INTERPRETATION OF SUSCEPTIBILITY PHENOMENON OF SOME MANGO CULTIVARS TO INFESTATION BY MEALYBUG *ICERYA SEYCHELLARUM* (WESTWOD) (HEMIPTERA: MONOPHLEBIDAE)

EL- BADAWY, S. S.

Plant Protection Research Institute, ARC, Dokki, Giza, Egypt

(Manuscript received 17 February 2014)

Abstract

A survey of scale insects and mealybugs on mango trees (*Mangifera indica* L.) (Anacardiaceae) were carried out in a private mango orchards located in El-Saff, Giza Governorate, during two successive years 2011-2012 for studying the role of mango leaf essential oil and their pure major constituents in interpretating the susceptibility phenomenon of five mango cultivar leaves to infestation with the white mealybug, *Icerya seychellarum* (Westwod) (Hemiptera: Monophlebidae).

The results indicated that, the mango trees were infested by 12 species of scale insects and mealybugs belonging to four families: Diaspididae, Coccidae, Monophlebidae and Pseudococcidae. From the monthly collection, the white mealybug, *I. seychellarum* was the most dangerous and abundant of the collected species. The selected five mango cultivars are not equally susceptible to *I. seychellarum* infestation. Fagrikalan mango trees cultivar (cv.) were heavily infested and severely damaged, while trees of Dabsha cv. are approximately complete free from *I. seychellarum* infestation. However, the susceptibility levels of mango cultivars to *I. seychellarum* could be arranged in descending order as follows: Fagrikalan >Alphonso-Naser >Baladi > Hendi >Dabsha. The essential oils of Fagrikalan and Dabsha mango cvs. leaves were determined by hydrodistillation and analyzed by GC-MS. A total of 38 components of the essential oil were identified. The insecticidal or repellent activities of some essential oils of the high susceptible Fagrikalan cv. and the completely tolerable Dabsha cv. against *I. seychellarum* nymphs revealed that, the chemical composition of essential oils plays an important role in this concern. This hypothesis was confirmed by the results of toxicity and repellent activities of the pure major compounds limonene, β-caryophyllene, and α-pinene of Dabsha cv. and the attraction effect of the high quantity of β–ocimene in Fagrikalan cv. against *I. seychellarum* nymphs.

Keywords: *Mangifera indica*, *Icerya*. *seychellarum*, essential oils, susceptibility, repellent and insecticidal activities.

INTRODUCTION

Mango trees, (*Mangifera indica* L.) (Anacardiaceae) are considered of the most popular and economic fruit trees in Egypt. They occupy the third rank after citrus and grapes from the commercial point of view. Mango trees are liable to be infested with many serious pests during their growth stages including the phloem-feeding mealybug, *Icerya seychellarum* (Westwod) (Assem, 1990). The mealybug, *I. seychellarum* infests different parts of mango trees (leaves, branches and fruits) (Dreistadt *et. al.,* 1994). Essential oils from different plant species possess ovicidal, larvicidal and repellent effects against various insect species and were regarded as environmentally compatible pesticides (Isman, 2000). Several studies conducted in different Egyptian localities showed that different mango cultivars express varying levels of tolerance to scale insects and mealybugs infestation in general and *I. seychellarum* in particular. (Monzer *et. al.,* 2006 and Salem *et. al.,* 2007). However, up to date, few studies determine the biological activities such as toxicity and repellency effects of essential oil of mango trees against scale insects and mealybugs. The present study was conducted to investigate the possible role of essential oil in susceptibility of five mango cultivars leaves (Fagrikalan, Baladi, Hendi, Alphonso-Naser and Dabsha) to *I. seychellarum* infestation and evaluate insecticidal and repellent effects of certain essential oils and their pure major compounds against the nymphs of mealybug, *I. seychellarum*.

MATERIALS AND METHODS

Study site:

This study was conducted in a private mango orchard (called Fisher orchards) located at El-Saff, Giza Governorate, Egypt. This 200-feddan orchard contains. 2000 trees of more than 10 mango cultivars, grow next to each other. Most trees were planted in 1935 and were more than 4 m high at the time of the study.

Sampling of survey:

Five local important mango cultivars were chosen (Fagrikalan, Baladi, Hendi, Alphonso-Naser and Dabsha). The cultivars type of the experimental trees were identified by taxonomy experts of Egyptian National Botanical Institute (Dokki, Giza, Egypt).The collection of samples was carried out monthly for two successive years from January 2011 to December 2012. Three representative trees from each cultivar were chosen at random and marked. Selected trees were similar in size, shape, height and vegetation. The marked trees and four trees adjacent to each of them were excluded from any chemical treatment applied to the rest of the orchard. A sample comprising 10 leaves was collected randomly from each of the four cardinal directions (East, West, North and South) of the middle crown parts from each of the three marked trees for each cultivar *i.e.* 120 leaves per sample (3 trees x 4 directions x 10 leaves). Leaves of each tree were packed separately in paper bags and transferred into the laboratory for carefully inspection in the same day.

Identification of insects and population density:

In the laboratory, scale insects and mealybugs were identified in department of scale insects and mealybugs. Population densities of *I. seychellarum* on Fagrikalan, Baladi, Hendi, Alphonso-Naser and Dabsha cultivar leaves were estimated on three trees for each cultivar. Total numbers of a live nymphs and adults on each leaf were counted and their total numbers for each cultivar on each inspection date were calculated.

Chemical analysis:

Leaves of Fagrikalan, and Dabsha cultivars were sampled using the same procedure described above. Leaves were carefully examined and insect damaged, old and infested leaves were removed. Healthy leaves were maintained at –20°C until extraction. The pure compounds (limonene, *α*-pinene, β-caryophyllene, terpinolene and *β*-ocimene) were purchased from Sigma-Aldrich Chemical Co.

