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COMBATTING HONEYBEE *VARROA* MITES BY PLANT OILS ALONE OR IN AN IPM PROGRAM

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Abstract:

Eleven volatile oils were tested against *Varroa* mites in vitro. These oils were spearmint, thyme, eucalyptus, marjoram, cumin, garlic, basil, orange, geranium, menthol and eugenol. Basil, geranium and eugenol exhibited noticeable varroacidal activity and were selected for different in vivo tests during winter, spring, and autumn seasons. Candidate oils were separately added to a pollen supplement integrated with formic acid, oxalic acid and queen caging techniques. To evaluate these treatments, % infestation of brood and bees, counts of dropped mites and dead bees, and brood rearing activity were recorded. Results showed that all treatments were significantly effective against *Varroa* in treated colonies compared to untreated ones. Counts of dropped mites were also significantly high. Some adverse effects including reduced brood area, colony disturbance and bee mortality occurred in some treatments, especially those with eugenol, which seemed to be harmful to bees. Therefore, the IPM approach is recommended to combat *Varroa* mites.

Key Words: Honeybee - *Varroa* – Control – Plant oils – IPM.

Introduction:

The serious ectoparasitic mite, *Varroa destructor* Anderson & Trueman (formerly *V. jacobsoni* Oud.), is a subject of concern to beekeepers worldwide. This mite which feeds on haemolymph of brood and adult bees causes colony disorder, weakness, decrease in brood and deformation of bees. It also reduces colony ability to pollinate plants (De Jong *et al*. 1984). Besides the economical loss of bees and honey production, infested colony may die or migrate (Needham, 1988). The parasite destroys the mechanical protective barriers of the integument and impairs the immune system of the bees (Glinski, 1991). Different pathogens such as acute bee paralysis virus, deformed wing virus, and fungi are probably transferred to bees by *Varroa* (Allen & Ball, 1996). More recently, six bee viruses were detected in bees infested by *Varroa*, so amplifying pathogenisity in honeybees is expected (Tentcheva *et al.* 2004).

In Egypt, the first detection of this parasite was in September 1987 at El-Arish region, became wide spread in 1989 and heavy infestations were found in 1990 in many regions

where apiaries were almost destroyed. For the control of bee pests and diseases including *Varroa* mites, chemicals were used, which must be constrained and legally recommended otherwise bee products will be contaminated enough to be dangerous for humans (Delaplane, 1997). Also, the repetition of pesticides use caused severe problems such as bee toxicity, and increased probability of disease-resistance (Milani, 1995 and Watkins, 1997). For these reasons, the more recent approach to control this mite was by the use of relatively safe natural products, especially volatile oils, and/or mechanical and biological managements. Therefore, as a contribution to *Varroa* control, this work was carried out to minimize *Varroa* mite population by applying cheap, available and almost safe plant materials alone or by amplify their effectiveness via an IPM program. The impact of these treatments on bees and colony performance was also observed.

Materials and Methods:

This work was conducted in the apiary of the Faculty of Agricultue, at Fayoum Governorate, Egypt, during 2000-2001 season as follows:

A. Selected materials:

Nine botanical volatile oils, extracted by water distillation, were tested to control *Varroa* mites. These oils were namely: spearmint; *Mentha viridis*, thyme; *Thymus vulgaris*, eucalyptus; *Eucalyptus spp*.; basil; *Ocimum basilicum*; orange; *Citrus sp.,* cumin; *Cuminum cyminum*, garlic; *Allium sativum*, marjoram; *Majorana hortensis,* geranium; *Plergonium graveolens*. Also, two products; eugenol (a synthetic product of Winlab, England) and menthol (a natural product of El-Gomhoria Chemicals Co., Egypt). Besides, a mixture of eugenol, geranium and basil (1 : 1 : 1) were tested.

