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Effect of Italian and Carniolan honey bee hybrids rearing colony within bar's level on acceptance mean and royal jelly production

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Abstract:

The honey bee queen diet is a white creamy substance consists mainly of proteins, sugars and lipids called royal jelly. It's one of the most valued products of honey bee colonies, it secreted in the acceptance royal cub. This study aimed to investigate some factor affecting the acceptance mean and royal jelly quantity production**.** Twelve Italian and Carniolan honey bee hybrid colonies (six/each) were used through two years (2017-2018). The Italian hybrid recorded the highest acceptance means (64.0). As the highest royal jelly quantity mean (148.1 mg/cup) recorded in the Carniolan hybrid. Also, the second year recorded the highest acceptance mean (61.8) more than the first years. As the lower bar showed the highest royal jelly quantity means (154.2 mg/cup)**.** It could be concluded that, the best hybrid for producing royal jelly was the Carniolan while the Italian hybrid has the highest acceptance mean.

Introduction

The honey bee queen is an integral part of a normal honey bee colony, the singularity of its presence makes its a subject of special scientific interest of equal or greater importance is that the queen performs the major female reproductive role in the colony and therefore it is essential for the welfare and development of a normal colony. The relationship between the queen's reproductive capacity and honey production in the colony is extremely important to the bee keeping industry itself (J. rangel *et al.,* 2013). The queen also controls the production of queen cells and laying workers by means of her scent and the secretion of "queen substances" from her mandibular glands. The amount of produced royal jelly increased by increasing the number of grafted queen cell cups to reach the maximum. Thus a fuller know ledge of queen rearing is desirable in order to understand her physiology, functions, and performance. The queen rearing industry is an essential part of commercial beekeeping in Egypt every year, thousands of queens are reared and used to found new colonies for honey, royal jelly, which is secreted from the

hypopharyngeal gland and mandibular gland of the worker honey bee, is the exclusive food for the queen honey bee and larvae (Satomi *et al*., 2004). Royal jelly has an important commercial appeal and nowadays it is used in many industries like food and pharmaceutical characteristics (Sabatini *et al*., 2009) as anti-tumor (Tamura and Kuboyama, 1987), anti-bacterial (Sauerwald *et al.,* 1998), anti-hypercholesterolemic (Nakajin *et al*., 1982), anti-allergic (Kataoka *et al*., 2001 and Oka *et al*., 2001), anti-fatigue (Kamakura *et al*., 2001), insulin-like (Okuda *et al*., 198) , wound-healing properties (Fujii *et al.*, 1990) and wax production. Therefore, it has become increasingly necessary to better understand the factors which influence the production of high quality queens so that recommendations can be made to queen breeders concerning the best queen rearing procedures to follow. The purpose of this paper is to study the efficacy of Italian and Carniolan honey bee hybrids rearing colonies under grafting bar's level on royal jelly production and the queen cub acceptance rates.

Material and methods

The experiments were conducted under the conditions of Kafr Elsheikh Governorate during two years of 2017 and 2018. Twelve honey bee colonies were used in the experiment. Six colonies for each hybrid (Italian and Carniolan).

The bee colony under the study was as follows:

1. Choose the parent colony of the Italian and Carniolan bee hybrid to lay the eggs between the folded brood tablets to force the queen to lay eggs and follow until the hatching was completed three days later. The fourth day is the first larval age (larvae of one day) which takes the shape of the crescent.

2. Processing of breeding frames with three bars arranged in three different locations (upper, middle, bottom) each frame carrying 15 plastic cups. The breeding frame is then exposed to the breeding colony for two hours before the grafting.

3. Colonies : Each colony has 8 comps covered with bee divided as follows: five sealed brood comps plus three honey and pollen comps plus plastic honey bee feed.The queens of the breading colony were removed for 48 hours. The method of Doolittle was obtained in 1909 - wet method of grafting (1 gram of royal food: 1 cm distilled water).

4. Grafting: The one day larvae were transferred into the plastic cup by the grafting needle and then the breeding frame that carried three wooden par placed between the sealed brood comps in the breeding chamber. The Italian grafting larvae was rearing in Italian colony and the Carniolan grafting larvae was rearing in Carniolan colony.

5. The nutrition: Sugar solution with concentrate of 1kg sugar: 1.5 water was used. Energized feed was done before grafting. Each colony fed on half a liter of the solution every three days until the experiment was finished.

On the day following the grafting, we collect the number of acceptable royal cups and calculate the acceptance ratio. On the same date of grafting, after 72 hours the breeding frames were raising from the breeding colonies and removing the larvae from the plastic cups, then collecting the royal jelly from the successful royal cups with a wooden spoon according to the location of the the bar (upper, middle, lower). The royal jelly stored in plastic containers which were weighed empty and full with royal jelly and numbered with a code number, the capacity of each container was five grams. Each bar cubs were weighed according to its location. The royal jelly was stored in the fridge. The grafting process is repeated every three days.

Statistical analysis using Duncan's Multiple Range Test (Duncan, 1955).

Results and discussion

Effect of different hybrids within bar's level in two different years:

1. Queen acceptance:

Data in Table (1) showed the grafted queen cups acceptance mean under Italian and Carniolan hybrids rearing colony with bar's level. At the first year, the highest acceptance means were (66.1 and 56.7%) recorded in the Italian and Carniolan hybrids colonies with middle bar's level, respectively. while the lowest acceptance means were (50 and 57.9%) recorded in the Carniolan and Italian hybrids colonies with upper bar's level, respectively. At the second year, the highest acceptance percentage means were (67.6 and 62.2%) recorded in the Italian and Carniolan hybrids colonies in middle bar's level, respectively. while the lowest acceptance means were (53.1 and 59.6%) recorded in the Carniolan and Italian hybrids colonies with upper bar's level, respectively.

Table (1): Queen cups acceptance means under Italian and Carniolan hybrids rearing colony within bar's level.

Bar Level	First Year			Second Year		
	Hybrids		Mean ±S.E	Hybrids		Mean ±S.E
	Italian	Carniolan		Italian	Carniolan	
Upper	57.9	50	54±3.783	59.6	53.1	56.4±7.540
Middle	66.1	56.7	61.4±4.273	67.6	62.2	64.9±2.525
Lower	65.6	54	59.8±5.323	67.2	61.2	64.2±2.885
Mean	63.2	53.6	58.4±2.893B	64.8	58.8	61.8±21.424A

Mean / Hybrids Colony Italian 64.0±1.973A Carniolan 56.2±2.147B

Mean in each factor designated by the same letter are not significantly different at 5 % level using Duncan's Multiple Range Test.

The results concluded that, the Italian hybrid has acceptance mean more than the Carniolan with significant differences between them. As the middle bar showed the highest acceptance mean with significant differences between it and upper bar and non- significant differences between middle and lower bars. Moreover, the second year showed the highest acceptance mean with significant differences between first and second years. Many researchers had discussed these findings , ex. Sharaf El-Din *et al*. ,2000; Osipitan *et al*. ,2012 and Shah ,2000.

2. The royal jelly quantity:

Data in Table (2) showed the royal jelly quantity mean (mg/cups) from bar's level in Italian and Carniolan hybrids rearing colony under queenright and queenless colonies. At the first year, the highest royal jelly quantity means were (152.8 and 152) recorded in the Italian and Carniolan hybrids colonies with lower bar's level respectively. while the lowest royal jelly quantity means were (128 and 134.5) recorded in the Carniolan and Italian hybrids colonies with upper bar's level respectively. At the second year, the highest royal jelly quantity means were (157.4 and 151) recorded in the Carniolan and Italian hybrids colonies with lower bar's level respectively. while the lowest royal jelly quantity means were (143 and 146.9) recorded in the Italian and Carniolan hybrids colonies with upper bar's level respectively.

The following conclusions can be drawn; the Carniolan hybrid has royal jelly quantity means more than the Italian with non-significant differences between them. As the lower bar showed the highest royal jelly quantity means with non-significant differences between treatments. Additionally, the second year showed the highest royal jelly quantity means with significant differences

between first and second years. Many researchers had discussed these findings and found that, there are several factors affecting royal jelly production (Pahinler, 2005 and Sharaf El-Din, 2010). The most important of them are the age of transferred larvae (Sahinler and KaftanoÛlu, 1997), feeding (Fuhai *et al*., 1993), number of transferred queen cell cups (Pahinler and Pahinler, 2002), harvesting interval (Ali, 1994 and Sharaf El-Din, 2010), whether the colony is queenless or queenright (Van-Toor and Littlejohn, 1994), age of nurse workers (Ali, 1994) and bee race (Shibi *et al*., 1993 and Lian-Fei *et al.* ,2016).

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Table (2): Royal jelly quantity mean (mg/cups) from bar level in Italian and Carniolan hybrids rearing colony under queenright and queenless colonies.

Bar Level	First Year			Second Year		
	Hybrids		Mean ±S.E	Hybrids		Mean ±S.E
	Italian	Carniolan		Italian	Carniolan	
Upper	134.5	128	131.3±4.233	143	146.9	146.0±4.705
Middle	149.9	151.5	150.7±2.176	149.9	153	151.4±3.143
Lower	152.8	152	152.4±3.585	151	157.4	154.2±3.646
Mean	145.7	143.8	144.8±5.007B	148	152.4	150.2±2.832A

Mean / Hybrids Colony Italian 146.8±3.408A Carniolan 148.1±4.966A

Mean in each factor designated by the same letter are not significantly different at 5 % level using Duncan's Multiple Range Test.

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Susceptibility of certain cantaloupe hybrids to major pests infestation in Sohag Governorate

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Aphid, whitefly, mite, leafminer, population density, cantaloupe, hybrids, susceptibility, yield and hairs.

Abstract:

Cantaloupe (*Cucumis melo* var. cantalupensis, galia type) is an important cucurbitaceous vegetable crop sown in Egypt. The present study was conducted at Shandweel Agricultural Research Station, Sohag Governorate during two summer successive seasons of 2017 and 2018 to investigate the population density of the aphid**,** *Aphis gossypii* Glover (Hemiptera: Aphididae); the whitefly**,** *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae); the spider mite, *Tetranychus urticae* (Koch) (Acari: Tetranychidae) and the leafminer**,** *Liriomyza trifolii* (Burgess) (Diptera: Agromyzidae) on cantaloupe crop and the susceptibility of ten local cantaloupe hybrids to infest by the previous pests, in addition to determine some yield characters. Also, the effect of hair density on the infestation with piercing sucking pests was studied. Data revealed significant differences between the tested hybrids in case of all pests and all horticultural characters during two successive seasons. It was evident that number of hairs on cantaloupe leaves had positive effect on the aphid and two spotted spider mite, and negative effect on whitefly**.** The present study can be suggest that the best hybrids are Shahd Zaman followed by Hybrid No. 8 and Hybrid No. 22, because of their high yield and their moderate infestation by most pests. This recommendation was built up on the data of two seasons under experimental conditions at Sohag Governorate .

Introduction

Cantaloupe (*Cucumis melo* var. cantalupensis, galia type) is one of the most important and popular fruity vegetable grown in Egypt. The cultivated area reached 69,000 feddan. Most the mentioned area is cultivated with imported seeds paid foreign hard currency. Improving of local cantaloupe hybrids with good fruit quality and high yield under high temperature at Sohag conditions is very important (El-Murabaa, 1971 and Shalaby, 1975). However, this vegetable is liable to attack by several pests, i.e., the aphid**,** *Aphis gossypii* Glover (Hemiptera: Aphididae); the whitefly**,** *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae); the spider mite,

Tetranychus urticae (Koch) (Acari: Tetranychidae) and leafminer**,** *Liriomyza trifolii* (Burgess) (Diptera: Agromyzidae) that reduce its quality and quantity. (Ibrahim, 2005; Gameel, 2013 and Abdel-Rahman *et al*., 2016). To avoid the damage caused by previous pests and decrease using pesticides, the use of resistant hybrids is the ideal alternative as a component of Integrated Pest Management programs. Many authors searched the effect of plant resistance on the level of plant infestation. Boissot *et al.* (2003) identified the resistance in 80 C. melo genotypes for *B. tabaci* infestation. Metwally *et al.* (2013) studied the susceptibility of six cantaloupe cultivars to infestation by *B. tabaci* and *T. urticae*. Abdallah (2015) studied the control of *T. urticae* on three melon cultivars. De Oliveira *et al.* (2017) evaluated 54 accessions and four commercial hybrids of melon in regard to resistance to leafminer. The morphological features of the plant such as leave hairs and trichomes play important role on water control and resistance against herbivory in some plants (Cipollini and Bergelson, 2002; Molina-Montenegro *et al.*, 2006 and Gonzales *et al.*, 2008). The simple trichomes of these genotypes probably act as mechanical barriers that hinder insect movement and/or feeding (Le Roux *et al.*, 2008), also, chemical compounds in glandular trichomes can be deterrent or toxic to several herbivores (Buta *et al.*, 1993). So, the current study was mostly built up to select the best hybrids which gave high yield and most resistant to aphid, whitefly, spider mite and leafminer infestation as one of the control options to suppress heavy infestation in integrated pest management programs. Also, the population density and the effect of hair density on the infestation with the perivous pests was studied.

Materials and methods

The present study was conducted at Shandweel Agricultural Research Station, Sohag Governorate during two successive summer seasons of 2017 and 2018.

1.Population density:

To study the population density of the main pests attacking cantaloupe plants, an area of about 54 m^2 was divided into plots of equal size (18 m^2) and was sown with cantaloupe (Shahd Zaman) on March 30th in 2017 and 2018 seasons and three replicates were used. The normal agricultural practices were performed and no insecticides used.

2.Susceptibility of different cantaloupe hybrids to infestation of pests:

Ten cantaloupe hybrids namely, Super Quality, Yathreb 100, Hybrid No. 2, Hybrid SQ, Hybrid No. 100, Hybrid No. 22, Hybrid No. 7, Shahd Zaman, Hybrid No. 3 and Hybrid No. 8 were sown to study their susceptibility to infest by *A. gossypii*, *B. tabaci*, *T. urticae* and *L. trifolii* during summer seasons of 2017 and 2018. Seeds were sown on March 30th in randomized complete block with three replicates, each replicate contained 10 experimental plots. Each plot was presented by three beds, 1.5 m width, 4 m length (18 m^2) and the plants were spaced at 50 cm. Land preparation fertilizer application and other field practices were carried out according to recommendation of Egyptian Ministry of Agriculture and all plots were kept without any pesticidal treatments.

3.Collecting data:

3.1.Pests data:

Depending on timing of infestation, initiation of counts varied for each trial. One hour after the sunrise, adults of whitefly were counted weekly, on abexial surface of ten randomly leaves/ plot in the field. After that, samples of ten leaves/ plot were randomly chosen, then transferred in polyethylene bags to laboratory. The population of *B. tabaci*

(nymphs and eggs) and *T. urticae* (adults, nymphs and eggs) were estimated by counting numbers per two square inches. However, the total individuals of *A. gossypii* were determined by counting the total number per the whole underside leaf, also, the number of mines of *L. trifolii* was recorded.

3.2.Leave hair density:

To estimate leave hair density, 30 leaves were randomly taken from three plant levels (upper, middle and lower) from each plot, transferred to laboratory, then the number of hairs on the abaxial leave surface was counted in a 1 mm^2 area using a compound microscope during the second season.

3.3.Horticultural data:

3.3.1.Main stem length:

A random sample of ten plants from each plot was used for evaluating main stem length (cm, 60 days after sowing).

3.3.2.Flowering date:

Ten plants were chosen for every hybrid from each replicate and the number of days from sowing to anthesis of first female flower was determined.

3.3.3.Number of fruits per plant:

It measured as an average of fruits of 10 randomly chosen plants per plot.

3.3.4.Fruit weight:

A random sample of ten plants from each plot was used for evaluating average fruit weight (g).

3.3.5.Total yield:

It was weight of all fruits harvested at the yellow netted ripe stage from 10 randomly chosen plants per plot.

4.Statistical analysis:

Statistical analysis was conducted by using one – way analysis of variance. 'F' test used to evaluate the differences significance between haybrids and mean separation was conducted using the least significant difference (L.S.D.) procedure at P = 5% (Snedecor and Cochran, 1971). To determine the effect of leaf trichome density on the level of infestation by *A. gosypii*, *B. tabaci* and *T. urticai*, the simple correlation (r) was also used (Gomez and Gomez, 1984).

Results and discussion

1. Population density:

1.1. *Aphis gossypii*:

Data illustrated in Figure (1) demonstrated that the infestation of this aphid started after 15 days from sowing date in both seasons. Aphids population increased gradually to reach its peaks of 132.3 and 23.3 aphid/ 10 leaves in 13th May and 10th June, respectively, in 2017 season, however, one peak was recorded in 2018 season, in 6th May by 118,7 aphids / 10 leaves. After that the population decreased gradually to the end of the two seasons. These results are in agreement with those of Abou El-Saad (2015) and Ibrahim *et al*. (2017).

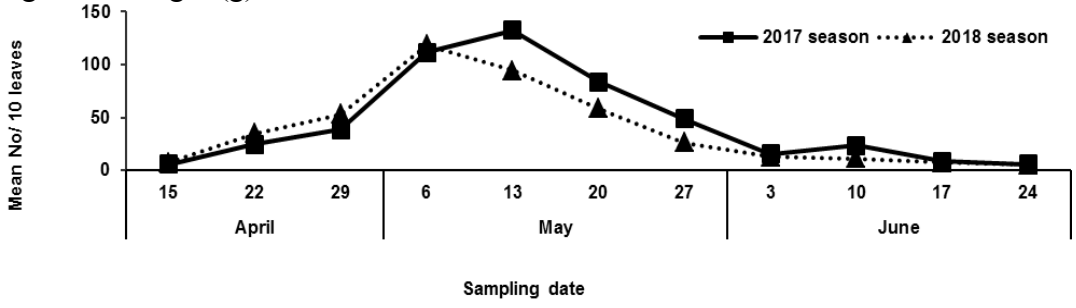

Figure (1): Population density of Aphis gossypii on cantaloupe crop in Sohag Governorate during 2017 and 2018 seasons.

1.2.*Bemisia tabaci*:

In general, the three stages of the whitefly , *B. tabaci* (adult, nymph and egg) started to take place from the first week of inspection in both seasons of the study (Figures, 2 and 3). Adults formed 2 and 3 peaks in 2017 and 2018 seasons, respectively. The peaks were 397.0 and

100.0 adults/ 10 leaves in 29th April and 27th May, respectively in 2017 season and 314.0, 340.0 and 220.3 adults/ 10 leaves in 22nd April and 6th and 27th May, respectively, in 2018 season. However, the nymphs recorded 4 peaks of activity in both seasons, in 22nd April, 13th and 27th May and 10th June with 262.3, 327.7, 267.7 and 365.3 nymphs/ 10 leaves, respectively, in 2017 season, and in 22nd April, 6th and 27th May and 10th June with 148.0, 300.7, 351.3 and 447.3 nymphs/ 10 leaves, respectively, in 2018 season. Meanwhile, two peaks were observed for egg stage during the two seasons. The peaks were recorded in 22nd April and 13th May with 260.7 and 157.3 eggs/ 10 leaves, respectively, in 2017 season and 240.0 and 158.7 eggs/ 10 leaves, respectively, in 2018 season. Then, the population decreased to the end of the two seasons. These findings are in partial agreement with the results of Metwally *et al.* (2013); Abou El-Saad (2015) and Hegab (2017).

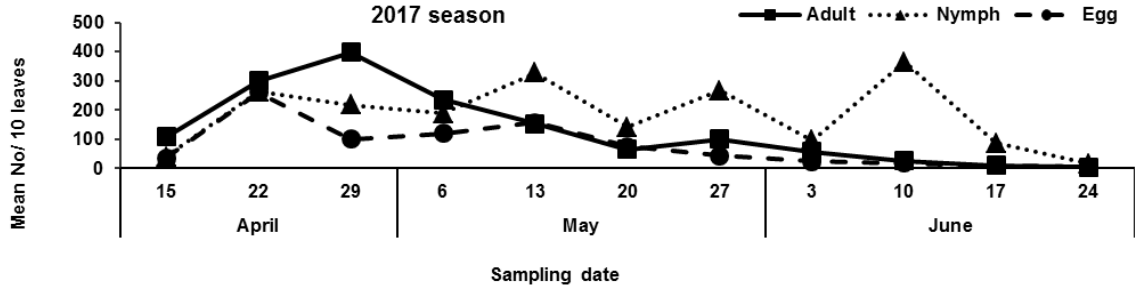

Figure (2): Population density of *Bemisia tabaci* adult, nymph and egg stages on cantaloupe crop in Sohag Governorate during 2017 season.

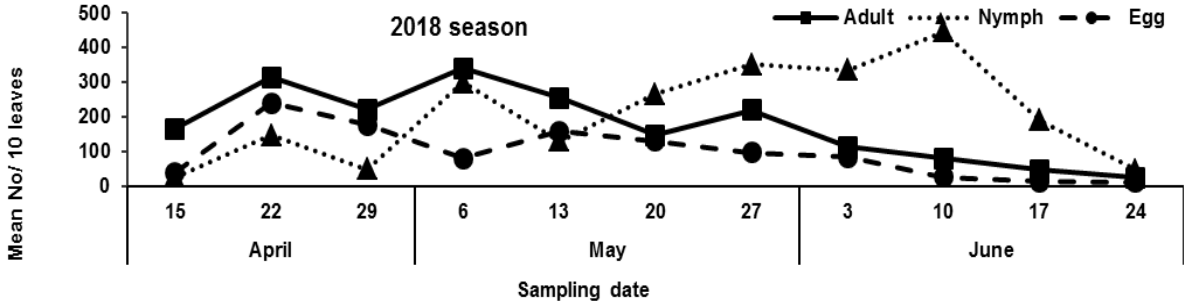

Figure (3): Population density of *Bemisia tabaci* adult, nymph and egg stages on cantaloupe crop in Sohag Governorate during 2018 season.

1.3.*Tetranychus urticae*:

Data in Figures (4 and 5) generally indicated that *T. urticae* (mobile and egg stages) started to take place from the first week of inspection in both seasons of the study. Two and three peaks of activity were found for mobile stages in 2017 and 2018 seasons, respectively. The peaks were recorded in 22nd April and 20th May with 72.3 and 31.7 individuals/ 10 leaves, respectively, in 2017 season. However, the mean numbers of 28.7, 38.3 and 29.7 individuals/ 10 leaves were recorded as peaks in 29th April, 20th May and 3rd June, respectively, in 2018 season. Then the population decreased to the end of the two seasons. On the other hand, egg stage number recorded two peaks in both seasons. The peaks were recorded in 6th and 20th May with 60.0 and 47.3 eggs/ 10 leaves, respectively, in 2017 season. While, in 2018 season, the peaks were observed in 6th May and 3rd June with 30.7 and 45.3 eggs/ 10 leaves, respectively. Then the population decreased to the end of the two seasons. The same results were obtained by Metwally *et al.* (2013); Aiad *et al.* (2014) and Abou El-Saad (2015).

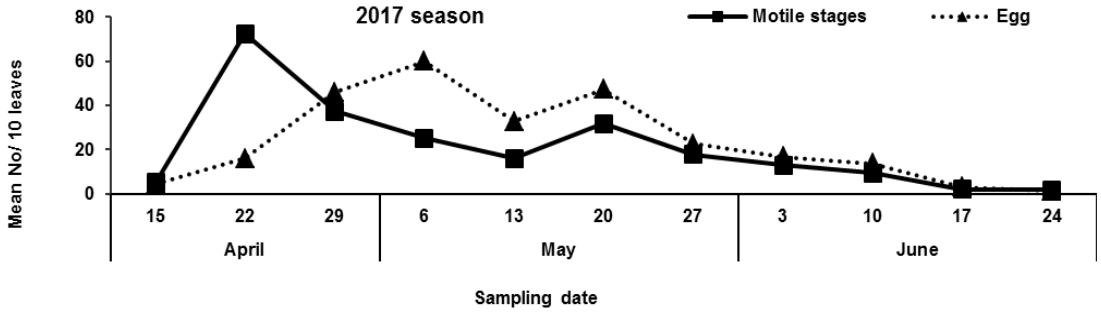

Figure (4): Population density of *Tetranychus urticae* mobile and egg stages on cantaloupe crop in Sohag Governorate during 2017 season.

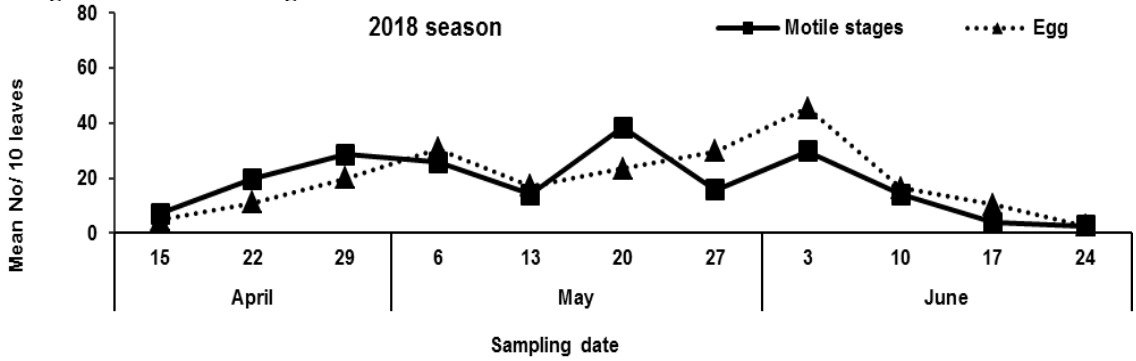

Figure (5): Population density of *Tetranychus urticae* mobile and egg stages on cantaloupe crop in Sohag Governorate during 2018 season.

1.3.*Lirimyza trifolii*:

Data illustrated in Figure (6) clearly indicated that the leafminer, *L. trifolii* population (determined as number of mines) attacked cantaloupe leaves from the first week of inspection. The number of mines peaked three and two times in 2017 and 2018 seasons, respectively. These peaks were observed at 6th and 20th May and 3rd June with 21.7, 18.7 and 21.7 mines/ 10 leaves, respectively, during the first season. While, in the second season, the mean numbers of 20.0 and 27.7 mines/ 10 leaves were detected as peaks in 29th April and 3rd June, respectively.

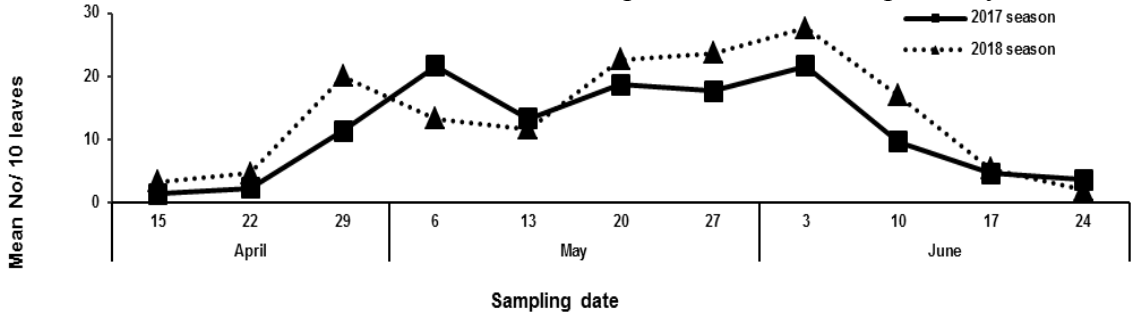

Figure (6): Population density of *Liriomyza trifolii* (Mines) on cantaloupe crop in Sohag Governorate during 2017 and 2018 seasons.

2.The susceptibility of ten cantaloupe hybrids to pests:

Data in Tables (1 and 2) showed that the mean numbers of aphid, whitefly (adult, nymph and egg), spider mite (mobile stages and egg) and leafminer (mines) populations on the ten tested cantaloupe hybrids in 2017 and 2018 seasons, respectively. Statistical analysis of data revealed significant differences between mean numbers of the previous pests populations on cantaloupe hybrids during two successive seasons.

2.1. *Aphis gossypii*:

According to the mean number of aphids, Hybrid SQ showed the lowest mean number of aphids with 22.76 aphids/ 10 leaves, while, the highest mean was observed in Yathreb 100 hybrid with 41.52 aphids/ 10 leaves followed insignificantly by Super Quality Hybrid in 2017 season. In 2018 growing season, the lowest and the highest mean number of aphid were recorded in Hybrid No. 22 and Yathreb 100 hybrid, respectively, with 16.61 and 39.58 aphids/ 10 leaves, respectively, with insignificant difference between the first one and Hybrid SQ. The other hybrids arranged between the lowest and the highest susceptible hybrids in both seasons of the study. The same results were obtained by Rashwan (2015) on melon and Hegab (2017) on cucumber.

2.2. *Bemisia tabaci*:

For adults, it is clear that, Super Quality was the most resistant hybrid to *B. tabaci* adult in both seasons, followed insignificantly by Hybrid No. 2 in 2017 season and Hybrid SQ in 2018 season. On the other hands, Shahd Zaman was the most infested hybrid in both seasons, followed insignificantly by Hybrid No. 22 and Hybrid No. 100 in the first season. In case of *B. tabaci* nymphs, Hybrid No. 7 was the lowest infested one in both seasons, followed insignificantly by Yathreb 100 in both seasons and Hybrid No. 8 in 2018 season. While, Hybrid No. 2 was found to be the most infested one in both seasons, followed insignificantly by Hybrid No. 100 in both seasons, and Hybrid SQ in 2018 season only. The lowest mean number of *B. tabaci* eggs was recorded in Shahd Zaman, followed insignificantly by Hybrid No. 7 and Yathreb 100 in 2017 season, and in Hybrid No. 7 in 2018 season. However, Hybrid No. 2 recorded the highest number in both seasons, with no significantly differences between the last and Hybrid No. 3 in both seasons and Hybrid No. 22 in the second season. Many authors had attention to whitefly resistance; Boissot *et al.* (2003) screened the resisitance in 80 *Cucumis melo* genotypes to *B. tabaci* adults and nymphs. They found that the studied genotypes differed significantly in order of adult and nymphs infestation. Also, Metwally *et al.* (2013) studied the susceptibility of six cantaloupe cultivars to *B. tabaci*.

2.3. *Tetranychus urticae*:

In the first season, Hybrids Super Quality, Hybrid No. 2 and Hybrid No. 8 were the lowest infested with mobile stages of *T. urticae*, however, Hybrid SQ was the highest infested. In the second season, Hybrid No. 8 was the lowest infested followed insignificantly by Hybrid No. 7, however, Hybrids Yathreb 100, Hybrid SQ and Hybrid No. 100 was the highest infested and the other hybrids arranged between the highest and the lowest. For *T. urticae* egg stage, data showed that the lowest mean number was observed in Hybrid No. 8 followed insignificantly by Hybrid Super Quality in both seasons. While, the highest mean number of egg was recorded in Hybrid SQ followed insignificantly by Hybrid Yathreb 100 in both seasons of the study. Similar findings were reported by Metwally *et al*. (2013), who mentioned that the six tested cantaloupe cultivar varied significantly in respect of *T. urticae* mobile stages. Also, Aiad *et al.* (2014) found that Galia2 and Ananas France were the highest and the lowest infested by *T. urticae*. In (2015), Abdallah concluded that cultivating the melon Shahd cultivar is preferable than Ananas or Galia cultivar.

2.4. *Lirimyza trifolii*:

It is clear that Hybrid No. 22 was the lowest infested in both seasons, followed insignificantly by Super Quality, Yathreb 100 and Hybrid SQ in the second season. On the other hand, the most susceptible hybrid was Shahd Zaman in both seasons. The susceptibility of rest arranged between

the lowest and the highest hybrids. Similar findings were obtained from De Oliveira *et al*. (2017) who found that the evaluated melon genotypes widely varied in regard to resistance to vegetable leafminer. They suggested that the lighter melon leaves are less oviposited by vegetable leafminer. Also, Celin *et al*. (2017) determined the resistance to leafminers in 52 melon accessions and 4 commercial hybrids as controls. They identified four resistant genotypes to leafminers, CNPH 11-1072 and CNPH 11-1077 (by antixenosis), and CNPH 00-915(R) and 'BAGMEL 56(R) (by antibiosis).

Table (1): Susceptibility of ten cantaloupe hybrids to infestation of pests in Sohag Governorate during 2017 season.

Hybrids	Mean No./ 10 leaves						
	Aphis gossypii	*Bemisia tabaci*			*Tetranychus urticae*		*Liriomyza trifolii*
		Adult	Nymph	Egg	Mobile	Egg	
Super Quality	39.06 **ab**	114.67 **e**	201.18 **cd**	101.52 **c**	15.09 **f**	18.06 **e**	5.36 **c**
Yathrib 100	41.52 **a**	136.42 **b**	147.61 **f**	83.52 **e**	25.30 **b**	34.24 **a**	5.58 **c**
Hybrid No. 2	34.70 **cd**	120.79 **de**	317.45 **a**	123.76 **a**	15.76 **f**	22.12 **cd**	5.58 **c**
Hybrid SQ	22.76 **g**	129.03 **bc**	298.27 **b**	104.91 **c**	27.73 **a**	35.00 **a**	5.27 **c**
Hybrid 100	31.70 **de**	151.24 **a**	301.76 **ab**	103.03 **c**	22.24 **c**	26.21 **b**	5.58 **c**
Hybrid No. 22	27.73 **f**	155.79 **a**	184.70 **e**	117.09 **b**	20.30 **d**	24.97 **bc**	4.55 **d**
Hybrid No. 7	29.58 **ef**	130.36 **b**	136.88 **f**	82.24 **e**	17.21 **e**	21.82 **d**	6.97 **b**
Shahd Zaman	37.48 **bc**	158.15 **a**	187.82 **de**	78.58 **e**	22.55 **c**	22.79 **cd**	10.36 **a**
Hybrid No. 3	29.58 **ef**	135.48 **b**	214.09 **c**	121.39 **ab**	20.21 **d**	23.27 **bcd**	7.15 **b**
Hybrid No. 8	32.52 **de**	122.55 **cd**	185.27 **de**	90.73 **d**	15.61 **f**	17.67 **e**	5.33 **c**
F. value	**28.51***	**36.38***	**143.37***	**67.37***	**153.33***	**34.66***	**51.15***
L.S.D.	**3.16**	**7.45**	**16.18**	**6.01**	**1.04**	**2.98**	**0.69**

Means followed by different subscript letters within columns are significantly different from each other (P < 0.05).

Table (2): Susceptibility of ten cantaloupe hybrids to infestation of pests in Sohag Governorate during 2018 season.

Hybrids	Mean No./ 10 leaves						
	Aphis gossypii	*Bemisia tabaci*			*Tetranychus urticae*		*Liriomyza trifolii*
		Adult	Nymph	Egg	Mobile	Egg	
Super Quality	31.33 **b**	119.33 **g**	221.76 **bc**	110.12 **c**	12.15 **bc**	13.18 **ef**	8.18 **cd**
Yathrib 100	39.58 **a**	132.21 **de**	158.42 **de**	95.12 **d**	17.58 **a**	28.94 **a**	8.30 **cd**
Hybrid No. 2	29.97 **bc**	128.39 **ef**	349.52 **a**	127.70 **a**	11.82 **bc**	17.30 **cd**	9.18 **bc**
Hybrid SQ	18.06 **ef**	116.52 **g**	309.88 **a**	107.82 **c**	17.58 **a**	30.88 **a**	8.27 **cd**
Hybrid 100	25.48 **cd**	163.73 **b**	320.61 **a**	112.70 **bc**	17.00 **a**	19.42 **bc**	8.97 **bc**
Hybrid No. 22	16.61 **f**	160.36 **b**	192.00 **cd**	120.21 **ab**	13.45 **b**	20.55 **b**	7.79 **d**
Hybrid No. 7	23.79 **de**	134.67 **cd**	140.67 **e**	80.91 **e**	10.67 **cd**	16.21 **de**	10.09 **b**
Shahd Zaman	31.52 **b**	169.33 **a**	192.42 **cd**	95.27 **d**	13.06 **b**	17.06 **cd**	12.18 **a**
Hybrid No. 3	26.88 **bcd**	138.94 **c**	237.73 **b**	125.09 **a**	13.58 **b**	18.67 **bcd**	10.03 **b**
Hybrid No. 8	29.15 **bcd**	126.79 **f**	143.79 **e**	96.42 **d**	9.61 **d**	12.09 **f**	9.15 **bc**
F. value	**12.21***	**137.28***	**25.03***	**28.43***	**15.88***	**33.30***	**11.27***
L.S.D.	**5.74**	**4.77**	**45.26**	**8.40**	**2.13**	**3.15**	**1.15**

Means followed by different subscript letters within columns are significantly different from each other (P < 0.05).

3. Hairs density:

Data in Table (3) showed the hair density on upper, middle and lower levels and mean of ten cantaloupe hybrids leaves during 2018 season. It is obvious that as age of the leaves increase, increase in the size of leaves and decreases the hair density. The analysis of data showed that the differences between the tested hybrids are significant at the three plant levels and their mean. The highest and density of hairs was found on leaves of Shahd Zaman in upper plant level, and on leaves of Yathreb 100 in middle and lower plant levels and mean number. However, the lowest density of hairs was found on leaves of Hybrid No. 3 in all plant levels and mean number. While the rest hybrids arranged between the highest and the lowest values. Data in Table (3) showed that the number of

hairs on cantaloupe leaves had weak, positive and insignificant effect on the aphid, *A. gossypii* population in all plant levels. Doryanizadeh *et al*. (2017) demonstrated that antixenosis of *Cucumis* correlated positively with leaf trichome density. On contrary, the relationship between the whitefly, *B. tabaci* and hair density was negative, varied according to the pest age and the plant level. The correlations between mean number of whitefly adult and nymph on side and hair density on the other side were found to be negatively weak and insignificant in all plant levels. Values of correlation coefficient between mean number of whitefly egg and number of hairs on upper plant level and mean of hairs were significantly and insignificantly negative, respectively (r= -0.7282* and -0.5420, respectively).

Concerning the influence of hair density on cantaloupe infestation with mite (Table, 3), the correlation coefficient values clear positive relationships, almost, varied according to the pest age and the plant level. Significantly positive correlation values was found between the numbers of mite mobile stages and hair density in middle and lower plant levels (r= 0.6329* and 0.6179*, respectively), while, insignificantly positive correlation value was recorded in upper plant level and mean (r= 0.2018 and 0.4892, respectively). For the effect of hair density on the number of mite egg, similar results were obtained. The correlation coefficient values, 0.2882, 0.7192*, 0.7690* and 0.6067 were recorded in upper, middle and lower plant levels and mean, respectively. Van Haren *et al*.(1987) stated that the leaf hairs and trichomes have an imprtant effect on searching ability of predator mite, *Phytoseiulus persimilis* Athias-Henriot (Acari: Phytoseiidae) on *T. urticae* on tomato. These findings are in harmony with Ibrahim *et al*. (2008) who revealed that the highest population of mite was recorded on Sudanian watermelon which has thick hairy leaves, whereas squash leaves which nearly hairless recorded the lowest population.

Table (3): Density of hairs on lower surface of ten cantaloupe hybrids leaves and their correlation with the pests in Sohag Governorate during 2018 season.

Hybrid		Mean No./ 1 mm^2/ 10 leaves			
		Upper level	Middle level	Lower level	Mean
Super Quality		248.89 **e**	101.44 **de**	22.89 **ef**	124.41 **de**
Yathreb 100		471.44 **abc**	336.67 **a**	235.78 **a**	347.96 **a**
Hybrid No. 2		280.78 **de**	200.67 **bc**	144.56 **b**	208.67 **cd**
Hybrid SQ		456.44 **bcd**	293.89 **ab**	170.56 **b**	306.96 **ab**
Hybrid No. 100		323.78 **cde**	192.11 **cd**	83.89 **c**	199.93 **cd**
Hybrid No. 22		177.57 **e**	99.89 **de**	52.44 **cde**	109.96 **e**
Hybrid No. 7		517.78 **ab**	144.78 **cd**	44.00 **def**	235.52 **bc**
Shahd Zaman		644.00 **a**	214.11 **bc**	46.33 **de**	301.48 **ab**
Hybrid No. 3		177.56 **e**	26.00 **e**	11.33 **f**	71.63 **e**
Hybrid No. 8		262.22 **e**	122.00 **cde**	63.33 **cd**	149.19 **cde**
F. value		**6.57***	**8.25***	**42.80***	**9.90***
L.S.D.		**182.47**	**97.32**	**33.00**	**87.57**
Correlation coefficient for					
Aphis gossypii		0.2476	0.2910	0.2926	0.3148
Bemisia tabaci	Adult	0.1668	-0.1128	-0.3084	-0.0243
	Nymph	-0.2129	0.1640	0.2268	-0.0057
	Egg	**-0.7282***	-0.3305	-0.0712	-0.5420
Tetranychus urticae	Motile stages	0.2018	**0.6329***	**0.6179***	0.4892
	Egg	0.2882	**0.7192****	**0.7690****	0.6067

Means followed by different subscript letters within columns are significantly different from each other (P < 0.05).

4. Horticultural data:

The mean numbers of main stem length and some yield component recorded in ten local cantaloupe hybrids are presented in Tables (4 and 5). In general, the data in previous Tables indicated that the tested local cantaloupe hybrids differed significantly for all studied characters in both seasons.

4.1.Main stem length:

The highest main stem length was recorded in Shahd Zaman in both seasons, respectively. However, the lower values were recorded in Hybrid No. 3 and Super Quality in both seasons, respectively, with insignificant difference between them. The rest hybrids arranged between the highest and the lowest values.

4.2.Flowering date:

The tested hybrids can be arranged in two significant groups, the earliest one contained Yathreb 100, Hybrid No. 2, Hybrid SQ, Hybrid No.100 and Hybrid No. 7, however, the latest one consisted of Super Quality, Hybrid No. 22, Shahd Zaman, Hybrid No. 3 and Hybrid No. 8 in the two seasons.

4.3.Number of fruits per plant:

The highest number of fruits per plant was obtained from Hybrid No. 3 Hybrid No. 8 in both seasons, respectively. While, the lowest number was recorded in Yathreb 100 in both seasons. The rest hybrids arranged between the highest and the lowest values.

4.4.Fruit weight:

The highest weight of the fruit was observed in Shahd Zaman in both seasons, followed insignificantly by Hybrid No. 22 and Hybrid No. 8 in the second season. While, the lowest weight was recorded in Super Quality and Hybrid SQ in both seasons, respectively, with insignificant differences between them. The rest hybrids arranged between the highest and the lowest values.

4.5.Total yield:

The highest and the lowest yield per plant were observed in Shahd Zaman and Super Quality, respectively, in both seasons. While, the rest hybrids arranged between them in both seasons. These results are in agreement with Hussien and Selim (2014) who studied 20 different hybrids of cantaloupe, resulting from 5x5 diallel under high temperature conditions, and reported that average fruit weight (gm) ranged from 558 to 986 gm for 25 genotypes, and the best crosses for total yield ranged from 12.2 to 13.7 ton/ feddan, while, premo commercial had 18.8 ton/ feddan.

Also, Hussien and Selim (2015a) found that average fruit weight ranged from 465 gm to 955 gm, while total yield ranged from 9.55 to 20.60 ton/ feddan, when evaluated four commercial hybrids. Hussien and Selim (2015b) studied the productivity and fruit quality of local resources of sweet melon, and found that the highest value for total yield was 23.7 ton/ feddan.

Depending on the susceptibility of ten local cantaloupe hybrids to aphid, whitefly, spider mite and leafMiner and their yield component, the present study can be suggest that the best hybrids are Shahd Zaman followed by Hybrid No. 8 and Hybrid No. 22, because of their high yield and their moderate infestation by most pests. This recommendation was built up on the data of two seasons under experimental conditions in Sohag Governorate .

Table (4): Mean number of main stem length and some yield component of ten loacal cantaloupe hybrids at Sohag Governorate during 2017 season.

	Main stem length (cm)	Flowering date (days)	Number of fruits/ plant	Fruit weight (gram)	Yield/ plant (KG.)
Super Quality	95.78 **e**	35.33 **a**	2.28 **d**	369.44 **g**	0.84 **e**
Yathrib 100	119.44 **c**	33.33 **b**	2.27 **d**	570.00 **c**	1.29 **d**
Hybrid No. 2	118.78 **c**	33.00 **b**	2.30 **d**	541.11 **cd**	1.24 **d**
Hybrid SQ	96.00 **e**	32.00 **b**	2.60 **c**	381.67 **fg**	0.99 **e**
Hybrid 100	104.78 **d**	33.00 **b**	2.70 **bc**	533.33 **cde**	1.43 **d**
Hybrid No. 22	115.22 **c**	35.67 **a**	2.31 **d**	779.44 **b**	1.80 **c**
Hybrid No. 7	130.78 **b**	33.00 **b**	2.90 **ab**	462.22 **def**	1.34 **d**
Shahd Zaman	179.00 **a**	35.33 **a**	2.80 **bc**	875.56 **a**	2.45 **a**
Hybrid No. 3	90.00 **e**	35.00 **a**	3.10 **a**	446.11 **efg**	1.38 **d**
Hybrid No. 8	131.67 **b**	35.33 **a**	2.77 **bc**	766.22 **b**	2.12 **b**
F. value	**122.43***	**5.97***	**9.94***	**35.31***	**40.62***
L.S.D.	**6.94**	**1.64**	**0.28**	**88.28**	**0.23**

Means followed by different subscript letters within columns are significantly different from each other ($P < 0.05$).

Table (5): Mean number of main stem length and some yield component of ten loacal cantaloupe hybrids at Sohag Governorate during 2018 season.

	Main stem length (cm)	Flowering date (days)	Number of fruits/ plant	Fruit weight (gram)	Yield/ plant (KG.)
Super Quality	93.56 **d**	35.67 **a**	2.49 **de**	395.00 **c**	1.01 **d**
Yathrib 100	127.78 **bcd**	33.00 **b**	2.34 **e**	583.33 **bc**	1.38 **cd**
Hybrid No. 2	123.78 **bcd**	33.00 **b**	2.39 **e**	547.78 **bc**	1.31 **cd**
Hybrid SQ	101.89 **cd**	32.33 **b**	2.60 **cde**	392.22 **c**	1.02 **d**
Hybrid 100	110.89 **bcd**	33.00 **b**	3.00 **abc**	542.78 **bc**	1.65 **bcd**
Hybrid No. 22	123.89 **bcd**	36.00 **a**	2.44 **e**	787.22 **a**	1.95 **abc**
Hybrid No. 7	137.67 **bc**	33.00 **b**	3.08 **ab**	477.22 **bc**	1.48 **cd**
Shahd Zaman	184.11 **a**	35.67 **a**	2.86 **bcd**	852.78 **a**	2.44 **a**
Hybrid No. 3	95.33 **d**	35.67 **a**	2.71 **bcde**	457.22 **c**	1.27 **cd**
Hybrid No. 8	139.67 **b**	35.00 **a**	3.39 **a**	672.22 **ab**	2.28 **ab**
F. value	**4.50***	**10.30***	**6.22***	**5.67***	**4.66***
L.S.D.	**37.53**	**1.37**	**0.41**	**196.22**	**0.68**

Means followed by different subscript letters within columns are significantly different from each other ($P < 0.05$).

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Effect of different grafting larvae genotype under cub's position and bar's level on acceptance percentages and royal jelly quantity mean

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Abstract:

Royal jelly is a yellowish-white, creamy, acidic secretion, with a slightly pungent odor and taste, produced by the hypopharyngeal and mandibular glands of worker honey bees [*Apis mellifera* L. (Hymenoptera :Apidae)]. In this study, the effect of the different grafting larvae genotype under different cub's position and bar's level on the acceptance percentages mean and the royal jelly produced quantity mean were examined. The fourth day is the first larval age (larvae of one day) which takes the shape of the crescent was used for grafting. Two different grafting larvae genotype were used (Italian and Carniolan hybrids). The Italian grafting larvae, middle bar and the medium cubs have the highest acceptance percentages mean more than the other treatments (71.3). As the Italian grafting larvae, right cubs and the middle bar have the highest royal jelly quantity mean more than the other treatments (167.2). For the best results of high acceptance percentage and the highest royal jelly quantity mean the Italian hybrid grafting larvae and were recommended. As the middle bar should be used for the best results.

Introduction

The queen bee holds the most important position in a colony. The performance of a honey bee colony is the result of its queen's function as well as of that of the drones that mated with her. Commercialization of queen breeding requires the mass production of large numbers of high quality queens (Büchler *et al*., 2013). Periodical requeening with young queens less than one year old, results in more honey production than colonies headed by old queens. Moreover, the loss of a queen represents a serious threat to the survival of the honey bee colony and beekeepers frequently require new queens to start new colonies and replace dead or failing queens. Royal jelly is widely consumed in the community and has perceived benefits ranging from promoting growth in children and improvement of general health status to enhancement of

longevity for the elderly (Leung *et al*. ,1997). Royal jelly has a much larger market in Asia than in the USA or Europe, and in Asia it is commonly found in products including cosmetics, food supplements, and beverages and is used in commercial medical products (FAO, 2007). Rearing of a quality queen is depends on many factor, the most important of which is the queen cub acceptance percentage and the produced of royal jelly quantity mean. The aim of this work is to study the effect of different grafting larvae genotype under cub's position and bar's level on acceptance percentages mean and the royal jelly quantity mean.

Material and methods

The experiments were conducted under the conditions of Kafr Elsheikh Governorate during year of 2017. Twelve honey bee colonies were used in the experiment of the Italian (six replicates) and Carniolan (six replicates) honey bee hybrids

The bee colony under the study was as follows:

1. Choose the parent colony of the Italian and Carniolan honey bee hybrids to lay the eggs between the brood frames to force the queen to lay eggs and follow until the hatching is completed three days later. The fourth day is the first larval age (larvae of one day) which takes the shape of the crescent was used.

2. Processing of grafting frames with three strips, 7 cm away from each other arranged in three different locations (upper, middle, lower) and each frame carrying 45 plastic cups (fifteen cups/strip). The rearing frame is then exposed to the rearing colony two hours before the grafting. Each grafting larvae genotype was rearing in the same rearing colony genotype.

Each colony has 8 comps covered with bee divided as follows: five sealed brood comps plus three honey and pollen comps + plastic honey bee feed.

The queens of the breading colony were removed for 48 hours. The method of Doolittle was obtained in 1909 - wet method of grafting (1 gram of royal food: 1 cm distilled water).

3. Grafting: The one day larvae were transferred into the plastic cup by the grafting needle and then the breeding frame that carried three wooden par placed between the sealed brood comps for both the queenless and queenright in the breeding chamber. The Italian grafting larvae was rearing in Italian colony and the Carniolan grafting larvae was rearing in Carniolan colony.

4. Nutrition: Sugar solution with concentrate of 1 Kg. sugar: 1.5 water was used. Energized feed was done before grafting. Each colony fed on half a liter of the solution every three days until the experiment was end.

On the day following the grafting, the number of acceptable royal cups were collected and the acceptance percentage were calculated. On the same date of grafting, after 72 hours the breeding frames were raising from the breeding colonies and removing the larvae from the plastic cups, then collecting the successful royal cups with a wooden spoon according to the location of the bar (upper, middle, lower). The royal jelly stored in plastic containers which were weighed empty and full with royal jelly and numbered with a code number, the capacity of each container was five grams. Each bar cubs were weighed according to its location and bar's level. The royal jelly was stored in the fridge. The grafting process is repeated every three days. Statistical analysis using Duncan's Multiple Range Test (Duncan, 1955).

Results and discussion

Effect of different grafting larvae within cub's position and bar's level:

1.Acceptance percentages mean:

Data in Table (1) showed that the grafted queen cups acceptance percentages mean

of using Italian and Carniolan grafting larval in cups position and bar level. In the Italian larvae, the highest acceptance percentages mean was (71.3, 70.4 and 57.9%) recorded in the medium, right and left cubs position with middle bar respectively. while the lowest acceptance percentages mean was (48.7, 62.8 and 64.8%) recorded in the left, right and medium cubs position with upper bar respectively. In the Carniolan larvae, the highest acceptance percentages mean were (65.2, 63.3 and 50.1%) recorded in the medium, right and left cubs position with middle, middle and lower bars respectively. while the lowest acceptance percentages mean were (44.9, 56.1 and 56.3%) recorded in the left, right and medium cubs position with upper bar respectively.

Table (1): Grafted queen cups acceptance percentages mean using Italian and Carniolan grafting larval in cups position and bar level.

Bar Level	Acceptance percentages						
	Italian larvae			AV±S.E	Carniolan larvae		
	Cub position				Cub position		
	Left	Medium	Right		Left	Medium	Right
Upper	48.7	64.8	62.8	58.8±5.120	44.9	56.3	56.1
Middle	57.9	71.3	70.4	66.5±4.477	50.1	65.2	63.3
Lower	57.9	71.2	70.4	66.5±4.344	50.5	62.7	60.1
Mean	54.9	69.1	67.8	63.9±2.961A	48.5	61.4	59.8

Mean / cups position on stripe Left 51.7±2.448C Medium 65.3±22.586A Right 63.8± 2.552B
Mean in each factor designated by the same letter are not significantly different at 5 % level using Duncan's Multiple Range Test.

2. The royal jelly quantity mean:

Data in Table (2) showed the grafted queen cups royal jelly quantity mean of using Italian and Carniolan grafting larval in cups position and bar level. In the Italian grafted larvae, the highest royal jelly quantity means were (167.2, 160 and 139.8) recorded in the right, medium and left cubs position with middle, middle and lower bars respectively. while the lowest royal jelly quantity means were (123.7, 143.9 and 145.7) recorded in the left, right and medium cubs position with upper bar respectively. In the Carniolan grafted larvae, the highest royal jelly quantity means were (162.1, 153.5 and 137.8) recorded in the right, medium and left cubs position with lower, middle and lower bars respectively. While the lowest royal jelly quantity means were (126, 145.1 and 145.2) recorded in the left, medium and right cubs position with upper bar respectively.

Table (2): Royal jelly quantity mean (mg/cups) using Italian and Carniolan grafting larval in cups position and bar level.

Bar Level	Royal jelly quantity mean (mg/cups)						
	Italian larvae			AV±S.E	Carniolan larvae		
	Cub position				Cub position		
	Left	Medium	right		Left	Medium	Right
Upper	123.7	145.7	143.9	138.1±7.936	126	145.1	145.2
Middle	139.8	160	167.2	155.7±8.524	134.3	156.4	158.9
Lower	140.2	157.7	154.2	150.7±6.198	137.8	153.5	162.1
Mean	134.6	154.8	155.1	148.2±5.119A	132.7	151.7	155.4

Mean / cups position on stripe Left 133.7±3.576B Medium 153.3±3.395A Right 155.3± 4.263A
Mean in each factor designated by the same letter are not significantly different at 5 % level using Duncan's Multiple Range Test.

From the obtained data the following conclusions can be drawn; The Italian grafting larvae has acceptance percentages mean more than the Carniolan with significant differences between both. As the medium cubs showed the highest acceptance percentages mean with significant differences between all cub's position. Additionally, the middle bar showed the highest acceptance percentages mean with significant differences between upper and middle/lower bars respectively. The Italian grafting larvae has royal jelly quantity mean more than the Carniolan with non-significant differences between both. As the right cubs showed the highest royal jelly quantity mean with significant differences between it and left cub's position. Additionally, the middle bar showed the highest royal jelly quantity mean with significant differences between upper and middle/lower bars respectively. Many researchers had discussed these findings i.e. Weiss (1967) , Garcia and Nogueira-Couto (2005), Sahinler and Kaptanoglu (2005) , Albarracin *et al*. (2006) , Macicka (1985), Sarling (1992), Ali (1994), Ibrahim (1997), Li (2000) , Shah (2000), Albarracin *et al.* (2006), El-Barbary (2007) and Sharaf El –Din (2010).

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An annotated list of species of Genus *Encarsia* in Egypt with new records of species and hosts (Hymenoptera: Chalcidoidea: Aphelinidae: Coccophaginae)

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Abstract:

Species of the genus *Encarsia* Förster (Hymenoptera: Chalcidoidea: Aphelinidae: Coccophaginae) are some of the most widely used and successful entomophagous biological control agents of whiteflies (Aleyrodidae) and armored scale insects (Diaspididae). This paper includes an annotated list of 22 species of the genus *Encarsia* as well as new hosts and distribution records for these species in Egypt.

Introduction

Encarsia Förster (Hymenoptera: Chalcidoidea: Aphelinidae: Coccophaginae) is the largest genus of the family Aphelinidae and currently contains 439 valid species known from all of the major zoogeographic regions of the world (Noyes, 2016). Most species of *Encarsia* are primary endoparasitoids of Aleyrodidae and Diaspididae and a few species are parasitoids of hormaphidine aphids or eggs of Lepidoptera (Polaszek, 1991). Diagnostic characters for the genus *Encarsia* include: all tarsi 5-segmented (tarsus of middle leg with 4 segments in certain species); antenna 8-segmented in females and 7-8 segmented in males, and usually is not spindle-shaped; submarginal vein with 1-3 setae (usually 2); scutellum with 2 pairs of setae; each axilla with 1 seta; hypopygium of female not prominant, usually not extending to the apex of the gaster; most species are parasitoids of whiteflies or armored scale insects, and a few species are parasitoids of hormaphidine aphids, eggs or other hosts. Species of the genus *Encarsia* are some of the most widely used and successful entomophagous biological control agents. *Encarsia formosa* Gahan (Hymenoptera: Aphelinidae) has been used to control *Trialeurodes vaporariorum* Westwood (Hemiptera : Aleyrodidae), a serious pest of numerous horticultural plants in greenhouses. The aim of the present work is to report on a survey of the genus *Encarsia* in Egypt and provide an annotated list of the species and their hosts that are known to occur in Egypt.

Materials and methods

Specimens of this faunistic research were collected from different localites in Egypt by rearing the puparia of the whiteflies under optimum conditions (28±3 ºC, 70±5 RH%, 16: 8 L: D). Specimens were dissected and mounted dorsally in Canada balsam on

slides following the method of Noyes (1982). All of the material examined is deposited in the PPRI (Plant Protection Research Institute), Dokki, Giza, Egypt.

Results and discussion

An annotated list of the species of *Encarsia* known to occur in Egypt.

Genus *Encarsia* Förster

Currently, there are 22 species of the genus *Encarsia* known to occur in Egypt including a new distributional record of *Encarsia cibcensis* Lopez Avila and *Encarsia gennaroi* Pedata and Giorgini in Egypt. The distribution of each species in Egypt as well as the hosts they have been recorded on are given below. One asterisk (*) is used to indicate a new host record and 2 asterisks (**) are used to indicate a new distribution record of the parasitoid species in Egypt.

1. *Encarsia acaudaleyrodis* Hayat

Distribution: New Valley Governorate .

Hosts: *Tetraleurodes leguminicola* (Hemiptera: Aleyrodidae).

Remarks: This species was reported for the first time in Egypt by Polaszek *et al*. (1999).

2. *Encarsia aurantii* (Howard)

Distribution: Beheira, Fayoum, Ismailia, Matruh, Northern Coast, Qalyubiya and Sharqiya Governorates.

Hosts: *Aonidiella aurantii*, *Aulacaspis tubercularis* * (new host in Egypt and world), *Aonidiella orientalis*, *Hemiberlesia lataniae* and *Parlatoria oleae* (Hemiptera: Diaspididae).

Remarks: This species was recorded for the first time in Egypt by Hafez (1988).

3. *Encarsia berlesei* (Howard)

Distribution: Alexandria Governorate .

Hosts: *Pseudaulacaspis pentagona* (Hemiptera : Diaspididae).

Remarks: This species recorded for the first time in Egypt by Priesner and Hosny (1940).

4. *Encarsia citrina* (Craw)

Distribution: Cairo, North Sinai (El-Arish), Giza and Qalyubiya Governorates.

Hosts: *Aonidiella aurantii, Aulacaspis tubercula*ris* (new host in Egypt and world), *Aspidiotus. hedreae*, *Chrysomphalus aonidum, C. dictyospermi, Lepidosaphes gloveri*, *L. beckii*, *Lindingaspis floridana* and *Parlatoria ziziphi* (Hemiptera : Diaspididae).

Remarks: This species was recorded for the first time in Egypt by Priesner and Hosny (1940).

5. *Encarsia cibcensis* Lopez Avila**

Distribution: Qalyubiya Governorate.

Hosts: *Aleuroclava psidii** (new host in Egypt and world) (Hemiptera: Aleyrodidae)

Remarks: This is the first record of *E. cibcensis* parasitizing this host species.

6. *Encarsia davidi Viggian*i and Mazzon

Distribution: Aswan, Giza, Qalyubiya, North and South Sinai Governorates.

Hosts: *Acaudaleyrodes rachipora*, *Aleurolobus marlatti*, *Aleyrodes proletella*, *Bemisia tabaci* and *Siphoninus phillyreae* (Hemiptera: Aleyrodidae).

Remarks: This species was first recorded in Egypt by Polaszek *et al.* (1999).

7. *Encarsia elegans* Masi

Distribution: Assiut, Aswan, El-Minya, Cairo**, Giza**, Sharqiya and North Sinai Governorates.

Hosts: *Aleurolobus marlatti, A. olivinus* and *Bemisia tabaci* (Hemiptera: Aleyrodidae).

Remarks: This species was first recorded in Egypt by Priesner and Hosny (1940).

8. *Encarsia formosa* Gahan

Distribution: Beni-Suef, El-Minya**, Qalyubiya and Gharbiya Governorates.

Hosts: Bemisia tabaci and Trialeurodes ricini (Hemiptera: Aleyrodidae.(

Remarks: This species was recorded for the first time in Egypt by Abd-Rabou (1998).

9. *Encarsia galilea* Rivnay and Gerling

Distribution: North Sinai (El-Arish) and South Sinai ** Governorates.
Hosts: Siphoninus phillyreae (Hemiptera: Aleyrodidae).
Remarks: This species was recorded for the first time in Egypt by Abd-Rabou (1998).

10. *Encarsia gennaroi* Pedata and Giorgini
Distribution: Demmyate** and Behira** Governorates.
Hosts: *Bemisia tabaci, Trialeurodes vaporariorum*. (Hemiptera: Aleyrodidae).
Remarks: This species was recorded for the first time in Egypt by Simmons *et al.* (2002) as *Encarsia pergandiella;* however in a revision of the *Encarsia pergandiella* species complex by **Gebiola *et al.* (2017)**, they described *E. gennaroi*, which was introduced from California to Europe (Italy and Israel) in 1979 to control *Trialeurodes vaporariorum*; it is the only known species in this group known from the Western Palearctic region and is likely to be the species occurring in Egypt, especially since **Gebiola *et al.* (2017)** cited **Abd-Rabou's (2006)**, paper on the introduction of *Encarsia pergandiella* into Egypt in the discussion of *E. gennaroi.*

11. *Encarsia inaron* (Walker)
Distribution: Assiut, Behira**, Beni-Suef, Cairo, Kafr El-Sheikh, North Sinai and Qena Governorates.
Hosts: *Acaudaleyrodes rachipora*, *Siphoninus phillyreae, Aleyrodes proletella, Bemisia tabaci* and *Trialeurodes ricini* (Hemiptera: Aleyrodidae).
Remarks: This species was recorded for the first time in Egypt by Priesner and Hosny (1940).

12. *Encarsia lahorensis* (Howard)
Distribution: Qalyubiya Governorate .
Hosts: *Dialeurodes citri* (Homoptera: Aleyrodidae).
Remarks: This species was recorded for the first time in Egypt by Abd-Rabou (1997).

13. *Encarsia lounsburyi* (Berlese and Paoli)
Distribution: Assiut, Aswan, Beni-Suef, El-Minya, Fayoum, Giza and Sohag Governorates.
Hosts: *Aonidiella aurantii , Aspidiotus nerii , Duplachionaspis natalensis, Chrysomphalus aonidum, C. dictyospermi, Diaspis echinocacti, Fiorinia fioriniae*, *Hemiberlesia cyanophylli*, *H. lataniae, Lepidosaphes beckii, L. pallidula*, *Lineaspis striata* and *Mycetaspis personata* (Hemiptera: Diaspididae).
Remarks: This was recorded for the first time in Egypt by Priesner and Hosny (1940).

14. *Encarsia lutea* Masi
Distribution: Aswan, Alexandria, Behira, Cairo, Daqahilya, Giza and Qalyubiya Governorates.
Hosts: *Aleurolobus marlatti, Bemisia. tabaci, Parabemisia myricae*, *Siphoninus phillyreae* and *Trialeurodes ricini* (Hemiptera: Aleyrodidae).
Remarks: This species was recorded for the first time in Egypt by Abd-Rabou (1998).

15. *Encarsia mineoi* Viggiani
Distribution: Cairo, Giza, Qalyubiya and Sharqiya Governorates.
Hosts: *Acaudaleyrodes rachipora, Bemisia tabaci* and *Trialeurodes ricini* (Hemiptera: Aleyrodidae).
Remarks: This species was recorded for the first time in Egypt by Polaszek *et al.* (1992).

16. *Encarsia olivina* (Masi)
Distribution: North Sinai (El-Arish) and Fayoum** Governorates.
Hosts: *Aleurolobus olivinus* (Hemiptera:Aleyrodidae).
Remarks: This species was recorded for the first time in Egypt by Abd-Rabou (2000).

17. *Encarsia perconfusa* Evans and Abd-Rabou

Distribution: Aswan, Qena and New Valley Governorates.

Hosts: *Tetraleurodes leguminicola* (Hemiptera:Aleyrodidae).

Remarks: This species was recorded for the first time in Egypt by Evans and Abd-Rabou (2005).

18. *Encarsia perniciosi* (Tower)

Distribution: Sharqyia Governorate .

Hosts: *Lepidosaphes pallidula* (Hemiptera: Diaspididae).

Remarks: This was recorded for the first time in Egypt by Evans and Abd-Rabou (2005).

19. *Encarsia protransvena* Viggiani

Distribution: Qalyubiya Governorate .

Hosts: *Dialeurodes citri* (Hemiptera : Aleyrodidae)

Remarks: This species was recorded for the first time in Egypt by Abd-Rabou (1997).

20. *Encarsia ramsesi* Polaszek

Distribution: New Valley Governorate.

Hosts: *Ramsesseus follioti* (Hemiptera: Aleyrodidae).

Remarks: This species was recorded for the first time in Egypt by Polaszek *et al. (*1999).

21. *Encarsia sophia* (Girault and Dodd)

Distribution: Demyaate, Kafr El-Sheikh, Qalyubiya and Sharqyia Governorates.

Hosts: *Bemisia tabaci and Pealius mori* (Hemiptera : Aleyrodidae)

Remarks: This species was recorded for the first time in Egypt by Abd-Rabou (1998) as a *Encarsia transvena*.

22. *Encarsia strenua* (Silvestri)

Distribution: Qalyubiya and Behira ** Governorates .

Hosts: *Dialeurodes citri* (Hemiptera: Aleyrodidae)

Remarks: This species was first recorded in Egypt by Abd-Rabou (1998).

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Impact of ecological aspects on pests infesting tomato varieties and their control measure

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Abstract:

Tomato, *Lycopersicon esculentum* (Solanacea) is one of the most important vegetable crop in Egypt. Two Field experiments were conducted during two winter successive seasons of 2013/2014 and 2014 /2015 at Gizera Bele, Banha, Qalyubiya Governorate . The study included population fluctuation, the population growth rate of the whitefly**,** *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae); the aphid**,** *Aphis gossypii* Glover (Hemiptera: Aphididae) ; the spider mite, *Tetranychus urticae* (Koch) (Acari: Tetranychidae) and evaluation the susceptibility degree of three varieties of tomato (omnia, arika and safira) for infestation by previously mentioned pests. The study, also included the control of these pests by sesame oil; biofly (*Beauveria bassiana*) , abamactin and malathion. The obtained results revealed that the highest mean population fluctuation (for 15 weeks) of *B. tabaci* (22.1, 61.2, 88.6) and (10, 17.7, 28.9) nymph and adult stages/ 5 leaves, followed by mean population of *T.urticae* (14.9, 5.3, 26.3) and (10.7, 9.0, 14.9) individuals/ 5 leaves of the three tomato varieties omnia, arika and safira during the two seasons, respectively, while population of *A. gossypii* was lowest mean population. Also the results revealed clearly susceptibility degree of safira variety was susceptible to infest with the above mentioned pests of tomato plants during the two seasons, with mean 58.73 , 10.6 and 20.6 (individual) / 5 leaves , respectively . Present data indicated that *B. tabaci* recorded the highest growth rate (23.64 and 4.57), taking time difference 14 days at safira variety in the first and second season, respectively. Our data exhibited, sesame oil and abamactin gave the significant highest reduction against *B. tabaci, A. gossypii* and *T. urticae* on tomato plants, as the average of their reductions after 14 days of spraying were (80.9% , 78.1% , 69.8%) and (77.6, 86.3, 71.4%), respectively .

Introduction

Tomato, *Lycopersicon esculentum* (Solanacea), is economically one of the most important and widely grown vegetables in the world vegetables (Polston and Anderson, 1999 and Peralta and Spooner, 2007), ranking second in importance next to potato (FAO STAT, 2005). Tomato is a good source of all nutrients especially vitamin C, B and K. Tomato plants are subject to infestation by the piercing sucking pests, such as the whitefly**,** *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae); the aphid**,** *Aphis gossypii* Glover (Hemiptera: Aphididae) and the spider mite, *Tetranychus urticae* (Koch) (Acari: Tetranychidae) (Ahmed, 2000). *B. tabaci* is one of the most important pests of tomato, it sucks the plant sap (Schuster *et al*., 1996) reducing the quality and quantity of the sap (Mound, 1965). This insect exists as an economic pest in most places of the world (Martin, 1987; Byrne and Houk, 1990 and Gerling, 1990). This pest also transmits various viral diseases (Bedford *et al.,* 1994 and Jones, 2003).

Aphids feed through phloem tissue. Those are important pests on agricultural crops (Blackman and Eastop, 2000). Aphids cause many losses on numerous crops and about of 13% agricultural outputs were recorded to be lost by insect pests (Van Emden and Harrington, 2007 and Faria *et al*., 2007). Economic agricultural losses resulted from aphid feeding, which returned to deficiency of essential plant nutrients through the plant development. Rapid reproduction of aphids is due to parthenogenetic reproductions which produce high population densities. While, winged aphids infest new host plants (Powell *et al*., 2006). In addition, aphids feeding, also, allow transition of more than 275 viruses; aphids cause also insufficiency of photosynthesis by producing honeydew on the leaf surface (Miles, 1989 and Sylvester, 1989).

The tetranychid mite species feed on the plant sap injuring the epidermis resulting in blotching, stippling or bronzing causing serious damage (Park and Lee, 2002). The mites consumed nearly all the chlorophyll causing decrease in leaves vitality and lead to a reduction or damage the crop. *T. urticae* causes much indirect damage by transmitting viral and fungal pathogens (Park and Lee, 2007).

The cultivation of pest-resistant plants is one way to counter pests. Resistant genotype can affect the morphology, biology and physiology of pests and can play a part in reducing the population of pests (Toscano *et al.,* 2002; Fancelli *et al*., 2003; Cunha *et al*., 2005; Bogorni and Vendramim 2005 and Baldin *et al*., 2007).

Pesticides produced from natural products have been recently attracting the attention of many scientists to avoid the problems caused by synthetic compounds they are deeply interested in their chemical constituents and biological properties (Abou-Yousef *et al.,* 2010).The significance of botanical pesticides/plant extracts is highly recognized in the field of agriculture as botanical pesticides are cheap, safe , hazardless, non-residual and highly effective.

The objectives of this research were to:

1. Study the population fluctuation and the population growth rate of *B. tabaci* , *A. gossypii* and *T. urticae* on three varieties of tomato.
2. Evaluate susceptibility degree of three varieties of tomato to infest by *B.tabaci* , *A. gossypii* and *T. urticae* .
3. Evaluate the efficacy of sesame oil, biofly (*Beauveria bassiana*) and abamactin (Cormat 1.8% E.C) in comparison to malathion 57% E.C (Coromandel)) against some pests on susceptible tomato variety in open field.

Materials and methods

Two field experiments were conducted at Gizera Bele, Banha, Qalyubiya Governorate . The first one conducted to study the population fluctuation, susceptibility degree and the population growth rate to *B.tabaci* , *A. gossypii* and *T. urticae* infestation on three tomato varieties. At second experiment the susceptible variety were used in evaluation the efficiency of some tested materials, against the three previous pests.

The first experiment was conducted for two successive winter seasons throughout 2013/ 2014 and 2014/2015 seasons. An area of about 525 m^2 was cultivated with the three tomato varieties (omnia, arika and safira) in 26 th and 30th of October during 2013/2014 and 2014/2015 seasons; respectively. The whole area was divided in 9 replicates, (each replicate of 58 m^2). Each variety was represented by 3 replicates. All the experimental area received the recommended and standard cultivation practices. The total area was kept free from any pesticides application. Weekly randomized samples continue for 15 weeks, sampling of 5 leaves were randomly taken from each replicate then each sample was kept in a tightly closed paper bag and transferred to the laboratory in the same day for inspection under stereomicroscope to count the numbers of *B.tabaci* (nymphs), *A. gossypii* (nymphs and adults) and *T. urticae* (Individuals) and direct count of the whitefly adults numbers was done in the field on random samples of 5 leaves.

For parameters, maximum population size and growth rate for *B.tabaci* , *A. gossypii* and *T. urticae* were recorded for three varieties tomato and the time taken to reach the maximum count (N_t) were used for comparing between varieties tomato. Population growth rate (GR) was calculated by using Odum's equation (Odum, 1971) as follow;

$GR= (N_t – N_\circ)/ \Delta t$

Where N_t = the number of each pest recorded at the maximum count of the population on a plant.

$N_\circ$= the initial number of each pest released on each plant.

Δt= the difference in time between N_t and $N_\circ$.

The classification the susceptibility degree of each variety to infestation with the previously mentioned pests was dependent on the general mean number ($\bar{X}$) of each pest and the standard deviation (SD) as reported by Chiang and Talekar (1980) The varieties that:-

Highly susceptible (HS) : had an average numbers of pest more than $\bar{X}$ +2SD

Susceptible (S): had an average numbers of pest between $\bar{X}$ and $\bar{X}$ +2S

Low resistant (LR): had an average numbers of pest between $\bar{X}$ and $\bar{X}$ -1SD

Moderately resistant (MR): had an average numbers of pest between $\bar{X}$ -1SD and $\bar{X}$-2SD

Highly resistant (HR): had an average numbers of pest less than $\bar{X}$ -2SD.

