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### Evaluation of the Insecticidal Activity of *Punica granatum* L. and *Olea europaea* L. against the Dry Bean Beetle, *Acanthoscelides obtectus* (Say) (Coleoptera:Bruchidae)



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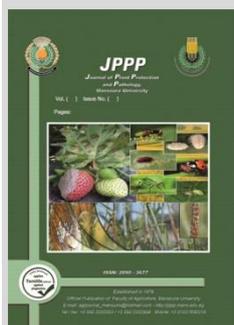
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#### ABSTRACT

A laboratory study was carried to determine the efficacy of *Punica granatum* Linn. and *Olea europaea* L. leaf powders against adults of the dry bean beetle, *Acanthoscelides obtectus* (Say) (Coleoptera:Bruchidae) under laboratory conditions. The death rate of *A. obtectus* increased gradually with increasing the concentration and period of exposure to powdered plant leaves. After 120 hours of exposure to the powdered plant leaves, *P. granatum* caused adult mortality 82.17%, while *O. europaea* caused adult death 68.14%. In case of *P. granatum* leaf powders, the LC<sub>50</sub> value was 16.73 g / 30 g white kidney bean while LT<sub>50</sub> recorded 69.01 hours at 25 g / 30 g white kidney bean. While the maximum effectiveness of inhibition oviposition was recorded by using *P. granatum* leaf powder (75.06 %) compared to *O. europaea* (53.11 %). A decrease in adult exit was reported among the treatments and the maximum decrease was noticed in the dry white beans with the highest dose rate leaves powder of *P. granatum* (68.19%) and *O. europaea* (52.38%). Results obtained from this study revealed that the used plant leaf powders significantly reduced the infestation and the weight loss for white kidney bean. However, *P. granatum* had higher activity against *A. obtectus* than *O. europaea*. There is no doubt that both plants under study have a significant effect on the pest, and accordingly, they can be used in the production of environmentally friendly plant-based insecticides.

**Keywords:** *Acanthoscelides obtectus*, *Punica granatum*, *Olea europaea*, plant leaf powder



#### INTRODUCTION

The widespread bean (*Phaseolus vulgaris* L.) (Fabaceae) is the most significant legume worldwide for direct human consumption and is a source of dietary fibre, calories, proteins, minerals and vitamins for millions of people worldwide (Shehata *et al.*, 2011). It is an important crop cultivated in Egypt, both economically and socially (Moghazy, 2014). Likewise, as in other cultivated grains and seeds, beans are usually stored after harvest for the purpose of consumption or marketing. The unsafe storage of such grains may cause many problems such as reducing the physiological quality of the planting seeds as well as reducing the quality of the seeds consumed.

Bruchids beetles represent a serious threat to the legumes that are spread throughout the world. At the present time, the dry bean beetle, *Acanthoscelides obtectus*, is considered one of the most important economic pests that infested dry beans and the most widespread in different regions of the world. It is a primary pest that pierces the seed and lives inside it to proceed with its development, feeding on the cotyledons in the process. Accordingly, those pills are more likely to be infested by secondary insect pests and microorganisms (Jiménez *et al.*, 2017). The beetle now occurs in Europe, Asia, North and South America, Africa, and Australia. Its spreading around the world is caused by both global warming and wrong practices in crop storage practices (Alvarez *et al.*, 2005 and Oliveira *et al.*, 2013). *A. obtectus* has from five to six generations per year. Both larvae and imagines can overwinter, though usually only in storage (Wightman and Southgate, 1982, Alvarez *et al.*, 2005). The

penetration of the seed and feeding on its contents results in many damages such as weight loss, a decrease in the nutritional value and cleanliness of the product, contamination with insect excrement and eggs, reduction in the germination rate of infected seeds (Gallo *et al.*, 2002).

Control methods of this pest and other stored grain pests depend mainly on the remaining insecticides and pesticides with gas fumes (Mondal and Khalequzzaman, 2006). Accordingly, the excessive use of such types of pesticides has led to many problems, such as the emergence of many resistant strains of pests, toxic residues in stored grains, and the continuous increase in the costs of this type of control (Riebeiro *et al.*, 2003). Therefore, it was necessary to resort to the use of safe, low-cost, environment-friendly alternatives to control these pests. At present, control efforts are focused on many natural substances extracted from plants that can later be used as commercial bioinsecticides.