Extraction of the essential oil:

The essential oil was extracted from the fresh leaves of Fagrikalan and Dabsha cultivars (500g fresh weight of each sample in 500 ml of distilled water) by hydrodistillation using a Clevenger type apparatus for 4 h. The distilled was extracted with ether after saturation with sodium chloride. The ether extract was dehydrated over anhydrous sodium sulfate. Solvent was removed under reduced pressure at 20 ^{o}C. The volatile constituents was packed in dark container and kept in refrigerator till analysis.

Chemical analysis of essential oil:

Gas chromatographic-mass spectrometric (GC-MS) analysis:

The prepared essential oil was subjected to GC / MS analysis using Shimadzu GC/MS–QP 5050A. Column: DB5, 30 m, 0.53 mm ID, 1.5 µm film. Carrier gas: Helium (flow rate 1.2ml / min.). Ionization mode: (70 ev). The injection volume was 0.5 µL (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^{o}C. The detector temperature was 290^{o}C, while, the injector temperature was 250^{o}C.

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.

Toxicity bioassays

A series of laboratory bioassays were conducted to determine the bioactivity of Dabsha and Fagrikalan leaf oil and their major constituents against *I. seychellarum* nymphs.

1- Spray toxicity assay:

The toxicity bioassay was conducted to evaluate toxicity of Dabsha and Fagrikalan leaf oil and their major constituents to *I. seychellarum* nymphs at three different concentrations (100, 1000, 10000 ppm). In spray toxicity assay, leaves containing nymphs were placed into plastic Petri dishes (10 cm dia×2cm ht). After solvent evaporation (acetone), for each oil leaf and their major constituents, ten infested leaves (each bearing not less than 10 *I. seychellarum* nymphs) of Baladi mango cultivar were sprayed with each oil and their major constituents, then, kept at room temperature. Control insects were sprayed with acetone. Five replicates were used and the experiment was repeated for three times. Mortality of insects was assessed after 48 hrs.

2- Contact toxicity assay (Thin Film Technique):

This technique was used according to Pascual and Robledo. (1997). 1 ml of different concentrations (100,1000,10000 ppm) of Dabsha and Fagrikalan leaf oil and their major constituents in acetone were sprayed from all sides to 9-cm petri dishes and the solvent was allowed to evaporate at 25^{o}C for 24h. For each concentration five replicates of ten mealybug nymphs, in addition to a control (solvent only) were used. Mortality records were taken after 48 hours. Number of dead nymphs were counted and converted to percent mortality for each oil and their major constituents.

Repellency assay (Filter paper test):

The repellency (or) attractiveness of Fagrikalan and Dabsha mango leaves, essential oil leaves and their major compounds against *I. seychellarum* nymphs was determined as described by (Wang *et. al.,* 2009). The crude essential oil and the pure major compounds were diluted in acetone to three concentrations (100, 1000, 10000 ppm). Filter paper (6 cm in diameter) was cut in half and to small pieces. The first half small pieces were placed near the side of Petri dish and the other half pieces were placed on the opposite side of the dish to serve as control. 150µL of each concentration was applied separately to the first half of the filter paper as uniformly as possible with a micropipette. The other half (control) was treated with 150 µL of absolute acetone. Both the treated half and the control half were then air dried to evaporate the solvent completely. *I. seychellarum* nymphs were collected from Baladi mango leaves before the experiment and were starved for 6 hrs. Ten nymphs were placed in the center of the dish, 4 cm away from both the treated half and the control half. Dish was covered and maintained at room temperature. After 24 hours, number of nymphs on the treated half and the control half was counted. Insects within 5 mm of the tested material were counted as being attracted to the source. Five replicates were used and the experiment was repeated for three times. The percent repellency of each Fagrikalan and Dabsha mango essential oil leaves and their major compounds was then calculated according to Thurston *et. al.* (1994) as follows:

$$PR\ (\%) = [(NC - NT)/(NC + NT)] \times 100$$

where: NC was the number of nymphs present in the negative control half, NT was the number of nymphs present in the treated half.

Statistical analysis

The population density of *I. seychellarum* on mango leaves were reduced to tree-specific means and these means were used in statistical analysis. Data of all experiments were evaluated statistically using ANOVA and means compared using Duncan's Multiple Range Test at P<0.05). All statistical analyses were done using the software package Costat. (Costat 2005).

RESULTS AND DISCUSSION

Survey of scale insects and mealybugs

The survey of scale insects and mealybugs infesting mango trees indicated that, mango trees were infested by 12 scale insects and mealybugs species: five species belonging to family Diaspididae, three species to family Coccidae, two species to families Monophlebidae and Pseudococcidae, (table 1). The highest abundant species was *I. seychellarum* (81.93%) of the total collected species, followed by *Lepidosaphes beckii* (Newman) (9.77%), then *Kilifia acuminata* (Signoret) (4.80%) and *Hemiberlesia lataniae* (Signoret) (1.91%). The less abundant species were *Ceroplastes floridensis* (Comstock) (0.51%), *Parlatoria oleae* (Colvee) (0.23%), *Aulacaspis tubercularis* (Newstead) (0.22%), *Aonidiella aurantii* (Maskell) (0.19%), *Planococcus citri* (Risso) (0.11%), *Pulvinaria vitis* (Linnaeus) (0.09%), *Icerya aegyptiaca* (Douglas) (0.10%) and *Planococcus ficus* (Signoret) (0.08 %). According to these results, the population densities of the most serious and abundant species *I. seychellarum* were studied on leaves of five mango cultivars Fagrikalan, Baladi, Hendi, Alphonso-Naser and Dabsha in this assay.