The second experiment was conducted during the second season 2014/2015 to evaluate the efficiency of four treatments, sesame oil (*Sesamum indicum* L.) (Pedaliaceae) was purchased from El-Captain Company (CAP PHARM) for extracting oils, Natural plantsand COSMECICS. Egypt; malathion 57% E.C (Coromandel); biofly (*Beauveria bassiana*) and Cormat 1.8% E.C (Abamactin), with rate of 500 ml, 500 ml, 425 ml and 40 ml /100 L water ,respectively. Sesame oil was formulated by addition of Pril detergent at 1% in water. Water was used as controls (or untreated plants) safira variety which infested by the highest numbers of these pests used in this experiment.

An area of about 1125 m^2 was cultivated with the tomato variety (safira) in 30 th and 25th of October during 2013/2014 and 2014/2015 seasons; respectively. The whole area was divided into 15 replicates (75 m^2 for

each replicates). Each treatment was represented by three replicates and control. All the normal of agricultural practices for tomato variety (safira) cultivation were followed except pesticidal treatment. The chosen treatments were sprayed in 1st of December during 2014/2015 by using a 20 L. knapsack sprayer with one nozzle.
The efficiency of treatments was determined by inspecting 5 randomly leaves from each replicate then each sample was kept in a tightly closed paper bag and transferred to the laboratory in the same day for inspection under stereomicroscope to count the numbers of *B.tabaci* (nymphs), *A. gossypii* (nymphs and adults) and *T. urticae* (individuals). In respect to *B. tabaci* adult, the direct count done in the early morning on random samples of 5 leaves.

Inspection of plants was carried out before spraying and after 3, 7 and 14 days from application to evaluate the efficiency of sesame oil; biofly (*Beauveria bassiana*) and abamactin, while malathion after 1, 7 and 14 days on the reduction rates of the pest populations.
The reduction percentage of population (% mortality) was calculated according the equation of Henderson and Tilton (1955).

Statistical tests were performed using SAS program computer and calculated LSD (Least significant difference) to find differences between mean numbers of three pests on the three tomato varieties studied (SAS Institute, 2003).

Results and discussion

1.Population fluctuation of *Bemisia tabaci, Aphis gossypii* and *Tetranychus urticae* on three tomato varieties during two successive seasons at Qalyubiya Governorate .

1.1.First season 2013/ 2014:

1.1.1.*Bemisia tabaci*:

Data in Table (1) and Figure (1) indicated that the population fluctuation of *B. tabaci* in omnia variety had three peaks in dates 26 November and 24 December 2013 and 21 January 2014 with 87.0, 29.0 and 6.0 (nymph and adult stages) / 5 leaves. On the other hand arika and safira varieties recorded single peak at the date of 26 November 2013 as 353.0 and 3 December 2013 with 298.0 (nymph and adult stages) / 5 leaves, respectively.

1.1.2.*Aphis gossypii* :

Data in Table (1) and Figure (1) was showed that the population fluctuation of *A.gossypii* in omnia variety recorded three peaks at the dates 26 November, 17 December 2013 and 28 January 2014 with 6.0, 16.0 and 5.0 (nymph and adult stages) / 5 leaves, respectively. In the first season, arika variety recorded single peak at the date of 17 December 2013. While safira variety had two *A. gossypii* peaks at the dates of 26 November 2013 and 21 January 2014 with 10.0 and 29.0 (nymph and adult stages) / 5 leaves, respectively.

1.1.3.*Tetranychus urticae*:

Data of omnia variety in Table (1) and Figure (1) illustrated that *T.urticae* population recorded two peaks at dates 26 November and 31 December 2013 with 67.0 and 25.0 individuals/ 5 leaves.
Data of arika variety showed that single peak of *T. urticae* at date of 31/12/2013 with 55.0 individuals. Mean while safira variety showed gradually increasing in *T. urticae* tell recorded single peak at the end of season at date 11 February 2014 with 70.0 individuals/ 5 leaves.

Table (1): Counts and population growth rate of some pests on three tomato varieties during of 2013 / 2014 season.

Dat of inspection	Plant age	Number of pests /5 Leaves								
		Omnia			Arika			Safira		
		B. tabaci	*A. gossypii*	*T. urticae*	*B. tabaci*	*A. gossypii*	*T.urticae*	*B.tabaci*	*A. gossypii*	*T. urticae*
		Nymph+ Adult stages	Nymph+ Adult stages	Indiv-iduals	Nymph+ Adult stages	Nymph+ Adult stages	Indiv-Iduals	Nymph+ Adult stages	Nymph+ Adult stages	Indiv-iduals
12/11/2013	17	15	2	7	22	2	3	25	2	0
19/11/2013	24	36	2	8	56	1	10	40	5	1
26/11/2013	31	87	6	67	353	5	15	137	10	3
3/12/2013	38	54	5	48	205	8	23	298	5	12
10/12/2013	45	38	8	10	107	8	29	248	9	15
17/12/2013	52	28	16	10	70	15	32	153	10	17
24/12/2013	59	29	10	15	46	10	35	130	5	22
31/12/2013	66	17	9	25	24	9	55	94	9	25
7/1/2014	73	15	7	15	12	6	22	89	10	32
14/1/2014	80	5	7	10	10	5	10	66	18	36
21/1/2014	87	6	4	5	5	3	9	42	29	40
28/1/2014	94	1	5	2	4	2	5	4	26	42
4/2/2014	101	0	3	1	1	2	3	2	12	58
11/2/2014	108	0	1	1	0	2	0	1	9	70
18/2/2014	115	0	0	0	0	2	0	0	5	22
Mean ± SE		22.07±6.31	5.67±1.1	14.93±4.86	61±24.38	5.33±1.03	16.73±4.07	88.6±23.58	10.93±2.0	26.33±5.26

1.2.Second season 2014/ 2015:

1.2.1.*Bemisia tabaci*:

Data of omnia variety in Table (2) and Figure (1) illustrated that *B. tabaci* population recorded single peak at date of 1 December 2014 with 50.0 (nymph and adult stages) / 5 leaves. It is notice that the weekly inspections from date 12 January 2015 tell the end of season at 23 February 2015 recording Zero. Arika variety recorded single peak of *B. tabaci* population at the date 8 December 2014 with 62.0 (nymph and adult stages) / 5 leaves. Meanwhile, safira variety had two peaks of *B. tabaci* population at dates of 1 December 2014 and 12 January 2015 with 88.0 and 22.0 (nymph and adult stages) / 5 leaves, respectively.

1.2.2.*Aphis gossypii*:

Data in Table (2) and Figure (1) showed that, omnia variety had four peaks in the second season. The peak dates of omnia variety were 1 December 2014 and 5, 26 January and 16 February 2015 with 29.0, 12.0, 9.0 and 8.0 (nymph and adult stages) / 5 leaves, respectively. Also, arika variety recorded three peaks were in 1, 15 December 2014 and in 12 January 2015 with 27.0, 20.0 and 12.0 (nymph and adult stages) / 5 leaves, respectively. The three peaks of safira variety were in 1, 22 December 2014 and in 5 January 2015 with 20.0, 25.0 and 29.0 (nymph and adult stages) / 5 leaves, respectively.

1.2.3.*Tetranychus urticae*:

Table (2) and Figure (1) illustrated that *T. urticae* population of omnia and safira varieties had two peaks at dates of 8 and 29 December 2014. The two peaks of omnia variety recorded 20.0 and 33.0 individual / 5 leaves, while safira showed 22.0 and 49.0 individual / 5 leaves, respectively. On the other hand Arika variety had single peak at date of 8 December 2014 with 30.0 individual / 5 leaves. In general, the three weeks from 1 December tell 22 December 2014 had the distinct peaks of three tomato varieties for *B. tabaci* and *A. gossypii*. On the other hand *T. urticae* peaks recorded in 8 and 29 December 2014.

Table (2): Counts and population growth rate of some pests on three tomato varieties during of 2014 / 2015 season.

Dat of inspection	Plant age	Omnia			Arika			Safira		
		B. tabaci	*A. gossypii*	*T. urticae*	*B. tabaci*	*A. gossypii*	*T. urticae*	*B. tabaci*	*A. gossypii*	*T. urticae*
		Nymph+ Adult stages	Nymph+ Adult stages	Indiv-iduals	Nymph+ Adult stages	Nymph+ Adult stages	Indiv-iduals	Nymph+ Adult stages	Nymph+ Adult stages	Indiv-iduals
17/11/2014	18	6	13	2	10	10	6	24	8	7
24/11/2014	25	18	17	6	25	19	14	49	13	14
1/12/2014	32	50	29	14	61	27	15	88	20	20
8/12/2014	39	35	20	20	62	15	30	62	19	22
15/12/2014	46	23	15	10	31	20	19	38	18	12
22/12/2014	53	10	12	12	28	18	13	37	25	17
29/12/2014	60	5	10	33	16	9	10	33	20	49
5/1/2015	67	3	12	22	10	7	10	20	29	34
12/1/2015	74	0	7	15	6	12	8	22	15	12
19/1/2015	81	0	6	8	5	8	4	20	8	9
26/1/2015	88	0	9	6	3	4	3	17	1	7
2/2/2015	95	0	2	5	4	3	2	10	0	5
9/2/2015	102	0	4	4	2	1	1	8	0	3
16/2/2015	109	0	8	2	2	0	0	4	0	8
23/2/2015	116	0	4	2	1	0	0	1	0	4
Mean ± SE		10.0±3.8	11.2±1.82	10.73±2.29	17.73±5.24	10.2±2.12	9.0±2.15	28.87±6.07	11.73±2.59	14.87±3.23

2.Evaluation of relative susceptibility degree of three tomato varieties to *Bemisia tabaci, Aphis gossypii* and *Tetranychus urticae* infestation during the two seasons under consideration (2013/ 2014-2014/2015 :

2.1.*Bemisia tabaci* :

Table (3) and Figure (1) showed that omnia variety was showed moderate resistance 22.07 (nymph and adult stages) / 5 leaves in the first season meanwhile recorded a low resistance 10.0 nymphs and adults / 5 leaves in the second season, respectively. The mean susceptibility degree of the two seasons recorded moderate resistance with 16.03 (nymph and adult stages) / 5 leaves. In respect to, arika variety had susceptible in the first season and low resistance in the second season with 61.0 and 17.73 (nymph and adult stages) / 5 leaves, respectively. The mean susceptibility degree of the two tested seasons showed low resistance with 39.37 (nymphs and adults) / 5 leaves. Susceptibility degree of safira variety was susceptible in the two seasons with mean 58.73 (nymph and adult stages) / 5 leaves.

2.2.*Aphis gossypii*:

Susceptibility degree of both omnia and arika varieties was recorded low resistance in both tested seasons, with mean 7.43 and 6.80 nymph and adult stages. While safira variety was susceptible in the two seasons with mean 10.63 nymph and adult stages [Table (3) and Figure (1)].

2.3.*Tetranychus urticae* :

In respect to *T. urticae* both omnia and arika varieties had a low resistance in both seasons, with mean 12.83 and 12.87 individual/ 5 leaves, respectively. Meanwhile safira variety recoded susceptible in the two tested season, with mean 20.60 individual/ 5 leaves[Table (3) and Figure (1)].

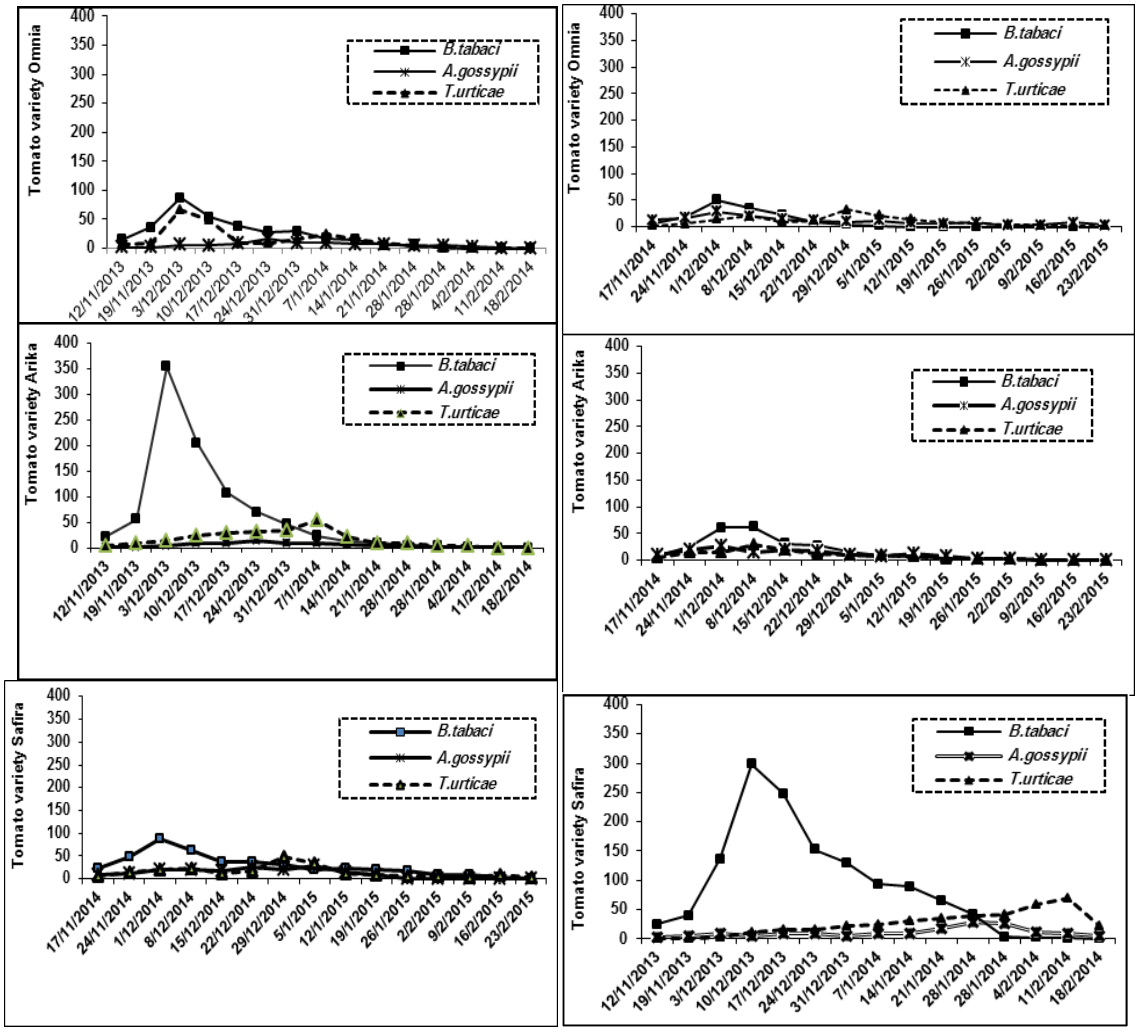

Figure (1): Counts and population growth rate of some pests on three tomato varieties during two seasons.

Table (3): Susceptibility degrees of three tomato varieties for *Bemisia tabaci, Aphis gossypii* and *Tetranychus urticae* during winter plantation of 2013/2014 and 2014/2015 seasons at Qalyubiya Governorate .

Pests varieties	*B. tabaci*						*A. gossypii*						*T. urticae*					
	1st season	S. degree	2nd season	S. degree	Mean	S. degree	1st season	S. degree	2nd season	S. degree	Mean	S. degree	1st season	S. degree	2nd season	S. degree	Mean	S. degree
Omnia	22.07	MR	10.00	LR	16.03	MR	5.67	LR	9.20	LR	7.43	LR	14.93	LR	10.73	LR	12.83	LR
Arika	61.00	S	17.73	LR	39.37	LR	5.33	LR	8.27	LR	6.80	LR	16.73	LR	9.00	LR	12.87	LR
Safira	88.60	S	28.87	S	58.73	S	10.93	S	10.33	S	10.63	S	26.33	S	14.87	S	20.60	S
Mean ± SD	57.22± 33.43		18.87± 9.48		38.04 ± 21.38		7.31± 3.14		9.27± 1.03		8.29± 2.05		19.33± 6.13		11.53±3. 04		15.43± 4.47	

Susceptible (S) = between $\bar{X}$ and $\bar{X}$ +2SD Low resistant (LR) = between $\bar{X}$ and $\bar{X}$ -1SD Moderately resistant (MR) = between $\bar{X}$ - 1SD and $\bar{X}$-2SD

3.Population growth:

Data in Table (4) is an attempt to study the population growth rate in length of recording the initial pests numbers (N_o) , maximum count of the pests populations (N_t) and the time difference them (Δt) of some serious pests that attacking some tomato varieties. Firstly, in respect to *B. tabaci*, data in Table (4) was showed that, arika

variety recorded the highest growth rate (23.64), taking time difference 14 days between initial *B. tabaci* number 22 and maximum count 353 in the first season. Data of the second season was showed the arika variety recorded the lowest growth rate 2.48 taking 21 day between the initial pest number N_o (10 nymph and adult stages) / 5 leaves) and maximum count N_t (62 nymph and adult stages) / 5 leaves). Secondly, in respect to *A. gossypii*, the three variety (omnia, arika and safira) had the lowest growth rate in the first season were 0.40, 0.37 and 0.39, respectively. Meanwhile, the safira variety recorded the lowest growth rate (0.43), where omnia and arika varieties had 1.14 and 1.21 in the second season. Thirdly, Data of *T. urticae* was showed fluctuated growth rate in the two tested seasons.

Table (4): Population growth rate of of some pests on three tomato varieties during 2013 / 2014 and 2014/2015 seasons.

Pests	Variety	First season				Second season			
		Population growth rate parameters			GR	Population growth rate parameters			GR
		N_o	N_t	Δt		N_o	N_t	Δt	
B. tabaci	Omnia	15	87	14	5.14	6	50	14	3.14
A. gossypii		2	16	35	0.40	13	29	14	1.14
T.urticae		7	67	14	4.29	2	33	42	0.74
B.tabaci	Arika	22	353	14	23.64	10	62	21	2.48
A.gossypii		2	15	35	0.37	10	27	14	1.21
T.urticae		3	55	49	1.06	6	30	21	1.14
B.tabaci	Safira	25	298	21	13.00	24	88	14	4.57
A.gossypii		2	29	70	0.39	8	29	49	0.43
T.urticae		1	70	84	0.82	7	49	42	1.00

GR= the population growth rate, N_t = the pest numbers at the maximum count of the population on a plant
N_o = the initial pest numbers on a plant, Δt = the time difference between N_o and N_t

4.Efficiency of different compounds for reducing the population density of *Bemisia tabaci, Aphis gossypii* and *Tetranychus urticae* during 2014/2015 season:

4.1.Initiale effect:

4.1.1.*Bemisia tabaci*:

In respect to initial effect (Table, 5) of the tested materials against *B. tabaci* nymph and adult, the abamactin categorized in first rank, followed by sesame oil and malathion in the second rank, while biofly occupied the third category, with % reduction were 85.7, 78.5, 78.5 and 61.9, respectively.

4.1.2.*Aphis gossypii* :

Dunkan analysis ranked the tested materials into three groups against *A. gossypii* . malathion, (sesame oil and abamactin) and biofly, where the initial reduction % recorded 100, (92.2, 90.5) and 73.3, respectively.

4.1.3.*Tetranychus urticae*:

Statistical analysis categorized the tested materials into four groups. The descending arrangements were sesame oil > abamactin > biofly > malathion, where the initial redction % showed 85.2, 79.0, 73.3 and 54.6, respectively.

4.2.Residual toxicity:

4.2.1.*Bemisia tabaci*:

It is worth to mention that sesame oil ranked in the first category after 7 and 14 days after application. The mean of residual toxicity (Table,5) of sesame oil recorded 82.1.

4.2.2. *Aphis gossypii* :

Abamactin occupied the highest reduction % after 7 and 14 days after application against *A. gossypii* , where recorded 100 and 68.3, respectively. In

respect to the mean of residual toxicity of abamactin was 84.2

4.2.3. *Tetranychus urticae*:

Sesame oil came in the first category after 7 days with reduction % was 79.8, while abamactin occupied the first category after 14 day with reduction % was 58.8. The mean of residual toxicity of abamactin was 67.7

4.3.General mean of reduction % (Table,5):

4.3.1. *Bemisia tabaci*:

The highest reduction % recorded by sesame oil and followed by abamactin with 80.9 and 77.6, respectively.

4.3.2. *Aphis gossypii* :

Abamactin and malathion showed the highest mean reduction % (86.3 and 79.9) with non significant differences.

4.3.3. *Tetranychus urticae*:

The best mean reduction % recorded by abamactin and sesame oil with 71.4 and 69.8, respectively with non significant differences.

In general the three tested tomato varieties, showed maximum population fluctuation of *B. tabaci* at the first fifth weeks from transplantation in the two growing tomato season. In the same field Metwally (1976) studied the seasonal fluctuation of *B. tabaci*, he found that the population peak took place by mid September on tomato.

In recent years, studies conducted in the field of production and use of crop varieties resistance to insects, has helped to significantly increase food production in major agricultural area. In most pests management programs the subject of plant resistance to insects (Smith *et al*., 1994 and Yasaikinici and Hincal, 1996), and the subject of the host preference of pests (Jounior *et al*., 2003) are important. If pest resistant varieties are used with chemical control methods, the costs of chemical control and problems related to insecticides which remain in the environment will be reduced. In particular, using substances of natural origin (as sesame oil in this study) in the chemical method will be very useful, because there are numerous known harmful effects of these substances on human health and animals. Consequently, the application of resistant plant varieties plays an important role in reducing environment pollution. Susceptibility degree studies of our varieties of three tomato varieties indicated that, the safira variety was susceptible for the three tested pests in both seasons. While arika variety had low resistance along the two study seasons. On the other hand omnia recoded low resistance for *A.gossypii* and *T. urticae* , while showed moderate resistance against *B. tabaci*.

Arika variety had fluctuated growth rate in the two test seasons in respect to *B. tabaci*. Safira variety recorded the lowest growth rate of *A. gossypii* during the two seasons. Lamiri *et al*. (2001) demonstrated that the insecticidal activity of an essential oil could be attributed either to the major compound present in the oil or to the synergistic and / or antagonistic effects of all the compounds of oil.

The chemical analysis of sesame oil (untabulated data) showed the components balance of sesame oil, were the Fatty acid 16.73 ug/ triolein/ ml, Triglycerides vales (212.3 mg%), total phenols values 372.3ug/ ml and Tannis values (130.3 ug tannic acid/ ml) exhibited a promise toxic effect against *B. tabaci* nymphs and adults and considerable results against *A. gossypii* and *T. urticae*. In the same field Homam and El Ghanam (2017) investigated the ovicidal effect of six plant oils, they concluded that Marjoram oil revealed the highest mortality % 89.1 against *Phthorimaea operculella* (Zeller) eggs and the predation efficiency of *Chrysoperla carnea* was 96.4 % with marjoram oil that treated *P. operculella* eggs and the predator lived for 15.3 days out of 16 days. Also, the results showed that, the abamactin recoded reduction %

Table (5): Effect of various treatments against *Bemisia tabaci, Aphis gossypii* and *Tetranychus urticae* infesting tomato variety (safira) during winter plantation at Qalyubiya Governorate .

Treatments		Mean number /5 Leaves and percent reduction at indicated periods (days)																	
		No. of	*B. tabaci*					No. of	*A. gossypii*					No. of	*T. urticae*				
	Rat/ 100L Water	Insect Pre-Spray	3	7	14	Residual Toxicity	Mean	insects Pre-Spray	3	7	14	Residual Toxicity	Mean	insects Pre-Spray	3	7	14	Residual toxicity	Mean
Sesame oil	500 ml	65	14	5	7	6	8.7	17	1	3	6	4.5	3.3	9	2	2	3	3	2.3
(*Sesamum indicum*)			78.5 b	89.1 a	75.1 a	82.1 a	80.9 a		92.2 b	81.4 b	60.8 b	71.1 b	78.1ab		85.2 a	79.8 a	44.4 c	62.1 ab	69.8 a
Bio fly	425 gm	92	35	16	13	14.5	21.3	10	2	3	4	3.5	3.0	10	4	4	3	3.5	3.7
(*Beauveria bassiana*)			61.9 c	75.3 b	67.3 bc	71.3 b	68.2 c		73.3 c	68.4 d	55.6 c	62.0 b	65.8 b		73.3 c	63.6 c	50.0 b	56.8 ab	62.3 ab
Cormat 1.8% E.C	40 ml	84	12	15	10	12.5	12.3	14	1	0	4	2.0	1.7	89	28	23	22	22.5	24.3
(Abamactin)			85.7 a	74.7 b	72.4 ab	73.6 b	77.6 ab		90.5 b	100 a	68.3 a	84.2 a	86.3 a		79.0 b	76.5 b	58.8 a	67.7 a	71.4 a
Control		88	88	62	38	50	62.7	20	15	19	18	18.5	17.3	20	30	22	12	17	21.3
Treatments	Rat/100L Water	No. of insects Pre-Spray	1	7	14	Residual Toxicity	Mean	No. of insects Pre-Spray	1	7	14	Residual Toxicity	Mean	No. of insects Pre-Spray	1	7	14	Residual toxicity	Mean
Malathion 57% E.C	500 ml	79	17	14	12	13	14.3	18	0	4	6	5	3.3	15	10	7	5	6	7.3
(Coromandel)			78.5 b	74.9 b	64.8 c	69.9 b	72.7bc		100 a	76.6 c	63.0 b	69.8 b	79.9 a		54.6 d	57.6 c	44.4 c	51.0 b	52.2 b
Control		88	88	62	38	50	62.7	20	23	19	18	18.5	20.0	20	15	22	12	17	16.3
LSD			3.261	4.738	6.679	6.771	6.464		5.069	2.977	3.882	12.62	12.82		5.242	3.919	1.883	14.12	12.22

were 77.6, 86.3 and 71.4 for *B. tabaci, A. gossypii* and *T. urticae*, respectively. Some author as, Soliman and Tarasco (2009) in Egypt stated that abamectin (Vertemic 1.8% EC) reduced significantly whitefly and aphid populations on cucumber and tomato plants, in field experiments. Siti Hajar *et al.* (2016) found that effect of malathion at 50µg/ml concentration on reproduction and feeding activity of aphids. The total number of new born nymphs produced and the relative development stage of nymphs were significantly reduced compared to untreated leaves.

This is primary study for sesame oil against some piercing sucking pests need more efforts and to apply in suitable method and tactics in the field to study its effect on predators and parasite to be become an item in integrated pest management.

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Effectiveness of microwave radiation, high temperatures and cooling degrees in control of dry wood termite, *Cryptotermes brevis* (Isoptrea: Kalotermitidae)

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Abstract:

Dry wood termite, *Cryptotermes brevis* Walker (Isoptrea: Kalotermitidae) is a serious pest attack the different kinds of hard and soft wood of wooden structures, standing trees, flooring, furniture, wooden works within buildings in different images and shapes , etc. Three different methods to control *C. brevis* termite within two kinds of wood (pine and beech) were used in this work; these are microwave radiation, high temperature and cooling degrees. The effects of these methods on the percentages of mortality are correlated with exposure times of infested wood (pine and beech) to each method. The least exposure time which gave 100% mortality differed according to used method. In microwave method, the shortest exposure time which gave 100% mortality recorded 20 sec. with power level 100 wattages for termite within pine wood while, required 80 wattages with beech wood, the longest exposure time which appeared100% mortality was 50 sec. for pine wood at 50wattages, while it was 60sec. at the power level 10 wattages for beech wood. In cooling method, the percentage of mortality recorded 100% at -27°C with 18Min.exposure time for pine wood and 20Min. for beech wood, while at -21°C the exposure time was 30 and 32Min. for pine and beech wood, respectively, whereas at -18°C the exposure time was 36 and 40Min. for pine and beech wood, respectively. In heating method, the shortest exposure time (for 100% mortality) recorded 28 Min. at 75°C for both pine and beech wood, while at 70°C the time was 34 Min. for both pine and beech wood, whereas the exposure times at 65 °C recorded 34 and 35Min.for pine and beech wood respectively. The longest exposure time reached 35Min at 60°C for two kind wood. The lethal time values (LT50 and LT95) were deterMined for the different treatments in different methods of control. The lost moisture content from infested wood exposure to both microwave irradiation and different temperature degrees was significant correlated with exposure time and showed changes in mortality percentages. Therefore, the combined effect of each temperature and lost moisture content play an important role beside exposure times and wood kind in effect on mortality of termite within infested wood.

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Introduction

Dry wood termites are social and cryptic insects which live entirely in the wood which cause significant damage to attacked wooden constructions. Previous studies by many investigators, such as, Light (1934); Coaton (1948); Edwards and Mill (1986); Myles *et al.*(2007) and Nunes *et al.*(2010) indicated that dry wood termites infest both hard wood and soft wood of wooden structures which cause a serious damage to furniture, painting, frames, poles, wooden works within buildings, standing trees of different wooden and ornamental trees, flooring and other wooden articles.

Cryptotermes brevis Walker (Isoptrea: Kalotermitidae) is one of most widespread pests in the world including, Africa, North Europe, Australia and most tropical islands (Scheffrahn *et al*., 2009). The same authors remember that the distribution of this pest is vast due to secondary introductions since it present in all continents except Antarctica, they added that *C. brevis* was introduced to Port Said, however this insect did not exist in Cairo, Egypt. While El-Hemaesy (1976) mentioned that this species is recently introduced to Egypt through Port Said Sea Port prior to 1965 by international trade.

Microwaves are radio waves with high frequency electromagnetic field which changed about 2 trillion times per second. These are unionized rays whose range of microwave frequency approximately ranges from 0.3 Ghz (Gigahertz) to 30 GHz with corresponding wavelength in vacuum from one meter to one millimeter. Microwave heating is heating inside the material and it is more effective than conventional methods of heating where the heat is generated outside the treated product and conveyed by conduction or convection. Materials containing free water, such as wood and organisms are capable of absorbing microwave energy with consequent increase in temperature. Water is an ideal material that absorbs radiation with wave length 12.25 Cm (corresponding to the frequency of 2.45GHz) (Merenda, 2006).

In this respect, Burdette *et al*. (1975) reported that microwave technology using for wood products to control borers infestations, while Lewis and Haverty (1996); Lewis *et al*. (2000); Peters and Creffield (2002) and Evans (2002) showed that microwave irradiation used to control for dry wood termite. Also, Fleming *et al.* , 2005 and Kisternaya and Kozlov, 2007 showed that the irradiation of microwave has been successfully used for many years for attacked wood treatment by wood borers. The heat treatment was used to control the infestations of dry wood termite *C. brevis* in timber and furniture (Horner and Bowe, 1934 and Forbes *et al*., 1988). The temperatures between 50 to 65 C° were used to control dry wood termites in infested timber < 5.1cm thick (Ehrhorn, 1934). The effects of low temperatures on termites have scarcely been investigated. Lund (1962) determined that workers of the eastern subterranean termite, *Reticulitermes flavipes* (Kollar) (Isoptera: Rhinotermitidae), succumbed after less than 5 Min exposure at -9.5 °C to -13.0° C. All *Coptotermes formosanus* Shiraki (Isoptera: Rhinotermitidae) died when maintained at 10° C, whereas *R. flavipes* survived (Smythe and Williams, 1972). This work carried out to study the effectiveness of physical methods such as microwave irradiation, high temperature and cooling degrees to control dry wood termite, *C. brevis* within pine and beech wood.

Materials and methods

1.Culture of termite:

Infested wood samples with dry wood termite, *C. brevis* were collected from cumulated wooden combinations at

different places and coastal regions of Port Said town (Mediterranean sea port) during the summer season of 2018. These samples were transferred to wood borers and termite laboratory, Plant Protection Research Institute, Dokki, Giza Governorate, Egypt. The samples contained termite was kept in suitable containers under ordinary laboratory conditions.

2.Used laboratory apparatus:

2.1.Microwave device:

Apparatus radiating microwaves of the frequency 2.45 GHz (wave length 12.2Cm) was for Tornado kind (domestic microwave oven), Model TM-25 SD., Rated voltage 230V.

2.2.Standing deep freezer:

To cooling the tested samples on different degrees used Toshiba freezer, Model GF-22h.

2.3.Electric heat oven:

To generate thermal energy on the different thermal degrees used electric oven (Veb Mlw Medizinische; Type, 116 -0100).

3.Experiment equipments:

3.1.Kind of wood blocks:

Used wooden blocks were made from pine and beech wood. The prepared blocks were in small boxes shape sized 4x2x2 cm with walls and covers of the same thickness (5mm). Inside each block, tunnel measured 3cm long x 0.5cm width x 1cm depth (Figure,1).

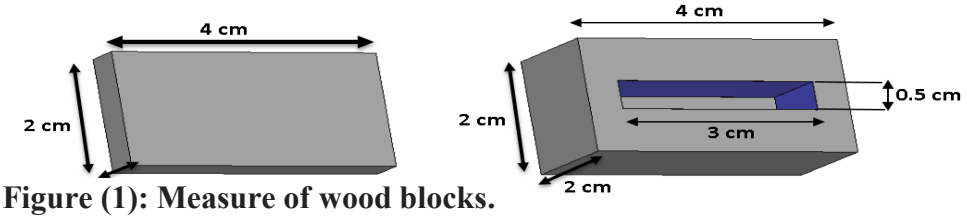

Figure (1): Measure of wood blocks.

4.Tested termite:

Extracted termite from infested wood samples for work the different experiments were distributed on manufactured tunnels inside wooden blocks, at rate 8 termite nymphs of each tunnel for 10 wooden blocks, these infested wooden blocks were applied for study the effect of microwave radiation, heating and cooling on mortality of termites at various exposure times. Microwave radiations were generated at power level of 10, 30, 50, 80 and 100 Wattage, while tested cooling degrees were -18,-21 and -27C°, whereas heating carried out heat at degrees 60,65,70 and 75 C°.

5.**Statistical analysis**:

The mortality percentages were calculated and corrected by using Aboott' Formula (Abbott, 1925) and the lethal time 50 and 95 was deterMined for established regression lines according to method of Finney (1971). Simple correlation and partial regression for obtained data were deterMined by SAS program (2001).

Results and discussion

The different effectiveness of each microwave radiation, cooling degrees and high temperature on mortality percentages of dry wood termite nymphs (*C. brevis*), introduced in two species of wooden blocks under different exposure time, obviously detected the following results:

1. Influence of microwave radiation:

1.1. On mortality percentages of *Cryptotermes brevis* termite:

Data illustrated in Table (1) showed the mortality percentages of *C.brevis* termite in two kinds of wooden blocks and treated by microwave radiations generated from microwave system under various wattages at different exposure times.

1.1.1. Pine wood:

Pine wooden blocks infested by *C.brevis* nymphs showed that percentages of mortality varied

according to exposure times and power of microwave device. The mortality percentage at 10 wattage was 0 % for all exposure times. At 30 wattage, the mortality percentage recorded 2.5 23.8, 67.5, 83.8 and 100% mortality for 20, 30, 40, 50 and 60 Sec. of exposure time, respectively. At 50 wattage, the percentage recorded 37.5, 68.8 and 91.3% for 20, 30 and 40 Sec. of exposure time, respectively; while it was 100% mortality for the other exposure times (50-60 Sec.). At 80 wattage, the mortality percentages were 22.5, 76.3 and 95%, for 10, 20 and 30 Sec. respectively, while the exposure time for 40 Sec. and more gave 100% mortality. While, at 100 wattage, the mortality percentage was 41.3% of exposure time 10 Sec., whereas all other exposure times recorded 100% mortality.

1.1.2. Beech wood:

Beech wood was more influence with microwave radiation, where the mortality percentages recorded 7.5, 31.3, 77.5, 96.3 and 100% for 20, 30, 40, 50 and 60 Sec. of exposure time, respectively, at 10 wattages. While, at 30 wattage the parentages of mortality were 25, 73.8, 93.8 and 100% for 20, 30, 40 and 50 Sec. of exposure time, respectively. At 50 wattage, mortality percentage reached 48.8, 86.3 and 100% for 20, 30 and 40 Sec. of exposure time, respectively. At 80 and 100wattages the all mortality percentages recorded 100% for all exposure times of more than 10 Sec. Previous results indicated that the mortality percentages for termite nymphs (exposing to microwave radiation) in beech wooden blocks were higher than pin wooden blocks at same measurements. These results revealed that wood kind play an important role in tolerance or protection of the internal stages of insect against the opposite external factors (circumstances).

The data presented in Table (2) showed that the lethal time (LT50 and LT95) values of the drywood termite*, C. brevis* in both pine and beech wood under power levels. Whereas, the lethal time values (LT50) for pine wood under power levels 30,50 and 80 wattage were 36.27, 23.3 and 14.43 Sec., while the lethal time values (LT95) recorded 59.29, 47.24 and 33.05 sec. respectively under same power levels. For beach wood the lethal times (LT50) were 32.4 and 25sec. while, the lethal time values (LT95) recorded 51.7 and 45.5 Sec., under power level 10 and 30 wattage, respectively.

Table (1): Effect of microwave irradiation on mortality rate of dry wood termite, *Cryptotermes brevis* in both pine and beech wood at different percentage exposure times.

Power level (Wattage)	Kind of wood	Mortality percentage at different exposure times (Sec.)					
		10	20	30	40	50	60
		M.±SE	M.±SE	M.±SE	M.±SE	M.±SE	M.±SE
10	Pine	0 ±0.0	0 ± 0.0	0 ± 0.0	0 ± 0.0	0 ±0.0	0 ±0.0
	Beech	0± 0.0	7.5 ±2.03	31.3 ± 2.08	77.5 ±1.67	96.3 ±1.81	100 ± 0.0
30	Pine	0± 0.0	2.5 ± 1.67	23.8±2.64	67.5±2.41	83.8 ±1.91	100 ± 0.0
	Beech	0± 0.0	25 +1.86	73.8 ±2.23	93.8 ±2.08	100 ± 0.0	100 ± 0.0
50	Pine	0± 0.0	37.5± 3.23	68.8 ± 2.80	91.3 ±3.75	100 ± 0.0	100 ± 0.0
	Beech	0± 0.0	48.8 +1.25	86.3 ±2.89	100 ± 0.0	100 ± 0.0	100 ± 0.0
80	Pine	22.5 ±1.67	76.3 ± 2.22	95+ 2.04	100 ± 0.0	100 ± 0.0	100 ± 0.0
	Beech	0± 0.0	100 ± 0.0	100 ± 0.0	100 ± 0.0	100 ± 0.0	100 ± 0.0
100	Pine	41.3 ±1.91	100 ± 0.0	100 ± 0.0	100 ± 0.0	100 ± 0.0	100 ± 0.0
	Beech	0± 0.0	100 ± 0.0	100 ± 0.0	100 ± 0.0	100 ± 0.0	100 ± 0.0

Table (2): Values of Lt50 and Lt95 for *Cryptotermes brevis* termite within pine and beach wood exposed to different power levels of microwave irradiation.

Power level (wattage)	LT50	LT95	Slop	P.value
Pine wood				
30	36.27	59.29	7.7 ± 0.68	0.45
50	23.3	47.24	5.36 ±0.68	0.33
80	14.43	33.05	4.57±0.47	0.48
Beach wood				
10	32.4	51.7	8.12 ±0.63	0.06
30	25	45.5	6.34 ± 0.75	0.16

1.2. On percentage of lost moisture content :

Date illustrated in Table (3) appear the percentages of lost moisture content for two kinds of wood blocks treated by microwave irradiation generated from microwave device under varies wattages at different exposure times.

1.2.1. Pine wood:

Obtained results on lost moisture content percentages indicated that the increase of power level (Wattage) led to increase of lost moisture content percentage at different exposure times. The minimum percentages of lost moisture content recorded 0.38, 0.41, 0.57, 0.64 and 0.67% at 10 Sec. under all wattages values, while, the maximum percentages of lost moisture recorded 7.12% and 7.26% at 60 Sec.under 80 and 100 wattages, respectively. Also, the lost moisture recorded 6.05%, and 6.17%, at 60 Sec. under 30 and 50 wattages, respectively. While, it recorded 6.07 and 6.9 0% at 50 Sec. under 80 and 100 wattages.

1.2.2. Beech wood:

The lost moisture content percentages increased with the increase exposure time at values of oven power level. At time exposure 10 Sec., the least percentages recorded 0.14%, 0.20%, 0.50%, 052% and 0.59% under tested wattages values, respectively. Also, the least percentages at time exposure 20 Sec recorded 0.31%, 0.48% and 0.95% under 10, 30 and 50 wattage respectively. At exposure time 30 and 40 Sec. the least percentage was 0.58% and 0.94% under10 wattage. While, the maximum percentage of lost moisture was 4.56% and 4.73% under 80 and 100 wattages, respectively at time exposure 50 Sec. As at exposure time 60 Sec. the percentage of lost moisture recorded 4.25, 4.56, 4.89, 5.07 and 5.53 under tested wattages values (Table, 3). Highly significant correlations were clear obviously between percentages of lost moisture content of both pine and beech wood with different exposure time to microwave irradiation at different power levels (Table, 4).

Table (3) : **Effect of microwave irradiation on percentages of lost moisture content of both pine and beech wood at different exposure times.**

Power level (wattage)	Kind of wood	Percentages of lost moisture content at different exposure times (Sec.)					
		10	20	30	40	50	60
		M.±SE	M.±SE	M.±SE	M.±SE	M.±SE	M.±SE
10	Pine	0.38±0.008	1.29±0.014	1.76±0.019	3.33±0.028	5.08±0.035	5.27±0.038
	Beech	0.14±0.015	0.31±0.020	0.58±0.026	0.94±0.018	1.87±0.015	4.25±0.024
30	Pine	0.41±0.012	1.36±0.013	2.31±0.018	3.43±0.016	5.41±0.020	6.05±0.028
	Beech	0.20±0.013	0.48±0.021	1.39±0.015	2.15±0.021	2.79±0.019	4.56±0.024
50	Pine	0.57±0.014	1.44±0.022	3.07±0.042	3.88±0.021	5.73±0.036	6.17±0.032
	Beech	0.50±0.018	0.95±0.033	1.55±0.025	2.78±0.030	3.31±0.023	4.89±0.031
80	Pine	0.64±0.023	1.48±0.028	3.31±0.032	4.65±0.021	6.07±0.043	7.12±0.051
	Beech	0.52±0.024	1.16±0.028	1.91±0.040	3.28±0.034	4.56±0.030	5.07±0.039
100	Pine	0.67±0.031	1.66±0.033	3.63±0.047	4.93±0.072	6.90±0.031	7.26±0.021
	Beech	0.59±0.025	1.61±0.040	1.94±0.036	3.60±0.081	4.73±0.042	5.53±0.066

Table (4): Simple correlation and regression values between different power levels and percentages of lost moisture content of both pine and beech wood at different exposure times.

Power level (wattage)	Kind of wood	Percentages of lost moisture content	
		Simple correlation (r)	simple Regression (b)
10	Pine	0.980	0.11
	Beech	0.883	0.73
30	Pine	0.991	0.12
	Beech	0.974	0.08
50	Pine	0.989	0.12
	Beech	0.987	0.08
80	Pine	0.997	0.14
	Beech	0.991	0.10
100	Pine	0.989	0.14
	Beech	0.989	0.10

2. Influence of the cooling on mortality percentages of *Cryptotermes brevis* termite:

The mortality percentages of dry wood termite *C.brevis*, resulting from three cooling degrees, for two kinds of infested wood by exposing to different periods were showed in Table (5). The obtained results indicated that the percentages of mortality differed with differences of each cooling degrees, exposure time and attacked wood kinds.

2.1. Pine wood:

The effect of tested cooling degrees (-18, -21 and -27°C) on mortality of termite nymphs showed that the mortality percentages increased with increase in cooling degrees at different exposure times. At -18°C, the percentages of mortality ranged between 5 % mortality at 26 Min. exposure time to 100% mortality at exposure time 36 Min. at -21°C, the least mortality percentage was 11.3% at 20 Min., while the highest percentage recorded 100% at 30Min exposure time, whereas at -27°C, the mortality percentage recorded 100% on all exposure times.

2.2. Beech wood :

The mortality percentages of termite nymphs increased with the increase of exposure time to different cooling degrees. The exposure time which gave 100% mortality was 20 Min. recorded at -27°C, while the longest exposure time to obtain 100%mortaiilty was 40 Min. at-18°C exposure time. The obtained data in Table (5) showed that the mortality percentages of termite nymphs were affected according to infested wood kind. The mortality percentages of termite nymphs existent enter pine wooden blocks were higher than the others enter beech wooden blocks through different exposure times to the three cooling degrees (-18,-21,-27°C). The highest percentage of mortality (100%) was nearly similar in each pine and beech wood on -18°C degree at exposure time of 40 Min.at -21°C degree, pine wood recoded 100% mortality on 30-40 Min., while beech wood showed 100% mortality on 32-40 Min. exposure time. At -27°C degree, the highest mortality percentages were 100% enter pine wooden blocks at exposure time of 18Min. and more, whereas beech wooden blocks showed 100% mortality at exposure time of 20 Min. and more.

The primary experiments carried out by Forbes and Ebeling (1986) detected that termite workers killed within 5 Min. at temperatures between -18.5 to -19.4 °C (-1.3 to 2.9 °F), whereas Rust *et al.* (1997) corroborated the previous experiences and found that exposure of workers (sic) in wooden blocks to temperatures below -21.4° C resulted in 100 percent mortality. Abd-El Malak (2002), in Egypt found that the mortality percentages of *C. brevis* increased with increasing exposure time

for cooling degree, whereas, the exposure at -18°C gave 26.67, 46.67, 60, 83.33 and 100 % mortality under exposure time 30,31,32,33, and 34 Min., respectively. The data in Table 6 appeared that the lethal time (LT50 and LT95) values of both pine and beech wood were as following: for pine wood the LT50 and LT95 recorded 30.83 and 36.29 Min. at -18°C, respectively. While the lethal time (LT50 and LT95) values were 23.36 and 29.06 Min. at -21°C, respectively. The beech wood give lethal time (LT50 and LT95) values 32.28 and 38.81Min. under -18 °C, receptively, whereas lethal time (LT50 and LT95) values were 24.16 and 30.46 Min. under -21 °C, respectively (Table,6).

Table (5): Effect of cooling degrees on mortality percentages of drywood termite *Cryptotermes brevis* in both pine and beech wood at different exposure times.

Cooling degree	Kind of wood	Mortality percentages affected by cooling degrees											
		Time (Minutes)											
		18	20	22	24	26	28	30	32	34	36	38	40
		M.±SE	M.±SE	M.±SE	M.±SE	M.±SE	M.±SE	M.±SE	M.±SE	M.±SE	M.±SE	M.±SE	M.±SE
-18 °C	P	0± 0.0	0± 0.0	0± 0.0	0± 0.0	5± 2.04	17.5± 2.76	36.3± 1.25	62.5± 1.86	86.3± 1.25	100± 0.0	100± 0.0	100± 0.0
	B	0± 0.0	0± 0.0	0± 0.0	0± 0.0	0± 0.0	8.8± 1.90	22.5± 2.5	56.3± 2.79	66.3± 2.67	81.3± 2.08	92.5± 2.76	100± 0.0
-21 °C	P	0± 0.0	11.3± 2.24	35± 1.67	56.3± 1.88	77.5± 2.5	92.5± 2.76	100± 0.0	100± 0.0	100± 0.0	100± 0.0	100± 0.0	100± 0.0
	B	0± 0.0	6.3± 2.79	27.5± 2.5	43.8± 2.08	60± 1.66	82.5± 2.04	98.8± 1.25	100± 0.0	100± 0.0	100± 0.0	100± 0.0	100± 0.0
-27 °C	P	100± 0.0	100±0. 0	100± 0.0	100± 0.0	100± 0.0	100± 0.0	100± 0.0	100± 0.0	100± 0.0	100± 0.0	100± 0.0	100± 0.0
	B	28.8± 1.91	100± 0.0	100± 0.0	100± 0.0	100± 0.0	100± 0.0	100± 0.0	100± 0.0	100± 0.0	100± 0.0	100± 0.0	100± 0.0

Table (6): Values of LT50 and LT95 for *Cryptotermes brevis* termite within pine and beach wood exposed to different cooling degrees.

Cooling Degrees	LT50	LT95	Slop	P. value
Pine wood				
-18°C	30.83	36.29	23.2±1.87	0.76
-21°C	23.36	29.06	17.36±1.39	0.86
Beach wood				
-18°C	32.28	38.81	20.53±1.48	0.30
-21°C	24.16	30.46	17.11±1.15	0.06

3.Effect of high temperature degrees:

3.1. On mortality percentages of *Cryptotermes brevis* termite:

Data illustrated in Table (7) detected the effect of four temperature degrees (60, 65,70 and 75 °C) on mortality percentages of *C. brevis* termite inside two kinds of wood (pine and beech) which subjected to different times.

3.1.1. On pine wood:

At 60 °C, 65°C and 70°C the lowest percentages of mortality recorded on different exposure times of 28, 29 and 30 Min., while the highest mortality percentages recorded at 33,34 and 35Min. At75 °C, the percentages of mortality were 100% at all exposure times.

3.1.2. On beech wood:

The same trend of mortality percentages on each 60 °C, 65 °C and 70°C for pine wood recorded also for beach wood. Previous results indicated that the infested wood with *C. brevis* termite required temperature degrees 60-70°C for exposure times 34-35 Min. to obtain 100% mortality except pine and beech wood for 60°C at 34 Min. gave 96.3% and 93.8% mortality, respectively. While, at 75°C the exposure time 28 Min. was enough to obtain 100% mortailty

In Florida, Scheffrahn *et al*. (1997) estimated that the temperature and heating time requirements to control *Cryptotermes brevis* in structural infestations. Furthermore, they found that exposure time 54.4°C for 60 Min., was suitable to control of dry wood termite. While, Forbes and Ebeling (1987), showed that the heat treatment was applied to control dry wood termite incicitermes minor at 49ºC under exposure time 30Min. Ebeling (1997), reported that temperature of 55ºC for 60 Min. was used in many applications. While, the exposure time 30 Minutes to 49ºC was suitable to controlled drywood termite *C. brevis* in wood (Woodrow and Grace, 1998 a, b). In Egypt, Abd-El Malak (2002) showed that the percentage of mortality of *C.brevis* termite increased by increasing the temperature degree of heating and exposure time, furthermore, he found mortality percentage was recorded 100% under exposure time 34 Minutes for 50, 60 and 70°C. The obtained data in Table (8) elucidated that the lethal time (LT50) values of the *C. brevis* termite, recorded 31.18, 30.79 and 30.27 Min. at 60° C, 65C° and70C°, respectively, while lethal time values (LT95) were 34.75, 34.70 and 34.34 Min. at 60° C, 65C° and70C° ,respectively for pine wood. Whereas, beech wood exposed to 60° C, 65C° and 70C° gave (LT50) values equal 31.74, 31.18 and 30.76 Min., respectively, also, (LT95) values were 35.57, 34.82 and 34.71 Min. respectively at same temperature degrees.

Table (7): Effect of high temperature degrees on mortality percentages of dry wood termite *Cryptotermes brevis* in both pine and beech wood at different exposure times.

Temperature degree	Kind of wood	Mortality percentages affected by heat degrees							
		Time (Minutes)							
		28	29	30	31	32	33	34	35
		M.+SE	M.+SE	M.+SE	M.+SE	M.+SE	M.+SE	M.+SE	M.+SE
60°C	Pine	6.3± 2.08	18.8 ± 2.79	27.5 ± 2.5	38.8 ± 2.42	56.3± 3.36	83.8± 2.66	96.3 ± 1.90	100± 0.0
	Beech	3.8 ± 1.91	12.5± 2.64	21.3± 3.3	30 ± 2.04	48.8± 3.46	78.8± 1.91	93.8± 2.08	100± 0.0
65°C	Pine	15± 1.67	21.3± 3.3	32.5± 2.76	41.3± 3.25	66.3± 3.09	93.8± 2.65	100± 0.0	100± 0.0
	Beech	8.8± 1.90	18.8± 2.08	26.3± 2.24	35± 1.67	57.5± 2.76	82.5± 2.04	97.5± 1.66	100± 0.0
70°C	Pine	23.8± 1.25	28.8± 1.91	37.5± 1.86	48.8± 2.92	77.5± 2.35	96.3± 1.91	100± 0.0	100± 0.0
	Beech	15± 1.67	22.5± 2.5	31.3± 2.08	43.8± 2.79	67.5± 2.04	92.5± 2.67	100± 0.0	100± 0.0
75°C	Pine	100± 0.0	100± 0.0	100± 0.0	100± 0.0	100± 0.0	100± 0.0	100± 0.0	100± 0.0
	Beech	100± 0.0	100± 0.0	100± 0.0	100± 0.0	100± 0.0	100± 0.0	100± 0.0	100± 0.0

Table (8): Values of LT50 and LT95 for *Cryptotermes brevis* termite within pine and beach wood exposed to different temperature degrees.

Temperature degree	LT50	LT95	Slop	P. Value
Pine wood				
60°C	**31.18**	**34.75**	**34.92±2.30**	**0.02***
65°C	**30.79**	**34.70**	**31.69±2.55**	**0.0007****
70°C	**30.27**	**34.34**	**30.04±2.49**	**0.0001****
Beach wood				
60°C	**31.74**	**35.57**	**33.27±2.81**	**0.133**
65°C	**31.18**	**34.82**	**34.27±2.27**	**0.002****
70°C	**30.76**	**34.71**	**31.04±2.54**	**0.004****

**: highly significant * : significant

3.2. On percentages of lost moisture content :

The percentages of lost moisture content from pine and beech wood exposed to high temperature degrees for different times are clarified in Table (9). Obtained data detected that the percentages of lost moisture content increased with the increase of temperature degrees and also with the increase of exposure times of both pine and beech wood.

3.2.1. Pine wood:

The percentages of lost moisture content was 1.63% at 60°C with 28 Min. exposure time, reached to 2.81% at 75°C with same exposure time, while it recorded 3.47% at 60 °C with 35 Min.exposure time, whereas it reached 3.94% at 75°C with 35 Min.

3.2.2 Beech wood:

The data showed that the lost moisture content percentages at 60°C and 28 Min. exposure time was1.32%, reached 2.38% at 75°C with same exposure time. Whereas, the percentages of lost moisture content at 60°C with 35 Min., exposure time recorded 2.30%, increased to 3.53% at 75°C with same exposure time.Highly significant correlations were clear between exposure time and percentages of lost moisture content from pine and beech wood at different temperature degrees, also highly significant correlations were determined between temperature degrees (60-75°C) and percentages of lost moisture content at different exposure times (Table,10).

3.3. Combined effect of temperature and moisture content of wood:

Previous results in Tables (9 and 10) revealed that the temperature showed changes on moisture content of wood with the changes in percentages of mortality, it means that the moisture content play an important role with temperature beside exposure time and wood kind in the effect on mortality of termite inside the infested wood.

Table (9): Effect of high temperature degrees on percentages of lost moisture content of drywood termite *Cryptotermes brevis* in both pine and beech wood at different exposure times.

Temperature degree	Kind Of Wood	Percentages of lost moisture content at different exposure times (Min.)							
		28	29	30	31	32	33	34	35
		M±SE	M±SE	M±SE	M±SE	M±SE	M±SE	M±SE	M±SE
60°C	Pine	1.63± 0.029	1.82± 0.020	2.19± 0.022	2.39± 0.027	2.70± 0.035	2.93± 0.037	3.21± 0.034	3.47± 0.027
	Beech	1.32± 0.028	1.47± 0.017	1.60± 0.025	1.73± 0.023	1.89± 0.013	2.04± 0.044	2.18± 0.023	2.30± 0.011
65°C	Pine	1.79± 0.024	1.99± 0.022	2.21± 0.037	2.48± 0.015	2.86± 0.018	3.10± 0.017	3.38± 0.30	3.71± 0.027
	Beech	1.49± 0.032	1.68± 0.015	1.81± 0.026	1.99± 0.017	2.13± 0.023	2.27± 0.042	2.33± 0.022	2.49± 0.017
70°C	Pine	2.26± 0.026	2.46± 0.025	2.60± 0.015	2.78± 0.030	3.02± 0.027	3.29± 0.023	3.53± 0.28	3.80± 0.018
	Beech	1.76± 0.012	1.89± 0.011	1.98± 0.017	2.15± 0.017	2.28± 0.012	2.40± 0.011	2.63± 0.013	2.75±0 .026
75°C	Pine	2.81± 0.030	3.08± 0.044	3.29±00. 016	3.40± 0.050	3.55± 0.033	3.64± 0.036	3.76± 0.035	3.94± 0.061
	Beech	2.38± 0.021	2.50± 0.024	2.63± 0.027	2.82± 0.020	3.09± 0.040	3.21± 0.033	3.34± 0.023	3.53± 0.027

Table (10): Simple correlation and regression values between different temperature degrees and percentages of lost moisture content of both pine and beech wood at different exposure time.

Temperature degree)	Kind of wood	Percentages of lost moisture content	
		Simple correlation (r)	Simple Regression (b)
60°C	Pine	0.999	0.27
	Beech	0.999	0.14
65°C	Pine	0.997	0.28
	Beech	0.995	0.14
70°C	Pine	0.995	0.22
	Beech	0.859	0.24
75°C	Pine	0.988	0.15
	Beech	0.996	0.17

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Effect of sprayer, nozzle types and spraying volume on efficacy of chemical compounds against tomato leafminer *Tuta absoluta* (Lepidoptera: Gelechiidae) infesting tomato

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Abstract:

Tomato leafminer, *Tuta absoluta* (Meyrick) (Lepidoptera: Gelechiidae) is a highly destructive insect pest to tomato plants and fruit and is also reported to infest other plants in the Solanacaeae family. In the present study, three equipments were conducted to control *T. absoluta*, at New Salhyia city, Sharqia Governorate. The first equipment, Cawzer sprayer with 3 nozzles type flat fan 02F-110, with spray volume 60L/fed., Matabi sprayer with Full cone Tx-6 with spray volume 35 L/fed., and conventional motor fitted with spray gun at spray volume 300 L/fed., fitted with local spray gun produce hollow cone. The aim of this work is to determine the effects of application method on initial and late biological efficacy of three insecticides , emamectin benzoate (Proclaim) 20% SC., at rate 60 gm/fed.; indoxacarb (Avant) 15% EC at rate 100 ml/fed and alverde (Metaflumizone) – SC. 24% at rate 400ml/fed., against *T. absoluta.* Results in two seasons (2016 and 2017) indicated that, the high efficacy gives by Matabi sprayer with volume mean diameter (VMD) 68.7-94.4µm, and N/cm2 3.94-37.8 and Cawzer sprayer with VMD 51.36-125.35µm. and N/cm2 18-115.5, while less efficient was obtained with conventional Motor, with VMD 480-950µm, and N/cm2 5-21. Otherwise, the highest toxic effect obtained with emamectin benzoate, metaflumizone and indoxacarb with Matabi and Cawzer sprayers.

Introduction

Tomato, *Lycopersicon esculentum* Mill. (Solanales: Solanaceae) is an important vegetable in Egypt and planted throughout the year. Egypt considered the fifth largest tomato producer in the world with about 9.2 million tons produced from about 454,000 Feddans planted with tomato (WPTC, 2011). Tomato infested by more than 200 arthropods (Anonymus, 2001). So, it was of great importance to combat tomato pests since emergence till harvesting to prevent huge damage caused by these pests (Moussa *et al.*,2013). Recently, tomato leafminer borer, *Tuta absoluta* (Meyrick) (Lepidoptera: Gelechiidae), considered the most quick devastating pest of tomato plantations (Shalaby *et al*., 2012). Their larvae were infested leaves,

flowers, stems and fruits leads to reduce photosynthesis, impairs fruits and paves the way for pathogens to enter through lesions caused by the pest (Colomo *et al*.,2002) and might lead to complete loss of the yield. In Egypt, the pest was detected for the first time in early 2010 at Marsa Matrouh Governorate. Then other reports confirmed pest presence in other parts of the Delta Valley (EPPO, 2006).

Nowadays pest management had been depended mainly on pesticides applications (Gonza´lez-Cabrera *et al*.,2011). Several insecticides belong to different insecticides classes were recommended for pest control. Emamectin benzoate, indoxacarb and metaflumizonewere the most commonly recommended insecticides used against *T. absoluta* in Egypt according to (Egyptian Agricultural Pesticides Committee).

Pesticide sprays aimed to maximize pesticide efficacy and minimized their adverse effects. So, factors such as optimum droplet size for killing, number of droplets per unit area might be optimized (Sundaram *et al*.,1985). Effective application obtained by increasing coverage percent per unit area (Ferguson *et al.*, 2016). Droplet size may be influence the biological efficacy of the applied pesticide as well as environmental hazards and spraying application could be affect toxicant efficacy, through affecting on the spraying parameters, such as optimum volume mean diameter (VMD) which greatly effects on mortality and residual effects (Omar and Matthew, 1991). The insecticidal effect of spraying droplets were dependent not only on droplet diameter but also on droplet number/cm^2 and concentration of active ingredient of used insecticides (Elbert *et al*., 1999). The previous studies suggested that the optimal droplet size varies according to insecticide, target pest and application method.

The aim of this work is to determine the effects of application methods (sprayer, nozzle types and spraying volume) and spray coverage distribution on initial and late efficacy of emamectin benzoate (Proclaim) , indoxacarb (Avant) and alverde (Metaflumizone) against tomato leafminer, *T. absoluta* .

Materials and methods

1. Tested sprayers :

In the current study, three different types of sprayers were evaluated and their technical data illustrates in Table (1).

Table (1): Technical data of tested equipment.

Equipment Parameter	Cawzer (compretion sprayer)	Matabi (hydraulic Hand-Heldsprayer)	Conventional Ground Motor (hydraulic)
Nozzle type	Flat fan 02F-110	Full cone Tx-6	Spray gun
Total Tank capacity (L)	8	20	400
Spray volume (L/fed)	60	35	300
Working Speed (Km/h)	2.4	2.4	2.4
Swath width (m)	2.5 (Hand lance fitted with 3 nozzles)	2	0.75
Flow rate (L/Min)	0.484	0.32	2.16
Spraying height (m)	50 at all treatments		
Spray system	Target spray		

2. **Pesticides used:**

Three insecticides were tested in the current study include: Emamectin benzoate (Proclaim 20% SC., Syngenta, Germany, rate dose 60 gm/feddan), Indoxacarb (Avant 15% EC-Du pont de Nemoursp-USA- rate dose 100 ml /fed) and alverde (Metaflumizone ¬-SC % 24-Basf-Germany- rate dose 400ml/ fed.) as recommended dose for this pest.

3. Field experiments:

Field experiment were conducted during the growing tomato season of March, 2016 and 2017 at New Salhyia city, Sharqia Governorate, Egypt. The experimental area was divided into plots, each is $40m^2$ (1/100 fed.) and arranged in randomized complete blocks with four replicates. Four plots were left untreated to serve as control. The normal agricultural practices were done. The tested insecticides were applied at the recommended rate of the aforementioned insecticides. The control plots were sprayed only with water. Also, care was taken to avoid any drift among the treated plots. Samples of 25 leaves were chosen at random from each replicate before treatments and at 1,3,5,7,9,11 and 14 days after pesticides application. The number of *T. absoluta* was counted. Percentage of reduction of the insect population was calculated according to Henderson and Tilton (1955).

4.Collection of spray deposit:

To collect the spray deposit and for a precise evaluation of spray coverage of the tested atomizers in the field, sensitive papers of water were placed on tomato plants canopy before spraying at three trends right, left, middle ones. In addition, the cards were fixed in a wire holders to measure lost spray on the ground. Sensitive papers were given code numbers before being stuck to the plants and the wire holder. About 50 Minutes had elapsed after spraying operation to allow dryness of deposits. So, the sensitive papers were carefully collected. There were transferred to the laboratory for qualitative analysis.

5. Laboratories tools:

5.1. Strubinlense (15x) was used to measure the sprayed spots (Gabir *et al*., 1982).

5.2. Calculator was used to calculate VMD of droplets cards at all trends of tomato plants.

5.3. Computer system was used to analyze data by using statistical analysis system SPSS.

Results and discussion

1. Spray coverage on tomato plants against *Tuta absoluta* by using pesticides during (2016/2017) seasons:

1.1. Cawzer sprayer (compretion sprayer):

Cawzer sprayer used to spray volume of 60L/fed. at the first season, produced volume diameters in the range of 60 - 98µm. The smallest droplet size obtained with proclaim , VMD mean diameter of 60µm followed by avant 75µm and then alverde 98µm. Similarly, at the second season VMD range was 61-97µm with mean diameter volume of 61µm with proclaim, 75µm for avant and 97µm for alverde (Tables, 2 and 3).

1.2. Matabi sprayer (Hand Heldhydraulic sprayer):

Matabi sprayer used spray volume of 35L./fed. and generated in the first season VMD with the range of 80-86µm. The tested pesticides gave mean volume diameter of 83µm with proclaim, 86µm with avant and 80µm with alverde. In the second season VMD recorded 85µm with proclaim, 86µm with avant and 82µm with alverde (Tables, 2 and 3).

1.3. Conventional ground motor:

Conventional motor used spray volume of 300L./fed. and generated in the first season VMD range of 725-825µm. with proclaim mean volume diameter recorded 725µm, 825µm with avant and 745µm with alverde. Similarly, in the second season VMD recorded 718,789µm and 735µm for

proclaim, avant and alverde, respectively (Tables, 2 and 3).

Table (2): Spray coverage means as obtained from three sprayers by using insecticides on tomato plants for controlling *Tuta absoluta* during season 2016.

Equipment		Cawzer sprayer			Matabi sprayer			Conventional motor		
Spraying volume		60 L.			35 L.			300 L.		
Spectrum Insecticide		VMD (µm)	N/cm²	N%	VMD (µm)	N/cm²	N%	VMD (µm)	N/cm²	N%
2016										
Proclaim	Right plant	62	111	44	91	33	29.7	850	12	24.5
	Middle plant	65	54	21.3	85	37	33.3	650	8	16.3
	Left plant	61	43	17.1	87	35	31.5	900	10	20.4
	Wire holder	52	44	17.6	70	6	5.4	500	19	38.8
Avant	Right plant	95	29	18	94	32	35.6	900	7	15.6
	Middle plant	84	32	20	94	24	26.7	800	9	20
	Left plant	65	47	29.3	81	28	31.1	950	11	24.4
	Wire holder	56	53	32.7	73	61	6.7	650	18	40
Alverde	Right plant	126	18	14.5	85	38	33.6	870	11	22
	Middle plant	85	39	31.3	81	36	31.9	780	5	10
	Left plant	86	31	25.3	87	35	31	850	13	26
	Wire holder	95	36	28.9	69	4	3.6	480	21	42

Table (3): Spray coverage means as obtained from three sprayers by using insecticides on tomato plants for controlling *Tuta absoluta* during season 2017.

Equipment		Cawzer			Matabi Sprayer			Conventional Motor		
Spraying volume L/fed.		60			35			300		
Spectrum Insecticide		VMD (µm)	N/cm²	N%	VMD (µm)	N/cm²	N%	VMD (µm)	N/cm²	N%
2017										
Proclaim	Right plant	64	108	43.21	93	32	29.79	820	12	25.5
	Middle plant	66	53	21.44	86	36	33.89	644	8	16.6
	Left plant	60	44	17.8	89	34	31.78	876	10	21.1
	Wire holder	53	44	17.56	72	5	4.54	530	18	36.8
Avant	Right plant	97	28	17.6	93	32	35.76	880	7	15.3
	Middle plant	83	32	20	93	24	26.9	756	9	20.4
	Left plant	67	45	28.18	81	28	30.84	870	12	25.7
	Wire holder	54	55	34.25	75	6	6.51	650	18	38.6
Alverde	Right plant	121	19	14.92	87	37	33.51	850	11	22.3
	Middle plant	87	38	30.36	83	35	31.85	770	5	10
	Left plant	87	31	24.75	89	34	31.06	840	13	26
	Wire holder	91	37	29.96	70	4	3.58	480	21	41.6

2. Biological efficacy of the tested treatments:

Biological efficacy was carried out for three insecticides (Proclaim, alverde and avant) belong to different insecticides classes. The treatment was accomplished with three different equipment for successive two seasons. Percent reduction was recorded after 5

and 10 days and the findings were as follow:

2.1. The first season:

2.1.1. Percent reduction after 5 days:

Matabi sprayer showed that the highest biological efficacy and proved the highest percent reduction with the tested insecticides. Proclaim recorded the highest percent reduction followed by alverde and finally avant with values of 98.85, 96.27 and 92.12%, respectively. Cawzer sprayer followed matabi in biological efficacy with percent reduction values of 89.85, 85.09 and 82.94 %for proclaim, alverde and avant, respectively. While, conventional motor recorded the least efficacy with percent reduction values of 85.69, 79.49 and 75.81% for proclain, alverde and avant, respectively (Table, 4).

2.1.2. Percent reduction after 10 days:

Unlike to 5 days results, cawzer sprayer showed that the greatest biological efficacy with the three tested insecticides. Percent reduction values were 93.60, 94.36 and 97.63 % for proclaim, alverde and avant, respectively. The follower was matabi sprayer with Percent reduction of 91.31, 90.91 and 77.41 %for proclaim, alverde and avant, respectively. The least efficacy obtained with conventional motor and recorded 87.20, 77.02 and 74.35 % for proclaim, alverde and avant, respectively (Table, 4).

2.2. The second season:

2.2.1.Percent reduction after 5 days:

Results showed that the same trend obtained in the first season. Matabi sprayer showed that the greatest percent reduction and recorded 95.30, 83.40 and 93.10 %for proclaim, alverde and avant, respectively. Also, Cawzer sprayer showed the same trend with percent reductionof 85.89, 70.86 and 78.76% for proclain, alverde and avant, respectively. Finally, the least efficacy obtained with conventional motor with percent reductionof 89.40, 57.26 and 59.35% for proclain, alverde and avant, respectively (Table,4).

2.2.2. Percent reduction after 10 days:

Percent reduction obtained with Matabi sprayer was recorded 96.10, 85.73 and 82.27 for proclaim, alverde and avant, respectively. While, Cawzer sprayer proved percent reduction of 93.97, 84.28 and 91.62 % for proclaim, alverde and avant, respectively. In contrast, conventional motor showed the least efficacy with percent reduction values of 87.38, 69.81 and 67.58 % for proclaim, alverde and avant, respectively (Table,4).

3. Pairwise correlation analysis:

Correlation analysis between volume mean diameter (VMD) and percent reduction was carried out. The analysis performed for the tested insecticides with the used equipment and in the range of the used VMD.

3.1.Correlation analysis between volume mean diameter (VMD) and percent reduction after 5 days:

Correlation analysis showed significant negative correlations (at 0.01 level) for the following: Proclaim and avant with Cawzer sprayer ; proclaim, avant and alverde with Matabi sprayer ; proclaim with conventional motor. In contrast, there was positive correlations for the following: Alverde with Cawzer sprayer and alverde and avant with conventional motor (Table,5).

3.2. Correlation analysis between volume mean diameter (VMD) and percent reduction after 10 days:

Correlation analysis for the aforementioned factors showed significant negative correlations (at 0.01 level) for the following: Avant with cawzer sprayer , avant and alverde with Matabi sprayer and proclaim with conventional motor. In contrast, analysis showed positive correlations for the following: Proclaim and alverde with Cawzer sprayer; proclaim with Matabi sprayer and avant and alverde with conventional motor (Table,5).

Table(4): Percent reduction of leafminer numbers (*Tuta absoluta*) after treated with pesticides used by three equipments during 2016 and 2017seasons .

Equipment		Matabi Sprayer			Cawzer Sprayer			Conventional Motor			LSD
Spraying volume (L/f)		35			60			300			
Pesticides & dosages		Proclaim 60 gm/fed.	Alverde 400ml/fed.	Avant 100ml/fed.	Proclaim 60 gm/fed.	Alverde 400ml/fed.	Avant 100ml/fed.	Proclaim 60 gm/fed.	Alverde 400ml/fed.	Avant 100ml/fed.	
Season 2016											
Reduction %	5 days	98.85 ±1.15	96.27 ±1.32	92.12 ±3.39	89.85 ±2.80	85.09 ±4.54	82.94 ±3.26	85.69 ±3.90	79.49 ±6.77	75.81 ±7.15	6.07
	10 days	91.31 ±3.72	90.91 ±4.14	77.41 ±8.11	93.6 ±1.28	94.36 ±2.30	97.63 ±1.36	87.2 ±1.98	77.02 ±2.62	74.35 ±1.76	5.15
Season 2017											
Reduction %	5 days	95.3 ± 2.99	83.4 ±5.63	93.1 ± 4.45	85.89 ± 7.61	70.86 ± 3.71	78.76 ± 5.58	89.40±2.89	57.26 ± 8.54	59.35±12.01	9.3
	10 days	96.10± 0.79	85.73± 2.05	82.27± 2.53	93.97 ± 1.16	84.28± 0.66	91.62 ± 1.78	87.38 ±1.87	69.81 ±1.86	67.58 ± 1.86	4.45

Table (5): Pairwise correlation analysis between volume mean diameter (VMD) and percent reduction .

Equipment		Reduction								
		Cawzer Sprayer			Matabi Sprayer			Conventional Motor		
Insecticides		Avant	Proclaim	Alverde	Avant	Proclaim	Alverde	Avant	Proclaim	Alverde
VMD	5 days	-1.000**	-1.000**	1.000**	-1.000**	-1.000**	-1.000**	1.000**	-1.000**	1.000**
	10days	-1.000**	1.000**	1.000**	-1.000**	1.000**	-1.000**	1.000**	-1.000**	1.000**

****. Correlation is significant at the 0.01 level (2-tailed).**

In the current study, *T. absoluta* showed variable responses to different insecticides treatments. Results indicated that the highest toxic effect obtained with emamectin benzoate, metaflumizone and indoxacarb with Matabi and Cawzer sprayers. On the other hand, conventional motor produced the least effect. Our results in line with Moussa *et al*. (2013), who stated that, emamectin benzoate gave satisfactory *T. absoluta* control five days after treatment. Similarly, other studies proved the efficiency of emamectin, metaflumizone and indoxacarb in *T. absoluta* control (Gacemi and Guenaoui, 2012; Hanafy and El-Sayed, 2013 and Santos *et al*., 2011).

In this study, spraying tool affected seriously pesticides efficacy. The highest percent reduction were obtained with either Matabi sprayer or Cawzer sprayer. While less efficient was obtained with conventional motor. Droplet size plays important role in pesticides efficacy. Small droplets (≤ 20 µm in diameter) do not impact efficiently on targets (Mason, 1971). In dry climates, the evaporation of the diluent will cause the droplets to shrink, slightly larger droplets are thus desirable. Also, too large droplets, i.e. ≥300 µm, they not only give poor coverage of the spray plot but also do not penetrate into the sites of the micro-habitat of the target insect (Joyce *et al.*, 1977 and Barry *et al.*, 1978). Obtained results indicates that

equipment with small droplets have a greater effect. This interpreted as a result to small droplets might produce a more uniform dose and, greater number of contact points (Symmons *et al.*, 1991). In addition, the volume of water used to carry the pesticide to the target is one key parameter of sprayer operation that can be varied by the grower to improve the level of coverage of the target crop (Landers, 2004).

