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Evaluation of some natural oils and formic acid for controlling varroa mite (Varroa destructor) in honey bee colonies

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Abstract

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Keywords

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*Varroa destructor* is a dangerous pest directly for beekeeping and indirectly for crops that require insect pollination. V. destructor spread from the Asian honey bee (Apis cerana Fabricius) to Apis mellifera L. (Hymenoptera : Apidae). Mites feed on haemolymph of brood and adult bees cause colony weakness, a decrease in brood and bees. The application of integrated pest management (IPM) techniques is more recent development in V. destructor control because its more effective than pesticides. This study was carried out to assess the efficacy of three natural oils (Camphor oil -mint oil - anise oil) and formic acid (65%) used at three concentrations (25%, 50% and 75%) against V. destructor in honey bee colonies (A. mellifera) during the period of January till September 2021 in Sidi Salem, Kafr El-Sheikh Governorate, Egypt. Data indicated that formic acid and the highest concentrations (75%) of tested essential oils caused effective control of varroa mites, whereas the infestation reduction percentage with formic acid, camphor oil recorded more than 80% after treatments on both brood and adult. while anise oil recorded the lowest one.

#### Introduction

The varroa mite (Varroa destructor) is an ectoparasitic mite on honey bee brood and adults .it considered one of the most pests in honey bee colonies (Sammataro et al., 2000 and Anderson and Trueman, 2000). It causes big losses to honey bee , Apis mellifera L. (Hymenoptera : Apidae) and great economic loss to the worldwide beekeeping industry (Rashid et al., 2012) and also, reduces the colony ability to pollinate plants (De Jong et al., 1984), the varroa mite is considered as the parasite with the most pronounced economic impact on the beekeeping industry (Guzman-Novoa *et al.*,2010). There is current concern about contamination of bee products with synthetic substances against the varroa (Howis and Nowakowski, 2009). The mite is an ectoparasite, that transfers entomopathogens such as viruses, thus causing death of brood and adult bees in colonies (De Jong *et al.*,1982). Natural products such as essential oils and their components or organic acids, especially formic acid and citric acid were used for controlling varroa mites (Mutinelli *et al.*,1997).

Formic acid and thymol are effective in the control of varroa mites without any side effects (Soroker *et al.*, 2019). In a similar way, smoke from plants has produced mixed success in increasing varroa reduction. Varroa mites need to be controlled because untreated colonies collapsed within a few years due to damage to both brood and adult bees (Elzen et al., 2000). In previous studies, plant essential oils have been examined and applied as alternatives to synthetic pesticides for varroa management (Rosenkranz et al., 2010 and Plettner et al., 2017). These allow circumstances for the development and testing of the effectiveness of new preparations (Gurgulova et al., 2004). Little is known about the electrophysiological detection of essential oils and essential oil components by acarine (Soroker et al., 2019).

The aim of the present study is to evaluate some natural products for controlling varroa mite on honey bees, these natural materials generally have no side effects on honey bees and are less hazardous to beekeepers.

# Materials and methods

### 1. Experimental site:

The experiment was carried out in an apiary at Sidi Salem district, Kafr El- Sheikh Governorate during the period from January to September 2021 to study the effect of some natural products for controlling *V. destructor* in honey bee colonies. Whereas the study required eighteen colonies of carniolan hybrid nearly of equal strength. The colonies have been divided into 6 groups (Each group 3 colonies).

#### 2. Materials:

- **2.1.** Camphor oil (25%-50% -75%)
- **2.2.** Mint oil (25% -50% -75%)
- **2.3.** Anise oil (25% -50% -75%)
- **2.4.** Formic acid (65%)

The bottom board of the hive was covered with a plastic sheet coated with Vaseline to capture the fallen mites.

# 3. Determination of varroa infestation:

The percent infestations of varroa mite on adult before and after treatments were determined according to Korneili (1988), whereas the number of mites\100 was calculated and the fallen varroa on the plastic sheet was counted periodically every 3 days till the end of treatment. The percent infestations of varroa mites on brood cells was determined by square inch. Reduction percentage in mite infestation was calculated according to Henderson and Tilton (1955)

Reduction % =

Treatment before Control after **4. Statistical analysis:** 

Data collected were statistically analyzed according to SAS Institute (1998) computer program.

