

## EFFECT OF COLONY MANAGEMENT AND PLANT EXTRACTS ON VARROA MITES ATTACKING HONEY BEE COLONIES

[41]

Elbassiouny<sup>1</sup>, A.M.; M.I. Abdel-Megeed<sup>1</sup>; M.O. EL-Shaarawi<sup>2</sup> and  
Gehan M. Mohammed<sup>2</sup>

1- Plant Protec., Dept. Fac. of Agric., Ain Shams Univ., Shoubra EL-Kheima, Cairo, Egypt

2- Honey Bee Res. Dept., Plant Protec. Instit., Dokki, Cairo, Egypt

**Keywords:** Honey bee, Varroa, Thymol, Santonica, Apistan, Mavrik

in beeswax 2 weeks, one month and two months after treatments, respectively.

### ABSTRACT

*Varroa jacobsoni* mite has become a major problem to Egyptian beekeeping. Nowadays, the pest management in infested honey bee colonies and alternative tools by using some natural products extracted from medicinal and ornamental plants are considered more save to human being and bees. Thymol and mixture of Thymol and Santonica as Natural compounds and Apistan and Mavrik as Traditional acaricides were evaluated against *V. jacobsoni*. The data clearly showed the superiority of Thymol and Santonica mixture for controlling *Varroa* mites at moderate or low infestation levels, and Apistan at sever infestation. Thymol and Santonica mixture caused an obvious reduction of the rate of mite infestation either in sealed brood cells or on adult bees, an obvious increase in the number of *Varroa* fallen on the floor of treated hive, apparent increase in the number sealed worker brood cells and, lastly, an increase in the number of combs covered from both sides with adult worker bees. Moreover, treating the colonies with compressed bees (reducing hive space) was more efficient than that of colonies received traditional beekeeping. Chemical analysis showed that fluvalinate residues (the active ingredient of Apistan and Mavrik) were 1.2, 1.5 and 1.8 ppm in honeybee workers, 0.6, 1.4 and 1.9 ppm in bee honey and 0.48, 0.96 and 1.3 ppm

### INTRODUCTION

Since the discovery of the ectoparasite mite *varroa jacobsoni* (Oudemans) in Egypt in 1987, it has become a major problem to Egyptian beekeeping. Damage to individual bees that emerge from infested brood cells reduced flight frequency of resulted drones and loss in weight of workers by 6-25% depending on the degree of infestation level (De Jong *et al* 1982). At high levels of *Varroa* infestation, there is a rapid decline in the number of adult bees, severe damage to the brood and death of the colony usually occur, (Elbassiouny *et al* 2004). Also at high infestation level, bees have reduced their life span (Elbassiouny, 1998). The mites also feed on adults between reproductive periods in brood. Their feeding leads to the possible spread of viral pathogens (Sammataro, 1997) and bacteria (Glinski and Jarose, 1992). These various effects of feeding lead to a complex of symptoms due to a parasitic mite syndrome (Shimanuki *et al* 1994), which proved as a sudden loss in number of worker bees in a colony and subsequent death of the colony.

Several years ago, the common acaricide registered to control varroa was Apistan. This single and repeated treatment strategy induced Apistan resistance in *V. jacobsoni* at many countries (Colin *et al* 1997; Baxter *et al* 1998 and Pettis *et al* 1998 Elbassiouny, 2003). In addition, fluvali-

(Received October 14, 2006)

(Accepted November 1, 2006)

nate use risks contamination hive products (Sla-bezki *et al* 1991 and Wallner, 1995). Therefore, trials were made to seek alternative control strategies based on natural products in integrated pest management such as plant oils and essential oils components in laboratory and field trials as control agents for the *Varroa* mite. These materials are relatively inexpensive, and consumer perceive them as natural compounds that will not contaminate hive products (Colin, 1990; Imdorf *et al* 1995; Calderone *et al* 1997 and Sammataro *et al* 1998).

The present study aimed to through some light on the relation between acaricides and the population density of *varroa* mite in infested honeybee colonies, as well as, to find out alternative tools for controlling *Varroa* mite with certain natural products extracted from medicinal and ornamental plants.

## MATERIALS AND METHODS

The present work was carried out during 2003 / 2004 season in the apiary belonging to Ministry of Agriculture, in Qaulub, Qalubia governorate, Egypt.

On December 20, 2003, forty colonies headed with natural mated local *carnica* queens (*Apis mellifera carnica*) were divided according to the infestation level into two main groups (20 colonies each). The first group (received normal beekeeping) was treated with natural compounds (4 colonies for each of thymol and mixture of thymol and santonica); chemical compounds (4 colonies) for each of Apistan and Mavrik and the last 4 colonies were left untreated (control). In the second group, the colonies were applied with the same application but the surplus combs were removed, therefore bees were compressed on the lower number of combs to reduce the colony space. Moreover, good wintering by covering the combs with gunny and plastic sheet, was done

### Colony vigor

The measures considered in this study were, square centimeters of sealed worker brood area and number of frames covered from both sides with adult bees as colony strength. The measures of colony strength were determined every week through the experimental period.

### *Varroa* mite infestation level

*Varroa* mite infestation levels were estimated using approximately 50 adult worker bees collected from each colony and placed into jars containing warm water with a few drops of liquid soap. They were then transferred to the laboratory. The jars were shaken vigorously and the contents strained through 8 per cm screen. The screen retained the bees but let varroa individuals pass where they were collected on a fin nylon cloth located below the screen. The process was repeated until no more mites were collected. Mites were then counted and the infestation level was estimated by dividing the number of varroa mites recovered by number of bees.

### Mites recovered on bottom boards (*Varroa* fallen)

A cardboard painted with thin layer of Vaseline was inserted in each colony on the bottom board and covered with 2.5-3 mm wire mesh. This application may catch the alive varroa. The wire mesh retained the bees but let the fallen mites pass where it collected on the cardboard. Counts of fallen mites were carried out before and after the treatment. Cardboards were removed every week and mites were counted.

### Varroacides used

#### Chemical compounds

The efficiency of the following two acaricides belonging to one chemical group was evaluated.

- **Apistan** ® strips 10% (fluvalinate) pyrethroid group.

- **Mavrik** 10% Ec (fluvalinate) pyrethroid group used at concentration of 5cm/liter- Water.