Matthews (1977) stated that ,one of the most important factors controlling deposition pattern within the canopy is the droplet size spectrum. Drops with diameters of approximately 100 µm maximize deposition on the pest or foliage, improve the level of control, reduce pesticide costs and minimize ground contamination but are prone to drift. Much lower droplet size may pose a greater environmental threat through spray drift, contamination of soil water and toxicity to nontarget species outside the crop environment (Cooke and Hislop, 1987). Although spray drift is most likely to occur by the smaller drops of ~80µm (Miller, 1993), producing small drops (c. 100µm) within a narrow drop size spectrum utilizes their greater target accuracy and may thereby lower off-target effects. Droplet size affected seriously droplets movement; whereas, droplet falls down increases proportionally with the force of gravity. So, terminal velocity is normally reached in less than 25 mm by droplets smaller than 100µm diameter and in 70 cm for a 500µm droplet (Matthews *et al*., 2014). Ideally there is an optimum droplet size (Himel, 1969) or spectrum which gives the most effective coverage of the target with minimum contamination of the environment.

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Qualitative and quantitative survey of mite and insect pests infesting ornamental plants in Giza Governorate

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Abstract:

Ornamental plants are grown for decorative purposes in gardens and landscape design and play important role in human health and psychology. The aim of this research paper is to study the survey mites and insects infesting three ornamental plants, *Lantana camara* L., *Hibiscus rosa-sinensis* L. and *Acalypha wilkesiana* Müll. Moreover, biweekly sampling was done for two successive years extending from January 2014 to December 2015 to evaluate the population density and the population dynamic of phytophagous pests and their natural enemies infesting these previous plants in Giza Governorate. The survey revealed the presence of 28 mite species and 24 genera belonging to 12 families and four orders collected from leaves and debris under the previous plants; while survey of insect were 23 insect species and 18 genera belonging to 11 families and five orders. The frequent pests were the two spotted spider mites (TSSM), *Tetranychus urticae* Koch (Acari : Tetranychidae), the mealybug, *Ferrisia virgata* (Cockerell) (Hemiptera : Pseudococcidae) and the aphid, *Aphis gossypii* Glover (Hemiptera: Aphididae) and the predatory mites, *Phytoseiulus persimilis* (Athias-Henriot) and *Amblyseius swirskii* (Athias-Henriot) (Mesostigmata: Phytoseiidae). The results showed that the mealybug was the most dominant species on hibiscus followed by the TSSM on acalypha then the aphid on lantana, while the predators were abundant on hibiscus shrubs.

Introduction

Ornamental plant is grown for decoration, rather than food or other by-products, flowerbed, shaped into a hedge. They are most often intentionally planted for aesthetic appeal. *Lantana camara* L. plants are known for their drought tolerance, cold hardiness and colorful flowers that generally bloom from March through October. However, there are several sucking pests that attack lantana.They are spider mites (Acari :

Tetranychidae), aphids (Hemiptera: Aphididae), whiteflies (Hemiptera: Aleyrodidae) and mealybugs (Hemiptera : Pseudococcidae). The aphids, whiteflies and mealybugs excrete a sweet, sticky material called honeydew, which drips down on the plant and accumulates on the leaves and stem of the plant cause sooty mold, a black fungus, often grows on the honey dew and cause problems to plants (Mott and Merchant, 2015). *Hibiscus rosa-sinensis* L. trees or shrubs have attractive flowers and grow relatively easily in sunny areas with good soil drainage, although they occasionally suffer from insect infestations. Hibiscus are susceptible to a variety of insect pests including aphids, scale insects ((Hemiptera: Coccoidea) , mealybugs, thrips (Thysanoptera: Thripidae), whiteflies and mites. Dyer (2018) recorded aphids and whiteflies as serious pests attacking hibiscus plants.

Acalypha wilkesiana Müll is an evergreen shrub which can grow to 3 meters tall. The stem is erect with many branches of closely arranged crown. Leaves are about 12cm long and 5cm wide are coppery green. Mealybugs and red spider mites attack the acalypha plant and cause damage to it.

The objective of this work is to survey and study the population density and the population dynamics of spider mites and ,insect pests and their associated predators on the three ornamental shrubs, *L. camara*, *H. rosa-sinensis* and *A. wilkesiana.* Moreover, the relationship between the population of the piercing sucking pests infesting the three ornamental plants and the maximum and minimum temperature was studied.

Materials and methods

1. Ecological and field study:

1.1. Survey study:

Survey of spider mites and insect pests infesting the ornamental plants *L. camara*, *H. rosa-sinensis* and *A. wilkesiana* in the Orman Park and in different public gardens scattered in the District of Giza Governorate were reported. Moreover, seasonal abundance of the phytophagous pests and their natural enemies were studied in the Land of the agricultural experiments of the Agriculture Research Centre in Giza Governorate throughout two successive years extending from January 2014 to December 2015.

1.2.Population study:

Three ornamental plants were selected for this study, the first was *L. camara* located in an area of about 100 m^2 in the form of hedge each side of 10 m^2long and two m^2 height and a set of these shrubs scattered in the middle of the area; the second plant was *H.rosa-sinensis* found as a set of trees composed of 30 shrubs of 2-3.5m. height around a building for decoration and beautification the place, the third plant was *A. wilkesiana*, a total of 20 shrubs were located on either side of the road of the Agriclture Research Centre entrance, each was 3-4 m high.

1.3. Counting of the phytophagous pests and their natural enemies:

Fifteen leaves from three selected shrub trees of the three mentioned ornamental plants were randomly picked up every two weeks and continued for two successive years 2014 and 2015 to study the population density of the different phytophagous pests and their associated predators. Moreover, debris under every investigated shrub was obtained by taking half kilograms of debris dry weight. Samples of leaves and debris were kept each alone in paper bags tightly closed including all necessary information concerning habitat, locality and date of collection was stuck on each bag, then transferred to the laboratory. Mites were extracted by using very thin spin then mounted in Hoyer's medium on glass microscopic

slides and preserved for identification, while the collected insects were kept in glass vials containing 70% ethyl alcohol and some droplets of glycerine. Sampling continued every two weeks for two successive years. The general climate registered concerning the minimum and maximum temperatures obtained from the meteorological station of Central Laboratory for Agricultural Climate, Agriculture Research Centre.

1.4. Preservation and identification:

Identification of mites of the family level was done according to the key given by Krantz (1978), Zaher (1986) and further segregated to the genus and species level by using different specific keys. The collected insects were identified in the Entomological Collection of the Classification and Pests Survey Research Department, Plant Protection Research Institute, Agriculture Research Centre, Egypt.

2. Population density and population dynamic of the phytophagous pests and their associated predators:

Population dynamic of the phytophagous pests, the mite, *Tetranychus urticae* Koch (Acari : Tetranychidae), the mealybug, *Ferrisia virgata* (Cockerell) (Hemiptera : Pseudococcidae) and the aphid, *Aphis gossypii* Glover (Hemiptera: Aphididae) and their associated predators, *Phytoseiulus persimilis* (Athias-Henriot) and *Amblyseius gossypii* Elbadry (Mesostigmata: Phytoseiidae) and the meteorological studied factors within the inspected periods through the examined years, were discussed.

3.Statistical analysis:

Statistical analysis using Pearson Simple Correlation Coefficient Calculator (Social Science Statistics, 2019).

Results and discussion

1. Survey studies:

1.1. Survey of mites inhabiting *Lantana camara*, *Hibiscus rosa-sinensis* and *Acalypha wilkesiana* plants:

Obtained results of mites inhabiting the mentioned ornamental shrubs indicated the occurrence of 28 mite species and 24 genera belongs to 12 families and four Orders (Table, 1).

1.1.1. Functional group of collected mites:

The collected mite fauna from the three shrubs were classified into the main functional groups: Herbivores, detrivores and predators.

1.1.2. Phytophagous mites (herbivores):

The phytophagous mites are plant feeders of economic importance, comprise four genera, five species belonging to 2 families Teranychidae and Tenuipalpidae of the Order Prostigmata.

1.1.3. Predaceous mites (carnivores):

It considers the biotic factors or the predators associated with the pests in ornamental plants and reduce the degree of infestation by different pest. The identified predatory mites were belonging to 13 genera and 15 species of the families Phytoseiidae, Ascidae and Laelapidae (Mesostigmata) and families Cheyletidae, Stigmaeidae and Eupodidae (Prostigmata).

1.1.4. Miscellaneous mites (fungivores and detrivores):

The miscellaneous are group of different mites, each of which has a different diet i.e. they are saprophytic, fungivores or pollen grain feeders. Represented this group Ameroseiidae (Mesostigmata), Acaridae (Astigmata) and two families of Cryptostigmata (Oribatulidae and Oppiidae). Most mite species in our survey consistent with the results of Embarak and Aiman (2010), they collected the phytophagous mites and their associated predators on lantana plants in Assuit Governorate .

1.2. Survey of insects inhabiting *Lantana camara*, *Hibiscus rosa-sinensis* and *Acalypha wilkesiana* plants:

The total of insects inhabiting *Lantana camara*, *Hibiscus rosa-sinensis* and *Acalypha wilkesiana* plants: belonged to five Orders, 11 families, 18 genera and 23 species (Table, 2) .

1.2.1. Functional group of collected insects:

The collected insect fauna invaded the three ornamental shrubs were classified into the main functional groups: Herbivores, detrivores and carnivores (predators).

1.2.2. Phytophagous insects (herbivores):

The phytophagous insects are plant feeders, provide some important insect pests causing economic damage to different plant Table (2). They comprise 12 genera, 15 species belonging to 8 families and 3 Orders. The most common families were Aphididae, Margarodidae, Pseudococcidae, Aleyrodidae and Cicadellidae.

1.2.3. Predator's insects or the carnivores:

The predators represented by families Coccinellidae and Scarabaeidae (Coleoptera) and Vespidae (Hymenoptera) found on leaves of the three-ornamental plant under investigation.

1.2.3.The miscellaneous insects:

Noted in few numbers in debris under the three ornamental plants and represented by (Formicidae) of the Order Hymenoptera. Embarak and Aiman (2010) surveyed the phytophagous insects and their associated predators on lantana plants in Assuit Governorate , most of their collection was coincide with our results.

2. Population density and population dynamic of phytophagous pests infesting ornamental plants and their associated predators:

The Experimental field of the Agricultural Research Centre was visited twice monthly for two years (January 2014 To December 2015) in Giza Governorate . The seasonal abundance of three pests(*T. urticae*, *F. virgata* and *A. gossypii*) and their associated predators (*P. persimilis* and *A. swirskii*) were determined by counting the number of individuals, occurred on 15 leaves for each of the three selected plants (lantana, hibiscus and acalypha) and placed in paper bags then transferred to the laboratory for examination. Motile stages of the different pests were estimated by counting their numbers per leaves on the low surface and calculating the monthly average number of collected arthropods (Tables, 3-7). Also, the monthly average values of temperature and relative humidity were given in the same Figures.

Table (1): Survey of mites attacking *Lantana camara*, *Hibiscus rosa-sinensis* and *Acalypha wilkesiana* plants and their associated predators during two successive years 2014 and 2015.

Taxonomic rank	Habitat and Habits					
	Lantana		Hibiscus		Acalypha	
	Leaves	Debris	Leaves	debris	leaves	debris
Order: Mesostigmata						
Family:Phytoseiidae						
Amblyseius swirskii Athias-Henriot	15	----	45	----	30	----
Amblyseius hutu Pritchard and Baker	8	----	----	----	----	----
Euseius scutalis Athias-Henriot .	13	----	8	----	15	----
Phytoseiulus persimilis Athias-Henriot	11	----	9	----	8	----
Typhlodromus athiasae Porath and Swirski	10	----	22	----	15	----
Family :Ascidae						
Blattis ociustarsalis (Berlese)	----	6	----	5	----	----
Arctoseius citrates(sellinck)	----	11	----	3	----	6
Proctolaelaps aegyptiaca Nasr	----	7	----	5	----	2
Proctolaelaps orientalis Nasr	----	14	----	8	----	3
Family :Laelapidae						
Laelaspis astronomicus (Koch)	----	4	----	2	----	2
Hypoaspis orientalis (Hafez, El-Badryand Nasr)	----	6	----	2	----	4
Family :Ameroseiidae						
Kleeman niaplumosus (Oudemans)	----	12	----	3	----	5
Order: Prostigmata						
Family :Tetranychidae						
Tetranychus urticae Koch	65	----	55	----	50	----
Oligonychus mangiferus	14	----	20	----	16	----
Eutetranychus orientalis (Klein)	6	----	----	----	9	----
Family :Tenuipalpidae						
Brevipalpus californicus (Banks)	12	----	6	----	8	----
Brevipalpus obovatus Donnadieu	5	----	----	----	----	----
Family :Stigmaeidae						
Agistemus exsertus Gonzalez	8	----	5	----	----	----
Agistemus vulgaris Soliman and Gomaa	4	----	----	----	----	----
Family :Cheyletidae						
Hemicheyletia bakeri (Ehara)	----	16	----	----	----	----
Eucheyletia bakeri Volgin	----	20	----	----	----	----
Family :Eupodidae						
Euopodes niloticus Abo-Awadand El-Bagoury	----	4	----	2	----	2
Order: Astigmata						
Family: Acaridae						
Tyrophagus putrescentiae (Schrank)	----	3	----	1	----	4
Rhizoglyphus robini Claparède	----	6	----	3	----	----
Acarus siro L.	----	14	----	----	----	----
Order: Cryptostigmata						
Family:Oribatulidae						
Siculobata sicula (Berlese)	- 9		- 2		---- 5	
Zygori batulasayedi (El-Badryand Nasr)	- 11		- 6		---- 4	
Family:Oppiidae	4		1		1	

Table (2): Survey of insects attacking *Lantana camara*, *Hibiscus rosa-sinensis* and *Acalypha wilkesiana* plants and their associated predatrs during two successive years 2014 and 2015

Taxonomic rank	Habitat and Habits					
	Lantana		**Hibiscus**		**Acalypha**	
	leaves	debris	leaves	debris	leaves	debris
Order: Thysanoptera						
Family:Thripidae						
Thrips tabaci Lindeman	54	----	45	----	35	----
Order: **Hemiptera**						
Family:Aphididae						
Aphis gossypii Clover	60	----	56	----	32	----
Aphis nerii (Boyer)	11	----	25	----	10	----
Aphis craccivora Koch	8	----	5	----	6	----
Myzus persicae (Sulz.)	24	----	9	----	4	----
Macrosi phumrosae (L.)	----	----	5	----	----	----
Macrosi phumpisi (Harris)	----	----	3	----	----	----
Family:Margarodidae						
F. virgata(Cockerell)	112	----	103	----	90	----
Icerya purchasi Maskell	24	----	----	----	16	----
Family:Pseudococcidae						
Planococcus citri (Risso)	8	----	----	----	----	----
Maconellicoccus hirsutus (Green)	6	----	----	----	----	----
Family: Aleyrodidae						
Bemisia tabaci(Gennadius)	100	----	75	----	65	----
Family: Cicadellidae						
Cicadulina bipunctata (Melichar)	----	20	----	----	----	12
Empoasca decipiens Paoli	----	16	----	9	----	8
Order: Hymonoptera						
Family: Vespidae						
*Vespa orientalis*F.	16	----	24	----	10	----
Family: Formicidae						
Cataglyphis bicolor Fab	----	5	----	3	----	7
Monomorium pharaonis L.	----	13	----	----	----	----
Order: Lepidoptera						
Family: Noctuidae						
Spodoptera littoralis	16	----	5	----	----	----
Order: Coleoptera						
Family: Coccinellidae						
Coccinella septempunctata L	16	----	8	----	11	----
Coccinella undecimpunctata L.	10	----	6	----	4	----
*Cydonia vicina*Mulsant	6	----	2	----	3	----
Family:Scarabaeidae						
Potosia cuprea (Fabricius)	2	----	8	----	1	----
Tropino tasqualida Scopoli	4	----	7	----	1	----

2.1. Monthly fluctuation of the spider mites *Tetranychus urticae*:

Data recorded in Table (3) showed the infestation levels of *T. urticae* motile stages to the three shrub plants mentioned previously throughout two successive years 2014 and 2015.

Two peaks of infestation were recorded, the first occurred in May; when the number of the spider reached 262, 166 and 337 individuals on the three shrubs, lantana, hibiscus and acalypha , respectively; while the second peak of 190.5, 96.5 and 199.5 individuals in September on lantana, and in October on hibiscus and acalypha, respectively.

In the second year 2015, the population density of *T. urticae* motile stages followed the same trend of the first year with slight change. The motile stages of *T. urticae* were peaked in May and June of mean 273 and 243 individuals, on lantana shrub of the first peak; acalypha plant recorded three peaks on hibiscus plant of mean 106.5, 189.5 and 50.5 individuals in March, June and November, respectively. The highest population in the second peak of mean count 71,50.5 and 84.5 individuals, on lantana, hibiscus and acalypha, respectively. The plant acalypha recorded the highest infestation by the spider mites which peaked in June accounted to 352.5 individuals and the second peak recorded 84.5 individuals in November. Pooled data of the two years indicated that the lowest population of the *T. urticae* were recorded in January when max. and Mini. temp., were (19.8 and10.6℃) and (17.6 and 9.9℃) and RH 65 and 50.3%, in the two years respectively; while the highest population recorded when max. and Mini. temp. were (31.2 and 20℃) and (31.6 and 21.4℃) and RH 42.5 and 49.6% in May and June in the two years, respectively.

These results was registered before by El-Halawany *et al.*(1990) who reported a gradual increase in the population of *T. urticae* from February till May, followed by a steady decline in the population through the summer months till the beginning of the next autumn. Moreover, El-Saiedy *et al.* (2011) and El-Halawany and Abou-Setta (2013) indicated that the mite populations reached its peak during May and June then decreased in July to October.

Table (3): Monthly fluctuation of *Tetranychus urticae* on lantana, hibiscus and acalypha in 2014 and 2015 in Giza Governorate.

Date	*Tetranychus urticae* in 2014 on			Temperature		RH%	*Tetranychus urticae* in 2015 on			Temperature		RH%
	Lantana	Hibiscus	Acalypha	Max.	Mini.		Lantana	Hibiscus	Acalypha	Max.	Mini.	
January	29	19	13	19.8	10.6	65	29	17	16.5	17.6	9.9	50.3
February	56	30.5	50	20.5	11.1	61.2	70.5	41	35.5	19.2	10.4	50
March	122.5	67	110.5	23.9	13.7	49.1	120	106.5	88.5	23.7	13.2	53.4
April	201.5	85.5	216	28.5	16.3	45.9	147	64.5	121	26.2	12.6	44.8
May	262	166	337	31.2	20	42.5	273	78	249	31.4	18.9	45.8
June	160	96	207.5	32.4	22	44.9	243.5	189.5	352.5	31.6	21.4	49.6
July	38	15.5	58	34.2	23.3	49	92.5	87	161	34.3	23.1	52.2
August	106	7.5	37	35	24.4	53.6	9.5	13.5	31.5	33.6	25	51.9
September	190.5	20	145	33.4	23.4	53.9	24	13.5	20	35.3	22.4	51.15
October	172	96.5	199.5	28.9	19	52.5	47	35	80	30.6	21.1	58.2
November	77.5	61.5	96.5	24.2	15.4	52.3	71	50.5	84.5	25.1	16	64.1
December	37.5	15.5	33	21.7	12.6	56.9	37	13	38	17.8	11.3	64.1
Total	1452.5	680.5	1503				1164	709	1278			

2.2. Monthly fluctuation of the pest *Ferrisia virgata*:

In the first year, the population of the mealybug (Table, 4) recorded two peaks. The first peak was sharp, observed in June of monthly mean 218.5, 364 and 238 individuals infested lantana, hibiscus and acalypha, respectively, a second peak in October of mean population 113.5 and 162 individuals infested the leaf plants of lantana and acalypha, respectively; while194 individuals infested the plant hibiscus in November. It was observed in the second year 2015, that the infestation rate of the mealybug was higher than the first season. Moreover, leaves of hibiscus shrub suffered the highest infestation in June recorded 493.5 and 344.5 individuals for the two peaks; while those of lantana and acalypha recorded the first peak of the mealybugs population in June of mean count 342 and 352.5 individuals, and 264.5 and 297.5 individuals for the second peak in November and October for the two plants, respectively. Pooled data of the two years indicated that the lowest population of the mealybug was recorded in January (54, 46.5 and 40 individuals) and (77.5, 97.5 and 57.5 individuals) in the two years on lantana, hibiscus and acalypha, respectively; when max. and Mini. temp. was (19.8 and 10.6℃) and (17.6and 9.9℃) and R.H. 65 and 50.3% in 2014 and 2015, respectively. While the highest population recorded in June when max. and Mini. temp. were (32.4 and 22℃) and (31.6 and 21.4℃) and R.H. 44.9 and 49.6% in the two years, respectively. High temp. during July and August and low temp. during November, December and January reduce the populations of the mealybug, *F. virgata* .

Data obtained for *F. virgata* in accordance with that obtained by Abd-El-Said (1997) and Balboul (2003) who revealed the occurrence of two peaks of the Egyptian mealybugs population; the first peak was recorded in June and the second was in October and November. The result showed that the environmental conditions through the months, June and November caused significant increase in the mealybug population. The maximum temperature during July, August and September and minimum temperature during January, February and March reduced the population.

Table (4) : Monthly fluctuation of *Ferrisia virgata* on lantana, hibiscus and acalypha in 2014 and 2015 in Giza Governorate.

Date	*Ferrisia virgata* in 2014 on			Temperature		RH%	*Ferrisia virgata* in 2015 on			Temperature		RH%
	Lantana	Hibiscus	Acalypha	Max.	Mini.		Lantana	Hibiscus	Acalypha	Max.	Mini.	
January	54	46.5	40	19.8	10.6	65	77.5	97.5	57.5	17.6	9.9	50.3
February	50	75	57.5	20.5	11.1	61.2	84.5	157.5	52	19.2	10.4	50
March	44.5	118.5	76	23.9	13.7	49.1	95.5	241.5	111	23.7	13.2	53.4
April	59.5	178.5	127	28.5	16.3	45.9	145	287	152.5	26.2	12.6	44.8
May	103	265.5	185	31.2	20	42.5	242.5	416	256	31.4	18.9	45.8
June	218.5	364	238	32.4	22	44.9	342	493.5	352.5	31.6	21.4	49.6
July	184.5	197.5	204.5	34.2	23.3	49	259	335.5	308.5	34.3	23.1	52.2
August	100	110	135	35	24.4	53.6	180	242.5	326	33.6	25	51.9
September	60	96.5	138	33.4	23.4	53.9	207.5	222	360.5	35.3	22.4	51.15
October	113.5	159	162	28.9	19	52.5	246	344.5	297.5	30.6	21.1	58.2
November	110	194	151.5	24.2	15.4	52.3	264.5	322.5	212	25.1	16	64.1
December	83	85.5	118	21.7	12.6	56.9	202.5	289	132	17.8	11.3	64.1
Total	1180.5	1890.5	1632.5				2346.5	3449	2618			

2.3. Monthly fluctuation of the pest *Aphis gossypii*:

Table (5) revealed that the aphid had one sharp peak of high population in April of monthly mean 181.5, 113.5 and 154.5 individuals for lantana, hibiscus and acalypha, respectively. In November, a second peak was observed of moderate population recorded 52, 55 and 73 individuals for the three-shrub mentioned before, respectively. In the second year, the population of the aphid fluctuated and reached its maximum infestation in May of count 175.5, 136.5 and 140.5 individuals infested lantana, hibiscus and acalypha, respectively. A second peak was observed in December 35.5 and 45 individuals on hibiscus and acalypha plants, respectively. These results are similar to data obtained by Habashi *et al*. (2007), in which the population of aphids reach its peak in the third week of May.

2.4. Monthly fluctuation of the predator *Phytoseiulus persimilis*:

As indicated in Table (6), the population of the predator *P. Persimilis* reached to its maximum population in June on lantana, hibiscus and acalypha of monthly mean (19.5, 16.5 and 16 individuals) and (15, 23.5 and 20 individuals), respectively, when max. and mini. temp. recorded (32.4 and 22℃) and (31.6 and 21.4℃) in the two years, respectively. A second peak was observed in October recorded (12.5 and 9 individuals) and (7 and 5.5 individuals) in lantana and hibiscus, when max. temp. rises to 28.9 and 30.6 ℃ in the two years 2014 and 2015, respectively. Rasmy *et al*. (2003) and Abou-Awad *et al*. (2005) reported that the phytoseiid mites seemed to be important to suppress the population density of *T. urticae.*

2.5. Monthly fluctuation of the predator *Amblyseius swirskii:*

The population of the predator, A. swirskii increased to the first peak in June on the three shrubs of monthly mean 47.5, 98.5 and 87.5 individuals on lantana, hibiscus and acalypha, respectively (Table,7). A gradual decreased observed in the following months then the population increased again to the second peak in October accounted 32, 77.5 and 83.5 individuals on lantana, hibiscus and acalypha, respectively. In the second year, the same observations were indicated, two peaks were observed, the first peak counted 50.5 individuals in June on lantana shrub and 100.5 individuals in July on both other plant's hibiscus and acalypha. The second peak was in October of mean count 35 and 59 individuals on lantana and hibiscus, respectively and 92 individuals in November on acalypha plants. Pooled data of the two years indicated that the lowest population of the predator, *A. swirskii* occurred when the Mini. and max. temp. ranged between 10.6– 19.8℃ and R.H. 65 % in the first year and 9.9-17.6 ℃ and R.H. 50.3% in the second year. The population reached its maximum infestation; when the temperatures ranged between 32.4 – 22℃ and 31.6 – 21.4℃ and relative humidity 44.9– 49.6 % in 2014 and 2015, respectively. The obtained results agree with the finding of Ismail *et al*. (2005) who proved that the predator *A. swirskii* appeared in early of May associated with the sucking pests, whiteflies and aphids and increased gradually till July. In addition, El-Halawany and Abou-Setta (2013) recorded this predator associated with the spider mites on guava trees and reached its peak in August at max. and mini. temp. 35.3 and 24.7℃. Abdel-Gawad *et al.*(1990) detected the highest population of the predator, *A. swirskii* which associated with phytophagous pests [(*Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae) and *Thrips* sp.], during September on different plants in the field (okra, bean, cotton and soya bean plants).

Table (5): Monthly fluctuation of *Aphis gossypii* on lantana, hibiscus and acalypha in 2014 and 2015 in Giza Governorate.

Date	*Aphis gossypii* in 2014 on			Temperature		RH%	*Aphis gossypii* in 2015 on			Temperature		RH%
	Lantana	Hibiscus	Acalypha	Max.	Mini.		Lantana	Hibiscus	Acalypha	Max.	Mini.	
January	13	24.5	45.5	19.8	10.6	65	24	39	55.5	17.6	9.9	50.3
February	46.5	45	59.5	20.5	11.1	61.2	54.5	56.5	42	19.2	10.4	50
March	111.5	85	101	23.9	13.7	49.1	107	96	99.5	23.7	13.2	53.4
April	181.5	113.5	154.5	28.5	16.3	45.9	94.5	73	95.5	26.2	12.6	44.8
May	132	85.5	111	31.2	20	42.5	175.5	136.5	140.5	31.4	18.9	45.8
June	93	32	39	32.4	22	44.9	104	88.5	97.5	31.6	21.4	49.6
July	16.5	8	0	34.2	23.3	49	17.5	22	30	34.3	23.1	52.2
August	0	0	14.5	35	24.4	53.6	0	0	0	33.6	25	51.9
September	27	13	32	33.4	23.4	53.9	6.5	0	0	35.3	22.4	51.15
October	54.5	36	51.5	28.9	19	52.5	58	5.5	7	30.6	21.1	58.2
November	52	55	73	24.2	15.4	52.3	21	21	29	25.1	16	64.1
December	14.5	35	49	21.7	12.6	56.9	8.5	35.5	45	17.8	11.3	64.1
Total	742	532.5	730.5				671	573.5	641.5			

Table (6): Monthly fluctuation of *Phytoseiulus persimilis* on lantana, hibiscus and acalypha in 2014 and 2015 in Giza Governorate.

Date	*Phytoseiulus persimilis* in 2014 on			Temperature		RH%	*Phytoseiulus persimilis* in 2015 on			Temperature		RH %
	Lantana	Hibiscus	Acalypha	Max.	Mini.		Lantana	Hibiscus	Acalypha	Max	Mini	
January	0	0	0	19.8	10.6	65	0	0	0	17.6	9.9	50.3
February	3	1	0	20.5	11.1	61.2	4	1.5	2	19.2	10.4	50
March	6.5	7.5	1.5	23.9	13.7	49.1	6.5	7	4.5	23.7	13.2	53.4
April	11	14	9	28.5	16.3	45.9	11	12.5	10	26.2	12.6	44.8
May	13.5	16.5	11.5	31.2	20	42.5	12.5	17	16.5	31.4	18.9	45.8
June	19.5	16.5	16	32.4	22	44.9	15	23.5	20	31.6	21.4	49.6
July	2.5	4	7	34.2	23.3	49	2.5	15	12	34.3	23.1	52.2
August	0	0	1.5	35	24.4	53.6	0	2.5	2	33.6	25	51.9
September	5	1	1	33.4	23.4	53.9	35	1	0	35.3	22.4	51.15
October	12.5	12.5	9	28.9	19	52.5	7	5.5	1.5	30.6	21.1	58.2
November	1.5	6.5	5.5	24.2	15.4	52.3	1.5	7.5	1.5	25.1	16	64.1
December	0	0	0	21.7	12.6	56.9	0	0	0	17.8	11.3	64.1
Total	75	79.5	62				95	93	70			

Table (7): Monthly fluctuation of *Amblyseius swirskii* on lantana,hibiscus and acalypha in 2014 and 2015 in Giza Governorate.

Date	*Amblyseius swirski* in 2014 on			Temperature		RH%	*Amblyseius swirski* in 2015 on			Temperature		RH%
	Lantana	Hibiscus	Acalypha	Max.	Mini.		Lantana	Hibiscus	Acalypha	Max.	Mini.	
January	5.5	11.5	1.5	19.8	10.6	65	4	5.5	6	17.6	9.9	50.3
February	6.5	16	10	20.5	11.1	61.2	11	12.5	7.5	19.2	10.4	50
March	13.5	32.5	25	23.9	13.7	49.1	20.5	32	20.5	23.7	13.2	53.4
April	14	54	41.5	28.5	16.3	45.9	28	62	37	26.2	12.6	44.8
May	35.5	76.5	68.5	31.2	20	42.5	34.5	83	65.5	31.4	18.9	45.8
June	47.5	98.5	87.5	32.4	22	44.9	50.5	99.5	92.5	31.6	21.4	49.6
July	32	87.5	63.5	34.2	23.3	49	38	100.5	100.5	34.3	23.1	52.2
August	20	62.5	40	35	24.4	53.6	27.5	54.5	69	33.6	25	51.9
September	20.5	51.5	43.5	33.4	23.4	53.9	24.5	49.5	44.5	35.3	22.4	51.15
October	32	77.5	83.5	28.9	19	52.5	35	59	45	30.6	21.1	58.2
November	31	49	39	24.2	15.4	52.3	30.5	52.5	92	25.1	16	64.1
Dec.	12.5	11	14.5	21.7	12.6	56.9	16.5	22	23	17.8	11.3	64.1
Total	270.5	628	518				320.5	632.5	603			

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Efficacy of thermal fog and partial spraying techniques for controlling *Bactrocera zonata* and *Ceratitis capitata* (Diptera: Tephritidae) on mango in Qalyubiya Governorate

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Abstract:

This investigation may be considered one of the rare attempts to make evaluation between thermal fog and partial spray techniques to control both of *Bactrocera zonata* (Saunders) and *Ceratitis capitata* (Wiedemann) (Diptera: Tephritidae **)** on tall mango orchards in Egypt . Thermal fog as a new technique gave promising results represented in good distribution and penetration of fog clouds at various parts of tree during the evening or at sun rise under climatic inversion condition, saving time , water consumptions and labors required for chemical application when compared with partial spray technique . Also a drastic difference in the rate of performance (fed. / day) between IGEBA TF 35 handle fogger machine and Hand Heldsprayer CP-3 (17.12 and 1.5 fed./ day), respectively . Data showed that *C. capitata* was more sensitive than *B. zonata* represented in percentages reduction by using thermal fog and partial spray techniques . A highly significant difference was recorded between recommended dose of thermal fog and partial spray techniques for controlling both *B. zonata* and *C. capitata* represented in mean number of captured flies in Mcphail traps . No significant difference between half recommended dose with thermal fog technique and partial spray technique for controlling fruit flies was also noticed . Moreover significant difference in mean numbers of captured flies inside Mcphail traps were recorded four weeks than between two weeks after application of *B. zonata*.

Introduction

Mango orchards areas in Egypt are approximately 150433 fed. Mean production about was 5.0 Tons / Feddan annually (Anonymous , 2014). True fruit flies (Diptera: Tephritidae) include over 4000 species, many of which constitute enormous threats to fruit and vegetable production worldwide (Benelli, 2015). Fruit fly control with full sprays started with inorganic insecticides , such as chlorinated hydrocarbons, organophosphates and synthetic pyrethroids. Addition of protein food baits to insecticide sprays

reduced the amount of pesticide needed for fruit fly control and has been used successfully in many eradication programs . Female flies, need of protein for full ovarian development and egg production (Roger *et al.*, 2015) . *Bactrocera zonata* (Saunders) (Diptera: Tephritidae) has been recognized as a serious insect pest during the last decade attacking a wide range of fruits in Egypt (Fahmy *et al.,* 2013) . *Ceratitis capitata* (Wiedemann) (Diptera: Tephritidae) has become invasive throughout the world (De Meyer *et al.,* 2007) . Modern control trends depending mainly on minimizing the hazards of insecticides to the environment , the costs time of control operations and in the mean time increasing the efficacy by using thermal fog technique (Brown and Watson ,1953) .They found that , fog was drifted across a swath of 150 m , but it coud be effective for 400 m . Mathews (1979) stated that fogging was particularly useful for the control of flying insect not only through contact with droplets , but also , by the fumigant effect of volatile pesticide . Lim and Abdul – Aziz Bin Kader (1978) evaluated ground fogging technique by using a new oil based fungicidal formulations against secondary leaf fall *Phytophthora* (Peronosporales : Peronosporaceae) and pod rot in comparison with duster sprayer to dust sulphur for controlling rubber leaf diseases . They concluded that , fogging technique was more rapid and economic at large scale treatments for controlling these diseases than dust sulphur technique . Hindy *et al.* (1995) compared between the traditional chemical spray method and a thermal fog technique for controlling *C. capitata* infested mango orchards in Egypt which gave good distribution and penetration of droplets at various parts of mango trees , saving time required for chemical application with a few amounts of chemical insecticides than traditional ground motor spray. Hindy *et al.* (1999) evaluated the bio-residual activity of summer oil 1 % KZ oil produced from three ground spray equipment against citrus leaf miner. Results indicated that thermal fog technique with a half dose rate and conventional ground motor sprayer as recommended dose rate showed higher mortality of thermal fog technique than the other treatments . Mc Govern *et al*. (1986) indicated that , trimedlure was a powerful attractant for males of *C. capitata* . Hafez and Ezzat (1967) used traps baited with 3 % solution of diammonium phosphate for *C. capitata* . Buttery *et al.* (1983) stated that Hand Heldthermal foggers were highly effective, with more than 90% reduction of both laid eggs and females mosquitoes (Boubidi *et al.,* 2016) .

The aim of the present investigation is to spot light on controlling fruit flies by using thermal fog technique in comparison with partial spraying technique as an attempt to save time of chemical application getting down insecticides , reducing water as a carrier and lowering cost of spray in mango orchards .

Materials and methods

1. Experimental area:

Experiments were conducted in mango orchards (Season, 2016) at El-Qanater Elkhairia district , Qalyubiya Governorate. Eleven feddans of mango trees were chosen and sprayed during summer , 2016 . The mean number of trees per feddan was about 50 trees. The height of trees in the area under investigation ranged between 6 – 8 meters . Two concentrations (full dose and half dose) of the recommended chemical insecticides for controlling *C. capitata* and *B.zonata* on mango trees (Malathion 57 % EC) + 100 ml of summer mineral oil (KZ- oil 95 % EC) and solar solution as a carrier were used by a Hand Heldthermal fog generator (IGEBA) TF 35. Each treatment was

conducted at 4.0 feddans sprayed cross wind at sun set . An area against wind direction of about half feddan was left untreated for each tested concentration as control area. Untreated ten rows mango trees were left between each two treatments to avoid over lapping due to fog drift. Spraying operations started about 10 days prior reaching physiological ripening stage of mango fruits where they well still less susceptible and / or to avoid infestation with med fly and peach fruit fly . Another spraying technique was partial spraying which consisted of mixed solutions consisted of 500 cm3 (Malathion 57 % EC) insecticide + one liter of food attraction bominal + 18.5 liters water used by Hand HeldKnapsack sprayer CP-3 . The solution was used for spraying trunks of trees at branching area using about 150 – 200 cm3 / tree. Each 20 liters solution sprayed downwind at sun set. Spraying operations were at all rows of mango trees used by target spraying technique.

2. Used Mcphail traps :

Four Mcphail traps (Mcphail, 1937) for each treatment , were used by putting about 200 ml of 3% diammonium phosphate solution in each trap. All prepared traps were distributed in a completely randomized design. The distance between two each adjacent traps was about 15 meters and the traps were hanged at about 1.5 – 2 meters in a shadow place of the trees was added. The traps were weekly inspected , where fresh baited solution along a period of 5 weeks was changed . Captured females and males of *B.zonata* and *C. capitata* were counted and recorded .

3.Statistical analysis:

Table (1) showed the techno operational data of two ground application techniques , calibration and performance rate for machines was according to Hindy (1992) . The percentages reduction of treatments calculating according to formula Hendrson and Tilton (1955) . Data was analyses by using ANOVA in SAS (SAS Institute, 1998) .

Table (1) : Techno- Operational data , spray parameters , insecticides application rates and rate of performance by using " IGEBA® thermal fog generator and CP-3 Hand Heldhydraulic sprayer .

Equipment / **Items**	**Thermal Fog IGEBA® TF 35**	**CP – 3 Knapsack Sprayer**
Manufacture	Germany	U K
Weight , empty in KG.	7.9	3.7
Dimensions .L . W . H in cm.	137.5.27.34	43.33.25
Solution Tank capacity in L.	5.7	20.0
Fuel consumption in L/H .	2.0	-
Effective Horizontal reach out doors , in m.	15.0	0.5
Power supply	4 dry batteries 1.5 V.	ManualPump
Flow rate , approx . in L/ Min	42	43.2
Machine speed km / hr	1.2	1.2
Atomization type	Pneumatic	Hydraulic
Kind of spray	Drift	Target
Direction of Travel	cross wind	Down wind
Rate of performance at (6) working hours (fed / day)	17.12	1 – 1.5
Quantity of Malathion (cc – in tank / treatment	1000 , 500	500 m Malathion + 1 L. Buminal
Quantity of KZ oil cc in tank / treatment	500 ml	-
Quantity of Solar as carrier solution / treatment (l.)	4.2 , 4.7 l / tank / treatment	-
Quantity of water as carrier solution in tank / treatment (l.)	-	18.5
spray time / fed. (Min.)	19.0	60.0

Weather conditions during spraying operations , Temp. 29 C , R. H. 70% WIND VELOCITY 1 – 1.5 m / sec.

Results and discussion

1. Comparison between thermal fog and partial spray techniques :

Data in Table (1) clearly showed that thermal fog technique offer tangible benefits when compared with partial spray technique. Less materials per feddan was applied . Consequently more safe time of application , less spray labor . No water was needed in treatments for fog , while in partial spraying large quantities of water is required . The swath width in fog was about 15 m. moved and penetrate the trees in 19 Min / feddan . But the swath width was about 0.5 meter in each tree in partial spray technique , the time to spray one feddan about one hour. Performance rate of one machine of thermal fog technique could covered 17.12 feddans / one tank but in other side partial spray technique made by Hand Heldspryer CP-3 covered from 1 – 2.5 feddans / day . Fog machines could be done at temperature inversion conditions , usually either early morning or at evening , so that , the fog remains close to the ground and drifted slowly to different levels of mango trees but partial spray technique a localized solution was used for spraying tree trunks at branching area using about 100 – 200 cm3 / tree . Technically thermal fog technique could gave good droplets distribution , penetration and full coverage on mango trees certainly if used pyrthroid – insecticides which induce knock down effect more than phosphorus – insecticides and give good protection against flying fruit insects than partial spray technique. These results agree with Hindy *et al* . (1995) who stated that thermal fog gave promising results represented in good distribution and penetration on droplets at various parts of tall mango trees , saving time required for chemical application and less consumed amount of chemical insecticide due to moving fog clouds on ground through mango trees and air drifted was about 37.1 % opposed to 77.5 % in case of using conventional sprayer as drift spray and lost spray between plants . Also data in Table (1) illustrated that , the productivity (fed. / hour) between thermal fogger machine and CP- 3 sprayer were 4.28 , and 0.14 respectively . On the other hand , the rate of performance (fed. / day) between thermal fogger machine and CP- 3 sprayer were 17.12 and about 1–1.5, respectively. Thermal fogger revealed a drastic differences in rate of performance which it had been made more quick control for large areas of mango orchards in short time than CP- 3 sprayer with spraying a partial sprayer .

2. Captured *Bactrocera zonata* flies inside Mcphail traps :

Mcphail traps were made of plastic powered with food attractant diammonium phosphate 3 % which attract both male and female of *B. zonata* and *C. capitata* into the orchards . It could not attract the fruit flies from vast distances out side the mango orchard, therefore these traps captured only the flies in mango orchards under investigation . Data indicated in Table (2) revealed that , no flies were captured into traps at recommended dose with thermal fog technique until the end of two weeks after applications . Data showed in Figure (1) mean reduction rates in flies population *B. zonata* at recommended dose rate with thermal fog technique and partial spraying technique was 80 % and 47. 5, respectively. After the first and second weeks of thermal fog at recommended dose of *B. zonata* was zero , so the percentage reduction 100 %.These results are in accordance with Mahmoud *et al*. (2017) who stated that, for controlling *B. zonata* and *C. capitata* in Egypt it was recommended to increase area and time of spraying from three to four or five times . No significant difference between half recommended dose treatment with thermal fog technique and partial spraying technique

treatment was recorded. The following mean reduction percentages were 37.73 and 47. 5, respectively (Figure, 1). In the third and fourth weeks after application a drastic increase in flies population was recorded after application in both of control and half recommended dose of thermal fog, and partial spraying technique, respectively. However in case of recommended dose of thermal fog technique the population of flies was very low at the same period. This may be attributed to the effect of space fumigation controlled flying insects such as *B. zonata*. Fogging at higher volume rates and with a greater proportion of larger droplets sometimes referred to as a wet fog will leave deposit on foliage. This may provide a longer residual effect, but at high flow rates foliage close to the nozzle is liable to be damaged by an overdose of large droplets. Although less effective as a space treatment. Fogging is particularly useful for the control of flying insects (Boubidi *et al*., 2016), moreover Matthews (1979) stated that mortality occurred through contact with droplets but also by the fumigation effect of a volatile pesticide. So the pesticide in a gas form is capable of reaching flies hidden on lower leaf surfaces inducing irritation of pest and hence slip out of their hiding place and receive a lethal dose from air borne fog droplets. The effect space fumigation was produced when stable aerosols smaller than 5 m was generated and coated on the leaves with very fine film of droplets ranged between 1–30 µm in both upper and lower surface of plant leaves through the good penetration of very fine droplets sizes under inversion condition occur .

3. Captured flies *Ceratitis capitata* inside Mcphail traps :

Data in Table (3) showed that *C. capitata* was more sensitive for treatments when compared with *B. zonata* in Table (2) . There was no significant difference among the two tested treatments of thermal fog technique and partial spraying technique . According Figure (2) the reduction percentages were 90.5 and 71.4 and 92.8 at recommended dose, half recommended dose and partial spraying technique, respectively . After the first , second and third weeks of treatments the population of *C. capitata* was zero , so the percentage reduction reached 100 % . The mean number of captured *C. capitata* flies inside Mcphail trap in control were less than the mean number of captured *B. zonata* inside traps . Also no significant difference among weeks after application were noticed . Boubidi *et al*. (2016) evaluated the efficiency of truck mounted ULV and thermal fogger chance that a droplet will come in contact with a mosquito in the sentinel cage in a thermal fog application versus a ULV application . We concluded that in the event of out breaks of fruit flies on mango orchards during summer season partial spray technique is unlikely to have significant impact on transmission but that despite being highly labor – intensive , thermal fog technique dispensed from portable sprayers was the method of choice. Clearly this was not practicable on any large scale but might be useful in the event of potential " hot – spots " of local transmission . Thermal fog technique for controlling insect pests infesting fruit trees gave promising results represented in less consumed amounts of chemical insecticides , saving time required for application and thorough spray droplet distribution penetration and deposition . Also from our data , could be concluded that data obtained from this study are promising , but may be , further investigations are required to make respective accumulation of fog spray's with little doses to obtained more control operations than one treatment .

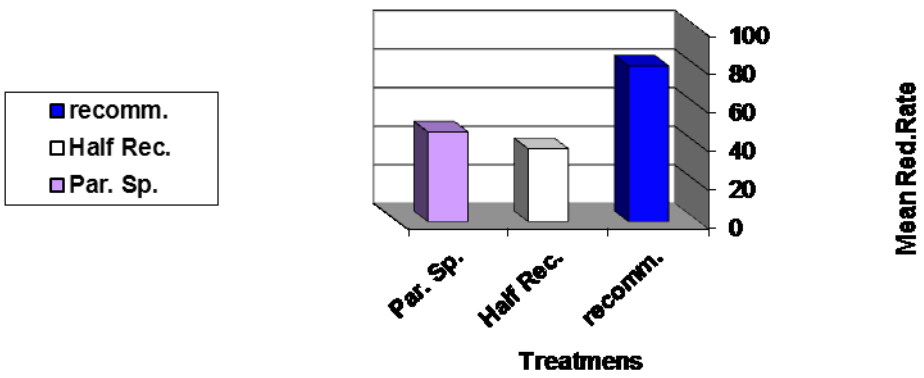

Figure (1): Mean reduction rate of *Bactrocera zonata* by using thermal fog techique compared to partial techique.

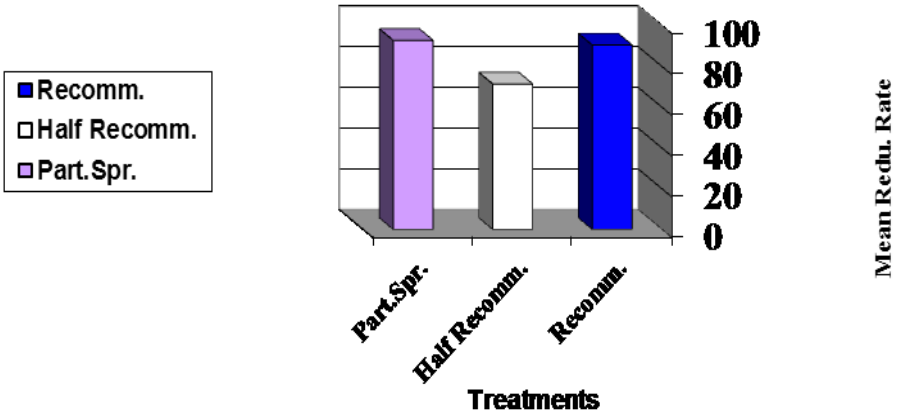

Figure (2): Mean reduction rate of *Ceratitis capitata* by using thermal fog techique compared to partial spray techique.

Table (2): Mean number captured and reduction rate of *Bactrocera zonata* inside Mcphail traps by using thermal fog techique compared to partial spray techique on mango orchard at Elkanater Elkhairia district , Qualubyia Governorate during season 2016.

Date		After applications					Mean	% Redu.
Treatments		One week	Two weeks	Three weeks	Four weeks	Five weeks		
Thermal fog	Recommended dose	0	0	3.25	3.25	1.25	1.55	80.7
	Half Recommended dose	1.75	2.75	10.75	9.25	0.5	5	37.7
Partial spray		1.25	0	11.25	8.5	0.5	4.3	47.5
Control		2.25	4.25	19.66	12.75	1.25	8.03	
LSD		1.963						

Table (3): Mean number captured and reduction rate of Ceratitis capitata inside Mcphail traps by using thermal fog techique compared to partial spray techique on mango orchard at Elkanater Elkhairia district , Qualubyia Governorate during season 2016

Date		After applications					Mean	% Redu.
Treatments		One week	Two weeks	Three weeks	Four weeks	Five weeks		
Thermal fog	Recommended dose	0	0	0	1	0	0.2	90.5
	Half Recommended dose	2.5	0.5	0	0	0	0.6	71.4
Partial spray		0.25	0	0.5	0	0	0.15	92.8
Control		2.75	1.5	3.75	1.25	1.25	2.1	
LSD		0.666						

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The effect of droplets distribution of insecticides on bioresidual activity of piercing sucking insects (Hemiptera) infesting eggplant by using ground spraying equipment

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Eggplant, bioresidual activity, *Bemisia tabaci* , *Aphis gosspyii*, Imidaclopride, Acetampirid, Lufenuron, low volume and ground equipment.

Abstract:

Eggplant is one of the most common tropical vegetables cultivated of the world. It contains a good amount of vitamins, minerals and fiber in few calories. Piercing and sucking insects (Hemiptera) damage crops by inserting their mouthparts into plant tissue and sucking juices. Heavily infested crops become yellow, wilted, deformed or stunted and may eventually die. Some sucking insects inject toxic materials into the plant while feeding and some transmit virus diseases. Field experiments were carried out in an area of about 19 Kirats planted with eggplant varity (Soma kafear) during two successive seasons 2017 and 2018 in 7th August at Qaha, Qalyubiya Governorate. The selected area was split into 9 plots and control plots. Three products were sprayed Imidaclopride, Acetampirid (Neonicotinoids) and Lufenuron (IGRs) of recommended dose rates and one treatment left without spraying as control by using Knapsack motor sprayer (Cifarilli) (20 L./ fed.), Economy Micron ULVA sprayer (15 L/Fed.) and Hand-Held compression sprayer (Kwazar) (94 L/Fed.). Data indicated that, all tested compounds induced significant negative influenced on both *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae) and *Aphis gossypii* Glover (Hemiptera: Aphididae) nymphs survival. Both Imidaclopride and Acetampirid revealed successful results followed by Lufenuron. It could be recommended that using those compounds with low volume spraying equipment with not less than (15L/ fed.). The data showed that Knapsack motor sprayer (Cifarilli) was the best equipment to control both *B. tabaci* and *A.gossypii* infesting eggplant. The rate of performance of Knapsack motor sprayer (Cifarilli) was 12 fed./day. It was the best equipment, but the lowest rate of performance was Hand Held compression sprayer (Kwazar) since it could spraying only 2.5 fed./day.

Introduction

Eggplant has a very low caloric value and is considered among the healthiest vegetables for its high content of vitamins, minerals and bioactive compounds for human health (Docimo *et al*., 2016). The top five producing countries are China (28.4 million tons), India (13.4 million tons), Egypt (1.2 million tons), Turkey (0.82 million tons) and Iran (0.75 million tons) (FAO, 2014). Piercing and sucking insects are dangerous pests which infested eggplant (*Solanum melongena* L.) and cause great hazarads to it. In Egypt, majority of interest was directed to the type, dosage rate of insecticides used, while a lesser attention was given to the application methods.

A comparative studies on the efficiency of different ground sprayers was carried out by (Hindy, 1992 and Hindy *et al*.,1997) who found a significant variation in the spray deposit due to arrangement of the nozzles, spray technique and rate of application. The world global attention was directed to minimization of spraying volumes and the control costs which may be achieved by using a cheap and effective insecticides or using developmental ground spraying technique with low application costs per feddan (Magdoline *et al*., 1992 and Matthews, 1992). Maintaining sprayers for pesticide application in a good state of repairing and proper working in order to reduce their harmful effects on human health and environment (Dokic *et al.*, 2018). The aim of this work is to determine the best insecticide and equipment controlling *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae) and *Aphis gossypii* Glover (Hemiptera: Aphididae) on eggplant with conservation of agricultural environment.

Materials and methods

1. Tested compunds:

1.1. Imidaclopride (Qwadoor ®), 20% S.L. , 100 cm^3/ 100L. water, (Neonicotinoids) , Acetylcholinesterase inhibitor .

1.2. Acetamiprid (Plan ex®), 70 % W.G. , 50 gm /fed. , (Neonicotinoids),Acetylcholinesterase inhibitor.

1. 3. Lufenuron (Match®), 5%E. C. , 160 / cm^3 / fed. (IGRs), Chitin synthesis inhibitor.

2. Spraying equipment tested on eggplant:

Three ground application equipments were selected to perform the scope of this work, as commonly used equipment in applying pesticides on eggplant. These are, Economy Micron ULVA sprayer, spraying volume (15L./fed.), UK made; Knapsack motor sprayer (Cifarilli), Spraying volume (20 L./fed.) ,Italy made and Hand- Heldcompression sprayer (Kwazar), Spraying volume (94 L./fed.), Poland made. The tested equipments could be represented according to the technical categorization mentioned in Table (1). Calculations of productivity and rate of performance were recorded as described by Hindy (1992).

Table (1): Techno-Operational data of certain ground sprayers applied on eggplant field during seasons (2017-2018).

Equipment	Motorized Knapsack sprayer (Cifarilli)	Spinning disc (ULVA) sprayer	Hand- Held compression sprayer(Kwazar)
Type of atomization	Pneumatic Mechanical	Rotary*	Pneumatic manual
Nozzle type	Air shear nozzle	Spinning disc	Hollow cone nozzle
Pump type	Centrifugal fan	-	Compression air pump
Number of nozzles	1	1	1
Pressure (bar)	-	-	From 7 to 1
Spray tank (L.)	20	1+10	8
Rate of application (L/fed.)	20	15	94
Working speed (Km/h.)	2.4	2.4	2.4
Swath width (m.)	5	1.0	1.0
Flow rate (L/Min.)	1	0.150	0.90
Spray height (m.)	0.5	0.5	0.5
Type of Spraying	Target spraying technique in all treatments .		
Productivity * (fed./h.)	2.85	0.571	0.425
Rate of performance* (fed./day)	12	3.04	2.5

* Number of spraying hours=8hours daily.
*Number of workers=2
* Hand carried-4 Battery operated spinning disc sprayer.
* Calculations of productivity and rate of performance after Hindy (1992).

3. Execution of field experiments:

3.1. Arrangements of the experiments:

Field experiments were carried out during two successive seasons 2017 and 2018 on 9th August in private eggplant field located at Qaha District, Qalyubiya Governorate . The eggplant cultivated varity was Soma Kafear planted at 10th of April in the two seasons, the experiments were done under local meteorological conditions of 37°C average temperature, 60% average R.H. and 2.5 m/sec. as an average wind velocity during spraying operations. The selected area of 19 Kirats was split into 9 plots and control plot. The area of each plot was 2 Kirats , two rows of eggplant plants between treatments were not sprayed as barrier zones to avoid drift spray between treatments, spraying operations have not been done with insecticides before execution the field experiment. The experimental fields were sprayed with recommended dose rate and one treatment left separated without spraying as a control, with three alternative insecticides Imidaclopride, Acetamiprid and Lufenuron, respectively. All treatments sprayed as target spraying technique. In each plot five eggplant plants were selected and remarked to define *B. tabaci* and *A. gossypii* nymphs numbers and follow

the results before and after one , five and seven days from spraying.

3.2. Bioassay procedure:

Field experiments were conducted on eggplant field highly infested with *B. tabaci* and *A. gossypii* nymphs. In order to evaluate the tested compounds on them, pre-treatment count was recorded before spraying at five marked plants for each treatment and post-treatment counts was recorded after 1,5 and 7 days from spraying treatments to determine the effect of the tested chemicals by different spraying equipment.

3.3. Phytotoxic effect:

Determined by recording any colour change, leaf curling or flaming up to 8 days after spraying, according to Badr *et al*. (1995).

4. Calculation and data analysis:

4.1. The reduction percentages in the field experiment was calculated according to Henderson and Tilton (1955).

4.2. The statistical analysis of results was achived according to SAS (1996) program for biological studies: Duncan's (Duncan, 1955) for biological evaluation of insecticides in field.

5. Calibration and performance adjustment of the tested equipment:

5.1. Collection of spray deposit:

Before spraying each eggplant field treatments, a sampling line was constructed of five wire holder fixed in diagonal line at each treatment to collect the lost spray between plants; each wire holder top has a fixed with water sensitive paper (Novartis Cards) on it. Also, each five eggplant plants, the water sensitive paper cards were put at plant; to collect the droplets deposit on eggplant leaves, were designed according to the method described by Hindy (1989). All cards were collected and transferred carefully to the laboratory for measuring and calculating the number of droplets/cm² and its volume (VMD) µm in all treatments.

5.2.Determination of spray deposit:

Number and size of blue spots (deposited droplets) on water sensitive papers (Novartis cards) measured with a special scaled monocular Japanies lens (Strüben)® (15X).The volume mean diameter (VMD) µm and number of droplets in one square centimeter (N/cm²) were estimated according to Hindy (1992).

Results and discussion

1. Bioresidual activity of Imidaclopride against *Bemisia tabaci* and *Aphis gossypii* infesting eggplant :

Efficiency of Imidaclopride represented as mortality percentages after 24 hours of spraying as presented in Tables (2 and 3). The highest reduction in population of *B.tabaci* nymphs was occurred by Economy Micron ULVA sprayer (15 L/fed.) the droplet sizes were 153 , 129 and 156 and N/cm^2 were 156, 312 and 176.The mean mortality percentages after one day of the two seasons (2017 and 2018) were 84,81.5 and 63% for initial for recommended dose sprayed with Economy Micron ULVA sprayer, Knapsack motor sprayer (Cifarilli) sprayer and Hand-Held compression (Kwazar) sprayer and the general mean reduction % of two seasons 94.7,93.8 and 84.3 for residual sprayed with Economy Micron ULVA sprayer, Knapsack motor sprayer (Cifarilli) and Hand-Held compression (Kwazar) sprayer , respectively . The highest reduction in population of *A.gossypii* nymphs were occurred by Economy Micron ULVA sprayer. The mean mortality percentages of *A.gossypii* nymphs of the two seasons (2017 and 2018) after one day of treatment by using Imidaclopride formulation were 92 ,88 and 72 % for initial and the general mean reduction % of two seasons were 97.3 , 96 and 88 for

residual for recommended dose sprayed with Economy Micron ULVA sprayer, Knapsack motor sprayer (Cifarilli) sprayer and Hand-Held compression (Kwazar) sprayer ,respectively.

2. Bioresidual activity of Acetamprid against *Bemisia tabaci* and *Aphis gossypii* infesting eggplant :

Efficiency of Acetamprid represented as mortality percentages after 24 hours of spraying as presented in Tables (2 and 3). The highest reduction in population of *B.tabaci* nymphs was occurred by Knapsack motor sprayer (Cifarilli) (20 L/fed.) the droplet sizes were 156 , 126 and 156 and N/cm^2 were 152, 344 and 183, respectively . The mean mortality percentages after one day of the two seasons (2017 and 2018) were 80, 81.5 and 66.5% for initial for recommended dose sprayed with Economy Micron ULVA sprayer, sprayer and Hand-Held compression (Kwazar) sprayer ,and 93.3 ,93.8 and 87 the general mean reduction % of two seasons for residual sprayed with Economy Micron ULVA sprayer, Knapsack motor sprayer (Cifarilli) and Hand-Held compression (Kwazar) sprayer , respectively . The highest reduction in population of *A.gossypii* nymphs were occurred by Knapsack motor sprayer (Cifarilli) (20 L/fed.) The mean mortality percentages of *A. gossypii* nymphs by using Acetamprid formulation after one day of the two seasons (2017 and 2018) were 85.5 ,87 and 74.5 % for initial for recommended dose sprayed with Economy Micron ULVA sprayer, Knapsack motor sprayer (Cifarilli) sprayer and Hand-Held compression (Kwazar) sprayer, the general mean reduction % of two seasons were 87.5 , 91 ,89.1 % for residual with recommended dose sprayed with Economy Micron ULVA sprayer, Knapsack motor sprayer (Cifarilli) sprayer and Hand-Held compression(Kwazar) sprayer ,respectively.

3.Bioresidual activity of Lufenuron formulation against *Bemisia tabaci* and *Aphis gossypii* infesting eggplant :

Efficiency of Lufenuron represented as mortality percentages after 24 hours of spraying as presented in Tables (2 and 3). The highest reduction in population of *B.tabaci* nymphs were occurred by Knapsack motor sprayer (Cifarilli) (20 L/fed.) the droplet sizes were 132 , 147 and 156 (VMD) µm and N/cm^2 were 148,329 and 176 the droplet sizes were 156 , 126 and 156 and N/cm^2 were 344,131 and 183. The mean mortality percentages after one day of the two seasons (2017and 2018) were 63.5, 66.5 and 63.5% for initial for recommended dose sprayed with Economy Micron ULVA sprayer, sprayer and Hand-Held compression (Kwazar) sprayer ,and 84.1 , 86.7 and 84.4% the general mean reduction % of two seasons for residual sprayed with Economy Micron ULVA sprayer, Knapsack motor sprayer (Cifarilli) and Hand-Held compression(Kwazar) sprayer, respectively .The highest reduction in population of *A.gossypii* nymphs was occurred by Knapsack motor sprayer (Cifarilli) (20 L/fed.) The mean mortality percentages of *A. gossypii* nymphs by using Lufenuron formulation after one day of the two seasons (2017 and 2018) were 71 ,73.5 and 73.5 % for initial for recommended dose sprayed with Economy Micron ULVA sprayer, Knapsack motor sprayer (Cifarilli) sprayer and Hand-Held compression(Kwazar) sprayer, the general mean reduction % of two seasons were 87.5, 91 ,89.1 % for residual with recommended dose sprayed with Economy Micron ULVA sprayer, Knapsack motor sprayer (Cifarilli) sprayer and Hand-Held compression(Kwazar) sprayer ,respectively.

Table (2): The relation between droplets distribution obtained by the tested ground spraying equipment and the corresponding mortality of *Bemisia tabaci* nymphs infesting eggplant during seasons (2017-2018) in Qalubyia Governorate.

Insecticide and dose rate/ fed.	Tested sprayer	VMDμm	N / cm^2	% Mortality	
				Initial mean *	Residual mean *
Imidaclopride (400 cm^3)	Micron ULVA	153	156	84	94.7
	Cifarilli	129	312	81.5	93.8
	Kwazar	156	176	63	84.3
Acetamprid (50 gm)	Micron ULVA	156	131	80	93.3
	Cifarilli	126	344	81.5	93.8
	Kwazar	156	183	66.5	87
Lufenuron (160 cm^3)	Micron ULVA	132	148	63.5	84.1
	Cifarilli	147	329	66.5	86.7
	Kwazar	156	176	63.5	84.4

VMD = Volume Mean Diameter. N / cm^2 = Number of droplets per square centimeter.
*Average of two seasons.

Table (3): The relation between droplets distribution obtained by the tested ground spraying equipment and the corresponding mortality of *Aphis gossypii* nymphs infesting eggplant during seasons (2017-2018) in Qalubyia Governorate.

Insecticide and dose rate/ fed.	Tested sprayer	VMDμm	N / cm^2	% Mortality	
				Initial mean	Residual mean
Imidaclopride (400 cm^3)	Micron ULVA	153	156	91	97
	Cifarilli	129	312	87.5	95.8
	Kwazar	156	176	71	87.7
Acetamprid (50 gm)	Micron ULVA	156	131	85.5	95.2
	Cifarilli	126	344	87	95.7
	Kwazar	156	183	74.5	89.8
Lufenuron (160 cm^3)	Micron ULVA	132	148	71	87.5
	Cifarilli	147	329	73.5	91
	Kwazar	156	176	73.5	89.1

VMD = Volume Mean Diameter. N / cm^2 = Number of droplets per square centimeter.
*Average of two seasons.

4.Relationship between lost spray on ground and the bioresidual activity of insecticides used:

Data in Tables (4 and 5) showed that there were a negative correlation between lost spray on ground equipment and the bioresidual activity of insecticides used.

4.1.Economy Micron ULVA spryer (15 L/fed.) :

Data in Tables (4 and 5) showed that the lost spray percentages were 6, 6.1 and 5.7 % from the total spray volume in the case of Imidaclopride, Acetamprid and Lufenuron and the general mean reduction % of two seasons (2017-2018) were 94.7 ,93.3 and 84.1 % *B.tabaci* nymphs at total recommended doses, respectively, in the case of the same insecticides and the general mean reduction % of two seasons of *A.gossypii* nymphs were 97 ,95.2 and 87.5 for the same insecticides, respectively.

4.2.Knapsack motor sprayer (Cifarilli) (20 L/fed.):

Data in Tables (4 and 5) showed that the lost spray percentages were 9.3, 9.2 and 9.4 % from the total spray volumes in the case of Imidaclopride, Acetamprid and Lufenuron and the general mean reduction % of two seasons (2017-2018) were 93.8 ,93.8 and 86.7 % *B.tabaci* nymphs at total recommended doses, respectively, in the case of the same insecticides and the general mean reduction % of two seasons of *A.gossypii* nymphs were 95.8 ,95.7 and 91 for the same insecticides,respectively.

Table (4): Lost spray on ground as produced by low volume ground spraying equipment against *Bemisia tabaci* nymphs during seasons (2017-2018).

Insecticide and dose rate / fed.	Tested sprayer and spray volume (L / fed.)	*N / cm^2 of total spray droplets	N / cm^2 droplets lost (on ground)	% N/cm^2 (ground) ——— x 100 N/Cm^2 (Plants+ground)	% Mortality	
					Intial mean *	Residual mean *
Imidaclopride (400 cm^3)	Micron ULVA (15)	166	10	6	84	94.7
	Cifarilli(20)	344	32	9.3	81.5	93.8
	Kwazar (94)	216	40	18.5	63	84.3
Acetamprid (50 gm)	Micron ULVA (15)	162	10	6.1	80	93.3
	Cifarilli(20)	379	35	9.2	81.5	93.8
	Kwazar (94)	216	35	16	66.5	87
Lufenuron (160 cm^3)	Micron ULVA (15)	157	9	5.7	63.5	84.1
	Cifarilli(20)	363	34	9.4	66.5	86.7
	Kwazar (94)	207	31	14.9	63.5	84.4

N / cm^2 = Number of droplets per square centimeter. * On Eggplant and lost spray on ground.
*Average of two seasons.

Table (5): Lost spray on ground as produced of low volume ground spraying equipment by using certain insecticides at total recomı Dar, 2019
***gossypii* nymphs during seasons (2017-2018).**

Insecticide and dose rate / fed.	Tested sprayer and spray volume (L / fed.)	*N / cm^2 of total spray droplets	N / cm^2 droplets (on ground)	% N/cm^2 (ground) ——— x 100 N/Cm^2 (Plants+ground)	% Mortality	
					Intial mean*	Residual mean*
Imidaclopride (400 cm^3)	Micron ULVA (15)	166	10	6	91	97
	Cifarilli(20)	344	32	9.3	87.5	95.8
	Kwazar (94)	216	40	18.5	71	87.7
Acetamprid (50 gm)	Micron ULVA (15)	162	10	6.1	85.5	95.2
	Cifarilli(20)	379	35	9.2	87	95.7
	Kwazar (94)	216	35	16	74.5	89.8
Lufenuron (160 cm^3)	Micron ULVA (15)	157	9	5.7	71	87.5
	Cifarilli(20)	363	34	9.4	73.5	91
	Kwazar (94)	207	31	14.9	73.5	89.1

4.3.Hand- Held compression sprayer (Kwazar) (94L/fed.):

Data in Tables (4 and 5) showed that the lost spray percentages were 18.5, 16 and 14.9 % from the total spray volumes in the case of Imidaclopride, Acetamprid and Lufenuron and the general mean reduction % of two seasons (2017-2018) were 84.3, 87 and 84.4% *B.tabaci* nymphs at total recommended doses, respectively, in the case of the same insecticides, and the general mean reduction % of two seasons of *A.gossypii* nymphs were 87.7, 89.8 and 89.1 for the same insecticides,respectively .

5.Relationship between the tested chemicals, techniques and the mortality percentages of *Bemisia tabaci* and *Aphis gossypii* infesting eggplant :

5.1.Bioassay evaluation:

To study the influence of various compounds and spraying equipment before and after application Hendresson and Tilton's formula (1955) was adopted to calculate the reduction percentages in the population. Tables (6,7,8 and 9) showed that, the percentages of reduction of *B.tabaci* and *A.gossypii* infesting eggplant affected by certain insecticides sprayed with certain ground application techniques during the seasons of (2017-2018) using total recommended dose rate. The performance rate of Knapsack motor sprayer (Cifarilli) was 12 fed./day. It was the best equipment, but the lowest performance rate was Hand-Held compression sprayer (Kwazar) since it could spray only 2.5 fed./day.

5.2.The following remarks and results were obtained:

5.2.1. There was no phytotoxic effect on eggplant leaves after treatments, no change in the leaves color, no leaf curling or flaming up phenomena was happened.

5.2.2. Insecticides treated plants revealed the lowest eggplant yield loss in comparison with untreated plots; their application reduced the incidence of whitefly and cotton aphid infestation on eggplant and decreased the percent loss of eggplant yield in all treatments and with all sprayers.

5.2.3. There was a significant differences between both the distribution percentages of droplets numbers/cm^2 (LSD=2.8255for Imidaclopride, 3.8257 for and Acetamprid 3.9958 for Lufenuron) , for droplet sizes (LSD=3.4605 for Imidaclopride, 2.5793 for Acetamprid and 2.8255 for Lufenuron) and for reduction percentages(LSD=1.8238 for Lufenuron, 1.9979 for Acetamprid and 1.2896 for Imidaclopride ,for white fly and(LSD=1.7302 for Lufenuron, 1.6313 for Acetamprid and 2.8255 for Imidaclopride ,for aphid.

Table (6): Reduction percentages in *B. tabaci* nymphs affected by certain insecticides sprayed with certain ground equipment during the season (2017) data were averages of five replicates.

Equipment / treatments	Counted nymphs before treatmen			% Reduction after spraying																							
				2 nd						5 th						7 th						General mean					
	Micron ULVA	Cifarilli	Kwazar	Micron ULVA (15 L/fed.)		Cifarilli (20 L/fed.)		Kwazar (94 L/fed.)		Micron ULVA (15 L/fed.)		Cifarilli (20 L/fed.)		Kwazar (94 L/fed.)		Micron ULVA (15 L/fed.)		Cifarilli (20 L/fed.)		Kwazar (94 L/fed.)		Micron ULVA (15 L/fed.)		Cifarilli (20 L/fed.)		Kwazar (94 L/fed.)	
				C	R %	C	R %	C	R %	C	R %	C	R %	C	R %	C	R %	C	R %	C	R %	C	R %	C	R %	C	R %
Imidaclopride (400 cm ^{3}/fed)	100	115	120	14	86	20	83	43	64	0	100	0	100	12	**90**	_	_	_	_	0	100	4.6	95.3	6.6	94.3	18.3	84.6
Acetampirid (50 gm/fed)	125	117	127	25	80	20	83	41	67	0	100	0	100	7	95	_	_	_	_	0	100	8.3	93.3	6.6	94.3	16	87.3
Lufenuron (160 cm^3/fed)	130	105	123	47	64	35	67	41	67	13	90	6	95	10	92	0	100	0	100	0	100	20	84.6	13.6	87.3	17	86.3
Untreated (control)	117	122	127	117	_	122	_	127	_	116	_	120	_	126	_	116	_	120	_	126	_	116.7	_	121.4	_	126.7	_

C = Count of life nymphs after treatment.

R = % Reduction of nymphs.

Table (7): Reduction percentages in *B. tabaci* nymphs affected by certain insecticides sprayed with certain ground equipment during the season (2018) data were averages of five replicates.

Equipment / treatments	Counted nymphs before treatmen			% Reduction after spraying																							
				2 nd						5 th						7 th						General mean					
	Micron ULVA	Cifarilli	Kwazar	Micron ULVA (15 L/fed.)		Cifarilli (20 L/fed.)		Kwazar (94 L/fed.)		Micron ULVA (15 L/fed.)		Cifarilli (20 L/fed.)		Kwazar (94 L/fed.)		Micron ULVA (15 L/fed.)		Cifarilli (20 L/fed.)		Kwazar (94 L/fed.)		Micron ULVA (15 L/fed.)		Cifarilli (20 L/fed.)		Kwazar (94 L/fed.)	
				C	R %	C	R %	C	R %	C	R %	C	R %	C	R %	C	R %	C	R %	C	R %	C	R %	C	R %	C	R %
Imidaclopride (400 cm ^{3}/fed)	127	122	115	23	82	25	80	44	62	0	100	0	100	12	90	_	_	_	_	0	100	7.6	94	8.3	93.3	18.6	84
Acetampirid (50 gm/fed)	118	126	130	24	80	26	80	45	66	0	100	0	100	7	95	_	_	_	_	0	100	8	93.3	8.6	93.3	17.6	86.6
Lufenuron (160 cm^3/fed)	120	118	127	45	63	41	63	51	60	15	88	10	92	10	92	0	100	0	100	0	100	20	83.6	17	86	25.3	82.6
Untreated (control)	119	120	125	119	_	120	_	125	_	118	_	118	_	124	_	118	_	118	_	124	_	118	_	118	_	124	_

Table (8):Reduction Percentages in *A.gossypii* nymphs affected by certain insecticides sprayed with certain ground equipment during the season (2017), data were averages of five replicates.

Equipment / treatments	Counted nymphs befor treatment			% Reduction after spraying																					
				2 nd						5 th						7 th						General mean					
	Micron ULVA	Cifarilli	Kwazar	Micron ULVA (15 L/fed.)		Cifarilli (20 L/fed.)		Kwazar (94 L/fed.)		Micron ULVA (15 L/fed.)		Cifarilli (20 L/fed.)		Kwazar (94 L/fed.)		Micron ULVA (15 L/fed.)		Cifarilli (20 L/fed.)		Kwazar (94 L/fed.)		Micron ULVA (15 L/fed.)		Cifarilli (20 L/fed.)		Kwazar (94 L/fed.)	
				C	R %	C	R %	C	R %	C	R %	C	R %	C	R %	C	R %	C	R %	C	R %	C	R %	C	R %	C	R %
Imidaclopride (400 cm ^{3}/fed)	55	60	70	6	90	8	87	21	70	0	100	0	100	6	92	_	_	_	_	0	100	2	96.6	2.6	95.6	9	87.3
Acetampirid (50 gm/fed)	67	75	64	10	85	11	86	17	74	0	100	0	100	4	95	_	_	_	_	0	100	33	95	3.6	95.3	7	89.6
Lufenuron (160 cm^3/fed)	59	69	72	18	70	17	75	19	73	6	90	4	95	5	93	0	100	0	100	0	100	8	86.6	7	90	8	88.6
Untreated (control)	65	71	58	65	_	71	_	58	_	64	_	57	_	62	_	62	_	68	_	55	_	63	_	69	_	56	_

C = Count of life nymphs after treatment.

