

Influence of Some Natural Substances for Control the Greater Wax Moth *Galleria mellonella* L. (Lepidoptera: Pyralidae)

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ABSTRACT

The present work studied the effect of some natural substances (Egyptian and Chinese propolis ethanolic extract-cinnamon- clove- peppermint ethanolic extract) against the 4th larval instar of *G. mellonella*. The experiments in this study were conducted in the experimental laboratory of Biology Department, Faculty of Science, Taibah University. Potential activities of active materials on mortality percent and duration of greater wax moth at four concentrations (0.5, 1.0, 2.0 and 4.0 % w/w) were studied, and it is obvious that the mortality effect depend on the kind of materials and concentration. Peppermint ethanolic extract recorded (53.33±1.86%) larvae mortality compare to other materials and control, on other hand the Egyptian propolis ethanolic extract and clove had no effect on great wax moth at 0.5%. the pupa mortality increased in concentration 4% in Egyptian and Chinese propolis ethanolic extract, cinnamon, clove and peppermint ethanolic extract (26.66±0.00, 23.33±1.00, 43.33±4.00, 33.33±2.00 and 8.33±10.01%, respectively). The adult stage was the more resistance stage. In the larval duration, the highest effect were recorded in peppermint ethanolic extract which recorded 26.67± 3.48 at 4% concentration with 31.62 % reduction of normal (control) duration and followed by Chinas propolis and cinnamon (30.33±1.33 and 30.08±0.08 day, respectively) with 22.23 and 22.87 % reduction respectively at 4%concentration.

Keywords: Control, *Galleria mellonella* L., Larvae, Propolis, Cinnamon, Clove, Peppermint

INTRODUCTION

Wax moths are serious pests of bees wax worldwide. The greater wax moth (GWM), *G. mellonella* (Lepidoptera: Pyralidae) is recognized to be harmful to deposited and stored bees wax. *G. mellonella* the most harmful for apiaries that cause great losses every year, wax combs damage by larval feeding and old frames in the hive. Adult and larvae of wax moths can also transport serious diseases pathogens, e.g. fowlbrood. Infested Colonies showed large amounts of causative bacteria spores, *Paenibacillus larvaein* in wax moth feces (Charriere and Imdorf, 1999).

Possibilities for dominant wax moth including some manipulations in hive and other stored combs treatments i.e. physical, technical, chemical and biological methods e.g. acetic acid, formic acid and para-dichlorobenzene (Owayss and Abd-Elgayed, 2007). However, these chemicals employ is relatively effective and easy some precautions of bee products safety and contamination were considered. Some of these chemicals showed no effect against wax moth eggs (Fraser, 1997).

In recent years, environmental problems caused by pesticides overuse became scientists and public matter of concern. Synthetic insecticides extensive employ led to beneficial species destruction such as predators and parasites of the pests beside the destruction of honey bees as a pollinator's that caused biological imbalance. Natural products are an excellent alternative to synthetic pesticides as a means to decrease negative impacts to human health and the environment (Kwon *et al.*, 1996 and Koul *et al.*, 2008).

Propolis or bee glue is a dark sticky resinous substance gathering by bees from plant sources (Sorkun *et al.*, 2001). The main chemical constituents of propolis are polyphenols (flavonoid aglycones, phenolic acids, and their esters, phenolic aldehydes, alcohols, and ketones), terpenoids, steroids, amino acids, and inorganic molecules (Kartal *et al.*, 2003).

One alternative to synthetic insecticides is the botanical pesticides i.e. insecticidal plants or plant compound and the utilize of natural compounds, such as essential oils. Essential oil and their constituents display to be a potent source of botanical pesticide. The toxicity of a large number of essential oils and their constituents has been estimate against a number of bruchid pests (Keita *et al.*, 2000, 2001 and Tripathi *et al.*, 2002).

In recent years, several essential plant oils had been lead to have repellent properties. Such plants included citronella, cedar, verbena, pennyroyal, geranium, lavender, pine, cinnamon, rosemary, basil, thyme, allspice, garlic and peppermint (Mohinder, 2001 and Rim and Jee, 2006).

Senanayake *et al.*, (1978) reported that the major components of *Cinnamomum zeylanicum* of sitting in stem-bark oil and root bark oil are cinnamaldehyde (75%) and camphor (56%) respectively, both were used to control mosquito (Cheng *et al.*, 2004).

Peppermints are the richest source of natural menthol (Sharma and Tyagi, 1991 and Shasany *et al.*, 2000). Peppermint leaves contain about 0.5-4% volatile oil that is contain of 50-78% free menthol, menthofurane, monoterpene and traces of jasmine (0.15%) to get remarkable oils quality (Dew and Evans, 1984). Peppermint was tested for toxicity and repellency against *G. mellonella*. (Owayss and Abd-Elgayed, 2007), and *Callosobruchus maculatus* (Aly and Sahar, 2010).

The essential oil of clove oil own a number of bioactive compounds. The chemical constituents of the clove oil had analyzed, and 9 of 18 compounds had distinguish. The main compound (83%) was Eugenol, the second most common compound (12%) was trans-caryophyllene. These two pure compounds and clove oil were examed for toxicity and repellency against *Rhyzopertha dominica*, *Sitophilus oryzae* and *Tribolium castaneum*. The pure compounds were examed at the dosages found in clove oil (Zeng *et al.*, 2010). Owayss and Abd- Elgayed (2007) also, used clove control against *G. mellonella*.

MATERIALS AND METHODS

The experiments were proceeded in the experiment laboratory of Biology Department, Faculty of Science, Taibah University located at Al-Madinah Al-Munawarah, Kingdom of Saudi Arabia.

1-Rearing of the greater wax moth

The artificial diet used for rearing the larvae was that described by (PDBC, 2007) mixing the whole wheat bran (100 g), Maize flour (100 ml), Skimmed powdered milk (100g), Dried yeast grains (100g), Honey (175 ml) and Glycerin (175ml) .

