

## EVALUATION OF SOME NATURAL SUBSTANCES AGAINST *VARROA DESTRUCTOR* ANDERSON AND TRUEMAN INFESTING HONEYBEE, *APIS MELLIFERA* IN EGYPT

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### **Abstract**

This study was conducted to investigate the effect of certain natural substances against *Varroa* mite. Four natural substances (Apiguard and Thyme, Camphor and Basil oils) were evaluated in the field as control agents against the parasitic mite, *V. destructor*. Four treatments of each tested material were applied to each colony during the treatment period. . Data showed that all four tested compounds were effective in controlling *Varroa* and Apiguard was the most effective substance as they had an efficiency of 81.3%, 74.6%, 71% and 67%, respectively.

### **INTRODUCTION**

*Varroa destructor* is considered one of the most serious pests affecting honeybee, *A. mellifera* on a worldwide basis, causing weight loss, malformation, and a shortened life span. It also serves as a vector of diseases that may lead to 100% bee mortality (Kanga and James, 2002). Chemical control using acaricides are still used to reduce mite populations in beehives, including Coumaphos, Apistan, Amitraz, Folbex, Folbex-Forte, Apitol and Sinecar (Henderson, 1988, Martin, 1994, Gregorc and Poklukar, 2003). These acaricides have significant drawbacks including contamination of honey, wax, and pollen (Calderone and Spivak, 1995), additionally resistant strains can be developed (Elzen *et al.*, 1999). Consequently, natural components might offer promising new avenues for the beekeeping industry. Many studies had been carried out using some extracts of natural essential oil of various plants such as rosemary, lemongrass, camphor, clove, ginger roots, fennel and eucalyptus (Gregorc and Poklukar, 2003; Batish *et al.*, 2008). Other scientists investigated the effectiveness of Apiguard and some organic acids (Bacandritos *et al.*, 2007). Therefore, this study was conducted to evaluate the use certain essential oils and Apiguard as alternative control methods that are safe, and environment friendly, against the parasitic mite, *V. destructor*.

### **MATERIALS AND METHODS**

The present study was carried out in an apiary at Giza Governorate in March and April 2008. Fifteen colonies of the honeybee, *A. mellifera* were identified as

being infested with *V. destructor*. Each colony consisted of 10 full-depth combs of worker bees and a queen. The colonies were divided into five equal groups. The first and second groups were treated with one of the following substance, Apiguard and Thyme, *Thymus vulgaris*, respectively, while the third and fourth groups were treated with camphor, *Eucalyptus* spp, and basil oils, *Ocimum basilicum*, the fifth group served as control. One application of the tested materials were applied weekly for four successive weeks (treatment period), using a filter paper (carrier) saturated with 5 ml of each tested material and placed on the top of colonies. Apiguard was applied in tray (two times, 50g) at 2 weeks interval. Control colonies received the carrier only without any test material.

To monitor *Varroa* populations before and after treatments, a "sticky board" was pushed in all hives's bottom prior to each treatment under the wire/wood frame, where falling mites were trapped and the wire kept bees from removing them (Sammataro *et al.*, 1998). These boards were removed, replaced by new ones daily and the trapped mites were counted. The hive entrances remained open during the experiment and applications were carried out after sunset, when all honeybees had returned to the hives. Surviving mites remaining in each colony after treatment period were killed by using formic acid (85%) for two consecutive weeks (treatment evaluation period) and counted.

The number of mites collected after each application was recorded and the efficiency percentage of each application of these compounds was determined.

The efficiency of each application was determined according to the formula of Colin (1990)

$$X_n (\%) = \frac{A_n \times 100}{Z - Z_{n-1}}$$

$A_n$ : number of mites collected throughout the days following each application,  $Z$ : total number of collected mites [mites collected after 4 applications + remaining mites collected after treatment evaluation period], and  $Z_{n-1}$ : number of collected mites before each application.

Data obtained in the *Varroa* control groups were analyzed statistically using analysis of variance (ANOVA) and means were separated by a least significant differences test in SAS.

## RESULTS AND DISCUSSION

Table (1) indicated that the mean number of mites collected from the colonies that received four applications of the Apiguard (during the treatment period) was significantly higher (151.3 mites) than those that have received thyme, camphor and Basil oils (117.6, 113 and 98 mites/ hive, respectively). The mean efficiency percentage of Apiguard was significantly higher ( $81\pm 3.2$ ) than thyme, camphor and Basil oils ( $74.6\pm 5.8$ ,  $71\pm 3.6$  and  $67\pm 6.2$ ) compared with the control group ( $14.6\pm 3.7$ ) at 5% significant levels.