Infestation levels and population densities of *I. seychellarum*:

Population densities of the *I. seychellarum* on leaves of mango cultivars Fagrikalan, Baladi, Hendi, Alphonso-Naser and Dabsha at the sampling date are shown in fig. (1), fig. (2) and table (2). The highest mean number of *I. seychellarum* individuals per 10 leaves was found on Fagrikalan cultivar (546) and (525) in the two successive years 2011 and 2012, respectively, flowed by Alfanso-Naser (213) and (208) then Hendi (184) and (180) and the lastly Baladi (43) and (40). Dabsha leaves were approximately clear from *I. seychellarum* infestation. Accordingly, the order of susceptibility levels of mango cultiva8989rs to *I. seychellarum* could be arranged in descending order as follows: Fagrikalan (highly susceptible) > Alfanso-Naser > Hendi >Baladi>Dabsha (highly tolerable). The calculated infestation rates of *I. seychellarum* were relatively low in winter, spring and summer months, whereas, the high rates were recorded with autumn months in both years. The highest abundance was recorded during October month (24.52 and 32.76 %) and the lowest abundance was recorded during April (2.49 and 0.62 %) in the two successive years, respectively. The overall population densities were not similar among the two studied years, since, the population density of the first year 2011was higher than the second year 2012. This difference in *I. seychellarum* activity is possibly attributed to the variation of the chemical composition (quantitvlly and qualitivlly) of the mango leaves cultivars or weathering conditions. In this study, influences of the weathering conditions and resource availability could not explain the differences in the level of cultivar susceptibility to *I. seychellarum* because trees of all the studied cultivars grow next to each other in Fisher mango orchards. This was supported by Assem (1990) mentioned that neither temperature nor relative humidity showed any significant correlation with population density of *I. seychellarum* on ornamental plants, In contrary, Abd–Elrahman *et. al.,* (2007), showed that, the difference in population density of *I. seychellarum* on mango trees is possibly attributed to the differences in the weathering conditions. El-Said (2006), reported that, the predation of *Rodolia cardinalis* (Muls.) on *I. seychellarum* had no influence in the level of infestation of mango trees.

Differences in susceptibility of various mango cultivars to scale insects had reported by several studies. Salem *et. al.,* (2007) showed that, population densities of the *I. seychellarum* on leaves of Sultani, Baladi, Hendi, Ewaisi and Alphonso mango cultivars were significantly different. They concluded that, Alphonso was completely resistant, while Sultani was highly susceptible to *I. seychellarum* and the order of susceptibility of the studied mango cultivars to *I. seychellarum* could be arranged in descending order as follows: Sultani >Baladi>Hendi>Ewaisi>Alphonso and the levels of susceptibility of mango cvs. for the mealybug, *I seychellarum* depend on the combined action of leaf nutrients, inhibitors, leaf properties and secondary metabolites. On other hand, Monzer *et. al.,* 2006 reported that, leaf nutrients, inhibitors and leaf properties cannot explain the completely resistant of

Alphonso leaves to *I. seychellarum* infestation. They proved that, Alphonso leaves contain certain repellant or toxic secondary metabolites responsible for their resistance to *I. seychellarum* infestation. However, the preference of *I. seychellarum* to Fagrikalan leaves and avoidance of Dabsha leaves can be interpreted by presences of leaf secondary metabolites especially essential oils, which are the most important secondary metabolites. To achieve this assumption, the chemical constituents of essential oils from leaves of Fagrikalan and Dabsha mango cvs. were analyzed and laboratory bioassays were conducted on Fagrikalan and Dabsha cvs. essential oil leaves and its effective compounds against the nymphs of mealybug, *I. seychellarum*. This is the first study that examines the relation between the differences in susceptibility of different mango cultivars to *I. seychellarum* and mango essential oils and its constituents.

Chemical analysis of constituents of oil.

The chemical composition of the essential oils of leaves of Dabsha and Fagrikalan cultivars are presented in table (3). The oil analysis by GC and GC/MS of the essential oil revealed the presence of 38 peaks, approximately all peaks were identified and representing 97.92 and 93.20 % of the essential oil of Dabsha and Fagrikalan cultivars, respectively. The oils of the two cultivars yields were 1.55% and 1.05%, respectively. Monoterpenes and sesquiterpenes hydrocarbons were found to be the most abundant volatiles of the two mango cultivars. This sort of compounds has also been found to be volatile components of many mango cultivars (Pino *et. al.,* 2005). These analyses also revealed that the major identified components in the leaves oil of Dabsha mango cv. were monoterpenes and sesquiterpenes. limonene (15.30%), α-pinene (14.39%), α-terpinolene (7.70%), 3-carene (5.26%), camphene (4.34%) and eugenol (3.45%) were the major monoterpenes and oxygenated monoterpenes, whereas, sesquiterpenes were β-caryophyllene (9.16%), α-gurjunene (5.10%) and humulene epoxide (1.45%). The monoterpene β-ocimene (36.80%) was the principal constituent in the leaves of Fagrikalan cv. whereas, the major identified components of sesquiterpenes and oxygenated sesquiterpenes were α-selinene (7.34%), β-selinene (6.45%), β-elemene (4.50%), α-caryophyllene (1.70%) and α-humulene (1.20%). Our results were confirmed by Pino *et. al.,* (2005), who showed that, the main compound classes of the essential oil of mango leaves were monoterpenes (*e.g*. β-pinene, α-pinene, limonene, α-terpinolene, myrcene, *cis* and *trans*-ocimene) and sesquiterpenes (*e.g.* β-caryophyllene, caryophyllene oxide, 3-carene, α-gurjunene, humulene epoxide, α and β-selinene)**.** In other study, Engel and Tressel, (1983) found that, cultivars from Egypt (Alphonso and Baladi) had limonene as main constituent**.**

Other compounds were also found in relative amounts, (Z)3-Hexenol is the main component (5.40%) in Fagrikalan cultivar and very small amount in Dabsha cultivar (0.10%), the main compounds of hydrocarbons in Dabsha and Fagrikalan mango cultivars were n-Decan (1.20%),(1.39%) and Eicosane (1.45%), (1.34%), respectively, two important fatty acids palmitic acid (1.23%) (1.98%) and oleic acid (1.13%), (1.67%) were also found in both cultivars, respectively. The essential oil of Fagrikalan and Dabsha cultivars are characterized by the unusual presence of benzene derivatives (1,2,3,4-tetrahydro-1-naphthalene,1,2 dicarboxylic benzoic acid and 3-methyl benzoic acid) these compounds were firstly isolated from Fagrikalan and Dabsha cultivars, however, these compounds does not seem to be widely distributed in the *M. indica*. table (3), also shows that there are important and appreciable differences between percentages of the leaf essential oil contents of the two cvs. Leaves of Dabsha cv. were rich with monoterpenes limonene, α-pinene and sesquiterpenes β-caryophyllene and α-gurjunene, while they presented as traces in Fagrikalan cv. Fagrikalan leaves produced mainly β-ocimene, α-selinene and β-selinene as their major constituents. These compounds represented 50.59% of the total Fagrikalan leaf essential oil contents compared with only 1.60% for Dabsha leaves.