#### Chemical analysis of Fluvalinate:

Pesticide used: Fluvalinate (Trade name Apistan & Mavrik)

#### Chemical name (IUPAC):

(*RS*) - $\alpha$ -yano - 3- phenoxy bezyl *N*- (2- chloro -  $\alpha$ - $\alpha$ - $\alpha$ - trifluoro - *p* - tolyl) - *D*- valinate.

#### Natural compounds

- Plant extracts from **Thymol**: (a crystalline phenol occurring naturally in thyme (*Thymus vulgaris*) packets of 2g thymol powder was placed inside the experimental colonies on the top bars

of the brood combs. The packets were replaced with a new one every 3 weeks.

- Plant extracts of *Santonica* (especially flowers) was prepared by boiling 500g in an equal volume of water, then an amount of 50 ml from the extract was added to the sugar syrup offered to the tested colonies. The plant extract was applied at four day interval.

All experimental colonies received sugar syrup (50%) two times weekly, immediately before the beginning of the application and every week after the application.

#### Extraction of bee honey and honey bees

Extraction of samples was carried out according to the official method of analysis (AOAC, 1990), as follows: 5 grams of bee honey or honey bees were extracted with 50 ml acetonitril and blended for 2 min, then filtrated through filler paper (whatman No.1) and transported into conical flask (500 ml in volume) to portioning with 50 ml petroleum ether three times. The filtrate (petroleum phase) was taken in order to clean up.

#### Extraction of bees wax

Extraction of samples was carried out according to the official method of analysis (AOAC, 1990), as follows: 5 grams of bee wax were extracted with 50 ml benzolen ether and blended for 2 min, then filtrated through filler paper (whatman. No. 1) and transported into conical flask 500 ml, to portioning with (50 ml) petroleum ether three times. The filtrate (petroleum phase) was taken in order to clean up.

#### Clean up

Clean up of samples were carried out according to the official method of analysis (AOAC, 1990).

#### Determination

The operating conditions for gas chromatography shimadzu (12-A) equipped with FID detector were as follows

- a- Column glass 3 mm x 1.7 m 2% Deil on sumi-kasorb HP 80/100 mesh.
- b- Typical operating conditions
 

Inj/Det. Temperature	250°C
Air pressure:	1 kg/cm <sup>2</sup>
Carrier Gas (N <sub>2</sub> )=	1 kg/m <sup>2</sup>

Hart speed:	2.5 mm/min
Oven temperature	220-250°C
Burner Gas (H <sub>2</sub> ) =	1 kg/cm <sup>2</sup>
Attenuation =	10 x 5
Injetion volume =	3 µl

### RESULTS AND DISCUSSION

In this work, comparative studies were made between experimental honeybee colonies received traditional beekeeping and the compressed bees, both were treated with natural compounds (Thymol and mixture of Thymol and *Santonica*) and traditional acaricides (Apistan and Mavrik) for 10 weeks.

#### 1- Worker brood infestation with *V. Jacobsoni*

As shown in Table (1), the rate of honeybee sealed worker brood infestation with *Varroa* mite in the experimental colonies just before treatments ranged between 11.5 -18.0% in colonies received normal beekeeping and (14.0 -16.5%) in colonies with compressed bees. Treating these colonies on Dec.,20 and every four days with different compounds gradually decreased the rate of sealed worker brood infestation with this mite to reach the lowest infestation rates 10 weeks after treatments on February 24.

The reduction percentage in the infestation rate in colonies received traditional beekeeping reached 62.0% after using Apistan followed by Thymol and *Santonica* mixture (54.10%), Thymol alone (45.0%) and the least reduction (20.8%) was, however, obtained after using Mavrik. The highest reduction percentage could be applied for experimental colonies with compressed bees. In this case, the reduction percentage was 71.60% after using Apistan, followed by Thymol and *Santonica* mixture (59.2%) and Thymol alone (51.3%). Mavrik gave the least reduction in infestation rate (43.7%).

#### 2- Adult infestation with *V. Jacobsoni*

The rate of honeybee adult infestation with *Varroa* mite ranged between 14.6 - 20.5% for traditional beekeeping and 13.2 - 20.2% for compressed bees just before treatments, Table (2). Ten weeks after treatments and in case of traditional beekeeping, the minimum infestation rate was recorded after using the recommended acaricide Apistan (4.9%), followed by the mixture of Thymol and *Santonica* (6.4%), and Thymol only

Table 1. Percentages of infestation with *Varroa* mite in honeybee sealed worker brood after treating the colonies with different compounds in both traditional beekeeping and compressed bees during 2003/2004 season

