treatments. There were three replications per treatment and these were kept under a temperature of 28±2°C, 75% relative humidity and 12:12, L: D photo regime. Mortality was recorded daily starting from 24 hours after treatment for seven days. An insect was considered dead if it did not respond to probing using a blunt probe.

3.8.2 Effect of petroleum ether extracts on the larvae.

Ten larvae were dipped in turns into the different concentrations (i.e. 0.1 g/mL, 0.3 g/ml, and 0.5 g/ml) of petroleum ether extracts prepared. These were then transferred into clean petri dishes containing maize for feeding. Hocklicombi 25 EC (2 ml/L) of water was used as a standard check whilst Petroleum ether was used as the control. There were three replications of each treatment and these were kept under a temperature of 28± 2°C and 65% relative humidity and 12:12, L: D photo regime. Mortalities were recorded after 24, 48 and 72 hours. Insects were considered dead if they did not respond to probing using a blunt probe.

3.8.3 Effect of extracts on adult insects in treated maize grains

Whole maize grains (1 kg) was weighed. The grains were sterilised and 100 g each was put into glass jars and treated with the different concentrations (0.1 g/mL, 0.3 g/mL, and 0.5 g/mL) of the plants extracts (Plate 3.5). Hocklicombi 25 EC applied at 2 ml/L of water and Petroleum ether were used as positive and negative control respectively. The grains were air-dried for one hour and 20 cohort of adult *E. cautella* were introduced into the treated maize (Plate 3.5).

There were three replications and these were kept under a temperature of 28±2°C, 65% relative humidity and 12:12, L: D photo regime. Mortality was recorded daily for seven days and insects were considered dead if they did not respond to probing using a blunt probe.

Plate 3.5: Insects in treated grains

3.8.4. Effects of extracts on progeny development and adult emergence

The effect of the extracts on progeny development and adult emergence was determined by modifying the method employed by Udo (2000). One hundred grams of the grains were put into separate glass jars and infested with twenty (20) adult *E. cautella* to allow oviposition after treating with the different concentrations (0.1 g/mL, 0.3 g/ml, and 0.5 g/ml) of the plant extracts. Hocklicombi 25 EC applied at 2 ml/L of water and Petroleum ether were used as positive and negative control respectively.

The adult *E. cautella* were removed from the treated grains after two weeks of infestation. The grains then were monitored for 14 days for larval and pupal stages. This follows the duration of the various developmental stages of the insects after Hoges (1986) and Obeng-Ofori (2008). There were three replications of each treatment. Response measured was adult emergence; the number of adults that emerged were counted and recorded.

3.8.5 Contact toxicity by topical application

Fifth instar larvae of mixed sexes were transferred to petri dishes and 0.1 g/mL, 0.3 g/ml, and 0.5 g/mL of the extracts concentrations each was topically applied (1µl) to the natal region of the larvae using a micro applicator. Hocklicombi 25 EC applied at 2ml/L of water and Petroleum ether used as the positive and negative control respectively. Each experimental unit consisted of 10 larvae and was replicated three times. The Completely Randomized Design was used for the experiment and the method used by Boateng *et al*. (2006) was adapted. The treated insect larvae were then transferred into glass petri dishes containing food. These were kept under a temperature of 28± 2°C and 65% relative humidity and 12:12, L: D photo regime. The larvae were examined for mortality after 24, 48 and 72 hours after the treatment. Insects were considered dead if they did not respond to probing using a blunt probe.

3.9 Field experiment

The method used by Boateng *et al*. (2006) was adapted. Maize grains were obtained from the University of Ghana Farm, Legon and sieved to remove all debris. One kilogram samples of maize were treated with 2 mL of *S. longependuculata*, *M. oleifera* extracts at a concentration of 0.5 g/mL and 2 ml/L of hocklicombi which served as a reference. This dosage had proven effective in laboratory experiments. Grains treated with distilled water served as a negative control. Each treatment was replicated three times. The treated grains were infested with thirty (30) adults of *E. cautella* in each polypropylene sack. The sacks were securely sealed by stitching and stored in the grain crib (plate 3.5 and 3.6) at the University farms for 60 days.

Plate 3.6. Crib at University of Ghana farm where grains were stored

After the 60 days, the content of the sacks were sieved. The number of live and dead adult insects was recorded. Germination test was also carried out at end of the storage period to test the effect of the treatments on viability of the maize seeds Data were analysed with analysis of variance Genstat 9.2 (Lawes Agricultural Trust, 2007). Means were separated using the Least Significant Difference at 5%.

3.10. Damage assessment

The damage caused by the insects in the treatments was determined and compared with the control. This was done by counting 100 and 200 grains at random for the laboratory and field trials, respectively. These grains were then separated into number of damage and undamaged grains from the setups in section 3.3 and 3.6. These damaged and undamaged grains were weighed separately for loss to be assessed Damage was assessed by using the method of FAO (1985) as modified by Udo *et al.*, (2004) as follows:

% Weight loss = [(Und – Dnu) / U (Nd + Nu)] x 100%

Where U = Weight of undamaged grains

D = Weight of damaged grains

Nd = Number of damaged grains

Nu = Number of undamaged grains

3.11. Data analysis

Data collected on insect counts were transformed using square root (x + 1) whereas percentage data were transformed using Arcsine transformation. Abbot's formula was also used to adjust the data where there were deaths in control treatment, before analysing with ANOVA in StatView for Windows v5.01 (SAS Institute Inc. Cary, NC, USA) statistical package at p=0.05. Fisher's Protected LSD was used to separate the means after ANOVA.

CHAPTER FOUR

4.0 RESULTS

4.1. Contact toxicity by topical application

All botanical extracts caused larvae mortality as shown in Fig. 4.1. The mean percentage mortality for the treated grains ranged from 29% - 90% and was significantly (P <0.05) higher than the control. Larvae treated with the various botanicals gave no appreciable mortality at lower dosage (0.3 g/ mL) but higher mortality was found with higher dosage of (0.5 g/mL) of the various botanicals, (Figs. 1 and 2). Higher mortality was recorded for *M. oleifera* (83.9, 90%), *O.canum* (77.7, 90%), *S. longepedunculata* (83.9, 90%), and *Z. xanthoxyloides* (77.7, 90%) in day 3 and 4, respectively. It was however observed that mortality increased with increasing dosage and days. There was also significantly (P < 0.05) higher difference in the reference than the botanicals. There were no significant differences between *M. oleifera*, *O.canum*, *S. longepedunculata*, and *Z. xanthoxyloides* as compared to the control.

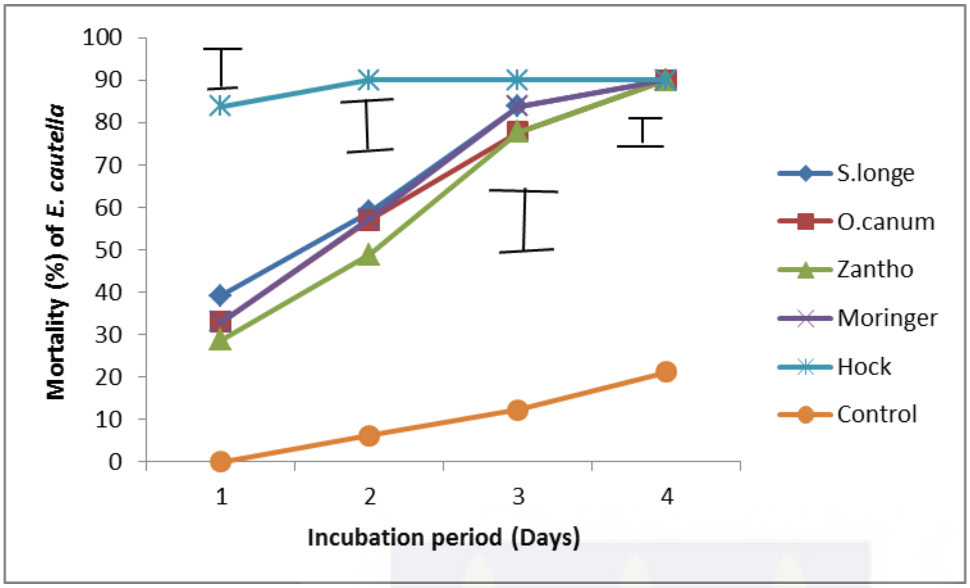


Fig. 4.1: Contact Toxicity (0.5 g/mL) of *M. oleifera*, *O. canum*, *S. longepedunculata*, *Z. xanthoxyloides* to larval *E. cautella* by topical application for 96 hours.

