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Evaluations of some extracted natural oils against *Bruchidius incarnates* and *Ephestia elutella*

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ABSTRACT

The repellency test of three extracted oils *Jatropha curcas*, canola and Jojoba Seed oils, were studied against two lepidopterous insect pests *Ephestia elutella* and *Bruchidius incarnates*. Results showed that, the tested oils, are promising for controlling the target stored product pests. Results also, showed that *Jatropha curcas* oil acted not only as oviposition deterrents but also adversely influence fecundity. Moths oviposited eggs on treated seeds with *Jatropha* oil but the numbers of eggs is always lower in treated seeds than in the control. The tested oils were significantly decreased the seeds infestations. The means number of eggs deposited/female were significantly decreased to 39.4 ± 1.7 , 34.4 ± 1.7 and 32.3 ± 3.7 egg/female after *E. elutella* treated with Jojoba Seed oil at concentrations 5, 2 and 3%, respectively. *Jatropha curcas* oils were gave the higher mortality of *B. incarnates* when treated at the corresponding concentrations. Accumulative mortality (%) of *E. elutella* and *B. incarnates* larvae increased gradually by increasing the period of exposure in case of treated foam with different tested oils. After seven days of treatments, the Jojoba Seed oil, *Jatropha curcas* and canola oil the accumulative mortality *B. incarnates* recorded 71.5, 73.4 and 40.1, respectively as compared to 33.1 in the control.

Keywords: *Ephestia elutella* and *Bruchidius incarnates*, *Jatropha curcas*, canola and Jojoba oil.

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INTRODUCTION

The tobacco moth *Ephestia elutella* An adult female lays 120-150 eggs, which are laid in the cracks and crevices of warehouses or food store facilities. Eggs hatch within 10-12 days. Larvae pass through 4-5 moults to attain full growth when they are 10-12 mm long. The larvae go to diapauses stage throughout the winter before pupation. *E. elutella* (Hübner) developed 3 generations in a year and over wintered with larvae in Egypt. The peak stages of emergence were in early May, early and middle July, late August to early September respectively. The results of the research showed that the egg stage of the first generation were 5-6 days, the larva stage 56 days, the pupa stage 8.5 days under the natural temperature it cause a lot of damage to chocolate beans (Emam et al., 2013; Mulungu et al., 2007 and Hany, 2001).

Legume seeds are considered a main source of protein for human and animal nutrition (Smartt, 1976). Bruchids are serious insect pests and cause considerable damage to seeds and grains (Shomar, 1963). Although numerous insect pests attacked all parts of bean plants, bruchids were the most important field and storage pests (Abat and Ampofo, 1996). Bruchid seed beetles are important parasites of legume seeds, but their effect on germination can be unpredictable. Beetles deplete seed resources and can kill the embryo but also scarify seeds (Fox et al., 2012). Weight losses caused by the bruchids and other factors after 7 months of storage averaged 8.5% in dry beans stored by small-scale farmers. The storage losses caused by insects and factors other than insects were estimated as 6.9 and 1.6%, respectively, and bruchids accounted for 24.5% of the combined losses (Espinal et al., 2004) and Emam, et al, 2013.

Bruchidius incarnates are major pests on wheat seed and other stored grains in Egypt. Most of the damage when larvae interfere with seeds. Using chemical control for these pests is undesirable, hence safe and yet effective control are being sought. Plant oils were used as toxicant, oviposition repellants, growth regulator or antifeedents for many insect pests (Abate and Ampofo, 1996). Almost all the insect pests of stored grains have a remarkably high rate of multiplication and within one season, they may destroy 10-15% of the grains and contaminate the rest with undesirable odours and flavours (Baby, 1994; Abate and Ampofo, 1996; Songa and Rono, 1998; Schmale et al., 2002). A wide range of seed beetles attack the grain of