Inspection date	Traditional beekeeping					Compressed bees				
	Natural compound		Traditional acaricides			Natural compound		Traditional acaricides		
	Thymol	Thymol & Santonica	Apistan	Mavrik	Untreated check	Thymol	Thymol & Santonica	Apistan	Mavrik	Untreated check
Before treat.										
20/12/2003	12.5±7.2	11.5±4.7	18.0±5.2	17.5±7.2	15.9±4.8	15.5±7.0	15.0±4.8	14±5.9	16.5±4.4	16.3±5.0
After treat.										
28/12	10.8±5.5 (12.7)	10.3±4.2 (13.7)	12.0±4.2 (36)	11.0±8.1 (33.2)	16.4±4.8	13.2±5.9 (9.9)	13±4.6 (7.8)	11.7±5.0 (12)	17.9±4.2 (12.7)	15.3±5.7
3/1/2004	9.2±5.3 (10.7)	9.3±3.1 (7.2)	11.6±4.2 (25.8)	15.2±6.3 (7.5)	13.7±3.8	12.5±5.5 (0)	12.1±4.8 (0)	9.8±4.3 (15)	14.0±3.6 (1.4)	13.0±5.0
10/1	7.8±3.5 (29.1)	8.8±3.3 (17.2)	9.2±3.3 (44.4)	13.9±5.0 (3.8)	14.5±3.0	11.3±5.7 (5.1)	10.9±3.9 (5.1)	6.5±1.9 (33.8)	11.5±2.5 (23.7)	11.3±1.3
16/1	7.3±3.5 (33.4)	7.9±4.1 (23.4)	8.1±2.4 (50)	12.2±3.9 (1.4)	14.3±4.5	9.0±4.1 (14.7)	9.6±3.3 (5.9)	4.4±1.4 (54.4)	12.8±3.5 (14.5)	11.1±1.4
23/1	6.7±2.7 (29.9)	6.9±1.7 (26.6)	7.2±3.3 (50.8)	10.2±3.1 (8.9)	12.9±4.4	7.5±3.1 (24.2)	7.3±2.4 (23)	2.9±1.3 (67)	10.1±3.4 (25)	10.4±5.4
29/1	6.5±2.6 (26.6)	5.8±1.3 (30.2)	6.4±2.6 (52.4)	8.2±2.6 (24)	11.7±3.5	7.0±2.3 (27.5)	6.6±2.4 (29.2)	2.1±0.3 (72.3)	8.2±1.4 (32)	10.1±5.9
5/2	5.8±2.5 (49.6)	5.2±1.3 (52.4)	6.0±2.3 (65.3)	7.2±2.6 (29)	15.1±6.0	5.8±1.7 (35.9)	4.7±2.4 (46.4)	2.3±0.5 (72.3)	7.0±0.6 (55.9)	9.4±1.3
11/2	5.2±1.4 (44.5)	4.7±1.4 (48.4)	5.9±2.2 (57.4)	7.7±2.7 (22.1)	12.3±3.8	5.5±2.2 (38)	4.1±1.5 (52.2)	2.2±0.7 (71.7)	6.6±1.1 (48.4)	9.2±4.5
18/2	5.2±1.4 (41.9)	4.1±1.1 (51.4)	5.4±2.4 (59.5)	7.2±3.2 (25)	11.8±2.9	4.3±1.7 (48.8)	3.1±1.1 (61.6)	2.0±1.0 (74.4)	7.1±1.3 (41.9)	8.9±2.6
24/2	5.0±2.3 <sup>a</sup> (45)	4.0±1.0 <sup>a</sup> (54.1)	5.2±2.2 <sup>a</sup> (62)	6.9±3.6 <sup>b</sup> (20.8)	12.1±4.1 <sup>c</sup>	3.7±2.3 <sup>b</sup> (51.3)	3.0±1.3 <sup>b</sup> (59.2)	2.0±1.0 <sup>a</sup> (71.6)	7.1±1.1 <sup>c</sup> (43.7)	8.0±2.6 <sup>d</sup>
F value			2.88*					3.21*		
LSD			1.41					0.81		

Table 2. Percentages of honeybee adult workers infestation with *Varroa* mite after treating the colonies with different compounds in both traditional beekeeping and compressed bees during 2003/2004 season

Inspection date	Traditional beekeeping					Compressed bees				
	Natural compound		Traditional acaricides			Natural compound		Traditional acaricides		
	Thymol	Thymol & Santonica	Apistan	Mavrik	Untreated check	Thymol	Thymol & Santonica	Apistan	Mavrik	Untreated check
Before treat.										
20/12/2003	14.6±2.9	18.5±2.4	20.5±5.9	19.2±2.7	18.9±3.6	13.2±3.4	20.1±3.1	17.5±4.2	20.2±4.8	20.4±6.7
After treat.										
28/12	12.3±2.3 (21)	16.5±3.6 (16.3)	17.5±4.3 (18.2)	17.3±3.2 (15.4)	20.1±4.9	10.5±2.8 (11.5)	18.2±3.1 (0.8)	15.9±6.6 (1.9)	17.5±3.4 (5.9)	18.2±4.9
3/1/2004	11.8±2.2 (19)	14.5±4.3 (22)	13.1±3.3 (35)	16.4±3.5 (15)	18.9±4.1	8.9±1.8 (24.3)	17.0±2.3 (5)	10.7±2.7 (31)	14.0±4.2 (22)	18.0±5.5
10/1	10.7±2.1 (23.3)	13.5±4.8 (23.3)	11.0±2.3 (42.2)	15.5±2.8 (14.9)	18.0±5.9	7.8±1.6 (26.2)	15.5±1.2 (3.7)	8.3±1.8 (41.2)	12.0±4.0 (26.2)	16.3±3.4
16/1	10.3±1.8 (27.9)	12.3±4.8 (32)	9.1±1.9 (53.6)	14.5±3.3 (22.7)	18.3±3.4	6.8±1.2 (28.6)	13.6±2.1 (4.8)	6.7±1.6 (46.8)	9.8±4.3 (32.8)	14.6±2.1
23/1	8.9±2.3 (34.6)	10.4±4.6 (38.9)	7.8±1.2 (57.5)	13.9±3.2 (21.5)	17.4±4.8	5.9±1.3 (37)	10.8±1.9 (23.9)	5.5±0.7 (56.3)	9.3±3.9 (35)	14.5±9.7
29/1	7.8±2.1 (43.3)	9.3±4.7 (38)	6.6±0.7 (59)	12.8±3.6 (18.2)	15.2±3.2	5.2±0.7 (35.3)	8.9±1.1 (27)	5.0±0.7 (53.5)	8.2±3.3 (33.6)	12.3±4.2
5/2	7.1±2.1 (39.5)	7.8±3.4 (47)	5.9±0.3 (63.5)	12.1±3.3 (20.6)	15.0±2.8	4.5±0.9 (35.3)	6.7±0.8 (39)	3.9±0.5 (59.3)	7.8±3.4 (27.8)	11.0±4.1
11/2	6.3±1.7 (41.9)	6.9±2.7 (50)	5.3±0.4 (64.9)	11.5±3.4 (19)	14.0±4.6	3.5±0.9 (53.5)	5.3±0.8 (53.5)	3.1±0.8 (67.8)	7.1±2.9 (37.3)	11.4±3.2
18/2	5.9±1.7 (49.2)	6.5±2.4 (55.5)	5.1±1.0 (68.2)	10.2±2.7 (32.7)	14.9±5.2	3.1±1.2 (57.3)	3.9±1.2 (63.1)	1.5±0.4 (84.5)	6.8±2.9 (34)	10.5±2.9
24/2	5.5±1.8 <sup>a</sup> (55.2)	6.4±2.3 <sup>b</sup> (59.9)	4.9±1.1 <sup>a</sup> (71.7)	9.8±2.6 <sup>c</sup> (39.8)	16.0±3.2 <sup>d</sup>	2.6±1.2 <sup>b</sup> (61.8)	3.8±1.6 <sup>c</sup> (63.7)	1.1±0.5 <sup>a</sup> (88.5)	6.3±3.3 <sup>d</sup> (40.8)	10.7±4.1 <sup>e</sup>
F value			4.83**					5.52**		
LSD			1.43					1.13		

(5.5%). The maximum (9.8%) was however, recorded after using Mavrik. In case of compressed bees, the rate of infestation with *Varroa* mite gradually decreased as the time after treatment progressed to reach 1.1, 3.8, 2.6 and 6.3% after using Apistan, Thymol and Santonica mixture, Thymol alone and Mavrik, respectively.