The percentage mortalities of larval *E. cautella* after topical application for 96 and 120 hours are presented in Figure 4.2 and 4.3, respectively. Generally, mortalities caused by the various botanicals increased with increasing number of days as compared to the negative control. However, mortalities were recorded from day 2-4. Again, the control recorded the lowest mortality (0.0%) whilst Hocklicombi which served as the reference recorded higher mortalities (83.9%) right from day 1. This was significantly ($P < 0.05$) different from that of *M. oleifera* (26.6%), *O. canum* (28.8%), *S. longepedunculata* (28.8%), *Z. xanthoxyloides* (28.8%) at 0.3 g/mL and *M. oleifera* (16.6%), *O. canum* (6.1%), *S. longepedunculata* (4.3%), *Z. xanthoxyloides* (8.6%) at 0.1 g/mL and the control (0.0%) in terms of percentage mortality. Also, there were no significant differences between *M. oleifera* (26.6%), *O.canum* (28.8%), *S. longepedunculata* (28.8%), and *Z. xanthoxyloides* (28.8%) as compared to the control (0.0%) and Hocklicombi

(83.9%) where significant (P <0.05) difference was observed. Similar results were recorded in day 2-3, but higher mortalities were recorded for all the treatments except the control.

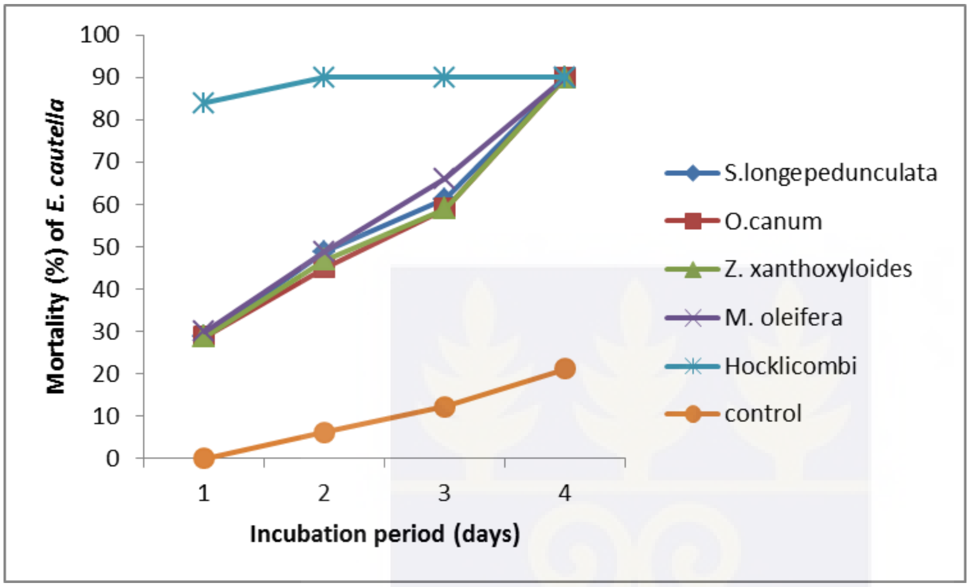


Fig. 4.2: Contact Toxicity (0.3 g/mL) of M. *oleifera*, *O.canum*, *S. longepedunculata*, *Z. xanthoxyloides* to larval *E. cautella* by topical application for 96 hours.

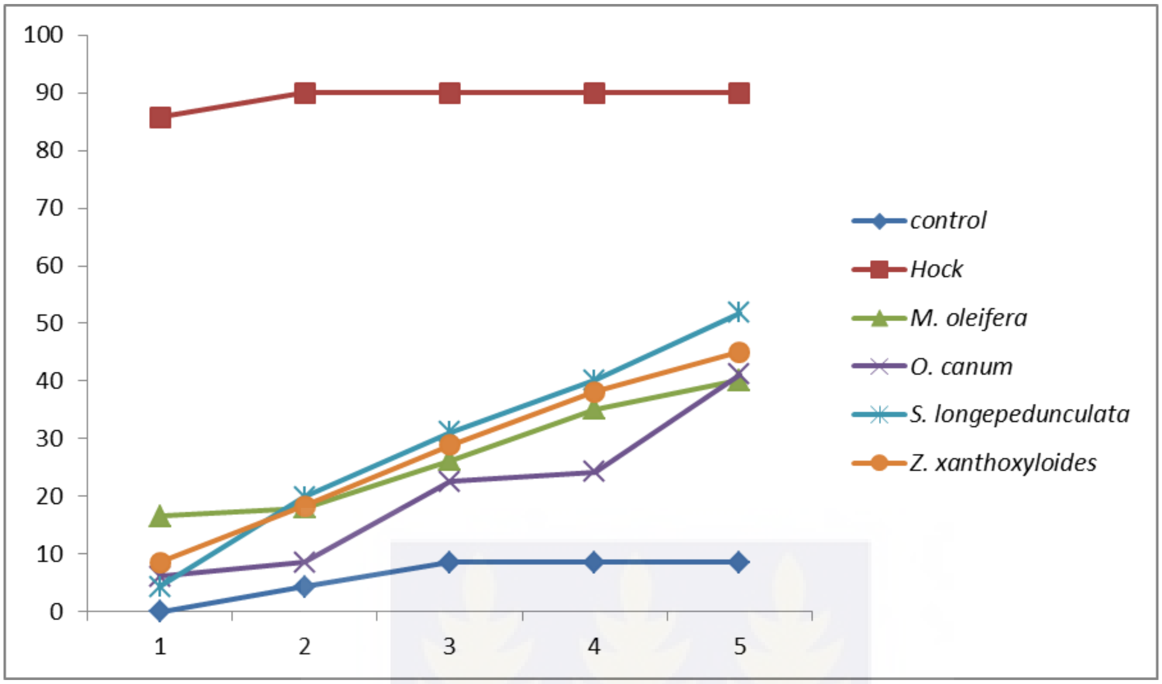


Fig 4.3. Contact Toxicity (0.1 g/mL) of M. *oleifera*, *O.canum*, *S. longepedunculata*, *Z. xanthoxyloides* tolarval *E. cautella* by topical application for 120 hours.

4.2. Effect of extracts on adult *E. cautella* in treated grains

The effect of extract (0.1 g/mL) on *E. cautella* in grains treated with the plant products after 7 days of infestation is presented in Figure 3. Generally, the control recorded the lowest mortality (0.0%), whilst Hocklicombi recorded the highest mortality (81.1%) at day 1. However, Hocklicombi was significantly different ($P < 0.05$) from *M. oleifera* (16.6%), *O.canum* (6.1%), *S. longepedunculata* (4.3%), *Z. xanthoxyloides* (21.3%) and the control in terms of percentage mortality. In day 2, Hocklicombi recorded the highest (90%) mortality which was significantly ($P < 0.05$) the same as the botanicals but different from the control which recorded the lowest percentage mortality. From day 2-7, all the botanical were significantly the same but different from the control.

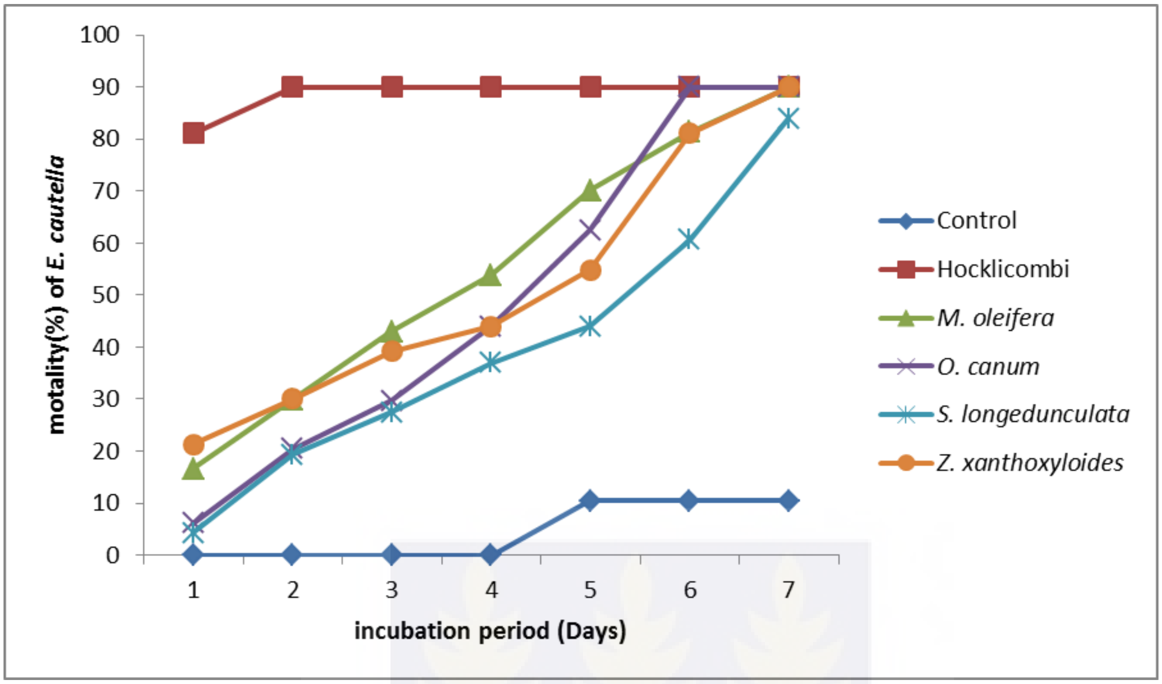


Fig. 4.4: Mortality (%) of adult *E. cautella* after treatment with (0.1 g/mL) *M. oleifera, O.canum, S. longepedunculata,* and *Z. xanthoxyloides* for 7 days of infestation of treated grains.