B. Lab. Screening of selected materials against *Varroa*:

The method of Koeniger & Fuchs (1989), with some modification, was used as follows: For each material tested, 10 alive local hybrid worker bees (*Apis mellifera carnica*) naturally infested with *Varroa* were kept inside each of 3 replicate plastic jars (15 cm ht. x 8 cm dia.). A cardboard (2 x 2 x 0.4 cm) impregnated with 0.1 ml of the test substance was hung inside the jar by means of a suitable wire. Each jar was covered with a plastic cup provided with a screen mech (6cm dia.) above the bottom that allows dead mites to fall through to a sticky paper below. The dead bees or mites were daily counted for 3 days. The confirmed bees were fed on sugar syrup (2:1) by a sponge placed in a suitable plastic cup used as cover for the jar. Three jars with untreated bees were used as control.

C. Field treatments:

Evaporation tests: Cardboard plates (4 x 4 x 0.4 cm) impregnated with 2 ml of each oil were placed on top of the combs.

Feeding tests: 1 ml of the tested oil was mixed well with a pollen supplement (2:1 wheat germ and honey). A cake of 100 g /colony was provided on top of combs and replaced evey 13 days. The treatments were repeated 4 times with weekly interval. Three replicate colonies (Langstroth's hives) were employed for each treatment, as well as untreated control.

D. IPM program and techniques:

An IPM program alone or combined with essential oils or oils plus organic acids were tested. Thirty naturally *Varroa*-infested colonies were tested and were divided into 10 groups i.e. 3 replicates / test, including control group. These groups were treated as follows:

Group1, IPM: Queens were caged, in their colonies, under hemispherical screen cages. The colonies were provided with cardboards impregnated with formic acid 60% (10 ml in 7x15x0.4 cm band), basil oil (1ml oil in 2 x 2 x 0.4 cm band), geranium oil (1 ml in 2 x 2 x 0.4 cm band), and oxalic acid 3% (2 ml/comb spray on bees) with seven day intervals after each treatment, while after the first three weeks the queens were released.

Group 2: Basil oil only. Group 3: Geranium oil only. Group 4: 60% formic acid only. Group 5: 3% oxalic acid. Group 6: Formic + basil. Group 7: Formic + geranium. Group 8: Oxalic + basil. Group 9: Oxalic + geranium. Group 10: Untreated (Control). Cardboards were placed on top of mid combs.

Evaluation of the tested materials and techniques was based on the efficiency (%) was evaluated according to Henderson & Tilton (1955) equation:

$$\% \text{ reduction of infestation} = 100 \times 1 - \{ T_a \times C_b \} / \{ T_b \times C_a \}$$

Where:

T = % infestation of treated mites and C = % infestation of untreated mites (a = after; b = before treatment).

Efficiency was measured using the following parameters:

1. Mite fall: Counts of dropped mites were recorded after 1, 7, 14, 21 & 30 day**s** of the application using plastic sheets (51.5 x 36.5 cm) coated with vaseline placed on the hive bottom board.

2. Brood measurement: Sealed worker brood areas (SWB) were recorded at 13-day intervals using a plastic sheet divided into square inches, converted cm^2 when multiplied by 2.54.

3. Infestation percent: The *Varroa* infestation (%) on worker bees and on brood were recorded before and after treatments as follows:

a) On adult bees: About 100 bees were collected, from combs with open brood, and dipped in water to which a detergent was added. The bees were collected in a wire net, and removed after shaking several times. Fallen mites found on bottom of the container (Ritter, 1981) were counted. The exact number of bees was counted as well and the infestation percentage (INFP) was calculated as No. of mites / No. bees x 100

b) On sealed brood: An area of 5 x 5 cm in the middle of a worker comb was selected. The cells were scratched and all stages of *Varroa* females in each cell were counted.

Data collected was statistically analyzed. Treatment means were compared at 5% probability levels by LSD test (Snedecor and Cochran, 1967).

Results and Discussion:

A. Winter control:

1. Infestation on brood:

Data, (Table 1) indicated that all the tested oils reduced the infestation with different rates. Infestation decreased from 37.33% to 14.00%, 32.67% to 26.67%, 40.00% to 27.00 % and 39.00% to 17.33 % with 64.0%, 21.4%, 35.0% & 57.2% reductions in colonies treated with basil, geranium, eugenol and a mixture, respectively.