Colonies received normal beekeeping were compared with those of compressed bees and the obtained data clearly indicate that bee compression increased the efficiency of compound used for controlling varroa mite in these colonies. The reduction rates due to varroa treatment irrespective of the type of material used were 71.7 and 88.5% for traditional beekeeping and compressed bees, respectively. The corresponding values for thymol and santonica mixture were 59.9 and 63.7%, for thymol alone were 55.2 and 61.8% and for Mavrik were 39.8 and 40.8%.

### 3- Number of *Varroa* fallen in treated colonies

Ten weeks after starting the experiment and in case of traditional beekeeping, the mean number of *Varroa* fallen on the floor of the untreated hive was 121.5 individuals per colony, **Table (3)**. This number increased significantly after treating the colonies with either natural compounds or traditional acaricides. Mean accumulated numbers of 789.4, 589.2, 533.5 and 491.3 mites/colony were recorded after treating the colonies with Apistan, thymol and Santonica mixture, Thymol alone and Mavrik, respectively. In case of compressed bees, highly significant increase in the number of *Varroa* fallen after treating the honeybee colonies with different tested compounds. The highest accumulated number of *Varroa* fallen per colony was recorded after using Apistan (881.6), followed by thymol and santonica mixture (788.7) and thymol alone (737.6). The least accumulated number of *Varroa* (551.9) was, however, recorded after treating the colony with Mavrik. It is important to notice that in case of Apistan all individuals of mites parasitized the bees of the colony were found failed seven weeks after treatment, after that no *Varroa* fallen were detected till the end of the experiment.

The data clearly show that, irrespective of the specific acaricide Apistan, which is recommended for *Varroa* control in the apiaries of many countries of the world, thymol and santonica mixture as natural compounds was found to be the most efficient materials for controlling these mites in the

honeybee colonies, as the highest accumulated numbers of *Varroa* fallen were found on the floor of treated hive.

### 4- Sealed worker brood cells

It is well known that, the increase in the number of sealed worker brood in the honeybee colony is considered as good indicator for queen vitality and, thus, the colony strength. Therefore, the mean numbers of sealed worker brood cells were obtained biweekly after treating the *Varroa* infested colonies with different compounds, **Table (4)**. In case of traditional beekeeping, the mean number of sealed brood cells before treatments ranged between 1207 - 2499 cells/colony. After treatments, this number increased gradually with two weeks interval. Ten weeks after treatments the number of sealed brood cells varied according to the type of compound used for controlling *Varroa* mites. The highest number of sealed brood cells per colony was obtained after using Apistan (5543), followed by thymol and santonica mixture (4328) and thymol alone (4118). The least number (3333) was obtained after using Mavrik. However, in the later case, the number of sealed brood cells was still significantly higher than that of the untreated colonies, which recorded (2190) cells/colony. In case of compressed bees, the average number of sealed worker brood cells per colony ten weeks after starting the experiment varied according to the type of compound used for controlling *Varroa* mites. The maximum account was 7526 cells/colony after using Apistan, followed by Thymol and Santonica mixture 6527 and Thymol alone 6011. The minimum amount of cells 5240 was obtained after using Mavrik. However, in the later case, the number of sealed brood cells was about the double in number as compared with that of the untreated colony, which recorded 2578 cells/colony.

### 5- Colony strength (combs covered with adult bees)

It is well known that an increase in the population density of the adult worker bees in the colony causes strength of this colony. Therefore, the numbers of combs covered with adult bees from both sides in the bee colonies were recorded weekly after treating these varroa-infested colonies with different compounds. The obtained data are given in **Table (5)**.

Table 3. Average numbers of *Varroa* fallen after treating the honeybee colonies with different compounds in both traditional beekeeping and compressed bees during 2003/2004 season

Inspection date	Traditional beekeeping					Compressed bees				
	Natural compound		Traditional acaricides		Untreated check	Natural compound		Traditional acaricides		Untreated check
	Thymol	Thymol & Santonica	Apistan	Mavrik		Thymol	Thymol & Santonica	Apistan	Mavrik	
Before treat. 20/12/2003	12.7±3.6	10.2±3.4	16.0±6.1	8±2.16	20±2.6	8±2.44	7.25±2.21	8.75±2.5	12.75±2.21	13.5±5.3
After treat. 28/12/2003	50.7± 5.9	40.2± 9.9	327.7±173.9	133±25.2	1.8±5.7	67.5±5	77.5±12.1	355±24.83	154.7±11.6	13.5±8.7
3/1/2004	58.7± 5.9	55.7±12.5	185.5±31.2	97.5±27.8	19.3±6.3	75.5±3.4	85.5±77	236.7±36.1	111.2±13.5	16.3±3.4
10/1	67.0± 5.5	67.7±15.9	105.7±34	76.2±18.9	14.1±9.4	88.2±7	112.2±13.2	144±26.3	89.7±5.7	15±6.7
16/1	71.0±13.5	85.5±21.6	57.5±23.9	63.7±18	14±3.6	97±4.2	136.2±25	76±19.5	61.2±7.4	15.4±6.0
23/1	79.0±13.7	92.0±19.9	43.7±16.6	48.5±11.9	15.4±2.7	109±10.7	104.7±15	43.7±11.08	55.2±6	19.8±7.6
29/1	78.0±10.1	98.7± 8.3	30.5±2.6	31±9.4	11±4.4	101±11.6	67.7±11.3	18±6.68	37±5.9	20.2±3.4
5/2	50.7± 6.7	64.7± 6.4	19±6.8	19±5.7	9±4.2	79.7±3.7	68.7±6.9	8.2±2.5	23.2±4.03	24.6±6
11/2	37.2± 2.5	42.0± 6.0	9.5±4.8	9.5±2.08	10.6±2.5	58.5±5.06	60.5±5.8	0	12.5±3.4	29.1±5.8
18/2	24.7±10.0	28.5± 7.4	7±4.08	6.7±4.9	12.3±4.2	39.5±1.3	45.7±2.7	0	4.7±2.2	27.5±4.9
24/2	16.0± 4.1	14.2± 3.9	3.3±1.52	6.2±4.3	14±5.8	21.7±5.6	30±2.6	0	2.5±0.70	28.3±6.3
Total (After treat.)	533.5 <sup>b</sup>	589.2 <sup>b</sup>	798.4 <sup>a</sup>	491.3 <sup>b</sup>	121.5 <sup>c</sup>	737.6 <sup>b</sup>	788.7 <sup>b</sup>	881.6 <sup>a</sup>	551.9 <sup>c</sup>	209.7 <sup>d</sup>
F value	3.21*					6.07**				
LSD	126.3					73.1				