Toxicity assays.

The results of toxicity assays (contact and spray toxicity) as represented in the tables (4 and 5), showed that, essential oil of Dabsha achieved significant high mortality percentages against *I. seychellarum* at the different concentrations (100,1000, 10000 ppm). Essential oil of Dabsha and Fagrikalan leaves cvs. exhibited toxicity rate with concentration dependent. The highest toxicity rate of spray and contact assays was recorded for Dabsha mango leaves cv. (93.0%) and (90.4%) at the maximum conc.10000 ppm, respectively, while, the Fagrikalan mango cv. caused (36%) and (14.73%) mortality at the same concentration. These findings were confirmed by Mesbah *et. al.,* (2009) who showed that, the evaluated volatile oils exhibited a high efficiency against the mealybugs *I. seychellarum* and the highest reducing mealybugs was recorded with both camphor and rose volatile oils, followed by Dill, Peppermint and the least efficient was Clove volatile oil.

Limonene exhibited the highest nymphcidal activity against *I. seychellarum* followed by *α*-pinene and *β*-caryophyllene then terpinolene, respectively, while, the lowest value was recorded with *β*-ocimene. All four compounds (limonene, β-caryophyllene, *α*-pinene, and terpinolene) were toxic to *I. seychellarum*. The toxicity assays indicates the order of nymphcidal activities in the essential oils and their compounds as: Dabsha leaves essential oil > limonene > *α*-pinene > *β*-caryophyllene > terpinolene > *β*-ocimene > Fagrikalan leaves essential oil.

Our results in agreement with the results of Monzer *et. al.,* (2006), they found that total extract of Alphonso mango leaves are highly toxic to *I. seychellarum*, while, there were no significant in mortality percentages of *I. seychellarum* nymphs treated with all extracts of Sultani mango leaves and suggested that, *p*-cymene, camphene and limonene play an important role in resistance of Alphonso mango cv. to *I. seychellarum* infestation. Also, Monzer *et. al.,* (2013) suggested that, the combined action of α-pinene, β-pinene and limonene could be responsible of non-preference of *Kilifia acuminate* to leaves of Alphonso mango cv. Robert, (2005) showed that, limonene is an effective natural alternative to mineral oils that can be used to wet and kill wax-covered insects such as mealybugs, scales, and whiteflies. The toxicity of terpinolene was recognized for several stored product insect pests such as *Sitophilus zeamais*, *Tribolium castaneum* (Herbst) (Wang *et. al.,* 2009).

Repellency study:

To our knowledge, no data are available on the repellent activity of Dabsha and Fagrikalan leaves essential oil and its pure major constituents against *I. seychellarum* nymphs. Fagrikalan and Dabsha leaves essential oil and its pure major constituents (limonene, α-pinene, terpinolene and β-caryophyllene from Dabsha leaves essential oil and β-ocimene of Fagrikalan leaves essential oil) were evaluated for their repellent activity against *I. seychellarum* nymphs.

The essential oils from Dabsha leaves and its pure major constituents exhibited good repellent activities against *I. seychellarum* nymphs at the given concentrations table (6). The repellent rate of Dabsha leaves cv. oil was clearly higher than Fagrikalan leaves oil at all concentrations. Data in table (6) also, showed that Dabsha cultivar essential oil possess the highest repellent activity against *I. seychellarum*, while, the lowest repellent activity was recorded with Fagrikalan leaves essential oil.

Our results indicated significant differences in repellence rates of the pure major constituents of Dabsha leaves essential oil against *I. seychellarum*, the highest rate recorded with limonene followed by α-pinene, β-caryophyllene then α-terpinolene, respectively, while, the lowest repellency rates was recorded with β-ocimene (the major constituent of Fagrikalan leaves essential oil) at the given concentrations. These findings confirmed the results of repellency of Dabsha and Fagrikalan leave essential oils. The decreasing order of repellency to nymphs was: Dabsha leaves essential oil > limonene > α-pinene > β-caryophyllene > terpinolene > β-ocimene > Fagrikalan leaves essential oil. These results suggested that, Dabsha cv. oil and its pure major constituents have positive repellent effect on nymph of *I. seychellarum*. Finally, there was a significant attraction of *I. seychellarum* nymphs to Fagrikalan leaves essential oil and its pure major compound and repellence from Dabsha leaves essential oil and their pure major compounds. These results confirmed by Alwala *et. al.,* (2010), they showed that**,** the essential oil of *M. indica* leaves showed a significant dose-dependent repellent effect on host-seeking female *Anopheles gambiae.* Many studies showed that pure major compounds of mango leaves essential oil have repellent activities against various insects of different families. Limonene is found to be repellent against several pest insects (Ibrahim *et. al.,* 2001). Terpinolene has been shown to possess repellent activity against many insects (Benelli1 *et. al.,* 2012). The high concentration of β-ocimene (36.80%) and the presence of (Z) 3-Hexenol (5.40%) in Fagrikalan leaves essential oil may be the reason of the attraction of *I. seychellarum* nymphs to Fagrikalan leaves essential oil. This conclusion was confirmed by Monzer *et. al.,* (2006), they hypothesized that, *β*-ocimene play important role in the high susceptibility of Sultani mango cv. leaves to *I. seychellarum* infestation. Wei *et. al.,* (2007), showed that, plants of pea leafminer, *Liriomyza huidobrensis* can effectively pull wasps, *Opius dissitus,* towards them by releasing a universally induced compound (Z)3-Hexenol and potentially keep these plants safe from parasitic assaults by leafminer pests, *L. huidobrensis.* This confirmed our results and supports our conclusion. The minor components like 3-carene, camphene, *α*-gurjunene and β-pinene may play an important role in repellent and insecticidal activities of Fagrikalan and Dabsha leaves essential oil to *I. seychellarum*. (see Nerio *et. al.,* 2010).