common bean varieties (Mulungu et al., 2007). However, the predominant damaging pests of stored grain legumes mainly in the tropics are *Callosobruchus maculatus* (Fab.), *C. chinensis* (L.), *Caryedonserratus* (Oliver), *Zabrotessub fasciatus* (Boheman) and *Acanthoselids obtectus* (Say) (Nahdy and Agona, 1995; Nichimbi-Msolla and Misangu, 2002; Emanu et al., 2003). Essential oils may have attractive or repellent effects and in some cases they showed an insecticidal action against insects. Essential oils isolated from plants and consisting of cyclic and monocyclic mono-terpenes are effective repellents against insects (Rodriguez and Levin, 1975). Oil carriers can also distribute the inoculum over the thin inter-segmental membranes, which are more readily penetrated by entomogenous fungi (Lisansky, 1989). During the past few decades, application of synthetic pesticides to control agricultural pests has been a standard practice. However, with growing evidence that many conventional pesticides can adversely affect the environmental requirements for safer means of pest management have become crucial (Rozman et al., 2007). *Simmondasia chinensis* (Link) (jojoba) is a semiarid evergreen shrub. The plant is cultivated in some parts of the middle-east and Latin-American countries (Habashy et al., 2005). Jojoba seeds are containing of some unique glucoside compounds that can cause food intake inhibition and repellency effect for the stored products pests (Bellirou et al., 2005). Chemical insecticides can cause pest resistance, environmental and food contamination and toxicity to non-target organisms (Pimentel et al., 2009; Tavares et al., 2010)

The present work aimed to explore the protective potency of some botanical oils against *E. elutella* and *B. incarnates*.

MATERIALS AND METHODS

The target insects were reared under laboratory conditions on semi artificial diet (fine wheat with some adherent endosperm), with 20% glycerin and 5% yeast powder. Population were held at $26 \pm 2^{\circ}$ C and 70-80% RH. The broad. Groups of 100 one-day old eggs were placed, each in 12 cm petri dishes comprising a thin layer of diet.

Extraction of *Jatropha* Seed Oil:

The seeds of *Jatropha curcas* were harvested from trees from Nobaryarigion. They were dried in a shade for seven days, shelled and the batches ground into a fine powder, Five hundred and fifty grams of the powder and 2.5 liters of petroleum ether (40°C) were used in the extraction of the oil with a oxhlet's extractor for 48 hours. This yielded 250 ml of clean yellow oil and the ether was recovered through a rotary evaporator. The oil was kept in the dark at 4°C until it was needed. Toxicity of the Oil on *E. elutella* and *B. incarnates* The rapeseeds are first ground up coarsely and heated slightly to break down oil viscosity and preliminarily expel some excess moisture. It will also compromise the integrity of the cellular walls of the seed, which will make the next step of de-hulling much easier. Extraction of canola oil begins with the rapeseed. The rapeseed comes from a yellow flowering plant from the Brassicaceae family (the same family of plant that produces mustard seeds). Rapeseed oil is the "crude oil" to canola oil's "gasoline." There is a refinement process used to create the finished product. The essential oils *jatropha*, *jojoba* and *canola* were isolated by steam distillation of the dried plants (Guenther, 1961). Canola oil extracted according to Unger (1990). The tested oil emulsions were prepared as follows: 5 drops of "Triton X-100 as emulsifier was mixed thoroughly with 5ml of each tested oil, then water was added to obtain the desired concentrations (0.2, 0.5, 2 and 3 %) in percent of (v/w). The emulsifier was mixed at the corresponding concentrations and used as check.

Repellency test :

The experiments were conducted in an arena in choice test . Disc of filter paper (Whatman No. 1) was treated with the tested oil at 1 % conc. and placed in cell A. While filter paper treated with distilled water and emulsifier only as control was placed in the cell B . Twenty newly emerged larvae were introduced into each arena. After 1,2,3,4,5,6 and 7 days, the number of beetles present in the cells A and B was recorded. The percentages of repellency values were calculated using the equation: $D = (1 - T/c) \times 100$ (Lwande et al., 1985) where T and C represent the mean number of larvae in cells A and B (Treated and untreated), respectively .

The insecticidal activity of tested oils:

Experiment was designed to test the initial as well as the persistent effect of the tested oils on moth as cumulative mortality during successive intervals (0, 2, 4, and 7 days). Foam granules about 1cm in diameter were treated at time (zero time) with tested oils, dried and provided with heat sterilized wheat seeds (100g/each) fastened each with a string. Then all treatments were used immediately as non-choice test. The foam granules treated with the tested oils were mixed with wheat seeds (2g foam/100g seeds) according to Abd El-Aziz (2001).

Ovipositional deterrent effect of tested oils (no choice test):

To evaluate the oviposition deterrent of the tested oils, a pair of newly emerged larvae, was placed with treated or untreated broad seeds in glass jars (250 cc capacity) covered with muslin. The moths were left to lay eggs, and then the

deposited eggs were counted on the seeds in the treated and untreated jars. Each experiment was repeated five times, (Abd El-Aziz and Ismail, 2000). The number of deposited eggs was used as a criterion for the evaluation of reduction percentages.