Samples of wax combs infested with the greater wax moth were collected from various apiaries in Al-Madinah Al-Munawarah. The samples were confined in glass jars, which were tightly locked with plastic cover set up with wire screens to cancel escaping of the insect larvae and were transferred to the insectary and then identified. Rearing larvae were kept under certain laboratory conditions of 30 ± 2 °C and 65 ± 5 % R.H. in dark (Controlled by incubator). The emerged moths of *G. mellonella* were collected daily and kept in separated glass jars. Jars used for adult mating were provided with soft tissue and piece of cotton moistened with honey bee and small pieces of artificial diet for larvae, all were closed with plastic covers provided with wire screens. The newly hatched larvae burrow tunnels and feed immediately on the artificial diet until they pupate outside and inside the tunnels. Soon after emergence, moths were collected again and provided with fresh artificial diet for larvae.

2- Collection and preparation of the tested materials.

Cinnamon (*Cinnamomum zeylanicum* L.) Inner bark of trees, Clove *Syzygium aromaticum* L. Flowers buds, Peppermint (*Mentha apiperita* L) Leaves were obtained from a commercial market as 1/4 kilogram of each and thoroughly cleaned with distilled water to remove debris and later shade dried at room temperature (Mulungu et al., 2007) after drying they were then separately pounded into powder using a pestle and mortar and sieved to obtain the fine powder particles (Mukhtar and Tukur, 2000). saved in the refrigerator until using.

Peppermint leaf extract, extraction procedures put up with a previous report of Yi and Wetzstein (2010). 10g powdered dried leaves, were homogenized and extracted at room temperature under darkness in 100 mL of 80% ethanol with agitation on an orbital shaker at 200 rpm. The crude extract solutions were filtered and supernatants were collected. The extract obtained by ethanolic solution was incubated at room temperature until ethanol evaporated.

Ethanolic Chinese and Egyptian bee propolis. Chinese and Egyptian propolis was obtained from commercial bee products markets and stored in the refrigerator until using Propolis ethanolic extract was prepared according to (Ildeniz et al., 2004) as follows: Propolis was extracted by maceration at room temperature, with occasional shaking in the proportion of 10 gm. of (Chinese and Egyptian) propolis to 100 ml of solvent (ethanol 80% v/v). Extracts were obtained after 7 days of maceration and filtered. The extracts

obtained by ethanolic solution were set at room temperature until ethanol evaporated and the product obtained a honey-like consistence's are named as propolis ethanolic extract.

Four serial concentrations of each materials and ethanolic extract were prepared as followed 0.5, 1.0, 2.0 and 4.0 g/100g artificial diet.

3- Effect of tested materials on mortality percentage and duration of different life stages of *G. mellonella* .

The purpose of this experiment was to study the effect of the tested materials against different stages of wax moth (larvae, pupae, adults).Four concentrations of each extract were prepared 0.5%, 1.0% 2.0 % and 4.0 %.

Prepare 6 groups (control- Egyptian propolis ethanolic extract- Chinese propolis ethanolic extract- peppermint ethanolic extract - cinnamon- clove) each group was treated with the 4 concentrations (0.5%, 1.0%, 2.0 % and 4.0 %) except control in three replicates. Each replicate contain 10 individuals of 4th larval instar of greater wax moth. They put in glass jars which contain artificial diet. Daily observations for all groups to record the subsequent biological aspects, e.g. mortalities of larvae, pupae and adults. Also, larval duration and pupal duration were recorded.

4. Statistical analysis.

The obtained data was statistically analyzed using analysis of variance (ANOVA) at 0.05% probability also correlation coefficients were calculated according to the methods given by Snedecor and Cochran (1967).

RESULTS AND DISCUSSION

1-Effect of tested materials on larval , pupal and adult wax worm mortality percentage %.

The greater wax moth, *Galleria mellonella*, is a major pest of beekeeping industry The present study was conducted to explore the effects of five natural materials on the developmental biology and mortality of the life stages of the greater wax moth *Galleria mellonella* in the laboratory trial.

The potential activities of active materials on mortality percent of the mortality percent of larvae of greater wax moth at four concentration were reported in (Table 1), and it is obvious that the mortality effect depend on the kind of materials and concentration ,the Chinese propolis ethanolic extract (3and 4%) , cinnamon, clove and peppermint ethanolic extract at concentration 4% were the highest effective materials on the larval mortality %, The peppermint ethanolic extract was (53.33±1.86%) mortality compare to other materials and control, on other hand the Egyptian propolis ethanolic extract and clove had no effect on great wax moth at 0.5%.

In addition , in pupa stage the concentration 0.5% in propolis ethanolic extract, cinnamon and peppermint ethanolic extract had no effect on the pupa stage of great wax moth (0.00±0.00), but the mortality increased in concentration 4% in Egyptian and Chinese propolis ethanolic extract , cinnamon, clove and peppermint ethanolic extract (26.66±0.00, 23.33±1.00, 43.33±4.00, 33.33±2.00 and 8.33±10.01, respectively).

Table 1. Effect of tested materials on larval , pupal and adult wax worm mortality percentage %.