CONCLUSION

Overall results indicated that the repellence and toxic effects of Dabsha leaves essential oil on *I. seychellarum* could be related to the high contents of limonene, *α*-pinene, β-caryophyllene and α-terpinolene. The presence of monoterpene *β*-ocimene with high concentration and (Z)3-Hexenol with slightly high concentration in essential oil of Fagrikalan leaves may be the reason for the high susceptibility of Fagrikalan mango cv. to *I. seychellarum* infestation,

Table 1. The collected scale insects that infest mango trees at Fisher mango orchards, Giza Governorate during 2011 and 2012 seasons.

No.	Insect scientific name	Family	Mean counts		Total	%
			2011	2012		
1	*Icerya seychellarum* (Westwod)	Monophlebidae	40266	29189	69455	81.93
2	*Lepidosaphes beckii* (Newman)	Diaspididae	5236	3048	8284	9.77
3	*Kilifia acuminata* (Signoret)	Coccidae	2305	1765	4070	4.80
4	*Hemiberlesia lataniae* (Signoret)	Diaspididae	927	685	1612	1.91
5	*Ceroplastes floridensis* (Comstock)	Coccidae	268	163	431	0.51
6	*Parlatoria oleae* (Colvee)	Diaspididae	254	-	254	0.23
7	*Aulacaspis tubercularis* (Newstead)	Diaspididae	188	-	188	0.22
8	*Aonidiella aurantii* (Maskell)	Diaspididae	60	98	158	0.19
9	*Planococcus citri* (Risso)	Pseudococcidae	48	46	94	0.11
10	*Icerya aegyptiaca* (Douglas)	Monophlebidae	89	-	89	0.10
11	*Pulvinaria vitis* (Linnaeus)	Coccidae	71	-	71	0.09
12	*Planococcus ficus* (Signoret)	Pseudococcidae	-	64	64	0.08
		Total	49712	35058	84770	100

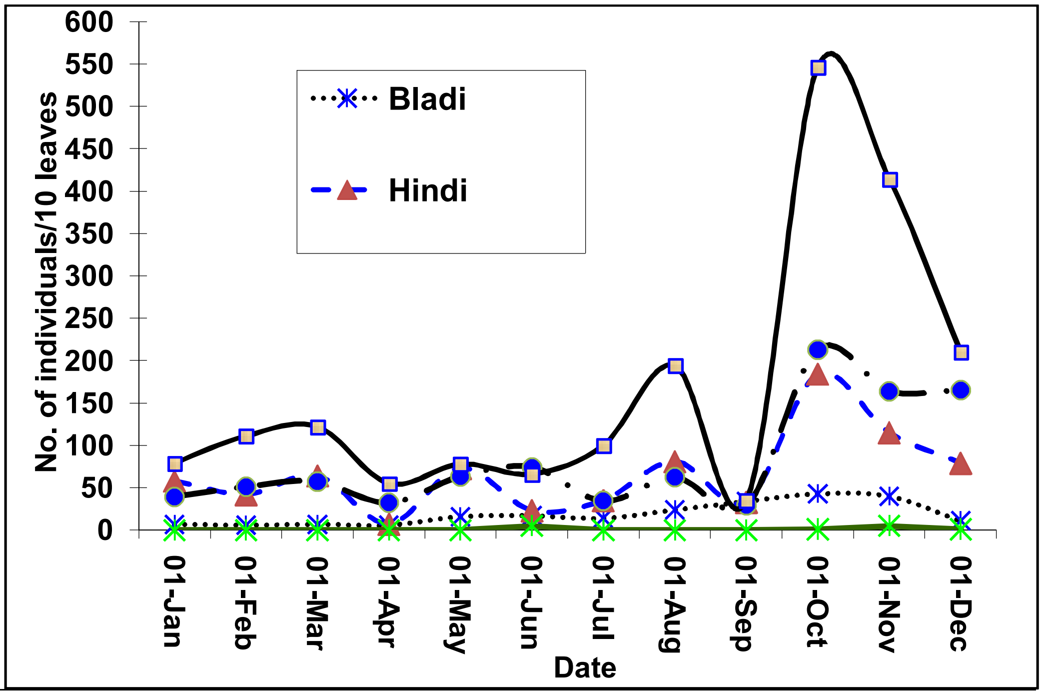

Fig. 1. Monthly mean counts of *I. Seychellarum* on five mango cultivars at Fisher mango orchards, Giza Governorate during season 2011.