#### **Results and discussion**

Three natural oils namely (Camphor oil, mint oil and anise oil) and formic acid were evaluated against V. destructor under field conditions. Data in Tables (1 and 2) showed that the reduction percentage of varroa mite infestation on adult was clearly reduced after the treatments in all tested oils and formic acid. In the colonies treated with essential oils and formic acid the reduction percentage of infestation with varroa on adult reduced gradually from the first treatment to the fourth (End of treatment) after the treatments. formic acid 65% and camphor oil caused a highly effective in controlling varroa mites, whereas the reduction percentage of infestation of Formic acid, camphor oil, mint oil and anise oil in January recorded (88.5%), (87.2%), (72.1%), (61.7%) after treatment on adult, respectively. Whereas, anise oil was the lowest percentage of infection reduction. The reduction percentage of infection of formic acid, camphor oil, mint oil and anise oil in September was recorded (94.5%), (89.6%), (65.3%),

(56%), respectively. The obtained data are in agreement with Allam *et al.* (2003) who found that formic acid killed 91.7% of varroa mites and Abd El-Wahab and Ebada (2006) and Hamaad *et al.* (2008) found that thyme oils resulted in 65.9% of varroa mite mortality for varroa control under Egyptian conditions. Data in Tables (1 and 2) showed that the highest number of varroa falling on the sheet was recorded after the first treatment and clearly decreased until the end of treatments.

| Table (1): The reduction percentages of infection on adult in honey bee colonies treated with essential oils |  |              |     |   |  |     |  |  |  |
|--|--|--------------|-----|---|--|-----|--|--|--|
| and formic acid in January 2021.   |  |              |     |   |  |     |  |  |  |
|  |  | <b>a</b> , 1 | D A | G |  | 1.0 |  |  |  |

| and formic acid in January 2021. |                |         |       |                          |  |     |     |                         |             |
|----------------------------------|----------------|---------|-------|--------------------------|--|-----|-----|-------------------------|-------------|
| Date                             | Treatment      | Control |       | Before<br>treat-<br>ment | Concentrations<br>No of fallen of varroa |     |     | After<br>treatm-<br>ent | Reduction % |
| January                          |                | Before  | After | Adult                    | %25                                      | %50 | %75 | Adult                   | Adult       |
| First<br>treatment               |                | 32      | 28    | 28                       | 22a                                      | 28b | 30c | 7                       | 71.4        |
| Second                           | Camphor        | 30      | 27    | 26                       | 25b                                      | 26b | 27b | 5                       | 78.6        |
| Third                            | oil            | 27      | 25    | 20                       | 20a                                      | 20a | 10a | 3                       | 83.8        |
| Fourth                           |                | 23      | 20    | 18                       | 7a                                       | 5a  | 3a  | 2                       | 87.2        |
| First                            |                | 32      | 28    | 30                       | 23a                                      | 25b | 28b | 10                      | 61.9        |
| Second                           |                | 30      | 27    | 28                       | 20a                                      | 23a | 25a | 8                       | 68.3        |
| Third                            | Mint oil       | 27      | 25    | 26                       | 15a                                      | 18a | 16a | 6                       | 75.1        |
| Fourth                           |                | 23      | 20    | 21                       | 13a                                      | 12a | 11a | 5                       | 72.6        |
| First                            |                | 32      | 28    | 27                       | 20a                                      | 21a | 22a | 12                      | 49.2        |
| Second                           |                | 30      | 27    | 25                       | 18a                                      | 20a | 20a | 10                      | 55.6        |
| Third                            | Anise oil      | 27      | 25    | 23                       | 17a                                      | 18a | 19a | 8                       | 62.4        |
| Fourth                           |                | 23      | 20    | 21                       | 15a                                      | 14a | 12a | 7                       | 61.7        |
| First                            | ]              | 32      | 28    | 24                       | 30c                                      | 30c | 30c | 6                       | 71.5        |
| Second                           | Formic<br>acid | 30      | 27    | 22                       | 28b                                      | 28b | 28b | 4                       | 79.8        |
| Third                            |                | 27      | 25    | 20                       | 15a                                      | 15a | 15a | 3                       | 83.8        |
| Fourth                           |                | 23      | 20    | 20                       | 2a                                       | 2a  | 2a  | 2                       | 88.5        |

| Table (2): The reduction perce | ntages of infection | on adult in honey | bee colonies trea | ated with essential oils |
|--------------------------------|---------------------|-------------------|-------------------|--------------------------|
| and formic acid in September   | 2021.               |                   |                   |                          |