The Pomegranate, *Punica granatum* L. is one of the ancient cultivated plants, and this type of plant is distinguished by the production of edible fruits (Lye, 2008). In addition, this plant is distributed in different regions of the world, including tropical and subtropical areas, and one of the most important countries famous for growing this plant is Iran, Malaysia, Turkey, California, Egypt, Italy, India, China, Chile, and Spain (Still, 2006 and Holland *et al.*, 2009). Many important chemicals compounds were recorded from different parts of the pomegranate plant, for example, the following chemicals were obtained from leaves: triterpenoids, steroids, alkaloids, flavonoids, tannins, polyphenols, and some anthocyanins (Li *et al.*, 2006 and Bhandary *et al.*, 2012). The plant extracts of

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the pomegranate plant have shown their effectiveness as insecticides against many different types of insects such as *Spodoptera litura* (Fabricius), *Anopheles pharoensis* (Theobald), *Culex pipiens* (L.), *Rhyzopertha dominica* and *Musca domestica* L. have been reported ( Mansour *et al.*, 2004, Sharma and Rajguru 2009, Mahmood, 2010, Gandhi and Pillai, 2011 and Eldiasty *et al.*, 2014).

The olive (*Olea europaea* L.) plant is one of the most famous and important agricultural crops in the Mediterranean region. Olive trees are grown for the purpose of extracting olive oil and obtaining table olives. Olive trees also annually produce many secondary products during and after harvesting the crop, such as leaves and fruit squeezing products that are wasted without using them for optimal use in practical applications. Olive leaves are considered one of the most important by-products of olive crops available throughout the year. Leaves can also be obtained in large quantities during separation from fruits after harvesting and before squeezing to obtain oil (approximately 10% of the weight of olives). Since ancient times, olive leaves and their extracts have been widely used for many medicinal purposes in the Mediterranean region, due to many, olive leaves contain many active substances that are used as an antioxidant, anti-hypertensive, anti-atherogenic, anti-inflammatory, hypoglycaemic and hypocholesterolaemic treatment (Boss *et al.*, 2016). However, there are very few studies that have been conducted on the effectiveness of olive leaves as insecticides. Olive leaf powder and extract have the ability to interfere with the development and reproduction of insects and therefore can be used successfully as insecticides ( Khemais *et al.*, 2018) .

The main objective of this study presented here was to measure the oviposition and progeny disincentive activity; damage and infestation estimate of the dry bean beetle, *Acanthoscelides obtectus* on the white kidney bean, *Phaseolus vulgaris* exposed to *Punica granatum* and *Olea europaea* leaves powders and the identification of phytochemical elements of plants used in this study that may be in charge of toxic effect.

## MATERIALS AND METHODS

### 1- Insect rearing:

All experimental studies were carried out in laboratories of Plant Protection Department, Faculty of Agriculture, South Valley University, Qena, Egypt. Adults of *A. obtectus* were gained from Infected white kidney bean from a general shop in Qena Market and sieved out from the infested white kidney bean. Fresh, healthy white kidney beans were used as food media for the insects.

The white kidney beans were subjected to dry heat treatment in an oven at 200 °C for 48 hours to get rid of any kind of insects or microorganisms that may be present inside the beans. A sample of 250 g of the disinfected white kidney beans was placed in each of five rearing plastic jars (1 liter) covered with the muslin cloth to keep the insects from escaping and also to allow the insects to breathe in a good way, after which 20 pairs of adult *A. obtectus* were introduced. Insects were raised and experiments were conducted at an average temperature of 28 ± 2 °C and relative humidity of 75 ± 5%. After that, the jars were preserved for insect breeding and reproduction. The recently emerging adults were then used in the following experiments.