Table 4. Average numbers of sealed worker brood in honeybee colonies after treating them with different compounds in both traditional beekeeping and compressed bees during 2003/2004 season

Inspection date	Traditional beekeeping					Compressed bees				
	Natural compound		Traditional acaricides		Untreated check	Natural compound		Traditional acaricides		Untreated check
	Thymol	Thymol & Santonica	Apistan	Mavrik		Thymol	Thymol & Santonica	Apistan	Mavrik	
Before treat.										
20/12/2003	2499±855	1207±961	2117±1727	2271±1898	1980±323.8	1860±1738	2515±673	2276±1132	1642±594	2010±476
After treat.										
3/1/2004	2479±1964	1322±637	2162±1774	1688±379	1490±621.8	1631±1062	2206±708	2302±813	1826±357	1870±594.4
16/1	2140±1298	2111±1115	2506±689	1941±807	1520±516.3	2333±796	2299±503	2720±91	2012±732	1790±496.6
29/1	2906±901	2715±637	2817±496	2328±975	1670±476	3541±869	3981±83.5	3419±321	2945±678	1985±365
11/2	3880±1283	3585±649	4580±717	2931±304	1960±516.3	4895±930	4986±1454	6186±1328	3844±642	2198±258
24/2	4118±934 <sup>b</sup>	4328±704 <sup>b</sup>	5543±989 <sup>a</sup>	3333±752 <sup>b</sup>	2190±416.3 <sup>c</sup>	6011±1228 <sup>b</sup>	6527±1039 <sup>b</sup>	7526±1248 <sup>a</sup>	5240±964 <sup>b</sup>	2578±439.7 <sup>c</sup>
F value	3.691*					14.44**				
LSD	1083					1104				



Table 5. Average numbers of combs covered with bees (colony strength) in honeybee colonies after treating them with different compounds in both traditional beekeeping and compressed bees during 2003/2004 season

Inspection date	Traditional beekeeping					Compressed bees				
	Natural compound		Traditional acaricides		Untreated check	Natural compound		Traditional acaricides		Untreated check
	Thymol	Thymol & Santonica	Apistan	Mavrik		Thymol	Thymol & Santonica	Apistan	Mavrik	
Before treat. 20/12/2003	3.25±0.5	2.7±0.5	3.5±0.57	3.25±0.95	3±0.82	3±0.81	2.5±0.57	3±1.15	2.7±0.5	3±0.81
After treat. 28/12/2003	3±0	2.7±0.5	3.5±0.57	2.75±0.5	2.5±0.58	3±0.81	2.5±0.57	3±1.15	2.7±2.5	3±0.5
3/1/2004	2.5±0.6	2.7±0.5	3.5±0.57	2.75±0.5	2.5±0.58	3±0.81	2.7±0.5	3.5±0.9	3±0	2.5±0.6
10/1	2.5±0.6	3±0.81	3.5±0.57	2.75±0.5	2.2±0.5	3±0.81	3±0	3.7±0.9	3.5±0.6	2.5±0.5
16/1	3.5±0.6	3.2±0.5	3.5±0.57	2.75±0.5	2.5±0.58	3.2±0.9	3.7±0.5	4.2±0.9	3.5±0.6	2.5±0.5
23/1	3.5±0.6	3.7±0.9	3.7±0.5	3±0.81	2.5±0.58	3.2±0.9	4.5±0.6	5±1.15	3.5±0.6	3±0.5
29/1	3.7±0.9	3.7±0.9	4±0.81	3.2±0.95	2.5±0.58	3.7±0.9	5±0.81	5.2±0.9	4.2±0.5	3±0.5
5/2	4±0.81	4.2±1.2	4.7±0.5	3.7±0.5	2.7±0.5	4.7±0.9	5.5±1	6.2±0.9	4.5±0.6	3±0.5
11/2	4.5±0.6	4.5±1	4.7±0.5	4.2±0.9	2.7±0.5	5±0.81	6.2±0.95	6.7±0.9	4.7±0.9	3.5±0.5
18/2	4.5±0.6	5±1.41	5.2±0.95	4.2±0.9	3±0.82	6±0.81	6.7±1.2	7.2±0.9	5.2±0.9	3.7±0.5
24/2	4.7±0.9 <sup>b</sup>	5±1.41 <sup>a,b</sup>	5.2±0.95 <sup>a</sup>	4.2±0.9 <sup>c</sup>	3±0.82 <sup>d</sup>	6±0.81 <sup>c</sup>	6.7±1.2 <sup>b</sup>	7.2±0.9 <sup>a</sup>	5.2±0.9 <sup>d</sup>	4.5±0.5 <sup>e</sup>
F value	3.14*					5.98**				
LSD	0.392					0.431				

In traditional beekeeping, before treatment the mean number of combs covered with bees ranged between 2.7 - 3.5 combs/colony. Four weeks after treatments, the number of combs covered with bees did not apparently affect with any of the used compounds, which ranged between 2.75 - 3.50 combs/colony. After this period, gradual increase was found to reach the maximum at the end of the experiment (10 weeks after treatments). At this period, the mean number of combs covered with adult bees varied according to the type of compound used for *Varroa* control. The maximum number of combs/colony was obtained after using Apistan (5.2), followed by thymol and santonica mixture (5.0), thymol alone (4.7), Mavrik (4.2) and finally the untreated check (3.0).