2. Infestation on adult bees:

Infestation on adult worker bees (Table 1 also) decreased from 16.00 to 7.53%, 14.67% to 4.60%, 14.07% to 6.27% and 13.50% to 6.40% (reductions were 66.6%, 77.7%, 68.4%& 66.3%) in colonies treated with basil, geranium, eugenol and mixture, respectively. The higher % reduction of mites that occurred on bees compared to brood may be due to the direct exposure of mites to the released volatiles used, whereas mites on sealed brood get low doses due to slow release. In this respect, Chiesa (1991) found 96.77% reduction in mite population with thymol treatment. Abou-Zaid and Ghoniemy (1993), in Egypt, stated that infestation was reduced from 45.7% to 21.2% (61.28 % reduction) and from 38.6% to 28.2% (39.20 % reduction) in colonies treated with wormwood and cumin, respectively. Eshabah *et al*. (1995) found 35% - 45% reduction in infestation when sprayed chamomile, thyme and

pudding-pipe extracts, while dusting of neem and chamomile reduced infestation by 34% - 39%. Allam (1999) recorded 71.8% & 64.3%, 62.5% & 65.0% and 62.6% & 55.35 efficacy for clove, bay and origanum sprayed during Aug.-Sept. in two successive seasons, respectively, while somking eucalyptus, pepper mint and clove oil gave variable range (45.5%-92.2%). Haggag and El-Badawy (1999) reported 89.6% - 94.1% reduction in infested brood after the 3rd treatment with thymol, camphor, garlic and thymol+garlic, respectively. Ibrahim and Shoreit (1999) recorded 20.5%, 35.4%, 24.8% & 37.0% reduction after the 4th dusting with coriander, caraway, chamomile and neem, respectively.

3. Effect of treatment on mite fall:

The number of fallen mites / colony after treatment averaged 35.27, 42.27, 45.93, 36.27& 21.80 in basil, geranium, eugenol, mixture and control, respectively. The highest significant mite fall was observed after two weeks of treatment being 55.33, 88.00, 80.33, 71.33& 40.33 mites/colony for the previous treatments, respectively. On the other hand, the lowest mite fall was observed after one day of treatment (18.33, 19.00, 29.67, 13.33 & 7.33 mites/colony, respectively) which may be due to the slow effect of tested oils. Averages of mite fall were 37.33, 41.67, 42.33, 14.67 & 10.33 after one week, 51.67, 37.00, 41.33, 39.00 & 21.00 after 3 weeks and 13.67, 25.67, 36.00, 43.00 & 30.00 mites/colony after 4 weeks respectively (Table 1).

However, the present findings disagreed with those of Chiesa (1991) who found high mite fall in the hours immediately following each application influenced by outdoor temperatures. High mite fall was also reported by Egyptian researchers (Abou-Zaid and Ghoniemy, 1993) and this may be due to high intitial infestation. On the other hand, the present findings, in general, agreed with those of Rezk and Gadelhak (1997) who reported that the total cumulative numbers of fallen mites (after a month) were 54.33, 15.67, 10.67& 33.67 and 37.33, 14.0, 16.33 & 30.67 for eucalyptus, peppermint, clove and chamomile treatments in two treated apiaries, respectively.

4. Effect of treatment on brood rearing activity:

The obtained data in Table 1 showed that sealed worker brood areas (SWB) started 310.67, 322.00, 347.33, 296.00 & 370.00 cm^2 in colonies given basil, geranium oils, eugenol, mixture and control, rspectively. A disturbance was noticed in brood rearing, after about four weeks in eugenol-treated colonies, since SWB averaged 617.33, 758.33, 218.33, 509.00 & 999.00 cm^2 in the same colonies, respectively. It was shown that eugenol was the most material which limited brood rearing activity. After about six weeks the brood build up was

normal in treated colonies except in eugenol-treated colonies, which had the lowest brood area compared to other treatments or control where SWB averaged 1009.33, 1226.67, 107.00, 1256.67 & 1046.67 cm^2, respectively.

5. Effect of treatment on mortality of adult bees:

The average numbers of dead worker bees were 1.00, 2.67, 10.00, 0.00 & 1.33 /colony for basil, geranium, eugenol, mixture treatments and control, rspectively (Table 1). The average number of dead bees was significantly high in case of eugenol but with harmful impact on adult bees and also on brood. The highest bee mortality, for eugenol, was observed after one day, 1^{st} , 2^{nd}, 3^{rd} & 4^{th} weeks being 10.00, 16.67, 52.33, 55.33 & 47.33 /colony compared to low numbers in other treatments, which showed less bee mortalities between 0.00 – 5.67 /colony, or the control being 1.33, 0.67, 1.33, 1.67 & 0.33 /colony for the same periods, respectively.