R = % Reduction of nymphs.

Table (9):Rduction Percentages in *A.gossypii* nymphs affected by certain insecticides sprayed with certain ground equipment during the season (2018),data were averages of five replicates.

Equipment / treatments	Counted nymphs treatment			% Reduction after spraying																					
				2 nd						5 th						7 th						General mean					
	Micron ULVA	Cifarilli	Kwazar	Micron ULVA (15 L/fed.)		Cifarilli (20 L/fed.)		Kwazar (94 L/fed.)		Micron ULVA (15 L/fed.)		Cifarilli (20 L/fed.)		Kwazar (94 L/fed.)		Micron ULVA (15 L/fed.)		Cifarilli (20 L/fed.)		Kwazar (94 L/fed.)		Micron ULVA (15 L/fed.)		Cifarilli (20 L/fed.)		Kwazar (94 L/fed.)	
				C	R %	C	R %	C	R %	C	R %	C	R %	C	R %	C	R %	C	R %	C	R %	C	R %	C	R %	C	R %
Imidaclopride (400 cm ^{3}/fed)	72	65	69	6	92	8	88	19	72	0	100	0	100	6	92	_	_	_	_	0	100	2	97.3	26	96	8.3	88
Acetampirid (50 gm/fed	70	74	68	10	86	9	88	17	75	0	100	0	100	3	96	_	_	_	_	0	100	3.3	95.3	3	96	6.6	90
Lufenuron (160 cm^3/fed)	65	59	74	18	72	13	78	19	74	5	93	2	98	4	95	0	100	0	100	0	100	7.6	88.3	5	92	7.6	89.6
Untreated (control)	68	75	65	68	_	75	_	65	_	67	_	73	_	64	_	66	_	72	_	64	_	67.6	_	74	_	64.7	_

Field experiment was carried out on infested area with *B.tabaci* and *A.gossypii* adults at early season on eggplant. For evaluation the field performance of Low-Volume spraying machines; Economy Micron ULVA sprayer (15 L/fed.), Knapsack motor sprayer (Cifarilli) (20L/ fed.) and Hand-Held compression sprayer (Kwazar) (94 L/fed.); to spray Imidaclopride, Acetamprid and Lufenuron with full recommended dose . A satisfactory coverage was obtained on eggplant, the droplets spectrum was obtained in field experiment was agreed with the optimum droplet sizes which mentioned by Himel (1969). The best obtained result was 20 L/fed. as spray volume, 146 μm and 123 droplets/cm^2 , these results agreed with (Himel and Moore, 1969) in the optimum droplet size to control cotton leafworm in cotton fields by ground equipment. Acetamiprid revealed the best bioefficiacy results with the three tested sprayers. Also , Imidaclopride for whitefly and cotton aphid revealed the best bioefficiacy results with Economy Micron Ulva sprayer (15 L/fed.). Acetamiprid revealed higher mortality than Imidaclopride with Kwazar sprayer (94 L/fed) and these results agreed with Hindy *et al.* (2004) and Genidy *et al.* (2005) which recommended KZ oil and Pyriproxyfen followed by Agerin by using low volume spraying because of reducing the time lost in process filling the machines, improve the homogeneity of the spray solution on the plant leaves and saving the lost spray on the ground,these results also in agreement with Bakr *et al*. (2014) recommendation by using Profenofos followed by Pyriproxyfen and Spinosad with Agromondo motorized knapsack sprayer (20L/fed.) and Morsy *et al.*(2015) whom recommended using Carbosolvan ,Acetamprid and Deltamethrin with low volume machines not less than (15 L/fed.), also Dar (2016) recommendation whenever using Lufenuron followed by Spinosad in controlling cotton leafworm on Clover with low volume machines .

Finnally, the data showed that, low application technique revealed by Knapsack motor sprayer (Cifarilli) (20L/ fed.) and Economy Micron ULVA sprayer (15 L/fed.) were best equipment to control whitefly and cotton aphid infesting eggplant . Also , the lowest spray volume and the lowest percentages of lost spraying between plants, these results were agreed with Hindy *et al.* (1997) , who mentioned that, there was a positive correlation relationship between rate of application and lost spray on ground. There was a negative complete correlation between (VMD) and the mean residual mortality of *B. tabaci* and *A.gossypii* while there was a positive complete correlate between N/cm² and the mean residual of mortality of *B.tabaci* and *A.gossypii* in all treatments.

It could be concluded that , using Imidaclopride and Acetamprid followed by Lufenuron with low volume (LV) ground spraying equipment with not less than (15L./fed.) by using recommended doses which revealed successful management against piercing and sucking insects infesting eggplant under our local conditions.

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Insecticidal activity of citrus peel oil of navel orange against the striped mealybug *Ferrisia virgata* (Hemiptera: Pseudococcidae) and the mango shield scale *Milviscutulus mangiferae* (Hemiptera: Coccidae)

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Abstract:

Essential oil extracted from peels of citrus fruit namely navel orange (*Citrus sinensis* L.), belonging to family Rutaceae was tested for its insecticidal activity at four different concentrations (500, 1000, 2500, and 5000ppm) against nymphs and adults of the striped mealybug *Ferrisia virgata* Cockerell (Hemiptera, Pseudococcidae) and mango shield scale or mango soft scale *Milviscutulus mangiferae* (Green) (Hemiptera: Coccidae). Formulated oils of navel orange was bioassayed against mealybug *F. virgata* and *M. mangiferae*. The results revealed that, the two formulated oils of navel orange achieved high toxicity against nymphs and adults of *F. virgata* and *M. mangiferae* . The essential oil of navel orange was isolated by hydrodistillation and the analysis of essential oil by GC/MS revealed the presence of 35 peaks, approximately all peaks were identified. The chemical composition showed that limonene was the main constituent in citrus oil (78.15%). The results of the present study suggested that, formulated navel orange oil used as safe, potential natural products for control of *F. virgata* and *M. mangiferae* infesting mango and guava trees and may be used as alternatives to the reference products after application of these results in the semifield and field experiments.

Introduction

Fruits trees are liable to be infested with many serious pests during their growth stages, including striped mealybug *Ferrisia virgata* Cockerell (Hemiptera: Pseudococcidae) infested guava trees and *Milviscutulus mangiferae* (Green) (Hemiptera: Coccidae) infested mango trees. Mango trees (*Mangifera indica* L.) are considered of the most popular and economic fruit trees in Egypt, it occupy the third rank from the commercial point of view (Attia, 2010). Mango is tropical/sub tropical fruit with highly significant economic importance (Sivakumar and Yahia, 2011). Guava trees (*Psidium guajava*) has major commercial importance in India, Egypt, South Africa, Barazil, Colombia and the Caribbean region. The fruits are eaten

fresh or as preserves and processed for use in dairy and baked products (Richard, 2005).

The striped mealybug *F. virgata* is one of the most important pest attacking many different host plant, belongs to several plant families, it infests mulberry, fig, guava , pear, apple, grape and olive in Egypt (EL-Minshawy *et al*., 1972). The mealybug *F. virgata* can be found throughout the world, is known to feed on more than 100 plant species grown throughout the world. This mealybug mainly attacks the foliage, sucks a great amount of plant sap for its protein requirement and secretes honey dew. Most mealybug individuals are accumulated around the branches, foliage, leaves, twigs and at the base of fruits. Many of these species are covered with white wax and have a distinct fringe of waxy filaments around the circumference of their bodies and the long tails and the presence of two stripes on the body. This species produces an egg mass or ovisac (Ghose and Ghosh, 1990).

The mango shield scale or mango soft scale, Milviscutulus mangiferae (Green) (Hemiptera: Coccidae), a serious pest of mango trees in various parts of the world and is reported on *M. indica* in Egypt , represents the first record of this species in the country (Abd-Rabou and Evans, 2018). *M. mangiferae* an invasive coccid infested mango orchard profusely in Qaliobiya Governorate caused yellowing, defoliation, reduction in fruit set and loss in plant vigor, the insect excrete large amounts of honeydew which encourage the growth of sooty mould and the infested parts acquire the dirty black appearance that affect on photosynthesis (Attia *et al.,* 2018).

In general, control of the mealybugs and soft scale insects around the world relies heavily on use of insecticides and mineral oils. However, continuous and heavy use of these synthetic pesticides has created serious problems such as environmental pollution, toxicity to non-target organisms (parasitoids and predators), pest resistance and pesticide residues (Mohan and Fields, 2002). Therefore, there is an urgent need to develop new, convenient and safer alternatives to synthetic pesticides. Essential oils and their major constituents, attracted research attention in recent years as potential alternatives to synthetic insecticides (Aslan *et al*., 2004).

The genus *Citrus* includes several important fruits such as oranges, mandarins, lime, lemons and grapefruits. The essential oils of some citrus species have been reported to have insecticidal properties against insect pests (Elhag, 2000). The major active component of citrus oil is limonene and using 1% limonene mixture was safe for most plants and provided good control of mealybugs and scale insects (Hollingsworth, 2005).

The aim of this research work is to study the efficacy of Egyptian citrus peel essential oil of navel orange (*Citrus sinensis*) against nymphs and adults of mealybug *F. virgata* and soft scale insect *M. mangiferae* on mango and guava trees, respectively. Also, extraction and chemical analysis of essential oil was studied.

Materials and methods

1. Tested citrus species:

The experimental citrus species, navel orange (*C. sinensis*), belonging to family Rutaceae was selected for this study. This citrus species was obtained from a private citrus orchard.

2. Insects source:

Insects culture of *F. virgata* and *M. mangiferae* for laboratory experiments were obtained from a private orchards at El Mansouria region in Giza Governotare . Samples were collected randomly from each of the four cardinal directions (East, West, North and South). Leaves were packed in paper bags and

transferred to the laboratory and they were maintained at laboratory temperature about 25±1°C and 65±1% relative humidity. In the laboratory, *F. virgata* and *M. mangiferae* were identified by Department of Mealybug and Scale Insects, Plant Protection Researches Institute, Agriculture Researches Centre.

3. Extraction of citrus oil:

Citrus oil was extracted by Cavalcanti *et al.* (2004). The essential oil was extracted from the fresh peels (200g weight 400 ml of distilled water) by hydrodistillation using a modified Clevenger type apparatus for 4 h. The distilled was extracted with diethyl ether after saturation with Sodium Chloride. The extracted oil was dried over anhydrous Sodium Sulfate, then packed in dark container and stored at 4°C until used for GC-MS analysis and bioassays.

4. Chemical analysis of essential oil:

4.1. Chemical analysis of citrus peel oil constituents:

The extracted citrus oil was subjected to GC/MS analysis using Shimadzu GC/MS–QP-2010 Plus (Japan). Column: DB5 MS (30 m length, 0.25 mm thickness, 0.25mdiameter, 1.5µm film). Carrier gas: Helium (flow rate 1.2 ml/Min.). Ionization mode: (70ev). The injection volume was 0.5µl (split ratio of 1:100), temperature program: 50^{o}C (static for 2 Min) with gradually increasing (a rate of 4^{o}C/ Min) up to 200^{o}C then (10^{o}C/ Min) to 280^{0}C. The detector temperature was 290^{0}C, while, the injector temperature was 250^{0}C.

4.2. Identification of the chemical constituents:

Qualitative identification of the essential oil was achieved by library searched data base Willey 229 LIB as well as by comparing their retention indices and mass fragmentation patterns with those of the available references and with published data, (Adams, 2007). The percentage composition of components of the volatile was deterMined by computerized peak area measurements.

5. Preparation of formulated orange essential oil:

Four concentrations (500, 1000, 2500 and 5000ppm) of formulated citrus oil of navel orange were prepared by two emulsifiers, Triton-100 (TE) and local emulsifier (LE).

6. Toxicity bioassays:

Laboratory bioassays were conducted to determine the bioactivity of formulated citrus oil of navel orange against nymphs and adults of *F. virgata* and *M. mangiferae*.

The toxicity bioassay was conducted to evaluate toxicity of formulated citrus oil of navel orange to nymphs and adults of mealybug *F. virgata* and soft scale insect *M. mangiferae* at four different concentrations (500, 1000, 2500 and 5000ppm). In spray toxicity assay, guava and mango trees leaves containing nymphs and adults of mealybug *F. virgata* and mango shield scale *M. mangiferae* , respectively were placed into plastic petri dishes (10cm dia × 2cm ht). Ten infested leaves were sprayed with 1ml for five seconds of the two formulated citrus oils of navel orange (Triton-x100 and local emulsifier), then, kept at room temperature. Control insects were sprayed with Triton-x100 and local emulsifiers alone (without oil). Five replicates were used and the experiment was repeated for three times and mortality was recorded after 24, 48 and 72 hrs.

7. Statistical analysis:

The percentage of the mortality was recorded and the mortality was corrected with Abbot formula (Abbott, 1925). LdP-line program was used to deterMinate LC_{50} values. Data of all experiments were evaluated statistically using ANOVA and means compared using Duncan's Multiple Range Test (Duncan, 1955) at $P<0.05$). All statistical

analyses were done using the software package Costat.

Results and discussion

1. Toxicity bioassay:

The obtained two formulated citrus oil of navel orange species in this study were mainly conducted to investigate a relationship between the oil constituents and their potency towards nymphs and adults of *F. virgata* and *M. mangiferae*.

1.1. Toxicity of formulated essential oil of navel orange against *Ferrisia virgata*:

The results of toxicity assays (spray toxicity) as represented in Tables (1 and 2), showed that essential oil of citrus peel exhibited toxicity rate with concentration and time dependent. Formulated peel essential oil of (TE) achieved higher mortality percentages against nymphs and adults than formulated peel essential oil of (LE) at the different concentrations (500, 1000, 2500, 5000 ppm). The highest toxicity rates against nymphs and adults was recorded with formulated peel orange oil of (TE) were 98.00±4.00 and 95.70±4.00 %, respectively, at the maximum concentration (5000ppm) and the last day (72hr) of assay. The percentages of mortality achieved by formulated peel essential oil of (LE) were 92.30±4.00% and 87.50±4.95 % respectively, at the same concentration and time.

1.2.Toxicity of formulated essential oil of navel orange against *Milviscutulus mangiferae*:

The results of toxicity assays as represented in Tables (3 and 4), showed that essential oil of citrus peels exhibited toxicity rate with concentration and time dependent. Formulated peel essential oil of (TE) achieved higher mortality percentages against nymphs and adults than formulated peel essential oil of (LE) at the different concentrations (500, 1000, 2500 and 5000 ppm). The highest toxicity rates of formulated navel orange essential oil of (TE) against nymphs and adults were 81.69±2.48 and 83.47±2.48%, respectively, at the maximum concentration 5000ppm and the last day 72hr. While, the percentages of mortality of navel orange essential oil of (LE) were 81.43±7.08% and 73.33±2.45%, respectively, at the same concentration and time. The lowest mortality against nymphs and adults of the two insects was obtained with the lowest concentration (500 ppm) and at the first day of assay (24hr). Generally, the two formulated citrus oils were toxic to nymphs and adults of *F. virgata* and *M. mangiferae* at all concentrations. There were significant differences in mortality between the tested concentrations after 24hr, but, non-significant differences in mortality after 48 and 72hrs were observed. Also, there were significant differences in mortality between control and treated variants ($P< 0.05$).

Insecticidal effect of citrus species essential oils against mealybugs and soft scale insects were studied by many workers, Pumnuan *et al*. (2015) showed that, fresh peels essential oils of four citrus species recorded moderate toxicity at 2 ml/L air (fumigation) and high toxicity at 2 ml/L air against larvae of mealybug *Pseudococcus jackbeardsleyi* Gimpel and Miller (Hemiptera: Pseudococcidae) at 24hr. These findings are confirmed by Karamaouna *et al*. (2013), who showed that the citrus peel essential oils of lemon (*Citrus limon*) and navel orange (*C. sinensis*) were the most toxic of all the tested essential oils against 3rd instar nymphs and female adults of the vine mealybug *Planococcus ficus* (Signoret) (Hemiptera: Pseudococcidae). Also, El-Badawy (2015) found that, all tested citrus oils specially navel and baladi oranges achieved high insecticidal and repellent activities against mealybug *Icerya seychellarum* (Westwood) (Hemiptera:Monophlebidae).

2.Effect of formulation on potency of navel orange essential oil:

From the data of LC_{50} values presented in Tables (1, 2, 3 and 4), it could be demonstrated that, the formulated peel citrus oil of (TE) was more potent nymphicidal and adultscidal effect than the formulated peel essential citrus oil of (LE) against *F. virgata* and *M. mangiferae* after 24, 48 and 72hr of assay. From the data presented in Tables (1 and 2), it could be demonstrated that, the LC_{50} values of the formulated orange oil of (TE) against *F. virgata* ranged from 22.44 (Last day 72hr) to 778.70ppm (1^{st}day 24 hr) for nymphs and from 36.79 to 807.66ppm for adults after the same times, while, the formulated orange oil of (LE) recoded LC_{50} values ranged from 44.56 ppm (Last day 72hr) to 934.61 ppm (1^{st}day 24 hr) for nymphs and from 81.27 to 1146.49ppm for adults after the same time.

The data presented in Table (3 and 4), reported that, the LC_{50} values of the formulated citrus oil of (TE) against *M. mangiferae* ranged from 155 (Last day 72hr) to 790.20 ppm (1^{st} day 24 hr) for nymphs and from 193.29to 815.23ppm for adults, while, the formulated citrus oil of (LE) recoded LC_{50} values ranged from 230.74 (Last day 72hr) to 1044.73ppm (1^{st}day 24 hr) for nymphs and from 474.83 to 8105.53ppm for adults after the same time. The variation of the LC_{50} values of citrus oil against *F. virgata* and *M. mangiferae* depending on the toxicity of the formulation of citrus oil, the mealybug and scale insect life stage. LC_{50} values of each formulated citrus oil reveal significant differences between nymphs and adults.

These findings are confirmed by Karamaouna *et al*. (2013) who showed that, the LC_{50} values of citrus (*C. sinensis* and *C. limon*) oils ranged from 2.7 to 8.1mg/ml depending on the essential oil and the mealybug life stage. These LC_{50} values were significantly lower than the LC_{50} of the reference paraffin oil in the respective *P. ficus* life stages. Results of El-Badawy (2015) revealed that the oil of navel orange achieved the highest toxicity against nymphs and adults of mealybug *I. seychellarum* with LC_{50} values of (406.97 and 370.04 ppm), respectively.

3. Chemical analysis of citrus peel essential oil:

The essential oil yield of fresh citrus peels of *C. sinensis* was 4.30%. The chemical composition of the essential oil of citrus peels are presented in Table (5). The essential oil analysis by GC/MS revealed that, the presence of 35 peaks, all peaks were identified, representing 99.46 % of the essential oil of navel orange. The major constituents of this essential oil mainly belonged to two groups: Monoterpenes and oxygenated monoterpenes hydrocarbons, while the minor constituents belonged to: sesquiterpene and oxygenated sesquiterpene hydrocarbons. Oxygenated monoterpenes with contribution of 3.08% constituted the second major portion of the essential oil after monoterpenes (86.41%) from peel oil. Sesquiterpene hydrocarbon was present at very low levels in the oil of navel orange.

The chemical analysis of the citrus oil showed limonene as the main constituent (78.15%) for navel orange. The monoterpene hydrocarbons α-Phellandrene, β- Phellandrene, *α*-pinene, β-pinene, β-Myrcene, 3-Carene β-Ocimene and γ-Terpinene are present in studied citrus oil. The qualitative and quantitative composition of the essential oil of fresh citrus peels showed that, the most abundant ingredients beside to limonene, were β-myrcene (4.30%), linalool (1.59%) and *α*-pinene (1.55%) in the citrus peels oil of navel orange. Among other than monoterpenes, Bis (2-ethylhexyl) phthalate (7.60%) was

present in oil. Our results of the chemical composition of citrus peel oil are in agreement with many other studies (Mansour *et al*., 2004; Ahmad *et al*., 2006; Asekun *et al*., 2007 and El-Badawy ,2015). All these studies showed that, limonene was the main component with high variation in all citrus peel oils and also, there are considerable variations in the other constituents of the chemical composition of citrus oils. Such variation in chemical composition (Limonene content and other constituents) in citrus peel oils may be related to the time of harvesting, the degree of freshness, genetic makeup and the size of the fruit. Also, geographical location, fruit variety and method of extraction (Ahmad *et al*., 2006).

Regarding to potency of citrus oil against nymphs and adults of *F. virgata* and *M. mangiferae* the data presented in Tables (1-4) indicated that the potency of the tested formulated oils was related to the major component limonene content of that oils. These results are confirmed by El-Badawy (2014 and 2015), who showed that the toxic effect of five citrus oils on *I. seychellarum* could be related to the high content of limonene. Also, these results are in agreement with these obtained by Ibrahim *et al*. (2001) who stated that the monoterpene limonene showed deterrent and insecticide properties, which might used in pest control in organic agriculture. The best limonene mixture (1% limonene, 0.75% emulsifier APSA-80 and 0.1% surfactant Silwet) controlled from 69 to 100% of mealybugs and scale insects, depending on the species, insect stage and application method (Hollingsworth, 2005). Also Ware (2000) showed that, orange oil contains the monoterpenoid d-limonene, and the mode of action of d-limonene is similar to that of pyrethrum, affecting sodium flux in the peripheral neurons.

Formulated citrus oil of navel orange achieved high insecticidal activity against striped mealybug *F. virgata* and mango shield scale *M. mangiferae*, so, it can be used as an effective natural alternative to mineral oils and insecticidal soap. Overall results indicated that the toxic effects of citrus oil on *F. virgata* and *M. mangiferae* could be related to the high content of limonene. There are synergistic or antagonistic effects between limonene and the other minor constituents in the citrus oils.

It is recommended to expand such laboratory experiments to semifield and field conditions and determine the efficacy of orange citrus essential oil against *F. virgata* , *M. mangiferae* and other scale insects

Table (1): Toxic effect of formulated essential oil (Triton- x100 emulsifier) of navel orange peels against *Ferrisia virgata* nymphs and adults at different concentrations.

Corrected mortality(%)±SE						
Conc. (ppm)	**Nymph**			**Adult**		
	24hr	**48 hr**	**72 hr**	**24 hr**	**48 hr**	**72 hr**
500	36.70±2.45^{c}	79.00±3.43^{a}	83.67±4.00^{a}	38.70±5.09^{c}	72.69±1.37^{a}	80.00±8.90^{a}
1000	56.67±5.09^{b}	86.11±4.75^{a}	88.00±4.90^{a}	58.67±5.09^{b}	80.00±4.33^{a}	88.00±4.90^{a}
2500	83.33±2.45^{a}	90.00±2.45^{a}	92.00±4.90^{a}	66.67±5.09^{ab}	85.70±4.87^{a}	92.00±4.90^{a}
5000	86.67±2.45^{a}	95.00±2.45^{a}	98.00±4.00^{a}	76.70±2.45^{a}	88.89±2.45^{a}	95.70±4.00^{a}
Control	0.00	0.00	00.00	0.00	0.00	00.00
LC_{50}	778.70	35.87	22.44	807.66	46.23	36.79
F value	20.10***	2.78ns	1.27ns	19.10**	2.48ns	1.30ns
$LSD_{0.05}$	17.19	17.19	13.41	16.20	27.91	17.99

Table (2): Toxic effect of formulated essential oil (Local emulsifier) of navel orange peels against *Ferrisia virgata* nymphs and adults at different concentrations.

Corrected mortality(%)±SE						
Conc.	**Nymph**			**Adult**		
(ppm)	**24hr**	**48 hr**	**72 hr**	**24 hr**	**48 hr**	**72 hr**
500	53.33±5.09^{b}	57.14±5.39^{b}	70.00±6.32^{b}	36.70±3.00^{c}	63.16±8.43^{a}	70.83±8.00^{a}
1000	60.0±2.74^{b}	83.33±4.43^{ab}	84.62±9.80^{ab}	60.00±3.19^{b}	66.70±2.91^{a}	76.00±6.86^{a}
2500	76.60±2.13^{b}	85.71±9.24^{a}	91.60±8.00^{ab}	66.70±2.45^{ab}	77.70±6.53^{a}	80.00±6.32^{a}
5000	83.33±2.13^{a}	95.00±2.77^{a}	92.30±4.00^{a}	76.70±4.00^{a}	83.30±7.13^{a}	87.50±4.95^{a}
Control	0.00	0.00	0.00	0.00	0.00	0.00
LC_{50}	934.61	335.52	44.56	1146.49	218.67	81.27
F value	6.93*	4.01 ns	2.42ns	11.6**	1.11 ns	1.02ns
$LSD_{0.05}$	16.31	27.43	22.03	16.31	36.21	19.84

Table (3): Toxic effect of formulated essential oil (Triton- x100 emulsifier) of navel orange peels against *Milviscutulus mangiferae* nymphs and adults at different concentrations.

Corrected mortality(%)±SE						
Conc.	**Nymph**			**Adult**		
(ppm)	**24hr**	**48 hr**	**72 hr**	**24 hr**	**48 hr**	**72 hr**
500	54.17±6.00^{b}	58.58±0.90^{c}	69.64±8.70^{a}	41.76±7.12^{c}	58.94±7.50^{b}	60.66±6.12^{b}
1000	63.06±2.54^{b}	67.02±3.39^{b}	71.43±1.66^{a}	70.22±2.65^{b}	64.62±3.21^{b}	73.33±0.96^{ab}
2500	73.75±1.92^{a}	76.97±3.40^{a}	76.67±2.63^{a}	75.66±3.31^{a}	75.61±1.70^{a}	78.60±1.92^{a}
5000	74.40±1.92^{a}	80.91±0.60^{a}	81.69±2.48^{a}	76.35±4.17^{a}	76.60±2.87^{a}	83.47±2.48^{a}
Control	0.00	0.00	0.00	0.00	0.00	0.00
LC_{50}	790.20	317.20	155.00	815.23	502.87	193.29
F value	7.98	16.8**	1.46	10.60**	6.89	2.62
$LSD_{0.05}$	10.59	7.44	14.32	13.86	13.22	10.42

Table (4): Toxic effect of formulated essential oil (Local emulsifier) of navel orange peels against *Milviscutulus mangiferae* nymphs and adults at different concentrations.

Corrected mortality(%)±SE						
Conc.	**Nymph**			**Adult**		
(ppm)	**24hr**	**48 hr**	**72 hr**	**24 hr**	**48 hr**	**72 hr**
500	42.33±9.37^{b}	42.78±7.32^{b}	61.54±8.16^{b}	38.78±7.12^{a}	32.26±3.68^{a}	53.45±4.60^{b}
1000	44.79±4.61^{b}	51.83±7.42^{ab}	69.51±7.41^{ab}	41.48±5.25^{a}	48.05±9.99^{a}	63.75±10.20^{ab}
2500	64.68±8.42^{a}	67.22±6.90^{a}	72.34±4.77^{ab}	43.76±5.39^{a}	51.14±3.21^{ab}	71.22±1.92^{a}
5000	71.38±3.08^{a}	73.32±4.33^{a}	81.43±7.08^{a}	50.00±9.09^{a}	53.17±2.76^{a}	73.33±2.45^{a}
Control	0.00	0.00	0.00	0.00	0.00	0.00
LC_{50}	1044.73	1005.63	230.74	8105.53	3671.55	474.83
F value	5.25**	2.52ns	1.74ns	0.173ns	3.11ns	3.45ns
$LSD_{0.05}$	19.82	20.64	20.911	20.66	17.17	18.235

Table(5):Chemical composition of essential oil from peels of Navel orange citrus species.

No.	Components	RT(Min.)	Ratio (%)
1	α-Phellandrene	6.901	0.03
2	α-Pinene	7.125	1.55
3	β-Phellandrene	8.344	0.63
4	β-Pinene	8.45	0.63
5	β-Myrcene	8.85	4.3
6	Octanal	9.223	0.53
7	3-Carene	9.443	0.72
8	Benzyl alcohol, p,.alpha.-dimethyl-	9.632	0.03
9	D-Limonene	10.115	78.15
10	β-Ocimene	10.241	0.08
11	γ-Terpinene	10.846	0.16
12	1-Octanol	11.163	0.07
13	β-Terpinolene	11.604	0.02
14	α-Terpinolene	11.665	0.14
15	β-Linalool	11.952	1.59
16	Nonanal	12.066	0.06
17	Limonene oxide	12.999	0.01
18	β-Citronellal	13.333	0.18
19	α-Terpineol	14.005	0.43
20	Decanal	14.596	0.56
21	n-Octyl acetate	14.708	0.06
22	Carveol	14.993	0.04
23	β-Citronellol	15.14	0.18
24	β-cis-Citral	15.477	0.27
25	1-Carvone	15.602	0.03
26	Geranial	15.739	0.35
27	Lauraldehyde	18.979	0.08
28	Caryophyllene	19.417	0.12
29	α-Farnesene	19.61	0.20
30	β-Farnesene	19.924	0.01
31	Germacrene	20.613	0.04
32	Eremophila-1or Eremophila-1(10),8,11-triene	20.903	0.42
33	Cyclohexene,1-methyl-4(5-methyl-1-methylene-4-hexenyl	20.994	0.13
34	δ- Cadinene	21.343	0.06
35	Bis (2-ethylhexyl) phthalate	39.438	7.60
	Monoterpene Hydrocarbons		86.41
	Oxygenated Monoterpene Hydrocarbons		3.08
	Sesquiterpene Hydrocarbons		0.85
	Aldehydes		1.23
	Others		7.89
	Total		99.46

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Effect of the aphid rose *Macrosiphum rosae* (Hemiptera : Aphididae) infesting rose on physiological and natural characteristics of rose oil as well as population dynamics of this pest

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Abstract:

The aphid rose *Macrosiphum rosae* L. (Hemiptera : Aphididae) is stunts the growth of rose and reducing its market value. The aim of this reasrch work is to study the population fluctuations of the aphid *M. rosae* during successive season 2018 on three varieties (colors) of rose and the effect of insect infestation by *M. rosae* on the physiological and natural characteristics of rose oil . The results indicated that the activity period for adults and nymphs of aphid rose infested the varities Carmen, Golden gate and Dream were 23.5 adults/flower and 45.5 nymphs/flower; 25.6 adults/flower and 50.7 nymphs/flower and 20.5 adults/flower and 40.5 nymphs/flower on the mid of April, respectively. Also, this study was carried out to study effect of infested rose plants by *M. rosae* (in different stages of infestation) on the physiological and natural characteristics of rose oil at El-Orman Garden (Giza Governorate) under glasshouse conditions during successive seasons 2018. Data obtained showed that the most important components of rose oil such as (geraniol, citronellol, nerol, stearpoten, phenyl ethanol and bioflavonoids), acids such as Citric acid and Malik acid and vitamins A, B, C and D were changed its concentrations as result of infestation by *M. rosae*. High infestation by *M. rosae* affected on concentrations of these components more than medium and low infestation compared to control. Also, data obtained showed that the most natural characteristics of rose oil such as volatility, light rotation and refraction value were changed as result of infestation by *M. rosae*, but other natural characteristics like freezing point did not change after infestation by *M. rosae.*

Introduction

Rose (*Rosa gallica*) is one of the most important ornamental plants in Egypt and around all over the world which cultivated both in the open field

and under greenhouse conditions. So, it named king of flowers. It's found from oldest countries and it is the favorite flower for human in the world wide. Although developing live and highly technology but love human to roses still and increase. The human love for roses due to their beautiful colors, style of flowers, smiles and tolerant the inferable weather factors. Recently rose cultivated area increased gradually during the last years, especially in the new reclaimed areas for purposes local consumption and exportation to the foreign markets. So, rose became one of the important components for increase income for many countries all over the world, which producing and exporting these roses to different countries (Baydar ,2014 and Emam, 2009).

Rose plants infested with large scale of insects belong to many orders and families. *Macrosiphum rosae* L. (Hemiptera : Aphididae) commonly known as (Rose aphid) and is consider one of the most important insects of rose plants which infested both its leaves and flowers and also infested rose plants both in open fields and under green houses, Jaskiewicz (1997) who reported that the strong infestation by the rose aphid, *M. rosae* resulted in high deformation of stems, leaves and flowers of rose plants. Derek (2017) in Australia reported that *M. rosae* is a serious pest on rose and it is reproducing, parthenogenticcally and viviparous all year round. It feeds mainly on the young leaves and developing flower-buds of roses. The adults and nymphs of aphid infest the rose plants and suck cell sap from flowers, tender shoots and buds, ultimately decreasing the market value of rose flowers, infestation with aphid causes badly affects the flowering capacity of plants about 20-40% losses. Labanowski (1989) in Poland reported that the rose aphid, *M. rosae* is the most important insect infests rose plants. Many authors dealt with the population dynamics of the rose aphid, *M. rosae* i.e. Tomiuk and Wohrmann,1980 ; Rhomberg *et al*., 1985; Ghosh *et al*., 1994; Dixon, 1998 and Jaśkiewicz, 2004.

The temperature does not only have a direct influence on the rose aphid but it also changes the physiology of the rose plant, which results in the stagnation of the plant. It would not be a suitable food source for the rose aphid. Therefore, aphids have to migrate to their secondary host plants to avoid these unsuitable conditions (Maelzer,1977 and Jaskiewicz ,1997).

This study was carried out to study effect of infested rose plants by *M. rosae* (in different stages of infestation) on the physiological and natural characteristics of rose oil which found in rose flowers at El-Orman Garden, (Giza Governorate) under glasshouse conditions during successive seasons 2018. Therefore this study divided into two parts, first part study the population fluctuations of *M. rosae* during successive season 2018 on three varieties (colors) of rose and the second part included effect of insect infestation by *M. rosae* (in different stages of infestation) on the physiological and natural characteristics of rose oil .

Materials and methods

1.Experimental design:

This study was conducted on three varieties (Carmen, Golden gate and Dream) of rose plants grown in El-Orman Garden, Giza Governorate under glasshouse conditions during successive seasons 2018. Two glasshouses with an area of 27x45 m of each one, was divided into 9 plots (3x5 m^2), three plots for each variety of rose. The first one of these glasshouses contained infested rose plants and other one left as control. The first glasshouse was arranged in randomized block with three replicates to three varieties (colors) of rose and also the second glasshouse was arranged in randomized block with three replicates as control. The 1st glasshouse was

artificially infestation by *M. rosae* and the 2nd one was left as control. The two glasshouses were in an area isolated from other trees in the garden. Also, the first glasshouse was isolated from the second one. Rose plants were planted in glasshouse conditions at the same time on November (the planting time of rose plants). All agricultural operations of irrigation and fertilization and others are completely identical in the two glasshouses were done without application of any insecticide. Artificial infestation was done by the aphid *M. rosae* in the first glasshouse, with careful observation of the mean numbers of *M. rosae* during plant growth period and especially during the flowering stage from February – August and recorded mean numbers of aphid by direct counting biweekly, with examining the second glasshouse free as (control). With check up the physiological and natural characteristics of rose oil at three levels of infestations during three periods: winter during February month (medium infestation), spring during April month (high infestation) and summer during July month (low infestation).

2.Effect of insect infestation by *Macrosiphum rosae* on the physiological and natural characteristics of rose oil:

This study was carried out to study effect of infested rose plants by *M. rosae* on the physiological and natural characteristics of rose oil through determination the concentrations of most important components of rose oil such as (geraniol, citronellol, nerol, stearpoten, phenyl ethanol and bioflavonoids), acids such as Citric acid and Malik acid and vitamins such as A, B, C and D. Also, determination of the most important natural characteristics of rose oil such as volatility, light rotation, refraction value and freezing point.

3.Determination physiological and natural characteristics of rose oil:

3.1.Rose oil extraction:

Rose oil was extracted from 0.5 kg fresh tissue. The tissues were ground in liquid nitrogen with a mortar and pestle. Then few mls of tris buffer extraction were added (1:2, tissue: buffer). The medium of extraction contained tris-HCL buffer (0.1mM tris, pH 7.5, 4mM B-mercaptoethanol, 0.1mM EDTA-Na_2, 10mM KCl and 10mM $MgCl_2$). The crude homogenate was centrifuged at 10.000xg for 20Min. The supernatant was used for gel analysis by SDS-polyacrylamide gel electrophoresis (SDS-PAGE) according to the method of Laemmli (1970)

3.2. Loading on a gel:

3.2.1.Gel preparation:

Sodium dodecyl sulphate polyacrylamide gel electrophoresis (SDS-PAGE) was performed using 12.5% acrylamide and 0.8% bis acrylamide running gel consisting of 0.375 M Tris-HCl (pH 8.8) and 0.1% SDS. Stacking gel (10 mm) was made using 4.5% acrylamide containing 0.8% bis-acrylamide in 0.125 M Tris–HCl (pH6.8) and 0.1% SDS. The electrophoresis buffer contained 0.025 M Tris-HCl, 0.19 glycine and 0.1% SDS. The samples were homogenized in 0.12M Tris-HCl (pH 6.8), 0.4 SDS, 10 B-mercaptoethanol, 0.02% bromophenolbule and 20% glycerol. The samples were then heated for 3Min. in a boiling water bath before centrifugation. The gel was run under cooling at 90v for the first 15Min, then 120v the next 0.5 hr and finally 150v for the remaining 1.5hr (Sheri *et al*. ,2000).

3.2.2.Sample loading:

A known volume of protein sample was applied to each well by micropipette. Control wells were loaded with standard protein marker.

3.2.3. Electrophoresis conditions:

The running buffer was poured into pre-cooled (4°C) running tank. The

running buffer was added in the upper tank just before running, so that the gel was completely covered. The electrodes were connected to power supply adjusted at 100v until the bromophenol blue dye entered the resolving gel, and then increased to 250v until the bromophenol blue dye reaches the bottom of the resolving gel.

3.2.4.Gel Staining and distaining:

After the completion of the run, gel was placed in staining solution consisting of 1g of Coomassie Brilliant bule–R-250; 455 ml methanol; 90ml glacial acetic acid and completed to 1L with deionized distilled water. The gel was destained with 200ml destaining solution (100ml glacial acetic acid, 400ml methanol and completed to 1L by distilled water) and agitated gently on shaker. The destaining solution was changed several times until the gel background was clear.

3.2.5.Gel analysis:

Gels were photographed using a Bio-Rad gel documentation system. Data analysis was obtained by Bio–Rad Quantity one Software version 4.0.3, the sugar and protein were analyzed by High Pressure Liquid Chromatography (HPLC).

4.Statistical analysis:

In these experiments, effect of the infested rose plants by rose aphid *M. rosae* (in different stages of infestation) on the physiological and natural characteristics of rose oil was subjected to analysis of variance (ANOVA) and the means were compared by L.S.D. test at 0.05 level, using SAS program (SAS Institute, 1988).

Results and discussion

1. Population fluctuation of *Macrosiphum rosae* on rose flowers in Giza Governorate during 2018 season:

Data tabulated in Table (1) showed that the infestation by rose aphid, *M. rosae* adults began to appear in Carmen variety on flowers on the 1st February with 6.5 adults/flower, then the infestation increased gradually to reach 23.5 adults/flower (activity peak) on the mid of April then the infestation decreased until reached to 1.7 adults/flower on mid of August. As the same trend nymphs began to appear in Carmen variety on the 1st February with 15.7 nymphs/flower, then the infestation increased gradually to reach 45.5 nymphs/flower (activity peak) on the mid of April then the infestation decreased until reached to 6.4 nymphs/flower on mid of August. Whereas for Golden gate variety *M. rosae* adults began to appear on flowers on the 1st February with 8.5 adults/flower, then the infestation increased gradually to reach 25.6 adults/flower (activity peak) on the mid of April then the infestation decreased until reached to 3.9 adults/flower on mid of August. As the same trend nymphs began to appear on flowers on the 1st February with 18.4 nymphs/flower, then the infestation increased gradually to reach 50.7 nymphs/flower (activity peak) on the mid of April then the infestation decreased until reached to 9.5 nymphs/flower on mid of August.

Also, for Dream variety *M. rosae* adults began to appear on flowers on the 1st February with 5.3 adults/flower, then the infestation increased gradually to reach 20.5 adults/flower (activity peak) on the mid of April then the infestation decreased until reached to 1.5 adults/flower on mid of August. As the same trend nymphs began to appear on flowers on the 1st February with 13.5 nymphs/flower, then the infestation increased gradually to reach 40.5 nymphs/flower (activity peak) on the mid of April then the infestation decreased until reached to 4.8 nymphs/flower on mid of August.

Table (1): Population fluctuation of *Macrosiphum rosae* (adults-nymphs) infested different rose varieties in Giza Governorate during 2018 season.

Date	Carmen (Red)		Golden gate (Yellow)		Dream (Pink)		Mean Temp.	Mean Hum. %
	Adult	Nymph	Adult	Nymph	Adult	Nymph		
1/2/2018	6.5	15.7	8.5	18.4	5.3	13.5	16.5	72
15/2/2018	12.8	21.8	15.3	25.9	10.5	18.6	18.3	73
1/3/2018	15.3	32.5	17.8	37.5	12.6	26.8	19.5	74
15/3/2018	18.5	37.7	19.3	40.5	15.4	32.5	18.4	70
1/4/2018	20.7	42.3	22.4	45.3	17.3	37.8	18.9	67
15/4/2018	23.5	45.5	25.6	50.7	20.5	40.5	22.7	63
1/5/2018	18.3	37.6	21.5	42.5	16.3	35.2	24.5	60
15/5/2018	15.2	30.5	18.3	36.7	12.3	26.7	25.2	53
1/6/2018	12.7	25.7	15.6	30.4	9.8	20.5	26.4	50
15/6/2018	10.5	20.8	13.4	25.7	7.2	16.7	27.1	47
1/7/2018	8.4	15.3	11.7	20.3	6.8	12.9	27.8	49
15/7/2018	5.9	11.6	8.4	15.4	3.6	9.6	28.6	53
1/8/2018	3.8	9.5	5.6	12.8	2.2	6.5	29.5	58
15/8/2018	1.7	6.4	3.9	9.5	1.5	4.8	30.7	56
Total	173.8	352.9	207.3	411.6	141.3	302.6	-	-
Mean	12.4	25.2	14.8	29.4	10.1	21.6	-	-
F(0.05)	475.85						-	-
L.S.D	1.97						-	-

Means within columns bearing different subscripts are significantly different ($P < 0.05$)

The obtained results are in agreement who those obtained by Mohammad and Al-Mallah (1987) in Iraq who stated that the first appearance of *M. rosae* in large numbers was in mid-February, peaked in early April and disappeared completely by mid-June. Jaskiewicz (2003) studied *M. rosae* population dynamics and found that maximum numbers of this insect was observed during March-May. Hole *et al*. (2017) studied incidence of the rose aphid, *M. rosae* on 30 rose varieties and found that the pest build up started on the third week of February which increased gradually reaching its peak on the fourth week of March and declined thereafter.

2. Effect of infested rose plants by rose aphid, *Macrosiphum rosae* on the physiological and natural characteristics of rose oil:

2.1. Effect of infested rose plants by rose aphid, *Macrosiphum rosae* (in different stages of infestation) on the physiological characteristics of rose oil:

Data tabulated from Table (2) showed comparison between determinations of rose oil components (concentrations) in the rose flowers which infested by rose aphid, *M. rosae* in different stages of infestation (low, medium and high infestation) compared to control (non infested flowers). Data obtained showed that the most important components of rose oil (geraniol,

citronellol, nerol, stearpoten, phenyl ethanol and bioflavonoids), acids such as Citric acid and Malik acid and vitamins such as A, B, C and D were changed its concentrations after infestation by *M. rosae*. Concentrations of these components were more in control compared to its concentrations in infested rose flowers (high, medium and low infestation, respectively). Statically analysis showed were highly significant differences between concentrations of these components in control compared to its concentrations in infested flowers (high, medium and low infestation), respectively.

Table (2): Determination of rose oil components (concentrations) on the different stages of infestation by rose aphid, *Macrosiphum rosae* in Giza Governorate during 2018 season.

Compounds	Concentrations of rose oil components (mg/g)				F(0.05)	L.S.D
	Low infestation	Medium infestation	High infestation	Control		
Geraniol	30.42 [a]	27.65 [b]	25.43 [c]	35.75 [a]	34.29***	1.18
Citronellol	20.18[b]	17.35[b]	15.45 [c]	25.25 [a]	25.31***	1.23
Nerol	16.45[c]	14.21[b]	12.54 [c]	18.35 [a]	19.45**	1.31
Stearpoten	31.45 [b]	27.34 [b]	23.56 [c]	35.21 [a]	23.71*	1.15
Phenyl ethanol	13.32[a]	11.57[d]	10.35[c]	17.54[a]	21.35**	0.75
Bioflavonoids	6.67[b]	5.23[b]	4.12[c]	8.89[a]	64.12**	0.63
Citric acid	10.21[b]	8.67[b]	6.25[c]	12.23[a]	34.18**	1.72
Malik acid	7.12[c]	5.67[a]	4.24[c]	9.24[a]	53.21***	0.16
Vitamin A	5.34[a]	4.45[b]	3.65[c]	7.25[a]	34.17*	0.08
Vitamin B	4.12[a]	3.56[a]	2.89 [c]	5.23[a]	27.29***	1.15
Vitamin C	3.65[a]	2.15[b]	1.50 [c]	4.76[a]	23.12**	0.37
Vitamin D	2.45[a]	1.86[b]	1.20 [c]	3.75[a]	16.24*	1.42

Means within rows bearing different subscripts are significantly different (P< 0.05)

2.2. Effect of infested rose plants by rose aphid, *Macrosiphum rosae* (in different stages of infestation) on the natural characteristics of rose oil:

Data tabulated in Table (3) showed that comparison between natural characteristics of rose oil in rose flowers which infested by *M. rosae* in different stages of infestation (low, medium and high infestation) compared to control (non infested). Data showed that the most natural characteristics of rose oil such as volatility, light rotation and refraction value were changed as result of infestation by *M. rosae*, but other natural characteristics such as freezing point did not change after infestation by *M. rosae.* Statically analysis show were highly significant differences between natural characteristics (except freezing point) of rose oil in infested rose flowers (high, medium and low infestation) compared to its natural characteristics in control.

Table (3): Natural characteristics of rose oil as result of infestation by rose aphid, *Macrosiphum rosae* in Giza governorate during 2018 season.

Characteristics	Low infestation	Medium infestation	High infestation	Control	F(0.05)	L.S.D
Volatility	0.812 [a]	0.765 [b]	0.760 [c]	0.855 [a]	33.97***	0.79
Light rotation	0.52[a]	0.48[b]	0.43 [c]	0.55 [a]	21.15**	0.58
Refraction value	1.420[a]	1.400[b]	1.365 [c]	1.460 [a]	31.28***	0.73
Freezing point	20C	20C	20C	20C	-	-

Means within rows bearing different subscripts are significantly different (P< 0.05)

The obtained results are in agreement who those obtained by Emam (2009) in Egypt who studied effect of infestation by *M. rosae* on the interior components of rose flowers, and found that natural characteristics of rose oil changed as result to the infestation by *M. rosae.* Peng and Miles (1991) studied the changes in the internal components of rose flowers such as rose oil, protein, sugar and vitamins, which infested with some insects and decided that the most effective in these components was the infestation by *M. rosae*. Becker and Apel (1992) reported that the decrease in concentrations of rose oil components may be due to the infestation by *M. rosae*. Atwal and Dhingra (2018) reported that the infestation by *M. rosae* was changed in the concentrations of rose oil components in the rose petals. While, Jaskiewicz (2006) studied the changes which happened in protein pattern in the rose petals which infested by *M. rosae*.

Also, the obtained results are in agreement with those obtained by Decheva (2015) in Bulgaria who investigated the changes in the rose oil components in flowers of rose plants, and found that the level of 12 rose oil components identified decreased as result of infestation by *M. rosae*.

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Effectiveness of baited traps for controlling oriental hornet *Vespa orientalis* (Hymenoptera: Vespidae) in Sohag Governorate apiaries

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Abstract:

The oriental hornet *Vespa orientalis* L. (Hymenoptera: Vespidae) is one of the most important pests attacking apiaries in arab countries. The aim of this work is to modify a new effective trap and compared it with the beekeeper hornet traps. Also determine the best baits used and the population dynamics of the *V. orientalis* adults in Sohag Governorate. Data revealed that, hive new trap catches the high number of the hornet, *V. orientalis* followed significantly by the wooden trap, but plastic trap catch the lowest number of the hornet. The best bait attracts the hornet *V. orientalis* was honey followed not significantly by syrup (1sugar:1water) followed significantly with candy and sugar and the lowest number of hornet attracted to the pollen. The queens of *V. orientalis* appeared at the beginning of March and its number increased gradually to peak at the end of April. While, the workers of *V. orientalis* appeared during June and increased gradually to peak at November. It is concluded that hive trap with honey or syrup bait is the best way to control *V. orientalis* hornets at the apiaries.

Introduction

The oriental hornet *Vespa orientalis* L. (Hymenoptera: Vespidae) is one of the specific factors for the expansion of apiaries in the valley lands in Egypt. Where it affects the apiaries significantly, sometimes leads to the destroyed of the honeybees in areas severely infected with these hornets. Beekeepers refrain from the establishment of apiaries in places infected by the hornet and sometimes forced to transfer their apiaries when the attack increased (Abdelaal and El-Defrawy, 2014).

Hornets attack a honeybee colony which considers an important source of carbohydrates (honey) and protein substances (adults, brood and pollen) (Mattu and Sharma, 2017). The hornets attack guard bees at hive entrances and foraging workers and resulted in weakening strong colonies (Matheson *et al*., 1989). The hornets attack on the weak colonies, then attacks the

healthier ones (Ifantidis, 2003). Many methods had been done to find a way to control hornets like, the manual collection of the hornet queens and workers by using palm branches, destroying the hornet nest (Robin and Dupres, 1945); fumigating the nest with calcium cyanide, spraying it by insecticides (Subbiah and Mahadevan, 1958); burning it (Bhutani, 1950 and Singh, 1962); tried honey bait, mixed with different insecticides beside hornet nests (Aihara, 1980 and Walfa *et al*., 1969); modificate the hive entrance to prevent the hornet from entering to the hive by using a queen guard or queens gate (Dave, 1943) or a large cuboid queen excluder (Abd Al- Fattah *et al.*,2014); using a poison baits, like Fipronil insecticide mixed with the beef bait to control the oriental hornet (Al-Heyari *et al.*,2016); dusting hornets by insecticides and releasing it to return to nests and destroyed it (Ghania and Abdel-Aziem, 2015) and finally using baited traps (Ibrahim and Mazeed, 1967; Longo, 1980 and Walfa *et al*., 1969). But nowdays an effective control program for the oriental hornet has not been developed.

The aim of this work is to modify new effective trap and compared it with the beekeeper hornet traps. Also determine the best baits used, attract the largest number of hornets. Finally, to determine the population dynamics of the *V. orientalis* adults at Sohag Governorate.

Materials and methods

The present study was carried out in three private apiaries in Sohag Governorate, during two seasons of 2017 and 2018.

1.Traps description:

1.1. Hive new trap: was consisted of the Langstroth brood box where the base is replaced with a wire funnel that allows the hornets to enter and inhibit it to return again. The hive cover was replaced by a wire net with hole diameters that allow, the bees to exit and inhibit the hornets and placed on a wooden chair. A bowl of a bait is placed underneath the funnel **(Figure, 1)**.

1.2.Wooden trap: was made of wooden bars and wire screen with diameters of 45 X 45 X 85 cm (Abdelaal and El-Defraw , 2014).

1.3. Plastic trap: A plastic bottle with piece of queen excluder at both sides (Abdelaal and El-Defrawy, 2014).

2. Efficacy of new designated traps in order to control *Vespa orientalis* adults:

In the first apiary three traps of each type were randomly distributed between the experimental colonies (60 colonies). These traps established and baited with 250ml of sugar syrup (1sugar:1water) from the first week of August until the last week of November through the two years of the study, 2017 and 2018 (Abdelaal and El-Defrawy, 2014). The time of the bait placement was early in the morning. The baits were changed regularly every three days (Bacandritsos *et al*., 2006). Samples were collected weekly and the hornets attracted and caught within the given traps were counted and the average number of wasps/trap/week was calculated.

Figure (1): Hive new trap

3. Efficacy of some baits to attract *Vespa orientalis* adults:

In the second apiary five baits were examined , honey, sugar syrup (1sugar:1water), candy, sugar (granules) and pollen 250 g. of each bait put in the bowl underneath the funnel of hive trap and replaced every three days. Each treatment replicated three times. The hornets caught within the traps were weekly collected and counted.

4. Population dynamics of the *Vespa orientalis* adults during seasons of 2017 and 2018:

In the third apiary three hive traps were used to determine the population dynamics of the *V. orientalis* adults with the bait of 250 ml sugar syrup (1 sugar :1water) replaced every three days. Samples were collected weekly and the hornets attracted and caught within the trap were counted and the average number of wasps/trap/week was calculated.

5.Statistical analysis:

The data obtained were subjected to regular statistical analysis (one way ANOVA) and mean comparison were carried out using L.S.D. at 5% (Snedecor, 1956).

Results and discussion

1. Efficacy of hivenew designated traps in order to control *Vespa orientalis* adults:

Data in Table (1) showed that hive trap catch the highest number of the hornet, *V. orientalis* with averages (1478.7 and 1063.7 hornets) during the two seasons, respectively. However, plastic trap catches the lowest number of the hornet with averages (229.33 and 175.33 hornets) during the two seasons, respectively. While, wooden trap catches the moderate number of the hornet with averages (665.67 and 397.00 hornets) during the two seasons, respectively. There was a significant difference between all traps.

These data are in partial agreement with those of Dwara and Hatom (2013) who tested the effect of three types of traps on reducing the damage of red wasp (*V. orientalis*) on bees, the used traps were small trap, cylindrical trap and a trap consisted of a hive with honey chamber trap. Results showed that hive with honey chamber trap was the best, with a mean catch of 1191.94 insects per two months and the worst was the cylindrical trap, with a mean catch of 925.17 insects per two months. Abdelaal and El-Defrawy (2014) studied the efficiency of three traps, the wooden trap, plastic bottles (20L. water bottle with piece of queen excluder in the end) and a small plastic bottle(2 L. plastic bottle with piece of queen excluder in both sides). They found that the plastic bottle 20L. was the best trap for the *V. orientalis*.

Table (1): Efficacy of three traps on hornet attraction during 2017 and 2018 seasons in Sohag Governorate.

Traps	2017	2018
Hive trap	1478.7	1063.70
Wooden trap	665.67	397.00
Plastic trap	229.33	175.33
F. value	87.10	67.63
L.S.D.	235.10	194.57

2. Efficacy of some baits to attract *Vespa orientalis* adults:

The data in Table (2) showed that the best bait attracts the hornet *V. orientalis* was honey with hornet averages (826.67 and 713.33 hornets) followed insignificantly by syrup (1:1) with hornet averages (811.00 and 688.33 hornets) followed significantly with candy with hornet averages (344.33 and 275.67 hornets) and sugar with hornet averages (209.00 and 182.00 hornets) and finally the pollen with hornet averages (122.00 and 100.33 hornets) during the two seasons, respectively.

These data were in partial agreement with those of Spurr (1996) who found that both meat-based and sugar-based food materials can be used to trap many species of social wasps. Dwara and Hatom (2013) found that cow lungs were significantly the best attractive bait to *V. orientalis* and the poultry guts was the least attractive bait. Al Antary *et al.* (2016) studied the acceptance of the oriental hornet for the baits (beef, chicken, liver and sardine) and found that the best percentage of baits consumed by the hornet after two hours was beef meat.

Table (2): Number of hornets attracted to different baits during 2017 and 2018 seasons.

Baits	2017	2018
Honey	826.67	713.33
Syrup	811.00	688.33
Candy	344.33	275.67
Sugar	209.00	182.00
Pollen	122.00	100.33
F. value	33.5	73.9
L.S.D.	235.10	194.57

3.Population dynamics of the *Vespa orientalis* adults during 2017 and 2018 seasons:

Data illustrated in Figure (2) showed that during the two seasons, *V. orientalis* queens appeared at the beginning of March and its number increased gradually and peaked at the end of April, then decreased through May. At June the *V. orientalis* workers appeared and increased gradually and peaked at November then decreased through December and disappeared during January and February.

The obtained results confirmed partially the findings of Gomma and Abd El-Wahab (2006) who reported that there is marked increase in the hornets from the second week of August (51 hornets/ trap) to the fourth week of September (3301 hornets /trap). Taha (2014) revealed that *V. orientalis* queens were starting to appear from January to May, with a peak of activity during May. While *V. orientalis* workers were collected from June to February, with a peak in October, then November, respectively. Also, Islam *et al.* (2015) showed that *V. orientalis* were found attacking honeybees, *Apis mellifera* during June to November and reached to the highest number during the second week of October (220 hornets/week).

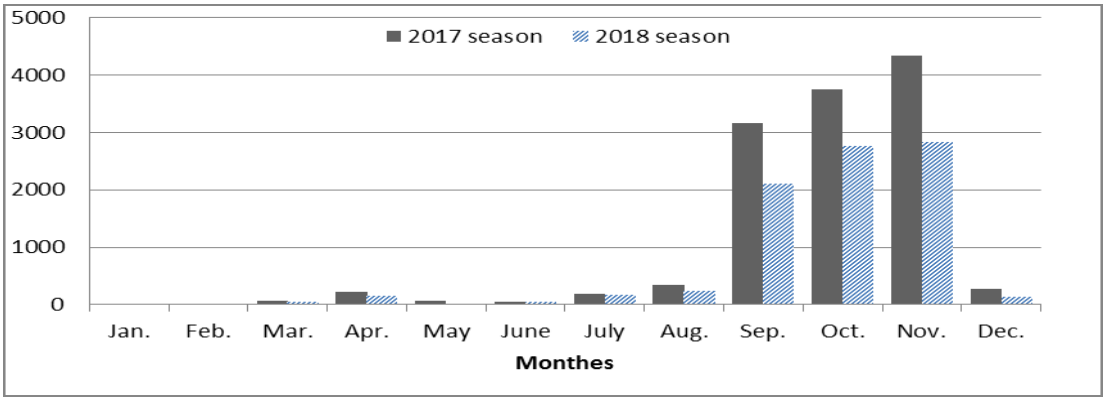

Figure (2): Population dynamics of *Vespa orientalis* adults during 2017 and 2018 seasons.

It is concluded that hive trap with honey or syrup bait is the best way to control *V. orientalis* hornets at the apiaries.

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Toxicity of some insecticides against the oriental hornet *Vespa orientalis* (Hymenoptera: Vespidae)

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Toxicity, insecticides , oriental hornet, *Vespa orientalis* , diluting and honeybee colony.

Abstract:

The present investigations were carried out at the laboratory of Shandaweel Agricultural Research Station, Sohag Governorate, Egypt during 2016 season to evaluate toxicity of Lambda-cyhalothrin 10%WP, Acetamiprid 25% SP, Diazinon 60% EC and Emamectin Benzoate 5% SG insecticides against the oriental hornet *Vespa orientalis* L. (Hymenoptera: Vespidae). Also to evaluate diluting these insecticides with sugar, flour and talc powder and its effect against the oriental hornet. Also to study indirect effects of Acetamiprid 25% insecticide with different concentration on *V. orientalis* adults. The results showed that Acetamiprid 25% and Lambda-cyhalothrin 10% recorded the lowest time required for killing the wasps with 1.88 and 3.04 Minutes, respectively, by no significant difference between them. However, the highest time was recorded for Emamectin Benzoate 5% with 122.40 minutes. Diluted all insecticides with sugar recorded the shortest time needed to kill oriental hornet with mean number (3.67, 4.48,100.10 and 159.10 Minutes) for Lambda-cyhalothrin 10%, Acetamiprid 25%, Diazinon %60 and Emamectin Benzoate %5, respectively. The indirect effects of Acetamiprid 25% recorded the shortest time needed to kill oriental hornet with 25% concentration (2.96 Minutes) for stage1. For stage 2, the 2.5% concentration with sugar recorded the shortest time needed to kill hornet with average (7.14 Minutes).

Introduction

The oriental hornet, *Vespa orientalis* L. (Hymenoptera: Vespidae) is considered a major pest to honey bees in many arab countries such as Egypt, Iraq, Saudi Arabia, United Arab Emirate, Lebanon, Oman ,Yemen , Sudan, Syria, Jordan and Palestine (Glaiim,2009; Khodairy and Awad, 2013; Abdelaal and El-Defrawy, 2014 and Al-Heyari *et al.*, 2016). Among many pests were found attacking *Apis mellifera* L. (Hymenoptera : Apidae) colony, *V. orientalis* was recorded to be one of the most destructive pests to the bees (Vishwakarma *et al.*, 2012). The oriental hornet, *V. orientalis* is considered one of the factors which effective for the honey bee colonies in

Egypt because it caused heavy losses in apicultural development (Ghania and Abdel-Aziem, 2015). It considered a predatory–carnivorous insects feeding mainly their brood with animal proteins (insects, pieces from fresh or spoiled meat and fish) while the adults are fed with carbohydrates (nectar, honeydew and ripe fruits). Bee hives constitute places where the wasps can find the best combination of proteins from animal origin (bees or larvae) and carbohydrates (nectar and honey) (Bacandritsos *et al*., 2006). At first they attack on the weak bee colonies, which are the most defenseless and then the damages are extended to the healthier ones. Bee colonies can be weakened by wasp, especially hornet predation (Adlakha, 1975 and Akratanakul, 1986).

In order to control wasps, various methods have been used based either on the use of insecticides or on the traps of free of insecticides. Baits mixed with insecticides were used for wasps control, in this case the adults become victims but before they are killed and transfered the poisoning insecticide to the brood (Tsanakakis and Katsogiannos, 1998 and Sackmann *et al*., 2001). In other cases, the nest can be detected and sprayed during the night with various insecticides (Ifantidis, 1995). Ghania and Abdel-Aziem (2015) found that dusting the trapped wasps by linnet insecticide 90% at the rate 1 gram to 100 gram powder sugar and return release it every week were reduced the wasps in the traps for three months after releasing. The objective of this study is to evaluate toxicity of Lambda-cyhalothrin 10%WP, Acetamiprid 25% SP, Diazinon 60% EC and Emamectin Benzoate5% SG insecticides against the oriental hornet. Also to evaluate diluting of these insecticides with sugar, flour and talc powder and its effect against the oriental hornet. Indirect effects of dilution Acetamiprid 25% insecticide by different substance (sugar, flour and powder) against *V. orientalis* adults in the laboratory.

Materials and methods

The present investigation was carried out at the laboratory of Shandaweel Agricultural Research Station, Sohag Governorate, Egypt during 2016 season.

1. **Hornets stock**: The hornets were collected from apiaries using a modified sweep net (with a transparent clothes). Each 5 hornets placed in a plastic jar with some sugar and a piece of cotton wet with water until to execute the experiments.
2. **Insecticides**: Table (1) showed four insecticides were used and evaluated the toxicity against the oriental hornet *V. orientalis*.

Table (1): **Insecticides used to evaluate the toxicity against the oriental hornet, *Vespa orientalis*.**

Common name	Family	Mode of action
Lambda-cyhalothrin 10%WP	Pyrethroid	contact and stomach action
Acetamiprid 25% SP	Neonicotinoid	contact and stomach action
Diazinon 60% EC	Organophosphor	contact and stomach action
Emamectin Benzoate5% SG	Avermectin	Acts on nerve cells

3. Effect of certain insecticides on *Vespa orientalis* adults:

In this experiment, 10 plastic jars were used, each with 5 wasps. These wasps were transferred to jars with the insecticide to be tested and shaken several times. It is then placed in a clean jar and inspected every minute for quick-acting insecticides and every five minutes for slow-acting insecticides.

4. Effect of insecticides diluted with different substance (sugar, flour and powder) on *Vespa orientalis* adults:

In this experiment, the previous insecticides were diluted using sugar,

flour and talcum powder then the wasps were dusted in the same way as before.

5. Indirect effects of Acetamiprid 25% insecticide with different concentration on *Vespa orientalis* adults:

In this experiment, the indirect effect of Acetamiprid 25% diluted with different substance (sugar, flour and powder) on *V. orientalis* adults was studied in two stages:

Stage 1: In which a wasp was dusted with the insecticide placed with 5 wasps not treated with insecticide. It was repeated 10 times.

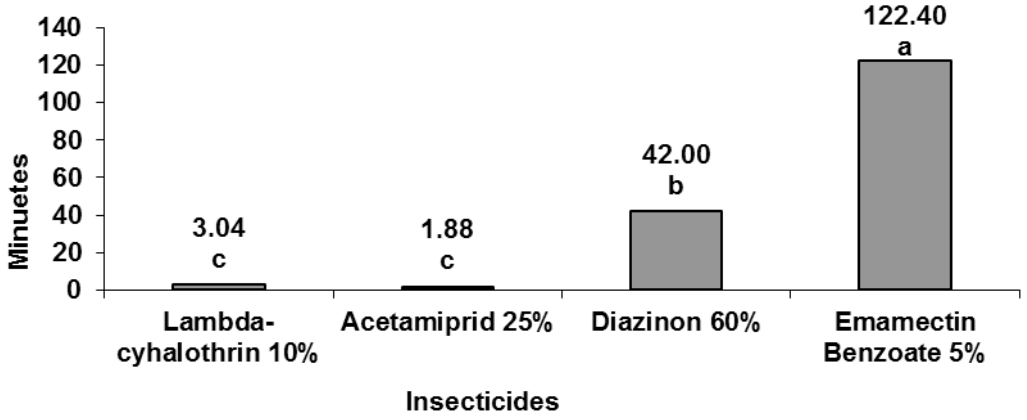
Stage 2: In which the wasps were placed from the previous 5 wasps after dead with 5 other untreated wasps. It was repeated 10 times.

Results and discussions

1. Effect of certain insecticides against *Vespa orientalis* adults:

Data in Figure (1) showed that the effect of certain insecticides on *V. orientalis* adults. It is clear that Acetamiprid 25% and Lambda-cyhalothrin 10% recorded the lowest time required for killing the wasps with 1.88 and 3.04 Minutes, respectively, by no significant difference between them. However, the highest time was recorded for Emamectin Benzoate 5% with 122.40 Minutes.

These results are in partially agreement with Al-Heyari *et al*. (2016) who tested 4 insecticides against the hornets and found that the total number of the dead oriental wasps counted after one hour of treatment were 80.42% , 78.50%,75.17% and 68.33 % for Fipronile, Imidacloprid, Diazinon and Deltamethrin, respectively.



F. value = 1291.594*

Figure (1): Effect of certain insecticides on *Vespa orientalis* adults.

2. Effect of insecticides diluted with different substance (sugar, flour and powder) against *Vespa orientalis* adults:

2.1. Effect of Lambda-cyhalothrin 10% diluted with sugar, flour and powder against *Vespa orientalis* adults:

Data in Table (2) showed that the effect of Lambda-cyhalothrin 10% diluted with different substance against *V. orientalis* adults. The average of all counts were calculated as shown in Table (2), which indicated that the dilution with sugar recorded the shortest time needed to kill hornet with mean time (3.67 Minutes) followed significantly by flour (4.89 Minutes) and powder (5.01 Minutes) with no significant difference between them. The highest concentration gives the lowest time and the inverse is correct. The concentrates 5% and 1% recorded (3.30 and 5.74 Minutes respectively).

The interaction between concentrates and the dilution substance was significant. The shortest time was found at concentrate 5% with sugar (3.14 Minutes) followed insignificantly by

flour and powder at 5% and sugar at 1%. However, the longest time was observed in concentrate 1% with powder with insignificant difference with flour at the same concentration with (6.64 and 6.38 Minutes, respectively). These results are in partially agreement with Raghavendra *et al.* (2011) whom found that in insecticide susceptibility tests, *Anopheles culicifacies* Giles (Diptera : Culicidae), registered 97% mortality to Malathion and 93% to Lambda-cyhalothrin.

Table (2): Effect of Lambda-cyhalothrin 10% diluted with sugar, flour and powder against *Vespa orientalis* adults.

Time per Minutes			
	5%	1%	Mean
Sugar	3.14 b	4.20 b	3.67 b
Flour	3.40 b	6.38 a	4.89 a
Powder	3.38 b	6.64 a	5.01 a
Mean	3.30	5.74	-----

F. value for matters= 6.1977*, F. value for concentrates= 46.5985* and F. value for interaction=3.7622*L.S.D. value for matters= 0.9177

2.2. Effect of Acetamiprid 25% diluted with different substance (sugar, flour and powder) against *Vespa orientalis* adults:

Data in Table (3) showed the effect of Acetamiprid 25% diluted with different substance against *V. orientalis* adults. The mean of all counts were calculated as shown in Table (3), which indicated that the dilution with sugar recorded the shortest time needed to kill hornet with mean number (4.48 Minutes), however, the longest time was recorded in powder (11.07 Minutes). The highest concentration gives the lowest time and the inverse is correct. The concentrates 12.5% and 2.5% recorded (5.13 and 9.50 Minutes respectively).

The interaction between concentrates and the dilution substance was significant. The shortest and the longest times were found in concentrate 12.5% with sugar (3.04 Minutes) and in concentrate 2.5% with powder, respectively. These results are in partially agreement with Iwasa *et al.* (2004) who conducted a laboratory bioassays to determine the contact honey bee toxicity of commercial and candidate neonicotinoid insecticides and found that the LD50 for acetamiprid was 7.1mg/bee.

Table (3): Effect ofAcetamiprid 25% diluted with different substance (sugar, flour and powder) against *Vespa orientalis* adults.

Time per Minutes			
	12.5%	2.5%	Mean
Sugar	3.04 d	5.92 c	4.48 c
Flour	4.64 c	8.18 b	6.41 b
Powder	7.72 b	14.42 a	11.07 a
Mean	5.13	9.50	-----

F. value for matters= 82.0548*, F. value for concentrates= 153.2809* and F. value for interaction= 11.1370* L.S.D. value for matters= 1.153

2.3.Effect of Diazinon 60% diluted with different substance (sugar, flour and powder) against *Vespa orientalis* adults:

Data in Table (4) showed the effect of Diazinon 60% diluted with different substance against *V. orientalis* adults. Data indicated that the dilution with sugar recorded the shortest time needed to kill hornet with mean number (100.1 Minutes) compared with, flour (113.00 Minutes) and powder (116.60 Minutes) with insignificant difference between the last two.

The highest concentration gives the lowest time and the inverse is correct. The concentrates 3% and 6% recorded (86.40 and 133.40 Minutes, respectively). The interaction between concentrates and the dilution substance was significant. The means can arrange in three significantly groups. The highest included concentrate 6% with flour and powder (138.80 and143.00 Minutes) , the moderate was the concentrate 6% with sugar and the lowest group consisted of concentrate 30% with sugar, flour and powder (81.80, 87.20 and 90.20 Minutes). These results are in partially agreement with Al-Heyari *et al*. (2016) who found that the total number of the dead oriental wasps counted after one hour of was 75.17% for Diazinon insecticide.

Table (4): Effect of Diazinon60% diluted with different substance (sugar, flour and powder) against *Vespa orientalis* adults.

Time per Minutes			
	30%	6%	Mean
Sugar	81.80 c	118.40 b	100.10 b
Flour	87.20 c	138.80 a	113.00 a
Powder	90.20 c	143.00 a	116.60 a
Mean	86.40	133.40	-----

F. value for matters= 9.9061*, F. value for concentrates= 328.8290*, and F. value for interaction= 4.0430
L.S.D. value for matters= 8.494

2.4. Effect of Emamectin Benzoate 5% dilutedwith different substance(sugar, flour and powder) against *Vespa orientalis* adults:

Data in Table (5) show the effect of Emamectin Benzoate %5 diluted with different substance against *V. orientalis* adults. Data indicated that the dilution with sugar recorded the shortest time needed to kill hornet with mean number (159.10 Minutes) compared with, flour (230.00 Minutes) and powder (233.90 Minutes) with insignificant difference between the last two. The highest concentration gives the lowest time and the inverse is correct. The concentrates 2.5% and 0.5% recorded (190.13 and 225.20 Minutes respectively). The interaction between concentrates and the dilution substance was significant. The concentrate 2.5% with sugar recorded the shortest time needed to kill hornet with mean number (136.60 Minutes), however, the longest time was recorded by concentrate 0.5% with powder (247.20 Minutes) followed insignificantly by the same concentrate with flour (246.80 Minutes).

These results are in partially agreement with Bengochea *et al.* (2014)who tested emamectin benzoate against the different stages of *Spodoptera exigua* (Hübner) (Lepidoptera: Noctuidae) and found that it progressive neonate mortality at all concentrations, culminating at 72 hours after hatching, when 100% of the larvae from the treated young eggs died. Also second and fourth instar *S. exigua* larvae did not exhibit significant mortality when exposed to the inert surfaces which were treated.

Table (5): Effect of Emamectin Benzoate 5% diluted with different substance (sugar, flour and powder) against *Vespa orientalis* adults.

Time per Minutes			
	2.5%	0.5%	Mean
Sugar	136.60 d	181.60 c	159.10 b
Flour	213.20 b	246.80 a	230.00 a
Powder	220.60 b	247.20 a	233.90 a
Mean	190.13	225.20	-----

F. value for matters= 92.3436*, F. value for concentrates= 207.1703*
F. value for interaction= 4.8439* L.S.D. value for matters= 13.50

3. Indirect effects of Acetamiprid 25% insecticide with different concentration against *Vespa orientalis* adults:

Data in Table (6) showed the indirect effects of dilution of Acetamiprid 25% insecticide by different substance (sugar, flour and powder) against *V. orientalis* adults. For stage1, the shortest time needed to kill hornet was recorded at 100% treatment (2.96 Minutes) followed insignificantly with sugar 12.5% and sugar 2.5% by means (4.26 and 5.70 Minutes). On the other hand, the highest time recorded with flour 2.5% by mean (113.00 Minutes).

For stage 2 the sugar 2.5% recorded the shortest time needed to kill hornet with mean numbers (7.14 Minutes) followed insignificantly with sugar 12.5% (8.30 minutes). However the highest time was recorded at 100% treatment with mean (11.12 Minutes). It is clear evidence that insecticide diluted with sugar recorded the least time of mortality may be it backs to hornets feeding on sugar so the insecticide effect by two ways as a contact poison and a stomach poison.

Table (6): Indirect effects of Acetamiprid 25% insecticide with different concentration against *Vespa orientalis* adults.