Concentration % (w/w)	control	larval stage						F	LSD _{0.05}
		Ethanollic extract				Cinnamon	Clove		
		Egyptian	Chinese	Peppermint	F				
0.5	0.00±0.00	0.00±0.00 ^{Cc}	13.33±1.16 ^{Ac}	13.00±9.01 ^{Ad}	7.00±9.01 ^{Bd}	0.00±0.00 ^{Cd}	216.88	1.40	
1.0		6.66±9.01 ^{Eb}	33.33±2.00 ^{Ab}	20.00±0.00 ^{Cc}	26.66±4.00 ^{Bc}	13.33±3.00 ^{Dc}	333.36	1.81	
2.0		13.33±3.00 ^{Da}	40.00±3.47 ^{Aa}	33.00±0.00 ^{Bb}	33.00±2.00 ^{Bb}	20.00±0.00 ^{Cb}	354.83	1.82	
4.0		13.33±9.01 ^{Da}	40.00±3.47 ^{Ba}	53.33±1.86 ^{Aa}	40.00±0.00 ^{Ba}	33.33±2.00 ^{Ca}	640.04	1.81	
F		137.57	475.30	942.17	942.17	770.25			
LSD _{0.05}	1.63	1.88	1.89	1.89	1.63				
Pupa stage									
0.5	0.00±0.00	0.00±0.00 ^{Bd}	0.00±0.00 ^{Bc}	0.00±0.00 ^{Bb}	0.00±0.00 ^{Bd}	6.66±9.01 ^{Ad}	133.66	0.81	
1.0		6.66±9.01 ^{Bc}	0.00±0.00 ^{Cc}	0.00±0.00 ^{Cb}	7.00±9.01 ^{Bc}	17.00±3.00 ^{Ac}	242.93	1.40	
2.0		13.33±3.00 ^{Cb}	11.11±1.76 ^{Db}	0.00±0.00 ^{Eb}	16.66±3.00 ^{Bb}	20.00±0.00 ^{Ab}	217.69	1.62	
4.0		26.66±0.00 ^{Ca}	23.33±1.00 ^{Da}	8.33±10.01 ^{Ea}	43.33±4.00 ^{Aa}	33.33±2.00 ^{Ba}	501.66	1.81	
F		518.32	742.38	208.16	208.16	362.37			
LSD _{0.05}	1.63	1.33	0.94	0.94	1.88				
Adult stage									
0.5	0.00±0.00	0.00±0.00 ^{Aa}	0.00±0.00 ^{Aa}	0.00±0.00 ^{Ab}	0.00±0.00 ^{Ab}	0.00±0.00 ^{Ab}	1.0	0.04	
1.0		0.00±0.00 ^{Aa}	0.00±0.00 ^{Aa}	0.00±0.00 ^{Ab}	0.00±0.00 ^{Ab}	0.00±0.00 ^{Ab}	1.0	0.05	
2.0		0.00±0.00 ^{Aa}	0.00±0.00 ^{Aa}	0.00±0.00 ^{Ab}	0.00±0.00 ^{Ab}	0.00±0.00 ^{Ab}	1.0	0.04	
4.0		0.00±0.00 ^{Ca}	0.00±0.00 ^{Ca}	8.33±1.67 ^{Ba}	41.16±5.01 ^{Aa}	8.33±1.67 ^{Ba}	1455.3	1.40	
F		1.0	1.0	208.16	5082.4	208.16			
LSD _{0.05}	0.05	0.05	0.94	0.94	0.64				

Means followed by the same letter in each column are not significantly different at (p> 0.05)

The adult stage was the more resistance stage to all materials under studied and at all concentration accept cinnamon , clove , peppermint ethanollic extract at 4% recorded (41.16±5.01, 8.33±1.67 and 8.33±1.67, respectively)

As shown in the same table, there are a significant differences among all mean values of tested materials on mortality percent of larval , pupal and adult of great wax moth at four concentration and The percentage observed mortality mortality were significantly increase by increasing the materials under study concentrations

2- Effect of tested materials on larval and pupal duration /day.

Table (2) reported the effect of tested materials under investigation on the duration of larval and pupa stage of great wax moth. In the larval stage the highest effect were recorded in peppermint treatment which recorded 26.67± 3.48 at 4% concentration with 31.62 % reduction of normal (control) duration and followed by Chinese propolis and cinnamon (30.33±1.33 and 30.08±0.08 day, respectively) with 22.23 and 22.87 % reduction respectively at 4%concentration. On the other hand the Egyptian propolis and clove treatment had no effect on larval duration at 0.5% concentration. In addition, in pupa stage the Egyptian propolis at 0.5%, Chinese propolis at 0.5, 1.0%, cinnamon at 0.5%and peppermint at 0.5, 1.0, 2.0 % had no effect on the pupa duration of great wax moth , but the more effectible were cinnamon and clove treatment (8.35±1.45 and 9.33±0.67 , respectively, at 4% concentration with 56.05 and 50.89 reduction %. The clove activity showed high significant differences among all concentration from 0.5% to 4.0% concentrations.The statistical analysis showed asignificant differences among all mean values of tested materials on pupal duration/day of *G. mellonella*.

Our natural materials provided effective control to wax moths pest, this may be due to the phytochemical composition of secondary metabolite diversity in the samples under investigation , it can

inhaled, ingested or skin absorbed by insects. And The fumigant toxicity repellency and toxicity at higher concentration accelerated larva and/or pupa development stage/stages. The abnormally higher rate of development may lead to malformed and immature individuals.

In recent years, searching for environmentally safe methods to control insect pests has been carried out using plant derivatives with significant insecticidal effects, which have been considered as new source of pesticides with negligible side effects on the environment (Balandrin *et al.*, 1985). Several wild plant extracts or isolated active compounds have been shown to act as potent acute or chronic insecticides (Emara *et al.*, 2002; Tripathy and Singh, 2005 and Moawad and Ebadah 2007). Antifeedant, (Salem *et al.*, 2003; Shishir *et al.*, 2004 and Erturk, 2006) and insect growth regulators (Abou El-Ghar *et al.*, 1996) against a variety of insect species. The use of gamma radiation and plant extract are the most promising new approaches for controlling this insect. The combined effects of gamma irradiation and bioinsecticides on Lepidopterous insects have been studied by several authors (Sallam *et al.*, 1991; Mohamed, 2004; Mohamed *et al.*, 2004; El-Shall and Mohamed, 2005 and Mohamed, 2013). Plants may provide potential alternatives to currently used insect-control agents because plants constitutions were considered as rich source of bioactive chemicals (Wink, 1993). Additionally, plant-derived materials are found to be highly effective against insecticide resistant insect pests (Kwon *et al.*, 1996), so that many investigators initiated a large screening efforts poisonous effects to use them as insecticides(El-Shazly and Hussien, 2004; Prowse *et al.*, 2006; Malarvannan and Subashini, 2007 and Khalaf *et al.*, 2009).