Honeybee, *Apis mellifera* is considered one of the most important and beneficial insects, whose damage has serious negative economic implications for both beekeeping industry and agriculture (Melathopoulos *et al.*, 2000). In the present study, several natural substances were used as they are biodegradable, safe for human and relatively inexpensive, for the control of *Varroa destructor*.

Several authors have evaluated essential oils and their components as control agents for *Varroa* (Sammataro *et al.*, 1998, Imdorf *et al.*, 1999, Ariana *et al.*, 2002 and Ismail *et al.*, 2006), and indicated that many of these compounds are biologically toxic to *Varroa* without injuring bees. However, significant bee mortality was reported when powdered thymol was used *against V. destructor*, which was probably a consequence of the high surface area of thymol or high ambient temperatures (Chiesa, 1991 and Gal *et al.* 1992). Calderone (1999) indicated that Apistan is more effective than thymol-based blend. Therefore, numerous factors contribute to the overall efficacy of acaricides such as the concentration of compounds, delivery method, ambient temperature, and colony and apiary environment (Calderone and Spivak, 1995). On the other hand, Apiguard reduced *Varroa* mite populations on honeybees. There is no indication that this compound has adverse effects on the bees, where no apparent increases in number of dead bees at hive bottoms. Similarly, several authors have evaluated this compound as control agent for *Varroa destructor* mite (Mattila *et al.*, 2000, Emsen and Dodologlu, 2009). In addition, May-Itza *et al.*, (2007) indicated that the Apiguard (thymol gel) could eliminate ~95% of the mites in honeybee colonies under tropical conditions and thus could be used as an alternative method for controlling this parasite.

Generally, using natural substances to control *Varroa* mites in recent years, support the need to overcome resistance issues with currently registered acaricides.

Table 1. Results of field evaluation of different natural extracts against *Varroa* mite

Group	Colony	Number of collected mites after indicated applications :				AT	M	B	Z	X <sub>n</sub>	M±SD
		1st	2nd	3rd	4th						
Apiguard	1	41	23	35	17	116		21	137	85	81.3±3.2
	2	67	59	39	26	191	151.3	49	240	80	
	3	55	46	30	16	147		38	185	79	
Thyme	1	28	49	10	13	100		46	146	68	74.6±5.8
	2	51	38	27	26	142	117.6	37	179	79	
	3	49	22	19	21	111		33	144	77	
Camphor	1	27	34	18	10	89		34	123	72	71±3.6
	2	48	36	39	25	148	113	51	199	74	
	3	30	32	29	11	102		50	152	67	
Basil (50%)	1	42	25	16	7	90		47	137	65	67±6.2
	2	18	25	12	11	66	98	40	106	62	
	3	33	49	27	29	138		48	186	74	
Control	1	18	13	10	8	49		189	238	19	14.6±3.7
	2	13	4	6	2	25	35	154	179	13	
	3	8	10	6	7	31		171	202	12	

AT: Number of collected mites after the treatment period

B: Number of mites remaining in colonies after the 4 applications (treatment evaluation period)

Z: Total number of mites in each colony (AT + B)

X<sub>n</sub>: Percentage of Efficiency of the treatments after the four applications

M: Mean

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## تأثير بعض المواد الطبيعية على طفيل الفاروا *VARROA DESTRUCTOR*

غادة رفاعي

معهد بحوث وقاية النباتات - مركز البحوث الزراعية - الدقى - الجيزة

يعتبر طفيل الفاروا من اخطر الطفيليات التي تصيب نحل العسل و تسبب الكثير من الأضرار لصناعة نحل العسل ومنتجاته المتنوعة. من هنا إهتمت هذه الدراسة بإقتراح طرق وأساليب مكافحة فعالة وآمنة. لذلك أجريت التجارب الحقلية بهدف دراسة تأثير كلا من مادة الأبيجارد وزيت الزعتر، الكافور و البردقوش فى مكافحة طفيل الفاروا . حيث تمت معاملة خلايا نحل العسل المصابة بطفيل الفاروا ٤ مرات طوال فترة التجربة، وفى كل الخلايا تم فحص وتقدير عدد الأكاروسات الميتة المتواجدة أسفل كل خلية على حدة وكذلك حساب كفاءة هذه المواد . وقد اوضحت نتائج هذه الدراسة أن كل هذه المعاملات كانت فعالة معنويا ضد الفاروا فى الطوائف المعاملة مقارنة بالغير معاملة حيث وصلت كفاءة مادة الأبيجارد فى خفض طفيل الفاروا إلى ٨١,٣ % بينما بلغت كفاءة المواد الأخرى ٧٤,٦ % ، ٧١% و ٦٧% على التوالى .