Time per minutes		
	Stage 1	**Stage2**
100%	2.96 e	11.12 a
Sugar 12.5%	4.26 e	8.30 b
Sugar 2.5%	5.70 e	7.14 b
Flour 12.5%	90.40 b	No
Flour 2.5%	113.00 a	No
Powder 12.5%	29.50 d	No
Powder 2.5%	41.40 c	No

No: Mean that no death in hornets till three days.
F. value for first worker= * F. value for second worker= *

If beekeepers, know the hornets' nest place or expected it is near the apiary they can use the fast killer insecticides Lambda-cyhalothrin 10%WP and Acetamiprid 25%SP insecticides. While if they, don't know the hornets' nest place or expected it is far from the apiary they can use the slow killer insecticides , Diazinon 60% EC and Emamectin Benzoate5% SG by dusting the hornets and rerelease it.

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Field evaluation for response of Mediterranean fruit fly *Ceratitis capitata* and peach fruit fly *Bactrocera zonata* (Diptera: Tephritidae) by tested attractant materials

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Abstract:

Recently, the peach fruit fly *Bactrocera zonata* (Saunders) (Diptera: Tephritidae) is recognized as causing fruit damage on a range of fruits and other economic host plants . Mediterranean fruit fly , *Ceratitis capitata* (Wiedemann) (Diptera: Tephritidae) is recorded in Egypt early last century and considerd as a serious pest of fruits. The present investigation was conducted in two locations with two crops (peach and mango) , to study response of fruit flies *B. zonata* and *C. capitata* to different chemical compounds. The obtained data indicated that 3 % Di-amunum phosphate recorded the highest mean numbers of weekly captured to *B. zonata* and *C. capitata*, followed by 10 % Amunum- Acetate and Trimethylamine (TMA) recorded a moderately attractiveness and other chemical compounds treated was less attractants . While 5 % Buminal recorded the highest attractants to attract female *C. capitata* captured . But 10 % Urea Fertilizer [CO(NH2)2] recorded the highest attractants to attract female *B. zonata*. Data presented showed that the females number of *B. zonata* and *C .capitata* were more attracted than male flies to tested different chemical compounds , also the number of attracted flies of *B. zonata* and *C. capitata* to plastic bottles traps (liquid trap) was highest than to using plastic containers (dry trap) .

Introduction

Peach fruit fly *Bactrocera zonata* (Saunders) (PFF) and Mediterranean fruit fly *Ceratitis capitata* (Wiedemann) (MFF) (Diptera: Tephritidae) are the most serious pests of fruits. The peach fruit fly *B. zonata* is considered one of the most important economic pests for several kinds of fruits in temperate, tropical and subtropical countries (Younes *et al.*, 2009). Also *B. zonata* has been recorded in Egypt since 1990 and it is widely spread recently in all Egyptian provinces attacking several fruits (Shehata *et al.,* 2006). It has been recognized as a serious insect pest during the last decade attacking a wide range of fruits in Egypt (Fahmy *et al.,* 2013) and is

a highly polyphagous species infesting more than 50 host plants, including peach, guava, mango, apricot, fig and citrus (Peňa *et al*., 1998). This pest is difficult to be controlled by the traditional chemicals due to the behavior of its larvae that hide inside the fruits or its pupae that pupate in the soil (Saleh *et al*., 2018). The Mediterranean fruit fly, *C. capitata* is a serious pest and worldwide pest attacking a wide range of different crops. It attacks many fruit species and some vegetable crops such as tomato, pepper and eggplant (Kapoor and Agrawal 1982; El-Minshawy *et al*., 1999 and and Ghanim, 2009) . Lures for capturing female fruit flies are based on food or host odours. Historically, liquid protein baits have been used to catch a wide range of different fruit fly species , liquid protein baits capture both females and males, with a higher percent of females captured , trimethylamine results in a highly attractive female lure for medfly which is being used in early detection trapping networks. This synthetic food lure is more specific than the liquid protein baits and is capable of detecting female medfly at a lower level compared to the male specific attractant (IAEA , 2003) . The aim of the present work is to study response of fruit flies *B. zonata* and *C. capitata* to different chemical compounds.

Materials and methods

1. Experimental area:

This investigation was conducted in two locations in the desert back in Shebin El-Qanater and Tookh districts, Qalyoubia Governorate with two crops, peach and mango , respectively, cultivated in the sandy soil .The first experiment was carried in Shebin El-Qanater district in the farm cultivated peach 6 years age during the period from the end of third week of May to 18th June 2018 with high population density of Mediterranean fruit fly, *C.* 40 males / trap / day , while the number of captured flies per trap per day (CTD) for peach fruit fly, *B. zonata* 0.28 males / trap / day.The second experiment was carried in Tookh district in the farm cultivated mango 20 years age , during the period from 12th July to 9th August 2018 , with CTD 35.7 and 10 males / trap / day for , PFF and MFF respectively

2. Used Trap :

Two trap types were used to catch fruit flies , attractant materials were used in two forms (dry paste and liquid form). The first one , plastic bottles (measuring 16 cm in height and 7 cm in diameter) used four holes numbers were made at the end of the upper third of the bottle were fixed by a metallic wire , as the liquid trap. The second trap was plastic containers (measuring 13 cm in height and 11 cm in diameter) with four holes numbers were made at the end of the upper third of the bottle fixed by a metallic wire , as the dry trap (Figure, 1). Inner cover of plastic containers was covered with 2% Lmbada pesticide to catch fruit flies .The dry paste is composed using cotton wick and clink paper saturated with the same concentration inside perforated paper bags put inside trap.

3 . Attracting compounds :

Six attractant materials were used in two locations on the peach and mango crops , every crop with two form liquid and dry traps, (10% Ammonium acetate , 10 % Urea Fertilizer [CO(NH2)2], Trimethylamine (TMA) (using cotton wick saturated with TMA), 1 % Dab Fertilizer (H9N2O4P) , 3 % Di-ammonium phosphate and 5 % Buminal .The experimental conducted by using 4 traps × 6 tested materials and in each location, each treatment was replicated four times and distributed in a completely randomized design .

4 . Statistical Analysis:

Data was analyses by using ANOVA in SAS (SAS Institute 2003) .

Figure (1): Trap types

Results and discussion

1 . Response of fruit flies to the tested chemical compounds attractants in peach orchard :

This experiment was carried in peach orchard with high population density of Mediterranean fruit fly *C. capitata* CTD , 40 males / trap / day , while CTD for Peach fruit fly, *B. zonata* 0.28 males / trap / day , by using two traps types , plastic bottles with liquid form and plastic containers with dry form . The obtained results are summarized in Tables (1 and 2) as :

1.1. The peach fruit fly *Bactrocera zonata*:

Data in Tables (1 and 2) showed that mean of weekly captured *B. zonata* by different chemical compounds was nil because population abundance of *B. zonata* was obviously low in this period of the year according the environmental conditions especially temperature degrees. From the obtained results temperature plays a significant role in the population of *B. zonata* especially in summer season. These results support those obtained by (Amin, 2003) and (Afia, 2007) mentioned the lowest abundance was recorded in winter months at Fayoum Governorate and during spring season at Qualyobia and Giza Governorates when temperature degrees higher 30°C; was suitable to increase the numbers of *B. zonata* , but below 30°C. *C. capitata* was usually the successful species. Hashem *et al.* (2007) reported that population density of *B. zonata* is highly affected by two factors, presence of the suitable host plant in addition to the adequate climatic factors especially mean temperature. Also *B. zonata* is more sensitive to low temperature than *C. capitata*, according to Nelson and Greeff (2009).

1. 2 . The Mediterranean fruit fly *Ceratitis capitata*:

Data presented in Tables (1 and 2) showed that the mean of weekly captured *C. capitata* by different chemical compounds during the period from the end of third week of May to 18th June 2018 . Firstly, data in Table (1) showed the mean of weekly captured of *C. capitata* in peach orchard by using plastic bottles (liquid trap). Statistical analysis were highly significant between the different tested food attractants ; the highest mean numbers of weekly captured *C. capitata* was 111.7 and 90.5 flies / trap for Di-amunum phosphate and Amunum-Acetate attractants, respectively ; so it can be say 3% Di-amunum phosphate and 10 % Amunum- Acetate attractants preference to attract *C. capitata* . While 5 % Buminal , 1 % Dab Fertilizer (H9N2O4P) and Trimethylamine recorded a moderately attractiveness to attract *C. capitata* with mean numbers of weekly captured 87.5 , 78.31 flies and 68.75 / trap, respectively . While 10 % Urea Fertilizer [CO(NH2)2] was the less efficient in attracting of *C. capitata* , the mean numbers of weekly captured was 26.81 flies . Also the obtained data indicated that in Table (1) the females number of MFF were more

attracted than male flies to tested different chemical compounds with general average; the first level recorded highest mean females number of *C. capitata* 127.12 , 118.62 , 99.25 and 93.62 female / trap for 5% Buminal , 3% Di-amunum phosphate , 1 % Dab Fertilizer (H9N2O4P) and 10 % Amunum- Acetate attractants, respectively and while the males of *C. capitata* were 47.87 , 104.75 , 53.37 and 87.37, respectively . The second level recorded less efficient in attracting mean females number of *C. capitata* 80.75 and 42.87 female / trap for Trimethylamine and Urea Fertilizer [CO(NH2)2], respectively ; but mean males number of *C. capitata* were 56.75 and 10.75, respectively . Secoundly, the obtained data in Table (2) indicated that by using plastic containers (dry trap); the numbers of attracted fruit fly to it was much lower than the numbers that were attracted to the plastic bottles traps (liquid trap); the mean of weekly captured of *C. capitata* in peach orchard and statistical analysis showed that highly significant between the different tested food attractants. Data were divided into three groups ; the first one was high attraction , the highest mean numbers of weekly captured *C. capitata* was 4.97 flies / trap Trimethylamine ; the second one was moderately attraction , the mean numbers of weekly captured *C. capitata* was 3.78 , 3.28 and 3.03 flies / trap for 3% Di-amunum phosphate , 10 % Amunum-Acetate and 5 % Buminal, respectively and the third one was less attraction recorded that 2.35 and 1.09 flies / trap for 10 % Urea Fertilizer [CO(NH2)2] and 1 % Dab Fertilizer (H9N2O4P), respectively .

2. Response of fruit flies to the tested chemical compounds attractants in mango orchard :

This experiment was carried in mango orchard with population density of ctd for peach fruit fly *b. Zonata* 35.7 males / trap / day and while mediterranean fruit fly *c. Capitata* ctd , 10 males / trap /day by using two trap types as already mentioned. .

2.1. The peach fruit fly *Bactrocera zonata*:

Data obtained in Table (3) showed that the mean of weekly captured *B. zonata* by different chemical compounds by using plastic bottles (liquid trap) showed that 3% Di-amunum phosphate was highly significant attractant to *B. zonata* than other chemical compounds attractants recorded 15.43 flies / trap during two weeks , while there were no significant differences between these other attractants chemical compounds. Mean of weekly captured *B. zonata* for 10 % Urea Fertilizer [CO(NH2)2] recorded 7.56 flies / trap relatively high comparing with other attractants chemical compounds. While 5 % Buminal recorded lower attractants to attract *B. zonata* than other chemical compounds attractants recorded 1.25 flies / trap . The obtained data in Table (4), the using plastic containers (dry trap) indicated that the numbers of attracted fruit fly to it was much lower than the numbers that were attracted to the plastic bottles trap , showed that 10 % Amunum-Acetate was highly significant attractant to *B. zonata* than other chemical compounds attractants recorded 2.78 flies / trap and while there were no significant differences between these other attractants chemical compounds . The second high attractant to attract *B. zonata* , 10 % Urea Fertilizer [CO(NH2)2] recorded 1 fly / trap .

2 .2 . The Mediterranean fruit fly *Ceratitis capitata*:

Data obtained in Table (3) showed that using plastic bottles (liquid trap), the highest mean of weekly captured of *C. Capitata* was recorded for 5 % Buminal

and 3% Di-amunum phosphate gave the same result 1.43 flies / trap , followed by Trimethylamine recoded 1.25 flies / trap , while 10 % Amunum-Acetate , 1 % Dab Fertilizer (H9N2O4P) and 10 % Urea Fertilizer [CO(NH2)2] recorded lower attractants to attract *C. Capitata* , 0.75 , 0.43and 0.31 flies / trap respectively . Data obtained in Table (4) showed that using plastic containers (dry trap), the highest mean of weekly captured of *C. Capitata* was recorded for Trimethylamine recoded 0.71 flies / trap , followed by 10 % Amunum- Acetate , recorded 0.56 flies / trap , while 3% Di-amunum phosphate and 5 % Buminal recorded less of weekly captured of *C. Capitata* 0.37 and 0.28 flies / trap respectively , but 1 % Dab Fertilizer (H9N2O4P) and 10 % Urea Fertilizer [CO(NH2)2] recorded the lower attractants to attract *C. Capitata* . In this investigation , the females number of *B. zonata* and *C. capitata* were more attracted than male flies to tested different chemical compounds. These results are agreement with those obtained by Saafan (2005), Afia (2007) and Moustafa and Ghanim (2008) reported the number of *B. zonata* and *C. capitata* females attracted to food attractants are significantly higher than males, also Sameh (2009) mentioned that all of the tested preparations were attracted *B. zonata* and *C. capitata* females with a significantly high numbers in comparison to males. However, Makkar *et al.* (2017) mentioned that females of *C. capitata* were more attracted than males in all treatments , also Ben Jemaa *et al.* (2010) found that the percentage of captured *C. capitata* females was significantly higher than that of males in mandarin and washengton navel orange orchards. The present data here showed that the number of attracted flies to plastic bottles traps (liquid trap) was highest compared to plastic containers (dry trap). These results are agreement with El-Abbassi *et al*. (2014) found that the performance of tested attractant materials in a liquid form was better than the attractant materials prepared in a dry form. Also in the present investigation , in peach orchard Di-amunum phosphate recorded the highest mean numbers of weekly captured *C. capitata* was 111.86 flies / trap in the plastic bottles (liquid trap) .While 5 % Buminal recorded the highest attractants to attract female *C. capitata* captured 127.12 females / trap weekly compared with 47.87 males . While in mango orchard in the same traps (plastic bottles) 3 %Di-amunum phosphate recorded the highest attractants to attract *B. zonata* recorded 15.43 flies / trap weekly , in the same trend 3 % Di-amunum phosphate and 5 % Buminal recorded the highest attractants to attract *C. capitata* recorded 1.43 flies / trap weekly . But 10 % Urea Fertilizer [CO(NH2)2] recorded the highest attractants to attract female *B. zonata* recorded 14.47 females / trap weekly compared with 0.25 males. However, 5 % Buminal recorded the highest attractants to attract female *C. capitata* captured 2 females / trap weekly compared with 0.87 males . These results are agreement with those obtained by Hanafy *et al.* (2001) and Amin (2003) found that Di-ammonium phosphate was the best compound for attracting PFF in comparison with the other tested compounds. Also Amin (2003), Saafan (2005) and Afia (2007) found that Buminal was the best food attractant used in attracting fruit flies *B. zonata* and *C. capitata*.

Table (1): Mean number of fruit flies *Bactrocera zonata* and *Ceratitis capitata* captured in the plastic bottles infesting peach orchard during the period from 21st of May and 18th of June 2018.

Date	Mean No. of Flies									
	PFF					MFF				
Treatments	after one week		two weeks		Mean	after one week		two weeks		Mean
	♂	♀	♂	♀		♂	♀	♂	♀	
10%Ammon. acetate	0	0	0	0	0	101.5	54.75	73.25	132.5	90.5
10 % Urea	0	0	0	0	0	12.25	24	9.25	61.75	26.81
TMA	0	0	0	0	0	97.75	55.75	15.75	105.75	68.75
1 % Dab	0	0.25	0	0	0.06	81.5	56.5	33.25	142	78.31
3%Di-ammo. phos.	0	0	0	0	0	145.25	66	64.25	171.25	111.7
5%Buminal	0	0	0	0	0	48	55.75	47.75	198.5	87.5
Mean	0	0.04	0	0	0.01	81	52.13	40.58	135.33	
L S D SEX						24.5				
L S D Treat.						42.46				

PFF : *Bactrocera zonata* , MFF : *Ceratitis capitata*, 10%Ammon. Acetate: 10 % Amunum- Acetate, 10 % Urea: 10 % Urea Fertilizer, TMA: Trimethylamine, , 1 % Dab : 1 % Dab Fertilizer, 3%Di-ammo. phos.: 3 % Di-amunum phosphate

Table (2): Mean number of fruit flies Bactrocera zonata and Ceratitis capitata captured in the plastic container infesting peach orchard during the period from 21st of May and 18th of June 2018.

Date	Mean No. of Flies																		
	PFF									MFF									
	after one week		two weeks		three weeks		four weeks		Mean	after one week		two weeks		three weeks		four weeks		Mean
Treatments	♂	♀	♂	♀	♂	♀	♂	♀		♂	♀	♂	♀	♂	♀	♂	♀	
10%Ammon. acetate	0	0	0	0	0	0	0	0	0	6.25	5.5	2.5	6	1.75	3.75	0	0.5	3.28
10 % Urea	0	0	0	0	0	0	0	0	0	2.5	5.5	1	4.75	1.5	2.25	0	1.25	2.35
TMA	0	0	0	0	0	0	0	0	0	7.25	8.25	6	6.25	1.5	8.5	0	2	4.97
1 % Dab	0	0	0	0	0	0	0	0	0	1.75	1.25	0.75	1.5	1.25	1.75	0	0.5	1.09
3%Di-ammo. phos.	0	0	0	0	0	0	0	0	0	2.75	8	3	5.5	0.5	9	0.25	1.25	3.78
5%BuMinal	0	0	0	0	0	0	0	0	0	2	8.5	1.5	6.25	0.25	5	0.25	0.5	3.03
Mean										3.75	6.16	2.46	5.05	1.12	5.04	0.08	1	
L S D SEX										0.654								
L S D Treat.										1.133								

PFF : *Bactrocera zonata* , MFF : *Ceratitis capitata*, 10%Ammon. Acetate: 10 % Amunum- Acetate, 10 % Urea: 10 % Urea Fertilizer, TMA: Trimethylamine, , 1 % Dab : 1 % Dab Fertilizer, 3%Di-ammo. phos.: 3 % Di-amunum phosphate

Table (3): Mean number of fruit flies *Bactrocera zonata* and *Ceratitis capitata* captured in the plastic container infesting mango orchard during the period from 12th of July and 9th of August 2018.

Date	Mean No. of Flies									
	PFF					MFF				
	after one week		two weeks		Mean	after one week		two weeks		Mean
Treatments	♂	♀	♂	♀		♂	♀	♂	♀	
10%Ammon. acetate	0.5	4.75	0.5	6.25	3	0.5	2	0.25	0.25	0.75
10 % Urea	0.5	4.5	0	25.25	7.56	0	1.25	0	0	0.31
TMA	2.25	6	0.25	2.5	2.75	1.25	3.75	0	0	1.25
1 % Dab	2	4.75	1.25	8.75	4.18	0.25	0.25	1.25	0	0.43
3%Di-ammo. phos.	8.5	24.5	4.75	24	15.43	1.5	2.25	0.75	1.25	1.43
5%BuMinal	1.75	3	0.25	0	1.25	1.5	4	0.25	0	1.43
Mean	2.58	7.91	1.16	11.12		0.83	2.25	0.41	0.25	
L S D SEX	4.92					0.75				
L S D Treat.	8.53					1.31				

PFF : *Bactrocera zonata* , MFF : *Ceratitis capitata*, 10%Ammon. Acetate: 10 % Amunum- Acetate, 10 % Urea: 10 % Urea Fertilizer, TMA: Trimethylamine, , 1 % Dab : 1 % Dab Fertilizer, 3%Di-ammo. phos.: 3 % Di-amunum phosphate

Table (4): Mean number of fruit flies *Bactrocera zonata* and *Ceratitis capitata* captured in the plastic containers infesting mango orchard during the period from 12 July to 9 August 2018.

Date	Mean No. of Flies																		
	PFF									MFF									
	after one week		two weeks		three weeks		four weeks		Mean	after one week		two weeks		three weeks		four weeks		Mean	
Treatments	♂	♀	♂	♀	♂	♀	♂	♀		♂	♀	♂	♀	♂	♀	♂	♀		
10%Ammon. Acetate	4.75	3.75	4.75	2.75	2.5	1.75	0.25	1.75	2.78	1.25	1.25	0.25	0.75	0	1	0	0	0.56	
10 % Urea	5.5	0.75	0.5	0.75	0.25	0.25	0	0	1	0	0	0	0	0	0	0	0	0	
TMA	0.75	0.5	0.25	0.25	0.25	0.25	0	0	0.28	1.75	1	0	1	0.75	1.25	0	0	0.71	
1 % Dab	2	0	0.75	0	0.25	0.5	0	0.25	0.46	0	0.25	0	0.25	0	0	0	0	0.06	
3%Di-ammo. phos.	0.25	0	1.25	0.5	0.5	1	0.25	0.5	0.53	1.75	1	0	0	0	0.25	0	0	0.37	
5%BuMinal	1.25	0.5	1.25	0.75	0.5	0.5	0.5	0	0.65	1	0.75	0.25	0.25	0	0	0	0	0.28	
Mean	2.41	0.91	1.45	0.83	0.7	0.7	0.16	0.41		0.95	0.71	0.08	0.37	0.25	0.41	0	0		
L S D SEX	0.61									0.2									
L S D Treat.	1.07									0.35									

PFF : *Bactrocera zonata* , MFF : *Ceratitis capitata*, 10%Ammon. Acetate: 10 % Amunum- Acetate, 10 % Urea: 10 % Urea Fertilizer, TMA: Trimethylamine, , 1 % Dab : 1 % Dab Fertilizer, 3%Di-ammo. phos.: 3 % Di-amunum phosphate

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Survey and distribution of weasels (Carnivora: Mustelidae) in popular and rural human habitats in Alexandria and Sohag Governorates

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Abstract:

The Egyptian weasel *Mustela subpalmata* Hemprich and Ehrenberg **(**Carnivora**:** Mustelidae) is omnivorous and includes vegetables and fruit in its diet as well as waste human food and animals. Under different human habitats in Alexandria and Sohag Governorates, Egyptian weasel , found in houses and streets of popular and rural habitats. The male rate was about 60% according to months, habitats and location. The abundance of weasels in Alexandria houses and streets more than Suhag's in both habitats. The higher index of weasels was recorded in rural human habitats and more disturbances could be found in the streets of both Governorates. In the winter months, weasels was limited in the streets while limited in houses during autumn months in rural habitats at both Governorates. In popular human habitats, weasel index limited to autumn months in streets and houses at the two Governorates. With high temperature months, weasels index increased earlier in Sohag compared with Alexandria and reached its peak in streets in summer and spring months in houses. Based on the previous results we can expect the disturbance weasels induces during high temperature months and could development suitable control program to get rid of weasels according to their abundance and distribution in houses and streets of rural and popular human habitats.

Introduction

The Egyptian weasel *Mustela subpalmata* Hemprich and Ehrenberg (Carnivora: Mustelidae) is a small mustelid with a distribution restricted to the lower Nile Valley and the Nile Delta. Traditionally considered a subspecies of the least weasel (*M. nivalis* L.), it is currently recognized as a separate species based on morphology (Rodrigues *et al.*, 2016) .The Egyptian weasel has short legs, a small head and small ears. Its tail is long and thin. The description of Egyptian weasels was reported by McDonald and Hoffmann (2016). The atmospheric factors (temperature, humidity, wind and rain) and level of environmental hygiene an important role in the presence and spread of weasels (Samson and Raymoned ,1995) and Zalewski ,2000). Brandt and Lambin

(2005) studied the rhythm of activity of weasels in summer and mentioned that this is the prevailing rhythm found for weasels under natural conditions. They also found that, climatic condition influenced weasels' activity, with weasels decreasing activity under rainy conditions. In certain habitat, by pass levels of weasels' activity increased with increasing field vole *Microtus agrestis* (L.) (Rodentia : Cricetidae) density. Sundell *et al.* (2000) observed that the least weasel activity was highest at vole densities of 8 voles per ha. Moreover, declined at densities of 12 voles per ha., probably because hunting become more efficient the weasels belongs to order Cornivora (Canidae and Mustelidae) is distributed worldwide contains, more than 200 species. In particular Mustelidae consist of 64 extant species and for this reason, they are considered one of the most highly differently group of mammals (Hosoda *et al.,*1993).

In Egypt (1980) Osborn and Helmy recorded weasels, *Mustela nivalis subpalmata* in Cairo, Alexandria and Fayoum Governorates, now the weasels can be seen easily in many habitats with its spread and increase numbers began to cause a lot of problems carrying serious human and animal diseases and feeding on small domestic animals and birds causing economic losses for home owners and breeders.

The aim of this work is to study the effect of temperature on weasel numbers in Alexandria and Sohag Governorates as well as the movement and survey of weasels in streets and houses in the popular and rural human habitats during year months

Materials and methods

Under the climatic conditions of the tested area in Alexandria 31.2°N 29.91667°E and Sohag 26°33'38"N, 31°41'30"E Governorates, the abundance of weasels was recorded monthly for two consecutive years (2017 and 2018) inside houses and on the streets. Relative abundance of weasels was estimated as well as weasel's index (average number of weasels/trap/night) each year.

1. Tested area:

According to the different climatic conditions (average temperature, wind speed and sunny or cloudy days) between Alexandria and Sohag Governorates, poplar and rural human habitats were chosen in each Governorate.

1.1. Popular human area:

To areas were chosen: El-Agamy 31° 5' 55.8132" N, 29° 45' 59.9976" E at Alexandria and Nag Abu Shagara 26° 32' 59.9964" N, 31° 42' 0.0036" E at Sohag Governorates. These popular areas consist of unregulated residential houses including shops selling poultry, meat and vegetables. The area are not crowded with little transportation and almost quiet at night. Poultry birds are raised on resident roofs residents, with much complains of bird's losses due to weasels attacks.

1.2. Rural habitats:

Two areas were chosen: El Ameriya 31° 6′ 16.34″ N, 29° 45′ 58.42″ E at Alexandria and in El-Maragha 26°40'51.2"N 31°37'45.5"E at Sohag Governorates. Rural habitats are informal dwellings close to canals, water banks and agriculture lands. There are nearby barns for raising cattles and poultries. Most of the streets are dusty with garbage scattered everywhere. Residents complain of bird losses due to weasels attack.

2. Survey experiment:

Three separated plots (5000 m^2, 50*100 m) were selected for each tested area at each habitat, approximately 25000 m^2. A hundred traps, for each plot/area were baited every evening for three consecutive days monthly. The mechanical traps used (40 X 15 X 20 cm) are divided into to parts. The first part represent one – third of the trap for live baits (Figure,1) and the rest were baited with liver as attractive bait. Fifty traps (five traps at each house) were distributed in ten houses on the roofs and skylights. Another fifty traps were

distributed in the streets near by crash and garbage collection box. Every morning traps were examined and cached weasels weighted, sexed and identified according to sex using the key of weasels by Osborn and Helmy (1980). Weasel index was recorded monthly and population densities were calculated for different seasons.

Results and Discussion

During 2017–2018 in all tested area, the trapped weasels were Egyptian weasel *M. subpalmatia* according to Wozencraft (2005). Until recently, the Egyptian weasel considered a subspecies and named *Mustela nivalis subpalmata* (Osborn and Helmy, 1980). It has been treated as its own species *Mustela subpalmatia*in the literature and was given species status. The weasel index and distribution in rural and popular human habitats of Sohag and Alexandria Governorates recorded monthly at houses and streets in Tables (1 and 2).

1. Distribution of weasel numbers in the streets and houses of the tested human habitats of Sohag Governorate:

Data in Table (1) showed that, the average weasel numbers were (115.5 and 128.4) and (97.5 and 105.3) animal in Sohag rural and popular areas for 2017 and 2018, respectively. In the first year, the weasel index at rural area was highest in June and August (10 animals) in the streets and 8.6 animals in June, in houses. This index recorded the lowest numbers in street (1-6 animals) and 1 animal in houses during June and December. In the second year, the indexes were higher in June (10.3 and 8.6 animals) and the lower in December (1 and 1.6 animals) at the street and the houses, respectively. During the same two years weasel index was high in popular area (8 and 6 animals) in June at the first year and in the second year were 9 animals in June and 6 animals in July at houses. The lowest number of weasels were 1 and 1.3 animals in December in the first year, while reached to 1 animal in June and 1.6 animal in November – December and June in the second year at street and houses, respectively.

2. Distribution of weasel numbers in the streets and houses of the tested human habitats in Alexandria Governorate:

Data in Table (2) exhibit that, at Alexandria Governorate in rural streets and houses, weasel indexes ranged between (1.6–11animals) and (2–10 animals) in the first year, respectively. Weasel indexes ranged between (1 – 11.6 animals) in December and July. At streets and between (2.3 animals in October to 9 animals in July, at houses during the second year. Another poplar area, during the first year, weasels indexes ranged between zero animal in December and 12 animals in June and July at streets, while recorded 1.6 animals in December and 6.3 animals in June at houses. Weasel in second year recorded 1 animal in December and 12.3 animals in August at street and ranged between 2 animals in January and 6.3 animals in April and May in houses. The total average of weasels during the two year were (138.3 and 117.6) and (134.7 and 121.9 animals) in rural and poplar areas, respectively.

3. Average seasonal number of weasel in houses and streets:

The average numbers of weasels' indexes during the four seasons summarized in Table (3). Data cleared that in poplar habitats of Alexandria streets, weasels' were 30.45 in summer with sex ratio of 37.2%, and 17.45 in houses with sex ratio of 46%, during Spring, In Sohag Governorate, the average number of weasels were 14.6 with 50.1 % sex ratio and 20.8 with sex ratio 46.2%, during Spring at houses and street, respectively. The peak of weasels number in rural area were 23.1 with sex ratio 38.1% in spring and 29.75 with sex ratio 40.2% in summer at Alexandria

houses and street, respectively. They also recorded 19.4 and 25.95 with sex ratio 46.4% and 50% for the same season at Sohag houses and street, respectively. The average number decreased in poplar area and reached 15.95, 9.3 and 8.9 in Alexandria house compare with 13.95, 8.75 and 6.2 in Sohag houses during summer, winter and autumn, respectively. The weasels in poplar streets were 25.15, 6.15 and 6.4 in Alexandria during spring, autumn and winter, respectively compare with 18.2, 8.8 and 8.45 in Sohag during summer, winter and autumn, respectively. McDonald and Hoffman (2008) reported that, the range of *M. subpalmata* is restricted to the lower Nile valley from Alexandria to the west Port Said in the east and from the delta south to Beni-Suef. Basuony *et al.* (2010) reported that weasels were trapped from the city of Aswan (650 K.M. south of Beni-suef). In all studied areas, the results in Figure (1) showed that the highest number of weasels recorded in summer where the mating season and the increasing rate of weasels in Sohag were less than Alexandria Governorate due to the thermal stress on animals as results of high temperature, which affects the fertility and production. The temperature degree in Sohag is higher than Alexandria by 10 ^{0}c during different months. The weasels record highest number in houses where the mating season begins and the construction of nests in safe place. Brown and Lasiewski (1972) found that metabolism of weasels to be inversely related to ambient temperature. Zub *et al.* (2013) reported that high ambient temperatures (T_a) was related in a "hump-shaped" (i.e. convex) manner to activity time (AT) , daily energy expenditure (DEE), resting metabolic rate (RMR) and metabolic scope (the ratio of DEE to RMR). This results supported the HDL hypothesis because in response to warm ambient high temperatures T_as male weasels reduced their AT, DEE and RMR.

Although the activity and energy expenditure of large endotherms are most likely to be constrained in response to warm T_{as} because they are less able to dissipate heat. Our results suggested that small endotherms might also experience constraints consistent with the HDL hypothesis. In the houses and streets of Alexandria the number of weasels decreased significantly with the month of September to December and this decline may be due to strong winds and rainfall that hinder the weasel number (Figure,2). The rate of decline in the street was higher than the houses then gradually adapted numbers with the improvement of the atmosphere and beginning of biological activity in March to August. Eisa (2001) reported that the highest number of weasels recorded during July in Cairo and June in Giza but the lowest number in January of both governorates

In Sohag Governorate, the number of weasels started in the gradual titration early from February to August. Weasel numbers in Alexandria were highest compared with Sohag due to the thermal stress to animals in Sohag. Regarding of the achieved results, it was clear that the number of weasels in the streets was greater than in the houses in the two Governorates, by means of reproduction and the search for prey. As well, the weasel numbers in the rural areas were more than in the poplar area as a result of the availability of food, rodent, small birds and domestic birds. Moreover, abundance of females in houses were highly compared with street and the male percent more than 60% compare with females in all areas.

It is concluded that the disturbance weasels induces during high temperature months and could development suitable control program to get rid of weasels according to their abundance and distribution in houses and streets of rural and popular human habitats.

Table (1): Average weasel index at Sohag Governorate during 2017 and 2018 in the two tested human habitats.

Months	Poplar habitat				Rural habitat			
	First year		Second year		First year		Second year	
	House	Street	House	Street	House	Street	House	street
Janauary	1.6	1.6	1.6	1	3.3	1.6	3	2.3
Febraury	3	3	4	4	3.3	2.6	2.6	4
March	4	4	3.3	4	4	4	5	3
April	4	5	4	6.3	4.6	4	4	5
May	4	6	5.6	7.3	6	1.3	7	7.3
June	6	8	5.6	9	8.6	10	8.6	10.3
July	5	6.3	6	6.6	8	9.3	7.6	10
August	4.6	5.6	5	7	5.3	10	6	9
Septembe	4	5.3	3.3	5.6	6	6	6.3	7.6
October	3.3	5	3	4.3	5	4	5.6	6.3
November	1.6	4.3	1.6	3	2.3	3.3	3.3	2
December	1.3	1	1.6	2.6	1	2	1.6	1
Total	42.4	55.1	44.6	60.7	57.4	58.1	60.6	67.8
	97.5		105.3		115.5		128.4	

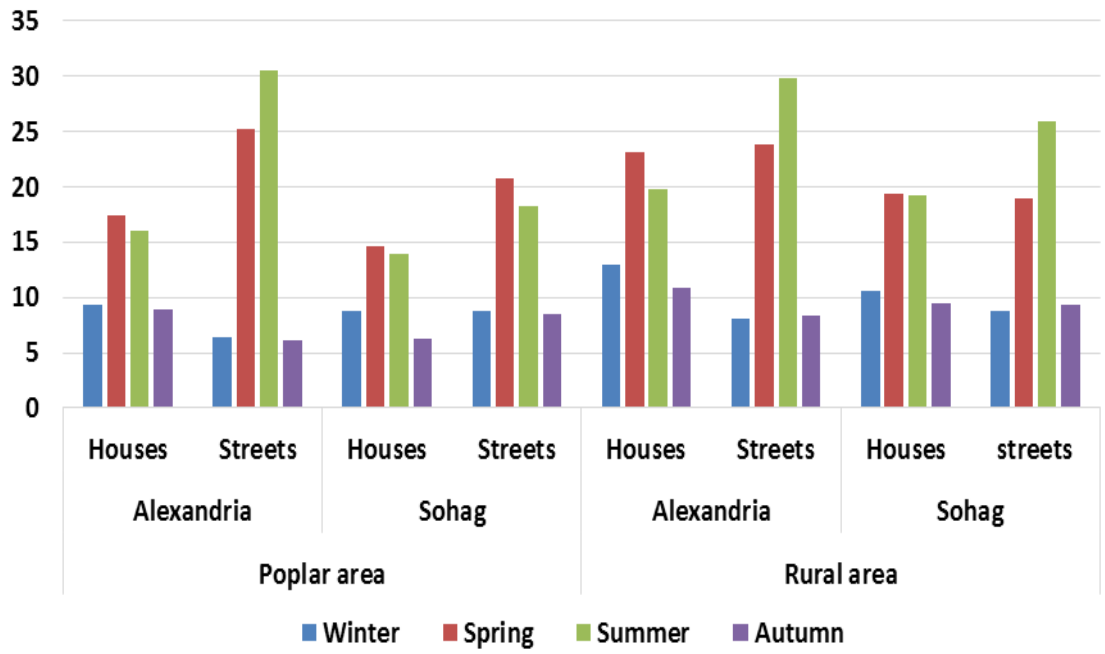

Figure (1) :Survey weasel during during the 4 seasons of the year in tested human habitats/ areas

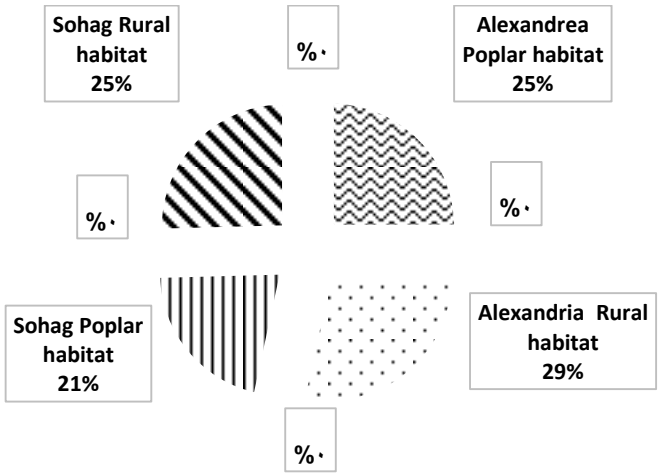

Figure (2): Weasel numbers in different area of Alexandria and Sohag Governorates

Table (2): Average weasel index at Alexandria Governorate during 2017 and 2018 in the two tested human habitats.

	Months				Popular habitat			
	First year		Second year		First year		Second year	
	Houses	Streets	Houses	Streets	Houses	Streets	Houses	Streets
Janauary	3	1.6	2	1.3	4	2.3	3	1.3
Febraury	3.3	1.3	2.3	1.6	4.3	4	3.6	1.3
March	4	4	4	3	5.6	4	5.3	3.3
April	5	4	6.3	5.3	6.3	6.3	6	5
May	5	8	6.3	9	9	10	8	6.6
June	6.3	12	6	12	8.6	9.3	8.3	10.3
July	5.6	12	6	12	10	11	9	11.6
August	6	11.6	6	12.3	7	8	8.6	11.3
September	4	7	4.3	6	2	8	3	9.6
October	3	4	4.6	3.3	2	5	2.3	4
November	3.3	2	3	2	4	2	4	3
December	1.6	0	2.3	1	4	1.6	5.3	1
Total	50.1	67.5	53.1	68.8	66.8	71.5	66.4	68.3
	117.6		121.9		138.3		134.7	

Table (3): Average seasonal numbers and sex ratio of weasels during 2017 and 2018 in tested human habitat .

Months	Poplar habitat								Rural habitat							
	Alexandria				Sohag				Alexandria				Suhag			
	Houses	Sex ratio	Streets	Sex ratio	Houses	Sex ratio	Streets	Sex ratio	Houses	Sex ratio	Streets	Sex ratio	Houses	Sex ratio	Streets	Sex ratio
Winter	9.3	42.1	6.4	40.5	8.75	45.7	8.8	42	12.9	38.4	8.1	36.2	10.6	41.2	8.75	42.3
Spring	17.45	46	25.15	39	14.6	50.1	20.8	46.2	23.1	38.1	23.75	37	19.4	88.2	18.95	36.4
Summer	15.95	50	30.45	37.2	13.95	50	18.2	50	19.8	46.4	29.75	40.2	19.2	51	25.95	50
Autumn	8.9	38.2	6.15	40.3	6.2	40.1	8.45	44.1	10.8	41	8.3	38.8	9.4	46	9.3	42.4
Total	51.6		68.15		43.5		57.9		66.6		69.9		59		62.95	
	59.875				50.7				68.25				60.975			

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Efficacy of predatory phytoseiid mites and biopesticides for controlling *Tetranychus urticae* (Acari: Tetranychidae) infesting *Phaseolus vulgaris*

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Abstract:

The kidney bean *Phaseolus vulgaris* L. is one of the most important economic vegetable crops cultivated in Egypt and many countries of the world as a main source of protein. This study dealt with the efficiency of two predatory phytoseiid mites, *Phytoseiulus persimilis* Athias-Henriot, *Typhlodromips swirskii* (Athias-Henriot) (Acari: Phytoseiidae); fungal pathogen *Beauveria bassiana* (Balsamo) and the biopesticide Abamectin 1.8 EC. + mineral oil Cable against the eggs, immature and adults of *Tetranychus urticae* Koch (Acari: Tetranychidae) infesting two cultivars of both *P. vulgaris* (Hama and Bolista) under net house conditions at Beheira Governorate during 2016 season. Highly reduction percentage was achieved by the predatory mite *P. persimilis,* the biopesticide Abamectin 1.8 EC. + mineral oil Cable and fungal pathogen *B.bassiana* , followed by the other predatory mite *T. swirskii* when compared with the untreated plants. Also, the results indicated that accarine pests are only a part of biological complex of which predacious mites, particularly phytoseiid group, could be of value in checking infestations.

Introduction

The kidney bean *Phaseolus vulgaris* L. is the popularity of the crop originates from the fact that it is relatively easy to produce, lavorful and versatile and a good source of nutrition. According to the report of economic affairs sector, Department of Agricultural Economics, Ministry of Agriculture (2007), the cultivated area was about 73022 feddens with production 330257 tones for green beans and 49639 feddens with production 60794 tones for dry beans in many Governorates for exportation and local consumption. The kidney bean plants are infested by many pests which cause a great damage in both quantity and quality. The spider mite, *Tetranychus urticae* Koch (Acari:

Tetranychidae) and some piercing sucking pests' viz. *Thrips tabaci* (Lindeman) (Thysanoptera: Thripidae), *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae) and the leaf miner *Liriomyza* spp. (Diptera: Agromyzidae) are the majority pests infested kidney bean cultivars (El-Saiedy *et al*., 2012). These pests cause great damage to the plant feeds on the plant sap causing serious damage varying according to the degree of infestations (Habashy, 2000; Iskandar *et al.*, 2002; Mahgoub, 2006; Abd El Gawwad, 2008 and El-Saiedy *et al.,* 2011).

The aforementioned pests have been rapidly developing resistance to a series of pestisids and have recently assumed a new aspect of multiple resistances (Pree *et al.,* 2002). The extensive use of pesticides can promote negative impacts on human health and on ecosystems, besides reducing the number of species and density of natural enemies, developing resistance and increasing production costs. To reduce these problems, it is necessary to reduce the chemical control by replacement of such pesticides by using biocides with releasing predatory mites. Biological control means the control of pests with predators, parasitoids and pathogens. Successful biocontrol can be obtained in many cases (Messelink *et al.,* 2006 and 2008). *Phytoseiulus persimilis* Athias-Henriot and *Typhlodromips swirskii* (Athias-Henriot) (Acari: Phytoseiidae) are a polyphagous predators capable of preying on a number of spider mites (Momen and El-Saway, 1993; Bolckmans *et al*., 2005; Van Houten *et al.,* 2007; Abdallah *et al*., 2012; El-Kholy and El-Saiedy, 2009; Calvo *et al*., 2011; Elmoghazy *et al*., 2011; Dimetry *et al*., 2012; Abdallah *et al*., 2014 and Abou-Awad *et al*., 2017).

The present study aims to evaluate the efficiency of the predatory mits *P. persimilis* and *T. swirskii* as well as the fungus, *Beauveria bassiana* and the biocide Abamectin+ mineral oil Cable on two varieties (Hama and Bolista) of kidney bean plants under controlled net house conditions against the egg, immature stages and adults of *T. urticae* during 2016 plantation season at Behaira Governorate.

Mateials and methods

1. Experimental design:

To study the effect of different types of biocontrol agents, four treatments were carried out on kidney bean plants in Beheira Governorate , using the two predatory mites, *P. persimilis* and *T. swirskii*, the fungus, *B. bassiana* + mineral oil Cable oil, and the biocide Abamectin 1.8EC + mineral oil Cable oil. The aforementioned experimental treatments were compared with untreated plants (control).

Kidney beans (*P. vulgaris*) Hama and Bolista cultivars seeds were planted in the net house on 16th October, 2016. The net house was divided into ten equal plots. Each plot was divided into three separated replicates (each one represented by five rows about 25 m^{-} long). The trail plots were arranged in randomized complete block design for each treatment. All the experimental plots received the standard cultivation practices of that area including organic and mineral fertilization, drip irrigation and mechanical control was applied to remove weeds. Pesticides were avoided entirely.

2. The efficiency of predators mites and fungal pathogen to control *Tetranychus urticae*:

2.1. *Phytossulus persimilis* and *Typhlodrompis swirskii* predators:

2.1.1. Prey culture:

T. urticae, was reared on kidney been plants, *P. vulgaris* planted in plastic pots at the rate of 20 seeds/pot

and put in isolated greenhouse of 2m width, 2m length and 2.5m height covered with a fine mesh (anti-virus) plastic net (500 holes/inch). When bean seedling were 7-10 days old inoculated with leaves infested with *T. urticae* collected from cucumber plants presented from Giza Governorate and when individuals of the *T. urticae* moved to the new foliage of bean plant the dried leaves of cucumber were removed 2 days later. Pots were planted and infested every 3-4 days to provide continuous spider mite production.

2.1.2. Predators rearing:

The phytoseiid predators, *P. persimilis* and *T. swirskii* were reared using methods modified by (McMurtry and Seriven, 1965), large plastic box 25x25x10 cm were used, cotton pad were put in the middle of each box on 2cm thick piece of sponge, leaving a space provided with water as a barrier to prevent predatory mites from escaping. Excised bean leaves highly infested with *T. urticae* were provided every other day as food source. The plastic boxes were kept in an incubator at 25 ±1ºC and 70±5% R.H., water was added daily to maintain suitable moisture for the predators rearing.

2.1.3. Mass rearing of the predatory mites:

Mass rearing was conducted in two separated small greenhouses for each predator with dimension 5m width, 7m length and 2.5m height. Bean plants were cultivated as host plants and infested with two spotted spider mite *T. urticae* as a prey when the population of spider mite increased to suitable population. Then, the predator mites were transferred to each infested bean plants. The various subjects were carried out at average temperature 25-30 ºC and relative humidity 65 ± 10%. When the population of the predator mites increased gradually where the rate of population investigated daily and supported with prey until the populations of the predators become suitable for collecting.

2.1.4. Releasing of the predatory mites:

P. persimilis was released with level 1:10 predators: prey one time, while *T. swirskii* was released with level 1:7 predators: prey two times thought the experimental time (15th of February and 22nd of March). The required population numbers of predatory mite individuals were calculated according to the following formula (EI-Saiedy, 2003):

$$\text{Released number} = \frac{\text{Total no. of } \textbf{\textit{Tetranychus urticae}} \text{ in treatment X predator level}}{\text{Prey level}}$$

The infestation bean leaves with the predatory mite were transferred in an ice-box to the greenhouse and then distributed on infested kidney bean plants except the treatment which kept free from any controlling agents.

2.2. *Beauveria bassiana* spraying:

Also, *B. bassiana* was sprayed three times through the experiment The commercial pathogen compound biofly, *B. bassiana* was used at the rate of 75cc/100 liter of well water (free chlorine). The fungal pathogen and Abamectin were sprayed thrice on 15th of February, 22nd of March and 27th of April 2016).

3. The efficiency of biopesticde to control *Tetranychus urticae*:

Abamectin 1.8 EC.: Formulation Type: Emulsifiable concentrate derived from a soil actinomycete (*Streptomyces avermitilis*) and has strong insecticidal, nematicidal and acaricidal activity (Wang and Wu, 2007) was also used at the rate of 100cc + 250cc mineral oil Cable 2/100 liter of water. The fungal pathogen and Abamectin were sprayed thrice on 15th of February, 22nd of March and 27th of April 2016).

4. **Sampling Procedure:**

For estimating the effect of four treatments of two cultivars of kidney bean plants, samples of 30 leaves from the three replicates of each treatment were randomly picked up just before release were mite population were counted as pre-count. This procedure was repeated as post-counts. Samples were also obtained from adjacent non-released treatment (control) at weekly intervals from the time of application until the end of this experiment. Each sample was kept in a tightly closed paper bag and transferred to the laboratory for inspection using a stereomicroscope. Number of eggs, immatures (larvae and nymphs) and adult stages of *T. urticae* were counted and recorded for each treatment.

5. Statistical analysis:

The percentages of reduction in the number of the previously mentioned pests were calculated by using the reduction equation of Henderson and Tilton (1955). The statistical analyses (ANOVA) of the obtained data were performed by using SAS program (SAS Institute, 2003) which runs under WIN. Also the difference between means was conducted by using Duncan's (Duncan, 1955) multiple range tests in this program.

Results and discussion

The reducing effect of the two phytoseiid predators *P. persimilis* and *T. swirskii*, fungal pathogen *B. bassiana* and biocide Abamectin+ mineral oil Cable against *T. urticae* eggs, immature and adults infested two beans cultivars (Hama and Bolista) were evaluated during 2016 season.

1. Egg stage:

Data organized in Table (1) indicated that the examined sample of kidney bean leaves collected just before applying the four controlling agents (*P. persimilis* and *T. swirskii*, *B. bassiana* and Abamectin+ mineral oil Cable) (pre-count) were harbored with averages 9.80, 9.70, 9.75 and 9.80 eggs/leaf of *T. urticae* , respectively and 9.90 for untreated Bolista leaves while Hama leaves had 21.5, 21.4, 21.38, 21.62 and 21.35 for the previously mentioned treatments, respectively .

After one week, the obtained results indicated reductions in number of the target pest in applied four treatments on both cultivars, as *T. urticae* eggs averaged 6.3, 8.1, 4.40 and 0.10 eggs/leaf, on the examined Bolista plants and 16.35, 23.5, 3.56 and 0.9 on Hama cultivar of the *P. prisimilis*, *T. swirskii*, *B. bassiana* and Abamectin + mineral oil Cable treatments, respectively. While it averaged 13.5 and 24.35 eggs/leaf for the control treatment on the two respectively arranged cultivars. The corresponding reduction percent in mite counts after one week from application were 68.07, 35.47, 80.52 and 88.27%, on Bolista and 49.26, 18.57, 85.14 and 95.62 % on Hama variety, respectively.

By increasing the time after application, the releasing of *P. persimilis* showed reduction of eggs to reach zero egg/leaf with general seasonal average 2.67 and 11.42 eggs/inch^2, while for the second treatment it reached after ten weeks of release with seasonal average 11.70 and 44.68 eggs/inch^2 on both Bolista and hama, respectively. Also, in the other two treatments (*B. bassiana* and Abamectin + mineral oil Cable) the infestations with the mite eggs were fluctuated until the end of the season during the 17 week of evaluation with allover mean 18.64 and 4.61 eggs/ inch^2 on Bolista and 9.72 and 6.08 for Hama , respectively.

Statistical analysis of the reduction percentages (Table, 2) showed that, controlling of *T. urticae* eggs on Bolista and Hama bean plants with the *P. persimilis,* Abamectin

+mineral oil Cable and *B. bassiana* led to lower infestation rate with reduction percentages (88.98, 89.51 and 84.04% and 80.07, 94.25 and 89.13%, respectively), followed significantly by the application of *T. swriskii* with percentages 66.89 and 51.52% of both the two planted cultivars. (L.S.D = 10.78 and 11.31).

These data proved that releasing of *P. persimilis* at level 1:10, *B. bassiana* and Abamectin +mineral oil Cable oil gave the best results of controlling of *T. urticae* eggs infesting the tested plants in net house, as statistically, this treatment occupied the lowest degree of infestation.

Table (1): Mean numbers of *Tetranychus urticae* eggs / leaf on two kidney bean varitites under net house conditions affected by releasing two predatory mites, *Beauveria bassiana* and the biocide abamactin +mineral oil cable spraying at Beheira Governorate during 2016 season.

Varitites	Bolista					Hama				
Inspections date / Treatment	*P.prisimilis*	*T. swirskii*	*B. bassiana*	Abamactin +Mineral oil cable	control	*P.prisimilis*	*T. swirskii*	*B. bassiana*	Abamactin +Mineral oil cable	control
Feb.,15th *	9.90	9.70	9.75	9.80	9.80	21.50	21.40	21.38	21.62	21.35
22nd	6.30	8.10	4.40	0.10	13.50	16.35	23.50	3.56	0.90	24.35
28th	5.20	10.40	6.40	1.90	16.20	15.40	24.60	4.48	1.34	30.14
Mar.,7th	5.30	11.60	9.40	4.20	18.10	17.20	30.60	8.43	3.11	37.11
15th	4.80	10.40	12.60	8.20	20.30	16.35	31.50	12.40	7.90	44.90
22nd	4.90	12.3**	7.10**	0.30**	24.80	17.23	32.00	17.30**	11.34**	58.70
29th	3.30	9.90	9.40	1.40	29.10	16.40	33.20	2.95	1.11	70.50
Apr.,6th	2.40	13.10	12.20	3.20	33.60	18.32	36.80	5.33	4.33	83.40
13th	1.20	11.60	14.60	5.20	37.11	16.30	34.90	7.60	6.20	95.30
20th	1.00	12.50	17.70	7.80	40.90	14.90	43.11	10.33	7.18	115.50
27th	0.80	13.90	23.50**	11.10**	50.40	13.80	48.10	16.80**	11.35**	142.60
May,4th	0.00	14.80	20.00	0.30	76.30	7.40	50.11	3.00	0.84	173.00
11th	0.00	11.60	26.70	0.20	98.00	2.18	52.00	5.70	1.35	194.70
18th	0.00	9.20	31.00	3.50	110.00	0.85	78.30	8.80	3.14	204.50
25th	0.00	11.80	33.90	4.20	115.90	0.00	70.33	1.90	5.42	210.70
Jun, 2nd	0.00	14.60	37.60	7.10	123.60	0.00	74.00	14.70	7.18	217.50
10th	0.00	13.20	40.60	9.90	118.90	0.00	75.14	20.60	9.11	234.60
Average ± SE	2.67± 0.72 C	11.70± 0.46 BC	18.64±2.81 B	4.61±0.91 C	55.12±10.18 A	11.42±1.87 C	44.68±4.65 B	9.72±1.55 C	6.08±1.30 C	115.23±18.61 A
F =	20..18					28.41				
L.S.D. =	13.4					24.39				

Values singed by the same letter of the same varity are not significantly different at alpha = 0.05 % level.
**Re-spray

Table (2): Reduction persentages of *Tetranychus urticae* eggs / leaf on two kidney bean varitites under net hous conditions affected by releasing two predatory mites, *Beauveria bassiana*, and the biocide abamactin +mineral oil cable spraying at Beheira Governorate during 2016 season.

Varitites	*Bolista*				*Hama*			
Inspections date / Treatment	*P.prisimilis*	*T. swirskii*	*B. bassiana*	Abamactin +Mineral oil cable	*P.prisimilis*	*T. swirskii*	*B. bassiana*	Abamactin +Mineral oil cable
Feb.,22nd	53.52	39.02	89.94	98.96	33.32	3.72	85.38	96.36
28th	68.07	35.47	80.52	88.27	49.26	18.57	85.14	95.62
Mar.,7th	70.85	35.22	63.61	76.90	53.97	17.74	77.28	91.74
15th	76.35	48.19	49.84	59.76	63.84	30.01	72.38	82.65
22nd	80.32	49.84**	94.93 **	98.99 **	70.85	45.61	70.53 **	80.95 **
29th	88.57	65.78	89.80	95.25	76.90	53.02	95.82	98.45
Apr.,6rh	93.03	60.57	84.73	90.56	78.19	55.98	93.61	94.88
13th	96.83	68.39	80.53	85.92	83.02	63.46	92.03	93.58
20th	97.69	69.02	75.40	81.03	87.19	62.76	91.06	93.87
27th	98.42	72.11	70.85**	77.89**	90.39	66.35	88.22 **	92.15 **
May,4th	100.00	80.38	98.15	99.65	95.75	71.10	98.27	99.52
11th	100.00	88.07	96.78	99.82	98.89	73.35	97.07	99.32
18th	100.00	91.56	95.52	96.86	99.59	61.80	95.70	98.49
25th	100.00	89.70	94.05	96.40	100.00	66.70	99.10	97.46
Jun, 2nd	100.00	88.05	92.57	94.23	100.00	66.06	93.24	96.74
10th	100.00	88.81	87.48	91.68	100.00	68.05	91.22	96.17
Average	88.98 A	66.89 B	84.04 A	89.51 A	80.07 B	51.52 C	89.13 AB	94.25 A
F =	7.74				22.76			
L.S.D. = 10.28	10.78				11.31			

Values singed by the same letter of the same varity are not significantly different at alpha = 0.05 % level.
**Re-spray

2. Immature stages:

As previously indicated in case of *T. urticae* eggs, the relative population density of immature stages infesting the two bean varieties was also affected, significantly, with the application of the tested control agents during experimental time (Tables, 3 and 4).

The differences in seasonal mean counts of immature stages infestation of *T. urticae* to bean leaves were significantly higher on the untreated plants as it harbored the highest mean number of immature stages (42.37 and 97.34 immature/inch^2 of Bolista and Hama, respectively), being differ from which controlled with *T. swriskii* which showed moderate immature infestation (16.62 and 10.53 immature/inch^2 of both varieties), the leaves of other three remaining applications, *P. persimilis* , *B. bassiana* and Abamectin_+mineral oil Cable were subjected to the lowest number of *T. urticae* immature stages

(3.06, 6.56 and 4.62 immature/inch^{2}on Bolista variety and (10.69, 10.53 and 6.45 immature/inch^2 on Hama leaves, respectively. (L.S.D. =9.61 and 20.35)

As it obvious from Table (4), the general mean in reduction percentages in *T. urticae* immature due to releasing of *P. persimilis, B. bassiana* and Abamectin +mineral oil Cable gave the highest mean percentage of reduction (85.44, 81.40 and 86.47% for Bolista and 79.59, 87.54 and 86.72% with Hama cultivar, respectively) followed significantly by the treatment of *T. swriskii* (49.28 and 54.17%) being significantly different from the previously mentioned three treatments on the two cultivated varieties. (L.S.D. values were 11.57 and 11.94, respectively).

Table (3): Mean numbers of *Tetranychus urticae* immature / leaf on two kidney bean varitites under net house conditions affected by releasing two predatory mites, *Beauveria bassiana* and the biocide abamactin +mineral oil Cable spraying at Beheira Governorate during 2016 season.

Varitites	Bolista					Hama				
Inspections date / Treatment	*P.prisimilis*	*T. swirskii*	*B. bassiana*	Abamactin +Mineral oil cable	control	*P.prisimilis*	*T. swirskii*	*B. bassiana*	Abamactin +Mineral oil cable	control
Feb.,15th	9.93	9.43	9.00	9.17	9.90	18.52	18.48	18.43	18.50	18.56
22nd	6.35	9.38	1.10	0.32	11.10	15.40	19.35	4.32	1.35	23.70
28th	7.35	10.30	3.40	1.50	14.13	16.32	20.18	6.35	3.11	28.71
Mar.,7th	5.37	10.40	6.50	3.50	17.18	13.56	26.18	8.33	5.22	34.30
15th	6.70	12.11	8.34	7.44	19.90	12.04	28.14	14.35	9.18	41.40
22nd	4.40	13.15**	1.25**	0.20 **	21.14	17.30	30.18	2.84**	1.09**	50.14
29th	3.86	15.13	1.46	11.30	24.90	15.20	32.18	4.58	1.48	62.13
Apr.,6rh	3.60	14.30	3.15	2.65	30.80	13.65	30.34	7.14	4.26	70.40
13th	2.14	16.60	7.40	3.50	34.70	14.90	34.11	9.18	7.00	85.13
20th	1.35	17.70	9.50	6.35	38.60	16.31	31.11	15.33	10.53	100.13
27th	0.95	18.70	13.80**	8.90**	41.50	17.23	36.30	3.11**	0.48**	115.90
May,4th	0.00	20.11	1.90	0.14	48.90	9.23	40.35	5.22	1.18	123.40
11th	0.00	19.50	3.70	1.80	53.80	2.14	45.35	7.18	2.00	145.90
18th	0.00	20.60	5.60	2.35	68.30	0.00	50.13	11.10	4.58	160.80
25th	0.00	23.70	8.50	3.90	78.50	0.00	48.50	16.38	8.13	175.13
Jun, 2nd	0.00	24.80	11.60	6.45	95.30	0.00	54.13	20.13	14.52	200.14
10th	0.00	26.70	15.30	9.13	111.60	0.00	60.14	25.00	17.35	218.90
Average ± SE	3.06 ±0.77 C	16.62 ±1.33 B	6.56 ±1.07 C	4.62 ±0.87 C	42.37 ±7.35 A	10.69 ± 1.74 C	35.6 ± 3.01 B	10.53 ± 1.60 C	6.45 ± 1.41 C	97.34 ± 15.65 A
F	23.02					27.96				
L.S.D.	9.61					20.35				

Values singed by the same letter of the same varity are not significantly different at alpha = 0.05 % level. **Re-spray

Table (4): Reduction persentages of *Tetranychus urticae* immature /inch2 on two kidney bean varitites under net hous conditions affected by releasing two predatory mites, *Beauveria bassiana* and the biocide abamactin +mineral oil Cable spraying at Beheira Governorate during 2016 season.

Varitites	*Bolista*				*Hama*			
Inspections date / Treatment	*P.prisimilis*	*T. swirskii*	*B. bassiana*	Abamactin +Mineral oil cable	*P.prisimilis*	*T. swirskii*	*B. bassiana*	Abamactin +Mineral oil cable
Feb.,15th	43.08	11.47	89.10	96.90	34.99	18.09	81.66	94.84
22nd	48.24	23.63	73.53	88.58	43.13	29.48	77.75	90.19
28th	68.90	36.58	58.38	78.08	60.45	23.43	75.57	86.22
Mar.,7th	66.50	36.25	53.90	59.77	70.90	31.81	65.13	79.92
15th	79.29	34.83**	93.50 **	98.98 **	65.48	39.61	94.30 **	98.03 **
22nd	84.58	36.34	93.55	51.17	75.52	48.04	92.58	97.84
29th	88.37	51.36	88.75	90.74	80.60	56.76	89.80	94.52
Apr.,6rh	93.86	49.88	76.54	89.15	82.49	59.80	89.15	92.56
13th	96.52	51.96	72.93	82.30	83.70	68.83	84.60	79.63
20th	97.72	52.79	63.42 **	76.92 **	85.13	68.58	97.30 **	54.32 **
27th	100.00	56.92	95.73	99.69	92.52	67.20	95.74	41.17
May,4th	100.00	62.03	92.43	96.40	98.53	68.82	95.05	98.76
11th	100.00	68.40	90.98	96.30	100.00	68.72	93.06	97.42
18th	100.00	68.37	88.09	94.65	100.00	72.22	90.59	95.80
25th	100.00	72.74	86.61	92.72	100.00	72.87	89.88	93.43
Jun, 2nd	100.00	74.94	84.92	91.20	100.00	72.44	88.51	92.82
10th	85.44 A	49.28 B	81.40 A	86.47 A	79.59 A	54.17 B	87.54 A	86.72 A
F	18.76				13.73			
L.S.D.	11.57				11.94			

Values singed by the same letter of the same varity are not significantly different at alpha = 0.05 % level.
**Re-spray

3.Adult stage:

Efficacy of *B. bassiana* and Abamectin + mineral oil Cable and the two predacious mites (*P. persimilis* and *T. swriskii*) against the population density of the adults of *T. urticae* infesting leaves of two bean cultivars are arranged in Tables (5 and 6). Revealed data clear that, the examined bean leaves before the application of the previous controlling agents against the adults as pre-count ranged between 3.5 and 3.6 individuals/ inch^2 on Bolista and 7.00 and 7.34 on Hama. Twenty-four hours after treatment, the count of the mite showed varied efficacy of the used treatments as the highest efficacy occurred due to *B. bassiana* and Abamactin + mineral oil Cable which reduced the infestation to 0.17 and 0.01 individuals/ inch^2, followed by *P. persimilis*, (2.8 individuals/ inch^2) then

T. swriskii releasing, (3.00 individuals/ inch^2) also the same trend observed with Hama as the corresponding counts were, 5.11, 1.00, 6.35 and 8.13 individuals/ inch^2, respectively.

With respect to the allover mean of all the previously seasonal inspections after 17 weeks of application, recorded data in Table (5) illustrated that, the highest reduction were observed with using the three treatments; *P. persimilis,* Abamactin +mineral oil Cable and *B. bassiana* as the bean leaves harbourd with 1.37, 2.27 and 3.08 individuals/ inch^2 on Bolista variety while with Hama the corresponding seasonal means were 4.91, 4.80 and 12.08 adults/ inch^2 respectively. For the efficacy of *T. swriskii* on the two cultivars, the seasonal average showed statistically moderate effect (10.83 adults/ inch^2 on Bolista and 18.30 adults/ inch^2 on Hama cultivar. On the other hand, the untreated plants infested with the highest means, 19.72 and 40.20 individuals/ inch^2 for the two cultivars, respectively.

Regarding the mean reduction for the four treatments, higher reduction percentages were recorded by Abamactin +mineral oil Cable followed by the phytoseiid predator *P. persimilis* and *B. bassiana.* After 17 weeks of treatment the statistical arrangement of their mean percentages were 88.04, 85.09, 83.69% on Bolista and 87.15, 82.81 and 82.17% on Hama for the aforementioned controlling agents, respectively. Also the statistical analyses showed no significant differences among them. (L.S.D. = 9.47 and 9.51). While the reduction percentages were the lesser of the other application, *T. swirskii,* which averaged 40.76 and 48.77% of adult stage for the same bean cultivars, respectively.

These results are an almost at the same direction with those of many researchers. El-Saiedy (2003) mentioned that *Neoseiulus californicus* (McGregor) (Acari: Phytoseiidae) and *P. persimilis* were the best predators for controlling *T. urticae* on strawberry. El-Saiedy *et al.* (2008) on two eggplant cultivars in open field. Who evaluated the efficacy of three predatory phytoseiid mites, *P. persimilis*, *Neoseiulus cucumeris* (Oudemans) (Acari: Phytoseiidae) and *N. californicus* and a biocide for controlling *T. urticae.* Also our results were in agreement with Rhodes *et al.* (2006) who observed that among the combination treatments, *P. persimilis* / *N. californicus* treatment significantly reduced *T. urticae* numbers compared with the untreated treatment, but was not as efficient as *N. californicus* alone. Xu and Enkegaard (2010) who stated that, *A. swirskii* consumed the same amount of the various types of spider mite nymphs except of the active deutonymphs of which significantly fewer were consumed. The latter is presumably in part a reflection of deutonymphs being larger and more active and thus more difficult to conquer and in part a reflection of a more pronounced congregating and web producing habit of deutonymphs compared to protonymphs there by slow downing the movements of *A. swirskii*. The latter is in accordance with notes by Van Houten *et al.* (2007) that *A. swirskii* was hardly found in the webbing of *T. urticae*. In nature, accarine pests are only a part of biological complex of which predacious mites, particularly phytoseiid group, could be of value in checking infestations. Many workers have reported that phytoseiids have a role to play in the control of acarine pests (Rasmy *et al.*, 2003; Abou-Awad *et al*., 2009 and Abou-Awad *et al*., 2017).

Table (5): Mean numbersof *Tetranychus urticae* adults / leaf on two kidney bean varitites under net hous conditions affected by releasing two predatory mites, *Beauveria bassiana* and the biocide abamactin +mineral oil Cable spraying at Beheira Governorate during 2016 season.

Varitites	Bolista					Hama				
Inspections date / Treatment	*P.prisimilis*	*T. swirskii*	*B. bassiana*	Abamactin +Mineral oil cable	control	*P.prisimilis*	*T. swirskii*	*B. bassiana*	Abamactin +Mineral oil cable	control
Feb.,15th	3.60	3.56	3.47	3.50	3.55	7.22	7.25	7.00	7.34	7.23
22nd	2.80	3.00	0.17	0.01	4.35	6.35	8.13	5.11	1.00	9.44
28th	2.65	4.11	1.18	0.45	6.00	5.44	9.33	6.33	1.95	13.50
Mar.,7th	2.40	5.00	2.18	2.00	7.33	8.43	10.11	7.18	3.90	16.55
15th	3.40	6.98	4.18	3.95	9.11	9.33	12.50	11.11	6.18	20.33
22nd	3.00	7.00**	0.65**	0.36**	10.90	7.11	12.50	18.23**	8.33**	25.14
29th	2.13	8.14	0.98	0.84	12.30	6.33	16.22	6.11	0.95	28.60
Apr.,6th	1.40	9.50	1.25	1.00	16.33	6.50	15.22	5.42	2.30	33.00
13th	0.94	10.85	3.15	2.13	18.33	7.14	18.35	8.33	4.13	37.14
20th	0.88	13.90	6.90	4.11	21.11	6.30	21.11	12.30	7.30	42.90
27th	0.14	12.80	7.15**	6.11**	24.35	5.11	23.90	18.50**	11.60**	50.80
May,4th	0.00	14.60	0.35	0.25	26.13	4.22	26.40	9.40	0.13	52.70
11th	0.00	15.30	0.95	0.75	29.11	3.10	28.33	13.50	0.90	58.90
18th	0.00	16.40	2.14	1.13	32.11	0.95	27.80	15.30	2.11	61.14
25th	0.00	17.60	3.90	2.30	35.90	0.00	22.90	17.60	4.55	68.90
Jun, 2nd	0.00	18.50	5.84	3.50	37.17	0.00	26.90	20.50	7.30	73.40
10th	0.00	16.90	7.97	6.17	41.16	0.00	24.22	23.50	11.56	83.71
Average ± SE	1.37±0.33 C	10.83±1.29 B	3.08±0.62 C	2.27±0.48 C	19.72±2.99 A	4.91±0.74 C	18.30±1.80 B	12.08±1.43 BC	4.80±0.89 C	40.20±5.75 A
F	27.03					26.89				
LSD	4.23					7.93				

Values singed by the same letter of the same varity are not significantly different at alpha = 0.05 % level.
**Re-spray

Table (6): Reduction persentages of *Tetranychus urticae* aduls /inch2 on two kidney bean varitites under net hous conditions affected by releasing two predatory mites, *Beauveria bassiana* and the biocide abamactin +mineral oil Cable spraying at Beheira Governorate during 2016 season.

Varitites	*Bolista*				*Hama*			
Inspections date / Treatment	*P. prisimilis*	*T. swirskii*	*B. bassiana*	Abamactin +Mineral oil cable	*P. prisimilis*	*T. swirskii*	*B. bassiana*	Abamactin +Mineral oil cable
Feb.,15th	36.53	31.03	96.04	99.77	41.05	13.76	71.07	89.58
22nd	56.45	31.50	80.05	92.39	64.69	30.79	75.01	85.79
28th	67.71	31.79	69.83	72.33	55.36	38.83	76.70	76.82
Mar.,7th	63.20	23.38	53.46	56.02	59.78	38.43	70.76	70.10
15th	72.86	35.78**	93.95**	96.65**	75.22	50.21	61.02 **	67.41 **
22nd	82.92	33.82	91.92	93.07	80.60	43.21	88.48	96.73
29th	91.55	41.82	92.24	93.79	82.74	53.81	91.24	93.14
Apr.,6th	94.94	40.81	82.57	88.21	83.15	50.52	87.89	89.06
13th	95.89	34.15	66.85	80.25	91.22	50.72	84.64	83.26
20th	99.43	47.43	70.22 **	74.5**	94.63	52.89	80.55 **	77.54 **
27th	100.00	44.13	98.64	99.03	97.97	49.84	90.40	99.76
May,4th	100.00	47.44	96.69	97.39	98.66	51.83	90.45	98.50
11th	100.00	48.93	93.24	96.43	99.93	54.47	90.15	96.61
18th	100.00	50.97	88.98	93.50	100.00	66.72	86.36	93.50
25th	100.00	50.23	84.06	90.45	100.00	63.30	85.01	90.22
Jun, 2nd	100.00	58.94	80.36	84.80	100.00	71.03	84.93	86.42
10th	85.09 A	40.76 B	83.69 A	88.04 A	82.81 A	48.77 B	82.17 A	87.15 A
F	40.77				27.98			
LSD	9.97				9.51			

Values singed by the same letter of the same varity are not significantly different at alpha = 0.05 % level.
**Re-spray

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Monitoring the carpenter worm *Paropta paradoxa* (Lepidoptera:Cossidae) infesting apple orchards in Egypt

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Abstract:

In apple orchards, the carpenter worm *Paropta paradoxa* (Herrich-Schäffer) (Lepidoptera:Cossidae) became recently a serious pest in Egypt. Monitoring the population fluctuation was conducted at Nobaria district, Beheira Governorate during the two successive years (2017 and 2018). The rate of infestation approximated 14% (11.5 – 16.5% infested trees), while the degree of infestation reached 1.235 (1.47 – 2.47) moths per tree. The seasonal abundance of the adult moths prevailed from early or late March to early or late November, with mostly two or three flight peaks. The major moths ' flight period was in summer months (July-September) (1.10 moths in 2017 to 1.46 moths / tree in 2018), while spring months (April-June) recorded moderate numbers, being 0.79 to 1.01 moths /tree. Autumn was the minor (0.12 to 0.16 moths/tree), however winter was scant (0.02 – 0.04 moths / tree). The total numbers per year were 2.05 and 2.33 moths / tree during 2017 and 2018, respectively. There was two brood of moths' activity in 2017, but three broods in 2018. There were 8.5 months of moths' activity each year. Effect of weather factors on the moths' activity was mostly positive and significant with daily maximum, daily minimum and daily mean temperatures but mostly negative and insignificant with daily mean relative humidity. Infestation was much doubled during only one year, thus needed urgent integrated control.

Introduction

Apple plantations (*Pyrus malus*, Rosaceae) in Egypt, became profitable cash crop. The area under cultivation extended through the old Delta and valley lands as well as new reclaimed lands. Frequent field observations allover the Governorates of Egypt , in both old valley lands and new reclaimed desert lands , indicated that in addition to *Paropta paradoxa* (Herrich-Schäffer) (Lepidoptera: Cossidae) (Kinawy *et al.,* 1991), the other major lepidopteran stem boring insect pests in apple orchards were, *Zeuzera pyrina* (Linnaeus) (Lepidoptera :Cossidae) (Tadros *et al.,* 2003) and *Synanthedon myopaeformis*

Borkhausen (Lepidoptera :Sessidae) (Tadros *et al.,* 2004).

In addition to apple, *P. paradoxa* is a dangerous pest in Egypt in fig orchards (Willcocks, 1937), apple orchards (Kinawy *et al*., 1991), grape vineyards (Tadros, 1982) and mandarin trees (El-Assal *et al*., 2008).

Young larvae *P. paradoxa* bore directly into twigs, branches and main trunks immediately after hatching. Young larvae bored sapwood, while old larvae bored heartwood. Larval tunnels that might reach 8 mm. in diameter were always kept open and enlarged gradually. Mature larvae lined their tunnels with loose dim silky webbing. Pupation occurred at the end of the tunnels and the pupal skins partially protruded from the opening after moth emergence (Kinawy, 1981). Moths started flight in fig orchard at Alexandria Governorate, from mid-April / late May until late October / late November resulting in two peaks of activity during early July and late August / early September (Mesbah *et al.*, 1993).