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Table 2. Effect of tested materials on larval and pupal duration /day.

Concentration % (w/w)	Control	larval stage										F	LSD _{0.05}
		Ethanollic extract											
		Propolis bees		peppermint		cinnamon		clove					
		Egyptian	Chinese										
		Duration	% red	Duration	% red	Duration	% red	Duration	% red	Duration	% red		
0.5		39.00±2.3 ^{Aa}	0.00	36.67 ±0.67 ^{Ba}	5.97	35.75±2.41 ^{Ba}	8.33	35.80±1.47 ^{Ba}	8.21	39.00±0.67 ^{Aa}	0.00	8.11	1.81
1.0	39.00±0.88	36.00±1.00 ^{Ab}	7.69	35.33±0.33 ^{Aa}	9.41	33.08±1.47 ^{Bb}	15.18	34.75±0.63 ^{ABa}	10.20	36.13±0.13 ^{Aa}	12.49	3.58	1.82
2.0		33.67±0.88 ^{ABc}	13.67	33.67±0.88 ^{ABa}	13.67	32.67±0.33 ^{Bb}	16.23	35.33±0.48 ^{Aa}	9.41	34.08±0.31 ^{ABa}	12.62	2.77	1.81
4.0		33.67±0.33 ^{Ac}	13.67	30.33±1.33 ^{Ba}	22.23	26.67±3.48 ^{Cc}	31.62	30.08±0.08 ^{Ba}	22.87	33.73±0.22 ^{ABa}	13.51	32.86	1.82
F		20.27		22.79		44.07		72.75		17.54			
LSD _{0.05}		1.917		1.87		1.88		1.88		1.88			
		Pupa stage											
0.5		19.00±0.00 ^{Aa}	0.00	19.00±0.88 ^{Aa}	0.00	19.00±1.86 ^{Aa}	0.00	19.00±2.33 ^{Aa}	0.00	13.00±0.58 ^{Ba}	31.58	21.60	1.819
1.0	19.00±1.16	14.33±0.88 ^{Bb}	24.58	19.00±0.67 ^{Aa}	0.00	19.00±2.08 ^{Aa}	0.00	14.67±0.88 ^{Bb}	24.58	10.33±1.33 ^{Bbc}	45.63	39.96	1.82
2.0		11.33±0.88 ^{Bc}	40.37	11.00±0.58 ^{Bb}	42.11	19.00±0.58 ^{Aa}	0.00	10.67±0.67 ^{Bc}	43.84	10.75±1.28 ^{Bb}	43.42	42.12	1.82
4.0		10.33±0.38 ^{Bc}	45.63	10.00±0.58 ^{Bcb}	47.37	13.00±0.58 ^{Ab}	31.58	8.35±1.45 ^{Cd}	56.05	9.33±0.67 ^{Bcc}	50.89	12.18	1.81
F		45.45		97.00		54.00		131.44		14.42			
LSD _{0.05}		1.88		1.63		1.33		1.33		1.33			

Means followed by the same letter in each column are not significantly different at (p> 0.05)

Aly and Sahar (2011) proved that the peppermint oil and powder are promising as alternatives to chemical control used against *Sitophilus oryzae* in wheat grains. Also, these peppermint products submit a solution of resistance development by insect due to the presence of a lot of bioactive components rather than the single insecticide. Swamy et al. (2006) observed that, the highest reduction in larval population was observed in pongamia oil (56.42%) followed by neem oil (62.57%) and mahua oil (63.09%). The lowest larval mortality of *G. mellonella* was obtained with pongamia seed kernel extract which recorded (9.28%) and neem seed kernel extract that recorded (11.78%).

Similar results were obtained in a study by Abdelrahman et al. (2012) whose proved that, peppermint and ginger were the most toxic oils against middle and late larval instars of *Galleria mellonella* than lemon and camphor.

The present findings agree with Abdelrahman et al. (2012) they demonstrated that peppermint and ginger were the most toxic oils against pupal stage of *G. mellonella* than lemon, camphor. Assegid et al. (2004) found that 2% and 4% propolis recorded 40% and 100% abortion of pupal metamorphosis and then died. Peppermint were supposed to make good companion plants, repelling pest insects and attracting beneficial ones. Peppermint are susceptible to whitefly and aphids (Ortiz, 1992). Abdelsalam (2009) stated that higher reduction percentages of *G. mellonella* fecundity (90%) were obtained when treated with the higher doses of either peppermint or lemon oil. Mortality of 100% were maintained using any tested doses of lemon oil or the higher doses of peppermint oil. Only peppermint and lemon oils caused significant decrease of emerged adults when used at the higher doses.

The discovery of natural materials in propolis, which has no side effects, but in the fact this substances interfere with the biological processes of insects as development, moulting, reproduction and hibernation, and its role is currently only breach of physiological and biochemical processes in insect (Browsers, 1984 and Klocvke et al., 1986).

Propolis extract dissolved in 55% ethanol at higher concentrations caused significantly higher

mortality to wax moth larvae than the lower concentrations and untreated controls 24 hrs after treatment the larvacidal action of propolis increases with the concentrations. However, the larvae of wax moth responded similarly to all concentrations 48 hrs but significantly more larvae up to 90% were killed in propolis treated than the controls. On the other hand, adult emergence was observed in treatments of higher concentrations. This may suggest propolis extract at higher concentration accelerated larva and/or pupa development stage/stages. from different concentration of propolis, 8 and 10 % w/v were the most toxic causing 90% and 80% mortality. These results indicate higher concentrations were more toxic Zewdu and Gemechis (2016). Assegid et al. (2004) investigated that the treatment of the late 7th stage with non-lethal doses of propolis shortened the duration of pupal metamorphosis significantly. Chandel et al. (2003) demonstrated that the duration of the pupal stage was 19.4 days in active generations and 147.9 days in overwintering generation of *G. mellonella*. Pastagia and Patel (2007) stated that pupal stage lasts for 12 to 19 days. Propolis accelerates the development of the larval/pupal stage of *Galleria mellonella*. The unusually higher rate of metamorphosis may lead to malformed and immature individuals (Assegid et al., 2004).