Mortality of *E. cautella* in grains treated with the plant products after 7 days of infestation is presented in Fig. 4.5. The extracts of *Moringar oleifera* (47.9%), *O.canum* (47.9%), *S. longepedunculata* (43.8%), *Z. xanthoxyloides* (53.8%) were significantly (P<0.05) toxic to the insect when grains were treated with the botanicals compared to the control. This was however different from Hocklicombi. Generally, the control recorded no mortality (0.0%), whilst Hocklicombi which served as the reference recorded the highest mortality (81.1%) at day 1. However, Hocklicombi was significantly different (P < 0.05) from *M. oleifera* (47.9%), *O.canum* (47.9%), *S. longepedunculata* (43.8%), *Z. xanthoxyloides* (53.8%) and the control in terms of percentage mortality. In day 2, Hocklicombi recorded the highest (90%) mortality which was significantly (P < 0.05) the same as the botanicals but different from the control which recorded the lowest percentage mortality. From day 2-7, all the botanical were significantly the same but different from the control.

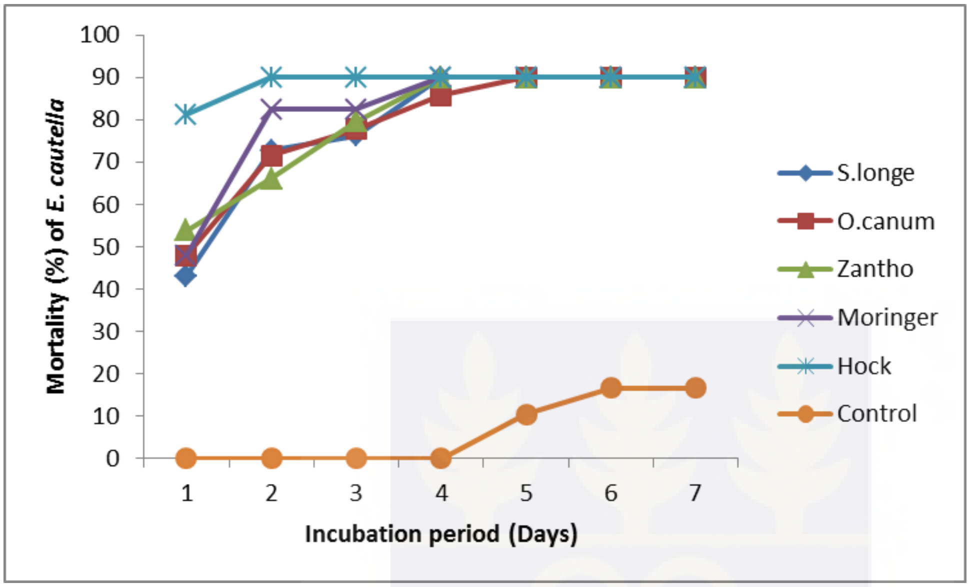


Fig. 4.5: Mortality (%) of adult *E. cautella* after treatment (0.3 g/mL) with *M. oleifera*, *O.canum*, *S. longepedunculata*, and *Z. xanthoxyloides* after 7 days of infestation of treated grains.

The toxicity of petroleum ether extracts of *M. oleifera*, *O.canum*, *S. longepedunculata*, and *Z. xanthoxyloides* at concentration of 0.5 g/mL on adult *E.cautella* in treated grains is illustrated in Fig.4.6 Mortality increased with increasing dosage and days. All treatments caused significantly ($P < 0.05$) higher mortality than the control. The highest concentration (0.5 g/mL) of the botanicals gave the highest mortality of 64.6% and 90% for *M. oleifera* and Hocklicombi respectively in day 1. There were also significant ($P<0.05$) differences between Hocklicombi and the botanicals. Meanwhile, there were no significant differences between *M. oleifera* (64.6, 85.7%), *O. canum* (53.7, 78.1%), *S. longepedunculata* (57.0, 81.4%), and *Z. xanthoxyloides*

(63.5, 85.7%) compared to the botanicals and the control (0.0%) where significant (P<0.05) difference was observed.

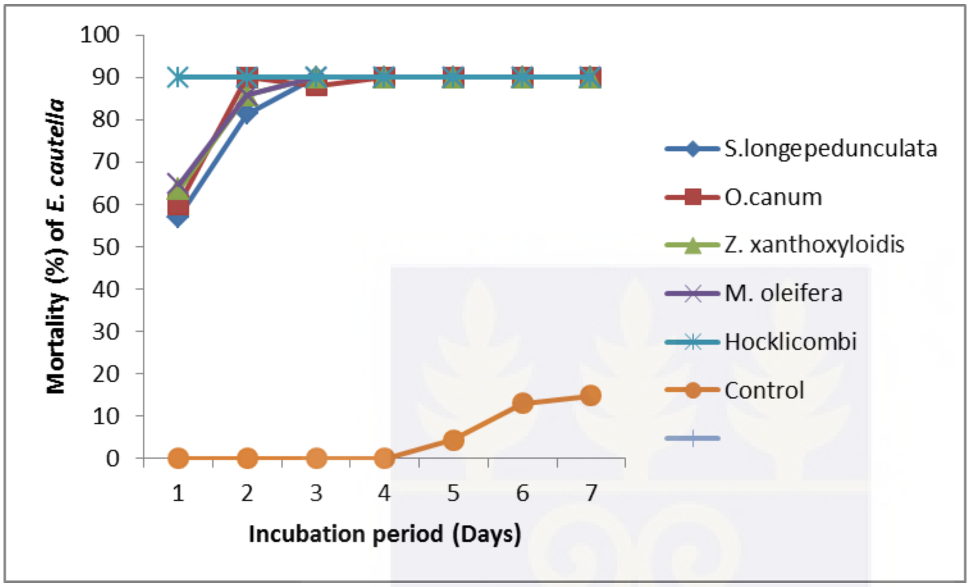


Fig 4.6: Mortality (%) of adult *E. cautella* after treatment with (0.5 g/mL) of M. *oleifera*, *O.canum*, *S. longepedunculata*, *Z. xanthoxyloides* after 7 days of infestation in treated grains.

All concentrations of botanical powders evaluated caused significant mortality of *E. cautella* which ranged from 51.8% to 90% for *M. oleifera*, 46.0 to 90% for *O.canum*, 46.0 for *S. longepedunculata* and 46.9 to 90% for *Z. xanthoxyloides* after 4 days on treated grains. Also, adult mortality observed in all botanical powders was significantly (P < 0.05) different from the control and Hocklicombi where 0.0% and 83% mortality were recoded after day 1 and 4, respectively. However, there was no significant (P < 0.05) difference between the percentage mortality of adult treated with 50% wt/wt of the botanical powders after 7 days of exposure although the highest mortality increased with increasing number of days.