Successful integrated pest control depends largely on monitoring studies especially the seasonal fluctuation in the target pest population, the progress of infestation, the seasonal cycle and the effect of the main weather factors on the target pest. In an attempt to contribute to such a gap in the knowledge, the present comparative ecological studies are aimed. The broad objective of investigation is to add new information that may help in planning of rather effective "Integrated Control Programs" for the management of *P. paradoxa* in apple orchards.

Materials and methods

Monitoring studies of *P. paradoxa* were conducted in apple orchards located in the reclaimed lands, at Nobaria district, Beheira Governorate. Monitoring studies extended during two successive years form early January, 2017 until late December, 2018. Only specific safe control treatments were applied in the selected areas throughout the studies.

Rate and degree of infestation:

Assessment of the rate of infestation was calculated as the percentage of numbers of randomly distributed infested trees with *P. paradoxa* in two apple orchards each year. The degree of infestation was estimated by the mean number of adult moths per tree (indicated by the newly protruding pupal skins) that completed their life cycle and emerged from apple trees each year. Estimation of rate and degree of infestation was carried out in 100 random trees.

1. Population fluctuation of *Paropta paradoxa*:

1.1. Seasonal abundance:

The seasonal abundance of *P. paradoxa* was carried out in apple orchards, about 5 feddans in area with trees approximately 12 years old located at Nobaria district, Beheira Governorate. Monitoring *P. paradoxa* covered two successive years (2017 and 2018).

During December 2016, the old pupal skins were removed and from January 1st , 2017 until December 31, 2018, the newly protruding pupal skins indicating emergence of moths of *P. paradoxa* were counted at half monthly intervals on the 15th and last day of every month. To avoid repeated counting newly protruding pupal skins were immediately removed after counting.

1.2. Progress of infestation and seasonal cycles:

Data of the seasonal abundance were accumulated from January 1st , 2017 until December 31, 2018 for each half monthly interval. The total numbers of moths (pupal skins) represented the accumulated number for the two years together. The presented Figures indicated the periods of the seasonal cycles of moths' activity and inactivity. Progress of infestation also indicated the rate of increase in the borer infestation year after another.

2. Effect of weather factors on the activity of *Paropta paradoxa*:

Four main weather factors, the day maximum temperature (DMxT), day minimum temperature (DMnT), day mean temperature (DMT) and day mean relative humidity (DMRH) were considered in this study. Necessary weather data were obtained from the Central Laboratory of Climate and Meteorology, Agricultural Research Centre, Giza. Population data of *P. paradoxa* taken into account and the meteorological data, both at half monthly intervals, were presented. The relationship between the four weather factors and the moths' population data during the activity season was investigated for two successive years (from January 2017 until December 2018) in the two apple orchards.

To determine the direct effect of each weather factor on the borer activity, population counts were plotted against the corresponding weather data. The simple correlation coefficients "r" for the relationship between each weather factor and *P. paradoxa* population was then worked out according to Snedecor and Cochran (1990).

Results and discussion

1. Rate and degree of *Paropta paradoxa* infestation:

Data in Table (1) clarified that *P. paradoxa* infestation markedly increased in apple orchards year after another. In two apple orchards at Nobaria district, Beheira Governorate, the percentages rate of infestation increased in the 1st orchard from 12% in December 2017 to 21% in December 2018 (mean, 16.5%), but in the 2nd orchard the percentages rate of infestation increased from 7% in December 2017 to 16% in December (mean, 11.5%). The approximated grant mean rate of infestation was 14%.

Although the number of larvae completed their development and emerged per tree (indicated by the pupal exuvia protruding on trees) in the 2nd orchard was only 0.67 (0.43 – 0.91), yet it was 1.80 (1.57 – 2.03) in the 1st orchard. The grand mean degree of *P. paradoxa* infestation approximated 1.235 (1.00 – 1.47) moths per apple tree. This is a serious parameter of the progress of the rate and degree of *P. paradoxa* infestation in apple orchards in Egypt.

Table (1): Rate and degree of *Paropta paradoxa* infestation in apple orchards at Nobaria district, Beheira Governorate during 2017 and 2018 activity seasons.

Year		**2017**	**2018**	**Total**	**Mean**
Rate of infestation (%)	1st orchard	12	21	33	16.5%
	2nd orchard	7	16	23	11.5%
	Grand Mean	**9.5**	**18.5**	**28**	**14%**
Degree of infestation (number of pupal skins /tree)	1st orchard	1.57	2.03	3.60	1.80
	2nd orchard	0.43	0.91	1.34	0.67
	Grand Mean	**1.00**	**1.47**	**2.47**	**1.235**

2. Population fluctuation of *Paropta paradoxa*:

2.1. Seasonal abundance:

As shown in Tables (2 and 3) and Figure (1), the moth's activity of *P. paradoxa* prevailed during the period from early or late March to early or late November in apple orchards at Nobaria district, Beheira Governorate during the two years of study (2017 and 2018 seasons).

The flight commencement of moths started between the 1st half of March (0.01 moths / tree) in 2017, and the 2nd half of March (0.02 moths / tree) in 2018 at Nobaria district, Beheira Governorate. Two or three flight peaks of *P. paradoxa* moths' emergence were recorded at Nobaria district, Beheira Governorate. Table (3) clarified that the first peak of moths' activity mainly fluctuated between the 1st half of March 2018 (0.19 moths / tree) and 2nd half of March 2017 (0.18 moths / tree). A second peak of moths' activity occurred during the 2nd half of June 2018 (0.30 moths / tree). The second peak in 2017 was noticed in the 2nd half of July (0.25 moths / tree). The third peak of moths' activity fluctuated during the same 2nd half of July 2018 (0.35 moths / tree).

At Nobaria district, Beheira Governorate, the last flight of moths was recorded during the 1st half of November in 2017 (0.02 moths / tree) and the 2nd half of November in 2018 (0.01 moths / tree). Tables (2 and 3) further indicated that the maximum *P. paradoxa* moths' flight was during summer months (July - September) showing 1.10 and 1.46 moths / tree in 2017 and 2018 seasons, respectively.

Spring months (April - June) recorded moderate numbers, being 0.79 moths / tree in 2017 and 1.01 moths / tree in 2018 seasons. Autumn months (October - December) showed few numbers, 0.12 and 0.16 moths /tree during 2017 and 2018 seasons, respectively.

Scant numbers of moths' activity was during winter months (January - March), showing 0.04 a nd 2.02moths / tree during 2017 and 2018 seasons, respectively. Moreover, the total numbers of moths emerged during the whole year were 2.05 and 2.33 moths / tree at Nobaria district, Beheira Governorate during 2017 and 2018 seasons, respectively. The respective means per month were 0.171 and 0.194 moths / tree during 2017 and 2018 seasons, respectively.

Data in Table (3) and Figure (1) emphasized that at Nobaria district, Beheira Governorate, *P. paradoxa* had two brood of moths' activity in 2017 or three broods in 2018. In 2017, the first brood prevailed from the 1st half of March to the 1st half of September and the second from the 2nd half of April to the 1st half of November. In 2018, the first brood prevailed from the 2nd half of March to the 1st half of August and the second from the 1st half of May to the 1st half of October and the third brood from the 1st half of June to the 2nd half of November.

Table (2) : Mean number of *Paropta paradoxa* moths in apple orchards at Nobaria district, Beheira Governorate during 2017 and 2018 activity seasons with the corresponding day mean temperature (DMT) and relative humidity (DMRH).

Date of inspection		2017 season		2018 season		2017 Season		2018 Season	
		Actual	Cumulative	Actual	Cumulative	DMT °C	DMRH %	DMT °C	DMRH %
January	1-15	0.0	0.0	0.0	2.05	16.5	72	16.1	70
	16-31	0.0	0.0	0.0	2.05	17.3	70	16.6	71
February	1-15	0.0	0.0	0.0	2.05	19.5	65	17.5	67
	16-28	0.0	0.0	0.0	2.05	20.3	55	22.1	57
March	1-15	0.01	0.01	0.0	2.05	23.5	59	23.5	59
	16-31	0.03	0.04	0.02	2.07	25.6	62	24	61
Winter		**0.04**		**0.02**					
April	1-15	0.06	0.10	0.05	2.12	28.3	52	27	52
	16-30	0.09	0.19	0.10	2.22	29.5	47	30.8	46
May	1-15	0.11	0.30	**0.19**	2.41	31.7	53	28	51
	16-31	**0.18**	0.48	0.16	2.57	33.9	42	30.1	43
June	1-15	0.15	0.63	0.21	2.78	35.3	51	33.6	52
	16-31	0.20	0.83	**0.30**	3.08	35.6	49	34.3	48
Spring		**0.79**		**1.01**					
July	1-15	0.22	1.05	0.26	3.34	36.1	52	35.2	53
	16-31	**0.25**	1.30	**0.35**	3.69	36.5	60	35.5	62
August	1-15	0.21	1.51	0.28	3.97	36.2	60	36.9	60
	16-31	0.19	1.70	0.24	4.21	35.8	63	34.2	63
September	1-15	0.13	1.83	0.20	4.41	33.6	61	32.8	60
	16-30	0.10	1.93	0.13	4.54	32.1	57	30	58
Summer		**1.10**		**1.46**					
October	1-15	0.07	2.00	0.09	4.63	29.1	61	29.3	62
	16-31	0.03	2.03	0.04	4.67	28.2	60	27.6	60
November	1-15	0.02	2.05	0.02	4.69	26.3	62	27.1	63
	16-30	0.0	2.05	0.01	4.70	23.5	59	25.5	58
December	1-15	0.0	2.05	0.0	4.70	21.5	58	22.2	58
	16-31	0.0	2.05	0.0	4.70	19.2	63	20.0	59
Autumn		**0.12**		**0.16**					
Grand Total		**2.05**	**2.05**	**2.33**	**4.70**				
Mean / month		**0.171**		**0.194**					

Table (3): Commencement, peak, last dates and broods of *Paropta paradoxa* moths in apple orchards at Nobaria district, Beheira Governorate during 2017 and 2018 activity seasons.

	2017 season	2018 season
Flight Commencement	1st half of March	2nd half of March
Peaks	2nd half of May	1st half of May
		2nd half of June
	2nd half of July	2nd half of July
Last flight	1st half of November	2nd half of November
Broods	1st half of March to the 1st half of September	2nd half of March to the 1st half of August
		1st half of May to the 1st half of October
	2nd half of April to the 1st half of November	1st half of June to the 2nd half of November

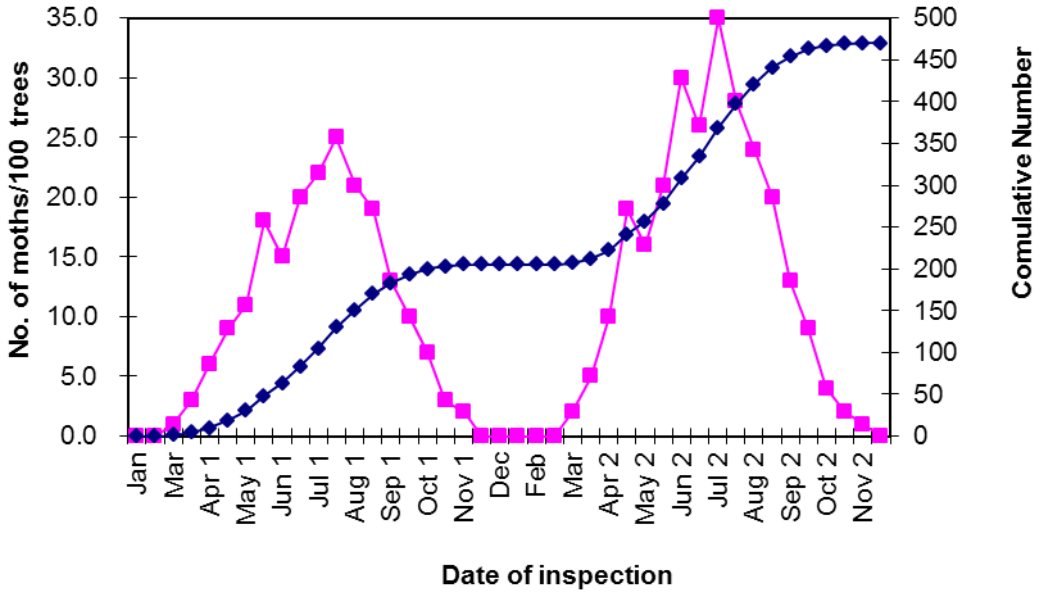

Figure (1): Mean numbers of *Paropta paradoxa* moths in apple orchards at Nobaria, Beheira Governorate during 2017 and 2018.

2.2. Progress of infestation:

The cumulative numbers (seasonal cycle) of emerged moths (Figure, 1) indicated that there was 8.5 months of moths activity at Nobaria, Beheira Governorate followed by 3.5 months of moths inactivity. Infestation was more than doubled each only one year. Infestation / tree / year increased from 2.05 moths at the end of 2017 to 4.70 moths at the end of 2018. This serious parameter imposed urgent need of controlling the pest.

2.3. Effect of temperature and relative humidity on moths activity:

Statistical analysis revealed that the fluctuation in *P. paradoxa* moths' population was significant and positively correlated with the temperature in the two years of study, that day maximum temperature (DMxT) showed "r" from 0.8133 to 0.8271, day minimum temperature (DMnT) resulted in "r" from 0.8065 to 0.8098, and day mean temperature (DMT) calculated "r" from 0.7753 to 0.7949). On the contrary, the

effect of day mean relative humidity (DMRH) on moths' population was insignificant and negative where "r" resulted in -0.1987 to -0.12031. Statistically, there were combined effect of all the weather factors: the DMxT, DMT, DMnT and DMRH on *P. paradoxa* moths' population fluctuation in the studied apple orchards than the effect of each single factor. The combined effect of these weather factors on moths' activity (explained variance "E.V.") ranged between 61.3% and 66.9%. This may be due to the hidden larval and pupal stages inside the wood of the trees not exposed to the direct weather factors. However, these weather factors strongly affect the whole ecosystem of the apple orchards.

Monitoring studies (especially the seasonal fluctuation of the pest population, progress of infestation, seasonal cycle and effect of the main weather factors on the target pest) are essential in planning successful and effective "Integrated Control Programs" for the management of pest. Survey studies indicated that *P. paradoxa* became recently dominant and economically important boring pest in apple orchards. In apple orchards moths' activity started from March and emergence was stopped by November. The activity seasons (8.5 months) were mostly in summer and spring, but few in autumn and late winter season showed scant moths activity. There were two to three broods of the borer activity each year. Infestation was more doubled each one year, thus needed urgent integrated control. Generally, there were positive and significant effects of major weather factors: daily maximum, daily minimum and daily mean temperatures on the borer activity, but this effect was mostly negative and insignificantly with daily mean relative humidity.

Literature is few concerning studies on *P. paradoxa* in apple orchards as well as other fruit hosts, yet in Egypt, researches focused on fig (Kinawy, 1981; Mesbah *et al.*, 1993 and Hashim, 2004), grapevine (Tadros, 1982), mandarine (El-Assal *et al.*, 2008) and apple (Kinawy *et al.*, 1991) trees. Theses researches stated that immediately after egg hatching the young larvae of *P. paradoxa* bore directly into twigs, branches and main trunks. Young larvae bored sapwood, while old ones bored heartwood. Mature larvae lined their tunnels with loose dim silky webbing. Pupation occurred at the end of the tunnels and the pupal skins partially protruded from the opening after moth emergence. Population studies indicated that moths' population fluctuation of *P. paradoxa* on the previous fruit host trees emerge prevailed from the April / May until October / November, with one or two major peacks. Mesbah *et al.* (1993) stated that the life cycle of *P .paradoxa* on fig trees recorded 370 -422 days. They stated that emergence of *P. paradoxa* moths highly affected with the temperature but the relative humidity had less effect. Abroad, Plaut and Tsour (1975) in the Jordan valley and the Shomorn foothills, found that in grapevine yards emergence of *P. paradoxa* adults began during first or second ten days of April, while in the Yizreil valley emergence of adult moths began late April or early May. The main emergence period ended in early July in Jordan valley and in September in the Shomorn foothills.

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Survey and population dynamic of spiders infesting faba bean, with emphasis on acaricide effect on biological aspects of the spider *Kochiura aulica* (Araneae: Theridiidae)

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Abstract:

Spiders are beneficial predators and ancient animals. They are abundant and widespread in all ecosystems and constitute one of the most important components of global biodiversity. They are voracious predators and combined with their high abundance, may play an important role in the reduction of different pests populations. Spiders are among the most abundant predators recorded in faba bean (*Vicia faba* Harz) in Beni-Suef Governorate. Field trials were conducted in Beni-Suef Governorate during 2017 and 2018 to survey and population dynamic of some families and species of the spiders. Results indicated that recorded predaceous spiders were identified in 6 families as follows: Uloboridae (orb-weavers), Theridiidae (mostly space web weavers), Salticidae (jumping spiders), Thomisidae, Philodromidae (crab spiders) and Cheiracanthiidae. The high percentage mortality appeared in different species spider treated with recommend of Wonder 36%SC resulted after 7 days from treatment averaged 32.8 and 38.7% mortality for Thomisus spinifer, 30 & 60% mortality of Theridion melanostictum, followed by Theridion spinitarse (26.6 & 39.7%) mortality for female and male, respectively compared with (0.0%) in control. The effect of acaricide on the biological aspects of *Kochiura aulica* (Koch) (Araneae: Theridiidae) was studied under laboratory condition. The first and 2nd instar spiderling females and males were high affected when treated with recommended compound, the both instars filled to complet the duration. Whereas, high significantly prolonged the duration of the 3rd, 4th and 5th spiderlings female than untreated. The compound increased the duration female by 16.3 days on contrast decreased the duration male by 5.7 days compared with control. Female resulted from treated 3rd instar spiderlings deposited 17.0 eggs/ sac, compared with 26.6 eggs/ sac in control. The hatching percentages were 65.0 %, when female resulted from treated, compared with 96.0 in control. The results also revealed that Wonder was the most effective acaricide on duration, fecundity and hatching.

Introduction

Spiders (Araneae) are considered one of the most important natural control agents in a wide range around the world. Members of this order are different in size small, medium or big size, they are usually found hanging upside down in an irregular web suspended on plants or hidden in rocks or fissures in soil. Many of them use the threads that often hard to notice unless they occasionally glitter in the sun light or covered with dust (EL-Erksousy, 2003 and EL-Erksousy *et al*., 2006). The true spiders recorded associated with different agricultural pests. All adult spiders are predaceous and play very important role in the decreased of numbers pest pulsations (Greenstone, 1999; Riechert, 1999; Whitehouse and Lawrence, 2001 and Huseynov, 2007). However, individual spider species lack many of the characteristics suggested as necessary for a successful biological control agent (Murdoch *et al.*, 1985). The significance of spider assemblages for biological control of pests is largely unknown and spiders have been the subject of very few investigations (Bishop, 1978 and 1980 and Bishop and Blood, 1981).

Therefore, The aim of this work is to study the effect of the recommended compound on different instars spiderling and studies on biological aspects of the predaceous spider, *Kochiura aulica* under laboratory conditions.

Materials and methods

1. Survey of predator spiders on faba bean:

Survey and abundance of predator spiders were conducted during the period of October until April for two seasons (2017–2018) at Beni-Suef Governorate in Middel Egypt. Samples were collected from 25 plants in four groups of faba bean. Predaceous spider species were collected from one location of cultivated faba bean (*Vicia faba*) and found on different the ages of plant and associated with flower and fruits. Spiders were collected by hand picking or/ and shaken for each sample the spiders after then brought to the laboratory for identification. Samples were conducted weekly during the surveying period. The surveyed spiders were kept in glass vials containing 75% ethyl alcohol and droplets of glycerin. Identification of the collected spiders was available for adults only Sallam (2002). Identification of adult females is depending on shape of eyes and epigyneal plate of female or on the palp in case of male.

2. Effect of acaricide on biological aspects of the spider *Kochiura aulica* :

2.1. Spider as a predator reared for treatment:

Spiders of the family Theridiidae were obtained from field by means of hand picking with collected from plants. The spider adults identified in the laboratory. Adult females and males were confined together in a test tube (20 cm long and 0.5 cm in diameter) and closed with a cotton pad. The female was observed daily until laying the egg sac and immature emergence. Each spiderling was isolated separately in a test tube with a surplus number of prey individuals. Thirty predator individuals (spiderlings) from each instar were noticed spray by recommended the compound were estimated for each stage of *K. aulica* males and females.

2.2.Chemical used:

Common name: Wonder 36% SC
Trade name: Chlorfenapyr, 36% SC
Chemical name: -Bromo-2-(4-chi lorophenyl)- 1 ethox methyl- 5- tryfluem ethyl – H- pyrrolc- 3- carbonitrile
Rate of application: 180 cm^3/ 200 L

2.3.Preparation the pesticides used:

The preparation of Wonder 36% SC was tested against different immature stages of *K. aulica* spiderling. Recommend concentration for compound was prepared as follow: (750ppm).

2.4. Effect of Wonder 36% SC compound on different immature stages of spiderlings:

Three replicates from each instars of spiderling for the recommend concentration were used, each replicate contained 30 individual (one day old), for each instars, they were placed in a jars (1/4kg.) and sprayed by recommend the acaricide. Another three replicates (20 individual (one day old)) from each, were sprayed by water as a check. Treated were placed in a jars (1/4kg.) under the previous conditions. After 1 to 7 days of each treatment to determine the reduction percentages by using Henderson and Tilton equation (1955).

2.5.Biological aspects:

To estimate the effect of the tested preparations compound against the biological aspects of the 3rd instar spiderling males and females predaceous spider; after treated 3rd instar spiderling with recommend Wonder compound, *K. aulica* was studied as follows: percent of mortality, spiderlings duration (days), mating and longevity (from adult emergence to death in days) and resulted from treated the third instar spiderlings of the spider females and males. In addition to pre-viposition (days), oviposition (days) , post-oviposition (days) periods, number of egg sac/female and total number of egg/sac of the spider females.

3.Statistical analysis:

One way Anova was calculated by using SAS statistical software (SAS Institute, 2003). In addition, LSD (Fisher's Significant Difference Test) was chosen to identify the significant difference within group.

Result and discussion

1. Survey of predator spiders infesting faba bean:

Data presented in Table (1) recorded the number of families collected from faba bean in Beni- Suif Governorate throughout 2017 and 2018 seasons. Resulted recorded 6 families collected from leaves or different parts of plant of faba bean. The collected predaceous family (Arachnida : Araneae) spiders were identified as follows: Theridiidae (mostly space web weavers), Salticidae (jumping spiders), Philodromidae (crab spiders), Thomisidae, Uloboridae (orb-weavers) and Cheiracanthiidae. The obtained results as shown in Table (1) found that the spider families and species are nocturnal collected comb footed spiders, the total number family estimated by 6 families as followed: 3 individuals in family Theridiidae, (*Theridion spinitarse* O. Pickard-Cambridge*, Kochiura aulica* Koch and *Theridion melanostictum* O. Pickard-Cambridge; one individual in different families was collected, Uloboridae (*Uloborius* sp), Thomisidae (*Thomisus spinifer* Blackwall), Philodromidae (*Thanatus albini* Audouin), Miturgidae [(*Chieracanthium inclusum* (Hentz)] and Salticidae *(Euophrys granulata* Denis*)* .

Data in Table (1) showed that the population of spider families was generally different throughout 2017 and 2018 seasons. The overall total of spider (3 in family Theridiidae) throughout two seasons were 118 and 440 individual on *Vicia faba*, respectively, recorded the high percentage of totally collected spider family (individuals adults or immature stages) while the lowest families were 4 and 5 individuals in Uloboridae and 7 and 10 individuals adults in Miturgidae families, respectively, during two seasons. The most abundant species was noticed for associated wit *E. granulata* (40 and 43) individuals, associated with leaves and flower faba bean in two location.

Table (1): List of collected families and species of spider associated with faba bean at Beni- Suif Governorate during 2017 and 2018 seasons.

Spider families /plant	Collected species	No. collected	
		2017	2018
Theridiidae	*Theridion spinitarse*	9	110
	Kochiura aulica	6	112
	Theridion melanostictum	12	111
Philodromidae	*Thanatus albini*	21	40
Thomisidae	*Thomisus spinifer*	16	12
Uloboridae	*Uloborus* sp.	4	5
Salticidae	*Euophrys granulate*	43	40
Cheiracanthiidae	*Cheiracanthium inclusum*	7	10
Total families= 6	Total species= 8	118	440

From these data can be concluded that the total number of spider species *T.spinitarse, K. aulica* and *T. melanostitctum* and families Philodromidae and Salticidae were collected from Beni- Suif Governorate is higher during 2018 than were collected during 2017 season, in the previous studies associated with leaves and flower. Evans (1985) collected 33 spider species from 12 families infesting soybean, while, Bishop (1978 and 1980) collected 25 species from ten families infesting cotton. A review of Australian literature recorded in cotton farming systems lists 41 species from 13 families (Johnson *et al.*, 2000)

2. Effect of recommended Wonder compound on mortality of different species:

Data represented in Table (2) showed that mortality different species of spider after treated from 1 to 7 days. The high percent of mortality recorded after 24hr in all families and species and the most species' affected were *T. melanostictum, T. spimitarse* and *T. spinifer* the percent mortality estimated by 20 and 25; 20 and 30 and 20 and 25 % mortality (for two species) females and males, respectively, the high total percentage mortality appeared in different species spider resulted after 7 days estimated by 32.8 and 38.7% mortality for *T. spinifer*, 30 and 60 % mortality for female and male *of T. melanostictum* respectively, compared with (0.0%) no mortality rerecorded in control, followed by, *T. spinitarse* (26.6 and 39.7) mortality for female and male, respectively, compared with (0.0%) in control. On the other hand the low total percentage mortality recorded with *T. albini* 10 and 25% mortality for female and male, respectively, compared with (5 and 5 %) in control, and (10 and 20.8%) mortality for female and male, respectively, compared with (0.0 and 4%) in control.

Table (2): Effect of recommended woner compound on percent of mortality different species of adult stage.

Spider families /plant	Species of spider treated	% Mortality after								Total mortality	
		24h		48h		72h		7days			
		♀	♂	♀	♂	♀	♂	♀	♂	♀	♂
Theridiidae	*Theridion*	20	25	0	0	0	7	6	7	27	20
	Kochiura aulica	12	12	0	0	5	2	2	2	17	12
	Theridion	20	30	0	5	5	10	5	10	30	20
Control		-	-	-	-	-	-	0	0	0	0
Philodromidae	*Thamoty albini*	5	10	0	5	0	5	5	5	10	25
Control		-	-	-	-	-	-	5	5	5	5
Thomisidae	*Thomisus*	20	25	6.2	6.6	0	0	6.6	7.1	32.8	38.7
Control		-	-	-	-	-	-	-	-	-	-
Uloboridae	*Uloborius sp*	5	10	11	2	0	0	5.5	6.2	21.5	18.2
Control		-	-	-	-	2	-	5	2	7	2
Salticidae	*Eudphrys*	5	10	0	5	0	5	5	5	15	25
Control		-	-	-	-	-	-	5	5	5	5
Cheiracanthiidae	*Cheiracanthium*	5	10	0	5.8	0	0	5	5	10	20.8
Control		0	0	0	1	0	0	0	3	0	4

3. Effect of recommended compound on some biological aspects of *Kochiura aulica*:

Data in Table (3) recorded that the first and 2nd instar spiderling females and males high affected when treated with recommended compound, the percent mortality estimated by 95% for 1st instar spiderling and 88% for 2nd instar spiderling and don't completed the development, compared with 3% in control, on the contrary, when the 3rd, 4th and 5th spiderlings treated with recommended compound the percent mortality estimated by 69, 49 and 20% mortality, respectively, compare with 2, 0 and 3 % in control.

Table (3) showed that the spider females and males have five spiderlings stages. The third and fifth spiderlings were longer in their duration than other spiderlings in both females and males, it averaged 26.6 and 21.6 days/ female and 15.3 and 14.0 days/ male when third and fifth spiderlings treated with recommended compound, respectively, compared with 15.0 and 15.0 days/ female and 25.6 and 23.0 days/ male for third and fifth spiderlings, respectively, in control, followed by 4th spiderlings in both sexes, respectively. As tabulated, the treated different instars spiderlings had higher duration (from 11 to 12 days) in female compared with spiderlings control.

The duration of the spiderlings increased when exposed to the recommended compound than untreated as shown in Table (3) and when exposed the first and 2nd instars of both females and males spiderlings to compound, the duration of the spiderlings high affected and no completed the duration, all the first and 2nd spiderling dead because its small size.

Table (3): Treated of different immature stages of *Kochiura aulica* with recommended Wonder compound under laboratory condition.

Spiderling	Conc.	% Mortality Trated	% Mortality control	Duration			Duration		LSD
				Female treated	Female control	LSD	Male treated	Male control	
1st	Recommend 1620 used ppm	95	3	---	15.6±0.9	---	---	13.3±0.5	--
2nd		88	3	--	19.0±1.6	---	--	16.6±0.9	----
3rd		69	2	26.6±1.2b	15.6±0.3a	0.549	25.3±0.9a	25.6±1.3a	ns
4th		49	0	16.3±1.4b	10.0±0.1a	1.022	19.6±1.3	18.0±1.2	0.311
5th		20	3	21.6±1.6b	15.3±0.4a	1.141	24.0±2.1	23.0±1.5	ns

The means with the same letters at the same row are not significantly different at 0.05%

3.1. Spiderling stage:

It is clear that the tested compound cussed high significantly prolonged the duration of the female stages spiderling than that of the untreated check. Table (4) revealed that the duration of 3rd, 4th and 5th spiderlings were 24.3, 13.1 and 19.7 days/ females resulted from treated 3rd spiderling, compared with 15.6, 10.0 and 15.3 days/ female untreated respectively, while the duration decreased in male to 23.3, 15.6 and 22.0 days / male treated compared with 25.6, 18.0 and 23.0 days/ male in control and 23.3, 15.6, respectively Also, data in Table (4) recorded that the compound increased the duration female by 16.2 days on contrast decreased the duration male by – 5.7 days compared with control.

Table (4): Biological effect of Wonder on biological aspects after treated 3rd instars spiderling.

Spiderling	Conc.	% Mortality	Duration			Total immature	Increased in duration times
			3rd	4th	5th		
3rd treated	Recommend used 1620 ppm	Female	24.3±1.2b	13.1±1.3b	19.7±1.6b	57.1±1.6b	+16.2
		Male	23.3±0.9a	15.6±1.3	22.0±2.1	60.9±4.6	- 5.7
3rd control		Female	15.6±0.3a	10.0±0.1a	15.3±0.4a	40.9±2.7	---
		Male	25.6±1.3a	18.0±1.2	23.0±1.5	66.6±3.5	---

The means with the same letters at the same column are not significantly different at 0.05%

3.2. Ovipostion period:

Table (5) showed that high significant differences occurred between pre-ovipostion, ovipostion and post ovipostion periods for *K. aulica* when treated and untreated, these periods averaged 20.0, 21.3 days for pre-ovipostion, in treated and control, respectively, while, prolonged to 65.0 days/ female treated compared with 45 days/ female untreated, also, the post ovipostion increased to 28.3 days when adult female treated compared with 20.0 days in control. Compared with 45 days/ female untreated, also, the post ovipostion increased to 28.3 days when adult female treated compared with 20.0 days in control.

3.3.Number of deposited eggs/female:

Data in Table (5) revealed that the number of deposited eggs/sac was affected by treatment. This average was higher for female untreated. Also, data analyses showed a highly significant

differences between the average number of deposited eggs, whereas, female deposited 17.0 eggs/ sac in treated, compared with 26.6 eggs/ sac in control, high significant differences occurred between eggs/sac hatchability percentages resulted when female treated and untreated., the hatching percentages were 65.0 %, when female resulted and fed on the above mentioned prey treated, compared with 96.0 in control. Edwared (1958) recorded 112 eggs in a single egg/sac produced by *C. inclusum,* while, Peck and Whitcomb (1970) reported the occurrence.

3.4.Adult longevity:

Table (5) illustrated that longevity of spider females result from treated was longer about (17.3 days) which estimated by 113.3 days/ female compared with females untreated which was 96.0 days. On contrast, the adult male longevity decreased to 21.6 days when male resulted from treated, its longevity, was decreased (by 9 days) in case of spider males treated male compared with 30.6 days/ male longevity in control.

Table (5): Effect of Wonder compound on fecundity and longevity of *Kochiura aulica***.**

Adults spiderling period	Treated	Control	LSD at 5%
Pre-oviposition	20.0±1.2b	21.0±3.1a	0.25
Oviposition	65.0±5.6b	45.0±12.4a	2.476
Post-oviposition	28.3±3.2b	20.0±1.7a	1.026
Longevity female	113.3±11.7b	96.0±1.2a	2.114
Longevity male	21.6±2.5b	30.6±3.1a	2.681
No of sac/ female	2-3	3-5	Ns
No of egg/sac/ female	17.0±2.5a	26.6±1.9b	1.31

The means with the same letters at the same row are not significantly different at 0.05%

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Survey of insect pests and spiders infesting medicinal and aromatic plants

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Absract:

Medicinal and aromatic plants have now become the main source for medicines, seasonings, colorings , preservatives and represent the oldest and most widespread form of medication. This study was conducted on twenty five of the most important medicinal and aromatic plants in two gardens Zoheria in Cairo and Orman in Giza Governorates. The results indicated that presence of twenty four species of insect pests belongs to fourteen families and five orders. Moreover, eighteen species of spiders belong to nine families and one order (Araneae) were recorded. All these insect pests were presented in low to medium numbers causing moderate damage. From Zoheria garden the largest number of insect pests belonging to orders, Hemiptera, Thysanoptera and Lepidoptera were recorded on sweet marjoram, mints, *Jasminum azoricum* and rosemary. The dominant spider families recorded with largest number of species . These are : Philodromidae and Theridiidae on jasmine flower, lavender and night-blooming jasmine at Zoheria garden whereas, the same families was dominant on rose, rose geranium and chamomile at Orman garden. The dominant insect orders Hemiptera, Thysanoptera, Diptera and Lepidoptera were recorded on rose, sweet basil and calendula at Orman garden.

Introduction

According to the World Health Organization, 80 percent of the population of developing countries relies on traditional plant based systems of medicine to provide them with primary health care needs (Agarwal and Upadhyaya, 2006). The large scale cultivation of these plants in countries may face the problem of sudden appearance of large populations of variety of insect pests in a single crop. Like other plants, medicinal plants too have to bear the devastating effects of injurious insect pests, which are not only harmful for the plant but also, deteriorate the quality of the produce, thus hampering its medicinal value. The majority of the pests caused in feeding damage on reproductive or vegetative organs (those under Thysanoptera and

Heteroptera, including the Thripidae, Miridae, Pentatomidae and Pseudococcidae families), whereas others cause erosion or tunnels on heads (Noctuidae), leaves (beetles) or roots (Elateridae) (Conti, 2003). El-Gendi (2007) in Egypt, recorded eighteen insect species on marjoram and chamomile. *Nysus cymoides* Schill, *Nezara viridula* L., *Lygus gomellatus* H.S., *Nesidocoris tenuis* Reut., *Bourletiella horttensis* (Fitch), *Empoasca decipiens* Paoli, *Trupanea stellata* Fuessly, *Myzus persicae* (Sulz.) and *Aphis gossypii* Glover were the main insect pests on chamomile. Thirteen species of insect pests belong to eight families and five orders were recorded on Calendula plants. While twelve insect pests belong to nine families under six orders were recorded on chamomile plants (Solaiman, 2015).

True spiders are one of the most abundance predatory groups in terrestrial ecosystems. Spiders have proved to be beneficial in regulation of agricultural pests and their role as natural enemies has recently been more and more stressed (Ghabbour *et al.,* 1999). The presence of spiders on some ornamental plants in Egypt was studied at the first time by (Shereef *et al.*, 1996); after that (Rizk *et al.,* 2012) studied the incidence of medicinal and ornamental plants in El-Fayoum Governorate. Ghallab (2013) was collected one family of spider from lantana and croton ornamental plants from Orman garden and the most abundant families were Miturgidae, Philodromidae, Salticidae, Theridiidae and Araneidae. Zoheria and Orman gardens were the most harbored spider. The most dominant families' recorded with the largest number of species were Salticidae, Gnaphosidae, Thridiidae and Oonopidae (Hassan *et al.,* 2016).

The information regarding the occurrence of insect pests on medicinal and aromatic plants of the state is scanty. Hence the present study was undertaken to record the insect pests and spiders associated with important medicinal and aromatic plants in countryside.

Materials and methods

This work was conducted on medicinal and aromatic plants during two successive years 2017 and 2018 in two locations, Zoheria garden in Cairo and Orman garden in Giza Governorates in Table (1). Twenty five double strokes were randomly from medicinal and aromatic plants (twigs, leaves and flowers) from January to December. Samples were collected in a polyethylene bags and transferred to laboratory. To kill insects, piece of cotton moistened with chloroform put in each sample and left for 15 minutes. The sample was emptied in Petri dish and cleaned from plant residues. Then it was examined under stereomicroscope to separate and count the major insect pests. This process was performed at weekly intervals throughout the entire period of investigation. Specimens were mounted for light microscopy according to the procedure detailed by Kosztarab and Kozár (1988).

The spider individuals were collected biweekly during two hours from (9-11) during summer and 10-12 in winter on fine silky traps, collected true spiders from branches, leaves and trunks of different trees and bushes from 25 medicinal and aromatic plants. The spiders were isolated and counted in glasses individually and transferred in the same day to the laboratory at the Plant Protection Research Institute for counting and identification. The collected spiders were kept in small test tubes containing 70% ethyl alcohol. The necessary information (locality, host plant and date) was recorded by a pencil on a slip of paper attached to each specimen inside the tube for identification. The characteristics of families, genera and species were examined using the related keys. In contrast, some specimens were identified

just on the possible genus level. Identification of specimens followed the descriptions of (Petrunkevitch, 1939; Kaston, 1978 and Jocqué and Dippenaar-Schoeman, 2007).

Table (1) : Common and scientific names of the medicinal and aromatic plants in Zoheria and Orman gardens:

Garden name	Medicinal and aromatic plants	
	Common name	**Scientific names**
Zoheria garden	Aloevera	*Aloe vera* (L.)
	Calotropis procera	*Calotropis procera* (Aiton)
	Jasminum azoricum	*Jasminum azoricum* L.
	Jasmine flower	*Jasminum sambac* (L.) Aiton
	Jasmine shami	*Jasminum officinale* L.
	Lavende*r*	*Lavandula angustifolia* Mill
	Mints	*Mentha spp.*
	Murraya	*Murraya paniculata* (L.) Jack
	Myrtus	*Myrtus communis* L.
	Night-blooming jasmine	*Cestrum nocturnum* L.
	Pencil tree	*Euphorbia tirucalli* L.
	Rosemary	*Rosmarinus officinalis* L.
	Sweet marjoram	*Origanum majorana* L.
	Vinca	*Catharanthus roseus* (L.) G.Don.
Orman Garden	Calendula	*Calendula officinalis* L.
	Carnation	*Dianthus caryophyllus* Lim
	Chamomile	*Chamaemelum nobile* (L.)
	Jasminum	*Jasminum grandiflorum* L
	Marigold	*Tagetes erecta* L.
	Mentha piperata	*Mentha piperata* L.
	Neem	*Azadirachta indica Juss.*
	Rose	*Rose* spp
	Rose geranium	*Pelargonium graveolens* L.
	Sweet basil	*Ocimum basilicum* L.
	Thyme	*Thymus vulgaris* L.

Results and discussion

A general survey was conducted on twenty five medicinal and aromatic plants at two gardens Zoheria in Cairo and Orman in Giza Governorates during two successive years 2017-2018. The results indicated that presence of twenty four species of insect pests belongs to fourteen families and five orders. Moreover, eighteen species of spiders belong to nine families and one order (Araneae), were recorded in 2017 and 2018 seasons (Tables, 2-5).

1. Survey of insect pests and spiders infesting medicinal and aromatic plants at Zoheria garden in Cairo Governorate:

The results in Tables (2 and 3) proved that fourteen true spiders belonging to seven families was recorded on fourteen medicinal and aromatic plants (aloevera, calotropis procera, *Jasminum azoricum*, jasmine flower, *Jasmine shami*, lavende*r*, mints, murraya, myrtus, night-blooming, jasmine, pencil tree, rosemary, sweet marjoram and vinca). Members of Philodromidae and Theridiidae were the dominant spider families record 63 and 55 individuals. The highest numbers of their occurrence was collected from jasmine flower, lavender and night-blooming jasmine. The family Philodromidae was presented by two species *Thanatus albini* Audouin and *Phildromous* sp. while, the family Theridiidae presented by four species *Theridion melanosticum* O. P. Cambridge, *Kochiura aulica* (Koch), *Theridion spinitarse* O. P. Cambridge and *Theridion* sp. Three families were representing by only a single individuals. These were Dictynidae, Eutichuridae and Uloboridae, members of the remaining families were found in few numbers. The obtained results are in harmony with that detected by Rizk *et al.,* 2012 and Hassan *et al.,* 2016.

Also, the obtained results in Table (2 and 3) indicated that sixteen insect species belonging to eight families and four orders on fourteen medicinal and aromatic plants. Members of Hemiptera, Thysanoptera and Lepidoptera were the dominant insect families recorded 88, 52 and 22 individuals, respectively . The highest numbers of their occurrence were collected from sweet marjoram, mints, *Jasminum azoricum* and rosemary recorded 30, 26, 16 and 15 individuals, respectively. Hemiptera was presented by five families: Aphididea, Aleyrodidae, Cicadellidae, Diaspididae and Monophlebidae. The most dominant insect from family Monophlebidae is *Icerya aegyptiaca* Douglas on aloevera and night-blooming jasmine; while the highest percent of members of the family Aphididae is *Aphis gossypii* Glover on mints and murraya. Thysanoptera presented by single family Noctuidae including four species *Agrotis ipsilon*, *Agrotis* spp**.,** *Spodoptera exigua* and *Spodoptera littoralis*. Whereas the order Thysanoptera presented by a single family Thripidae which including four species *Frankliniella tritici, Haplothrips cottei, Thrips orientalis* and *Thrips palmi.*

Table (2): Survey of insect pests and spiders infesting medicinal and aromatic plants at Zoheria garden in Cairo Governorate during 2017.

Order	Family Species	Aloevera	Calotropis procera	Jasminum azoricum	Jasmine flower	Jasmine Shami	Lavender	Mints	Total
Araneae	**Cheiracamthiidae** *Cheiracanthium inclusum* O. P. Cambridge					+	++		**10**
	Philodromidae *Phildromous* sp.	+							**2**
	Thanatus albini (Audouin)		++	+	+		++	+	**22**
	Salticidae *Thyene* sp.					+			**2**
	Thyene imperialis (Rossi)				+				**2**
	Euophrys sp.				+				**2**
	Theridiidae *Theridion melanosticum* O. P. Cambridge	+							**3**
	Kochiura aulica (Koch)	++			+++				**13**
	Theridion sp.						+	++	**10**
	Theridion spinitarse O. P. Cambridge				+		++		**10**
	Thomisidae *Thomisus spinifer* O. P. Cambridge			+	+				**4**
	Synema sp.				++				**8**
	Uloboridae *Uloborus* sp.	+							**2**
Total		**14**	**8**	**4**	**33**	**4**	**26**	**10**	
Hemiptera	**Aphididea** *Aphis gossypii* Glover							++ +	**8**
	Aphis nerii (Boyer)		+						**2**
	Myzus persicae (Sulz.)							+	**3**
	Diaspididae *Aonidiella aurantii* (Maskell)					+			**2**
	Diaspis echinocacti (Bouche)	+							**3**
	Monophlebidae *Icerya aegyptiaca* (Douglas)	++							**8**
Lepidoptera	**Noctuidae** *Agrotis ipsilon* (Hufnagel)				+				**3**
	Agrotis spp.							+	**3**
	Spodoptera exigua (Hubner)				+			+	**6**
	Spodoptera littoralis (Boisduval)							+	**3**
Orthoptera	**Gryllotalpidae** *Gryllotalpa gryllotalpa* l.							+	**3**
Thysanoptera	**Thripidae** *Frankliniella tritici* (Fitch)			++				++	**16**
	Haplothrips cottei (Vuill.)			++					**9**
	Thrips orientalis (Bognall)					++			**9**
Total		**11**	**3**	**16**	**6**	**11**	**-**	**26**	

+ = low number (1 to 3 individuals); ++ = midum no. (3 to 9 individuals); +++ = high no. (up to 9 individuals)

Table (3): Survey of insect pests and spider infesting medicinal and aromatic plants at Zoheria garden in Cairo Governorate during 2018.

Order	Family Species	Murraya	Myrtus	Night-blooming jasmine	Pencil tree	Rosemary	Sweet marjoram	Vinca	Total
Araneae	**Dictynidae** *Dictyna* sp.							+	**2**
	Philodromidae *Thanatus albini* (Audouin)			+++		++	+		**17**
	Phildromous sp.	+							**2**
	Theridiidae *Theridion melanosticum* O. P. Cambridge	+	+		+				**6**
	Kochiura aulica (Koch)							+	**2**
	Theridion sp.		+			+	+		**6**
	Theridion spinitarse O. P. Cambridge	+			+		+		**6**
Total		**6**	**4**	**15**	**2**	**10**	**8**	**10**	
Hemiptera	**Aleyrodidae** *Bemisia tabaci* (Gen.)					+	++		**12**
	Aphididea *Aphis durantae* Theobald						+		**3**
	Aphis gossypii Glover	++					+		**11**
	Myzus persicae (Sulz.)					+	+		**7**
	Cicadellidae *Empoasca lybica* **(de Bergevin and Zanon)1922**						++		**9**
	Diaspididae *Chrysomphalus ficus* (Ashmead)	+							**3**
	Monophlebidae *Icerya aegyptiaca* (Douglas)			++				+	**13**
	Icerya purchase Maskell				+				**3**
Lepidoptera	**Noctuidae** *Agrotis ipsilon* (Hufnagel)						++		**4**
	Spodoptera littoralis (Boisduval)						+		**3**
Thysanoptera	**Thripidae** *Thrips palmi* Karny		+ +			++			**18**
Total		**12**	**9**	**8**	**3**	**15**	**30**	**3**	

+ = low number (1 to 3 individuals); ++ = medium no. (3 to 9 individuals); +++ = high no. (up to 9 individuals)

2. Survey of insect pests and spiders infesting medicinal and aromatic plants at Orman garden in Giza Governorate:

This survey is considered an essential work in studying the survey of spiders and insects that attacked eleven medicinal and aromatic plants at Orman garden in Giza Governorate (Calendula, carnation, chamomile, *Jasminum grandiflorum*, marigold, *Mentha piperata*, neem, rose, *Rose geranium*, sweet basil and thyme). These plants received eight spider families consists of sixteen species. In Tables (4 and 5) the number of collected spiders associated members of Theridiidae, Philodromidae and Thomisidae were the dominant spider families recorded 94, 45 and 42 individuals, respectively. While other remaining families Salticidae, Eutichuridae, Dictynidae, Lycosidae and Filistatidae composed of 21, 16, 10, 6 and 2 individuals, respectively. The highest numbers of spider occurrence were collected from rose, *Rose geranium*, chamomile, sweet basil, neem and *Mentha piperata* composed of 52, 39, 35, 28, 20 and 20 individuals, respectively. While marigold and carnation received the lowest number of spider of 10 and 8 individuals, respectively.

As similar results in Zoheria garden the family Philodromidae was presented by two species while, Theridiidae by three species *T. melanosticum*, *K. aulica* and *Theridion* sp. whereas, Thomisidae by four species *Misumena atrocincta, Synema candicans* , *Thomisus spinifer* and *Thomisus* sp. Four families were representing by only a single individual. These were Dictynidae, Eutichuridae, Filistatidae and Lycosidae members.

Tables (4 and 5) indicated that twenty two insect species belonging to thirteen families and five orders on eleven medicinal and aromatic plants. Members of Hemiptera, Thysanoptera, Diptera and Lepidoptera were the dominant insect families composed of 80, 61, 45 and 38 individuals, respectively . The highest numbers of their occurrence were collected from rose, sweet basil, calendula, mentha, carnation and marigold recorded 45, 38, 35, 29, 23 and 22 individuals, respectively. Hemiptera was presented by six families: Aphididea, Aleyrodidae, Diaspididae, Miridae, Monophlebidae and Pentatomidae. The most dominant insect from family Aphididea is *A. gossypii* on rose, sweet basil, calendula and mentha. Thysanoptera presented by two families Noctuidae including four species and Pieridae including one species. Whereas the order Thysanoptera presented by a single family Thripidae which including five species the most dominant insect *Frankliniella occidentalis* on marigold, rose and carnation.

Similar results were obtained by (Abd El-Raheem and Abd EL-Wareth, 2015) who reported that chamomile crop infesting *Myzus persicae* (Sulzer) was the numerous pest as compared with the other insect pests (54.17% and 71.70%). The highest level of abundance was recorded during March and April. Also, the results indicared that 15 insects included seven sap-suckers, seven predators and one parasitiod on German chamomile, where the common insect pests were *M. persicae*, *N. cymoides*, *L. gomellatus* and *N. tenuis*. Chamomile plants are known to harbor many species of insects and mites including *M. persicae, A. gossypii*, *E. decipiens* and *N. cymoides* (Etman *et al.,* 1990 and Rahil, 2005).

Table (4): Survey of insect pests and spiders on medicinal and aromatic plants at Orman Garden in Giza Governorate during 2017.

Order	Family Species	Calendula	Carnation	Chamomile	Jasminum grandiflorum	Marigold	Mentha piperata	Total
Araneae	**Dictynidae** *Dictyna* sp.			++				**8**
	Lycosidae *lycosa cingara* C.L. (Kock)			+				**2**
	Philodromidae *Thanatus albini* Audouin	++	+			+	++	**20**
	Phildromous sp.		+					**2**
	Salticidae *Thyene sp.*			+++				**15**
	Thyene imperialis (Rossi)	+						**2**
	Euophrys sp.				+		+	**4**
	Theridiidae *Kochiura aulica* (Koch)			++	++	++	++	**16**
	Theridion sp.	+			+			**4**
	Theridion spinitarse O. P. Cambridge			+	+	+		**12**
	Thomisidae *Thomisus sp.*		+		+		+	**6**
	Synema candicans O. P. Cambridge		+		+			**4**
Total		**12**	**8**	**35**	**10**	**10**	**20**	
Diptera	**Agromyzidae** *Liriomyza trifolii* (Burgess)	++						**9**
	Melanagromyza sojae (Zehntner)	++				++		**20**
	Tephritidae *Trupanea stellata* (F.)			+		+		**7**
Hemiptera	**Aphididea** *Aphis gossypii* Clover	+					+	**8**
	Myzus persicae (Sulz.)		++				+	**13**
	Miridae *Nesidocoris tenuis* Reut.	+						**4**
	Pentatomidae *Nezara viridula* L.			++				**9**
Lepidoptera	**Noctuidae** *Agrotis segetum* (Denis and Schiffermiller)	+						**4**
	Agrotis spp**.**						+	**3**
	Spodoptera exigua (Huebner)						+	**4**
	Spodoptera littoralis (Boisduval)	+					+	**8**
	Pieridae *Pieris brassicae* L.	+						**3**
Orthoptera	**Acrididae** *Aiolopus stripins* Latreille	+						**4**
	Gryllotalpidae *Gryllotalpa gryllotalpa* L.						+	**4**
Thysanoptera	**Thripidae** *Frankliniella occidentalis* (Pergand)		++				++	**18**
	Haplothrips cottei (Vuill.)		+		++			**12**
	Neohydatothrips samayunkur (Kudô)					++		**9**
Total		**35**	**23**	**13**	**9**	**22**	**29**	

+ = low number (1 to 3 individuals); ++ = midum no. (3 to 9 individuals); +++ = high no. (up to 9 individuals)

Table (5): Survey of insect pests and spiders on medicinal and aromatic plants at Orman Garden in Giza Governorate.

Order	Family Species	Neem	Rose	Rose geranium	Sweet basil	Thyme	Total
Araneae	**Cheiracamthiidae** *Cheiracanthium inclusum* O. P. Cambridge		++			+	**10**
	Filistatidae *Filista* sp.	+					**2**
	Lycosidae *Lycosa* sp.		+	+			**4**
	Philodromidae *Thanatus albini* Audouin		+++		+	++	**25**
	Salticidae *Thyene imperialis* (Rossi)				++	+	**10**
	Theridiidae *Theridion melanosticum* O. P. Cambridge	++	+++	+			**25**
	Kochiura aulica (Koch)	++	+	+++	+	++	**27**
	Theridion sp.		++		+		**10**
	Thomisidae *Thomisus spinifer* O. P. Cambridge	+		+	++		**12**
	Synema candicans O. P. Cambridge	+			+		**4**
	Synema sp.		+	++	+	+	**14**
	Misumena atrocincta Costa				+		**2**
Total		**20**	**52**	**39**	**28**	**14**	
Diptera	**Agromyzidae** *Liriomyza trifolii* (Burgess)				++		**9**
Hemiptera	**Aleyrodidae** *Bemisia tabaci* (Gen.)	+		++	+		**15**
	Aphididea *Aphis craccivora* Koch		++				**8**
	Aphis gossypii Glover		++		++		**19**
	Myzus persicae (Sulz.)		+				**4**
	Diaspididae *Aonidiella aurantii* (Maskell)		+				**3**
	Monophlebidae *Icerya aegyptiaca* (Douglas)	+	++	+			**4**
	Pentatomidae *Nezara viridula* L.				+		**3**
Lepidoptera	**Noctuidae** *Spodoptera exigua* (Huebner)	+			+		**7**
	Spodoptera littoralis (Boisduval)				++		**9**
Thysanoptera	**Thripidae** *Frankliniella occidentalis* (Pergande) (Pergand)		++				**8**
	Thrips palmi Karny		+			+	**7**
	Thrips tabaci Lind.		++				**9**
Total		**10**	**45**	**9**	**38**	**4**	

+ = low number (1 to 3 individuals); ++ = midum no. (3 to 9 individuals); +++ = high no. (up to 9 individuals)

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Histopathological and ultrastructural impacts of nuclear polyhedrosis virus on infected tissues of the leopard moth *Zeuzera pyrina* (Lepidoptera: Cossidae) using transmission electron microscopy

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Abstract:

Leopard moth, *Zeuzera pyrina* (L.) (Lepidoptera: Cossidae) is considered one of the most dangerous and destructive wood boring pests that attack several varieties of the horticultural fruit trees mainly the apple trees, *Malus domestica* B. (Rosales: Rosacease). In Egypt, this borer causes severe crop loss resulting in serious detrimental and economic damage thus continuous control trials are taken place aiming to avoid these key problems. Through the ongoing study surveying visits were done to collect and sample the natural infected stage of this cossid borer. Then a series of experiments was performed to isolate and trace of the viral pathogen which is identified as the *Zeuzera pyrina* nuclear polyhedrosis virus **(**ZPNPV). Then testing the susceptibility of *Z. pyrina* larvae to this virus was done at three different controlled temperatures (25±5°C) in the laboratory. Screening tests revealed high mortality rates for the larval stage at 20°C recording 85.7±0.38 %. Ultimate sequence of experiments was applied to conduct histopathological and ultrastructural studies using transmission electron microscopy. Gained results showed severe malformation and distortion of the mid-gut cells confirming the susceptibility of *Z. pyrina* to the nuclear polyhedrosis viral infection and possibility of ZPNPV to be applied within the control programs of this cossid borer. It is concluded that *Z. pyrina* revealed a definite susceptibility to the infection with ZPNPV and it is highly provoked to be involved in the integrated pest management programs against this dangerous moth.

Introduction

Leopard moth, *Zeuzera pyrina* (L.) (Lepidoptera: Cossidae) is a highly destructive wood boring pest attacking several horticultural fruit trees mainly the apple trees, *Malus domestica* B. (Rosales: Rosacease) in several global regions especially the Mediterranean area including Egypt causing serious damage and considerable crop loss (Navon, 1977; Katlabi, 1989; Ismail *et*

al., 1988 and 1992; Guario *et al.*, 2000; Hegazi and Khafagi, 2005; Kutinkova *et al.*, 2006; Merghem, 2012; Mani *et al.*, 2014 and Manja and Aoun, 2019). Thus it was of great importance to combat this devastating boring pest, control measures for this borer are mainly focused chemical applications.

In Egypt, the control and management programs against this borer are depending mainly on pesticides applications for many years and still up till now. Excessive chemical sprays lead to high hazard risk, resistance problems and dangerous health effects for human, livestock and beneficial insects (Tadros *et al.*, 1993; Haniotakis *et al.*, 1999; Sarto, 2001; Osuna and Patanita, 2006; Patanita *et al.*, 2009 and Almanoufi *et al.*, 2012).

Consequently, searching for safe and non-chemical or alternative agents is encouraged aiming at maximizing the efficiency of that insect boring pest's control program and preventing the detrimental, residual and adverse effects resulting from chemical pesticide applications such as biocontrol agents. Unfortunately, biological control elements were restrictedly applied against *Z. pyrina* such as entomogenous nematodes and ectoparasitoids in addition to some scanty trials using microbial bioagents such as bacteria and fungi (Abdel-Kawy *et al.*, 1992; Nashnosh *et al.*, 1993; Tawfik and Ramadan, 2006; Lawrence *et al.*, 2007; Japoshvili and Hansen, 2013; Merghem and Hassan, 2014 and Labaude and Griffin, 2018).

Viral disease infections, especially the nuclear polyhedrosis viruses (NPV), as microbial entomopathogenic agents are still used on a limited scale for the control of the wood boring pests especially this cossid borer thus there was a need to estimate the efficiency of such biocontrol agents against this boring pest.

Present study was undertaken to explore the susceptibility of the wood boring pest *Z. pyrina* to the viral disease as a biocontrol agent for it. Moreover, focusing on the isolation of viral diseases that naturally infect the stages of this cossid borer was undertaken. Ultimately concern with the viral impacts on the larval tissues of *Z. pyrina*, through histopathology and electron microscopy investigations was studied.

Materials and methods

1. Field surveyed visits:

During the current study, different localities of the host horticultural trees of *Z. pyrina*, mainly the apple trees, had been visited throughout 2016 to 2017 searching for the naturally diseased individuals in Qalubeia and Beheira Governorates.

2. Sampling, preservation and identification:

Collected intact, health and moribund specimens of this target boring pest were collected and preserved separately in glass vials containing 0.55% sterile saline solution. Then samples were subjected to a preliminary identification for the collected stages of the boring insect done at the Department of Wood Borers and Termite in the Plant Protection Research Institute (PPRI) followed by a confirmation of this borer identity undertaken at the Entomological Collection of the Classification and Taxonomy Research Department of the same research institute (*i.e.* PPRI) and finally it was revealed that it is the leopard moth, *Z. pyrina*.

Diseased specimens were collected from the surveyed localities of the fore-mentioned Governorates. Then the viral-shown specimens were separated in the saline solution for the further pathological experiments and the moribund symptoms were recognized according to the identification keys after Vlak and Grőner (1980) and Evans and Shapiro (1997). Dissection of infected larvae was done and the digestive guts

were obtained for the following microscopic examination elucidating the internal symptoms as the presence of the virions and inclusion bodies virus (IBV) in the infected tissues with considerable numbers, dispersion of air sacs and vacuolation within cells and destruction of the nuclear membrane of target cells. Both histological and electron microscopy studies confirmed the identity of a NPV infection to this cossid borer called *Zeuzera pyrina* nuclear polyhedrosis virus (ZPNPV).

3. Laboratory screening tests:

On identifying the viral pathogen type, a second series of experiments dealing with the susceptibility of this leopard moth borer to the infection of ZPNPV, was undergone. Firstly, a stock viral suspension that would be used further for the screening tests was prepared through using the diseased larvae. Moribund larval guts were grinded with a sterile mortar till complete dissolve in 2ml sterile distilled water (SDW) tube with a ratio of 1:1 to tissues used. Then, the obtained suspension after a skimming process was stored as a stock for the further viral infectious treatments against healthy larvae of *Z. pyrina* and was keep in 4°C. Then, a suspension solution of $3x10^{10}$ IBV/ 100ml SDW concentration (*i.e.* $3x10^{8}$ IBV/ml) was prepared and was applied for each individual larva of this moth borer and the exposure treatment was done within a suitable Petri dishes. The screening tests were done at three controlled temperatures (25±5°C) in the laboratory with inspection process for a week and a fortnight periods, and seven replicates were used for each temperature degree both to control and treated checks. Mortality rate percentages of the inoculated *Z. pyrina* larvae were then recorded and corrected with Abbott's formula according to Abbott (1925).

4. Histopathology and electron microscopy studies:

4.1. Histopathological study:

To achieve the purpose of the histopathological study and the further investigation with electron microscope, a sequence of laboratory steps was followed beginning with the dissection of the larval body, removal of visceral and fat tissues and getting the gut (*i.e.* the mid-gut). Then, a procedure of micro-technique preparation was provided following by the dehydration, clearing, embedding, sectioning, staining, and fixation processes till the full examination, this procedure was according to Hamm (1999).

4.2. Electron microscopy study:

IBVs were randomly selected then suspended in a 50:50 glycerin/water solution and measured at x1,250 (phase contrast) with a micrometer hence were pelleted from an aqueous suspension at 15,000 rpm for 10min in an Eppendorf centrifuge. The bodies were fixed in 1.5% glutaraldehyde (pH 7.2) at 4°C for 2 hr; washed in 5% sucrose-sodium cacodylate buffer (pH 7.2) for 4 hr; and then fixed in 2% osmium tetroxide for 1 hr), washed in sucrose-sodium cacodylate buffer (10 min), and dehydrated in a series of 20, 40, 60, 80,90, and 100% ethanol. After remaining in 100% ethanol for 30 min, they were pelleted at 15,000 rpm for 10 min. The pellets were infiltrated and embedded in Spurs medium, sections ranged from 0.7 to 1.0 mm were cut on an Ultratome and stained with 1% ethanolic uranyl acetate for 10min, followed by a deionized water rinse and lead citrate stain for 2 min. They were then rinsed in sterile SDW, dried, and observed with the electron microscope at 80 kV as transmission electron microscopy (TEM) studies. This offered electron microscopy procedure was presented by Bud and Kelly (1977), Mclntosh and Ignoffo (1986) and Tanada and Kaya (1993) and it was taken place at the Electron Microscopy Unit, National Research Center.

5. Statistical analysis:

Obtained data were statistically analyzed according to Finney (1971).

Results and discussion

1. The viral isolation and identification:

Figure (1) showed that an electron micrograph of a viral suspension smear, at different magnifying powers (x), prepared from the infected larval stages of *Z. pyrina*. It reveals the dimensional hexagonal shape of the inclusion bodies (IBVs) for ZPNPV; the nuclear polyhedrosis virus isolated from the infected larval stages of *Z. pyrina*.

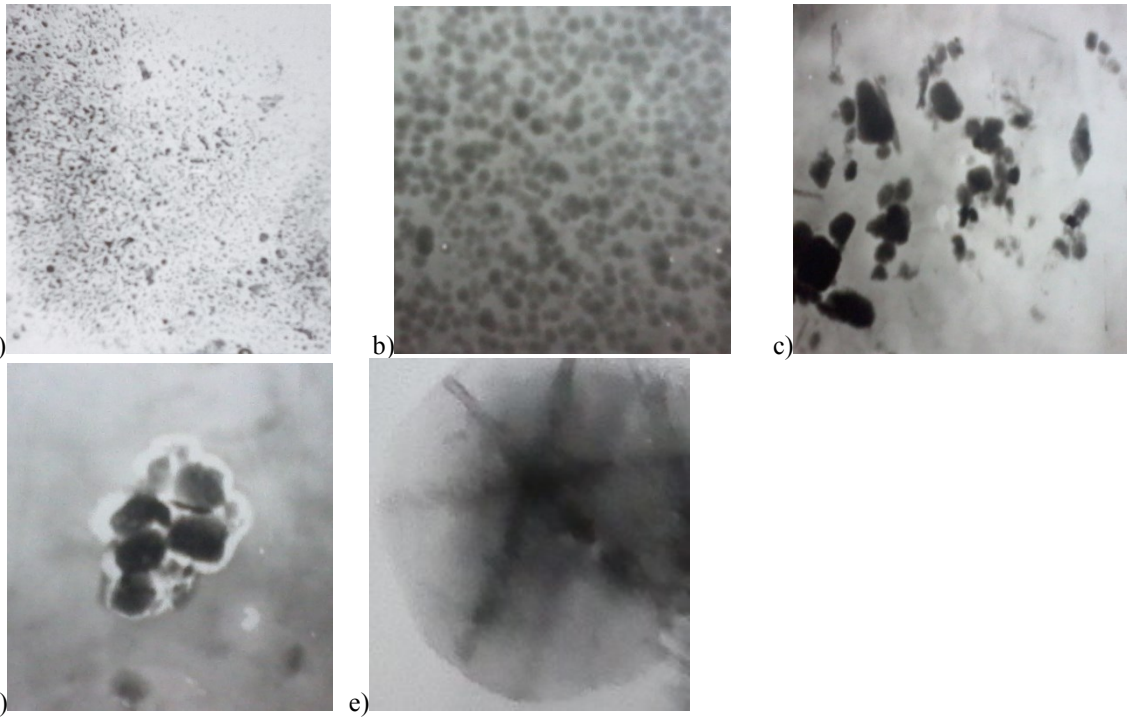

Figure (1): Electron micrograph of a smear of the viral suspension showing hexagonal inclusion bodies virus (IBVs) of the *Zeuzera pyrina* nuclear polyhedrosis virus (ZPNPV) at magnifying power ($x10^3$): a)12.5, b)16, c)20, d)40, e)60.

The finding that leopard moth borer, which belongs to the lepidopteran members, is susceptible to the NPV infection is matching with the results of Bud and Kelly (1977), Vlak and Grőner (1980) and Mclntosh and Ignoffo (1986) with noctuids and Shapiro (1992) with lymantriids which confirm the infectivity of the nuclear polyhedrosis viruses to lepidopterous families.

2. Laboratory screening tests:

Figure (2) elucidates the efficacy of ZPNPV; the nuclear polyhedrosis virus of *Z. pyrina* when inoculated to the larvae of this cossid borer at the controlled temperatures 25±5°C.

This efficacy is represented by the average mortality rates resulted from the inoculation treatments with the concentration of $3x10^8$ IBV/ml. Mortality rates were found to vary significantly at ($P<0.05$) as each temperature degree resulted in an average mortality rate, indicating the efficacy of the viral suspension, significantly different from the two other average rates of the two rest temperatures. It is also concluded that the increase of the temperature degree is diversely proportional with the viral efficacy as it is obvious that the highest controlled temperature (30°C) revealed the least mortality 42.9±0.54 % meanwhile the figure is reflected with the lowest degree (20°C) which recorded a rate of 85.7±0.38 %.

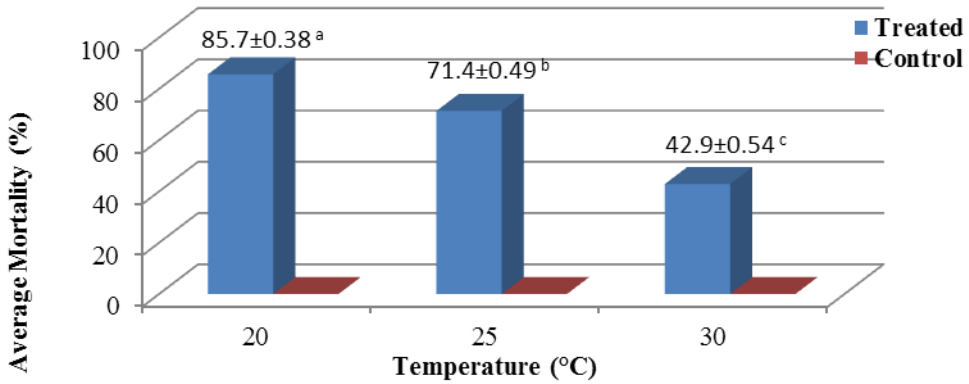

Figure (2): Efficacy of 3x10^8 inclusion bodies virus(IBV) /ml *Zeuzera pyrina* nuclear polyhedrosis virus (ZPNPV) concentration when inoculated to *Zeuzera pyrina* larvae at controlled temperatures (25±5°C).

3. Histopathological study:

Figure (3a) indicates the invasion of the nuclear polyhedrosis virus of the *Z. pyrina*; ZPNPV at the concentration of 3x10^8 IBV/ml. showing the viral replication and IBVs distribution through the mid-gut tissue cells of this cossid borer. The basement membrane of the epithelial tissue section reflects the impact of the viral replication and the invasions of the IBVs appearing the distortion of the epithelial cells near the membrane layer. The cytoplasm is filled with a lot of air sacs and vacuoles due to the viral infections.

Figure (3b) reveals an apparent contrast with the former Figure (3a) as it show an intact tissue lining with a firm in row of the adjacent cells. It is free from the IBVs presence in addition to the lack of either the air sacs or any vacuolation.

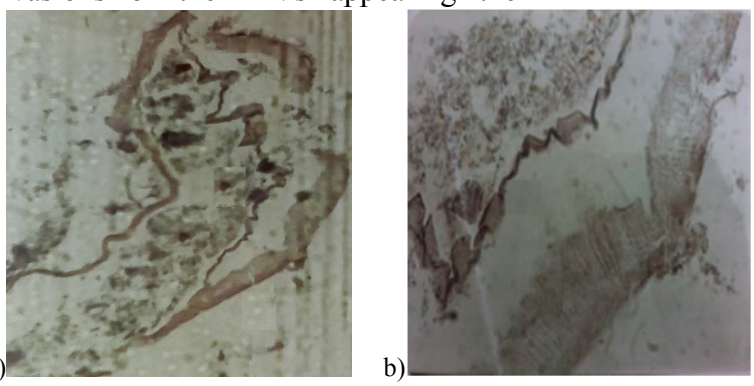

Figure (3): A photomicrograph of a longitudinal section of the mid-gut region of *Zeuzera pyrina* showing: a) the invasion of the inclusion bodies virus (IBVs) , distortion of the basement membrane and air sacs; and b) the normal intact membrane layer and lacking of vacuolation.

The photomicrograph in Figure (4) demonstrates the severe and detrimental effects of the advancing infection resulted due to the viral dispersion through the mid-gut epithelial cells of *Z. pyrina*. Figure (4a) represents the complete invasion of the IBVs to the epithelial cells of the mid-gut and Figure (4b) verified the destruction of the basement membrane layer with the bursting of the included epithelial cells with its cellular contents.

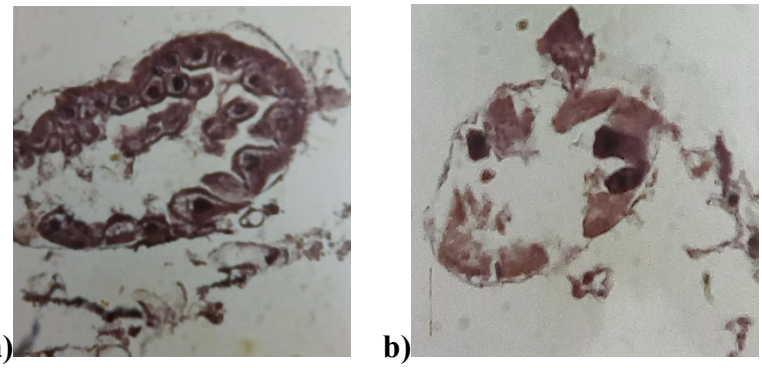

Figure (4): A photomicrograph of a longitudinal section of the mid-gut region of *Zeuzera pyrina* showing: a) the advanced and complete invasions to the epithelial cells; and b) the nearby detrimental symptoms of the inclusion bodies virus (IBVs) invasions leading to the distortion of the basement membrane and the epithelial cells.

The forementioned findings confirmed the susceptibility of this cossid borer to the infection by the viral attacks through its specific NPV *i.e.* ZPNPV. Similar findings were found by Hostetter *et al.* (1990) about the infectivity of a nuclear polyhedrosis virus of the yellow striped army worm (Lepidoptera: Noctuidae) encouraging the wider usage of the viral diseases infections to control such lepidopterous borers.

4. Electron microscopy study:

Figure (5) demonstrates the infection with the IBVs of ZPNPV to the mid-gut cells of *Z. pyrina* on the scale of ultrastructural technique using the transmission electron microscopy (TEM).

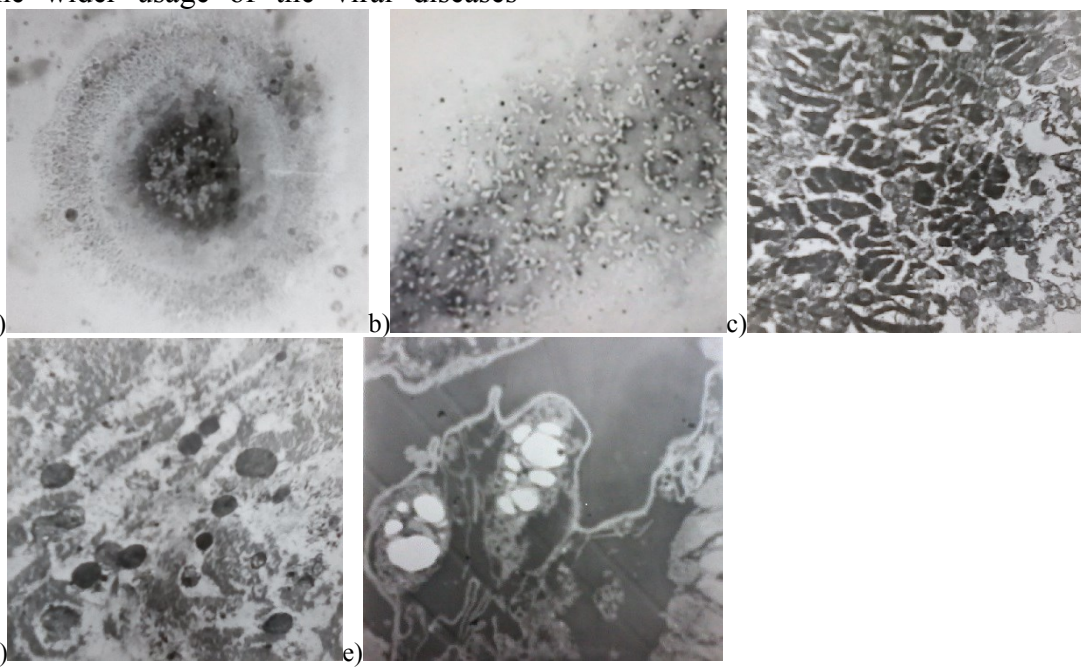

Figure (5): Electron micrograph of a transverse section of the infected mid-gut of *Zeuzera pyrina* with inclusion bodies virus (IBVs) of the *Zeuzera pyrina* nuclear polyhedrosis virus (ZPNPV) showing: a) infection of the mid-gut section with IBVs; b) magnified part of the basement membrane indicating the infection; c) scattering of the inclusion bodies virus (IBVs) invasions through the mid-gut tissues; d) multiplied IBVs within the nuclear matrix; and e) appearing of multiple vacuoles and air sacs; this was at magnifying power (x10^3): a)6.3, b)12, c)10, d)16, e)25.