The sixth and seventh larval instars were reported to be more sensitive to treatments with propolis concentrations of 10% propolis that was resulted in 100% mortality of seventh larval instars. The abnormally higher rate of development may lead to malformed and immature individual's. On the other hand, earlier adult emergence was observed in treatments of higher concentrations. This may suggest propolis extract at higher concentration accelerated larva and/or pupa development stage/stages. The abnormally higher rate of development may lead to malformed and immature individuals (Garedew et al., 2004).

In general, the use of propolis as an insecticide may help us to minimize the problem environmental pollutions as result of synthetic insecticide applications. It also helps to reduce the constantly increasing problem of insecticide resistance development. Since propolis is

a complex natural product, having different components with various modes of action is unlikely or very slow (Imdorf *et al.*, 1999).

Hany (2013) stated that the essential oils of Camphor, Castor, Cinnamon, Clove and mustard with 1% concentration repelled the adult *Callosobruchus maculatus* except the castor oil. Non-significant repellent effect was found in 0.01% concentration in all tested essential oils. As a result of the presence of the essential oil of clove the nerve cord protein was significantly affected as a result of the binding of 3H-octopamine with it. Accordingly, the lack of octopamine receptors in vertebrates likely accounts for the specific mammalian selectivity of essential oils as insecticides (i.e. they are toxic to insects but not to mammals) and thus the octopaminergic system of insects represents a biorational target for insect pest control (Enan *et al.* 1998). Clove bud oil has biological activities, such as antibacterial, antifungal, insecticidal and antioxidant properties, and are used traditionally as flavoring agent and antimicrobial material in food (Lee and Shibamoto, 2001; Huang *et al.*, 2002 and Velluti *et al.*, 2003).

Extract of leaves of *Ocimum viride* gave the most effective in *T. castaneum* control and *S. oryzae*, then that of *C. odorata*. On the other hand strong repellent activity gave by *O. viride* with less than 25% survival reduction at 0.1 mg ml⁻¹. (Owusu, 2000).

The essential oils toxic effect, commonly, essential oils can be ingested, inhaled or absorbed by insects. Their fumigant toxicity control 27 oils, the volatile monoterpenes, has been described (Netzurbanza, 1991; Regnault-Roger *et al.*, 1993 and Regnault-Roger and Hamraoui, 1995). Some essential oils components contain repellent and larvicidal action: *O. basilicum* exerted a larvicidal activity evaluated at EC50 was 81 ppm (Chokechajaroenporn *et al.*, 1994).

The plants produced secondary metabolites have great activities that affect cellular, tissue and organismal level of insects. In general, they disturb physiological and cellular processes responsible for homeostasis maintenance, and provoke sublethal changes within various tissues and organs, that lead to death. However, secondary metabolites also have sublethal implications, such as reduced fecundity, reduced viability or deformities in parental and filial generations which reduce populations as a result of death, disturb development, malformations or malfunctions, extend development duration (Weissenberg *et al.*, 1998; Friedman 2002; Nenaah 2011) or act as repellents (Chopa *et al.*, 2009, Dinesh *et al.*, 2014).

REFERENCES

- Abdelrahman, M. A.; Azza, M. A. and Abeer, I. E. (2012). Efficacy of some plant volatile oils against the greater wax moth, *Galleria mellonella* L. (Lepidoptera: *Galleriidae*). Egypt. J. Agric. Res., 90(2): 411-427.
- Abdelsalam, A. M. (2009). Efficacy of some essential oils for wax moths control. 6th international conference for Arabic beekeepers, Abha, KSA, 17-19 March.
- Abou El-Ghar, G. E. S., Khalil, M. E. and Eid, T.M. (1996). Some biochemical effects of plant extracts in the black cutworm, *Agrotis ipsilon* (Hufn.) (Lep., Noctuidae). J. Appl. Ent., 482-477 (8)120.
- Aly, S. D. and Sahar, I. A. (2010). Efficacy of spearmint oil and powder as alternative of chemical control against *C. maculatus* in Cowpea Seeds. Egypt. Acad. J. Biol. Sci., 2(1): 53-61.
- Aly, S. D. and Sahar, I. A. (2011). Oil and powder of spearmint as an alternative to *sitophilus oryzae* chemical control of wheat grains. J. Plant Prot. Res., 51(2): 146-150.
- Assegid, G.; Erik, S. and Ingolf, L. (2004). Effect of the bee glue (propolis) on the calorimetrically measured metabolic rate and metamorphosis of the greater wax moth *Galleria mellonella*. Thermochemica Acta, 413(1-2): 630-72.
- Balandrin, M. F.; Klocke J. A.; Wurtele, E. S. and Bollinger, W. H. (1985). Natural plant chemicals: sources of industrial and medicinal materials. Sci., 228: 1154-1160.
- Browers, W. S. (1984). Insect-plant interaction: endocrine deviances. In organic and development of adaptation. Pitman Book London, pp.119-137.
- Chandel, Y. S.; Sharma, S. and Verma, K. S. (2003). Comparative biology of the greater wax moth, *Galleria mellonella* L., and lesser wax moth, *Achoria grisella*. Forest Pest Management and Econ. Zool., 11(1): 69-74.
- Charriere, J. D. and Imdorf, A. (1999). Protection of honey combs from moth damage. Am. Bee J., 139(8): 627-630.
- Cheng, S. S.; Liu, J. Y.; Tsai, K. H.; Chen, W. J. and Chang, S. T. (2004). Chemical composition and mosquito larvicidal activity of essential oils from leaves of different *Cinnamomum mosmophloeum* provenances. J. Agric. Food Chem., 52(14): 4395-4400.
- Chokechajaroenporn, O., Bunyapraphatsara, N. and Kongchensin, S. (1994). Mosquito repellent activities of *Ocimum* volatile oils. Phytomedicine 1, 135-9.
- Chopa, C.S.; Benzi, V.; Alzogaray, R.; Ferrero, A.A. (2009). Repellent activity of hexanic and ethanolic extracts from fruits of *Solanum eleagnifolium* (Solanaceae) against *Blattella germanica* (Insecta, Dictyoptera, Blattellidae) adults. Bol. Latinoam. Caribe. 8, 172-175.
- Dew, M. J. and Evans, J. R. (1984). Peppermint oil for the irritable bowel syndrome; a multi center trial. Br. J. Clin. Pract., 38(11-12): 394-395.
- Dinesh, D.S.; Kumari, S.; Kumar, V.; Das, P. (2014). The potentiality of botanicals and their products as an alternative to chemical insecticides to sandflies (Diptera: Psychodidae): A review. J. Vector Borne Dis. 51, 1-7.
- El-Shall, S.S.A., Mohamed, H.F., (2005). The combined effect of gamma irradiation and plant extract (Barnoof) on the nutritional profile of the Black cutworm, *Agrotis ipsilon* (Hufn.) (Lepidoptera: Noctuidae), II—the effect on the F1 progeny during the 5th and 6th instars larvae. Arab J. Nucl. Sci. Appl. 38 (2), 289.
- El-Shazly, A. M. and Hussein, K. T. (2004). Chemical analysis and biological activities of the essential oil of *Teucrium leucocladum* Boiss. (Lamiaceae). Biochem. Syst. and Ecol., 32(7): 665-674.
- Emara, S., Bakr F. R., EL-Bermawy S., Abulyazid I. and Abdelwahab H. (2002). Biological effects of four botanical extracts on the different developmental stages of cotton leaf worm. Spodoptera littoralis. 2nd Inter. Conf. Plant Protection Research Institute, Cairo. vol.1, 904-916.