|           |                | Control |       | Before  | Concentrations         |     |     | After   |            |
|-----------|----------------|---------|-------|---------|------------------------|-----|-----|---------|------------|
| Date      | Treatment      |         |       | treatm- | No of fallen of varroa |     |     | treatm- | Reduction% |
|           | Treatment      |         |       | ent     |                        |     |     | ent     |            |
| September |                | Before  | After | Adult   | %25                    | %50 | %75 | Adult   | Adult      |
| First     |                | 30      | 26    | 26      | 25b                    | 27b | 29c | 6       | 73.4       |
| treatment | ~ •            |         |       |         |                        |     |     |         |            |
| Second    | Camphor        | 28      | 25    | 24      | 23a                    | 26b | 27b | 4       | 81.4       |
| Third     | oil            | 27      | 24    | 22      | 20a                    | 24a | 15a | 3       | 84.7       |
| Fourth    |                | 22      | 20    | 21      | 8a                     | 6a  | 2a  | 2       | 89.6       |
| First     |                | 30      | 26    | 28      | 24a                    | 26b | 28b | 12      | 50.6       |
| Second    |                | 28      | 25    | 24      | 22a                    | 24a | 26b | 10      | 53.4       |
| Third     | Mint oil       | 27      | 24    | 22      | 17a                    | 18a | 14a | 8       | 59.1       |
| Fourth    |                | 22      | 20    | 19      | 14a                    | 13a | 10a | 6       | 65.3       |
| First     |                | 30      | 26    | 26      | 20a                    | 23a | 25b | 13      | 42.4       |
| Second    |                | 28      | 25    | 23      | 18a                    | 20a | 22a | 11      | 46.5       |
| Third     | Anise oil      | 27      | 24    | 22      | 17a                    | 19a | 20a | 10      | 48.9       |
| Fourth    |                | 22      | 20    | 20      | 16a                    | 14a | 13a | 8       | 56         |
| First     |                | 30      | 26    | 27      | 29c                    | 29c | 29c | 7       | 70.1       |
| Second    | Formic<br>acid | 28      | 25    | 25      | 28b                    | 28b | 28b | 5       | 77.6       |
| Third     |                | 27      | 24    | 23      | 12a                    | 12a | 12a | 3       | 85.4       |
| Fourth    | aciu           | 22      | 20    | 24      | 2a                     | 2a  | 2a  | 2       | 94.5       |

Data in Tables (3 and 4) showed that the reduction percentages of infection on brood in colonies treated with formic acid and essential oils in January recorded (86.3% formic acid), (81.5% camphor oil), (65.7% mint oil) ,(56.7% anise oil ) and recorded in September the reduction of infection of formic acid, camphor oil, mint oil and anise oil (86.9%), (82.1%), (63.5%) ,(56.2%) after treatment, respectively. Results showed that camphor oil is effective against *V. destructor* and safe **Table (3): The reduction percentages of infecti**  for bees. Several authors have evaluated essential oils as control agents for varroa (Imdorf *et al.*, 1999; Ariana *et al.*,2002 and Ismail *et al.*, 2006). Data agreement with May-Itza *et al.* (2007). Statistical analysis showed that highly significance between treatments.

| Table (3): The reduction percentages of infection on brood in honey bee colonies treated with |
|---|
| essential oils and formic acid in January 2021.   |

| Date      | Treatment | Control |       | Before<br>Treatment | After<br>Treatment | Reduction % |
|-----------|-----------|---------|-------|---------------------|--------------------|-------------|
| January   |           | Before  | After | Brood               | Brood              | Brood       |
| First     |           | 33      | 30    | 25                  | 11                 | 51.6        |
| treatment | Comphon   |         |       |                     |                    |             |
| Second    | Camphor   | 31      | 27    | 23                  | 10                 | 50.1        |
| Third     | oil       | 28      | 25    | 22                  | 6                  | 59.3        |
| Fourth    |           | 26      | 21    | 20                  | 3                  | 81.5        |
| First     |           | 33      | 30    | 23                  | 12                 | 42.7        |
| Second    |           | 31      | 27    | 22                  | 10                 | 47.9        |
| Third     | Mint oil  | 28      | 25    | 20                  | 7                  | 60.8        |
| Fourth    |           | 26      | 21    | 18                  | 5                  | 65.7        |
| First     |           | 33      | 30    | 27                  | 14                 | 43          |
| Second    |           | 31      | 27    | 25                  | 12                 | 44.9        |
| Third     | Anise oil | 28      | 25    | 23                  | 10                 | 51.4        |
| Fourth    |           | 26      | 21    | 20                  | 7                  | 56.7        |
| First     |           | 33      | 30    | 26                  | 9                  | 62          |
| Second    | Formic    | 31      | 27    | 23                  | 7                  | 65.1        |
| Third     |           | 28      | 25    | 22                  | 5                  | 74.6        |
| Fourth    | acid      | 26      | 21    | 18                  | 2                  | 86.3        |