B. Spring control:

To evaluate the effect of basil and geranium oils as a long-term potential *Varroa* control agents, each oil was mixed with a pollen supplement (PS) meanwhile, enhances colony building up.

1. Infestation on brood:

Data presented in Table 2 showed that the infestation decreased from 26.00% to 5.67% and from 22.00% to 1.33% in colonies treated with basil+PS and geranium+PS, with 81.8% & 95.0% reductions in infestation, respectively. In this concern, Hassan (1998) reported that infestation was reduced after feeding colonies a pollen substitute contained coriander honey, eucalyptus honey, sugar solution compared to control. Sadov (1981) reported that pine essence added to a honey+sugar paste, candy or sugar syrup in winter and early spring increased brood rearing and honey harvest (15%-50%).

2. infestation on adult bees:

The infestation (Table 2) decreased from 13.13% to 8.77% and from 5.67% to 0.73% in colonies treated with basil+PS and geranium+PS; reduction in mite infestation averaged 60.0% & 92.3%, respectively.

3. Effect of treatments on mite fall:

Averages of mite fall were 100.67, 69.00 & 65.00 /colony for geranium+PS, basil+PS and control, respectively.

4. Effect of treatments on brood rearing activity:

The obtained data (Table 2) indicated that SWB areas started 1058.00, 875.33, 1421.00 cm^2 / colony for those given basil+PS, geranium +PS and PS, respectively. At the end of the test, averages were 1550.33, 1460.67& 1749.11 cm^2 / colony with 40.36%, 34.15% & 52.56 % insignificant increase, for the same treatments, respectively. It is worth noting that brood rearing in treated colonies was not highly affected when adding essential oil to PS, except some repellency of basil to bees in some cases.

C. Autumn control (IPM):

1. Infestation on brood:

Data presented in Table (3) showed that all the tested treatments reduced the infestation compared to control. Infestation level decreased significantly from 4.77%, 3.33%, 2.33%, 1.33%, 3.00%, 9.33%, 2.33%, 11.67%& 5.33% to 2.33%, 1.67%, 1.33%, 0.33%, 0.60%, 1.00%, 0.30%, 1.33% & 0.67% for IPM, geranium (G), basil (B), geranium+formic (GF), geranium+oxalic (GO), basil+formic (BF), basil+oxalic (BO), formic (F) and oxalic (O), respectively. The reduction averaged 72.6%, 71.9%, 68.0%, 86.1%, 88.8%, 94.0%, 92.8%, 93.6%& 92.9% for the same treatments, respectively.

2. Infestation on adult bees:

The obtained data (Table 3) explained that infestation decreased significantly from 5.10%, 5.03%, 2.57%, 3.27%, 4.10%, 5.17%, 1.80%, 3.80% & 3.67% to 0.40%, 2.37%, 1.47%, 0.83%, 0.00%, 1.77%, 0.37%, 1.10% & 0.43% (reduction averaged 95.3%, 71.6%, 65.6%, 84.7%, 100.0%, 79.4%, 87.6%, 82.6%& 92.9%) for IPM, G, B, GF, GO, BF, BO, F and O, respectively.

However, the present findings are, in general, agreed with those of Egyptian authors; Abo-Zaid and Ghoniemy (1992) who reported that oxalic acid treatment reduced infestation from 33.6% to 7.7%. The same authors (1993) reported that infestation was reduced from 51.60% to 7.45% (89.40% reduction) and from 41.82% to 10.90% (79.00% reduction) in two Egyptian governorates (Qalubia and Fayoum), respectively after four treatments with 60% formic acid. Also, Ghoniemy (1998), in Fayoum, recorded 77.53%, 69.63% & 39.62% reduction with formic acid in cardboard plates in different applications. El-Shaarawy (1999) recorded 84%, 77% & 75% efficacy at Qalubia gov. and 85%, 80% & 74% at Giza gov. for different Apiguard® treatments and 60% for formic acid. On the other hand, Mutinelli *et al*.(1996) found variable efficacies; 49.2% - 98.8%, 15.0% - 83.2%, 3.0% - 98.3% & 51.5% - 68.7% for formic acid, lactic acid, oxalic acid and Apilife-VAR®, respectively. Nanetti (1999) showed that 4.2% oxalic acid solution in 60% sucrose resulted in a remarkable effectiveness