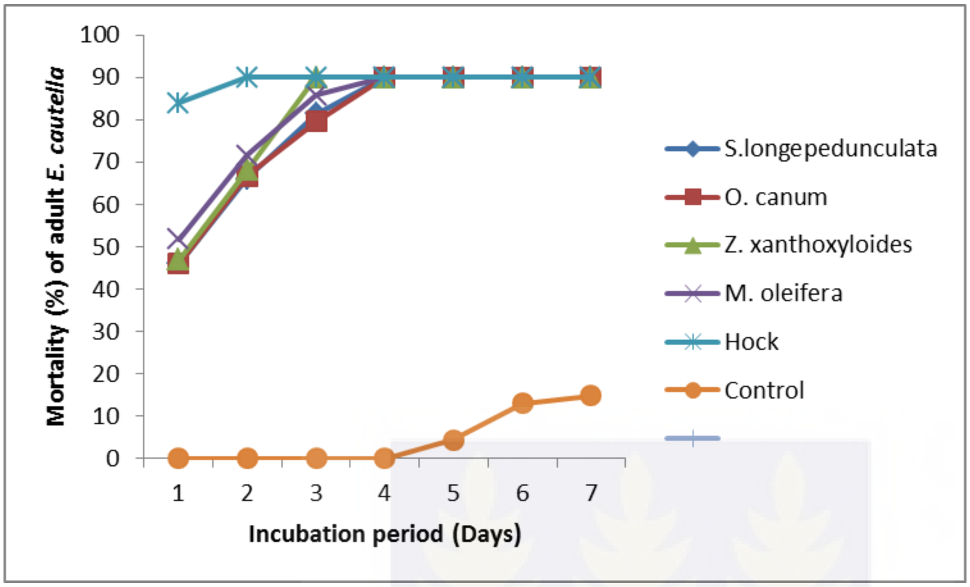


Fig. 4.7: Mortality (%) of adult *E. cautella* after treatment with (50% wt/wt) of *M. oleifera, O.canum, S. longepedunculata, Z. xanthoxyloides* after 7 days of infestation.

Figure 4.8 shows the percentage mortality of adult *E. cautella* in maize grains treated with different botanicals powders at 100% wt/wt. Mortality increased with increasing number of days. All treatments caused significantly (P <0.05) higher mortality than the control. The highest dosage (100% wt/wt) gave the highest mortality within the first 3 days. Again, adult mortality observed in all botanical powders was significantly (P <0.05) different from the control and Hocklicombi where 0.0% and 83% mortality, respectively were recorded.

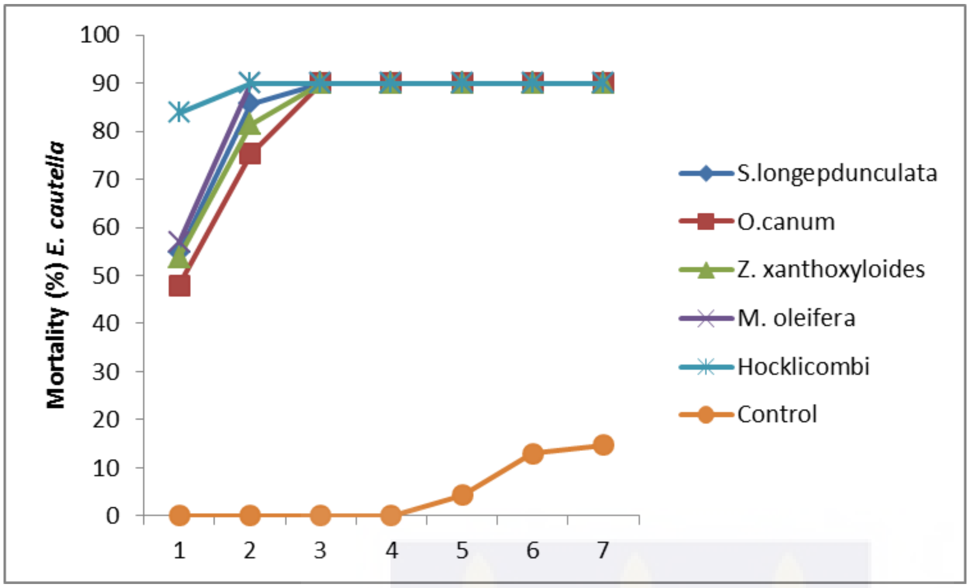


Fig 4.8: Mortality (%) of adult *E. cautella* after treatment with (100% wt/wt) of M. *oleifera, O.canum, S. longepedunculata,* and *Z. xanthoxyloides* for 7 days of infestation.

4.3. Effects of the extracts on immature stages

4.3.1 Effect on eggs

The number of eggs laid by *E.cautella* on grains treated with different levels of petroleum extracts and powders of *M. oleifera, O.canum, S. longepedunculata,* and *Z. xanthoxyloides* are presented in Tables 8 and 9. Eggs laid decreased with increasing levels for all treatments. There were significant (P <0.05) differences between the eggs laid on treated maize grains and the control. There were also significantly fewer number of eggs laid in grains treated with the Hocklicombi than those treated with botanical extracts and powders. No adult emerged from grain treated with 0.3 g/mL 0.5 g/mL for the botanical extracts and powders.

4.3.2 Effect on larvae

The botanical powder and petroleum extracts reduced the emergence of *E. cautella* adults when grains containing eggs were treated with the botanicals (Tables 4.1 and 4.2). There were significant (P <0.05) difference between the treatments and the control but no significant difference between the highest concentration of *S.longepedunculata* and Hocklicombi was observed.

4.3.3 Effect on pupae

The botanical powders and petroleum ether extracts reduced the emergence of *E. cautella* adults when pupae of *E. cautella* were treated with the botanical as compared to the untreated grains (Tables 4.1 and 4.2). There were significant (P <0.05) difference between the treatment and the control, but no significant difference between the concentration of *M. oleifera* at 0.5 g/mL and Hocklicombi was observed

Table 4.1. Effect of petroleum ether extract of *M. oleifera*, *O.canum*, *S. longepedunculata,* and *Z. xanthoxyloides* on the immature stages of *E.cautella*

Treatment	0.1 g/mL			0.3 g/mL			0.5 g/mL		
	Eggs	Larva	Pupa	Eggs	Larva	Pupa	Eggs	Larva	Pupa
Control	43.0	10.0	20.0	43.0	10.0	20.0	43.0	10.0	20.0
Hocklicombi	1.0	0.0	0.0	1.0	0.0	0.0	0.0	0.0	0.0
M. oleifera	7.0	2.0	3.0	2.0	0.0	1.0	0.0	0.0	1.0
O. canum	11.0	2.0	2.0	4.0	0.0	1.0	1.0	0.0	1.0
S.longepedunculata	6.0	3.0	2.0	2.0	1.0	1.0	1.0	0.0	0.0
Z. xanthoxyloides	7.0	3.0	2.0	2.0	1.0	1.0	1.0	1.0	0.0
LSD(0.05)	5.94	2.13	1.67	4.49	1.39	1.39	4.35	1.18	1.25

Table 4.2. Effect of the botanical powders of *M. oleifera, O.canum, S. longepedunculata,* and *Z. xanthoxyloides* on the immature stages of *E.cautella*

Treatment (powders)	Egg		Larvae		Pupa	
	50% wt/wt	100% wt/wt	50% wt/wt	100% wt/wt	50% wt/wt	100% wt/wt
Control	43.0	43.0	10.0	10.0	20.0	20.0
Hocklicombi	1.0	0.0	0.0	0.0	0.0	0.0
M. oleifera	1.0	1.0	1.0	1.0	2.0	1.0
O. canum	2.0	1.0	1.0	1.0	2.0	0.0
S. longepedunculata	7.0	3.0	2.0	1.0	1.0	1.0
Z. xanthoxyloides	13.0	3.0	3.0	1.0	1.0	1.0
LSD (0.05)	4.89	4.49	1.32	1.39	1.39	1.32

4.4. Effects of extracts on adult emergence

The effect of the various extracts and powders on the number of adult insects that emerged after treating the grains with the extracts is summarised in Table 4.3. There was no adult emergence recorded for *M. oleifera* and Hocklicombi which served as the reference whilst at 100% wt/wt of the botanicals, lower emergence was recorded. However, the highest mean adult emergence of 58-60 adults were recorded in the control. There were significant ($P < 0.05$) differences between the treatments and the control. The botanical powders also had a tremendous effect on adult emergence of *E. cautella* (Table 4.3). This ranged from 1.0 to 6 adults at 50% wt/wt and 1.0 to 2 adults at 100% wt/wt as against untreated grains which also ranged from 58 to 60 adults at 100%

wt/wt and 50% wt/wt, respectively. There were also significant ($P < 0.05$) differences between the various treatments and the control.

Table 4.3: F1 progeny from grains treated with petroleum ether extracts and powders of *M. oleifera, O.canum, S. longepedunculata,* and *Z. xanthoxyloides* and incubated for 60 days.