### Plant collection and preparation:

Fresh leaves of *P. granatum* and *O. europaea* is were collected from the Agricultural Research Experiments Center, Faculty of Agriculture, Assiut University, and the Agricultural Research Experiments Center, Faculty of Agriculture, South Valley University respectively. The leaves were washed well with water several times and then were dried in the shade away from sunlight. Using a mixer device, the dried leaves were thoroughly ground and then sifted with a 1 mm sieve. The powders were then separately kept in polythene bags at room temperature in the laboratory.

### 2- Collection of the white kidney beans:

The white kidney beans used in the experiment were obtained from a freshly stored bean crop and were not treated with any kind of pesticide. Damaged seeds were excluded by manual examination, and the healthy seeds were kept for use in the experiment. The seeds used in the experiment were exposed to a temperature of 100 °C for two hours inside the drying oven and then cooled at room temperature in order to get rid of any infection in the seeds coming from the field. Using a digital weigh, 30 g of heat-treated white kidney beans were weighed and placed in 250 g of plastic jars, with triplicates per concentration, then stored in a cool dry place.

### Bioassay experiment:

By using 250 gm plastic jars, 30 g of clean white kidney beans were mixed with separate concentrations (0 g, 10 g, 15 g, 20 g, and 25 g) of *P. granatum* and *O. europaea* leaves powder to evaluate the potency of this plant against *A. obtectus* that infecting white kidney beans. To ensure good mixing of the dry white kidney beans with the leaves powder, they were thoroughly stirred several times inside each plastic jar. In each plastic jar, twenty adults *A. obtectus* (10 days old) were placed and then covered to prevent insects escaping. The adult mortality rate was recorded after (24, 48, 72, 96 and 120 h) of treating white kidney beans with leaves powdered of both types of plants. Dead adult insects were recorded when they are not moving after applying slight pressure with fingertip, in order to avoid the phenomenon of death imitation. Three replicates were made from each treatment and three replicates were made from the control, and were placed on the bench in the laboratory. The mortality rate in adults of these insects was estimated using the method used by (Omotosho and Oso, 2004).

$$\% \text{ mortality in adults} = \frac{\text{Number of Dead Weevils}}{\text{Total Number of Weevils}} \times 100$$

### 3- Oviposition obstructive activities:

To assess the effectiveness of the tested plant leaves powder on the effect of Oviposition obstructive activities, 10 g, 15 g, 20 g and 25 g of plant leaf powders were mixed with 30 g of clean kidney beans in 250 g plastic jars. Ten adults for *A. obtectus* were placed in a plastic jar and covered with a gauze cover surrounded by rubber to prevent the insects from escaping. The insects were left in the plastic jars for seven days until they laid the eggs. The number of eggs that were laid on the treated seeds and the control was calculated after one week of laying eggs (after 14 days of treatment) by using a hand magnifying glass before removing the female insects accordingly, the percentage of oviposition obstructive activity was calculated using the equation assigned by (Arivoli and Tennyson , 2013).

**% oviposition obstructive activity = (Number of eggs laid in control - Number of eggs laid in treated) / (Number of eggs laid in control + Number of eggs laid in treated) × 100**

**4- Adult emergence:**

To measure the F1 progeny deterrence potency of plant leaves powder, In 250g plastic jars, it was placed 30g of white kidney beans blended with various dose rates of the plant leaves powders as it was mentioned previously. After the number of eggs has been calculated beforehand the experiment continued until the appearance of F1 adults. The number of adults in F1 that appeared in all of the white kidney beans replicates were recorded in control and treatments by using a pipette, 28 days after the injury. The number of appeared adults from treated beans and control appear was applied to estimate the percentage decrease in adult emergence.

**Percentage reduction in adult emergence = (no of the emerged adult from control - no of the emerged adult from the treated white beans)/ no of the emerged adult from control ×100**

**5- Damage and infection estimate:**

The total number of holes that adult individuals exit from was calculated in all treatments after 28 days had passed from the occurrence of the infestation, for use in calculating the infestation rate and thus assessing the damage.