(80%-92%) against *Varroa* mites in broodless colonies treated in the autumn/winter. Imdorf *et al.* (1999) reported 37%-96% efficacy for formic acid. They attributed this variable range to the method of application, hive type, acid concentration and climatic conditions. Allam (1999) suggested an IPM program for *Varroa* control including selection for tolerant bees, using natural materials such as mint, camphor leaves, and a mixture, exposing colonies to sunlight, destroying drone brood and strengthen colonies, and using volatile oils during suitable times. Sammataro *et al.* (2000) tested mite-tolerant queen lines, screens prevent fallen mites from climbing back on to comb, an essential oil, EO, (thymol mixture), and combinations of queens and EO, queens and screens, and screens and EO. They found that EO caused a significant quick drop of mites, while the queen/screen combination had the lowest one. Also, they added that colonies located in open field had significantly fewer mites than sheltered ones. Rice and Winston (2001) compared an IPM treatment (thymol + modified bottom boards + hygienic queens) to Apistan. They showed that all of the systems maintained mite populations to levels below an economically important threshold.

3. Effect of treatments on mite fall:

At the end of the treatment, mite fall averaged 24.33, 20.31, 28.33, 44.31, 49.13, 46.43, 27.37, 35.22, 100.41& 15.38 /colony for IPM, G, B, GF, GO, BF, BO, F and O, respectively. After one week, the highest mite fall averaged 433.0 /colony for O, while the lowest one was 6.0 /colony for G, however GO averaged 202.3 /colony with significant difference. The other treatments showed variable mite fall ranged between 12.7 - 87.0 /colony (Table 3).

In this respect, Egyptian authors reported gradual mite fall after treating colonies with organic acids or volatile oils and recorded variable ranges (Ghoniemy and Abo-Zaid, 1993, Ghoniemy, 1998 & El-Shaarawy, 1999).

4. Effect of treatments on brood rearing activity:

Data in Table (3) indicated that SWB areas started 1281.33, 1115.67, 1071.00, 1420.33, 804.67, 1534.67, 466.33, 715.67, 608.00 & 823.00 cm^2 / col. in IPM, G, B, GF, GO, BF, BO, F, O and (C), respectively. At the end of the test averaged 482.04, 399.33, 455.96, 426.88, 400.70, 338.38, 273.54, 309.17, 419.96 & 363.38 cm^2 / col. for the same treatments, respectively. The decrease in SWB in case of formic or oxalic may be due to their noticeable adverse effect on brood rearing and, also, to synergetic effect of organic acids and oils. Also, average SWB area recorded 0.00 cm^2 twice in the 3^{rd} and 4^{th} measurements in IPM-treated colonies and that due to caging of queens for 21 days to minimize mite population by

depriving female mites to reproduce in brood cells, but this procedure obviously caused queen damage or loss in some cases. In this concern, many researchers reported low brood area after treating with chemicals or organic acids (Alekseenok & Shutov, 1986 and Schulz, 1993).

5. Effect of treatments on mortality of adult bees:

As indicated in Table (3) dead bees averaged 0.85, 2.48, 5.89, 0.77, 1.04, 0.29, 0.59, 1.85, 1.37& 1.29 / colony for IPM, G, B, GF, GO, BF, BO, F, O and C, respectively. Significant average was recorded in colonies given B (4.0 & 22.0 /colony) after one and two weeks, respectively. After three weeks, the G treatment showed higher bee mortality (15.0 /colony) than other treatments or control during test period.