- Enan, E.; Beigler, M. and Kende, A. (1998). Insecticidal action of terpenes and phenols to cockroaches: effect on octopamine receptors. Paper presented at the International Symposium on
- Erturk, O. (2006). Antifeedant and toxicity effects of some plant extracts on *Thaumetopoea solitaria* (Frey.) (Lep., Thaumetopoeidae). *Turk. J. Biol.* 30: 51-57
- Fraser, H. W. (1997). The effect of different nonspecific male sex hormone component ratios on the behavior of the female greater wax moth, *Galleria mellonella* L. (Lepidoptera: Pyralidae). M.Sc.Thesis, Univ.Gueiph, Canada),
- Friedman, M. (2002). Tomato glycoalkaloids: Role in the plant and in the diet. *J. Agric. Food Chem.* 50,5751–5780.
- Garedeu A.A., Schmolz E., and Lamprecht I., (2004). Effect of the bee glue (propolis) on the calorimetrically measured metabolic rate and metamorphosis of the greater wax moth *Galleria mellonella*. *Thermochimica Acta*, vol. 413, 63-72,
- Hany A. F. (2013). Effect of five essential oils as repellents against the Cowpea Beetle, *Callosobruchus maculatus* (F.). *Bull. Env. Pharmacol. Life Sci.*, 2(5): 23-27.
- Huang, Y.; Ho, S. H.; Lee, H. C. and Yap, Y. L. (2002). Insecticidal properties of eugenol, isoeugenol and methyleugenol and their effects on nutrition of *Sitophilus zeamais* Motsch. (Coleoptera: Curculionidae) and *Tribolium castaneum* (Herbst) (Coleoptera: Tenebrionidae). *J. Stored Prod. Res.*, 38(5): 403-412.
- Ildenize, B. S.; Cunha, A.; Alexandra, C. H.; Sawaya, F. M.; Mario, T.; Shimizua, M. C.; Marcucci, C.; Flavia, T.; Drezza, A.; Giovanna, S.; Poviaa, p. O. and Carvalho, A. (2004). Factors that influence the yield and composition of Brazilian propolis extracts. *J. Braz. Chem. Soc.*, 15(6): 964-970.
- Imdorf, A., Bogdanov S., Ochoa R.I., and Calderone N.W., (1999). Use of essential oils for control of *Varroa jacobsoni* Oud. in honeybee colonies. *Apidologie*, vol. 30, 209–228.
- Kartal, M.; Yildiz, S.; Kaya, S.; Kurucu, S. and Topcu, G. (2003). Antimicrobial activity of propolis samples from two different regions of Anatolia. *J. Ethnopharmacol.*, 86(1): 69-73.
- Keita, S. M.; Vincent, C.; Belanger, A. and Schmit, J. P. (2000). Effect of various essential oils on *Callosobruchus maculatus* (F.) [Coleoptera: Bruchidae]. *J. Stored Prod. Res.*, 36(4): 355–364.
- Keita, S. M.; Vincent, C.; Schmit, J. P.; Arnason, J. T. and Belanger, A. (2001). Efficacy of essential oil of *Ocimum basilicum* L. and *O. gratissimum* L. applied as an insecticidal fumigant and powder to control *Callosobruchus maculatus* (Fab.) (Coleoptera: Bruchidae). *J. Stored Prod. Res.*, 37(4): 339-349.
- Khalaf, A. A.; Hussein, K. T. and Shoukry, K. K. (2009). Biocidal activity of two botanical volatile oils against the larvae of *Synthesiomyia nudiseta* (Wulp) (Diptera: Muscidae). *Egypt. Acad. J. biolog. Sci.*, 2(1): 89-101.
- Klocvke, J. A.; wagenea, B. V. and Bolandrin, M. F. (1986). The ellagitannin geranin and its hydrolysis products isolated as insect growth inhibitors from semiarid land plant. *Phytochemistry*, 25(1): 85-91.
- Koul, O.; Walia, S. and Dhaliwal, G. S. (2008). Essential oils as green pesticides: potential and constraints. *Biopesticides Int.*, 4(1): 63-84.
- Kwon, M.; Ahn, Y. J.; Yoo, J. K. and Choi, B. R. (1996). Potent insecticidal activity of extracts from *Ginkgo biloba* leaves against *Nilaparvata lugens* (Homoptera: Delphacidae). *Appl. Entomol. Zool.*, 31: 162-166.
- Lee, K. G. and Shibamoto, T. (2001). Antioxidant property of aroma extract isolated from clove buds [*Syzygium aromaticum* (L.) Merr. Et Perry]. *Food Chem.*, 74(4): 443-448.
- Malarvannan, S. and Subashini, H. D. (2007). Effect of *dodonaea angustifolia* crude extract on biochemical profile of *Helicoverpa armigera* (Hubner) (Noctuidae: Lepidoptera). *Biochem. and Cell. Arch.*, 7(1): 1-8.
- Moawad, S. S. and Ebadah I. M. A. (2007). Impact of some natural plant oils on some biological aspects of potato tuber moth *Phthorimaea operculella*, (Zeller) (Lepidoptera: Gelechiidae). *Res. J. Agric. and Biol. Sci.*, 3(2) 119-123.
- Mohamed, H.F. (2013). Bioenergetics growth model for the effect of gamma irradiation and plant extract (Barnoof) on the progeny of Black cutworm, *Agrotis ipsilon* (Hufnagel) *Applied Radiation and Isotopes* 73, 101–108.
- Mohamed, H.F., (2004). Inherited sterility induced by gamma irradiation and/or Barnoof plant extract on reproductive potential and mating ability of the black cutworm, *Agrotis ipsilon* (Hufngel) (Lepidoptera: Noctuidae). *Isot. Radiat. Res.* 36 (4), 713.
- Mohamed, H.F., EL-Naggar, S.E.M., Mohamed, A.Z., (2004). The combined effect of gamma irradiation and plant extract barnoof on the nutritional profile to the black cutworm, *Agrotis ipsilon*, III. The effect on some haemolymph digestive enzyme activities. *J. Egypt. Acad. Soc. Environ. Dev. (C-Mol. Biol.)* 5 (2), 99.
- Mohinder, S. J. (2001). Toxicity effect to garlic extracts on the eggs of *Aedes aegypti* (Diptera: Culicidae): A scanning electron microscopic study. *J. Med. Entomol.*, 38(3): 446-450.
- Mukhtar, M. D. and Tukur, A. (2000). Antibacterial activities of aqueous and ethanolic extract of *Pistia stratiotes*. *Niger J.*, 1(1): 51-59.
- Mulungu, L. S.; Lupenza, G.; Rieuben, S. O. W. and Misangu, R. W. (2007). Evaluation of botanical products as Stored grain Protectant against maize weevil *S. zeamais*. *J. Entomol. Sci.*, 4: 258-262.
- Nenaah, G. (2011) Toxic and antifeedant activities of potato glycoalkaloids against *Trogoderma granarium* (Coleoptera: Dermestidae). *J. Stored. Prod. Res.* 2011, 47, 185–190.
- Netzurubanza, L. (1991) Quelques applications d'*Ocimum canum*: revue de la littérature, *Rivista Italiana EPPOS*, 10mes Journées Scientifiques de Digne les Bains, 703–6.
- Ortiz, E. (1992). The encyclopedia of herbs, spices & flavorings. London: Dorling Kindersley. pp: 36–37.
- Owayss, A. A. and Abd-Elgayed, A. A. (2007). Potential efficacy of certain plant volatile oils and chemicals against greater wax moth, *Galleria mellonella* L. (Lepidoptera: pyralidae). *Bull. Ent. Soc. Egypt. Econ. Ser.*, (33): 67-75.
- Owusu, E. O. (2000). Effect of some Ghanaian plant components on control of two stored-product insect pests of cereals. *Journal of Stored Products Research*, Vol. 37, 1, 85–91.
- Pastagia, J. J. and Patel, M. B. (2007). Biology of *Galleria mellonella* L. on brood comb of *Apis cerana* F. J. of *Plant Prot. and Envi.*, 4(2): 85-88.