No. of deposited eggs in treatment

$$\text{Reduction \%} = [100 - \frac{\text{No. of deposited eggs in treatment}}{\text{No. of deposited eggs in control}}] \times 100$$

No. of deposited eggs in control

The percent reduction is an index of effectiveness of the applied oils in reducing infestation and was calculated according to, Su (1989).

The persistence of oils during storage

Experiment was designed to test the persistent effect of on foam as surface protectant at 20 day intervals over 120 days. All gunny sacks (20x20 cm each) were full of heat sterilized wheat seeds (100 g each), fastened, each with a string. The foam granules (about 1 cm in diameter) were sprayed with treatments, dried and provided as a layer between sacks. Following exposing to those treatments, two pairs of newly emerged moths (2–3 day) were placed in a jar (2 l capacity with four gunny sacks) and observed for egg laying. The laid eggs were counted on the seeds in the treated and untreated jars. Each experiment was repeated five times, (Abd El-Aziz 2001). The number of deposited eggs was used as a criterion for the evaluation of reduction percentages.

$$\text{Reduction \%} = \frac{(100 - \text{no of laid eggs in treatments})}{\text{no of laid eggs in control}} \times 100$$

no of laid eggs in control

The percent reduction is an index of effectiveness of the applied testedoils in reducing the pest infestation and was calculated according to Su (1989).Dead larvae were removed and the jars were kept under the same experimental conditions until the emergence of F1 progeny adults occurred. Percentage reduction in adult emergence or inhibition rate (% IR) was calculated as:

$$\%IR = \frac{(Cn - Tn)}{Cn} \times 100$$

where: Cn is the number of newly emerged larvae in the untreated (control) jar

Tn is the number of larvae in the treated jar (Tapondjouet *et al.* 2002).

RESULTS AND DISCUSSION

The effect of tested plant oil vapors on the reproduction of the *Ephestia cutella* and *B.incarnates* larvae were studied using the no-choice test (Table 1 & 2). The reproduction of larvae was reduced by the treatments with Jojoba Seed oil and canola oil vapors, but not inhibited completely. *Jatropha curcas* oil acted not only as oviposition deterrents but also adversely influence fecundity. Eggs laid by females on treated seeds with *Jatropha* oil but the numbers of eggs is always lower in treated seeds than in the control. Data in table (1) show that the tested oils were significantly decreased the wheat seeds infestations. The means number of eggs deposited / female were significantly decreased to 39.4±1.7, 34.4±1.7and 32.3±3.7egg/female after *E. elutella* treated with Jojoba Seed oil at concentrations,5, 2 and 3%, respectively.

Table 1. Oviposition deterrent effect of tested oils against 4th larval instar of *Ephestia elutella*

Type of oils	Mean number of eggs/female ±S.E.		
	0.5%	2%	3%
Jojoba Seed	39.4±1.7 (62.46)	36.5±3.5 (72.50)	34.3±4.3 (78.61)
<i>Jatropha curcas</i>	31.4±4.4 (73.63)	23.3±3.3 (84.66)	21.4±4.7 (90.57)
canola	55.4±4.5 (51.65)	63.4±5.7 (55.31)	67.1±2.7 (48.40)
Control	116.6±3.4		
F value	21.8	23.4	25.16
LSD	11.91	10.11	11.13

Jatropha curcas oils were gave the higher mortality of *B.Bruchidius* when treated at the corresponding concentrations. Shaaya *et al.*, (1997) reported that edible oils are potential control agents against *C. maculatus* and can play an important role in stored-grain protection. Abd El-Aziz (2001) mentioned that clove and eucalyptus oil vapors impaired the fecundity of the bruchid beetles, *Callosobruchus maculatus* .Data proved promising oviposition deterrence, toxicity and suppressing egg deposition and adult emergence (Emam, *et al.*, 2013; Mulungu *et al.*, 2007 and Hany, 2001).