Treatment	**Powders**		**Extracts**		
	50% wt/wt	100% wt/wt	0.1g/mL	0.3g/mL	0.5 g/mL
Control	60.0	58.0	58.7	59.0	59.0
Hocklicombi	1.0	0.0	0.3	0.0	0.0
M. oleifera	1.0	1.0	5.7	1.0	0.0
O. canum	1.0	2.0	9.7	4.0	2.0
S. longepedunculata	3.0	1.0	6.3	1.0	1.0
Z. xanthoxyloides	6.0	2.0	9.7	2.0	1.0
LSD (0.05)	10.19	10.38	9.98	9.81	9.81

4.5. Repellent effect of extracts on insects

The repellent effect of the botanical extracts is summarised in Tables 4 - 6. All botanicals showed effective repellency against the insect, however the higher the concentration of extracts, the higher the mean percentage repellency. The high repellency of 80% was recorded for *M. oleifera* and *Z. xanthoxyloidis*, after 72 hours of exposure. Meanwhile, at same concentration of 0.5 g/mL (Table 4.6) lower percentages of 66.7%, 60.0%, 40% and 17.3% were observed in *S.*

longepedunculata, *O. canum*, Hocklicombi and the control. There were significant (P<0.05) differences between Hocklicombi, the control, and the botanical treatment. At a concentration of 0.3 g/mL (Table 4.5) however, highest repellency were recorded for *M. oleifera* (56%), Z. *xanthoxyloides* (53%) and *O. canum* (50%) as compared to a lower repellency for the control (17.3%) after 72 hours. Generally, there was significantly (P< 0.05) fewer emergence between *M. oleifera, O.canum, S. longepedunculata*, *Z. xanthoxyloides* and the control.

Table 4.4: Mean % repellency of petroleum ether extracts of *M. oleifera, O.canum, S. longepedunculata, Z. xanthoxyloides* against *E. cautella* at 0.1 g/mL concentration.

Treatment	24 hours	48 hours	72 hours
Control	16.7	16.7	16.7
Hocklicombi	36.7	40.0	40.0
M. oleifera	26.7	40.0	46.7
O. canum	26.7	13.3	30.0
S. longepedunculata	20.0	23.3	30.0
Z. xanthoxyloides	20.0	33.3	36.7
LSD (P ≤ 0.05)	22.1	17.29	19.22

Table 4.5: mean percentage repellency of petroleum ether extracts of *M. oleifera*, *O. canum*, *S. longepedunculata*, *Z. xanthoxyloides*, at 0.3 g/mL against *E. cautella*.

Treatment	**24 (hours)**	**48 (hours)**	**72 (hours)**
Control	16.7	16.7	17.3
Hocklicombi	36.7	40.0	40.0
M. oleifera	50.0	40.0	56.7
O. canum	40.0	43.3	50.0
S. longepedunculata	25.0	40.0	43.3
Z. xanthoxyloides	33.3	43.3	53.3
LSD (0.05)	**19.6**	**21.8**	**19.51**

Table 4.6: Mean % repellency of petroleum ether extracts of *M. oleifera*, *O.canum*, *S. longepedunculata,* and *Z. xanthoxyloides* against *E. cautella* at 0.5 g/mL concentration.

Treatments	24 (hours)	48 (hours)	72 (hours)
Control	16.7	16.7	17.3
Hocklicombi	36.7	40.0	40.0
M. oleifera	66.7	66.7	80.0
O. canum	66.7	60.0	60.0
S. longepedunculata	50.0	60.0	66.7
Z. xanthoxyloides	53.3	73.3	80.0
LSD ($P \leq 0.05$)	35.33	25.16	27.70

4.6 Effect of extracts on damage assessment

Damage assessed in terms of percentage weight loss due to the feeding activities of the insect is summarised in Table 6. Grains treated with the botanical powders and extracts reduced damage caused by *E.cautella* significantly ($P < 0.05$) compared with the control. The petroleum extracts from *M. oleifera*, *O.canum*, *S. longepedunculata,* and *Z. xanthoxyloides* proved to be more potent than that of the powders. However, the higher the concentration of the extracts, the less damage recorded in terms of percentage weight loss.

Table 4.7: Weight loss (%) in maize grains as a result of *E. cautella* infestation after treatment with petroleum ether extracts and powders of *M. oleifera*, *O.canum*, *S. longepedunculata*, and *Z. xanthoxyloides* for 60 days.

Treatment	**50% wt/wt**	**100%wt/wt**	**0.1 g/mL**	**0.3 g/mL**	**0.5 g/mL**
Control	9.49	9.49	31.51	9.49	9.4
Hocklicombi	0.35	0.33	0.35	0.35	0.31
M. oleifera	1.71	0.46	3.78	0.82	0.4
O. canum	2.35	2.84	5.65	1.88	1.2
S. longepedunculata	4.89	0.39	6.80	0.80	0.0
Z. xanthoxyloides	3.99	2.23	5.54	0.65	0.07
LSD (P≤ 0.05))	3.46	2.29	7.21	1.52	1.2

4.7 Effect of extracts on maize seed viability

Studies on the effect of petroleum ether extracts and powders of *M. oleifera*, *O.canum*, *S. longepedunculata*, and *Z. xanthoxyloides* on seed viability revealed that the extracts and powders did not hamper seed viability as germination for the treatments ranged from 77.3% to 93.3% for 50% wt/wt and 100% wt/wt and 88.0% to 100% for 0.3 g/mL and 0.5g/mL for the powders and extracts, respectively. The control recorded the least percentage germination of 25.0%. There were significant ($P < 0.05$) differences between the treatments and the control.

Table 4.8: Maize seed germination (%) after treatment with petroleum ether extracts and powders of *M. oleifera, O.canum, S. longepedunculata*, and *Z. xanthoxyloides* for 60 days.

Treatment	**Powders**		**Extracts**	
	50% wt/wt	100% wt/wt	0.3 g/mL	0.5 g/mL
Control	25.0	25.3	25.0	25.0
Hocklicombi	95.0	100.0	95.0	100.0
M. oleifera	84.7	93.3	91.6	100.0
O. canum	87.0	90.0	90.0	97.0
S. longepedunculata	80.0	93.3	92.3	98.3
Z. xanthoxyloides	77.3	86.7	88.3	93.3
LSD (0.05)	5.77	6.48	3.88	3.85

4.8. Effects of extracts on adult emergence on the semi -field trials

The petroleum ether extracts of *M. oleifera, S. longepedunculata*, and the Hocklicombi had a tremendous effect on adult emergence of *E.cautella* (Fig. 4.9). This ranged from 0.3 to 5.0 as against untreated grains which also recorded 66 adults after 60 days of storage in the field. There were significant (P<0.05) differences between the various treatments and the control.

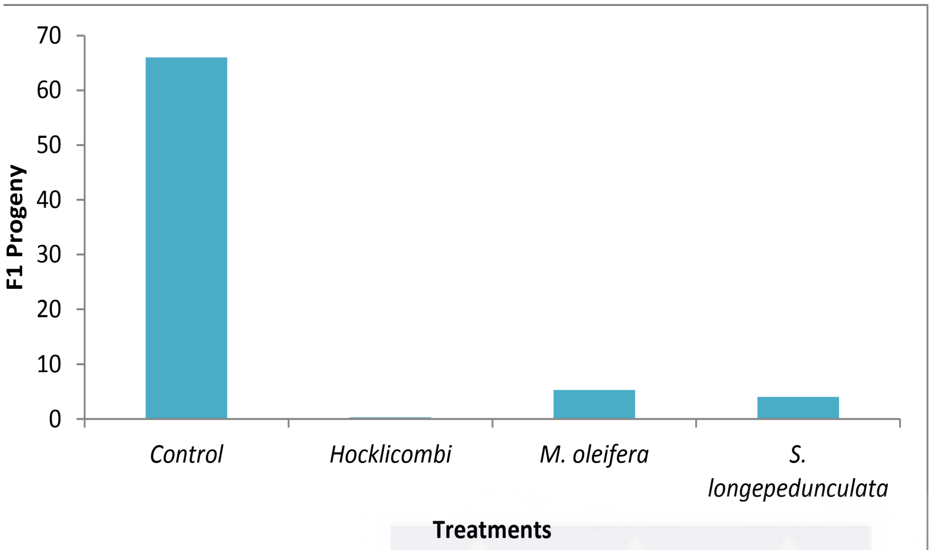


Fig 4.9: F1 progeny of *E. cautella* after 60 days of treatment with *M. oleifera*, and *S. longepedunculata*, in the field.

4.9 Effect of extracts on weight loss (%) in the field

Overall percentage weight loss was lower in the petroleum ether extracts treatments (0.5 g/mL) than that of the untreated control (18.78%) (Table 4.9). Grains treated with *M. oleifera*, *S. longepedunculata*, and hocklicombi in the field gave appreciable reduction in weight as compared with control. All treatments were also significantly ($P<0.05$) different from control in terms of weight loss.