This electron micrograph of the TEM in Figure (5) points out the infected mid-gut of *Z. pyrina* with IBVs of the ZPNPV showing different symptoms resulting from these viral infections to the mid-gut region and facilitates understanding the infection process of the NPV through the tissues of this lepidopterous borer. The TEM examinations revealed the presence of a

ZPNPV virus which is specific for *Z. pyrina* invading the nuclei of the epithelial cells of the mid-gut region. These observations are concordant with those of Mclntosh and Ignoffo (1986) who studied the impacts of the viruses on the cell structure. Thus, it is observed that both histopathological and electron microscopy studies confirmed the infection of a NPV to this leopard moth borer named ZPNPV.

Subsequently, it is concluded that the leopard moth borer revealed a definite susceptibility to the infection with ZPNPV and it is highly provoked to be involved in the integrated pest management programs against this dangerous boring moth, *Z. pyrina*.

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Classical classification and discrimination analysis of physicochemical characters of Sidr honey produced in some Arab countries

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Abstract:

Sidr honey is one of the best and most expensive monoflora honeys world wide. The aim of this work is to use discriminate analysis for classifying of blossom Sidr honey from Egypt, Libya, Algeria and Yemen countries. All data were statistically tested using analysis of variance. Discriminate analysis was used to identify the most important parameters in the classification. The development of partial least square discriminate analysis (PLS-DA) model on validation gave 100% correct classification of the samples. The results showed that, Sidr honey from Egypt and Algeria could not be assigned by 100% into their actual groups even when all parameters were used simultaneously in the analysis as well as Libya and Yemen. Different parameters were discussed in the light, electrical conductivity (EC), pH, free acidity, lactone, fructose, glucose, sucrose and maltose contents accounted for the most discrimination parameters between the different Sidr honey in Arab countries and the effectiveness of the chosen parameters to characterization and discrimination. All tested honey samples were within the level allowable by the international standards for honey quality. The application of the discriminate technique (PLS-DA) presented the best for discriminating Sidr honey in Arab countries.

Introduction

Sidr honey is famous in Arab countries, it was used for both nutritional and therapeutic purposes and its price attains quite high levels, the quality control of local and imported honey is completely inadequate. Properties and compositions of bee honey depend on its geographical floral origin, season, environmental factors and treatment of beekeepers (Kasˇkonien *et al*., 2010 and El-Metwally, 2015). Honey consists primarily of sugars, at most fructose (40–50%) and glucose (32–37%), little amounts of sucrose (<2%) and mineral constituents (ash less than 0.1%). Honey also contains water (13–20%) (Alvarez-Suarez *et al*., 2013). Moisture content of bee honey represents a major importance to its stability against granulation and fermentation. The low moisture content conserves honey from microbiological activity and thus it can be preserved for longer periods (Buba *et al.*, 2013; Akhtar *et al*., 2014 and El-Metwally, 2015).

Honey contains at least 181 components (White, 1975). Although the major fundamental of honey are nearly the same in all honey samples, physical properties and the precise chemical composition of natural honeys differ according to the plant species on which the bees forage (James *et al*., 2009 and Cantarelli *et al*., 2008). This situation does not allow a sufficient protection of the consumer and facilitates possible frauds. Indeed, at the scientific level, only a few data are available: a research carried out on Sidr honey samples, to contribute more to the knowledge of Sidr honey produced in some Arab countries (Egypt, Algeria, Libya and Yemen), further investigated the subject, with the aim of evaluating, the quality of Sidr honey, verifying their compliance with international standards (Codex Alimentarius Commission , 2001). This paper was carried out to classify and indication Sidr honey types produced at Egypt, Algeria, Libya and Yemen countries by using discriminate analysis.

Materials and methods

1.Honey samples:

Twenty Honey samples were collected from different geographical regions Sidr plant of Egypt ,Libya, Algeria and Yemen.

1.1. Physical properties:

1.1.1. Moisture determination:

Association of official Analytical Chemists (A.O.A.C.) (1990) official method 969.38 was emphasized by refractometer (Digital refractometer a tago, Germany). All measurements were performed at 20 °C, after waiting for 6 min for equilibrium and obtaining the analogous % moisture (g/100 g honey) from the refractive index of the honey sample by consulting a standard table for the purpose.

1.1.2. The specific gravity and viscosity:

The gravity and viscosity were estimated according to White (1978). Electrical conductivity was determined by conducterimetric experiment (WTW Inolabconducterimeter), from a solution containing 10 g of honey in 75 mL of distilled water, (Sancho *et al*., 1992) the total soluble solids (TSS%) was measured according to A.O.A.C. (1990).

1.1.3.Electrical conductivity (EC) and total soluble solids(TSS):

The EC and TSS were measured using a conductivity meter HI 98311 (Hanna Instruments, Mauritius) in a 20% (w/v) solution of honey suspended in Milli-Q water as recommended by Bogdanov *et al.* (1999). The EC and TSS of each sample were analyzed and the means are expressed as mS/cm and ppm, respectively. The EC of milli-Q water alone was less than 10 µS/cm.

1.2.The chemical properties:

1.2.1. Measuring of pH, free acidity, lactones and total acidity:

The pH, free acidity, lactones and total acidity were measured, with a combined pH glass electrode connected to pH meter Basic 20, in a solution prepared with 10 g of honey in 75 mL of distilled water (NP 1309/1976). Free acidity was determined by potentiometric titration (A.O.A.C. ,1990). Official method 962.19. Honey samples were homogenized in a water bath and filtered through gauze, prior to analysis. Ten grams of honey were then dissolved in 75 mL of distilled water and alcoholic solution of phenolphthalein added. The solution was titrated with 0.1 N NaOH. Acidity (milli equivalent of acid per kg of honey) was determined as 10 times the volume of NaOH used in titration.

1.2.2. Estimation of the sugar content:

The honey tested was similar with the findings of other previously studied by High Performance Liquid Chromatography (HPLC) measured the concentration of fructose, glucose, sucrose and maltose (%) according to Bogdanov and Bauman (1988).

2. Statistical analysis:

Analysis of variance (ANOVA) and linear discriminate analysis (LDA)

were used to the investigated set of data and calculated by the statistics software SPPS (version.20.0 for windows). ANOVA was applied to all the investigated physicochemical parameters, as a pre-treatment p rocedure, in order to point out the significant parameters that could discrimination Sidr honeys of Arab countries ($P< 0.05$). LDA, which is a supervised statistical technique, was then applied only to the significant parameters ($P< 0.05$) (independent variables) to determine a linear amalgamation of these group of subjects, which could provide a discrimination rate of Sidr honey according to different countries. The original and cross validation methods were considered. In the cross validation method, each case is classified by the functions derived from all cases other than that case. Lastly, the statistical criterion of Wilk's Lambda was also considered, since it evaluates the statistical significance (discriminatory ability) of the discriminate functions derived (Karabagias *et al.,*2017).

Results and Discussion

1. Physicalchemical properties of Sidr honey produced in Arab countries:

The measured values of physicochemical properties of Sidr honey in four Arab countries (Egypt, Algeria, Libya and Yemen) were shown in Table (1). Moisture content, a parameter related to the ripening degree, was ranged 17.70 ±0.224% to 18.00 ±0.447%, there were no significant differences, between Sidr honey samples, hence these results are indicated of good storage ability of these honeys. The sidr plants grow in same the approximately environmental condition nearly .The average moisture of the Sidr honey samples from Arab countries under study were found to be within the limit of not more than 20.0 g/100 g as prescribed by Codex Alimentarius Commission (2001). Moisture content is an important quality parameter, important above all for honey shelf-life (Bogdanov *et al*., 2008). Thus, honey having high water content is more probable to ferment (Bogdanov *et al*.,1999). A maximum value of 20.0 g/100 g was established by Codex Alimentarius Commission (2001) and European Commission (2002) commission as the international standard for honey moisture contents. The specific gravity were 1.415 ±0.018 to 1.417 ±0.073 of all samples were found this value meet honeys quality European Legislation, European Commission (2001). Viscosity,shows no significant difference between examined samples (P<0.05) were 69±0.08 to 69±0.36. But the electrical conductivity (EC%) is one of the most important factors for determining the physical characteristics of honey (Serrano *et al.,* 2004). It is also an important physical measurement for the authentication of unifloral honeys (Mateo and Bosch-Reig, 1998). The EC % values of the honey samples of Egypt and Algeria 0.046±0.009 and 0.036±0.004, respectively, were significantly different higher than honey samples of Libya and Yemen (P=0.0005***) which recorded 0.011±0.002 and 0.015±0.003, respectively. The EC% values of all tested honey samples were reported to be 0.21–1.61 mS/cm, in a previous study by Ouchemoukh *et al.* (2007). EC is a basic physicochemical parameter for the authentication of unifloral honeys (Mateo and Bosch-Reig, 1998). The EC value depends on the ash and acid content in honey in which the higher the content, the higher the resulting conductivity (Bogdanov *et al*., 2002). This parameter was recently included in the international standards, replacing the determination of ash content (Codex Alimentarius Commission, 2001). However, the results were similar to the findings previously reported by Saxena *et al.*(2010) and Alvarez-Suarez *et al*. (2010).

Table (1): Physicalchemical properties of Sidr honey produced in Arab countries (Egypt, Algeria, Libya andYemen).

Parameter	Egypt	Algeria	Libya	Yemen	F	P
Moisture (%)	17.70 ±0.224a	17.70 ±0.235a	18.00 ±0.292a	18.00 ±0.447a	1.54	0.243ns
Specific gravity	1.417 ±0.073a	1.417 ±0.043a	1.415 ±0.039a	1.415 ±0.018a	0.003	1.000ns
Viscosity(Poise)	69.00 ±0.316a	69.00 ±0.292a	69.00 ±0.100a	69.00 ±0.381a	0.000	1.000ns
EC (%)	0.046 ±0.004a	0.036 ±0.003a	0.011 ±0.002b	0.015 ±0.003b	184.04	0.000***
TSS (%)	82.00 ±2.345	82.00 ±2.236a	82.00 ±1.871a	82.00 ±2.121a	0.000	1.000ns
PH	5.30 ±0.332A	4.30 ±0.292b	3.500 ±0.200c	3.90 ±0.255bc	39.78	0.000***
Free Acidity(meq/ kg)	34.00 ±2.641b	47.00 ±1.797a	34.00 ±1.766b	49.90 ±1.105a	97.63	0.000***
Lactone(meq/ kg)	10.00 ±0.954a	3.50 ±0.381b	10.00 ±0.731a	2.000 ±0.283b	214.32	0.000***
Total acidity (meq/ kg)	43.14 ±4.384	49.76 ±2.483	43.66 ± 1.557	51.72 ±1.484	12.43	0.000***
Fructose(g/100g)	39.14 ±0.51b	39.70 ±0.39b	41.90 ±0.86a	38.00 ±0.89b	27.60	0.000***
Glucose(g/100g)	30.1 ±0.9b	32.0 ±0.40a	29.60 ±0.90b	30.00 ±0.60b	10.12	0.001**
Fructose/ Glucose Ratio	1.31 ± 0.066	1.24 ± 0.044	1.44 ± 0.104	1.28± 0.049	7.29	0.003**
Sucrose(g/100g)	1.10 ±0.25c	3.00 ±0.38b	5.10 ±0.29a	5.50 ±0.29a	218.38	0.000***
Maltose(g/100g)	6.00 ±0.39a	3.00 ±0.23b	6.30 ±0.33a	6.50 ±0.35a	123.18	0.000***
Glucose/Water Ratio	1.67 ±0.037	1.78 ±0.028	1.644 ±0.104	1.660 ±0.066	8.86	0.001**

Different letters in the same row indicate significan t differences.

Total soluble solids is a measure of the combined content of all inorganic and organic substances in honey in the molecular, ionized or micro-granular (colloidal solution) suspended forms. The present data in Table (1) revealed non-significant between all Sidr honey samples, were 82.0 ±1.41 to ±2.45, these results demonstrated a good correlation between EC and TSS, indicating that both parameters can be used to determine honey purity European Commission (2001). Chemical characteristic of all Sidr honey samples were acidic, the pH ranged 3.5±0.2 to 5.3±0.332, it was agreement with the standard limit pH 3.40-6.10 (Codex Alimentarius Commission , 2001). The acidic of Libyan Sidr honey was high significantly (pH 3.5±0.2) (*P* =0.0022**), than other samples followed by Yemen (3.9± 0.255) and Algeria honey (4.3± 0.292). The lowest acidity was detected in Egypt honey (5.3± 0.332). High pH value (6.23) was recorded for Sidr Aseer honey, while Sidr Albaha had a pH of 3.93 (Al-khalifa and Al-Arify, 1999). The pH values of honey samples were close to those previously reported in Indian, Algerian, Brazilian, Spanish and Turkish honeys (between pH 3.49 and 4.70) (Kayacier and Karaman, 2008 and Saxena *et al*., 2010). Acidity in honey is calculated as free, lactonic and total acidity. Specifics a free acidity of not more than 50 meq/1000 g (meq/kg) (European Commission, 2002). Some factors affecting bee honey acidity e.g. harvest seasons and floral types (El-Sherbiny and Rizk 1979 and Pe´rez-Arquillue´ *et al*., 1994).The average values for free acidity in samples were between 34.0±2.288 and 49.9±1.159 meq/kg and were highly significant among Yemen, Algeria Sidr honey samples and Egypt, Libya (*P*=0.0002***). Lactonic acidity ranged from 2.0±0.356 to 10.0±1.034 meq/kg and found highly significant between all samples (*P*=0.0000***). Total acidity detected highly significant between all samples (*P*=0.0000***), it's ranged from 44.03±5.02 to 51.93±1.59 meq/kg; The present invitigations are quite in agreement with Ouchemoukh *et al.* (2007). The high acidity of honey correlates with the fermentation of sugars present in the honey into organic acid, which is responsible for two important characteristics of honey: flavor and stability against microbial spoilage (Bogdanov *et al*.,2008). Illustrated

HPLC chromatography of the fructose, glucose, sucrose and maltose analysis of all Sidr honey samples in different Arab country under studies. The results indicated that there were significant differences of fructose value between Libyan Sidr honey (42.0±1.18) and others examined samples (*P*= 0.0204*).Furthermore, glucose content no significant in all Sidr honey samples. The glucose content was lower than the fructose content which indicated the natural feeding of honey colonies. In addition, the clear sucrose contents of Yemeni Sidr honey(5.5±0.29 g/100 g) and Libyan honey (5.1±0.29 g/100 g) were statistically significantly (*P*= 0.000***) higher than Algeria (3.0±0.29 g/100 g) and Egyptian honey (1.1g/100g).While maltose contents of Sidr honey samples shows highly significant (6.0±0.374, 6.3±0.37 &6.5±0.24 g/100 g) for Egypt, Libya and Yemen , respectively and Algeria Sidr honey recorded 3 ±0.22 g/100 g.(*P*= 0.000***).These results supported the previous several studies on different honey types (Buba *et al*., 2013 and EL-Metwally, 2015). Fructose/ glucose ratio indicates the ability of honey to crystallize. White and Doner (1980) reported that even though honey has less glucose than fructose, the honey were granulated because glucose less soluble in water than fructose. When the fructose/glucose ratio is high, honey remains liquid. Honey crystallization is slower when the fructose/glucose ratio is more than 1.3 and it is rapid when the ratio is below 1.0 (Amir *et al*., 2010). However, because honey contains others sugars (sucrose, maltose, turanose, etc.) and insoluble substances (like dextrin, colloids, etc) which can influence the crystallization process, the glucose/water (G/W) ratio is considered more suitable than the fructose/glucose (F/G) ratio for the forecast of honey crystallization. It has been stated that when the glucose/water ratio is <1.3 honey crystallization is very slow or even zero and it is complete and rapid when the ratio is >2.0 (Amir *et al*.,2010). Glucose, which is a major sugar in honey, can spontaneously crystallize from honey solutions in the form of its monohydrate (White and Doner,1980). This sometimes occurs when the moisture level in honey is allowed to drop below a certain level; i.e., when the musture content is very low. The international normal established by Codex Alimentarius Commission (2001) that a good quality honey should not contain more than 5 g/100 g sucrose. The apparent sucrose contents of the honey samples studied were in the range of 1.1 to 5.5 g/100 g. The values obtained for sucrose contents of the honey samples were all within the limits of international standards. According to White and Doner (1980) even though honey contains an active sucrose incision enzyme (sucrose and glucosidase), the sucrose level in honey never arrives at zero. The sucrose contents obtained in this realization are within the range of values stated for Argentine and Turkish (Cantarelli *et al.*,2008), Venezuelan (Vit *et al*.,2009), American (White and Doner,1980), Algerian (Makhloufi *et al*., 2007), Pakistani (Zafar *et al*.,2008) and Spanish (Cavia *et al*., 2006) honeys.

2. Discrimination of Sidr honey producing in Arab countries on selected physicochemical parameter values:

Results discriminate analysis showed that two discriminate functions were formed significant among these honey samples belong to country, hence, chemical parameters analysis showed significant differences on pH, lactone, total acidity, fructose , glucose , sucrose and maltose according to floral origin of honeys Wilks' Lambda between 0.028 to 0.345 (P = < 0.001 to 0.000) (Table, 2). The discriminate two functions was used for the classification of Sidr honey according to belong countries, since it

explained 100% of total variance and a good canonical correlation equal to 0.999. In addition, the standardized canonical discriminate function coefficients correlation for each of the significant physicochemical parameters that contributed to the country discrimination of Sidr honey show in Table (3). In the end summary regarding the identification of the variables with the highest discriminatory power, higher the absolute value of a standardized canonical coefficient, the more significant the variable is for the determination of honey origin. Remarkable, discrimination ability of conventional physicochemical parameters, ease of application and reproducibility, have been previously reported in the literature in studies involving Spanish (Serrano *et al*., 2004 and Karabagias *et al*., 2017), Moroccan (Chakir *et al*., 2016) and Greek (Karabagias *et al*., 2014) unifloral honeys, in agreement with the present results. Ruoff *et al*. (2007) stated that several exceptions are listed in the above-mentioned standards, thus indicating the limited value of this measure and for the discrimination of honey types. Thus multivariate data evaluation of traditional physical and chemical measures and may also be helpful to establish new criterion for a more reliable description of the honey types and for the determination of their botanical origin. The chemo-metric analysis of physical and chemical data demonstrated that the botanical origin of honey can be determined without considering pollen analytical results.

Table (2): Multivariate analysis of variance for testing the equality of the means of investigation of the physicochemical parameters according to Sidr honey of Arab countries.

Tests of Equality of Group Means					
Physicochemical Parameters	Wilks' Lambda	F	df1	df2	P
Moisture (%)	0.776	1.538	3	16	0.243 ^{ns}
Specific (gravity)	0.999	0.003	3	16	1.000^{ns}
Viscosity (poison)	1.000	0.000	3	16	1.000^{ns}
EC (%)	0.028	184.044	3	16	0.000***
TSS(%)	1.000	0.000	3	16	1.000 ^{ns}
PH	0.118	39.778	3	16	0.000***
Acidity	0.052	97.631	3	16	0.000***
Lactone	0.024	214.321	3	16	0.000***
Fructose (g/100g)	0.162	27.59 5	3	16	0.000***
Glucose (g/100g)	0.345	10.115	3	16	0.001**
Sucrose (g/100g)	0.024	b218.377	3	16	0.000***
Maltose (g/100g)	0.041	123.182	3	16	0.000***

F: Fisher's coefficient, ns: not significant, df: degrees of freedom, p: probability.

3. Summary regarding the identification of the variables with the highest discriminatory power:

The higher the absolute value of a standardized canonical coefficient, the more significant the variable is for the determination Sidr honey of countries. Table (3) showed physicochemical parameters of groups correlations between discriminating variables and standardized canonical discriminate functions. Variables ordered by absolute size of correlation within function. The standardized canonical discriminate function coefficients obtained in the developed statistical model for the discrimination of Sidr honey from Arab countries under study. Based on the aforementioned, the variables that most contributed to the discrimination of Sidr honey according to country origin .

Table (3): Physicochemical parameters of groups correlations between discriminating variables and standardized canonical discriminate functions. Variables ordered by absolute size of correlation within function.

Physicochemical parameters	Function	
	1	2
Moisture (%)[a]	0.064	0.496*
Specific (gravity)[a]	-0.050	0.007
Viscosity	0.000	0.000
EC (%)	0.055	-0.328*
TSS(%)[a]	0.004	0.069*
PH[a]	0.033	0.078*
Acidity	-0.185*	-0.001
Lactone	0.274*	0.001
Fructose(g/100g)[a]	0.342*	-0.184
Glucose(g/100g)[a]	-0.375*	-0.006
Sucrose(g/100g)	-0.120	0.332*
Maltose(g/100g)	-0.120	0.332*

a. This variable not used in the analysis.

*. Largest absolute correlation between each variable and any discriminate function.

4. External validation of the developed statistical model for the differentiation of Arab countries:

In order to investigate the robustness of the statistical model developed for the classification of honeys, data involving specific physicochemical parameters of Sidr honey from Arab countries were introduced into the set of data and a new statistical analysis was carried out. The common physicochemical parameter values taken into account from the present database were EC, pH, total acidity, lactone, fructose, glocose, sucrose, maltose. In Figure (1), it is also shown that Sidr honey Egyptian, Algerian, Libyan and Yemeni are well differentiated. The overall correct classification and cross validation method rate was 100% within and between the countries, which is considered a very satisfactory discrimination rate for this method. Table (4) lists the discriminatory power of the developed statistical model (Egypt, Algeria, Libya and Yemen Sidr honey) was taken as the dependent variable. The total number of honey samples was arrived to 20 prior to discriminate analysis. Based on chemical parameter analysis and electrical conductivity, content values could be classified as honeydew honeys. Discriminate analysis showed that two discriminate functions were formed: discriminate function 1 was the basic function for the classification of Egyptian and Algeria Sidr honey according to physiochemical parameters , since it explained 62.8% of the total variance providing a high eigenvalue (527.29) and a high canonical correlation (0.999) in comparison with those of the discriminate function 2 (eigenvalue of 282.886 and canonical correlation of 0.998). Respective group centroid values, representing the discriminate functions created, were (23.141,-12.870), (-18.676, -17.027), (17.680, 16.119) and (-22.146, 13.779) for Egypt, Algeria , Libya and Yemen Sidr honey, respectively (Figure, 1). The discriminate function 1 was used for the classification of Sidr honey according to countries, since it explained 62.8% of variance function 1 and a good canonical correlation equal to 0.999. In addition, the standardized canonical each of the significant physicochemical parameters that contributed to the country discrimination of Sidr honeys are given

in Table (3) and 100% for the cross validation method, considred a satisfactory discrimination rate for this method. Finally ,two methods for the determination of the botanical origin of Sidr honey are used. The first is to find and quantify specific parameters of unifloral honeys. In the second method different quality parameters are first determined and then physiochemical analysis is applied for honey classification. The first method is easier and more straight forward, but due to the major natural variation of honey composition, it may not be always possible to find specific markers for each unifloral honey. The second method yields statistical models for the classification of a known group of Sidr honey. The application of this method in future routine work needs statistical models that are based on the analysis of a representative number of authentic unifloral honeys. In these models polyflor al honeys, should also be included. Also, the physiochemical techniques should be correspond for routine use in control laboratories.

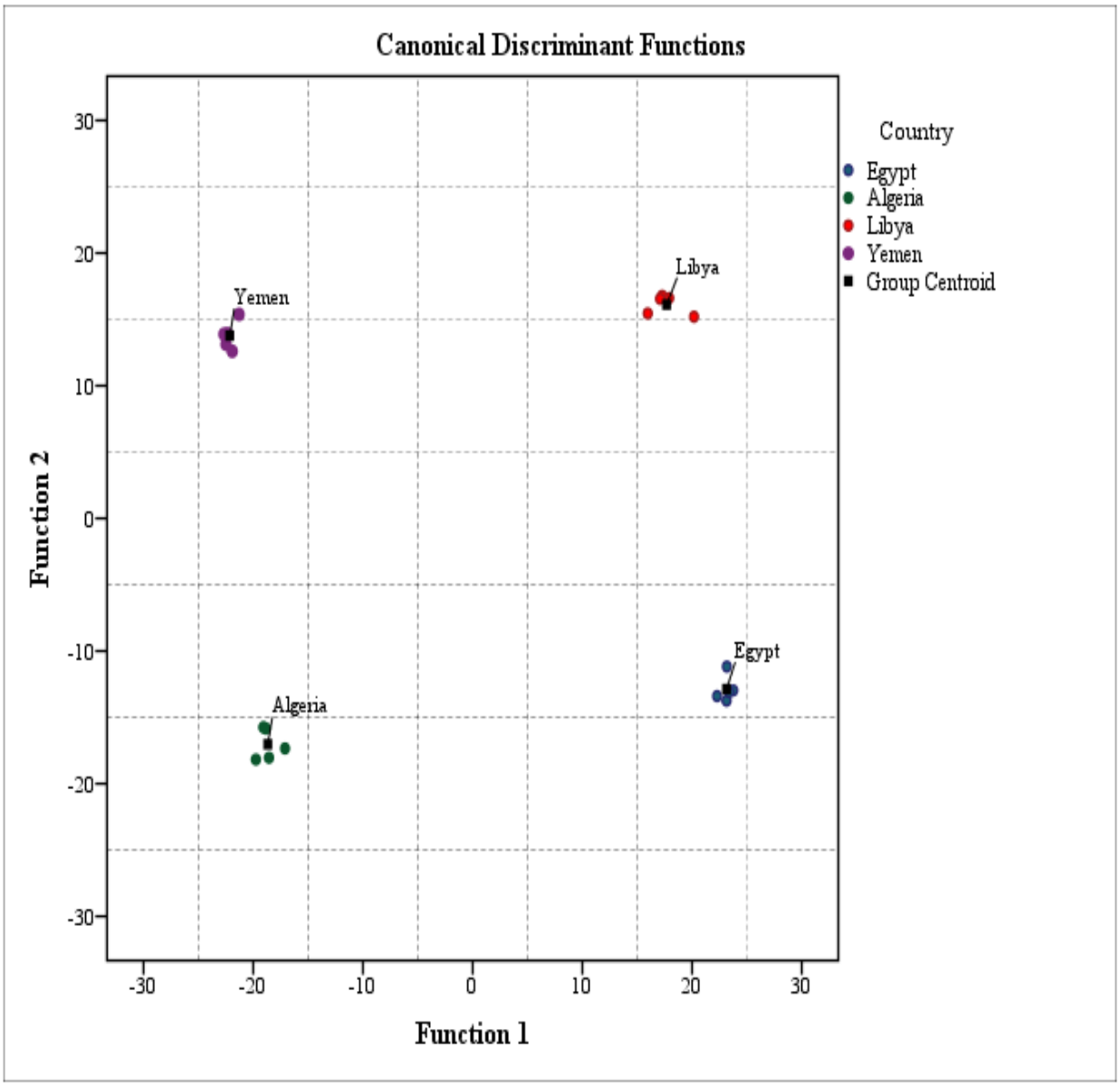

Figure (1): Discrimination of Sidr honey from Egypt, Algeria, Libya and Yemen based on 12 physicochemical parameters.

Table (4): Discriminatory power of the developed statistical model for the classification of Sidr honey from Arab countries

		Country	Predicted Group Membership				
			Egypt	Algeria	Libya	Yemen	Total
Original	Count	Egypt	5	0	0	0	5
		Algeria	0	5	0	0	5
		Libya	0	0	5	0	5
		Yemen	0	0	0	5	5
	%	Egypt	100.0	.0	.0	.0	100.0
		Algeria	.0	100.0	.0	.0	100.0
		Libya	.0	.0	100.0	.0	100.0
		Yemen	.0	.0	.0	100.0	100.0
Cross-validated[a]	Count	Egypt	5	0	0	0	5
		Algeria	0	5	0	0	5
		Libya	0	0	5	0	5
		Yemen	0	0	0	5	5
	%	Egypt	100.0	.0	.0	.0	100.0
		Algeria	.0	100.0	.0	.0	100.0
		Libya	.0	.0	100.0	.0	100.0
		Yemen	.0	.0	.0	100.0	100.0

a. Cross validation is done only for those cases in the analysis. In cross validation, each case is classified by the functions derived from all cases other than that case.

b. 100.0% of original grouped cases correctly classified.

c. 100.0% of cross-validated grouped cases correctly classified.

Classification Results b,c

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Laboratory and field evaluation of certain chemicals comparing with methomyl against land snail *Monacha* sp. (Stylommatophora: Hygromiidae) infesting Egyptian clover plant

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Abstract:

The land snails feed on leaves, roots, tubers and seeds of of ornamental plants, , citrus, peach, plam , cabbage, carrot and bean as well as thses crops lose their marketability and hence their export potential in many countries. Efficacy of some chemicals as Potassium hydroxide, Ferrous sulphate and Sulphonic acid compared with Methomyl as standard pesticide was evaluated against *Monacha* sp. (Stylommatophora: Hygromiidae) under laboratory and field conditions infesting Egyptian clover plant. The obtained results in the laboratory exhibit the following ; the all tested chemicals exerted substantial mortality against *Monacha* sp. after one day of evaluation where the general mean of mortality percentage values was 63.3, 63.3, 25.8 and 21.6 for Methomyl, Potassium hydroxide , Ferrous sulphate and Sulphonic acid , respectively. Methomyl treatment was more toxic than that appeared with other tested treatments after 3 days of treatments where values were 87.5, 72.5, 36.6 and 31.6 for Methomyl, Potassium hydroxide , Ferrous sulphate and Sulphonic acid , respectively (values increased as concentration and time increased). According to mortality percentages, the descending order of the tested chemicals was Methomyl, Potassium hydroxide , Ferrous sulphate and Sulphonic acid where the general mean was 87.5, 74.1, 45.8 and 41.6 , respectively after 21 days. The field experiment appeared that, the spray of Methomyl (1%) gave the highest reduction percentages 93 and 83% after 7 and 15 days of treatments, respectively; while it was 73 and 49% with Potassium hydroxide and was 49 and 50 % with Ferrous sulphate and the least values were 43 and 42 % with Sulphonic acid .

Introduction

In Egypt, land snails have been increased and distributed rapidly in different locations especially in the northern Governorates (Eshra *et al.*, 2016). They caused considerable damage especially in most areas where they found the optimum conditions for survival and dispersion (Kassab and

Daoud, 1964; Glen and Wilson, 1997 and Glen *et al.*, 2000). These animals attack almost all crops reducing their yields, marketing values and cause severe damages to all plant parts (El-Okda, 1980) as a result of mucous secretion and the particular structure of their mouth parts enabling scratching and crushing. In addition, some of these animals work as intermediate hosts for parasite trematodes, cestodes and nematodes which cause worm diseases in man and domestic animals. Therefore, they attack the attention of the biologists because of the great economic damage they do in agriculture and horticulture (Godan, 1983). The land snails *Monacha* sp. (Stylommatophora: Hygromiidae) were recorded to be harmful snails in many Egyptian Governorates attacking various parts (El-Wakil *et al*., 2000 and Eshra, 2013). There are three common methods for controlling these pests (mechanical, biological and chemical). Nowadays the control with chemical pesticides is still one of the most effective methods (Radwan *et al.,* 1992; Eshra, 2004; Moran *et al*., 2004; El-Shahaat *et al*., 2005, 2009 and Ghamry, 1997) for elimination of different pests (Hilmy and Hegab, 2010). Many investigators have drawn the attention to control the land snails using chemical compounds (Ebenso, 2004; Hegab *et al.*, 2013 and Abdel-Rahman, 2017). Although that, it causes many environmental problems. Therefore, searching for effective and safety agents for terrestrial snails control is very important. Using safety agents as urea fertilizer and New-Fort® for terrestrial snails control had been previously studied (El-Shahaat *et al.*, 2009 and Eshra, 2014). Therefore, this study is carried out to investigate the molluscicidal effects of Potassium hydroxide, Ferrous sulphate and Sulphonic acid as safety chemicals against *Monacha* sp. under laboratory and field conditions comparing to Methomyl as standard pesticide.

Materials and methods

1. Experimental animals:

Adults of the land snail *Monacha* sp. having approximately the same age and size were collected from infested Egyptian clover (*Trifolium alexandrium*) field at Zawar Abo-Elliel village, Awlad-Sakr district, Sharkia Governorate for laboratory study. These snails were collected during November, 2018. They were then transferred to plastic cups covered with cloth netting and maintained under laboratory conditions of 20°C and 75% R.H. The snails were daily fed on clover plants for acclimatization two weeks. Dead snails were removed immediately (Eshra, 2014).

2.The tested chemicals:

2.1. Methomyl (Lannate ® 90% SP):

Chemical group: Carbamates

Trade name: Neomyl 90% WP

Common name: Methomyl

2.2. Ferrous sulphate:

Scientific name: Ferrous sulphate

Chemical formula: FeSO

The formulation: As crystalline powder

Source: Ferrous sulphate was obtained from El- Gamhouria

Company for chemicals, Zagazig branch, Egypt.

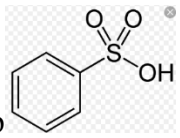
2.3. Potassium hydroxide :

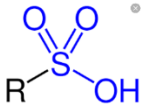
Chemical name: Potassium hydroxide

Molecular formula: KOH

Formulation: (WP) or (WC)

2.4.Sulphonic acid



General formula:  , where R is an organic alkyl or aryl group and the S (=O)2-OH group is a sulphonyl hydroxide .

3.Laboratory evaluation:

Toxicity of the tested chemicals against *Monacha* sp. was evaluated as a spray with concentrations (1, 2, 3, and 4) % of Potassium hydroxide , Ferrous sulphate and Sulphonic acid and (0.125, 0.25, 0.5 and 1) % of Methomyl that were prepared as solutions. Acclimatized adult snails were transferred into plastic cups where 10 healthy adult snails were placed into each cup. Each concentration had three replicates and untreated cups were used as a check treatment. Replicates were sprayed with the tested chemicals a single time. The cups were covered with muslin clothes and secured with rubber band to prevent snails from escaping (El-Okda, 1981). Mortality number was recorded after 1, 3, 7 and 21 days post-treatments according to Ghamry (1997). The means of died snails were calculated at the end of the experiment.

4. Field evaluation:

Field experiments were performed at Zawar Abo-Elliel village, Awlad-Sakr district, Sharkia Governorate. For each treatment, a quarter 3x3.5m at an area of about one feddan cultivated with Egyptian clover (*Trifolium alexandrium*) heavy infested with land snail *Monacha* sp . The field was irrigated only day before any treatment. The tested materials were applied as solution spray with one concentration (4%) and methomyl with (1%) by incorporating the tested material with tap water. Number of snails inside each quarter was estimated before just treatment and after 1, 3,7,15 and 21 days of spraying application. Reduction percentages were statistically calculated according to the formula of Henderson and Tillton (1955) as follows: % Reduction = 100 [1- t2r1/ t1r2] where r1 and r2 are the number of the alive snails before and after treatment respectively in untreated plots (control), t1 and t2 are the number of the alive snails before and after treatment respectively, in treated plots. Statistical analyses were designed using Costat statistical software, 2005 Version 6.311.

Results and discussion

1. Laboratory evaluation:

Mortality percentages of snail *Monacha* sp. sprayed with different concentrations of Methomyl, Potassium hydroxide , Ferrous sulphate and Sulphonic acid under laboratory conditions presented in Table (1). It can be seen that the all tested chemicals exerted substantial mortar effect against *Monacha* sp. after 1 day of evaluation where the general mean of mortality percentage values were (63.3, 63.3, 25.8 and 21.6) for methomyl, Potassium hydroxide , Ferrous sulphate and Sulphonic acid , respectively. Methomyl was the most effective one where, there were no survivals (100% mortality) at highest concentration (1%) after one day post-treatment. The corresponding values were 87, 30 and 30% at highest concentration (4%) of Potassium hydroxide , Ferrous sulphate and Sulphonic acid , respectively. The mortality percentages increased as time increased where the general mean was 63.3, 87.5, 87.5, 87.5 and 87.5 for Methomyl; 63.3, 72.5, 72.5, 73.3 and 74.1 for Potassium hydroxide ; 25.8, 36.6, 42.5, 45 and 45.8 for Ferrous sulphate and 21.6, 31.6, 39.1, 39.1 and 41.6 for Sulphonic acid after 1,3,7,15 and 21 days respectively. The mortality percentages increased as increasing concentration (20, 53. 3, 80 and 100) % mortality at concentration 0.125, 0.25, 0.5 and 1% of methomyl respectively. The descending order of the tested chemicals was Methomyl, Potassium hydroxide , Ferrous sulphate and Sulphonic acid according to mortality percentages where the general mean of % mortality was 87.5, 74.1, 45.8 and 41.6, respectively after 21 days. Table (2) revealed that there were significant differences between the treated and untreated *Monacha* sp. The differences

between the tested chemicals were significant.

2. Field evaluation:

Table (3) showed that the same trend was observed when the tested chemicals were applied under field conditions. The reduction percentages of *Monacha* sp. infesting Egyptian clover plant treated with Methomyl, Potassium hydroxide, Ferrous sulphate and Sulphonic acid increased gradually with time till 7 days then decreased. Methomyl exhibited higher molluscicidal efficiency than Potassium hydroxide, Ferrous sulphate and Sulphonic acid. After 7-days post-treatments, reduction percentages were, 93, 73, 49 and 43 consecutively. F.test showed significant differences between values of the reduction percentages at the four tested chemicals (Table,4).

The obtained results supported the findings mentioned before by other researches where the current data agree with Gouth *et al.* (1968); El-Okda *et al.* (1989); El-Shahaat *et al.* (1995, 2005 and 2007) and Abdel-Rahman and Al Akra (2012). They found that Oxime Carbamates Methomyl appeared to be an efficient chemical against land snails. Bailey (2002) said that the carbamates are feeding inhibitors leading to death. Godan (1983) said that the increasing of the mucous secretion is one of the first reaction of gastropods to many stressors including mechanical irritation caused by molluscicidal chemicals leading to death. Abdel-Rahman (2017) found that Ferrous sulphate appeared to be the most toxic in comparison with natural extracts against *Monacha cartusiana* under both field and laboratory conditions. .

Also, El-Shahaat *et al.* (2009) found that urea as a chemical fertilizer, was highly successful agent when sprayed directly on terrestrial snails at the resting or aestivation period, this fertilizer, also could be sprayed on weeds around trees for controlling snail. It is well known that the important way for controlling terrestrial mollusks (snails and slugs) is chemical control using certain traditional pesticides that have undesirable or detrimental effects on the environment and non-target organisms (Moran *et al.*, 2004). Finally, it is necessary to show that more research and attention are need to evaluate certain chemical fertilizers and understand their effects on molluscs.

Table (1): Mean values of mortality percentages of snail (*Monacha* sp.) treated with different concentrations of Methomyl, Potassium hydroxide, Ferrous sulphate and Sulphonic acid after indicated days under laboratory conditions.

Mean values of % Mortality of snail (*Monacha* sp.) after indicated days						
Compound	**Treatment conc. %**	**1 day**	**3 days**	**7 days**	**15 days**	**21 days**
Methomyl	0.125	20	50	50	50	50
	0.25	53.3	100	100	100	100
	0.5	80	100	100	100	100
	1	100	100	100	100	100
	General mean	63.3	87.5	87.5	87.5	87.5
Potassium hydroxide	1	37	43	43	43	47
	2	67	70	70	70	70
	3	63	90	90	90	90
	4	87	87	87	87	87
	General mean	63.3	72.5	72.5	73.3	74.1
Ferrous sulphate	1	10	13.3	13.3	13.3	13.3
	2	20	33	50	50	50
	3	43	43	43	43	43
	4	30	57	57	63	67
	General mean	25.8	36.6	42.5	45	45.8
Sulphonic acid	1	17	20	33	33	33
	2	10	30	30	30	30
	3	30	30	37	37	47
	4	30	47	57	57	57
	General mean	21.6	31.6	39.1	39.1	41.6
Control	General mean	0	0	0	0	0

Table (2): Statistical analysis of the general means of mortality percentages of snail (*Monacha* sp.) treated with different concentrations of Methomyl, Potassium hydroxide, Ferrous sulphate and Sulphonic acid after for 21 days under laboratory conditions.

Compound	The general means of % mortality of snails after indicated days				
	1 d.	3 d.	7 d.	15 d.	21 d.
Methomyl	63.3a	87.5a	87.5a	87.5a	87.5a
Potassium hydroxide	63.3a	72.5b	72.5b	73.3b	74.1b
Ferrous sulphate	25.8b	36.6c	42.5c	45c	45.8c
Sulphonic acid	21.6b	31.6c	39.1cd	39.1c	41.6c
Control	20b	20c	20d	16.6d	10d
F.test	***	***	***	***	***
L.S.D 0.05	2.939	2.358	2.587	2.392	2.167

Table (3) Population reduction percentages of *Monacha* sp. infesting Egyptian clover plant exposed to the tested compounds with one concentration as sprays for 21 days under field conditions.

Compound	After after indicated days population reduction%					
	Replicate	**1 day**	**3 days**	**7 days**	**15 days**	**21 days**
Methomyl (1%)	1	89	82	92	93	61
	2	77	73	94	83	54
	3	65	83	94	72	71
	Mean	77	79.33	93.33	82.66	62
Potassium hydroxide (4%)	1	54	42	98	71	60
	2	41	69	67	56	36
	3	57	71	55	20	22
	Mean	50.66	60.66	73.33	49	39.33
Ferrous sulphate (4%)	1	62	51	72	65	36
	2	26	56	64	68	36
	3	20	29	12	18	16
	Mean	36	45.33	49.33	50.33	29.33
Sulphonic acid (4%)	1	30	23	53	21	19
	2	31	41	56	69	15
	3	29	30	19	36	23
	Mean	30	31.33	42.66	42	19

Table (4): Statistical analysis of the general means of population reduction percentages of *Monacha* sp. infesting Egyptian clover plant exposed to the tested compounds with one concentration as sprays for 21 days under field conditions.

Compound	General means of % population reduction percentages of snails after indicated days				
	1 day	**3 days**	**7 days**	**15 days**	**21 days**
Methomyl	77 a	79.33 a	93.33 a	82.66	62 a
Potassium hydroxide	50.66 b	60.66 ab	73.33 ab	49	39.33 ab
Ferrous sulphate	36 b	45.33 bc	49.33 b	50.33	29.33 b
Sulphonic acid	30 b	31.33 c	42.66 b	42	19 b
F.test	* 0.0117	** 0.0066	* 0.0801	N.S 0.2237	*
L.S.D 0.05	25.49	22.69	41.86	44.02	22.82

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Intercropping Influence of aromatic plants and okra on the population fluctuations of *Earias insulana* and *Helicvorpia armigera* (Lepidoptera :Noctuidae) a well as the role of intercropping on predator population and okra yield

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Abstract:

This study was conducted at El-Riad region, Kafr El-Sheikh Governorate during two successive growing seasons (2017 and 2018) to investigate the role of intercropping of okra and aromatic plants on the infestation with *Earias insulana* (Boisduval), *Helicvorpia armigera* (Hübner) (Lepidoptera :Noctuidae) and the associated predators and okra yield. The aromatic plants were catnip, spearmint, lemongrass, rosemary and lemon balm plants. Results showed that intercropping reduced the infestation percentage with *E. insulana* , *H. armigera*, especially in case of rosemary + okra, catnip + okra, respectively and increased the associated numbers of predators especially in lemon balm+ okra. Results exhibited that lemon balm intercropping with okra was highest attractive for *Coccinella* sp. (Coleoptera: Coccinellidae) and *Nesidiocoris tenuis* (Reuter) (Heteroptera: Miridae). Intercropping between spearmint and okra was highest attractive to *Chrysoperla carnea* Stephens (Neuroptera: Chrysopidae), as so catnip plants intercropping with okra plants was more attractive to *Paederus alfierii* Koch (Coleoptera:Staphylinidae) and *Scymnus* spp. (Coleoptera: Coccinellidae). The highest abundance of true spiders was found on okra plants intercropped with lemongrass, followed by okra with catnip. The highest okra yield was obtained when okra was intercropped with lemon balm.

Introduction

Vegetables consist a major part of food consumed by the Egyptian population. One of the popular important vegetable crops in Egypt is okra [(*Abelmoschus esculentus* (L.) Moench. (Malvaceae)], which is a good source of protein, vitamin and mineral elements needed for the development and maintenance of human body. The fruit also lend itself well to freezing and canning products (Dike, 1983). Foliage of okra plants are known to provide good sources of fodder for livestock (BOSADP, 1998).

Okra is a popular summer vegetable crop in Egypt. The cultivated

area in Egypt for okra was nearly 17 thousand fedan (one fed. =4200m^2) and produced about 97 thousand ton in 2012 cropping season (Anonymous, 2013). Intercropping of compatible plants also encourages biodiversity, by providing a habitat for a variety of beneficial insects and soil organisms that would not be present in a single crop environment. This biodiversity can in turn help to limit outbreaks of crop pests by increasing the diversity or abundance of natural enemies, such as spiders or parasitic wasps. Increasing the complexity of the crop environment through intercropping also limits the places where pests can find optimal foraging or reproductive conditions. There are some different variants of intercropping: 1. Mixed intercropping. 2. Row intercropping 3. Relay intercropping (Infonet biovision).Okra is an annual Malvaceae crop and is susceptible to a large range of insect-pests and diseases. Various growth stages of the crops are susceptible to the different insect-pests and diseases(Baseline survey). Okra plants infested by the bollworms of *Helicvorpia armigera* (Hübner) (Lepidoptera :Noctuidae), which take it up as food. Pods and flowers are primary targets of spiny bollworm, *Earias insulana* (Boisduval) (Lepidoptera :Noctuidae), while the American bollworm caterpillar, *H. armigera* prefers the reproductive parts of the plant, including buds, flowers and fruits (Mudathir,2000). The American bollworm (*H. armigera*) caterpillars prefer the reproductive parts of the plant, including bands, flowers and fruits. Also, invade attack the ripped and pre-ripped fruits, contaminating them fraises and exposing them to fungi and bacteria (Ahmed, 2004). Planting insect pest repellent plants (PRP) as companion plants along with crops has been used as an alternative method in pest management. Many plant species have been identified to contain repellent effects on pests, such as, planting basil (*Ocimum basilicum* L.) with tomatoes repels T.tabaci. Coriander (*Coriandrum sativum* L.) repelled aphids, spider mites and potato beetles in potato. Garlic (*Allium sativum* L.) repelled aphids in roses, while mint (*Mentha cordifolia* L.) deterred white cabbage moths, ants, rodents, beetles, fleas and aphids in many crops (Anonymous, 2004a). Onion repelled cabbage lepidopterous pests in cabbage (Anonymous, 2004b). Marigolds repelled Mexican bean beetles in beans and tomato hornworms (Anonymous, 2004c) pest repellent plants may be an alternative method in controlling pests in organic agriculture as it needs to avoid the use of synthetic pesticides, growth regulators, livestock feed additives, etc. Insect predators and parasitoids were detected at variable levels, attacking mainly insect pests on okra (Barahoei *et al.*, 2012; Abdalla and Bilal, 2012; Saljoqi *et al.*, 2013; Khan *et al*.,1980 and El-Fakharany, 2016). In relay intercropping, two or more crops are grown in the same piece of land during part of the cropping season. A second crop (usually a cover crop) is planted in the same field as the first crop after the first has achieved reproductive maturity but before it has reached physiological maturity. This helps avoid competition between the main crop and the intercrop. It also uses the field for a longer time, since the cover crop usually continues to grow after the main crop is harvested.

The present investigation aimed to study the impact of intercropping of five aromatic plant species (catnip, spearmint, lemongrass, rosemary and lemon balm) with okra on the population fluctuations and abundance of *E. insulana* and *H. armigera*. In addition, the influence of intercropping on predator population and okra yield was investigated

Materials and methods

Experiments were carried out at okra field to study the impact of intercropping, between okra and aromatic plants (Table, 1) and its effect on okra infestation with *E.insulana* , *H.armigera*, predator population and okra yield.

Table (1): Aromatic plants intercropped with okra to manage the infestation with *Earias insulana* and *Helicvorpia armigera* in okra plants

Common name	Scientific name	Plant family	الاسم العربى
Catnip	*Mentha pulegium* L.	Lamiaceae	النعناع البرى
Spearmint	*Mentha viridis* L .	Lamiaceae	النعناع البلدى
Lemongrass	*Cymbopogon citratus (Dc.)*	Poaceae	حشيشة الليمون
Rosemary	*Rosmarinus officinalis L.*	Labiatae	حصالبان
Lemon balm	*Melissa officinalis* L.,	Lamiaceae	الترنجات

1. Experimental design:

The experiment was carried out during two okra growing seasons, 2017 and 2018 on pests *E. insulana and H.armigera,* in El-Raid region, Kafr El-Sheikh Governorate, Egypt. An area of one fedan was prepared (6 treatments × 4 rep.) and divided into 24 plots (each plot about 175 m2 13*13m, 10 row) separated by a border of 1 m path and 2 m between each replication .in a randomized complete block design. The seeds of okra (Ladies fingers Mansoura Red variety) were sowed (2 seeds/hole) on two sides of row with widths' of 100cm spacing 30 cm between plants during the two seasons on 20th Marsh

Seedlings of catnip, spearmint, lemongrass, rosemary and lemon balm were transplanted on 1st of April in the middle of rows at a space of 50 cm between plants and 100 cm row intercropping of each five aromatic plants sown with okra on alternating rows (1:1), (Individual terraces of the replicate. Horticultural practices were performed according to the recommendations of the Ministry of Agriculture and Land Reclamation of Egypt, but without pesticides.

2. Sampling procedure:

Weekly samples were taken randomly beginning from June 15th up to end of October . Each sample consisted of 25 plants/ replicate randomly chosen from each treatment, Population of *E. insulana* and *H. armigera* (number of larvae/25 plants) and percent shoot damage *by E. insulana* (number of bored shoots to healthy shoots in 10 plants) were observed starting from 30 days after till the end of the crop season at 7 days interval. Percent fruit damage by *E . insulana* and *H. armigera* (number of bored fruits to healthy fruits/ 25 plants) was recorded at each harvest starting from 60 days after dibbling till the end of the crop season.

3. Abundance of associated predators:

Coccinella sp. (Coleoptera: Coccinellidae) (eggs, larvae and adults), *Chrysoperla carnea* Stephens (Neuroptera: Chrysopidae) (eggs, larvae and adults), *Paederus alfierii* Koch (Coleoptera:Staphylinidae) (adults), *Scymnus* spp. (Coleoptera: Coccinellidae) (larvae and adults), spiders (spiderlings and adults) and *Nesidiocoris tenuis* (Reuter) (Heteroptera: Miridae) were counted weekly by the aid of lens on 25 plants/ replicate, repeated four times beginning from May 15th up to end of October.

4. Estimating the crop yield of different treatments:

The mean okra yield (in Kg) were estimated for each treatment per Karat or fedan; fruits of 25 plants / treatment were picked and weighed

5. Statistical Analysis:

Data were subjected to ANOVA and statistically different means were compared using Duncan Multiple Range Test **(Duncan, 1955)**.

Results and discussion

1. Effect of intercropping between okra and aromatic plants on *Earias insulana* and *Helicvorpia armigera* infestation:

1.1. Number of *Earias insulana* larvae infested shoot and fruit borers:

Data presented in Table (2) showed that the effect of intercropping between okra plants and some aromatic plants on number of *E . insulana* larvae infested on okra shoots and fruits. In 2017, okra solid plants (control) received the highest mean number of *E. insulana* larvae (78.42 /25 plants) compared to okra intercropped with aromatic plants that received ranging between 7.45 and 49.75 larvae / 25 plants. This means that intercropping between okra and aromatic plants achieved 36.56-90.50 % reduction in *E. insulana* larvae. Intercropping between okra and rosemary or lemon balm or catnip reduced larvae infested shoot and fruits borer by 90.50, 82.07 and 73.82 %, respectively. Regardless of the intercropping pattern, the highest numbers of larvae infested were those on 2nd week from August and the end sample, in first season. However, the least number of infested larvae were in 2nd week from September. In 2018 season, almost the results were the same of 2017 season.

1.2. Number of *Helicvorpia armigera* larvae infested shoot and fruit borers:

In 2017 and 2018 seasons (Table, 3)), control okra had the highest larval population; 57.95 and 62.44 larvae/ 25 okra plants, respectively. The second rank of larval population was detected in okra- catnip intercropping pattern; with values of 36.67 and 41.11larvae / 25 okra plants in the first and second seasons, respectively. The third rank of larval population was found in okra + spearmint intercropping pattern. On the other hand, the least larval population was detected in okra+ lemon balm intercropping, as this pattern achieved the highest reduction in *H. armigera* larval population; 93.39 and 91.76% reduction, in the first and second seasons, respectively. Okra- rosemary pattern occupied the second rank of efficiency in reducing larval population, while the third rank of efficiency was that of okra- Lemongrass pattern. Regardless of intercropping pattern, the highest population densities of *H. armigera* larvae were recorded on 2nd, 1st week- August, in first and second season, respectively. However, the least larval population densities occurred by late September in both seasons.

1.3. Percent fruit damage by *Earias insulana* and *Helicoverpa armigera* in intercropping system:

During 2017 season, the lowest percent fruit damage by *E. insulana* and *H. armigera* was in *A. esculentus* (2.25 %), intercropping with Lemon balm. However, the highest percent fruit damage by *E. insulana* and *H. armigera* in intercropping system was in okra control (83.56 %) (Table, 4). In 2018 season, almost the results were the same of 2017 season.

The current results showed that intercropping aromatic plants with okra plants reduced mean number of E. insulana and H. armigera larvae compared to okra sole. It was clear that aromatic plants were more attractive to predators than okra sole. The high yield of okra was in case of intercropping between okra and aromatic plants, while, the lowest yield was that of okra plants only. These results agree with those obtained by other authors who proved that intercropping has the potentiality to reduce the injuries of harmful insects and to increase the predatory populations. This finding was coinciding with Ofuya (1991) who recorded that damage by *H. armigera* was significantly higher in sole cowpea than in cowpea intercropped with tomato. Also our result was coinciding with that of Abro *et al*. (2004) who indicated that pest Infestation in mix crop was recorded two weeks later than monoculture; so, Earias sp. infestation in okra grown as mono crop remained higher than in okra grown as mix crop, recorded 24.9 and 12 %, respectively. In okra fields in Ghana, intercropping with basil caused 23% decrease in insect pests compared to pure okra stands (Amoatey and Acquah, 2010).

Table (2): Number of shoot and fruit borer, *Earias insulana* larvae/ 25 okra plants as affected by intercropping pattern.

Sampling Date	Intercropping pattern					
	Okra	**Okra+**	**Okra+**	**Okra+**	**Okra+**	**Okra+**
Mean number of larvae of *Earias insulana* / 25 plant during 2017 season						
July 1st	30.50	3.00	16.25	5.25	22.75	10.50
2nd	47.25	5.25	20.25	10.00	36.50	19.25
3rd	55.75	8.50	36.75	16.25	49.25	25.25
4th	69.00	13.75	41.50	20.75	55.75	29.75
August 1st	75.00	14.25	50.25	20.50	71.75	38.00
2nd	109.25	18.00	70.50	28.25	97.50	50.25
3rd	83.50	2.50	35.75	11.00	50.25	25.50
4th	57.75	1.75	19.50	6.00	20.00	10.25
September 1st	33.25	1.50	15.00	2.25	17.25	5.00
2nd	47.25	1.00	6.00	2.00	10.25	3.75
3rd	73.75	2.75	15.25	5.25	24.50	5.00
4th	91.00	3.50	22.50	8.00	37.75	9.25
October 1st	106.25	5.25	31.75	12.75	59.25	10.00
2nd	112.50	9.00	38.25	19.25	68.50	15.00
3rd	121.00	13.50	50.25	22.00	79.00	26.75
4th	141.75	21.00	65.75	35.50	95.75	45.00
Overall mean	78.42 f	7.45 a	33.53 d	14.06 b	49.75 e	20.53 c
Reduction %	-	90.50	57.24	82.07	36.56	73.82
Mean number of larvae of *Earias insulana* / 25 plant during 2018 season						
July 1st	40.25	5.00	17.25	6.25	24.00	10.00
2nd	49.50	7.25	23.00	10.75	30.25	15.25
3rd	61.25	9.25	27.25	13.25	43.25	20.25
4th	85.75	15.00	33.75	17.25	58.00	30.75
August 1st	111.75	17.25	58.25	24.50	76.75	46.50
2nd	92.00	3.75	33.00	10.50	60.25	17.00
3rd	86.25	2.25	18.25	5.75	36.00	10.00
4th	50.25	1.25	11.50	2.50	15.25	4.25
Septem. 1st	20.00	0.75	8.00	2.25	10.25	3.25
2nd	48.75	2.75	11.25	4.25	18.25	8.75
3rd	77.75	3.75	15.75	9.00	23.75	12.00
4th	96.00	6.00	23.25	15.25	44.25	14.00
October. 1st	98.75	5.75	43.25	21.5	65.00	28.25
2nd	133.00	10.25	60.75	29.50	97.25	33.75
3rd	143.75	20.00	84.25	35.75	113.75	54.25
4th	147.25	28.00	97.75	38.50	137.75	59.50
Overall mean	86.33	8.64	36.03	15.42	53.37	22.98
R.%.	-	89.99	58.26	82.14	38.18	73.38

Means followed by a common letter are not significantly different at the 5% level by DMRT

Table (3): Number of shoot and fruit borer, *Helicoverpa armigera* larvae/ 25 okra plants as affected by intercropping pattern.

Sampling date		Intercropping pattern					
		Okra solid	Okra+ rosemary	Okra+ lemongrass	Okra+ lemon balm	Okra+ spearmint	Okra+ catnip
Mean number of ***Helicoverpa armigera*** larvae / 25 plant during 2017 season							
July	1st	12.50	1.25	4.25	0.75	7.00	15.75
	2nd	31.75	7.75	10.00	1.00	8.25	23.25
	3rd	55.25	10.00	19.00	3.25	13.75	31.25
	4th	124.00	15.00	25.75	11.00	30.00	37.00
August	1st	108.25	27.25	33.25	13.00	41.50	48.00
	2nd	77.25	30.50	57.75	23.25	74.25	95.25
	3rd	41.25	25.25	49.00	4.75	66.00	83.25
	4th	85.00	20.25	40.00	2.00	51.75	63.50
September	1st	96.00	14.25	31.25	1.25	43.50	58.25
	2nd	110.75	9.00	22.50	0.75	31.25	50.00
	3rd	45.25	4.25	14.75	0.25	25.25	30.00
	4th	31.25	2.00	8.25	0.00	15.00	20.25
October	1st	40.50	1.25	4.25	0.00	13.00	13.25
	2nd	38.75	0.50	2.75	0.00	7.25	7.75
	3rd	20.25	0.25	1.25	0.00	4.75	6.75
	4th	9.25	0.00	0.75	0.00	2.25	3.25
Overall mean		57.95	10.55	20.30	3.83	27.17	36.67
R %		-	81.79	64.97	93.39	53.11	36.72
Mean number of ***Helicoverpa armigera*** larvae / 25 plant season 2018 season							
July	1st	16.75	2.25	6.00	0.25	10.25	17.50
	2nd	33.50	8.25	15.75	6.25	20.25	25.50
	3rd	41.25	11.00	21.00	10.50	25.00	35.25
	4th	172.75	23.75	52.5	21.75	53.75	58.75
August	1st	118.25	47.25	68.00	31.00	84.50	96.25
	2nd	72.25	39.00	58.00	15.00	74.25	80.00
	3rd	7.00	31.25	50.25	6.25	66.25	76.75
	4th	99.50	24.00	41.25	1.75	65.50	68.50
September	1st	109.75	15.75	33.50	1.50	44.50	59.25
	2nd	133.25	9.75	25.25	0.50	33.75	45.00
	3rd	89.25	5.00	15.75	0.00	22.25	34.75
	4th	63.50	2.25	9.00	0.00	18.75	25.25
October	1st	55.75	1.5	4.25	0.00	10.00	14.75
	2nd	43.50	0.75	2.00	0.00	4.00	10.75
	3rd	27.75	0.25	1.00	0.00	3.50	6.25
	4th	15.00	0.00	0.25	0.00	1.75	3.25
Overall mean		71.81	13.88	24.91	5.92	31.44	41.11
R %		-	80.67	65.31	91.76	56.22	42.75

Means followed by a common letter are not significantly different at the 5% level by DMRT

Table (4): Percent fruit damage by *Earias insulana* and *Helicoverpa armigera* / 25 okra fruit as affected by intercropping pattern.

Intercropping Pattern	Percentage fruit damage/ 25 okra fruit	
	2017 season	2018 season
Okra+ rosemary	9.33 b	10.55 b
Okra+ lemongrass	17.65 c	18.23 c
Okra+ lemon balm	2.25 a	4.33 a
Okra+ spearmint	35.55 e	36.23 e
Okra+ catnip	24.75 d	26.00 d
Okra control	83.56 f	85.55 f

Means followed by a common letter are significantly different at the 5% level by DMRT

The average population and shoot and fruit damage caused by *E. insulana* and *H. armigera* on okra were low when intercropped with cluster bean (*Cyamous tetragocalobe* L.), besides attracting comparatively high population of *Chrysoperla zastrowi sillemi* (Esben-Petersen) (Neuroptera: Chrysopidae) (Baskaran and Parthiban 2017). Khafagy (2011) reported that intercropping system of kidney bean with sweet basil, geranium, peppermint, spearmint and hot pepper showed highly reduction of *Bemisia tabaci* (Gennadius) (Hemiptera, Aleyrodidae) (eggs, nymphs and adults) compared to kidney bean solid. El-Gobary *et al*. (2014) found that okra plants intercropped with aromatic plants reduced *H. armigera* compared to control (okra solely). Khafagy (2015) reported that intercropping aromatic plants with tomato plants reduced the infestation with *Tuta absoluta* (Meyrick) (Lepidoptera: Gelechiidae), especially on geranium + tomato and increased the numbers of predators especially on sweet basil + tomato compared with tomato solid (control). The pooled average percent fruit infestation by fruit borer was lowest in the marigold intercrop treatment (9.69) followed by coriander intercropping (12.40) on weight basis. While the sole crop (32.14) recorded the highest percent fruit infestation (Surjayanand *et al*., 2016). The data on the pod damage percent revealed that all the intercrop treatments were significantly superior over the chickpea sole crop. Among all the intercrop treatments, lowest per cent pod damage was recorded in chickpea + coriander treatment with 7.74 found among and it was followed by chickpea + lentil, chickpea + safflower, chickpea + tomato and chickpea + mustard with 9.41, 10.19, 11.81 and 12.62 per cent pod damage respectively. The highest pod damage treatments were, chickpea + wheat and chickpea sole with 14.45 and 16.08 per cent pod damage respectively and they were found high damage than other. Chickpea sole was found to be predominantly affected by highest pod damage per cent compared to other treatments with 16.08 per cent (Aditya, 2018).

2. Effect of intercropping aromatic plants between okra on predator population:

Intercropping of okra with lemongrass or catnip encouraged almost all considered predatory insects and true spiders (Table, 5). In 2017 season, the highest population densities, of *C. carnea*; 50.00 and 36.50 individuals / 25 plant were obtained with okra + peppermint and okra + Lemon balm intercropping pattern, respectively. *Coccinella* spp. population densities were highest with okra + lemon balm and okra + catnip, followed by okra +spearmint, but low in plots of okra solid and okra +lemongrass pattern. The highest densities of *P. alfierii* were detected with okra +catnip and okra + lemon balm, with value, of 50.00 and 43.75 individuals / 25 plant, respectively. The same trend was found with *Scymnus* spp. The true spider populations proved to be highest in case of intercropping between okra and lemongrass (53.25) , followed by okra+ catnip (46.50 spiderlings and adults / 25 plants) . The least densities of true spiders were found in plots with okra control, or okra intercropped with spearmint. Other intercropping patterns resulted in intermediate population densities of true spiders. The highest population density of *N.tenuis*, intercropping between okra and lemon balm or spearmint encouraged the occurrence of the predator, with values of 37.25 and 30.75 individuals / 25 plants, respectively. Predatory population densities in 2018 season took a trend very similar to that 2017 season**.**

Table (5): Effect of intercropping okra with aromatic plants on population density of predators in okra fields .

Treatment	Mean No. / 60 leaflets					
	Chrysoperla carnea (eggs, larvae , adults)	*Coccinella* spp (eggs , larvae,adults)	*Paederus alfierii* (adults)	*Scymnus* spp (larvae & adults)	True spider (spiderlings , adults)	*Nesedicoris tenuis* (nymphs & adults)
2017 season						
Okra control	7.50 e	11.00f	12.50f	17.75f	20.50f	5.00f
Okra + lemongrass	15.75 d	16.25e	19.25e	23.50e	**53.25a**	11.25e
Okra + Spearmint	**50.00 a**	28.50c	35.75c	36.00c	26.00e	30.75c
Okra + Rosemary	24.25 d	22.25d	27.50d	30.25d	39.75b	17.00d
Okra + catnip	30.25 c	53.75b	**50.00a**	**55.50a**	46.50c	25.50b
Okra+ Lemon balm	36.50 b	**59.00a**	43.75b	42.25b	32.00d	**37.25a**
2018 season						
Okra control	9.25 f	13.50f	13.50f	20.50f	19.50f	7.75f
Okra + lemongrass	18.50 e	19.75e	20.75e	25.25e	55.25a	13.50e
Okra + Spearmint	**55.00 a**	30.00c	37.00c	40.75c	28.25e	33.25c
Okra + Rosemary	27.25 d	26.00d	29.50d	33.00d	41.00b	19.50d
Okra + catnip	34.25 c	45.25b	53.25a	56.00a	48.75c	26.00b
Okra+ Lemon balm	40.50 b	**60.00a**	46.50b	43.75b	33.50d	40.75a

Means followed by a common letter are not significantly different at the 5% level by DMRT

This finding in agree with Kares *et al.* (1993) and Shalaby *et al*. (1983), who proved that cotton surrounded by maize on the periphery of the plots refuge the highest numbers of predators followed by cotton and maize in alternating rows at the ratio (2:1), then cotton and maize at the ratio (1:1). Accordingly, some environmental manipulation could affect efficiency of a natural enemy during biological control programs of *Helicoverpa* spp. (Roome, 1975) suggested that increasing plant diversity in cropping systems by intercropping crops carrying nectars could enhance effectiveness of natural enemies. When different host plants of *H .armigera* are interplant, population of *H. armigera* and its natural enemies on a crop are influenced by neighboring crops, both directly and indirectly. Direct influences include preference for one crop over the other by ovipositing moths and the movement of larvae and natural enemies between interplant crops. Indirect influences arise when *H. armigera* infestation on one crop is influenced by the population build-up or mortality level on neighboring crops (van den Berg *et al.* ,1993). Hickman and Wratten (1996) refered to increasing in population of natural enemies was attributed to supplying access of nectar-producing plants such as alyssum (*Lobularia maritima* L.). Overall, flowering companion plants have been implemented in a variety of crops including cereals, vegetable crops and fruit orchards to improve conservation biocontrol (Landis *et al*., 2000 and Jonsson *et al*., 2008). Flowering companion plants have been used in different cropping systems to enhance the impact of natural enemies (Begum *et al*., 2004). In addition to food resources, companion plants can provide a shelter to pests away from predators and pesticides as well as favorable microclimates (Hossain *et al*., 2002). A wide variety of natural enemies utilize non-prey food sources. For example, pollen and nectar have been

demonstrated to be highly attractive to a variety of predators including syrphids (Diptera: Syrphidae) (Hickman and Wratten, 1996) and coccinellids (Pemberton and Vandenberg, 1993). Nectar is a source for carbohydrates and provides energy, while pollen supplies nutrients for egg production (Lee *et al*., 2004). Many natural enemies, including predators, require non-prey food items in order to develop and reproduce (Wackers *et al*., 2005).The availability of alternative prey and hosts is likely to mostly benefit generalist natural enemies. But, it has been shown that a better supply of pollen, nectar and honeydew might increase the effectiveness also of specialized predators and parasitoids. In addition, diversified communities provide better habitats for natural enemies because they have a larger variation in microclimates and microhabitats and thus provide better shelter to escape adverse condition. El-Gobary *et al*. (2014) found that okra plants intercropped with aromatic plants increased the associated numbers of predators compared to control (okra solely). Khafagy (2015) reported that intercropping aromatic plants with tomato plants increased the numbers of predators especially on sweet basil + tomato compared with tomato solid (control).

3. Intercropping between okra and aromatic plants on okra yield:

Data in Table (6) present the okra yield as affected by intercropping between okra and aromatic plants. Okra + lemon balm pattern proved to be the best combination. This combination produced 176.00 and 137.75 K.g./ Karat in 2017 and 2018 seasons, respectively. Thus, the yield advantages of okra + lemon balm were 212.89 and 227.83 % as compared to tomato solid in the first and second seasons, respectively. Okra + spearmint intercropping pattern proved to be the second best combination, followed by okra+ rosemary pattern, concerning obtained okra yield, with values of 157.75 and 155.50 and 143.50 and 139.00 K.g./ Karat .in the first and second seasons, respectively. However, the lowest tomato yields were obtained from okra+ lemongrass and okra + catnip patterns in both seasons. Mean of harvested okra pods yield in the previous cropping systems exceeded that of the control (untreated) which recorded the lowest total okra pods yield of 68.73 and 68.37 K.g. / Karat in the two regions (Mansour *et al.,* 2017). The highest tomato yield was obtained when tomato was intercropped with lemongrass than tomato plants only (Khafagy. 2018).

Table (6): Effect of intercropping okra with aromatic plants on okra yield.

Aromatic plant species intercropping+ okra	2017 season		2018 season	
	Yield production (K. g./Karat)	Increase in yield production %	Yield production (K. g./Karat)	Increase in yield production %
Okra control	56.25 f	-	53.00 f	-
lemongrass	115.50 e	105.33 e	113.75 e	114.62 e
Catnip	128.75 d	128.89 d	125.75 d	137.26 d
Rosemary	143.50 c	155.11 c	139.00 c	162.26 c
Spearmint	157.75 b	180.44 b	155.50 b	193.40 b
Lemon balm	176.00 a	212.89 a	173.75 a	227.83 a

Means followed by a common letter are not significantly different at the 5% level by DMRT

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Efficacy of the predatory mites and entomopathogenic fungi against *Thrips tabaci* (Thysanoptera: Thripidae) infesting strawberry in Egypt

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Abstract:

The strawberry is one of the most popular berry fruits of the world. Strawberries are an excellent source of vitamins C and K as well as providing a good dose of fiber, folic acid, manganese and potassium. *Thrips tabaci* Lindeman (Thysanoptera: Thripidae) is a serious insect pests affecting strawberries in all stages of growth. The aim of the present work is to study the efficacy of the predatory mites and entomopathogenic fungi (*Beauveria bassiana*) against *T.tabaci* infesting strawberry in El-Behera Governorate (Bader centre). The results indicated that the two predator mites, *Typhlodromus swirskii* Denmark and *Neoseiulus cucumeris* (Qudemans) (Acari: Phytoseiidae) used to control strawberry T.tabaci by releasing two times at a rate of 5-10 /m^2 during the season of productivity. Four varieties; winterstar, florida, fortuna and markez were used. The efficiency of the predatory mites was different according to the strawberry variety. The predator *T.swirskii* proved to be more efficient than the predator *N. cucumeris*. The use of entomopathogenic fungi, *B. bassiana* was less efficient than the predatory mites and the effect was varied according to the strawberry variety in control *T.tabaci*. The study affirmed that the use of the predatory mites is very important in the integrated control program for strawberry *T.tabaci*.

Introduction

In Egypt, strawberry (*Fragaria* x *ananassa*) is grown as a semi protected crop in open-sided polythene tunnels. *Thrips tabaci* Lindeman (Thysanoptera: Thripidae) is one of the mainly significant insect pests affecting strawberries in each stages of growth (Shakya *et al*., 2010). It generated significant yield loss globally (Lewis, 1997). Thrips are polyphagous nature, transmitted plant pathogens, tiny life cycle and insecticides resistance (Morse and Hoddle, 2006 and Diaz-Montano *et al*., 2011). *T.tabaci* is caused to delay growth of plant and attributed to

decrease yield as resulting for extensive feeding.

At the present time, traditional pesticides use is not feasible, especially with successful of some the predator mites against *T.tabaci*. Many insecticides were efficiency against this species, although not all were agreed for use on strawberry in all countries. However, all options use in pest management control for reducing pest numbers with precedence to the non-chemical control [International Organisation for Biological and Integrated Control (IOBC), 2008]. Accordingly, chemical applications should not be made on a standard protection program, except only when critical. Supremely, crop protection agents should be slight toxicity to non target insects because of used in control program (Cuthbertson, 2004 and Rosell *et al*., 2008). The predatory mites *Typhlodromus swirskii* Denmark and *Neoseiulus cucumeris* (Qudemans) (Acari: Phytoseiidae) are effective in controlling *T. tabaci* (Croft *et al*., 1998; Fitzgerald *et al*., 2007 and Khaliq *et al.*, 2018). However, biological control is only effective when pest populations were low to moderate (Croft *et al.,* 1998) whereas slow acting its (Fitzgerald *et al*., 2007). The main goal of biological control is to maintain environmental balance (Pedigo and Rice, 2015). In this context, the performance cost of natural enemies are cheap and safe (Buitenhuis *et al*., 2007). Also, use of entomopathogenic fungi for example, *Beauveria bassiana* had become very important in controlling *T.tabaci* (Abd El-Salam *et al*., 2013).

The main focus of these studies were measured the efficacy of the predators, *T. swirskii* and *N. cucumeris* against *T.tabaci*. Also, the resistance of strawberry varieties for *T.tabaci* was studied. *B. bassiana* fungi was used as comparison with the predatory mites in integrated pest management.

Materials and methods

1. Experimental design:

Tests were conducted on an area of 1400 m^2 at El-Behera Governorate (Bader centre) from 17th October 2015 to 6th March 2016. Four strawberry varieties were used winterstar, florida, fortuna, markez, each 350m^2 specialized to each variety. Each 350m^2 divided into 4 blocks, the treatment blocks were 100 m^2 for each block while the untreated block was 50 m^2 area and each block is divided into 5 pieces as replicates. Shit plastic separator placed between each treatment and other. The strawberry was cultivated on terraces (15x5m^2 terrace area).