In case of compressed bees, before treatment the mean number of combs covered with bees ranged between 2.5 - 3 combs/colony. Two weeks after treatments, an obvious increase in the number of combs covered with bees and gradually increase towards the end of the experiments was obtained to reach the maximum ten weeks after treatments. At this period, the number of combs covered with bees varied according to the type of compound used. The maximum number of combs/colony was obtained after using Apistan

(7.2), followed by thymol and santonica mixture (6.7), thymol alone (6.0) and Mavrik (5.2). In untreated colonies the mean number of combs/colony was 4.5. This number was significantly higher than that of the corresponding number in colonies receiving traditional beekeeping ( $3.0 \pm 0.82$ ).

Generally, comparing the experimental colonies, which were, received normal beekeeping with those of compressed bees, Table (6) and Fig. (1) clearly show that bee compression increased the efficiency of any compound used for *Varroa* control, except for Mavrik. Moreover, the superiority of Thymol and Santonica mixture for controlling *Varroa* mites irrespective of Apistan was noticed. This mixture caused an obvious reduction of the rate of infestation either in the sealed brood or on the adult bees, and obvious increase in the number of varroa fallen on the floor of treated hive, an apparent increase in the number of sealed worker brood cells and, lastly, an increase in the number of combs covered from both sides with adult worker bees (colony strength) in the treated colony. This findings coincide with those of Imdorf *et al* 1995; Calderone *et al* 1997; Baxter *et al* 1998; Pettis *et al* 1998; Sammataro *et al* 1998 and Elbassiouny, 2003.

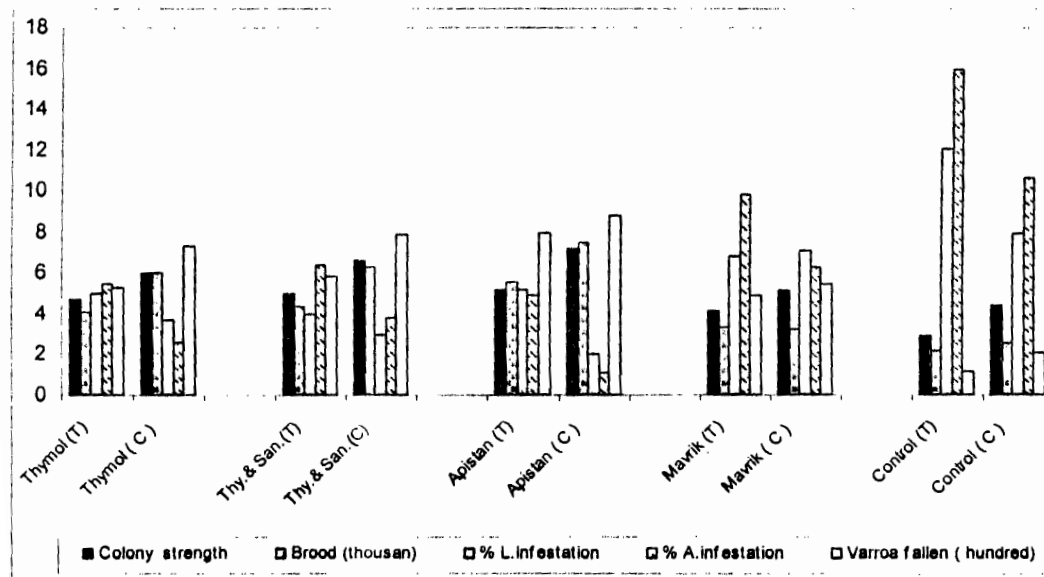


Fig. 1. Honeybee activities and *Varroa* infestation rates as being affected by different tested compound in both traditional beekeeping and compressed bees during 2003/2004 season.

Table 6. Honeybee activities and Varroa infestation rates as being affected by different tested compound in both traditional beekeeping and compressed bees during 2003/2004 season

Parameters	Traditional beekeeping					Compressed bees				
	Natural compound		Traditional acaricides			Natural compound		Traditional acaricides		
	Thymol	Thymol & Santonica	Apistan	Mavrik	Untreated check	Thymol	Thymol & Santonica	Apistan	Mavrik	Untreated check
Brood infes.										
Before tr.	12.5±7.2	11.5±4.7	18.0±5.2	17.5±7.2	15.9±4.8	15.5±7.0	15.0±4.8	14±5.9	16.5±4.4	16.3±5.0
After tr.	5.0±2.3 (45) <sup>d</sup>	4.0±1.0 (54.1) <sup>c</sup>	5.2±2.2 (62) <sup>b</sup>	6.9±3.6 (20.8) <sup>c</sup>	12.1±4.1	3.7±2.3 (51.3) <sup>c</sup>	3.0±1.3 (59.2) <sup>b</sup>	2.0±1.0 (71.6) <sup>a</sup>	7.1±1.1 (43.7) <sup>d</sup>	8.0±2.6
F value						4.92				
LSD						3.9				
Adult infes.										
Before tr.	14.6±2.9	18.5±2.4	20.5±5.9	19.2±2.7	18.9±3.6	13.2±3.4	20.1±3.1	17.5±4.2	20.2±4.8	20.4±6.7
After tr.	5.5±1.8 (55.2) <sup>c</sup>	6.4±2.3 (59.9) <sup>d</sup>	4.9±1.1 (71.7) <sup>b</sup>	9.8±2.6 (39.8) <sup>g</sup>	16.0±3.2	2.6±1.2 (61.8) <sup>de</sup>	3.8±1.6 (63.7) <sup>c</sup>	1.1±0.5 (88.5) <sup>a</sup>	6.3±3.3 (40.8) <sup>f</sup>	10.7±4.1
F value						6.43				
LSD						4.1				
Varroa fallen										
Before tr.	12.7±3.6	10.2±3.4	16.0±6.1	8±2.16	20±2.6	8±2.44	7.25±2.21	8.75±2.5	12.75±2.21	13.5±5.3
After tr.	533.5 <sup>de</sup>	589.2 <sup>c</sup>	798.4 <sup>b</sup>	491.3 <sup>d</sup>	121.5	737.6 <sup>b</sup>	788.7 <sup>b</sup>	881.6 <sup>a</sup>	551.9 <sup>c</sup>	209.7
F value						9.64				
LSD						60.2				
Brood										
Before tr.	2499±855	1207±961	2117±1727	2271±1898	1980±323.8	1860±1738	2515±673	2276±1132	1642±594	2010±476
After tr.	4118±934 <sup>e</sup>	4328±704 <sup>d</sup>	5543±989 <sup>c</sup>	3333±752 <sup>c</sup>	2190±416.3	6011±1228 <sup>b</sup>	6527±1039 <sup>b</sup>	7526±1248 <sup>a</sup>	240±964 <sup>de</sup>	5240±964
F value						10.54				
LSD						989				
Combs										
Before tr.	3.25±0.5	2.7±0.5	3.5±0.57	3.25±0.95	3±0.82	3±0.81	2.5±0.57	3±1.15	2.7±0.5	3±0.81
After tr.	4.7±0.9 <sup>de</sup>	5±1.41 <sup>dc</sup>	5.2±0.95 <sup>c</sup>	4.2±0.9 <sup>c</sup>	3±0.82	6±0.81 <sup>b</sup>	6.7±1.2 <sup>a</sup>	7.2±0.9 <sup>a</sup>	5.2±0.9 <sup>c</sup>	4.5±0.5
F value						4.32				
LSD						0.54				