**Percentage rate infestation = Number of white kidney beans that have the exit holes for adults / The total number of white kidney beans used in experiment × 100**

The final weight for each treatment was calculated for use in calculating the percentage of weight loss due to the infection with *A. obtectus*

**Percentage weight loss = (The premier white kidney beans weight - definitive white kidney beans weight) / The premier white kidney beans weight × 100**

**6- Phytochemical screening of plant leaves powder**

Phytochemical analysis of the leaves powder of *P. granatum* and *O. europaea* was conducted by Pharmacognosy Department, National Research Centre, Dokki, Cairo, Egypt.

**7- Statistical analysis**

The results obtained from the experiment were subjected to analysis of variance (ANOVA) using (SPSS

software (version 25). The number of eggs produced after the treatments, adult exit, damaged and undamaged white kidney beans, where significant differences subsist, treatment means were separating by using the Least Significant Difference (LSD) at 0.05% probability level. Also, data obtained from adult weevil's mortality were subjected to Probit analysis employing SPSS computer program for calculating LC<sub>50</sub> and LT<sub>50</sub> values.

**RESULTS AND DISCUSSION**

Table (1) showed the toxicity of plant leaves powder by contact on the survival of adult beetles after treatment.

The results revealed that in each treatment, no mortality was noted in the control. The death-rate of *A. obtectus* raised progressively with the time of treatment and concentration of plant leaves powders. Although none of the plant leaf powders used in the experiment were able to cause 100% death to adults; however, at 25 g/30 g of white kidney beans *P. granatum* caused 82.17% adult mortality and *O. europaea* exerted 68.14% adult mortality after 120 hours of the transaction, precisely followed by *P. granatum* used at 20 g, 15 g and 10 g which caused the death of adults. The results obtained from the experiment proved the powders of plant leaves that were tested as insecticides by contact greatly reduced the number of insects. In general, *P. granatum* leaf powder was shown to be more toxic on *A. obtectus* than on *O. europaea* leaf powder. Deaths that occurred among adults of insects may be due to the toxicity of plant leaves powders by contact. Most of the insects breathe through the trachea, which opens to the surface of the body by the spiracles (Adedire *et al.*, 2011). The used plant leaves powders may cause the spiracles to close and thus prevent air from reaching the trachea, which causes the insects to die by suffocation. Moreover, the high mortality rate may be due to the possibility that the tested plant leaves powder to possess the properties of the pesticides (Rajkumar and Jebanesan, 2004). This confirms the results obtained that prove that the tested plants can be considered a rich source of active chemicals that can be extracted and used as effective insecticides.

**Table 1. Mean percentage of the mortality of *A.obtectus* treated with of *P.granatum* and *O.europaea***

Treatment	<i>P.granatum</i>					<i>O.europaea</i>				
	24h	48h	72h	96h	120h	24h	48h	72h	96h	120h
Control (0g)	0	0	0	0	0	0	0	0	0	0
10 g	3.11	6.14	10.21	12.14	22.15	1.14	4.41	8.16	11.32	17.42
15 g	5.22	11.11	16.14	22.1	34.11	3.18	8.1	13.12	18.14	25.18
20 g	11.11	21.18	31.22	44.18	61.14	8.21	17.14	27.14	32.1	50.11
25 g	18.09	32.16	46.33	57.14	82.17	11.03	25.18	35.16	48.19	68.14
LSD	10.41	12.2	18.17	7.61	25.71	13.11	10.11	14.33	12.56	16.34

Four different concentrations (25 g, 20 g, 15 g and 10 g /30g bean) from each of *P. granatum* and *O. europaea* leaves powder were tested against the adult *A. obtectus*. The two leaves powder reduced the number of *A. obtectus* under laboratory conditions. The LC<sub>50</sub> values and slope of different leaves powder, *P. granatum* and *O. europaea* are presented in Table (2).

Lowest LC<sub>50</sub> value 16.73 g /30g bean was recorded at *P. granatum* treatment. Based on the LC<sub>50</sub> values, *P. granatum* leaves powder was the most effective against *A. obtectus* adults, followed by *O. europaea*.

LT<sub>50</sub> values decreased whenever the concentration increased. There was a difference among LT<sub>50</sub> of the plant leaves powder, *P. granatum* and *O. europaea*. At a concentration of 25 g/30g bean the lowest LT<sub>50</sub> (69.01 hours) was obtained by *P. granatum*, followed by *O. europaea* (91.19 hours) Table (3).