Table 4.9: Weight loss (%) in maize grains as a result of *E. cautella* infestation after treatment with petroleum ether extracts of *M. oleifera*, and *S. longepedunculata*, for 60 days in the field.

Treatment	**Weight loss (%)**
Control	18.78
Hocklicombi	0.42
M. oleifera	0.45
S.longepedunculata	1.76
LSD (0.05)	1.51

CHAPTER FIVE

5.0. DISCUSSION

5.1. Toxicity of the powders

In the present study, the powders of all the four botanical plants. i.e. *M. oleifera, O.canum, Z. xanthoxyloides* and *S. longepedunculata* exhibited various levels of bioactivities against the adult *E.cautella.* In a similar study by Udo (2011), it was reported that the roots, barks and leaves of *Z. xanthoxyloides* were very effective against *C. maculatus and Sitophilus zeamais.* This phenomenon suggests the presence of highly pungent secondary metabolites found in the roots of *Z. xanthoxyloides* (Adesina, 1986). One of the constituents secondary metabolites, Zanthoxylol has been identified as a phenolic compound reputed for insecticidal activity (Elujoba and Nagels, 1985; Wongo, 1988).

Another study by Belmain *et al*. (2001) indicated that the powder of *S.longepedunculata* was very effective against *Rhyzopertha dominica* and *Prostephanus truncatus.* The presence of eugenol which is a major essential oil in the plant as established by the above studies may account for these physiological effects exhibited against the insect (Obeng-Ofori and Reichmuth 1997). Prabhu *et al.* (2011) reported the larvicidal potential of *M. oleifera* against malarial vector, *Anopheles stephensi* Liston. Kamel (2010) also showed that *M. oleifera* was effective in the control of *Spodoptera frugiperda* Smith as it yielded antifeedant and toxic effect against the target insect. These authors indicated the presence of fatty acids and sterols in *M. oleifera* which played a synergistic effect as antifeedants against the insects (Kamel, 2010). Quercetin and Kaempferal are also found in the *M.oleifera* (Prabhu *et al.*, 2011). These are flavonoids and phenol oxides inhibitors which interfere with the bio-chemical processes involved in B-sclerotization, quinone tanning and melanin biosynthesis, which occupies a major role in insect development and immunity such as cuticle sclerotization, wound healing and defence against foreign pathogens (Kubo, 2001).

According to Ntezurubanza 1987, the large amount of linalool in the dried leaves of *O.canum* confirms the earlier reported percentage found in the essential oil of this species. The milled leaves of *O. canum* contain linalool and due to that the insects in the milled leaf trials are rapidly coated with sticky leaf particles (thus due to their movement) that adhere to them. The insects, therefore, received a topical chemical and physical treatment. After the insect becomes intoxicated and fall upon their dorsal surfaces frequently they may also become coated with residual linalool from the surface of the treated grains and hence the higher rate of mortality.

Under the same wt/wt powder treatment of botanicals, the powders of *M. oleifera, S. longepeduncul ata,Z. xanthoxyloides* and *O. canum* have been shown from this study to contain insecticidal, anti-ovipositant and ovicidal properties. Maize treated with the powders of *M.oleifera, S.longependunculata, Z. xanthoxyloides* and *O.canum* induced over 80% mortality in the treated gains. The results obtained suggest good potential for the use of the botanicals in stored product pest management system. The results of this study have established the scientific basis of the practice by farmers in northern Ghana in which the roots of botanicals are pounded and mixed into stored grains. Worldwide reports show that, when mixed with stored grains, leaves, barks, seed powder or oil extracts of plants reduce oviposition rate and suppressed adult emergence of stored product insects and also reduced seed damage (Talukder and Howse, 1994; Onu and Aligu, 1995, Keita *et al.*, 2001; Talukder *et al.*, 2004).

5.2 Toxicity of Petroleum ether extracts of *M. oleifera, S. longependunculata, O. canum, Z. xanthoxyloides* on adult *E. cautella* in treated grains.

The various concentrations of the petroleum ether extracts of *M. oleifera*, *O. canum*, *S. longopendunculata*, and *Z. xanthoxyloides affected* the survival of adult *E.cautella* in the treated grains after a seven day period of observation. The highest concentration yielded the highest rate of mortality of the insect. In another work conducted on some botanicals in Northern Ghana, it was recorded that 100% methanol extracts of *S.longependunculata* roots at 100000 ppm was able to induce close to 100% deterrence against *Sitophilus zeanais* in stored maize (Belmain, 2002) this may be attributed to the toxicant, repellent and antifeedant effect of the plant due to the methyl selicylate present in the roots and bark of the plant (Talukder, 2006). Olifontoye (2010) also recorded 90% mortality inflicted on *Bulimus globosus* at concentration of 10 ppm methanol extracts of *S.longepedunculata* for 24 hours. Taura *et al.* (2004) found that aqueous extract of *S.longependuculata* was active against *Culex* mosquito larvae by direct contact. The higher mortality rate could also be as a result of the toxic properties possessed by the botanicals and this could have arisen from the interference with normal respiration resulting in suffocation (Hall and Harma, 1991).

5.3. Effect of petroleum ether extracts of *M. oleifera, S. longepedunculata, O. canum* and *Z. xanthoxyloides* on adult emergence.

The various botanicals of the petroleum ether extracts reduced the rate of adult *E. cautella* emergence. The higher the concentration of the extracts, the lower the number of insects that emerged. The *M. oleifera* yielded no adult emergence at a concentration of 0.5g/mL while lower emergence of 1-2 adults were recorded for *O. canum*, *Z. xanthoxyloides*, and *S. longepedunculata* at that same concentration. Higher adult emergence ranging from 6-10 adults were recorded at a concentration of 0.1 g/mL and 59 adults for the control. This confirms work done by Tunc *et al*., (2000) who reported that ground plant parts, extracts, oil and vapour also suppressed fecundity and fertility of many insects. Many researchers have reported that plant parts, oils or extracts mixed with grains reduced insect oviposition, egg hatchability, post embryonic development and progeny production (Ivbijaro, 1983; Saxena and Yadav, 1984; Schmidt *et al*., 1991). The inhibition of the emergence of adults in the treated grains at high concentration indicates the presence of high amount of active ingredients which inhibit oviposition and progeny development in the stored grains. It is established that the methyl-salicylate and securidacasidae acted as oviposition deterrent and were toxic to the progenies of *E.cautella* Owusu E.O. (2001). Hubrect *et al.* (1989) reported that *S. longepedunculata* contains saponnins that cause high larval and nymphal mortality in *Spodoptera ferugiperda.*

In addition, the presence of eugenol which is a major essential oil in plants as established by the above studies may account for these physiological effects exhibited against the insect (Obeng-

Ofori and Reichmuth, 1997). The antifeedant, toxicant and repellent properties of the plants also contributed to the low adult emergence in the treated grains.

Again, the presence of the extracts serving as coating material around treated grains could cause fewer eggs to be laid since the laying of eggs is related to site and may also cause the fewer eggs laid not to be firmly attached effectively to the seed surface and hence hatching of first instar larvae may be prevented because penetration is more difficult if egg attachment is less secured. The current study showed that in addition to the mechanical effects, a combination of mechanical and toxic effect could occur to reduce the F1 emergence.

5.4. Repellency of petroleum ether extracts of *M. oleifera, S. longepedunculata,O. canum* and *Z. xanthoxyloides* to the larva of *E. cautella*.

The extracts of *M. oleifera, S. longepedunculata,O. canum* , and *Z. xanthoxyloides* were repellent to *E. cautella* at the different concentrations of the extracts that were used. The highest concentration of 0.5 g/mL of *M. oleifera* and *Z. xanthoxyloides* extract yielded 80% repellency against larval *E. cautella.*

The general repellency of the insect is an indication of the presence of chemicals in the plants that stimulates or causes the insect to make oriented movement away from the source stimulus (Deithier *et al.*, 1960). The high repellency at high concentration is an indication of high amounts of these plant chemicals in the treatment. It has been reported that the roots of *S. longepedunculata* are composed of over 90% methyl salicylate (Jayasekra et al., 2002) which is an ubiquitous aromatic ester with a well-known insect repellent properties (Hardie *et al*., 1994).

Owusu *et al.* (2007) found that methanol extracts of *Z. xanthoxyloides* was an effective repellent to stored product beetles. This was also confirmed by Udo (2011) who established that *S. zeamais* was least repelled by the extracts of roots and bark of *Z. xanthoxyloides* with the highest repellency of 14% observed in dry root and fresh bark extracts. However *Callosobrochus maculatus* was observed to be highly affected by the extracts with overall repellency of 45%.