- PDBC.(2007).Traning on Entomopathogenic nematode for insect pest control. Training manual for "Hands-on Training on Entomopathogenic nematode" 20th march to 29th March 2007, Project Directorate of Biological Control, Bangalore, India
- Prowse, G. M.; Galloway, T. S. and Foggo, A. (2006). Insecticidal activity of garlic juice in two dipteran pests. *Agric. Forest Entomol.*, 8(1): 1-6.
- Regnault-Roger, C. and Hamraoui, A. (1995) Fumigant toxic activity and reproductive inhibition induced by Monoterpenes upon *Acanthoscelides obtectus* Say (Coleoptera), bruchid of kidney bean (*Phaseolus vulgaris*). *Journal of Stored Product Research* 31, 291-9.
- Regnault-Roger, C., Hamraoui, A., Holeman, M., Theron, E. and Pinel, R. (1993) Insecticidal effect of essential oils from mediterranean aromatic plants upon *Acanthoscelides obtectus* Say, Coleoptera, bruchid of kidney bean (*Phaseolus vulgaris* L.). *Journal of Chemical Ecology* 19, 1233-44.
- Rim, I. S. and Jee, C. H. (2006). Acaricidal effects of herbal essential oils against *Dermatophagiodes farinae* and *D. pteronyssinus* (Acari: Pyroglyphidae) and qualitative analysis of a herbal *Mentha pulegium* (Pennroyal). *Korean J. Parasitol.*, 44(2): 133-138.
- Salem, Nagwa, Y., Hoda, A. Ramadan and Elham, A. Sammour (2003). Physiological and histopathological effects of some wild plant extracts on the cotton leaf worm *Spodoptera littoralis* (BOISD) (Lepidoptera Noctuidae). *Bull. Ent. Soc. Egypt, Econ.*, 29 (113).
- Sallam, H.A., Elnagar, S., Ibrahim, S.M., (1991). Effects of gamma radiation on reproductive potential of the black cut worm, *Agrotis ipsilion* (Hufn.). *Arab J. Nucl. Sci. Appl.* 24, 165-174.
- Senanayake, U. M.; Lee, T. H. and Wills, R. B. H. (1978). Volatile constituents of cinnamon (*Cinnamomum zeylanicum*) oils. *J. Agric. Food Chem.*, 26(4): 822-824.
- Sharma, S. and Tyagi, B. R. (1991) .Character correlation, path coefficient and heritability analyses of essential oil and quality components in corn mint. *J. of Gen. Virol.*, 45: 257-262.
- Shasany, A. K.; Khanuja, S. P. S.; Dhawan, S. and Kumar, S. (2000). Positive correlation between menthol content and in and in vitro menthol tolerance in *Mentha arvensis* L. cultivars. *J. Bio.*, 25(3): 263-266.
- Shishir-Tandon, Mittal, A. K., Kasana, V. K. and Pant, A. K. (2004). Effect of essential oil of *Elsholtzia densa* L. on growth and reproduction of *Spilosoma obliqua* (Walker). *Indian J. Ent.*, 66(3): 206-208.
- Snedecor, G.W. and Cochran, W.G. (1967): Statistical methods. The Iowa state Univ. Press. Ames, Iowa, USA. 6th ed.
- Sorkun, K.; Suer, B. and Salih, B. (2001). Determination of chemical composition of Turkish propolis. *Z. Naturforsch.*, 56(7-8): 666-668.
- Swamy, B. C. H.; Rajagopal, D. and Naik, M. I. (2006). Effect of plant products against larvae of greater wax moth. *Mysore J. of Agric. Sci.*, 40(1): 125-128.
- Tripathi, A. K.; Prajapati, V.; Verma, N.; Bhal, J. R.; Bansal, R. P.; Khanuja, S. P. S. and Kumar, S. (2002). Bioactivities of the leaf essential oil of *Curcuma longa* (Var. Ch-66) on three species of stored-product beetles (Coleoptera). *J. Econ. Entomol.*, 95(1): 183-189.
- Tripathy, M. K., Singh, H. N. (2005). Synergistic effect of certain vegetable oils to the efficacy of synthetic pyrethroids for the control of *Helicoverpa armigera* (Hubner). *Agric.Sci. Digest.*, 25(1): 1-5.
- Velluti, A.; Sanchis, V.; Ramos, A. J.; Mari'n, S. (2003). Inhibitory effect of cinnamom, clove, lemongrass, oregano and palmarose essential oil songrowth and fumonisin B1 production by *Fusarium proliferatum* in maize grain. *Int. J. Food Microbiol.*, 89: 145-154.
- Weissenberg, M.; Levy, A.; Svoboda, J.A.; Ishaaya, I. (1998). The effect of some solanum steroidal alkaloids and glycoalkaloids on larvae of the red flour beetle, *Tribolium castaneum*, and the tobacco hornworm, *Manduca sexta*. *Phytochemistry*. 47, 203-209.
- Wink, M. (1993) Production and application of phytochemicals from an agricultural perspective. In: "Phytochemistry and Agriculture" (T.A. van Beek, H. Breteler, eds.). Clarendon, Oxford, UK, Vol. 34. p. 171-213.
- Yi, W., and Wetzstein, H.Y. (2010) Biochemical, biological, and histological evaluation of some culinary and medicinal herbs grown under greenhouse and field conditions. *J. Sci. Food Agr.* 90:1063-1070.
- Zeng, L.; Lao, C. Z.; Cen, Y. J. and Liang, G. (2010). Study on the insecticidal activity compounds of the essential oil from *Syzygium aromaticum* against stored grain insect pests. 10th International Working Conference on Stored Product Protection, pp. 766-771.
- Zewdu A. and Gemechis L. (2016) Insecticidal action of honeybees propolis extract against larvae of lesser wax moth. *Agric. Biol. J. N. Am.*, 2016, 7(6): 302-306