When *B.incarnates* treated with *Jatropha curcas* at concentrations, 0.5, 2 and 3%the eggs laid per female showed a significant decreased reached to 29.4±5.4, 18.1±3.3, and 16.5±4.4 eggs/female as compared to 118.1±3.5 in the control (Table 2).In this respect, Deshpande *et al.* . (1974) reported that oleic and linoleic acid as insecticidal components from *Nigella sativa* which were found to be toxic to the pulse beetle, *C. chinensis* . In a choice test, filter paper strips treated with *Acorus*

calamus oil at 200,400 or 800 µg/cm² repelled *Tribolium castaneum* adults during the first 2 weeks, there after repellency decreased more rapidly, than neem oil (Jilani *et al.* , 1988) . Abd El-Aziz and Ismail (2000) mentioned that Nigella oil gave 45.5 and 40.2 % repellency during the first and second days, respectively. Nigella oil became attractive to *Bruchidius incarnatus* beetles and had little persistent. Pumpkin oil at 1 % conc., had strong repellent activity (88%) during the first day of observation and then decreased gradually to reach (0.0%) repellency during the last day of experiment. Frankincense oil indicated the more persistent. White Mustard oil was found to protect storage insects infesting stored pulses, especially the black gram and the green gram,(Prakash, 1982) . Black mustard seeds contain sinigrin and myrosin and yield after maceration with water 0.7–1.3% of volatile oil. The latter contains over 90% of allylisothiocyanate (Olivier *et al.*, 1999). The main chemical components of clove oil are eugenol, eugenol acetate, iso-eugenol and caryophyllene (Olivier *et al.*, 1999).

Table 2. Oviposition deterrent effect of tested oils against 4th larval instar of *B.incarnates*

Type of	Mean number of eggs/female ±S.E.		
	0.5%	2%	3%
Jojoba Seed	40.4±4.8 (67.56)	34.5±4.5 (73.51)	30.3±5.3 (78.61)
Jatropha	29.4±5.4 (73.63)	18.1±3.3 (85.66)	16.5±4.4 (95.50)
canola	66.4±2.4 (50.55)	72.4±1.7 (55.41)	78.1±5.6 (49.70)
Control	118.1±3.5		
F value	18.7	22.2	25.5
LSD	11.1	13.1	11.1

Data in Table (3) indicate that accumulative mortality (%) of *C. maculatus* and *B.incarnates* larvae increased gradually by increasing the period of exposure in case of treated foam with different tested oils. After seven days of treatments, the Jojoba Seed oil, Jatropha curcas and canola oil the accumulative mortality of *B.incarnates* recorded 71.5, 73.4 and 40.1 , respectively as compared to 33.1. In this respect, Chander and Ahmed (1986) applied different doses of the essential oil of *Acorusc alamus* to seeds of green gram *Vignaradiata* (Wilcz) to protect them against *B .incarnates* and found that 1ml/Kg offered a high degree of protection up to a period of 135 days. Prolonged protection of the seeds was mainly due to a high adult mortality besides reduced oviposition and low hatching. Foam sprayed with clove oil (5%) and placed between sacks caused the highest mortality (66.6%) of *C. maculatus* as compared with treated sacks or foam inside sacks (63.3% and 42%, respectively) after 6 days of storage (Abd El-Aziz, 2001) .The same results were obtained by Chander and Ahmed (1986); (Emam *et al.*, 2013; Mulungu *et al.*, 2007 and Hany, 2001), Saxena *et al.*, (1976), Surabaya *et al.*, (1994) and Maheshwari *et al.*, (1998).

Table 3. Accumulative mortality of *E.elutella* and *B.incarnates* adults during the first week of wheat seeds exposed to treated foam with different oils

Treated oils	Time(days)	Accumulative mortality	
		<i>E. elutella</i>	<i>B .incarnates</i>
Jojoba Seed oil	0	19.3	22.3
	2	32.6	35.5
	4	41.7	44.3
	7	59.7	71.5
Jatropha curcas	0	25.7	27.5
	2	47.8	48.9
	4	61.4	73.4
	7	66.9	73.1
Canola oil	0	23.3	1
	2	34.6	
	4	38.9	16.6
	7	43.7	24.8
Untreated (control)	0	15.3	27.7
	2	23.5	33.1
	4	26.8	
	7	29.9	

Data in (Table 4 and 5) show that the effect of oil tested against the target insect pests. The number of eggs laid / female *E. cutella* were decreased after oil treatment reached to 9±5.0 and 11±3.1eggs/ female as compared to 98.0±5.4 and 94.4±7.8eggs/female in control after 100 and 120 days of treatments with Jojoba oil. The percentage of reduction recorded 92,89 and 90% when *E. cutella* treated with Jojoba , canola and Jatropha curcas oil after 100 days, respectively (Table 4).