In this regard, these findings agreed with those of Alekseenok and Shutov (1986) who found that the number of dead bees were 1.8, 4.1& 5.4 for colonies treated with KAS-81(a mixture containing volatiles), Folbex and control, respectively. Comparing KAS-81 to formic acid, the number of dead bees were 1.4 & 1.9, respectively. On contrary, Moosbeckhofer (1993) mentioned that colonies treated with volatiles were adversely affected and produced 20% less honey than control. He added that 50% of bees had died by the end of winter compared to 10.7% in colonies treated with pyrethroid strips. Schulz (1993) stated that brood was damaged in 37 colonies given Apilife-VAR®.

Also, many researchers recorded some adverse effects on bees after volatile treatments; Lensky *et al*. (1996) found that the use of pure origanum oil or 30% thymol during summer was harmful to bees depending on dose and ambient temperature. This result was also found by Chiesa (1991) and Gal *et al*. (1992). Mattila and Otis (1999) showed that honey production was reduced by 30% during the Apiguard® treatment. On contrast, Mutinelli *et al.* (1996) reported low or absent bee mortality in all tests of formic acid, lactic acid or Apilife-VAR®, and also El-Shaarawy (1999) claimed that honey yield increased after colonies treated with Apiguard® or formic acid.

Conclusions and Recommendation:

The use formic acid, oxalic acid and essential oils such as geranium and basil alone or combined an IPM program is to control *Varroa* mites is recommended. These materials are inexpensive, efficient and relatively less toxic compared to very expensive and toxic acaricides presently used by beekeepers. No danger of honey contamination occurs if such treatments are carried out, properly, outside the period of production or storage of honey in the hive. In spite of reduced brood rearing in some cases, colonies could restore their normal strength gradually. It is worth mentioning that formic acid should not be used with weak

colonies. Moreover the present study showed that colonies received such treatment were severely damaged and most probably collapse thereafter, also eugenol exhibited lethal effects to bees and brood.

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الملخـــص العربـــــى

مكافحة حلم الفاروا فى نحل العسل بالزيوت النباتية، بمفردها أو من خلال برنامج مكافحة متكاملة

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قسم وقاية النبات - كلية الزراعة بالفيوم - مصر

اختبر أحد عشر نوعا من الزيوتً الطّيّارة معمليا ضد الفاروا. وكانت مـن نباتـات النعنـاع البلدى، الزعتر، الكافور، الريحان، البرتقال، الكمون، الثوم، البردقوش، العتر البلدى، بالإضـافة إلـى الإيوجينول والمنتول. وقد وجد أن أفضلها من حيث التأثير على الفاروا هى زيـوت العتـر البلـدى والريحان ومشتق الإيوجينول، والتى تم اختيارها لمعاملة طوائف النحل المصابة فى الشتاء، والربيـع ،والخريف وذلك باستخدامها بمفردها أو بإضافتها إلى بديل حبوب اللقاح أو مع حمضـى الفورميـك والأوكساليك والتقفيص على الملكات ولتحديد كفاءة هذه المواد، فقد تم دراسة تأثيرهـا علـى معـدل الإصابة بالطفيل على الحضنة والنحل البالغ وتساقط أفراد الطفيل وإنتاج الحضنة، وكذلك التأثير على حياة النحل البالغ. وقد اوضحت نتائج هذه الدراسة أن كل هذه المعاملات كانت فعّالـة معنويـا ضـد الفاروا فى الطوائف المعاملة مقارنة بالغير معاملة. كما كان تساقط أفراد الطفيل كبيـرا أيضـا فـى الأولى. وقد لوحظ بعض التأثيرات العكسية مثل انخفاض مساحة الحضنة وموت بعض النحل بنسـبة قليلة جدا وذلك فى بعض المعاملات خاصة فى الطوائف التى عوملت بالإيوجينول. واتضـح أيضـا أهمية التوصية بمكافحة هذا الطفيل من خلال برنامج متكامل.

Table 1. Efficiency of some volatile oils against *Varroa* mites during winter.