تأثير بعض المواد الطبيعية لمكافحة دودة الشمع الكبيرة

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تعتبر دودة الشمع الكبيرة من اهم الافات التي تصيب نحل العسل وتسبب ضررا اقتصاديا لتربية النحل ولذلك كان الهدف من هذه الدراسة تقييم كفاءة بعض المواد الطبيعية لمقاومة هذه الافة وهي المستخلص الكحولي للبروبوليس المصري والصيني والمستخلص الكحولي لاوراق النعناع وكذلك القرقة والقرنفل وذلك بعمل اربع تركيزات من هذه المواد (0.5 و1.0 و2.0 و4.0 %) باضافتها الى التغذية الصناعية المقدمة للافة وتم قياس نسب الموت للمراحل المختلفة في دورة حياة الافة. اوضحت النتائج ان تأثير هذه المواد يتوقف على نوع المادة المستخدمة وكذلك التركيز , فقد اظهر المستخلص الكحولي لاوراق النعناع بتركيز 4% نسبة موت لليرقات وصلت الى 53.33% ولكن انخفاض التركيز الى 0.5% لم يظهر اي تأثير للمستخلص الكحولي للبروبوليس المصري وكذلك القرنفل وكذلك سجلت نسبة موت كبيرة بتركيز 4% في طور العذرى للمستخلص الكحولي للبروبوليس المصري والصيني والقرقة والقرنفل والمستخلص الكحولي لاوراق النعناع (66.26-33.23-33.33-43.33 على الترتيب) واطهر طور الحشرة الكاملة مقاومة كبيرة للمواد تحت الدراسة. اما بالنسبة لفترة العمر لليرقات فقد سجل المستخلص الكحولي لاوراق النعناع نسبة تأثير تصل الى 31.62% بالمقارنة بالكونترول يليها القرقة والمستخلص الكحولي للبروبوليس الصيني .