Table 4. Effect of different oil tested on number *E. elutella* laid eggs/female and % of adult emergence (F1) larvae during storage periods

Storage	Control	Treatments with oils
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Interval [days]	Jojoba		canola		Jatropha curcas,	
	no. of eggs /♀±S.E.	% adult Emergence(F1)	no. of eggs /♀±S.E.	% adult Emergence (F1)	no. of eggs /♀±S.E.	% adult Emergence (F1)
20	39.8±3.5	82	0.0±0.0	0	0.0±0.0	0
40	49.2±2.3	83	1.7±1.0	0	3.0±0.0	0
60	69.0±1.4	83	5.0±1.0	2	3.0±0.0	2
80	78.0±1.3	90	7.0±0.0	4	4.0±0.0	3
100	98.0±5.4	88	9±5.1	8	12.0±1.1	7
%of reduction	-	-	92		89	
120	94.4±7.3	91	10±0.1	6	13±1.5	6
%of reduction	-	-	95		92	
						93

When *B. incarnates* were treated with different oils, the eggs laid /female after 100 days, 4±3.1, 12±3.5 and 11±3.1 when treated with Jojoba, canola and *Jatropha curcas*, oil respectively as compared to 98.0±1.0 in the control. The corresponding percentage of reduction recorded , 97, 89 and 90 %. Prolonged protection of the seeds was mainly due to a high adult mortality besides reduced oviposition and low hatching. Foam sprayed with clove oil (5%) and placed between sacks caused the highest mortality (66.6%) of *C. maculatus* as compared with treated sacks or foam inside sacks (63.3% and 42%, respectively) after 6 days of storage (Abd El-Aziz, 2001). Araya and Eman (2009), found that More than 90% mortality of adult *Z. subfasciatus* was also observed for bean seeds treated with *J. curcas*, *D. stramonium* and *B. dodecondra* 96 hour after treatment at the rate of 15g/ 150g of grain application. The same results were obtained by Chander and Ahmed (1986); Saxena et al, (1976); Surabaya *et al.* (1994) Kheradm et al., (2010) Emam et al, (2013) and Maheshwari et al, (1998). Abd El-Aziz and Ismail, (2000) mentioned that *Nigella* oil gave 45.5 and 40.2% repellency during the first and second days, respectively. *Nigella* oil became attractive to *Bruchidius incarnatus* beetles and had little persistent. Pumpkin oil at 1% concentration, had strong repellent activity (88%) during the first day of observation and then decreased gradually to reach (0.0%) repellency during the last day of experiment. Frankincense oil indicated the more persistent. White Mustard oil was found to protect storage insects infesting stored pulses, especially the black gram and the green gram (Prakash, 1982). Sabbour and Abd-El-Raheem, (2013) reported that, The means number of eggs deposited/female were significantly decreased to 39.4±1.7, 34.4±1.7 and 32.3±3.7 egg/female after *C. maculatus* treated with Jojoba Seed oil at concentrations 5, 2 and 3%, respectively. *Jatropha curcas* oils were gave the higher mortality of *C. maculatus* when treated at the corresponding concentrations. Accumulative mortality (%) of *C. maculatus* and *C. chinensis* beetles increased gradually by increasing the period of exposure in case of treated foam with different tested oils. After seven days of treatments, the Jojoba Seed oil, *Jatropha curcas* and canola oil the accumulative mortality of *C. chinensis* recorded 71.5, 73.4 and 40.1 , respectively as compared to 33.1.

Table 5. Effect of different oil tested on number *B incarnates* of laid eggs/female and % of adult emergence (F1) of larvae during storage periods of wheat

Storage Interval [days]	Control		Treatments with oils					
			Jojoba		canola		Jatropha curcas,	
	no. of eggs /♀±S.E.	% adult Emergence (F1)	no. of eggs /♀±S.E.	% adult Emergence (F1)	no. of eggs /♀±S.E.	% adult Emergence (F1)	no. of eggs /♀±S.E.	% adult Emergence (F1)
20	43.8±3.2	80	0.0±0.0	0	0.0±0.0	0	0.0±0.0	0
40	54.2±2.1	86	1.0±0.0	0	0.0±0.0	0	1.0±0.0	0
60	79.0±1.1	88	5.0±0.1	2	4.0±0.1	1	3.0±0.0	1
80	82.0±1.5	94	8.0±0.0	3	6.0±0.0	3	5.0±0.0	2
100	98.0±1.	98	4±3.1	4	12±5.3	5	11±3.1	9
%of reduction	-	-	97		89		90	
120	95.4±1.6	91	10±1.5	6	13.0±3.0	7	14.0±1.4	8
%of reduction	-	-	94		91		90	

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