Adebayo *et al*. (2007) showed that the essential oils of leaves collected in Nigeria are dominated by methyl salicylate (89.6%), major compound of the root bark. Methyl salicylate which is found in the *S. longepedunculata* roots and leaves extracts and the alkanoids and flavonoids with the other secondary metabolites found in the root and leave extracts of *Z. xanthoxyloides* (Adesina, 1986) have been found to be good repellents to these insects and these may account for the repellency observed in the treatments.

In this study, adult *E. cautella* demonstrated clear orientation choices between the volatiles generated by the control, Hocklicombi and the petroleum ether extract of *S. longepedunculata M. oleifera, Ocimum canum* and Z. *xanthoxyloides*. This suggests that *E. cautella* is able to detect active ingredients through olfaction and avoid it when given the choice. This could explain, at least in part, how the application of powdered of *S. longepedunculata*, *M. oleifera*, *Ocimum canum* and *Z. xanthoxyloides* protect grain from insect infestation.

5.5. Damage assessment

Grain damage by *E. cautella* was significantly minimized in all treatments. At higher concentrations of the petroleum ether extracts of *S. longepedunculata M. oleifera*, *Ocimum canum*, and Z. *xanthoxyloides* damage caused were significantly reduced. This could be attributed to the reduction in the progeny and to certain nutritional inhibitors in plant extracts (Dales, 1996). The reduction in F1 progeny could have resulted from ovicidal and larvicidal properties of plants and this could have arisen from the interference with normal respiration resulting in suffocation (Hall and Harma, 1991).The appreciable effect in damage caused is also evidenced by the high repellency observed in the treatments.

Udo (2011) indicates that damage to grain by *S. zeamais* and *C. maculatus* was reduced in grains with treatments which exhibited high repellency to the insect. Worldwide reports show that when mixed with stored grains leaves, barks, seed powder or oil extracts of plants reduce oviposition rate and suppressed adult emergence of stored product insects, and also reduced seed damage (Talukder and Howse, 1994; Onu and Aliyu, 1995, Keita *et al*., 2001; Talukder *et al*., 2004). The active compounds in these plants acted as feeding deterrents and repellent to the insect. These inhibited feeding on the grains by rendering it unattractive or unpalatable to the insect (Saxena *et al*., 1988).

5.6. The Effect of the petroleum ether extract of *M. oleifera, S. longepedunculata, O. canum,* and *Z. xanthoxyloides* on maize seed germination.

Studies on the effect of extracts and powders of *M. oleifera, S. longepedunculata,O. canum,* and *Z. xanthoxyloides* on seed viability revealed that the botanical did not hamper seed viability as germination for the botanical powders ranged from 77.3-87.0% and 86-93.3% (at 50% and 100% wt/wt), respectively whilst that of the petroleum ether extracts ranged from 88.3-92.3% and 93.3-100% for the concentrations of 0.3 g/mL and 0.5 g/mL. The control had the least percentage germination 25-25.3%. Therefore, such botanicals could reduce the infestation of stored grain pests without causing any adverse effect on grain quality. It is economically reasonable to use plants highlighted above to control *Ephestia cautella* as these products are relatively available and cheap when compared with conventional pesticides as possible protectants of stored grain (Enobakhare and Law-Ogbomo, 2002).

CHAPTER SIX

6.0. CONCLUSION AND RECOMMENDATION

6.1. Conclusion

The study has shown the bio-efficacy of *M.oleifera*, *S.longependunculata*, *Z. xanthoxyloides*, *O.canum* and Hocklicobi which served as the reference against *E. cautella*.

The leaf powder of *M.oleifera, S.longependunculata, Z. xanthoxyloides* and *O.canum* used in the ratio of 50% and 100% wt/wt increased the mortality of *E.cautell* in stored grains.

Petroleum ether extracts of leaves of *M.oleifera*, *S.longependunculata*, *Z. xanthoxyloides* and *O.canum* were also effective in controlling adult *E.cautella* in treated grains reducing adult emergence as well as repelling the insects.

Petroleum ether extracts of leaves of *M.oleifera*, *S.longependunculata*, *Z. xanthoxyloides* and *O.canum* minimized damage caused by *E.cautella* in treated grains.

The powder of the leaves and petroleum ether of *M.oleifera, S.longependunculata, Z. xanthoxyloides* and *O.canum* used in the ratio of 50% and 100% wt/wt and 0.3g/mL and 0.5 g/mL, respectively gave appreciable germination percentage compared to the standard check after the storage period.

The potential of the four plants for stored product protection has been established through laboratory bio-assays.

6.2. Recommendations

From the results obtained above, the following recommendations could be made.

- Residual analysis should be carried out to find out the level of the active ingredients residues in the treated grain after the storage period.

- The test for viability of treated grains must be continuous till the percentage germination drops in order to detect the period for which good protection can be given.

- Two of the botanicals should also be combined to evaluate their effect on the insect.

- For longer term storage, the concentration of the botanicals should be increased so as to achieve better control of storage pests.

- Further research on these botanicals should be conducted on other insects to broaden its spectrum of action.

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APPENDICIES

APPENDIX 1: Analysis for contact toxicity of petroleum ether extracts of botanicals against *E. cautella*.

Analysis of variance for Contact toxicity of petroleum ether extract of botanicals (0.1 g/mL) by topical application to *E. cautella.*

Variate: Day_1

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	15884.16	3176.83	65.64	<.001
Residual	12	580.74	48.39		
Total	17	16464.90			

Variate: Day_2

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	15074.75	3014.95	127.34	<.001
Residual	12	284.12	23.68		
Total	17	15358.87			

Variate: Day_3

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	12010.60	2402.12	124.47	<.001
Residual	12	231.59	19.30		
Total	17	12242.19			

Variate: Day_4

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	10611.90	2122.38	166.29	<.001
Residual	12	153.15	12.76		
Total	17	10765.05			

Variate: Day_5

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	10276.22	2055.24	136.73	<.001
Residual	12	180.38	15.03		
Total	17	10456.60			

Analysis of variance for Contact toxicity of petroleum ether extract of botanicals (0.3 g/mL) by topical application to *E. cautella.*

Variate: MortDay_1

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	11309.41	2261.88	86.19	<.001
Residual	12	314.90	26.24		
Total	17	11624.31			

Variate: MortDay_2

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	10579.79	2115.96	70.60	<.001
Residual	12	359.67	29.97		
Total	17	10939.45			

Variate: MortDay_3

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	9576.34	1915.27	64.03	<.001
Residual	12	358.97	29.91		
Total	17	9935.30			

Variate: MortDay_4

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	11852.533	2370.507	645.54	<.001
Residual	12	44.066	3.672		
Total	17	11896.599			

Analysis of variance for Contact toxicity of petroleum ether extract of botanicals (0.5 g/mL) by topical application to *E. cautella*.

Variate: MortDay_1

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	10996.71	2199.34	54.30	<.001
Residual	12	486.08	40.51		
Total	17	11482.79			

Variate: MortDay_2

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	10956.22	2191.24	44.95	<.001
Residual	12	585.04	48.75		
Total	17	11541.27			

Variate: MortDay_3

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	12685.09	2537.02	26.87	<.001
Residual	12	1132.82	94.40		
Total	17	13817.92			

Variate: MortDay_4

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	11852.533	2370.507	645.54	<.001
Residual	12	44.066	3.672		
Total	17	11896.599			

APPENDIX 2: Analysis of variance for the effect of botanical on adult emergence.