### Fluvalinate residues in bee honey, beeswax and honey bee workers

Data in Table (7) and Fig. (2) indicate fluvalinate residues, (ppm) in bee honey, beeswax and honeybee workers at different time after application. Examination of the obtained data reveal that, fluvalinate residues varied tremendously due to the source of detection as well as time elapsed after treatment. As a general, trend fluvalinate residues (ppm) seemed to be the highest in honeybee workers followed by bee honey and beeswax products, respectively. Also, the advancement of the after applications was positively correlated with the rate of fluvalinate accumulation.

Fluvalinate residues were 1.2, 1.5 and 1.8 ppm in honeybee workers, 0.6, 1.4 and 1.9 ppm in bee honey and 0.48, 0.96 and 1.3 ppm. in beeswax 2 weeks, 1 and 2 months after treatment, respectively.

In this respect, Lodesani *et al* (1992) proved that, bromopropylate, thymol and fluvalinate were

persisted in bee honey and wax samples from the brood chamber. Kubik *et al* (1995) showed that, the rate of fluvalinate release from Apistan strips is initially low but increases with time. Bees spread fluvalinate within the hive whereas the diffusion of vapours has a negligible effect. Moreover, they found that beeswax and propolis were contaminated with fluvlinate much more (up to 8 ppm) than honey or worker bees (0.010-0.036 ppm). Wallner (1999) mentioned that, fluvalinate accumulate in beeswax with years of treatment. Through the process of diffusion the ingredient migrate from the wax comb into the stored bee honey.

Chemical analysis of residual level of fluvalinate was measured in bee honey, beeswax and adult worker bees by Lodesani *et al* (1992). They found that this compound persisted in wax samples only. Bogdanov *et al* (1998) found that fluvalinate accumulated in brood comb wax of colonies treated continuously with Apistan for one year.

Table 7. Fluvalinate residues in bee honey, beeswax and honey bee workers at different time after treatment

Time after treatment days	Bee Honey		Beeswax		Honey Bee	
	Conc. ppm	% increase	Conc. ppm	% increase	Conc. ppm	% increase
2 weeks	0.6	-	0.48	-	1.2	-
1 month	1.4	133.3	0.96	100	1.5	25.0
2 months	1.9	216.7	1.3	170.8	1.8	50.0

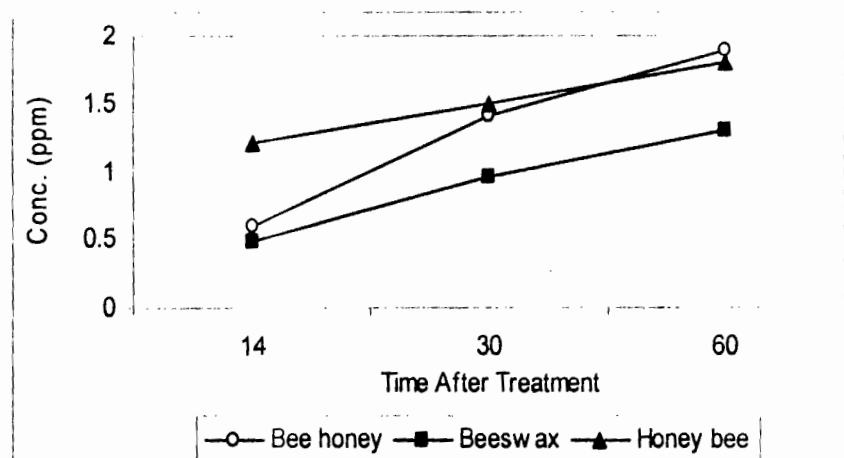


Fig. 2. Fluvalinate residues from acaricide in the bee honey, beeswax and honey bee workers at different time after application