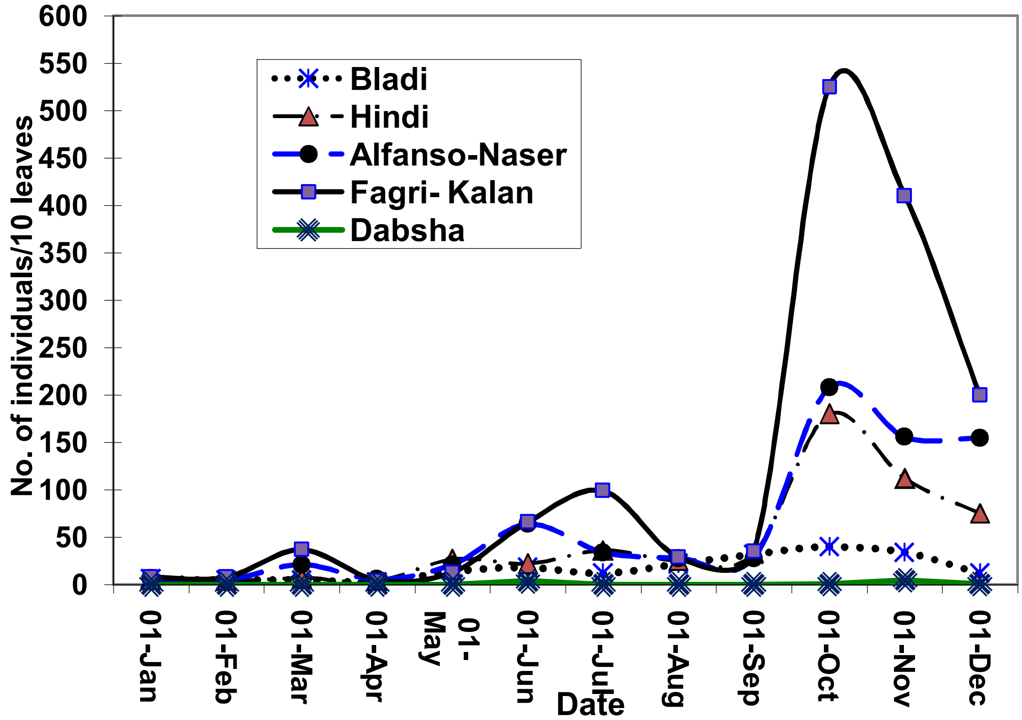

Fig. 2. Monthly mean counts of I. Seychellarum on five mango cultivars at Fisher mango orchards, Giza Governorate during season 2012.

Table 2 . Monthly mean counts of *I. Seychellarum* on five mango cultivars at Fisher mango orch Governorate during 2011 and 2012 seasons.

Date of inspection	Mean number of individuals/10 leaves											
	Fagri-Kalan		Alfanso-Naser		Hindi		Baladi		Dabsha		Total	
	2011	2012	2011	2012	2011	2012	2011	2012	2011	2012	2011	2012
Jan	79	8	39	7	58	5	7	6	0	0	183	26
Feb	111	8	51	5	42	4	6	5	0	0	210	22
Mar	122	37	57	21	64	7	7	5	0	0	250	70
Apr	55	5	32	6	7	4	6	3	0	0	100	18
May	78	13	63	21	73	27	16	14	0	0	230	75
Jun	66	66	74	64	23	22	17	18	5	4	185	174
Jul	100	99	34	34	35	36	14	12	0	0	183	181
Aug	194	29	62	28	81	25	24	22	0	0	361	104
Sep	35	35	29	28	33	33	34	32	0	0	131	128
Oct	546	525	213	208	184	180	43	40	1	1	987	954
Nov	414	410	164	156	115	112	40	34	5	5	738	717
Dec	210	200	165	155	79	75	11	12	1	1	466	443
Total	2010	1435	983	733	794	530	225	203	12	11	4024	2912
Avarage	167	120	82	61	66	44	19	17	1	1	335	243

Table 3. Chemical composition of the volatile oil from leaves of *M. indica* .

No.	Components	Rt (min)	Ratio (%)	
			(L_1)	(L2)
	Monoterpene hydrocarbons			
1-	α-pinene	7.13	0.20	14.39
2-	camphene	7.30	1.68	4.34
3-	β-pinene	7.47	1.56	3.19
4-	β-myrcene	8.23	3.10	1.20
5-	3 –carene	9.14	0.60	5.26
6-	β -ocimene	10.06	36.80	1.15
7-	p-Cymene	10.56	2.10	2.34
8-	Limonene	10.85	0.10	15.30
9-	α -Terpinolene	11.45	0.66	7.70
			46.70	54.87
	Oxygenated monoterpene			
10-	Eugenol	20.14	1.10	3.45
11-	Geranyl acetone	21.12	1.27	1.34
12-	Eugenyl acetate	25.95	ND	0.56
			2.37	5.35
	Sequiterpens hydrocarbons			
13-	β-Elemene	22.07	4.50	0.25
14-	β-Caryophyllene	22.53	ND	9.16
15-	α-Gurjunene	22.75	ND	5.10
16-	α -Caryophyllene	22.85	1.70	0.59
17-	α-Humulene	23.18	1.20	0.14
18-	β-chamigrene	23.75	0.20	0.30
19-	α-Selinene	23.17	7.34	0.35
20-	β -Selinene	23.24	6.45	0.10
21-	α--calacorene	23.93	0.40	0.50
			21.79	16.49
	Oxygenated sesquiterpenes			
22-	E-Nerolidol	25.12	0.70	0.30
23-	Spathullenol	25.65	0.70	1.20
24-	β-Caryophyllene oxide	26.33	1.29	0.52
25-	Viridiflorol	26.78	ND	0.70
26-	α-Humulene epioxid	27.10	1.10	1.45
27-	Cubenol	28.35	1.65	0.36
28-	α-Cadinol	28.49	1.78	0.40
			7.22	4.93
	Other compounds			
29-	(Z)3- Hexenol	3.56	5.40	0.10
30-	n Decan	10.84	1.20	1.39
31	Palmitic acid	31.44	1.23	1.98
32-	Oleic acid	32.21	1.13	1.67
33-	Eicosane	33.17	1.45	1.34
34-	Tetracosane	33.45	0.56	0.67
35-	1-methyl-7ethyl cyclopentane	35.97	1.82	1.20
36-	1,2,3,4 tetrahydro-1-naphthalene	38.05	0.67	1.24
37-	3- methyl benzoic acid	39.21	0.56	2.13
38-	1,2 dicarboxylic benzoic acid	40.53	1.10	4.56
			15.12	16.28
	Total identified		93.20	97.92

Rt : Retention time L1= Fagrikelan leaves L2= Dabsha leaves
ND: not detected Tr. : Trace

Table 4. Toxic effect (Spray assay) of Dabsha and Fagrikalan mango cvs. leaves essential oils and their major compounds against *I. seychellarum* nymph at different concentrations.