**Table 2. LC<sub>50</sub> values of the *P.granatum* and *O.europaea* leaves powder against *A.obtectus* adults after 120 hrs**

Plant leaves powder	LC <sub>50</sub> g / 30g bean	95% Confidence limits		Slope ± S.E
		lower	upper	
<i>P.granatum</i>	16.73	15.19	18.4	4.26 ± 0.62
<i>O.europaea</i>	19.86	17.9	22.72	3.78 ± 0.63

**Table 3. LT<sub>50</sub> values of the *P.granatum* and *O.europaea* Leaves powder against *A.obtectus* adults at 25g/30g (Whit kidney bean)**

Plant leaves powder	LT <sub>50</sub> (Hours)	95% Confidence limits		Slope ± S.E
		lower	upper	
<i>P.granatum</i>	69.01	59.32	80.85	2.34 ± 0.34
<i>O.europaea</i>	91.19	77.95	112.65	2.29 ± 0.36

Such increased mortality rates within exposure time increase are due to the slow-acting mode of action of plant leaves powder, *P. granatum* and *O. europaea* which are different from the traditional insecticide. Similarly, (Papachristos and Stamopoulos, 2002) and (Marcelo *et al.*, 2018) Proved that the activities of plants' extracts and plant products to control *A. obtectus* dependent on both increasing concentration and exposure period.

The results recorded in Table (4) proved that the inhibitory effect of laying eggs of *A. obtectus* and reducing the appearance of adults on white kidney beans treated with plant leaves powder.

**Table 4. Mean percentage of oviposition obstructive and percentage of the lowering in adult emergence by *A.obtectus* on treated white kidney been.**

Treatment	Percentage oviposition obstructive		percentage lowering in adult emergence	
	<i>P.granatum</i>	<i>O.europaea</i>	<i>P.granatum</i>	<i>O.europaea</i>
	Control (0g)	25.17	25.84	17.33
10 g	54.13	28.17	30.11	22.41
15 g	67.22	37.06	47.13	38.18
20 g	71.14	45.12	56.17	43.09
25 g	75.06	53.11	68.19	52.38
LSD	18.06	N.S	N.S	N.S

The leaves powder of *P. granatum* and *O. europaea* decrease or prohibit in a sure way laying eggs and thus the emergence of adults. The lowering in oviposition was raised with the rise in dose of each replicate. The higher concentration 25 g/30 g of beans (75.06 and 53.11) were achieved the highest percentage in reducing the rate of egg laying compared to the lower concentrations 10 g/30 g of beans (54.13 and 28.17) for *P. granatum* and *O. europaea* respectively. *P. granatum* leaves powder recorded the highest percentage to reduce oviposition. (Gandhi *et al.*, 2010) proved that high concentrations of pomegranate leaf powder are able to significantly reduce egg laying. One of the reasons for the lack of eggs for *A. obtectus* may be the physiological and behavioral changes that occur to adult insects as a result of the effect of the chemical properties of the natural plant leaf powder that these beetles handle, thus reducing the number of eggs that these beetles lay (Shukla *et al.*, 2007). In addition to the above, natural plant leaf powders may negatively affect insect movement and mating, and reduce the number of eggs produced (Akinkurolere *et al.*, 2009 and Ileke *et al.*, 2012). The results obtained are largely consistent with (Gandhi and Pillai 2011), who showed a significant decrease in the number of eggs for *Rhyzopertha dominica* (Coleoptera: Bostrichidae) after treating them with pomegranate leaf powder.