Treatments	No. of dropped mites after periods given						%*Varroa* on brood		%*Varroa* on bees		%Reduction on	
	1 day	1 week	2 week	3 week	4 week	Mean	BT Feb. 2	AT Mar. 3	BT Feb. 2	AT Mar. 3	Brood	Bees
Basil	18.33	37.33	55.33	51.67	13.67	35.27	37.33	14.00	16.00	7.53	64.0	66.6
Geranium	19.00	41.67	88.00	37.00	25.67	42.27	32.67	26.67	14.67	4.60	21.4	77.7
Eugenol	29.67	42.33	80.33	41.33	36.00	45.93	40.00	27.00	14.07	6.27	35.0	68.4
Mixture	13.33	14.67	71.33	39.00	43.00	36.27	39.00	17.33	13.50	6.40	57.2	66.3
Control	7.33	10.33	40.33	21.00	30.00	21.80	26.33	27.33	13.63	19.20		
$LSD_{5\%}$	**10.52**	**9.67**	**11.62**	**15.84**	**23.63**		**12.59**	**9.56**	**n.s.**	**2.80**		

Table 1. Continued. Effect of treatments on brood rearing and bee mortality.

Treatments	Sealed worker brood area (cm^2)					No. of dead bees after treatments					
	Feb. 2	Feb.14	Feb. 7	Mar. 3	Mean	1 day	1 week	2 week	3 week	4 week	Mean
Basil	310.67	473.33	617.33	1009.33	602.67	1.00	1.00	1.00	2.67	5.67	2.27
Geranium	322.00	403.00	758.33	1226.67	677.50	2.67	2.67	2.00	3.67	0.00	2.20
Eugenol	347.33	452.33	218.33	107.00	281.25	10.00	16.67	52.33	55.33	47.33	36.33
Mixture	296.00	379.33	509.00	1256.67	610.25	0.00	2.00	2.33	3.33	0.33	1.60
Control	370.00	657.00	999.00	1046.67	768.17	1.33	0.67	1.33	1.67	0.33	1.07
$LSD_{5\%}$	**62.56**	**95.04**	**102.84**	**204.44**		**4.20**	**7.61**	**7.28**	**6.37**	**4.24**	

BT = before treatment and AT = after treatment.

Table 2. Efficiency of tested volatile oils added to a pollen supplement against *Varroa* mites in spring.

Treatments	Sealed worker brood area (cm^2)				No. of dropped mites	% *Varroa* on brood		%*Varroa* on bees		%Reduction on	
	Apr. 16	Apr. 29	May 12	Mean	May 14	B.T.	A.T.	B.T.	A.T.	Brood	Bees
Basil + PS	1058.00	971.67	2621.33	1550.33	69.00	26.00	5.67	13.13	8.77	81.8	60.0
Geranium + PS	875.33	943.67	2563.00	1460.67	100.67	22.00	1.33	5.67	0.73	95.0	92.3
Control (PS)	1421.00	1122.67	2703.67	1749.11	65.00	15.33	18.33	2.83	4.73		
$LSD_{5\%}$	**140.31**	**205.28**	**808.72**		**n.s.**	**2.39**	**7.83**	**1.81**	**2.30**		

PS = a pollen supllement, BT = before treatment, AT = after treatment, and n.s.= not significant.

Table 3. Efficiency of IPM, essential oils and/or organic acids in autumn.

Treatments	%*Varroa* on bees			%*Varroa* on brood			No. of dropped mites									
	22/9	20/10	22/11	22/9	20/10	22/11	29/9	6/10	13/10	20/10	27/10	3/11	10/11	17/11	24/11	Mean
IPM	5.10	1.60	0.40	4.77	0.00	2.33	36.7	8.0	34.0	18.0	5.7	39.3	29.7	31.3	16.3	24.33
Geranium	5.03	2.57	2.37	3.33	2.33	1.67	6.0	6.3	13.7	9.7	15.0	25.0	42.7	24.7	39.7	20.31
Basil	2.57	1.87	1.47	2.33	1.33	1.33	12.7	21.7	18.0	35.0	27.3	35.0	33.0	29.3	43.0	28.33
Geranium +Formic	3.27	2.03	0.83	1.33	1.00	0.33	53.7	39.0	82.7	46.0	18.0	16.3	24.7	76.7	41.7	44.31
Geranium + Oxalic	4.10	0.00	0.00	3.00	1.00	0.60	202.3	52.0	31.7	17.3	6.3	6.7	56.3	32.3	37.3	49.13
Basil+ Formic	5.17	3.23	1.77	9.33	1.67	1.00	87.0	45.3	56.0	54.3	19.3	21.3	35.0	80.7	19.0	46.43
Basil+ Oxalic	1.80	0.67	0.37	2.33	0.33	0.30	37.3	39.7	31.7	12.3	7.0	5.0	55.0	42.7	15.7	27.37
Formic	3.80	1.50	1.10	11.67	3.00	1.33	43.7	41.3	38.7	83.0	7.3	16.0	18.0	57.7	11.3	35.22
Oxalic	3.67	2.67	0.43	5.33	1.67	0.67	433.0	108.7	67.3	50.3	39.0	19.0	78.7	62.7	45.0	100.41
Control	5.30	6.27	8.80	7.67	12.67	13.67	36.7	15.0	12.7	19.3	10.7	19.0	6.3	10.0	8.7	15.38
$LSD_{5\%}$	**2.30**	**1.25**	**1.39**	**1.93**	**1.91**	**2.94**	**34.9**	**22.0**	**18.0**	**13.9**	**7.4**	**8.1**	**16.2**	**16.6**	**17.0**	