2. Rearing and mass rearing the predator mites:

Two predator mites were used, *T. swirskii* and *N. cucumeris*. The predator mites were reared in Laboratory of Pests and Plant Protection Department in National Research Centre (El-Saeidy and Romeih, 2007). The predator mites were maintained and rearing on mulberry leaves highly infested with *Tetranychus urticae* Koch (Acari: Tetranychidae) previously and transmitting in large plastic boxes 26x15x10 cm inside car refrigerator under 20.0 Cº to the experimental region.

3. Compound used:

Bio-Power is a biological insecticide based on a selective strain of naturally-occurring entomopathogenic fungus *Beauveria bassiana*. The product contains spores and mycelial fragments of *B. bassiana* and is available in liquid (1x109 CFU's/ml). The compound is produced by T. Stanes Company limited.

4. Samples collection:

Thirty strawberry leaves were inspected from each area and the numbers of adult *T.tabaci* per leaf were counted by eye in the field using a x7 head lens (optimizer, Light Craft, London, UK). We used counts of adult *T.tabaci* rather than nymphs because this is more practical for growers and the

assessment of nymphs by eye in the field is unreliable (Gonzalez-Zamora and Garcia-Mari, 2003).The number of *T. swirskii* and *N. cucumeris* predatory per leaf was also counted. Leaves pooled and placed in 70 % alcohol so that *T.tabaci* could be extracted and identified to species. The predatory mites, *T. swirskii* and *N. cucumeris* were released by the growers in all the fields sampled, but with varying amounts and frequencies. The count of the *T.tabaci* individuals was carried out before and after the treated either the predators or bio-compound.

5.Release of the predatory mites :

Discs of mulberry leaves with predatory mite, *T. swirskii* and/or *N.cucumeris* contained 5-10 individuals put on one strawberry plant in 2.0 m^2 area containing 10 strawberry plants approximately. This process carried out 1-2 hours before sunset and 3-4 hours after crop irrigation to warrant suitable air temperature and relative humidity for introduction of predatory mites. The process of release carried out twice during the experimental when *T. tabaci* density reached to 2-3 individuals / leaf.

6. Application of bio-power formulation:

Three applications were carried out by bio-power (*Beauveria bassiana*) at rate of 5.0 ml /l.

7. Statistical analysis

The experimental data were analyzed by one-way Anova analysis of variance. Statistical analysis was carried out with Spss,11.

Results and discussion

1. Efficacy of predators of *Typhlodromus swirskii* and *Neoseiulus cucumeris* compared with *Beauveria bassiana* against *Thrips tabaci* in strawberry:

The results showed in Figure (1) the most susceptible varieties of *T.tabaci* were florida, fortuna and winterstar and markez was the least sensitive. In the first week, the *T.tabaci* numbers began to attack the leaves recording 5.2, 5.0, 4.8 and 4.0 individuals / leaf in florida, fortuna, markez and winterstar, respectively.

The populations fluctuated between rise and little decrease until reached 11.2, 10.2, 9.8 and 8.8 individuals /leaf with winterstar, florida, fortuna and markez, respectively, with the end of the twenty-first week. The results clarified that the highly peak of *T.tabaci* population was 12.4 , 10.6 ,9.6,9.4 individuals /leaf for florida, frotona, winterstar and markez varieties, respectively, during the seventeen and eighteen weeks .The mean number of *T.tabaci* during 21st week was recorded 9.2, 8.0, 7.8, 7.0 individuals / leaf in florida, fortuna, winterstar and markez, respectively. The results manifested that the varieties have different sensitivity for *T.tabaci* infestation.

The results in Table (1) indicated that a difference in the efficiency of both predators against *T.tabaci* individuals with different strawberry variety. The *T. swirskii* predator achieved more efficient with the winterstar variety to reduced the number of *T.tabaci* to 85.5%. Other varieties, the predatory achieved 77.14, 76.59 and 76.52% reduction in the *T.tabaci* numbers with markez, florida and fortuna, respectively. *N. cucumeris* predator achieved to reduce of the *T.tabaci* numbers reached 79.5, 75.3, 74.74 and 72.5% with winterstar, markez, florida and fortuna, respectively. This may refer to the physical of the strawberry leaves varieties.

Predator release for two times at a rate of 5-10 individuals/ m^2 during the twenty one weeks led to decline in the number of *T.tabaci* / leaf to 85.5 and 79.5% by *T. swirskii* and *N. cucumeris* ,respectively, in the winterstar variety. While the blocks which was treated twice time with *B. bassiana* fungi, led to achieved 71.8% reduction in T.tabaci numbers. This indicated that the process

of using predator mites is more efficient than the entomopathogenic fungi. Also, the *T. swirskii* predator was more efficient than *N. cucumeris*. The efficiency of *B. bassiana* was varied with variety difference; the *T.tabaci* numbers was decline to reaching 74.46, 72.5, 71.8 and 69.88% reduction in florida, fortuna, winterstar and markez varieties, respectively.

The population of *T.tabaci* after the predators release and spraying with entomopathogenic fungi illustrated in Figures (2, 3, 4 and 5). Radically differences were detected between the control and the three different treatments. The predators related with *T.tabaci* were phytoseiid mites (*Amblyseius* spp.). The findings suggest that feed more on little instars of *T. tabaci* and if these predators were free during early pest epidemic it would be success in controlling the *T.tabaci* population.

The surface of a host plant had a physical block such as waxy cuticles and/or epidermal structures including trichomes. Thrips damage were harmfully with the amount of epicuticular wax on gladiolus leaves (Zeier and Wright, 1995). Other studies did not recognize any connection between *T.tabaci* feeding harm and morphological constitution such as hairiness, age and area of leaf (Leiss *et al*., 2009 and Mirnezhad *et al*., 2009). Instead, the resistance of plant was mainly affected by chemical constitution in host plant. Plant chemical guard can to be high from both primary and secondary metabolites. Primary metabolites, as dietetic chemicals, are generally favorable for thrips . However, at low concentrations of perfumed amino acids in host plant were connected with reduced T.tabaci feeding damage (Mollema and Cole, 1996). Therefore, the common of studies focus on the plant defense by secondary metabolites. In the mean time, few studies had examined the chemical host resistance to *T.tabaci*. In a study on different chrysanthemum varieties, is obutylamide was recommended to be associated with thrips host plant resistance (Tsao *et al*., 2005). Leiss *et al*. (2011) was developed an ecometabolomic technique to identified resistant and sensitivity plants for thrips. Some compounds were identified for example, jacobine, jaconine and kaempferol glucoside in the wild plant species *Jacobaea vulgaris*, chlorogenic and feroluylquinic acid in chrysanthemum, acylsugars in tomato and sinapic acid, luteolin and β-alanine in carrot (Leiss *et al*.,2009 ; Mirnezhad *et al*.,2009; Tsao *et al.*,2005 and Leiss *et al*., 2011). In a review of Francisco *et al*. (2011) that dissection and discussed the strawberry plant defense mechanism. The authors confirmed that physiological responses of plants at a molecular level will provide valuable information to improve future breeding strategies for new strawberry varieties and to engineer strawberry plants for durable and broad-spectrum disease resistance. In turn, this will lead to a reduction in use of chemicals and in environmental risks. Mouden *et al*. (2017) stated that some metabolites of plants had shown harmful effect on thrips and also had attention for prevention of human health by their antioxidant functions. In strawberry, a downbeat relationship between the oviposition and survival of the two-spotted spider mite *T. urticae*, the number and density of glandular and non-glandular trichomes in leaves has been reported (Luczynski *et al.*, 1990b). However, Kishaba *et al.* (1972) proposed that foliar character might be related to spider mite susceptibility. The resistance of strawberry cultivars and other plant attributed the density of non-glandular trichomes and pre-formed glandular trichomes that containing oxidative enzymes. Also, cultivars that had yellow-green, glossy to semi-glossy leaf surfaces were less attractive to onion *T.tabaci* compared with other cultivars that had

blue green and waxy. Thus, semiglossy onion cultivars with low levels of epicuticular waxes to glossy should be important in onion thrips control strategies (Steinite and Levinsh , 2003; Diaz-Montano *et al.*, 2012 and Damon *et al*., 2014).

Brown *et al.* (1999) reported that the predation efficacy of predatory mites, *N. cucumeris* against *Frankliniella occidentalis* (Pergande) (Thysanoptera: Thripidae) was different on different plant species. The variation in predation efficacy might be due to differences in plant style, surface structure of leaves (Kareiva and Sahakian, 1990) and plant chemistry (Price *et al*., 1980). The susceptibility of herbivores to predators is often related to the nutritional quality and quantity of plants in orders to attractive the herbivores for feeding (Price *et al*., 1980).

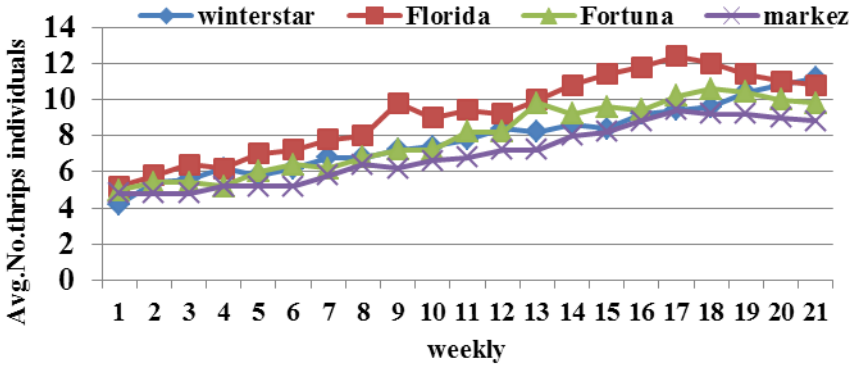
Several species of *Amblyseius* Berlese have been found as predators of *T.tabaci*. The first greedy mites used for *T.tabaci* control were *Amblyseius barkeri* Hughes (Acari: Phytoseiidae) and *N. cucumeris* which mainly eat upon larval first instar. Result to the insufficient control, a number of other mites had been deliberated to find a greater *T. tabaci* predator. A pair of *N. cucumeris* can feed on more than five *T.tabaci* /day and prefer tiny individuals (Riudavets, 1995 and Sabelis and van Rijn, 1997). *N. cucumeris* reduced *T. tabaci* numbers to more than 80.0 % on cucumber plants (Hassan *et al*., 2008), while *A. swirskii* favorite to eat on thrips larvae (Xu and Enkegaard, 2010 and Kutuk *et al*., 2011). An adult female of *Amblyseius fallacies* Garman (Acari: Phytoseiidae) devoured an average of 21.43 and 26.86 thrips at temperatures of 20 °C and 30 °C, respectively, during its life cycle (Abdel-Karim and Abd El-Wareth ,2012). *A. barkeri* is an oligophagous predatory mite against *T. urticae* and *T. tabaci* infestations (initial instars) on cucumber and pepper plants (Karg *et al*., 1987; Hansen, 1988; Bakker and Sabelis, 1989 and Fan and Petit, 1994). Metwally *et al*. (2008) found that *N. barkeri* females produce 1.9, 2.1, 2.3 eggs per day feeding on *T. urticae*, *T. tabaci* nymphs and eriophyid mites, respectively. Species such as *Amblydromalus limonicus* (Garman and McGregor) (Acari: Phytoseiidae), *A. swirskii*, *Amblyseius degenerans* (Berlese, 1889) and *Amblyseius montdorensis* (Schicha) confirm successful predators of thrips in sweet pepper and chrysanthemum (Messelink *et al*., 2008; Wimmer *et al*., 2008; Arthurs *et al*., 2009; Knapp *et al*.,2013; Buitenhuis *et al*.,2015; and Hewitt *et al*.,2015). Efficiency of *A. swirskii* against thrips in biocontrol agent is also influenced by increased trichomes densities hinder in host plant species (Buitenhuis *et al*.,2014). *T. tabaci* could devour *A. swirskii* eggs and female predators were observed preferentially to oviposit at sites in absence *T.tabaci*, or to kill more T.tabaci at oviposition sites to protect their young (De Almeida and Jansen, 2013). Thrips are not the best food source for mites. Therefore, the addition of supplemental food to *A. swirskii* has recently been investigated. Supplying pollen improved the performance of *A. swirskii* in thrips control on chrysanthemum (Vangansbeke *et al*., 2016). Efficient predator of T.tabaci, *A. swirskii* was easily reared that allowed economic mass production. Since , 2005, *A. swirskii* had become used for biological

control program of *T.tabaci* and whiteflies in crops worldwide. Khaliq *et al.* (2018) found that predatory mites, *N. barkeri* eat more on larval first instar than second larval instar and adults of *T.tabaci*. However, consumption rate of *N. barkeri* was rather higher during the initial 12 hours of feeding then slowed down later (24 h); this may be result to aggressive predation initially.

Table (1): Efficacy of predator mites and entomopathogenic fungi against *Thrips tabaci*.

Varieties	Winterstar			Florida			Fortuna			Markez		
Predatory & Fungus	Mean No. *Thrips tabaci* individuals/ leaf±SE		% Reduction	Mean No. *Thrips tabaci* individuals/ leaf±SE		% Reduction	Mean No. *Thrips tabaci* individuals/ leaf±SE		% Reduction	Mean No. *Thrips tabaci* individuals/ leaf±SE		% Reduction
	Before treatment	After treatment by 21 week		Before treatment	After treatment by 21 week		Before treatment	After treatment by 21 week		Before treatment	After treatment by 21 week	
T. swirskii	3.6±0.5a	1.2±0.2b	85.5	4.2±0.58a	2.2±0.8b	76.59	4.6±0.6a	1.8±0.5b	76.52	4.6±0.5a	1.6±0.5b	77.14
N. cucumeris	3.4±0.8a	1.6±0.24b	79.5	4.6±1.2a	2.6±0.5b	74.74	4.8±0.9a	2.2±0.3b	72.5	4.8±0.01a	1. 8±0.4b	75.35
B. bassiana	3.4±0.51a	2.2±0.58b	71.8	4.2±1.1a	2.4±0.6b	74.46	4.8±1.2a	2.2±0.5b	72.5	4.8±0.9a	2.2±0.5b	69.88
Cont.	3.4±0.7a	7.8±2.1a	------	4.2±1.59a	9.2±0.7a	------	4.8±0.7a	8.0±2.0a	------	4.6±0.7a	7.0±1.3a	------
F	0.02UN	7.5UN	------	0.02UN	20.04*	------	0.01UN	7.18UN	------	0.01UN	10.9UN	------
LSD $_{0.5}$	2.03	3.37	------	3.57	2.06	------	2.67	3.32	------	3.3	2.34	------

Means within columns followed by the same letter are not significantly different.
(UN means unsignificant)



Figure(1): Senstivity of differrent strawberry varities for *Thrips tabaci* infestation

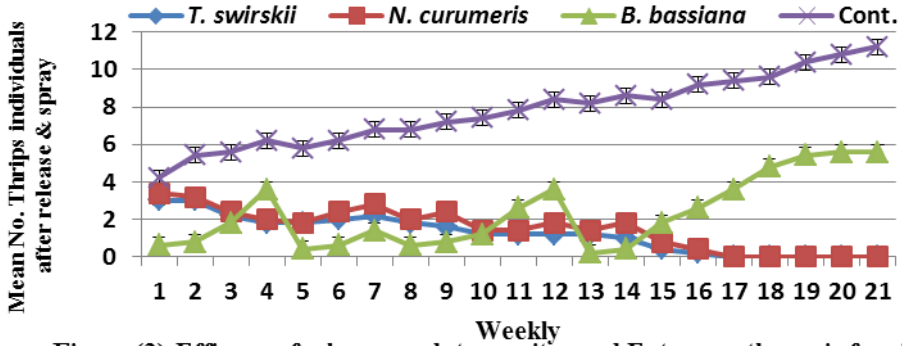
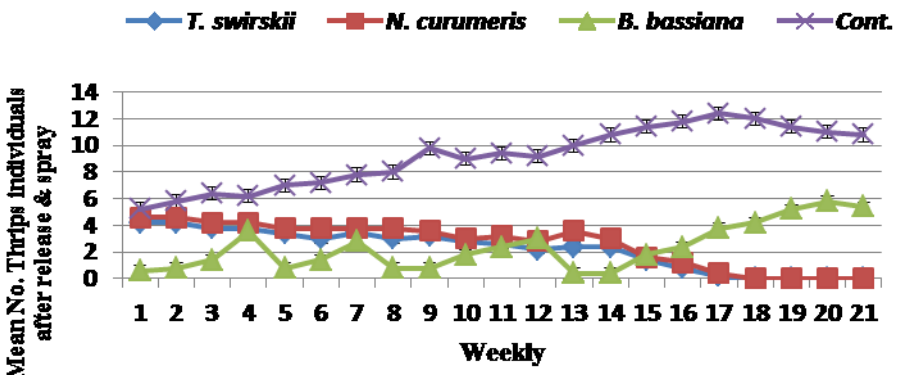
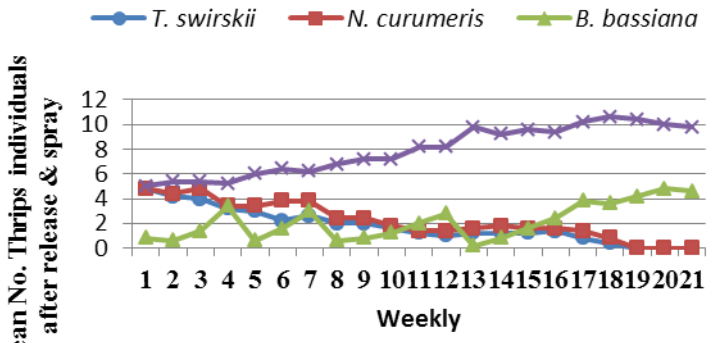


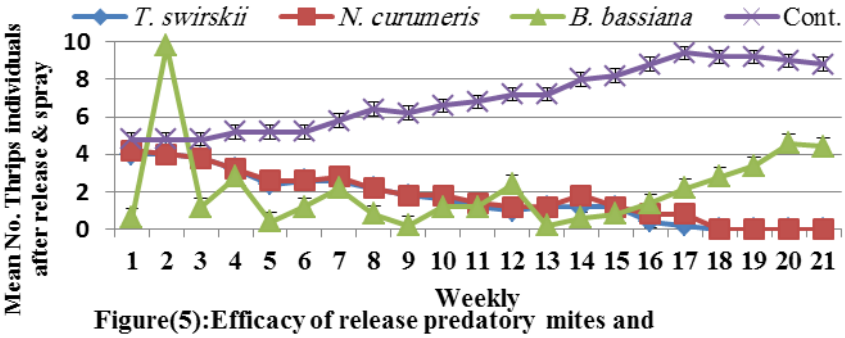
Figure (2):Efficacy of release predatory mites and Entomopathogenic fungi against *Thrips tabaci* individuals on strawberry (Winterstar variety)



Figure(3):Efficacy of release predatory mites and Entomopathogenic fungi against *Thrips tabaci* individuals on strawberry (Florida variety)



Figure(4):Efficacy of release predatory mites and Entomopathogenic fungi against *Thrips tabaci*individuals on strawberry (Fronta variety)



Figure(5):Efficacy of release predatory mites and Entomopathogenic fungi against *Thrips tabaci* individuals on strawberry (Markez variety)

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Exploration of gut bacteria in white grub *Anomala* sp. (Coleoptera: Scarabaeidae) , a major pest of vegetables and fruit trees in India

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Abstract:

White grubs are among the most destructive pests living in the soils causing severe economic loss to most of the agriculturally important crops. An attempt was made in the present study to isolate the gut bacterial communities of white grub, *Anomala* sp. (Coleoptera: Scarabaeidae) collected from citrus trees soil. A culture based approach was deployed for exploring the diversity of gut bacterial isolates in this pest. About 24 bacterial species were isolated from the gut of this insect by using diverse growth media and generic identification of the same was done based on 16s ribosomal RNA gene identification method. The proportional distribution of the gut bacteria revealed that bacterium *Ochrobactrum* sp. (25 %) was the most dominant one followed by *Bacillus* (21 %), *Citrobacter* (21 %), *Pseudomonas* (17 %), *Enterobacter* (8%) and *Paenibacillus* (8%). Phylogenetic association between these gut bacterial isolates, their possible functional role and scope and utility of these gut bacterial isolates in pest management have been discussed in this work.

Introduction

The grubs of scarabaeids are among the most destructive root feeders and of soil insect pests thus cause serious economic losses to diverse agricultural and horticultural crops across the world (Huang *et al*., 2010).

Insects signify as one of the largest reservoirs of bacterial diversity on earth and about 15 per cent of all insects harbour diverse communities of bacteria (Brooks, 1963 and Moran *et al*., 2008). The insect bacterial association has co-evolved for more than 250 million years and have resulted in manifold interactions between insects and bacteria, ranging from pathogenicity to highly complex symbiotic relationships (Douglas and Beard, 1996 and Oliver *et al*., 2005). The number of bacteria within an insect outnumbers the total number of cells within the insect body (Ann and Fergus, 2006). These gut bacteria may play a role in the nutrition, physiology, reproduction, overall, growth and development of the insect host (Dale *et al*., 2006).

Recent studies have shown that the native gut inhabitants drive the

intestinal immunity in order to regulate the colonization of the gut by other non-indigenous microbes including pathogens. The collective genome of indigenous microbiota is described by the term 'microbiome' (Lederberg and McCray, 2001). It has been recognized that some bacteria can incorporate their whole genome into the host DNA for successful transmission (Dunning-Hotopp *et al*., 2007). The composition of the gut flora reflects natural selection at both the microbial and host levels that forms a mutualistic relationship with each other. Petri (1910) described one of the first bacterial symbiotic associations in an insect species, the olive fly, *Bactrocera oleae* (Rossi) (Diptera : Tephritidae) .

The hindgut of scarabaeid grubs have a unique physiological structure called fermentation chamber, which is nothing but an enlarged and modified ileum housing a complex and dense aerobic and anaerobic microbial communities (Cazemier *et al*., 1997). Previous studies had shown that 25–65 per cent of the ingested pure cellulose were degraded by Scarabaeid grubs and the intestinal bacteria present in the hindgut were found to be associated with the cellulose degradation (Cazemier *et al*., 1997). Furthermore, several cellulolytic bacterial species have been successfully isolated from the gut contents of some Scarabaeids (Cazemier *et al*., 2003). These studies demonstrated that the hindgut of scarab larvae represent an ideal resource for identifying microorganisms and enzymes that can be used for biofuel production and to improve biofuel production technology (Huang *et al.*, 2010).

Usage of broad range 16SRNA gene as a tool for identification of bacteria is possible because the 16S ribosomal RNA (16s rRNA) gene is present in all bacteria (Woese, 1987). The 16S rRNA gene has a highly conserved nucleotide sequences, scattered with variable regions that are genus or species-specific. Bacteria can be identified by nucleotide sequencing of the PCR product followed by comparison of this sequence with the known sequences stored in a database (Clarridge, 2004). The previous works for bacterial isolation from Scarabaeid larval gut concerned about many species, while, little is known about the gut bacterial diversity of *Anomala* sp. a major pest of several fruits and vegetable crops.

The present work explores the gut bacterial diversity of this important pest of agricultural crops and the information generated may be of practical utility in exploiting the gut microbes for management of this pest. Studies were carried out on isolation and identification of bacteria culture by using the 16srRNA gene sequence from the gut of white grub, *Anomala* sp. Besides, phylogenetic analysis was done to understand the relationship between the gut bacterial isolates. The possible functional role and scope and utility of these gut bacterial isolates in pest management have been discussed.

Materials and methods

1. Larval collection:

Fully grown healthy third instar grub of *Anomala* sp. were collected from citrus trees from the experimental fields of Indian Agricultural Research Institute, New Delhi, (28° 38′ 5.430″ N; 77° 09′ 8.410″ E), India . Grubs were maintained individually in rearing jars containing sprouted potatoes in a soil medium at 25±2˚ C and 60 % RH inside the insect growth chambers.

2.Dissection and extraction of guts:

The grubs of *Anomala* sp. selected for this study were pre starved for 24h. and subsequently anesthetised at -20˚ C before extraction of gut compartments. The grubs were surface sterilized with 70 % ethanol for 60 sec. followed by immersion in 5 % sodium hypochlorite (NaOCl) solution followed

by thorough rinsing with sterilized water to remove the disinfectant. The surface sterilized larvae were then dissected under aseptic conditions to extract the intestinal tract. The gut was dissected to get the mid-gut and hind gut sections including the fermentation chamber. The extracted gut compartments of each grub were homogenised in 0.85% NaCl using a sterile motorised homogenizer. Care was taken to extract the gut contents under aseptic conditions to avoid contamination of gut sections.

3. Isolation and enumeration of gut bacteria:

The isolation of gut bacteria from *Anomala sp*. was done by using three different media: Nutrient Agar, Brain Heart Infusion Agar and *Pseudomonas* Isolation Agar. The media were autoclaved at 121˚ C for 20 min. The gut homogenate samples were serially diluted in NaCl solution and spread on agar plates. The inoculated plates were incubated at 37˚ C for 24h. The colonies were differentiated on the basis of their size, colour and morphology and a single isolate was transferred to an agar slant. Enumeration of gut bacterial isolates was performed by counting the colony forming units (CFU). The mean values of CFU were used to calculate the viable count of bacteria. After incubation of 24 h, the colonies were picked up from the spread plate and purified by streaking on respective agar plates. Streaking of gut bacteria was repeated for four to five times to ensure the purity of each bacterial culture. Gram Staining was differentiated as Gram positive or Gram negative bacteria. The purified strains were maintained in glycerol stock at -80 ˚C. For the experimental purposes, the bacteria were revived in nutrient broth containing 3.0 g/l beef extract and 5.0 g/l peptone.

4. DNA Extraction and PCR Amplification of 16S rRNA:

Isolated gut bacterial cultures were generically identified by using 16S rRNA gene sequencing technique. The bacterial isolates were grown in nutrient broth for 24h. at 37˚ C. The inoculated broth was then centrifuged at 10621g. to separate the pellet and the supernatant. The pellet of the broth was then used for DNA extraction using a modified cetyltrimethylammoniumbromide (CTAB) method. The quality of bacterial nucleic acid was checked on an agarose gel. Bacterial DNA was amplified by using universal primers 27F (5`-AGAGTTTGATCCTGGCTCAG-3`)1492R-(5'-AAGGAGGTGATCCAGCCGCA-3'). The PCR was carried out in an AB-Applied Biosystems thermal Cycler as follows: one cycle at 94.0 ˚ C for 5 min, 30 cycles at 94 ˚ C for 1 min, 58 ˚ C for 1 min and 72 ˚ C for 1 min 40 s, followed by 72 ˚ C for 10 min and 4 ˚ C forever. The PCR products were then examined on horizontal gel electrophoresis on a 0.8% agarose gel, and the bands were visualized by staining with ethidium bromide. Gels were visualized under UV Gel Documentation system of Alpha Imager™ gel imaging system. The PCR products were sequenced by Sanger's sequencing technique with M/s. Sci Genome Pvt Ltd, India. The high-quality curated sequences of bacterial isolates were compared with the 16S rRNA sequences retrieved from Gen Bank data base by using the Basic Logic Alignment Search Tool (BLAST) algorithm. Bacterial isolates were generically identified based on their similarity with existing sequences.

5. Phylogenetic tree analysis:

The high quality sequences were chosen and thus assigned to phylogenetic tree analysis. The phylogenetic tree for the gut bacteria isolated from *Anomala* sp. was constructed. The sequences were assembled and aligned BIOEDIT V. 7.0 (Hall, 1999) and tree was constructed using MEGA V. 6.0 software program (Tamura *et al*., 2011) by using the Maximum parsimony method. To

calculate the support for each clade, bootstrap analysis was performed with 1000 replications (Felsenstein, 1985). *Geothrix fermentans* (U41563.1) was chosen as an out-group and the sequence of which was obtained from National Centre for Biotechnology Information (NCBI) database.

6.Statistical analysis:

The data of colony forming unit was analyzed using one way analysis of variance and Tukey's honest significant difference post hoc test by using SPSS 16.0 program.

Results and discussin

1. Isolation of gut bacteria:

The isolation of bacterial flora from the intestinal tract of the *Anomala* sp. was carried out by deploying a culture-dependent approach coupled with 16S rRNA gene sequencing for generic identification of bacteria. A total of 35 pure colonies of gut bacteria were isolated from the larvae of *Anomala* sp. on different media. The gut bacterial counts were enumerated on different media and were expressed as colony forming units (CFU). The average CFU of gut bacteria was calculated from all three media and it was found to be 2.21± 0.15 on Nutrient agar media, 2.15±0.10 on Brain Heart media and 1.15±0.11 on *Pseudomonas* isolation agar media. The results revealed that the mean CFU of the gut bacteria was found to be significantly higher on Nutrient agar media as compared to other two media. However it was found that the maximum CFU was observed on the Brain Heart Infusion agar. The mean CFU of these gut bacterial isolates are given in the Table (1) .

Table (1): Colony forming unit (CFU) means of isolated bacteria from three different media of *Anamola* sp. gut.

No.	Media used for the isolation	Colony Forming Unit CFUml^{-1} (× 10^4)
1	Nutrient agar	1.15±0.11^c
2	*Pseudomonas* agar	2.21±0.10^a
3	Brain Heart Infusion Agar	2.15±0.15^b
	CV%	28.10
	SE(M)	0.13

CV is the Coefficient of Variation; SE (M) is the Standard Error of the mean; the values after ± indicates the standard deviation. Means of the Colony Forming Units (CFU) were calculated for the gut bacteria of 3rd instars of *Anamola* Sp. The analysis was done by using the SPSS v.16.0.

2.16srRNA gene sequencing:

The sequences of gut bacterial isolates obtained were checked for the quality and 24 unique, high quality, non-repetitive and no redundant sequences were shortlisted for further analysis. Comparative BLAST analysis revealed that most of the bacterial isolates had shown 99 per cent similarity and a few showed 88 % similarity to their closest relatives retrieved from the GenBank database. The 16S rRNA sequences of gut bacterial isolates generated from this study were submitted to GenBank (accession No. MK235187 to MK235210) (Table, 2).

The results showed that the gut of *Anomala* sp. consisted of diverse gut bacteria with *α-proteobacteria* being the most dominant group represented by the genus *Ochrobactrum* sp. constituting 25 per cent of the total gut bacterial isolates followed by the *γ-proteobacteria* group represented by the genus *Citrobacter* (20.8%) consisting of single species *viz., C. Koseri; Pseudomonas* (16.6%): *P. aeruginosa and Enterobacter* (8.33%) were the other bacteria belonging to the *γ-Proteobacteria. Firmicutes* were represented by genera such as *Bacillus* spp. (20.8%) and *Paenibacillus* (8.33%) represented by *P. Jamilae* (Figure, 1).

Table (2): Gen Bank accession details of gut bacterial isolates from the white grub *Anamola* sp.

Isolate No.	Organism of closest match identified from Genbank	Gene Bank Accession no.	Similarity %	Family	Class
1	*Pseudomonas aeruginosa*	MK235187	99%	*Pseudomonadaceae*	*γ-proteobacteria*
2	*Pseudomonas aeruginosa*	MK235188	99%	*Pseudomonadaceae*	*γ-proteobacteria*
3	*Bacillus pseudomycoides*	MK235189	99%	*Bacillaceae*	*Bacilli*
4	*Ochrobactrum* sp.	MK235190	99%	*Brucellaceae*	*α-proteobacteria*
5	*Ochrobactrum* sp.	MK235191	99%	*Brucellaceae*	*α-proteobacteria*
6	*Paenibacillus jamilae*	MK235192	99%	*Paenibacillaceae*	*Bacilli*
7	*Ochrobactrum anthropi*	MK235193	99%	*Brucellaceae*	*α-proteobacteria*
8	*Bacillus aryabhattai*	MK235194	99%	*Bacillaceae*	*Bacilli*
9	*Citrobacter koseri*	MK235195	99%	*Enterobacteriaceae*	*γ-proteobacteria*
10	*Citrobacter koseri*	MK235196	99%	*Enterobacteriaceae*	*γ-proteobacteria*
11	*Citrobacter koseri*	MK235197	88%	*Enterobacteriaceae*	*γ-proteobacteria*
12	*Bacillus aryabhattai*	MK235198	100%	*Bacillaceae*	*Bacilli*
13	*Citrobacter koseri*	MK235199	99%	*Enterobacteriaceae*	*γ-proteobacteria*
14	*Bacillus* sp.	MK235200	99%	*Bacillaceae*	*Bacilli*
15	*Ochrobactrum* sp.	MK235201	99%	*Brucellaceae*	*α-proteobacteria*
16	*Enterobacter* sp.	MK235202	99%	*Enterobacteriaceae*	*γ-proteobacteria*
17	*Pseudomonas aeruginosa*	MK235203	99%	*Pseudomonadaceae*	*γ-proteobacteria*
18	*Paenibacillus jamilae*	MK235204	99%	*Paenibacillaceae*	*Bacilli*
19	*Citrobacter koseri*	MK235205	99%	*Enterobacteriaceae*	*γ-proteobacteria*
20	*Bacillus* sp.	MK235206	100%	*Bacillaceae*	*Bacilli*
21	*Pseudomonas aeruginosa*	MK235207	99%	*Pseudomonadaceae*	*γ-proteobacteria*
22	*Ochrobactrum anthropi*	MK235208	99%	*Brucellaceae*	*α-proteobacteria*
23	*Enterobacter* sp.	MK235209	99%	*Enterobacteriaceae*	*γ-proteobacteria*
24	*Ochrobactrum* sp.	MK235210	99%	*Brucellaceae*	*α-proteobacteria*

3.Phylogenetic tree analysis:

A total of 24 non reductant sequences were curated and aligned with the outgroup sequence of *Geothrix fermentans* (Accession no. U41563.1). Phylogenetic tree was analysed using Maximum Parsimony algorithm and is shown in (Figure, 2). The phylogenetic tree of the gut bacterial isolates of *Anomala* sp. showed seven different clades. Four clades of the phylogenetic group belong to the group *γ-proteobacteria* with the genus *Enterobacter* (with 2 isolates) and *Pseudomonas* (with 4 isolates) and two clades represented by the genus *Citrobacter* (with five bacterial isolates). Another major clade belongs to the group *α-proteobacteria* with 6 bacterial isolates of the genus *Ochrobactrum* which is found to be the most predominant gut inhabitant of *Anomala* Sp. Other two clades represented by the group *Bacilli* with two isolates of genus *Paenibacillus* and *Firmicutes* with five isolates of *Bacillus.*

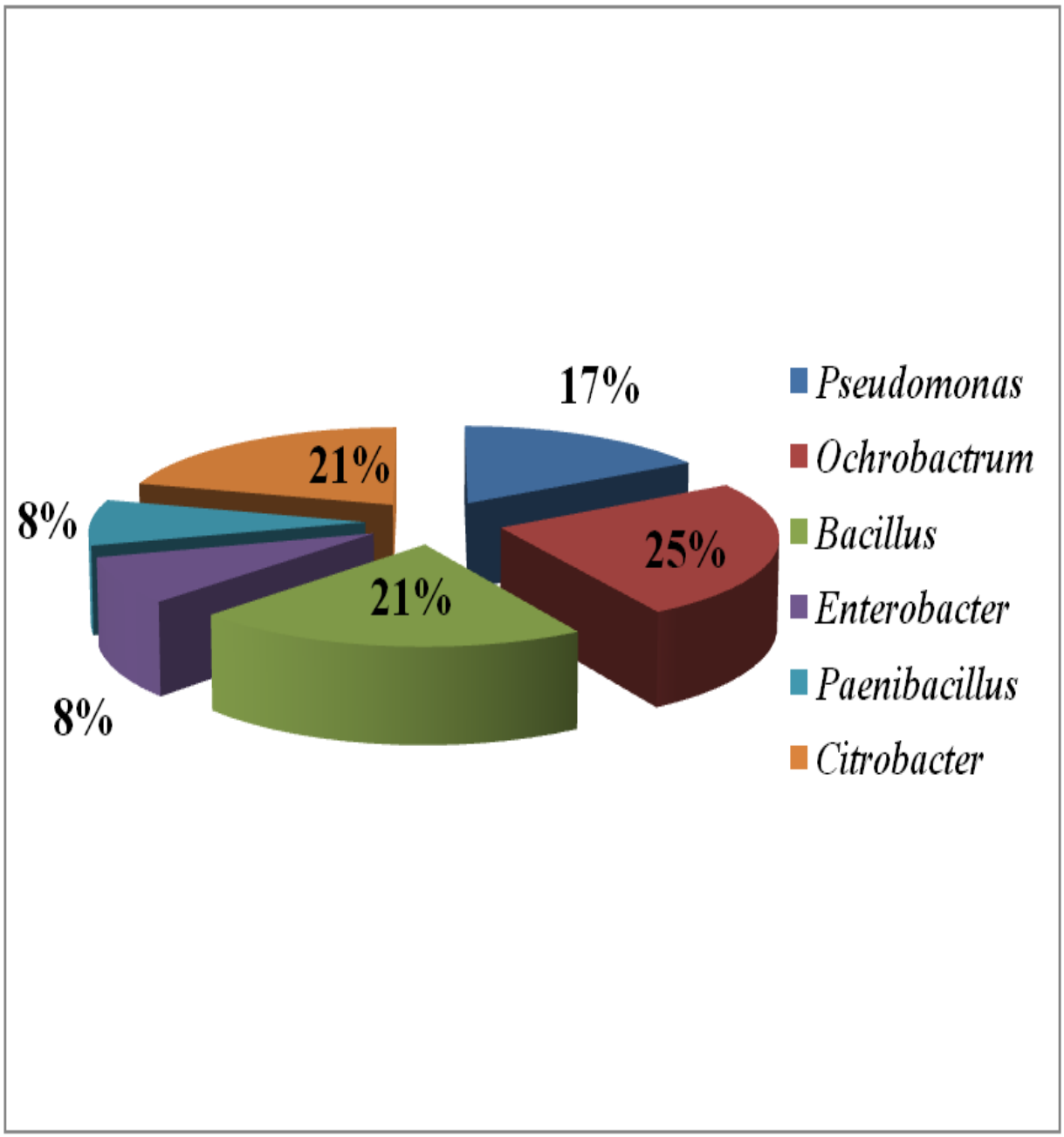

Figure 1: Proportionate distribution of 24 gut bacteria isolated and identified from *Anamola* sp. at genus level.Distribution of different bacteria among the gut of *Anamola* sp. Pseudomonas is 17 percent, Orchrobactrum is 25 percent, Bacillus is 21 percent, Enterobacter is 8 percent, Paenibacillus is 8 percent and Citrobacter is 21 percent. Among all the presence of Orchrobactrum was found more than other bacteria. The presence of Bacillus , Citrobacter, Paenibacillus and Enterobacter was the same.

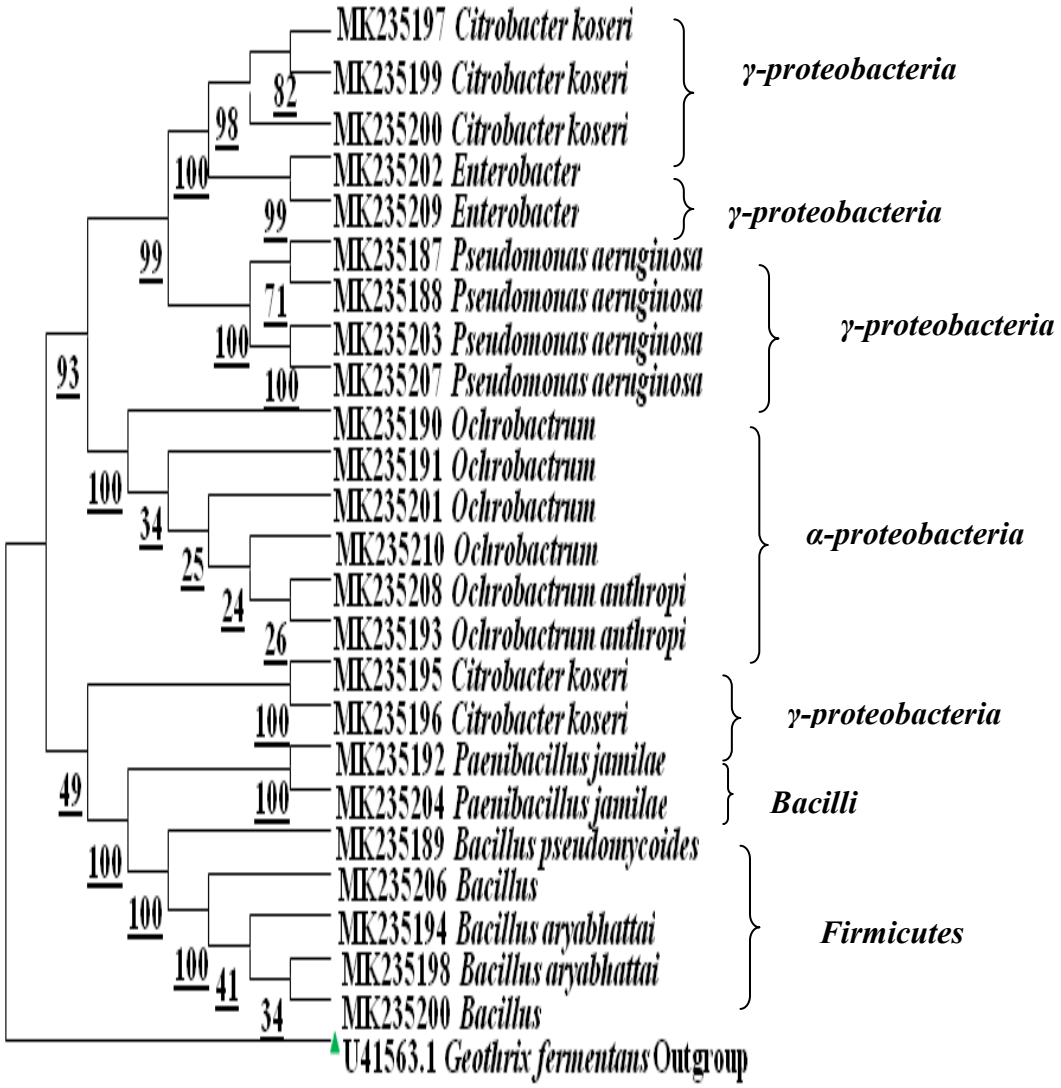

Figure (2): Phylogenetic tree of 24 bacteria from the gut of *Anamola* sp. 16S rRNA gene sequences aligned using Bioedit 7.0, with the ClustalW program, and a phylogenetic tree was constructed based on the Maximum parsimony algorithm supportrd by bootstrap values with 1000. symbol indicates the outgroup used in the phylogenetic tree.

Very little is known about the gut bacterial diversity of the *Anomala* sp. However, there are similar results were obtained by (Lehman *et al*., 2008), they reported the presence of *Citrobacter freundii* and *Pseudomonas sp.* inhabiting the digestive tract of ground beetle, *Poecilus chalcites*. We used culture-dependent 16S rRNA gene sequence-based approaches to identify six major genera of bacteria *Pseudomonas*, *Ochrobactrum*, *Paenibacillus*, *Citrobacter*, *Bacillus* and *Enterobacter.* Results of the present study revealed that a variety of gut bacteria belonging to different groups such as *γ-proteobacteria, Firmicutes, α-*proteobacteria inhabit the gut of *Anomala* sp. Studies with Lepidopteran insects showed that *Proteobacteria* and

Firmicutes were the dominant gut microflora (Broderick *et al*., 2004). The presence of *Pseudomonas* and *Bacillus* species has been documented as the dominant bacterial communities in the gut of the gypsy moth, *Lymantria dispar* (Lepidoptera), (Broderick *et al*., 2004). Similarly, Gayatri Priya *et al*. (2012) isolated and identified the members of *Bacillus firmus* and *Bacillus niabense*, *Paenibacillus jamilae*, *Cellulomonas variformis, Acinetobacter schindleri*, *Micrococcus yunnanesis*, *Enterobacter sp.* and *Enterococcus cassiliflavus* from the midguts of fifth-instar larvae of the lepidopteran moth *Helicoverpa armigera* by using cultural techniques. Cheng *et al*. (2017) reported that the gut symbiont, *Citrobacter* sp. isolated from the peach fruit fly *Bactrocera dorsalis* played a key role in the degradation of Organophosphate insecticides. Similarly, gut bacterial species isolated from the silkworm *Bombyx mori* were including *Pseudomonas vulgaris*, *Klebsiella pneumoniae* and *Citrobacter freundii* and they were known to have high cellulolytic activity, while, *Pseudomonas fluorescens* and *Erwinia sp*. showed good pectinolytic activity, moreover, *Aeromonas* sp. and *Serratia liquefaciens* were found to be cellulolytic and pectinolytic (Anand *et al*. , 2009). A similar study conducted by Desiely *et al*. (2010) reported the presence of 39 bacterial genera including *Serratia*, *Enterobacter*, *Klebsiella*, *Pantoea* and *Citrobacter* from the gut of the mosquito *Aedes aegypti*. Also, Park *et al.* (2009) isolated bacteria belonging to the genus *Paenibacillus* from the subterranean termite gut *Diestrammena apicalis* a novel bacterium capable of degrading pectin which was identified by 16srRNA gene sequence analysis. Results concerning *Ochrobactrum sp*. are in agreement with those obtained by Huang *et al.* (2010), it was identified as a potential cellulolytic bacteria associated with the fermentation chamber of a Scarabaeid beetle, *Holotrichia parallela.* Detailed functional characterization of the gut bacterial isolates and identified in this study offers scope for utilization some of these isolates as cellulose degraders in bioremediation, preparation of microbial consortia for decomposing of plant wastes, bio fuel potential and development of bacterial volatile based novel lures for monitoring and mass trapping of this pest. Further studies are needed for characterization and utilization of the gut bacterial isolates from *Anomala* sp.

Symbiotic microorganisms present one way to convene the expected demand for novel insect pest management strategies created by growing human populations and global climate change. Generic characterization of gut bacterial isolates is an essential step towards profiling the cultivable gut bacterial diversity in an organism. The present study outlines a detailed investigation of the composition of common gut symbionts of the white grub *Anomala* sp. and these gut bacteria may help in developing enhanced methods of biological control of this pest.

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Phenolic contents and antimicrobial activity of some Libyan honeys

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Abstract:

The phenolic contents of 6 Libyan honey varieties of different floral sources were determined. Honey samples included the 5 mono-floral honeys, *Ziziphus louts*, *Citrus medica*, *Thymus capitatus, Amygdalus communis* and *Commiphor myrrha,* while the multi-floral honey was Rabia (spring) honey. The analysis of phenolic compounds was performed using High Pressure Liquid Chromatography. Twenty three phenolic components in the different honeys were determined. The highest number of phenolic components were found in the darker honeys, *Thymus* and *Commiphor* followed by *Citrus*, Rabia and *Ziziphus*, respectively. The least number of phenolic components were detected in *Amygdalus* (only 4). *p*-Hydroxybenzoic acid was found in all studied honey varieties, while rutin was not detected in any of honey samples analyzed. Gallic acid and chrysin were found only in *Thymus* honey, Caffeic acid, salicylic acid and pinostrobin were only in *Commiphor* honey, while catechin, daidazein and pyro gallic were detected only in *Citrus* honey. The phenolic contents can be used as a marker for the studied honey varieties. The antimicrobial effect of on *Staphylococcus aureus, Bacillus subtilis, Klebsiella pneumoniae, Enterococcus faecalis, Pseudomonas aeuroginosa, Escherichia coli, Bacteriods* spp., *Sarcina* spp. and *Candida albicans* was studied. All honey samples inhibited the growth of *Escherichia coli* with different degrees, where $P<0.001$. Among all bacteria, *Bacteroids* spp. and *Klebsiella pneumoniae* were the most resistant against most honey samples.

Introduction

Honey is a complex natural food produced from the honey bee *Apis mellifera* feeding on plant nectar of blossoms, exudates of trees and plants, or from honey bees feeding on honeydew produced by hymenoptran insects.Honey is a saturated solution of sugar of 31% glucose and 38% fructose, and its colour and flavor vary considerably depending on it botanical and geographical origin

and of a moisture content of about 17.7% . In addition to minor component of phenolic acids, flavonoids, glucose oxidase, catalase, ascorbic acid, carotenoids, organic acids, and α-tocpherol . Honey contains at least 181 components (White, 1975). Phenolic compounds are common in plants and collected by honey bees with nectar (Scalbert *et al.,* 2005; Fiorani *et al.,* 2006 and Pyrzynska and Biesaga, 2009). Some phenolic compounds have been shown to exhibit antibacterial, antiviral, anti-inflammatory, anticarcenogenic, antiatherogenic, antithrombotic, Immune-modulating and analgesic activity (Evers *et al.,* 2005; Harris *et al.,* 2006; Nasuti *et al.,* 2006 and Viuda-Martos *et al.,* 2008). Phenolic contents, free amino acids, volatile compounds, trace elements as well as physiological and chemical characters have been used to determine the botanical and geographical origin of honey (Senyuva *et al.,* 2009; Ioannis *et al.,* 2014 and Youngsu *et al.,* 2015). Mohamed *et al.,* (2017) studied the physiological characteristics and total phenolic compounds contents of some Libyan honeys collected from the local markets of Banghazi city in east Libya. The samples included the four mono-floral honeys, *Ziziphus louts, Thymus capitatus, Eucalyptus sp.*and *Arbutus pavari,* and the multi-floral honey Al-Rabia. They found that the total phenolic compound content of the samples ranged from 97.67-123.50 mg gallic acid / 100g of honey, with a mean value 100.64 $\pm$ 11.93 mg gallic acid / 100 g.

The use of honey for the treatment of diseases and wounds has been mentioned since ancient time (2100-2000 BC), where Aristotle (384-322 BC) described pale honey for sore eyes and wounds (Mandal and Mandal 2011 and Vallianou *et al.,* 2014).The healing effect of honey could be due to its physical and chemical properties (Snow and Manley-Harris, 2004) and to its antioxidant and antimicrobial activity (Escuredo *et al.,* 2012; Isidorov *et al.,* 2015; Almasaudi *et al.,* 2017 and Leyva-Jimenez *et al.,* 2019). A possible reason for its activity depends on its ability to generate hydrogen peroxide by the bee derived enzyme glucose dehydrogenase (Saleh *et al.,* 2011). Microorganisms such as *Staphylococcus aureus, Staphylococcus epidermis*, *Micrococcus luteus, Streptococcus uberis, Enterococcus faecalis, Pseudomonas aeruginosa*, *Escherichia coli* and *Klebsiella pneumoniae* are frequently isolated from human and animal skin wounds (Nasser *et al.,* 2003 and Altoparlak *et al.,* 2005. Abd-ElAal *et al.* (2007) found that honey has stronger inhibitory effect (85.7%) than the commonly used antimicrobial agents on gram negative bacteria *Pseudomonas aeruginosa, Enterobacter* sp. and *Klebsiella.* A 100% inhibition was recorded for the methicillin-resistant gram positive bacteria *Staphylococcus aureus.*The antimicrobial activity of honey against *Bacillius cereus, Staphylococcus aureus, Pseudomonas aeruginosa, Klebsiella pneumoniae, Morganiella morganii, Micrococcus luteus, Escherichia coli and Candida albicans; Enterococcus faecalis* and the pathogenic fungi *Candidia albicans* has been studied by many authors (Mercan *et al.,* 2007; Isidorov *et al.,* 2015; Almasaudi *et al.,* 2017 and Leyva-Jimenez *et al.*, 2019).

The aim of the present work was to quantify the phenolic contents of 6 Libyan honeys of different floral sources and to evaluate their antimicrobial effects on *Staphylococcus aureus, Bacillus subtilis, Klebsiella pneumoniae, Enterococcus faecalis, Pseudomonas aeuroginosa, Escherichia coli, Bacteriods* spp*., Sarcina* spp. and *Candida albicans.*

Materials and Methods

The present investigation was carried out at the Beekeeping Research

Department, Plant Protection Research Institute, Giza, Egypt.

1. Honey samples:

Six types of Libyan honeys of mono and multi-floral source were collected from selected beekeepers during the harvesting periods and from local markets in western Libya. The honeys of mono-floral source were *Ziziphus louts*, *Citrus medica*, *Thymus capitatus, Amygdalus communis Commiphor myrrha,* while the honey of multi-floral source was Rabia (Spring) honey. Honey samples were kept in dark at room temperature prior to analysis. The samples were investigated microscopically to determine their containing of pollen grain types.

2. Determination of phenolic compounds contents:

The analyses of phenolic components in six Libyan honeys and their potential for floral authentication were evaluated. The analyses included 23 standard flavones (Gallic acid, *p*-Hydroxybenzoic acid, Caffeic acid, Phenol, *p*-coumaric acid, Salicylic acid, Ferulic acid, Cinnamic acid, Quercetin, Chrysin, Galangin, Pinostrobin, Vanillin, 3,5 dimethoxy benzyl alcohol, Catechin, Daidzin, Genstin, Daidazein Gestein, Pyro gallic, and kaempherol).Extraction of phenolic compounds from honey samples was carried out using ethyl alcohol, where one g of honey was dissolved in 10ml ethyl alcohol 70% to prepare a final concentration of 10 % honey solution, and then kept in closed glass tubes for analysis.

3. HPLC Identification:

Identification of phenolic compounds of the honey samples was performed by a JASCO, using a hypersil C_{18} reversed- phase column (250 X 4.66 mm) with 5 µm particle size.

Injection by means of a Rheodyne injection valve with 50 µl fixed loop was used. A constant flow rate of 1 ml min^{-1} was used with two mobile phases (A) 0.5 % acetic acid in distilled water at pH 2.65; and solvent (B) 0.5 % acetic acid in 99.5 % acetonitrile. The elution gradient was linear starting with (A) and ending with (B) over 35 min, using a µv detector set at wavelength 254 nm. Phenolic compounds of each sample were identified by comparing their relative retention times with those of the standards mixture chromatogram. The concentration of individual compound was calculated on the basis of the peak area measurements, and then converted to µg phenolic g^{-1}dry weight. All chemicals and solvents used were in HPLC spectral grade. 23 standard phenolic compounds were obtained from Sigma (St, Louis, USA) and from Merck-Schuchard + (Munich، Germany) chemical companies.

4. Estimation weight % of phenolic compounds:

The scanning of identified phenolic compounds extracted in honey samples by (HPLC) analysis are estimation of weight % for these compound was calculated as follows:

Weight % phenolic = 100 X (PH/PH^*) X (v/v^*) X (w^* x w)

Where: PH: area for sample

PH^*: area of standard

V: volume of sample

V^*: volume of standard

W^*: weight of standard

W: Weight of sample**.**

5.Bacterial strains:

Bacterial strains and *Candida albicans* were kindly donated by the Microbial Genetic Department, Genetic Engineering and Biotechnology Division, National Research Center, Giza, Egypt.

6.Assay of antimicrobial activity:

Antimicrobial activity of honey samples was determined by the disc diffusion method (Collins *et al.*, 1995). A concentration of 20% of each kind of honey in distilled water was prepared in clean sterile test tube and kept in refrigerator at 4^oC to be used for microbiological test.

7.Preparation of the microbial culture:

The tested organisms were inoculated in the appropriate liquid media and incubated at 37 ^{o}C for 24 hours. The microbial culture was used for the preparation of seed layer by inoculating the agar medium with 2% (v/v) of the microbial culture, thoroughly mixed, and immediately used as the seed layer of plates.

8.Preparation of plates:

The appropriate agar medium was distributed at the rate of 7 ml portion in Petri dishes. After solidification 5 ml of the seeded agar was distributed over the surface of the base layer and left for 15 min to solidify. The previously prepared filter paper discs (each disc was moistened with exactly 0.05 ml of the diluted honey) placed side down on the seeded agar and gently pressed with a tip of sterile forceps. Discs were placed symmetrically around the center of the dish. Plates were incubated at 37 ^{o}C for 24 hours. For *P. aeruginosa* and for *M. leutus* , plates were incubated at 30 ^{o}C. Antimicrobial activity was determined measuring the diameter of inhibition zones around the discs to the nearest mm.

Three replicates were prepared for each honey sample. As a positive control method, the antibiotic tetracycline (30 µg) was used, while sucrose sugar solution (20%) was used as a negative control method.

9.Statistical analysis:

Results are expressed as mean $\pm$ standard deviation. ANOVA were applied at a confidence level of 95%.

Results and discussion

The samples of analyzed honey, their local names and their floral sources are listed in Table (1). In our study 23 phenolic components were found in the different honey samples as shown in Table (2) and Graph (1). Gallic acid and traces of chrysin were found to be characteristic for *Thymus*. Caffeic acid, salicylic acid and pinostrobin for *Commiphor*. Catechin, daidazein and pyro gallic for *Citrus*, while *p*-Hydroxybenzoic was detected in all honey samples. The highest number of phenolic components were found in the darker honey *Thymus* and *Commiphor* followed by *Citrus*, Rabia and *Ziziphus*, respectively. Only 4 phenolic components were detected in *Amygdalus*.

In the present study *p*-Hydrobenzoic ranged from 83.85 µg/100 g in *Citrus* 1248.17 µg/100 g in *Commiphor*, phenol from 3416.59 µg/100 g in *Citrus* to 14737.98 µg/100g in *Thymus*, *p*-Coumaric acid from 513.37 µg/ 100g in *Thymus* to 2387.71 µg/ 100g in *Ziziphus*. Ferulic acid was found only in *Citrus* (269.13 µg/ 100g) and in *Thymus* (2520.43 µg/ 100g), while cinnamic acid was detected in both *Ziziphus* and *Commiphor* (4324.11 µg/100g and 3502.63 µg/100g, respectively). Traces of euganol were found in *Amygdalus* (0.81 µg/100g), while its amount in *Thymus* measured 82.41 µg/100g.Traces of galangin were found in both Rabia and *Amygdalus* (0.28 µg/100g and 1.99 µg/100g, respectively).The amount of detected vanillin ranged from 8.44 µg/100g in *Citrus* to 290.20 µg/100g in *Commiphor*, 3,5 dimethoxybenzyl ranged from 0.47 µg/100g in *Citrus* to 10.53 µg/100g in Rabia, daidazin ranged from 2626.99 µg/100g in *Commiphor* to 11943.0 µg/100g in *Amygdalus*, genstin ranged from 2456.45 µg/100g in *Ziziphus* to 1293.85 µg/100g in Rabia, gestein ranged from 75.02 µg/100g in *Thymus* to 295.61 µg/100g in *Commiphor* and kaempherol ranged from 17.44 µg/100g in *Commiphor* to 275.04 µg/ 100g in *Thymus*. The results of inhibition effects of different honey samples in comparison to control are shown in Table (3). Graph (1), show Phenolic contents a marker and discriminant of Libyan honeys.

It was observed that all honey samples inhibited the growth of

Escherichia coli with different degrees, where P<0.001. The lowest effect was recorded for the *Amygdalus* honey with an inhibition zone of 5.33±1.15 mm, while the greatest effects were shown by Rabia and *citrus* honeys with inhibition zones of 22.33± 0.57 mm and 21.0±1.17 mm, respectively. Among all bacteria, *Bacteroids* spp. and *Klebsiella pneumoniae* were the most resistant against most honey samples, while five out of the six honey samples inhibited the growth of *Sarcina* spp. Except *Commiphor,* all honey samples inhibited the growth of the fungus *Candida albicans. Commiphor* honey inhibited only 3 out of the nine tested microorganisms, while *Zizyphus* and Rabia honeys inhibited seven of them. *Escherichia coli, Pseudomonas aeruginosa* and *Bacteriods* spp. were found to be resistant to the antibiotic tetramycine (+ve control), while 20% sucrose sugar solution (-ve control) had no inhibitory effect on all bacterial strains.

Floral source, geographical origin, seasonal and environmental factors and processing affect the honey phenolic composition and antioxidant activities (Al-Mamary *et al.,* 2002; Yao *et al.,* 2003 and 2005; Ioannis *et al.,* 2014 and Youngsu *et al.,* 2015). In the present study b-oh benzoic was found in all studied honey varieties, while rutin was not detected in any of honey samples analyzed. Gallic and chrysin were found only in *Thymus* honey; caffeic acid, salicylic acid and pinostrobin only in *Commiphor* honey; while catechin, daidazein and pyro gallic acid were found only in *Citrus* honey. Quercetin was detected only in multi-floral honey. Our results showed that phenolic contents can be used as a marker for the studied honey varieties. Studying the phenolic contents of *Robinia* honey samples in Croatia, Kenjerić *et al.* (2007) reported that quercetin, kaemperol and chrysin raged from 2.9 to 29.9, 5.7 to 23.8, and 21.1 to 231.1 µg/100g, respectively. Myricetin was not detected in any of the analyzed honey samples. Martos *et al.,* (1997) studied the flavonoids composition of 13 Tunisian honeys (eucalyptus, thyme, rosemay, orange, rape, sunflower and multifloral honey) and propolis. They reported that flavonoid contents varied significantly between 20 and 2,400 µg/g. Quercetin and kaempferol were detected in linden and heather honeys studied by Michalkiewicz *et al.* (2008). Quercetin ranged from 2.0 to 2.6 mg/kg in linden honeys and 0.39 to 0.41 mg/kg in heather honeys. Respective values of for kaempferol were 1.5 to 1.9 mg/kg in linden honeys and from 0.28 to 0.32 mg/kg in heather honeys. Ioannis *et al.* (2014) studied phenolic compounds of Greek thyme honeys from different geographical origin and found that quercetin ranged from 0.58 mg/kg (in honey sample from Irakleio) to 69.00 mg/kg (from Hania), kaemperol ranged from 50.01 mg/kg (from Lakonia) to 61.38 mg/kg (from Hania), chrysin ranged from 0.01 mg/kg (from Hania) to 5.60 mg/kg (from Kefalonia), myricetin ranged from 0.74 mg/kg (from Hania) to 244.67 mg/kg (from Kefalonia) and syringic acid from 1.56 mg/kg (from Irakeio) to 195.4 mg/kg (from Hania).

Dark coloured *Commiphor* and *Thymus* honeys were found to have the highest number of phenolic compounds among the studies honey varieties (10 phenolic compounds). This result agree well with the findings of Bertoncelj *et al.* (2007), who stated that dark coloured varieties of honey have higher levels of phenolic compunds and antioxidant activities, and with the results of Youngsu *et al.* (2015), who found that the dark colour of chestnut honey showed the higher levels of total phenolics than light coloured acacia honey. Ferreira *et al.* (2009) studied the total phenolic contents of Portugues honeys and reported 132.17 mg/kg for

light coloured honeys, 168.44 mg/kg for amber honeys and 204.24 mg/kg for dark honeys. According to the study of Mohamed *et al.* (2017) on the total phenolic compounds contents of some Libyan honeys from Banghazi city (Eastern Libya), Arbutus honey (*Arbutus pavari*) which have the highest optical density value, exhibited the highest phenolic compounds content. Further research studied on physical and chemical characteristics, organic acids, proteins, enzymes and antimicrobial effects of Libyan honeys are recommended.

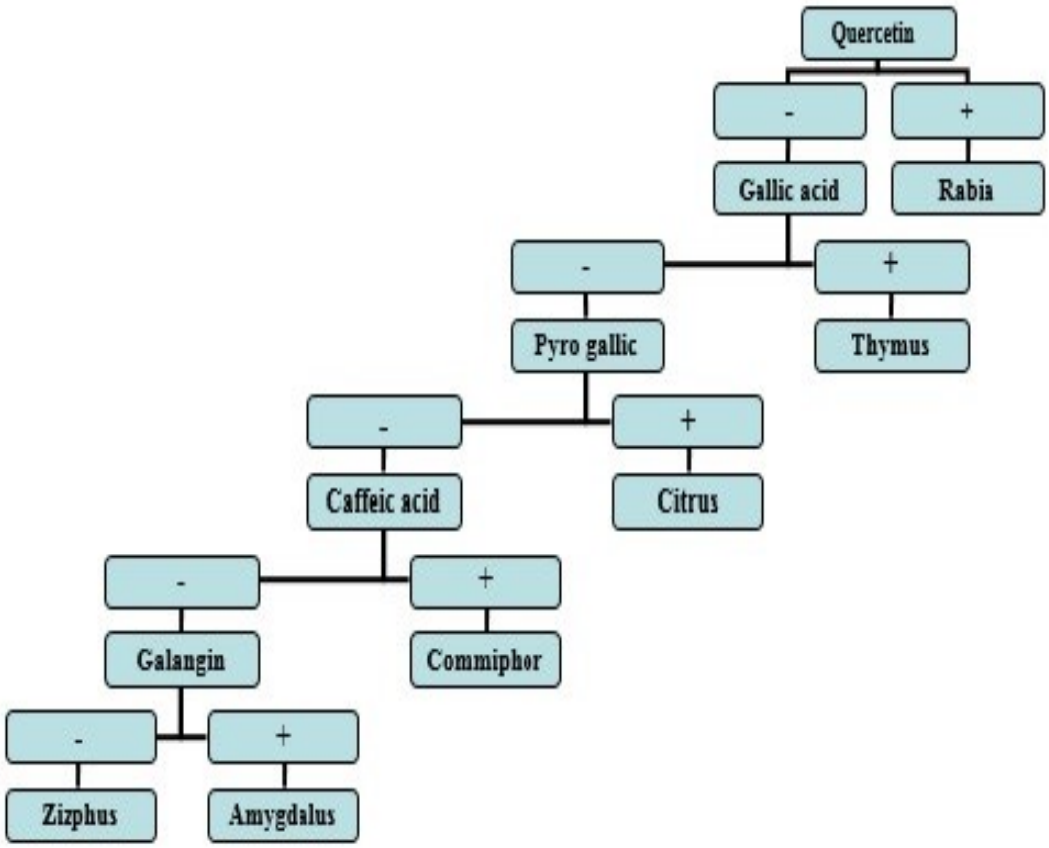
The antimicrobial activity of honey is mainly contributed to the high osmolarity and acidity. In addition, hydrogen peroxide, volatiles, organic acids, flavonoids, phenolic compounds, wax, pollen, propolis are important factors that provide antimicrobial properties to honey. Shin and Ustunol (2005) stated that the sugar composition of honeys from different floral source were responsible for the inhibition of various intestinal bacteria. According to Moumbe *et al.* (2013) the minor components of honey including proteins, minerals, phytochemicals and antioxidants are responsible for the antimicrobial activity of honey in the treatment of infections, burns, wounds and ulcers.

Our results are in agreement with other published studies, showing that some kinds of honey have an inhibitory effect against the fungus *Candida albicans* and the bacteria *Staphylococcus aureus, Bacillus subtilis, Klebsiella pneumoniae, Enterococcus faecalis, Pseudomonas aeuroginosa, Scherichia coli, Bacteriods* spp.*, Sarcina* spp. (Mercan *et al.,* 2007 and Leyva-Jimenez *et al.,* 2019). The results of this study are similar to the results obtained by Mohapatra *et al.* (2011), who reported that honey was effective against gram-positive bacteria *Staphylococcus aureus, Bacillus subtilis, Enterococcus faecalis* and gram-negative bacteria *Escherichia coli* and *Pseudomonas aeruginosa.*The inhibitory effect of honey against *S. aureus*, *E. coli* and *K. pneumonia* is of great importance due to the fact that Streptococcus species and coliforms are recognized pathogens. In this work the growth of *Pseudomonas aeruginosa* was inhibited by 3 honey samples. This type of bacteria is always found in wounds, especially those related to burns causing a variety of systemic infections, particularly in victims with severe burns (Yau *et al.,* 2001). Irish *et al.* (2011) noted that temperature, the time of storage, and the nature of flower's nectar may explain the different antimicrobial activities of different honeys.

Our data are in agreement with the findings obtained by McCarthy (1995), who reported that, honey from different floral sources varies greatly in their antibacterial activity. Rybak and Szczęsna (1996) found that the minimum concentrations of honey which inhibit the growth of *B.subtilis* were 5-10%. Molan and Russell (1988) reported significant differences between different kinds of floral honey in their activities on *S. aureus* at dilutions of 1/4, 1/8 and 1/16 original strength. Radwan *et al.* (1984) reported that honey from *Acacia mellifera* inhibits the growth of *E.coli*. Molan and Russell (1988) found that pollen present in honey could be the source of the antibacterial aromatic acids, which causes the component to act individually or synergically to prevent bacterial resistance (Cooper *et al.*, 2010). In addition to pollen, propolis is also found in honey. The antimicrobial and anti-inflammatory activity of European propolis is associated with the presence of flavonoids, flavones, and phenolic acids and their derivate (Bankova, 2005).

Table (1): Types and floral sources of Libyan honeys.

Nr. Of samples	Local name of honey	Floral source
Sample 1	Sidr	*Ziziyphus louts*
Sample 2	Limon	*Citrus medica*
Sample 3	Zater	*Thymus capitatus*
Sample 4	Lose	*Amygdalus communis*
Samples 5	Morr	*Commiphor myrrah*
Sample 6	Al Rabia	*Multiflora*



Graph (1): Phenolic contents a marker and discriminant of Libyan honeys.

Table (2): The phenolic contents detected in Libyan honeys (µg/100g).

Chemical Name:	Chemical formula	Sidr	Citrus	Zater	Lose	Morr	Al rabia
		µg/100g	µg/100g	µg/100g	µg/100g	µg/100g	µg/100g
Gallic acid	$C_7H_6O_5$	0.00	0.00	18.34	0.00	0.00	0.00
p-Hydroxybenzoic acid	$C_7H_6O_3$	251.30	83.85	154.44	69.07	1248.17	251.70
Caffeic acid	$C_9H_8O_4$	0.00	0.00	0.00	0.00	143.64	0.00
Phenol	C_6H_6O	0.00	3416.60	14737.98	0.00	9037.58	6173.74
p-Coumaric acid	$C_9H_8O_3$	2387.71	1055.94	513.37	0.00	0.00	2068.42
Salicylic acid	$C_7H_6O_3$	0.00	0.00	0.00	0.00	1524.34	0.00
Ferulic acid	$C_{10}H_{10}O_4$	0.00	269.13	2520.43	0.00	0.00	0.00
Cinnamic acid	$C_9H_8O_2$	342.41	0.00	0.00	0.00	350.26	0.00
Quercetin	$C_{15}H_{10}O_7$	0.00	0.00	0.00	0.00	0.00	45.05
Euganol	$C_{10}H_{12}O_2$	0.00	0.00	82.41	0.81	0.00	0.00
Chrysin	$C_{15}H_{10}O_4$	0.00	0.00	0.55	0.00	0.00	0.00
Galangin	$C_{15}H_{10}O_5$	0.00	0.00	0.00	1.99	0.00	0.28
Pinostrobin	$C_{16}H_{14}O_4$	0.00	0.00	0.00	0.00	40.13	0.00
Vanillin	$C_8H_8O_3$	522.23	8.44	0.00	0.00	290.20	0.00
3,5-Dimethoxybenzyl alcohol	$C_9H_{12}O_3$	0.00	0.47	0.00	0.00	0.00	10.53
Catechin	$C_{15}H_{14}O_6$	0.00	428.44	0.00	0.00	0.00	0.00
Daidzin	$C_{21}H_{20}O_9$	2746.43	0.00	0.00	11943.00	2626.99	0.00
Gestin	$C_{15}H_{10}O_5$	205.80	0.00	245.65	0.00	0.00	1293.85
Daidazein	$C_{15}H_{10}O_4$	0.00	1647.53	0.00	0.00	0.00	0.00
Genistein	$C_{15}H_{10}O_5$	0.00	0.00	75.02	0.00	295.61	0.00
Pyro gallic acid	$C_6H_6O_3$	0.00	46.22	0.00	0.00	0.00	0.00
Rutin	$C_{27}H_{30}O_{16}$	0.00	0.00	0.00	0.00	0.00	0.00
Kaempferol	$C_{15}H_{10}O_6$	0.00	0.00	27.50	0.00	17.44	0.00

Table (3): The diameter (in mm) of inhibition zones and standard deviation of different bacterial strains by honey samples compared to control.

Honey samples / Bactria strains	*Zizyphus*	*Citrus*	*Thymus*	*Amygdalus*	*Commiphor*	*Rabia*	*Tetracycline*	*Sucrose*
Escherichia coli	21.0 ± 1.17^{c}	11.31 ± 1.15^{b}	10.66 ± 0.57^{b}	5.33 ± 0.57^{a}	11.33 ± 1.15^{b}	22.33 ± 0.57^{c}	0.00	0.00
Enterococcus faecalis	0.00	0.00	0.00	22.66 ± 0.57^{c}	12.00 ± 1.00^{b}	12.0 ± 0.00^{b}	20.66 ± 1.15^{c}	0.00
Staphylococcus aureus	12.0 ± 0.0^{b}	0.00	5.33 ± 0.57^{a}	0.00	0.00	21.33 ± 1.15^{c}	21.0 ± 1.17^{c}	0.00
Pseudomonas aeruginosa	11.33±1.15b	11.31 ± 1.15^{b}	0.00	11.0 ± 0.00^{b}	0.00	0.00	0.00	0.00
Bacillus subtilis	0.00	0.00	6.33 ± 1.15^{a}	11.33 ± 0.57^{b}	0.00	5.00 ± 0.00^{a}	20.0 ± 0.55^{c}	0.00
Bacteroids spp.	6.00 ± 0.00^{a}	11.55 ± 1.12^{b}	0.00	0.00	0.00	0.00	0.00	0.00
Sarcina spp.	5.82 ± 0.43^{a}	0.00	19.8 ± 1.15^{c}	20.0 ± 0.55^{c}	21.33 ± 1.15^{c}	11.5 ± 0.50^{b}	22.0 ± 0.00^{c}	0.00
Klebsiella pneumoniae	19.8 ± 1.32^{c}	0.00	0.00	0.00	0.00	21.0 ± 0.00^{c}	5.66 ± 0.57^{a}	0.00
Candida albicans	5.66 ± 0.57^{a}	10.14 ± 1.55^{b}	20.66 ± 1.15^{c}	21.33 ± 1.15^{c}	0.00	5.66 ± 1.15^{a}	21.33 ± 1.15^{c}	0.00

Different letters indicate in the row significant difference ($P< 0.01$).

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• Negotiation research spans many disciplines (Abd-Rabou, 1996).

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Journal article

Abd-Rabou, S. (1998): The efficacy of indigenous parasitoids in the biological control of *Siphoninus phillyreae* (Homoptera: Aleyrodidae) on pomegranate in Egypt. Pan-Pacific Entomolgists, 74 (3): 169-173.

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Abd-Rabou, **S. and Simmons, A. M. (2012):** *Bemisia tabaci* (Hemiptera: Aleyrodidae) whitefly as a pest in Egypt. Advances In Agricultral Research In Egypt, 10 (1): 1-82.

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