Mango cv. and compounds	Dabsha mango cv.	Fagrikalan mango cv.	Limonene	α-Pinene	β-Caryophyllene	α-Terpinolene	β–Ocimene
Conc. (ppm)	Corrected mortality (%)						
100	٥٥.0±0.78^{c}	13.0±0.47^{c}	74.0±0.29^{c}	50.0±1.76^{c}	53.0±0.89^{c}	69.9±0.95^{b}	29.0±0.35^{c}
1000	77.0±1.90^{b}	22.0±0.21^{b}	85.0±1.42^{b}	63.0± 0.65^{b}	81.0±0.35^{b}	81.1±0.15^{a}	40.0±0.53^{b}
10,000	93.0±0.51^{a}	36.0±0.55^{a}	91.0±0.66^{a}	89.0±0.50^{a}	84.0±0.38^{a}	83.2±0.92^{a}	43.0±0.60^{a}
Control	0.0	0.0	0.0	0.0	0.0	0.0	0.0
F value	336.32***	162.05***	116.25***	564.2***	444.50***	168.51***	95.52***
LSD	3.767	2.902	2.978	2.978	2.825	2.153	3.193

Values (means ± SD) followed by similar letter within the same column do not differ significantly (P< 0.05)

Table 5. Toxic effect (Contact assay) of Dabsha and Fagrikalan mango cvs. leaves essential oils and their major compounds against *I. seychellarum* nymph at different concentrations.

Mango cv. and compounds	Dabsha mango cv.	Fagrikalan mango cv.	Limonene	α-Pinene	β-Caryophyllene	α-Terpinolene	β–Ocimene
Conc. (ppm)	Corrected mortality (%)						
100	71.8±0.64^{c}	11.80±0.46^{a}	71.22±1.5^{c}	65.220.39^{c}	٧٠.٠٠±054^{c}	٦٦.٧±0.32^{b}	23.7±0.36^{c}
1000	83.6±0.25^{b}	12.91±0.26^{a}	82.3±1.10^{b}	71.30±0.15^{b}	٨١.٢٠±0.60^{b}	٧٥.٢±0.89^{b}	34.8±0.31^{b}
10,000	90.4±0.45^{a}	14.73±0.45^{a}	89.30±0.45^{a}	٨٥.٢٠±0.45^{a}	٨٣.٢٠±0.45^{a}	٨١.٣±0.45^{a}	39.0±0.45^{a}
Control	0.0	0.0	0.0	0.0	0.0	0.0	0.0
F value	128.72***	1.72^{ns}	69.13***	146.98***	126.01***	22.37**	163.95***
LSD	2.884	1.992	3.265	2.862	2.205	7.990	2.210

Values (means ± SD) followed by similar letter within the same column do not differ significantly (P< 0.05)

Table 6. Repellency (%) of Dabsha and Fagrikalan mango cvs. leaves essential oils and their major compounds against *I. seychellarum* at different concentrations.

Mango cv. and compounds	Dabsha mango cv.	Fagrikalan mango cv.	Limonene	α-Pinene	β-Caryophyllene	α-Terpinolene	β–Ocimene
Conc. (ppm)	Repellency (%)						
100	97.1±0.40^{b}	13.7±0.35^{b}	٨٨.٢±0.10^{b}	٨٢.٢±0.11^{b}	٨١.٠±0.15^{b}	٧٥.١٠±0.60^{b}	13.80±0.11^{b}
1000	98.0±0.58^{ab}	14.8±0.25^{b}	٨٩.٣٠±0.26^{b}	٨٨.٣٠±0.10^{b}	٨٣.٣٠±1.0^{b}	٨٠.٤٥±0.16^{a}	14.91±0.06^{b}
10,000	99.4±0.62^{a}	23.0± 0.20^{a}	٩٤.٣٠±0.50^{a}	٩١.٣٠±0.10^{a}	٨٧.٣٠±0.21^{a}	٨٢.٤±0.25^{a}	١٦.٧٣±0.12^{a}
Control	0.0	0.0	0.0	0.0	0.0	0.0	0.0
F value	5.27*	173.47***	8.74	12.07**	11.04**	20.24**	8.10*
LSD	1.93	1.226	3.240	3.908	8.571	2.580	1.485

Values (means ± SD) followed by similar letter within the same column do not differ significantly (P< 0.05)

REFERENCES

1. Abd-Elrahman, M.M., Salem, M. S., Moussa, S. M., Nour, M. E. and EL-said, M. I. 2007. Seasonal fluctuation of the seychelles fluted scale, *Icerya seychellarum* (Westwood) on four mango cultivars in Egypt. *Egypt. J. Agnc. Res.,* 85 (1) : 77-88.
2. Adams, R.P. 2007. Identification of essential oil components by gas chromatography/ quadrupole mass spectroscopy; Allured: Carol Stream, IL, USA.
3. Alwala. J. O., Wanzala. W., Inyambukho, R. A., Osundwa. E. M. and Ndiege. O. 2010. Characterization and evaluation of repellent effect of essential oil of *Mangifera indica* L. on *anopheles gambiae*, from Kenya. *Journal of Essential Oil Bearing Plants*. 13(1): 85-96.
4. Assem, M.S. 1990. Survey and biological studies on some insects attacking certain ornamental plants. Ph.D. Thesis, Fac. Agric., Cairo Univ., Cairo, Egypt, 202p.
5. Benelli1, G., Flamini, G., Canale, A., Molfetta, I., Cioni, P. L. and Barbara. C. 2012. Repellence of *Hyptis suaveolens* whole essential oil and major constituents against adults of the granary weevil *Sitophilus granaries*. *Bulletin of Insectology*, 65 (2):177-183.
6. Costat. 2005. Version 6.311, Copyright(c), CoHort Software, 798 Lighthouse Ave. PMB 320, Monterey, CA, 93940, USA.
7. Dreistadt, S. H., Clark, J. K. and Flint, M. L. 1994. Pests of landscape trees and shrubs: An integrated pest management guide. Oakland: Univ. Calif. Div. Agric. Nat. Res., 3359.
8. El- Said. 2006. Studies on some Eco-physiological factors affecting resistance of five mango cultivars to the margarodid mealybug *Icerya seychellarum* (Westwood), *Ph.D. Thesis, Department of Economic Entomolgy and Pesticides*, *Fac. Agric., Cairo Univ.*120p. (50-55).
9. Engel, K. H. and Tressel, R. 1983. Studies on the volatile components of two mango varieties. *J. Agr. Food Chem*. 31:796–801.
10. Ibrahim, M. A., Kainulainen, P., Aflatuni, A., Tilikkala, K. and Holopainen, J. K. 2001. Insecticidal, repellent, antimicrobial activity and phytotoxicity of essential oils: With special reference to limonene and its suitability for control of insect pests. *Agric. Food Sci*. Finland, 10: 243-259.
11. Isman, M B. 2000. Plant essential oils for pest and disease management. *Crop Protection* 19 (8-10): 603-608.
12. Mesbah, H. A., Magda, Ael-K.., Mourad, A. K., Abdel-Razak, S. I., Weheda, M. L., Zaghloul, O. A. and Samar. E. A. 2009. Efficacy of five volatile oils and their