## REFERENCES

- A.O.A.C. (1990). Multiresidues methods: Federal Methods for organochlorine and organophosphorous pesticides. *Assoc. Official. Anal. Chem.* 13: 466-472.
- Bogdanov, S.; A. Imdorf and V. Kilchenmann (1998). Residues in wax and honey after Apilife/VAR treatment. *Apidologie* 29(6): 513 – 524.
- Baxter J.R.; F. Eischen; J. Pettis; W.T. Wilson and H. Shimanuki (1998). Detection of fluvalinate – resistant varroa mites in U.S honey bee. *Amer. Bee J.* 138 : 291.
- Calderone, N.W.; W.T. Wilson and M. Spivak (1997). Plant extracts used for control of the parasitic mites *Varroa jacobsoni* (Acari: Varroidae) and *Acarapis woodi* (Acari: Tarsonemidae) in colonies of *Apis mellifera* (Hymenoptera: Apidae). *J. Econ. Entomol.* 90 (5):1080-1086.
- Colin, M.E. (1990). Essential oils of labiate for controlling honey bee varroosis. *J. Appl. Entomol* 110(1): 19-25.
- Colin, M.E.; R. Vandame; P. Jourdana and S.Di. Pasquale (1997). fluvalinate resistance of *Varroa Jacobsoni* oudemans (Acari: Varroidae) in Mediterranean apiaries of France. *Apidologie* 28(6): 375-384.
- De Jong, D.; R.A. Morse and G.C. Elckwort (1982). Mite pests of honeybees. *Ann. Rev. Entomol* 27: 229-252.
- Elbassiony, A.M. (1998). Effect of the ectoparasitic mite, *Varroa Jacobsoni* on the longevity and hoarding behavior of honey bee workers. *Annals Agric., Sci., Ain Shams Univ., Cairo*, 43(2): 599- 605.
- Elbassiony, A.M. (2003). Maintaining and developing varroa tolerant parameters. *Arab. Univ. J. Agric. Sci.* 11(1): 427 – 437; 25.
- El-Bassiouny, A.M.; A.A. Gomaa; M.A. El-Banby and M.A.M. Ali (2004). Aproximated damage threshold level for varroa mite *varroa destructor* Inhabiting honey bee colonies. *Ann. Agric. Sci., Ain Shams Univ., Cairo*, 49(2): 787 - 792.
- Glinski, Z. and J. Jarose (1992). *Varroa Jacobsoni* as a carrier of bacterial infections to a recipient bee host. *Apidologie* 23: 25-31.
- Imdorf, A.; V. Kilchenmann; S. Bogdanove; B. Bachofen and C. Beretta (1995). Toxic effects thymol, camphor, menthol and eucalyptol on *Varroa jacobsoni* Oud, And *Apis mellifera*. L.in Laboratory test. *Apidologie* 26(1): 27-31.
- Kubik, M.; J. Nowacki; L. Michalczyk; A. Pidek and J. Marcinkowski (1995). Penetration of fluvalinate into bee products. *Journal of Fruit and Ornamental Plant Research*. 3(1): 13-22.
- Lodesani, M.; A. Pellacani; S. Bergomi; E. Carpana; T. Rabitti and P. Iasagni (1992). Residue determination for some products used against Varroa infestation in bees. *Apidologie*. 23(3): 257-272.
- Pettis, J.; H. Shimanuki and M. Feldlaufer (1998). An assay to detect fluvalinate resistance in Varroa mites. *Am. Bee J.* 138(7): 538-541.
- Sammataro, D. (1997). Report on parasitic honeybee mites and disease associations. *Am. Bee J.* 137(3): 301-303.
- Sammataro, D.; H. Degrandi; G. Needham and G. Warell (1998). Some volatile oils as potential control agents for Varroa mites (Acar.:Varroidae) in honeybee colonies. *Am. Bee J.* 138(9): 681-685.
- Shimanuki, H.; N.W. Calderon and D.A. Knox (1994). Parasitic mite syndrome. The symptoms. *Am. Bee J.* 134(10): 827-828.
- Slabezki, Y.; H. Gal and Y. Lensky (1991). The effect of fluvalinate application in bee colonies on population levels of *Varroa jacobsoni* and honeybee (*Apis mellifera*) and on residues in honey and wax. *Bee Science* 1: 189-195.
- Wallner, K. (1995). The use of varroacides and their influence on the quality of bee products. *Am. Bee J.* 135(12): 817-821.
- Wallner, K. (1999).Varroacides and their residues in bee products. *Apidologie*. 30(3): 235-248.



حوليات العلوم الزراعية  
جامعة عين شمس، القاهرة  
مجلد (٥١)، عدد (٢)، ٥٥٩-٥٧٢، ٢٠٠٦

## تأثير ادارة الطائفة والمستخلصات النباتية على حلم الفاروا الذى يهاجم طوائف نحل العسل

[ ٤١ ]

عادل محمد البسيونى<sup>١</sup> - محمد ابراهيم عبد المجيد<sup>١</sup> - محمد اسامة الشعراوى<sup>٢</sup> - جيهان محمود محمد<sup>٢</sup>

١- قسم وقاية النبات - كلية الزراعة - جامعة عين شمس - شبرا الخيمة - القاهرة - مصر

٢- قسم بحوث النحل - معهد بحوث وقاية النبات - مركز البحوث الزراعية - الدقى - القاهرة

البالغ وفى عش الحضنة مع زيادة اعداد الفاروا المتساقط وزيادة مساحة الحضنة واعداد الاقراص الشمعية المغطاة بالنحل البالغ بالمقارنة بباقي المكونات الاخرى فى حالة الاصابة المتوسطة والضعيفة ، اما فى حالة الاصابة الشديدة فينصح باستخدام المبيد الاكاروسى التقليدى الموصى به (الابستان). كما اظهرت النتائج ايضا ان ادارة الطائفة (العمليات النحلية) عن طريق ضغط النحل على اقل عدد من الاقراص (خفض مساحة الطائفة من الداخل) قد اعطت نتائج ايجابية وتفوق عن العمليات النحلية التقليدية.

وقد اظهر التحليل الكيماوى تراكم المادة الفعالة للمبيدات التقليدية (الفلوفينات) فى شغالات نحل العسل بجرعات ١,٢ ، ١,٥ ، ١,٨ جز فى المليون. كانت فى حالة العسل ٠,٣ ، ١,٤ ، ١,٩ جزء فى المليون. وكانت فى حالة الشمع كانت ٠,٤٨ ، ٠,٩٦ ، ١,٢ جزء فى المليون وذلك بعد المعاملة بأسبوعين وشهر وشهرين على التوالى.

أصبح حلم الفاروا من أهم مشاكل النحالة فى مصر لكونه طفيل خارجى على كل من الحضنة المقفولة والنحل البالغ حيث يتغذى على هيمولينف النحل فيضعفه، وقد تؤدى الاصابة الشديدة إلى تدهور الطائفة وفنائها. حاليا تعتبر ادارة الطائفة واستخدام بدائل علاجية من المنتجات الطبيعية المستخلصة من النباتات الطبية والعطرية ضرورية لآمانها على كل من النحل والانسان. لذلك تم تقييم الثيمول وخليط الثيمول والشيخ البلدى ضد طفيل الفاروا فى موسم ٢٠٠٣/٢٠٠٤ فى منطقت قليوب بمحافظة القليوبية، ومقارنة النتائج بتلك المتحصل عليها بعد استخدام المبيدات الاكاروسية التقليدية وهى الابستان والمافريك حيث تم تقدير نسب اصابة النحل البالغ والحضنة بالطفيل - اعداد الحلم المتساقطة - عدد عيون الحضنة - واعداد الاقراص المغطاه بالنحل نتيجة المعاملة اسبوعيا ولمدة ١٠ اسابيع فى منطقة قليوب .

وقد اظهرت النتائج تفوق خليط الثيمول والشيخ البلدى فى مقاومة هذا الطفيل على كل من النحل

تحكيم: أ.د أحمد على جمعه

أ.د محمد عطيه عويس