Table 3. Continued. Effect of treatments on reduction of mites and on bee mortality.

Treatments	%Reduction		No. of dead bees after treatments									
	On bees	On brood	29/9	6/10	13/10	20/10	27/10	3/11	10/11	17/11	24/11	Mean
IPM	95.3	72.6	0.67	0.00	0.00	0.67	0.33	0.33	3.67	2.00	0.00	0.85
Geranium	71.6	71.9	0.67	1.00	15.00	0.00	1.00	4.33	0.00	0.00	0.33	2.48
Basil	65.6	68.0	4.00	22.00	8.67	6.67	0.33	6.00	0.00	3.00	2.33	5.89
Geranium+Formic	84.7	86.1	0.00	3.00	2.00	0.33	0.67	0.67	0.00	0.00	0.33	0.77
Geranium+Oxalic	100.0	88.8	0.00	3.33	0.33	1.33	0.33	0.33	2.00	1.67	0.00	1.04
Basil + Formic	79.4	94.0	0.00	0.00	0.33	0.00	0.00	1.33	0.00	1.00	0.00	0.29
Basil + Oxalic	87.6	92.8	0.00	1.00	0.33	0.00	0.67	2.67	0.00	0.67	0.00	0.59
Formic acid	82.6	93.6	1.00	5.00	2.00	2.67	0.33	3.67	0.67	1.33	0.00	1.85
Oxalic acid	92.9	92.9	0.00	5.33	0.00	0.67	0.67	3.67	0.00	1.33	0.67	1.37
Control			1.33	1.00	2.33	0.00	0.00	4.67	0.00	2.33	0.00	1.29
$LSD_{5\%}$			**0.21**	**3.52**	**2.11**	**1.55**	**1.54**	**3.19**	**0.91**	**2.29**	**0.85**	

Table 3. Continued. Effect of treatments on brood rearing activity.

Treatments	Sealed worker brood area (cm^2)								
	22/9	5/10	18/10	31/10	13/11	26/11	9/12	22/12	Mean
IPM	1281.33	570.00	0.00	0.00	322.67	402.33	596.33	683.67	482.04
Geranium	1115.67	271.67	612.00	516.33	214.00	133.33	147.67	184.00	399.33
Basil	1071.00	438.67	719.67	375.00	364.00	75.00	145.00	459.33	455.96
Geranium+Formic	1420.33	509.67	557.33	400.33	187.67	68.00	62.67	209.00	426.88
Geranium+Oxalic	804.67	369.00	471.67	653.33	242.33	110.33	218.33	336.00	400.70
Basil+ Formic	1534.67	216.67	263.33	130.00	38.67	157.33	133.33	233.00	338.38
Basil+Oxalic	466.33	298.33	397.67	404.33	159.00	116.33	132.33	214.00	273.54
Formic	715.67	326.00	468.33	181.00	141.33	115.00	213.33	312.67	309.17
Oxalic	608.00	161.67	761.00	353.00	164.33	444.67	440.33	426.67	419.96
Control	823.00	598.67	385.33	265.33	243.00	209.00	257.67	125.00	363.38
$LSD_{5\%}$	**94.31**	**87.88**	**72.83**	**61.56**	**47.68**	**48.83**	**41.46**	**74.93**	