Table (4) also showed that the results obtained from the effect of two plant leaves powder on adult emergence of *A. obtectus* on white kidney beans. The results obtained also showed that the lowest number of adult emergence was recorded on white kidney beans treated with the highest concentrations of the plant leaf powders used. Based on that,

the lowering in the adult emergence depends to a large extent on the dose concentration of the plant leaf powder. Among the two plant leaves powder, *P. granatum* applied at 25 g/30 g of beans recorded the highest percentage reduction in adult emergence (68.19%) while the least percentage reduction in adult emergence (22.21%) was obtained from 10g/30 g of beans *O. europaea* treatment. There were about (17%) percentage reductions in adult emergence in the control. This confirms the results of (Gandhi *et al.*, 2010) who observed the ability of the evaluated plant leaves powder, *P. granatum* to significantly suppress adult emergence of *Tribolium castaneum* (Herbst.). The study results are in the same line of the findings of (Hategekimana and Erlor, 2020) who found that the inhibitory effect of EOs from *Eucalyptus camaldulensis*, *Mentha piperita* and *Pimpinella anisum* on fecundity and fertility of *A. obtectus*. Also (Raja *et al.*, 2001) demonstrated that volatile oils are taken from *Mentha arvensis* and *Cymbopogon nardus* prevent adult appearance in *Callosobruchus maculatus* in cowpea. (Jose and Adesina, 2014) reported that the ability of the evaluated plant extracts to show the evident effects on the survival of the young stages of the insect after hatching, which causes a decrease in the appearance of adult insects in all the concentrations. Also, the decrease in the adult emergence insect population may be due to the death of eggs or the death of larvae, or to the decrease in the rate of egg hatching. Finally, powders of plant leaves that have been mixed with seeds may prevent the larvae from penetrating into the seeds, thus reducing the adult emergence insect (Khalequzaman and Goni, 2009). The leaf powders of *P. granatum* and *O. europaea* used in this experiment proved their great ability to reduce the appearance of adult insects, which proves that these plants possess the properties of pesticides used to kill eggs and larvae. Hundreds of plant species have been counted and recorded, which have proven their high ability as inhibitors oviposition and hatching (Nuttavich and Mayura, 2019). (Shukla *et al.*, 2007) reported a decrease in the numbers of *Callosobruchus chinensis* beetle on stored green cowpea seeds which proved that the powders of *Murraya koenigii* and *Eupatorium cannabinum* effective as ovicidal.

The Percentage of infestation and weight loss were offered in Table (5).

**Table 5. Mean percentage of seed damage caused by *A.obtectus* and percentage weight loss recorded on treated white kidney bean.**

Treatment	Percentage infestation		percentage weight loss	
	<i>P.granatum</i>	<i>O.europaea</i>	<i>P.granatum</i>	<i>O.europaea</i>
Control (0g)	25.14	25.16	35.18	35.22
10 g	16.37	20.18	30.44	33.62
15 g	10.15	15.52	21.18	27.13
20 g	6.48	10.71	14.63	20.85
25 g	3.18	7.14	11.95	17.75
LSD	15.31	N.S	1.27	N.S

Untreated control recorded the highest (25 and 35%) percentage infestation and weight loss when compared to other treatments. Meanwhile, white kidney beans treated with *P. granatum* leaves powder was significantly different between treatments, white kidney beans treated with *O. europaea* was not significantly different in less infestation and weight lack, respectively. Percentage infestation and weight loss for all the treatment decreased as the

concentration of botanicals increased. At 25 g/30 g of beans, the Percentage infestation and weight loss were higher (7.14 and 17.75 %) in *O. europaea* than in *P. granatum* (3.18 and 11.95%), respectively. Through the results of the study, the reason leading to the decrease in infestation and lack of weight can be attributed to the decrease of progeny in the first generation resulting from the effect of plant leaf powders used as effective pesticides that prevent the hatching of eggs and the emergence of adults. Plant products have proven effective in controlling the different life stages of many pests by killing or prolonging the life stages (eggs, larvae, pupae) (Baskar and Ignacimuthu, 2012). Similar results were reported by (Suleiman and Suleiman, 2014) that leaves plant powders of *Euphorbia balsamifera* L. and *Lawsonia inermis* L. greatly reduced cowpea seed damage 28 days after treatment. This might be due to the limited contact of *C. maculatus* with the treated seed.

The results of the primary plant chemical components for each plant leaf powder separately are presented in Table (6).