Analysis of variance for the botanical powders (50% wt/wt) on adult emergence of *E. cautella*

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	8307.17	1661.43	50.69	<.001
Residual	12	393.33	32.78		
Total	17	8700.50			

Analysis of variance for the botanical powders (100% wt/wt) on adult emergence of *E. cautella*

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	7463.83	1492.77	43.83	<.001
Residual	12	408.67	34.06		
Total	17	7872.50			

Analysis of variance for the petroleum ether extract (0.1 g/mL) on adult emergence of *E. cautella*

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	7022.94	1404.59	44.59	<.001
Residual	12	378.00	31.50		
Total	17	7400.94			

Analysis of variance for the petroleum ether extract (0.3 g/mL) on adult emergence of *E. cautella*

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
TREATMENT	5	8186.28	1637.26	53.88	<.001
Residual	12	364.67	30.39		
Total	17	8550.94			

Analysis of variance for the petroleum ether extract (0.5 g/mL) on adult emergence of *E. cautella*

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
TREATMENT	5	8508.94	1701.79	57.04	<.001
Residual	12	358.00	29.83		
Total	17	8866.94			

Analysis of variance for effect of petroleum ether extracts of botanicals (0.1 g/mL) on egg of *E. cautella*

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	3466.00	693.20	62.08	<.001
Residual	12	134.00	11.17		
Total	17	3600.00			

Analysis of variance for effect of petroleum ether extracts of botanicals (0.1 g/mL) on larva of *E. cautella*

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	159.167	31.833	22.04	<.001
Residual	12	17.333	1.444		
Total	17	176.500			

Analysis of variance for effect of petroleum ether extracts of botanicals (0.1 g/mL) on pupa of *E. cautella*

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	805.8333	161.1667	181.31	<.001
Residual	12	10.6667	0.8889		
Total	17	816.5000			

Analysis of variance for effect of petroleum ether extracts of botanicals (0.3 g/mL) on egg of *E. cautella*

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	4091.333	818.267	128.08	<.001
Residual	12	76.667	6.389		
Total	17	4168.000			

Analysis of variance for effect of petroleum ether extracts of botanicals (0.3 g/mL) on larva of *E. cautella*

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	216.6667	43.3333	70.91	<.001
Residual	12	7.3333	0.6111		
Total	17	224.0000			

Analysis of variance for effect of petroleum ether extracts of botanicals (0.3 g/mL) on pupa of *E. cautella*

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	928.9444	185.7889	304.02	<.001
Residual	12	7.3333	0.6111		
Total	17	936.2778			

Analysis of variance for effect of petroleum ether extracts of botanicals (0.5 g/mL) on egg of *E. cautella*

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	4454.500	890.900	148.48	<.001
Residual	12	72.000	6.000		
Total	17	4526.500			

Analysis of variance for effect of petroleum ether extracts of botanicals (0.5 g/mL) on larva of *E. cautella*

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	230.6667	46.1333	103.80	<.001
Residual	12	5.3333	0.4444		
Total	17	236.0000			

Analysis of variance for effect of petroleum ether extracts of botanicals (0.5 g/mL) on pupa of *E. cautella*

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	954.2778	190.8556	381.71	<.001
Residual	12	6.0000	0.5000		
Total	17	960.2778			

APPENDIX 3: Analysis of variance for the effect of botanical powders treatment on developmental stages of *E. cautella*

Analysis of variance for the effect of botanical powders treatment (50% wt/wt) on eggs of *E. cautella*

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	3918.278	783.656	103.72	<.001
Residual	12	90.667	7.556		
Total	17	4008.944			

Analysis of variance for the effect of botanical powders treatment (50% wt/wt) on larva of *E. cautella*

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	191.6111	38.3222	68.98	<.001
Residual	12	6.6667	0.5556		
Total	17	198.2778			

Analysis of variance for the effect of botanical powders treatment (50% wt/wt) on pupa of *E. cautella*

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	871.1667	174.2333	285.11	<.001
Residual	12	7.3333	0.6111		
Total	17	878.5000			

Analysis of variance for the effect of botanical powders treatment (100% wt/wt) on eggs of *E. cautella*

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	4227.833	845.567	132.35	<.001
Residual	12	76.667	6.389		
Total	17	4304.500			

Analysis of variance for the effect of botanical powders treatment (100% wt/wt) on larva of *E. cautella*

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	218.4444	43.6889	71.49	<.001
Residual	12	7.3333	0.6111		
Total	17	225.7778			

Analysis of variance for the effect of botanical powders treatment (100% wt/wt) on pupa of *E. cautella*

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	947.7778	189.5556	341.20	<.001
Residual	12	6.6667	0.5556		
Total	17	954.4444			

APPENDIX 4: Analysis of Variance for repellent effect of extracts on the insect.

Analysis of Variance for repellent effect of petroleum ether extracts (0.1 g/mL) on *E. cautella* after 24 hours of exposure.

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	9644.4	1928.9	12.40	<.001
Residual	12	1866.7	155.6		
Total	17	11511.1			

Analysis of Variance for repellent effect of petroleum ether extracts (0.1 g/mL) on *E. cautella* after 48 hours of exposure.

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	10911.11	2182.22	23.11	<.001
Residual	12	1133.33	94.44		
Total	17	12044.44			

Analysis of Variance for repellent effect of petroleum ether extracts (0.1 g/mL) on *E. cautella* after 72 hours of exposure.

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	9850.0	1970.0	16.89	<.001
Residual	12	1400.0	116.7		
Total	17	11250.0			

Analysis of Variance for repellent effect of petroleum ether extracts (0.3 g/mL) on *E. cautella* after 24 hours of exposure.

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	2040.3	408.1	3.38	0.039
Residual	12	1450.0	120.8		
Total	17	3490.3			

Analysis of Variance for repellent effect of petroleum ether extracts (0.3 g/mL) on *E. cautella* after 48 hours of exposure.

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	1561.1	312.2	2.08	0.138
Residual	12	1800.0	150.0		
Total	17	3361.1			

Analysis of Variance for repellent effect of petroleum ether extracts (0.3 g/mL) on *E. cautella* after 72 hours of exposure.

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	3027.8	605.6	5.04	0.010
Residual	12	1442.7	120.2		
Total	17	4470.4			

Analysis of Variance for repellent effect of petroleum ether extracts (0.5 g/mL) on *E. cautella* after 24 hours of exposure

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	7694.4	1538.9	4.07	0.021
Residual	12	4533.3	377.8		
Total	17	12227.8			

Analysis of Variance for repellent effect of petroleum ether extracts (0.5 g/mL) on *E. cautella* after 48 hours of exposure

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	8427.8	1685.6	8.20	0.001
Residual	12	2466.7	205.6		
Total	17	10894.4			

Analysis of Variance for repellent effect of petroleum ether extracts (0.5 g/mL) on *E. cautella* after 72 hours of exposure

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	10116.7	2023.3	8.35	0.001
Residual	12	2909.3	242.4		
Total	17	13026.0			

APPENDIX 5: Analysis of variance ffect of petroleum ether extracts on weight loss caused by *E. cautella* after 60 days of treatment.

Analysis of Variance for the effect of petroleum ether extracts (0.1 g/mL) on weight loss by *E. cautella* after 60 days of treatment

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	1910.69	382.14	23.24	<.001
Residual	12	197.29	16.44		
Total	17	2107.99			

Analysis of Variance for the effect of petroleum ether extracts (0.3 g/mL) on weight loss by *E. cautella* after 60 days of treatment

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	188.4134	37.6827	51.55	<.001
Residual	12	8.7726	0.7310		
Total	17	197.1860			

Analysis of Variance for the effect of petroleum ether extracts (0.5 g/mL) on weight loss by *E. cautella* after 60 days of treatment.

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	208.8297	41.7659	90.88	<.001
Residual	12	5.5149	0.4596		
Total	17	214.3446			

Analysis of Variance for the effect of petroleum ether extracts (0.5 g/mL) on weight loss by *E. cautella* after 60 days of treatment for semi field trials.

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	3	724.8742	241.6247	371.44	<.001
Residual	8	5.2041	0.6505		
Total	11	730.0783			

Analysis of Variance for the effect of botanical powder (50% wt/wt) on weight loss by *E. cautella* after 60 days of treatment

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	155.944	31.189	8.23	0.001
Residual	12	45.491	3.791		
Total	17	201.435			

Analysis of Variance for the effect of botanical powder (100% wt/wt) on weight loss by *E. cautella* after 60 days of treatment

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	186.786	37.357	13.84	<.001
Residual	12	32.401	2.700		
Total	17	219.187			

APPENDIX 5: Analysis of variance effect of petroleum ether extracts Maize seed germination after 60 days of treatment.

Analysis of variance effect of petroleum ether extracts (0.3 g/mL) Maize seed germination after 60 days of treatment

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	11109.203	2221.841	466.72	<.001
Residual	12	57.127	4.761		
Total	17	11166.329			

Analysis of variance effect of petroleum ether extracts (0.5 g/mL) Maize seed germination after 60 days of treatment

Variate: F4

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	13304.825	2660.965	565.56	<.001
Residual	12	56.460	4.705		
Total	17	13361.285			

APPENDIX 6: Analysis of variance effect of botanical powders on Maize seed germination after 60 days of treatment

Analysis of variance effect of botanical powders (50% wt/wt) on Maize seed germination after 60 days of treatment

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	9493.20	1898.64	180.17	<.001
Residual	12	126.46	10.54		
Total	17	9619.66			

Analysis of variance effect of botanical powders (100% wt/wt) on Maize seed germination after 60 days of treatment

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Treatment	5	11729.00	2345.80	176.90	<.001
Residual	12	159.13	13.26		
Total	17	11888.13			