mixtures against the mealybug, *Icerya seychellarum* (Westw.) infesting Sago palm, Cycas revoluta in Alexandria, Egypt. *Commun Agric. Appl. Biol. Sci.*, 74(2):445-455.

13. Monzer, M. A., Salem, M. S., El-said M. I. and Melegi, A. 2006. Resistance of Alphonso mango-cultivar to the margarodid mealybug, *Icerya seychellarum* (Westwood) in relation to leaf quality: I. Leaf secondary metabolites. Egypt *J. Agric. Res.*, 84 (1): 17-29.
14. Monzer, M. A., Srour, H. A. and Melegi, A. 2013. Isolation and identification of volatile substances in Alphonso mango leaves repellent to *Kilifia acuminata* (Signoret) (Hemiptera: Coccidae). *New York Science Journal*, 6 (12):84-91.
15. Nerio L. S., Olivero-Verbel J. and Stashenko E. 2010. Repellent activity of essential oils: a review.- *Bioresource and Technology*, 101: 372-378.
16. Pascual, J. M. and Robledo, A. 1997. Screening for anti-insect activity in Mediterranean plants. *Industrial Crops Products,* 8:183.
17. Pino, J. A., Mesa, J., Muoz, Y., Mart, M. P. and Marbot, R. 2005. Volatile components from mango (*Mangifera indica* L.) cultivars. *J. Agric. Food Chem*. 53: 2213-2223.
18. Robert, G. H. 2005. Limonene, a citrus extract, for control of mealybugs and scale insects. *J. Econ. Entomol.*, 98(3):772-779.
19. Salem, M. I., El-Said, M. I., Melegi, A. and Monzer, M. A. 2007. Susceptibility of five mango cultivars to Seychelles fluted scale, *Icerya seychellarum* (West.) in relation to leaf quality: Leaf toughness and anatomical characteristics. Egypt*. 1, Agric. Res.,* 85 (1):89-100.
20. Thurston, G.S., Yule, W.N. and Dunphy, G.B. 1994. Explination for the low susceptibility of *Leptinotarsa decem/ineata* to *Steinernema carpocapsae.* Biological Control 4: 53-58.
21. Wang, J. L., Li, Y. and Lei. C. L. 2009. Evaluation of monoterpenes for the control of *Tribolium castaneum* (Herbst) and *Sitophilus zeamais* Motschulsky- *Natural Products Research*, 23:1080-1088.
22. Wei, J., Wang, L., Zhu, J., Zhang, S. and Nandi, O. I. 2007. Plants attract parasitic wasps to defend themselves against insect pests by releasing (Z) 3-Hexenol. *PLoS ONE* 2(9): e852. doi:10.1371/*journal.pone*.0000852.

تفسير ظاهرة قابلية إصابة بعض أصناف المانجو

بالبق الدقيقي *Icerya seychellarum*

سامي سيد البدوي

معهد بحوث وقاية النباتات – مركز البحوث الزراعية – الدقى جيزة.

يهدف هذا البحث إلي إجراء حصر للحشرات القشرية والبق الدقيقي في مزارع المانجو بمنطقة الصف–محافظة الجيزة في الفترة من يناير ٢٠١١ حتي ديسمبر ٢٠١٢ ، ودراسة مدي قابلية خمس أصناف من أوراق أشجار المانجو للإصابة بالبق الدقيقي *Icerya seychellarum* وكذلك دور الزيوت العطرية المستخلصة من أوراقها ومكوناتها الفعالة في تفسير ذلك من خلال دراسة التأثير السمى والطارد لهذه الزيوت ومكوناتها الفعالة ضد البق الدقيقي *I. seychellarum*. وقد أظهرت النتائج أن ١٢ نوع من الحشرات القشرية والبق الدقيقي والتي تنتمي إلي أربع عائلات مختلفة قد أصابت أوراق المانجو، وأن البق الدقيقي *I. seychellarum* كان الأعلي كثافة في التعداد والأكثر خطورة علي أوراق أشجار المانجو وأن معدل التعداد السنوي لحشرة *I. seychellarum* اختلفت بشكل ملحوظ في قابلية الأصناف الخمسة للإصابة، فوجد أن صنف الفجرى كلان هو اعلي الأصناف إصابة ثم يليه صنف ألفونسو ناصر ثم الهندي وأقلهم إصابة البلدي بينما خلى صنف الدبشة تقريبا من الاصابة. تم فصل والتعرف علي ٣٨ مركب من الزيوت المستخلصة من أوراق الصنف الفجرى كلان (الأكثر إصابة) والدبشة (الخالي تقريبا من الاصابة) بجهاز الكروماتوجرافي الغازي– الكتلي، وأوضحت النتائج أيضا أن الزيوت العطرية ومركباتها الفعالة لها دور كبير في تباين قابلية أصناف المانجو للإصابة بالبق الدقيقي *I. seychellarum*، وتأكد ذلك الإفتراض بنتائج التأثير السمي والطارد للمركبات الفعالة (limonene, α-pinene and β-caryophyllene) في صنف الدبشة والتأثير الجاذب للمركب الأكثر توجداً في صنف الفجري كلان(β–ocimene) ضد حوريات *I. seychellarum*.