**Table 6. Preliminary phytochemical screening of *P. granatum* and *O.europaea* leaves powder.**

phytochemicals	Methanol (70%) extract of	Methanol (70%) extract of
	<i>P.granatum</i> leaves	<i>O.europaea</i> leaves
Alkaloid	+	+
Tannins	+	+
Saponin	+	-
Steroids	+	+
Phenols	+	+
Anthraquinone	-	-
Flavonoids	+	+
Terpenoids	+	+
Caradiacglycosides	+	+

The phytochemical screening showed the existence of alkaloid, Tannins, Steroids, phenols, Flavonoids, Terpenoids and Cardiac glycosides in leaves powder of both plants under study. It was noted that the powders of the leaves of both plants did not contain Anthraquinone. While *P. granatum* leaf powder contained Saponins, it was not found in *O. europaea*. These results are in agreement with previous findings of (Lahmadi *et al.*, 2019), (Shah *et al.*, 2017) and (Boggula and Peddapalli, 2017).

The outcomes of the phytochemical structure of *P. granatum* and *O. europaea* provide good knowledge about the mechanism of the action of *P. granatum* and *O. europaea*. Phytochemicals, like tannins, have a strong harmful impact on phytophagous insects and influence the insects development by a link to proteins, decrease supplement retention absorption, and cause midgut injuries (Barbehenn and Peter, 2011) and (Sharma *et al.* 2009).

Tannins are contraction sharp polyphenols (mouth wrinkles) and act as nutrition obstructions to numerous insect pests. They accelerate protein sedimentation nonspecifically (including the stomach enzyme related to herbivores), by hydrogen bonds or covalent bonds of protein-NH<sub>2</sub> bunches (Abdul Rashid *et al.*, 2012). (Karamanoli *et al.*, 2011) detailed that tannins apply their activity by a group of mechanisms that incorporate iron chelation and enzyme inhibition. (Padin *et al.*, 2013) Moreover, demonstrated that tannin combine with protein represses the enzyme movement and decreases the existence

of protein in haemolymph insect and alkaloid dependable for harmful impact against *Tribolium castaneum*. In spite of the mechanism actions of *P. granatum* and *O. europaea* aren't however known, the dominance of tannins in its powder may responsible for the effect of phytochemicals. Most current studies referred that the insecticidal mechanism of saponins is concerned with the impact on the detoxification enzymes of insect. Saponins can decrease superoxide dismutase, catalase, acetylcholinesterase, and carboxylesterase exercises (Lin *et al.*, 2018) and (Cai *et al.* 2016). (Chaieb, 2010) broadly surveyed insecticidal impacts of saponins, connecting their insecticidal action their interaction with cholesterol, which causes a decline in ecdysteroid synthesis. Flavonoides can repress enzymatic action and restrain the development of larvae of diverse insect species. Many flavonoids affect the process of moulting and reproduction in many different species of insects through the ability of these compounds to prevent the formation of the hormone juvenile. Flavonoid compounds inhibit the ecdysone-dependent genes to transcription (Oberdorste *et al.*, 2001). Many types of flavonoids have been evaluated as insecticides with potent effects on oviposition, fecundity, mortality, weight decrease, and the emergence of adults (Salunke *et al.*, 2005). The results proved the effectiveness of Flavonoides compounds as insecticides on nymphs and adults of the aphid *Eriosoma lanigerum* Hausmann. Based on that, these compounds can be used successfully in an integrated management program for this type of aphids (Ateyyat, 2012).

Steroids are available in many different plant groups in varying proportions. These compounds are considered to be analogues of moulting steroid hormone, ecdysteroid. Accordingly, the steroids clearly control the growth and development of insects in all stages of their life cycle. Phytoecdysteroids give security to plants by changing the ordinary levels of the ecdysteroid hormone in adults and larvae in insects. Their most frequent and prominent role is their function as moulting hormones, thereby controlling not only insect and arthropod moulting, but also reproduction and other physiological processes such as embryonic development (Spindler *et al.*, 2001 and Truman and Riddiford, 2002). Subsequently, phytoecdysteroids can be a fabulous substitution of insecticides in insect pest management programs. Alkaloids control neuroendocrine functions by inhibiting acetylcholinesterase activity in larvae treated with these compounds. Alkaloids from some plants have a similar effect of blocking the acetylcholinesterase enzyme (Aniszewski, 2007). Several investigators reported that alkaloids are strong natural insect antifeedants (Cornelious *et al.*, 2009, Sani *et al.*, 2014 and Manosalva *et al.*, 2019).

Phenolic compounds, in general, are characterized by their high ability to precipitate proteins (Appel, 1993 and Al-yousuf *et al.*, 2010), accordingly these phenolic compounds have toxic effects on many types of insects by inhibiting digestive enzymes in insects or to the formation of insoluble compounds with dietary proteins and thus reduce the nutritional value of the food insects eat through the chemical decomposition of essential amino acids (Felton *et al.*, 1992). The overall results of this study demonstrated that two plant leaf powders were used against dry bean beetle *Acanthoscelides obtectus*; *Punica granatum* leaf powder is

effective than *Olea europaea* leaf powder used and our next approach will be targeted to isolate possible active compounds.

### CONCLUSION

In this study, it can be concluded that dry bean beetle *A. obtectus* in stored white kidney beans were exposed to leaf powders of two plants, *P. granatum* and *O. europaea*. Results indicated that, percentage mortality varied according to plant species and concentrations of the plant leaf powders. *P. granatum* had lower LC<sub>50</sub> values; hence the most toxic plant leaf powders to *A. obtectus*. Accordingly, higher concentrations of the treatments recorded lower number of eggs laid and progeny emergence. Therefore, these botanicals can be used as option for the control of *A. obtectus* in stored white kidney beans. However, further research is encouraged to study their pesticidal activities against other insect pests. Also, it is therefore recommended that the plant leaf powders of *P. granatum* and *O. europaea* could be used as alternative insecticides by our farmers especially for preserving the grains used as seeds for recent cultivations.

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## تقييم النشاط الابادى لمساحيق أوراق نباتات *olea europaea* L. و *Punica granatum* Linn. ضد خنفساء الفاصوليا *Acanthoscelides obtectus* (Say) (Coleoptera: Bruchidae)

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اجريت هذه الدراسة لتحديد فاعليه مساحيق أوراق نبات الرمان و الزيتون ضد خنفساء الفاصوليا الجافه تحت الظروف المعملية . واطهرت النتائج زياده معدل موت خنفساء الفاصوليا تدريجيا مع زياده كل من التركيز و فتره التعرض لمسحوق أوراق النباتات. بعد مرور 120 ساعة من التعرض لمساحيق أوراق النباتات تسبب مسحوق أوراق الرمان في موت الحشرات الكاملة بنسبه 82,17% بينما تسبب مسحوق أوراق نبات الزيتون في موت الحشرات الكاملة بنسبه 68,14%. كانت قيمه التركيز اللازم لقتل 50% من الحشرات المعامله في حاله استخدام مسحوق أوراق الرمان هي (16,73 جرام /30 جرام من الفاصوليا البيضاء) بينما كانت قيمه الوقت اللازم لقتل 50% من الحشرات المعامله هي (69,01)ساعه عند تركيز 25 جرام /30جرام من الفاصوليا البيضاء). سجل ايضا مسحوق أوراق الرمان أعلى فاعليه لتثبيط وضع البيض وهي 75,06% مقارنة بمسحوق أوراق الزيتون 53,11%. ايضا تم تسجيل إنخفاض تعداد خروج الحشرات الكاملة بين المعاملات و لوحظ الحد الاقصى من ذلك في الفاصوليا البيضاء مع أعلى جرعه من مسحوق أوراق الرمان 68,19% و 52,38% مع الزيتون. أظهرت النتائج المتحصل عليها من هذه الدراسه أن مساحيق أوراق النباتات المستخدمه قللت بشكل كبير من الاصابه و فقدان الوزن للفاصوليا البيضاء الجافه مع ذلك كان مسحوق الرمان أعلى نشاط ضد خنفساء الفاصوليا من الزيتون و لكن مما لاشك فيه أن كلا النباتين قيد الدراسه لهما تأثير كبير على الافه و بالتالي يمكن استخدامهما في انتاج مبيدات حشريه نباتيه صديقه للبيئه.