| Table (4): The reduction percentages of infection on brood in honey bee colonies treated with |
|---|
| essential oils and formic acid in September 2021.   |

| Date      | Treatment      | Control |       | Before    | After     | Reduction |
|-----------|----------------|---------|-------|-----------|-----------|-----------|
|           |                |         |       | Treatment | Treatment | %         |
| September |                | Before  | After | Brood     | Brood     | Brood     |
| First     |                | 30      | 27    | 28        | 10        | 60.4      |
| treatment | Commhon        |         |       |           |           |           |
| Second    | Camphor<br>oil | 29      | 25    | 25        | 8         | 62.9      |
| Third     | 011            | 27      | 23    | 24        | 6         | 70.7      |
| Fourth    |                | 25      | 19    | 22        | 3         | 82.1      |
| First     |                | 30      | 27    | 25        | 11        | 51.2      |
| Second    | Mint oil       | 29      | 25    | 22        | 9         | 52.6      |
| Third     |                | 27      | 23    | 20        | 8         | 53.1      |
| Fourth    |                | 25      | 19    | 18        | 5         | 63.5      |
| First     |                | 30      | 27    | 27        | 13        | 46.6      |
| Second    |                | 29      | 25    | 24        | 11        | 46.8      |
| Third     | Anise oil      | 27      | 23    | 23        | 9         | 54.1      |
| Fourth    |                | 25      | 19    | 21        | 7         | 56.2      |
| First     |                | 30      | 27    | 24        | 8         | 63        |
| Second    | Formic         | 29      | 25    | 23        | 6         | 69.8      |
| Third     |                | 27      | 23    | 22        | 4         | 78.7      |
| Fourth    | acid           | 25      | 19    | 20        | 2         | 86.9      |

References

- Anderson, D. and Trueman, J.W.H. (2000): Varroa jacobsoni (Acari : Varroidae) is more than one species. Exp. Appl. A. Carol., 24:165-189.
- Abd El-Wahab, T.E. and Ebada, M. A. (2006): Evaluation of some volatile plant oils and mavrik against Varroa destructor in honey bee colonies. J. APPI. Sci. Res., 2(8): 514 -521.
- Allam, S.F.; Hassan, M.F.; Rizk, M.
  A. and Zaki, A. U. (2003): Utilization of essential oils and chemical substances alone or in combination against varroa mite, a parasite of honey bees. Insect Pathogens and Insect parasitic Nematodes IOBC WPRS Bulletin,26:273-274.
- Ariana, A.; Ebadi, R. and Tahmaseb,
  G. (2002): Laboratory evaluation of some plant essences to Control Varro a destructor (Acari: Varroidae). Exp. Appi. A Carol., 27(4): 319-327.
- De Jong, D.; Andrea, D.D. and Goncalves, I.S. (1982): A comparative analysis of shaking solution for the detection of *Varroa jacobsoni* on adult honey bees. Apidologie, 13: 297-306.
- De Jong, D.; Goncalves, L. S. and Morse, R. A. (1984): Dependence on climate of the virulence of *Varroa jacobsoni*. Bee World, 65(3):117-121.
- Elzen, P.J.; Baxter, J.R.; Elzen, G. W.; Rivera, R. and Wilson, W.T. (2000): Evaluation of grapefruit essential oils for controlling *Varroa jacobsoni* and *Acarapis woodi*. Amer. Bee. J., 140 (8):666-668.
- Gurgulova, K.; Zhelyazkova, I. and Popova, V. (2004): Metican

against varroatosis among bees. Apiacta, 38:307-316.

- Guzman-Novoa, E.; Eccles, L.; Calvete, Y.; Mcgowan, J.; Kelly, P. G. and Correa-Benetez, A. (2010): Varroa destructor is the main culprit for the death and reduced populations of over wintered honey bee (*Apis mellifera*) colonies in Ontario, Canada. Apidologie, 41:443-450.
- Hamaad, R.F.M.; Eldoksch , H.A.; Abdel-Samed, A.M. and Abdel Moein , N .M . (2008): Effect of essential oils and athymol formulation for controlling Varroa destructor in honey bee (*Apis mellifera*) colonies, Egypt. J. Agric. Res., 86 (3): 951-961.
- Henderson, C.F. and Tilton, E. W. (1955): Test with acaricides against the brown Wheat mite. J. Econ. Entom., 48-157-161.
- Howis, M. and Nowakowski, P. (2009): Varroa destructor removal efficiency using bee vital hive clean preparation. Journal of Apicultural Sciences, 35(2): 15-20.
- Imdorf, A.; Bodanov, S.; Ochao, R.I. and Calderone, N. W. (1999): Use of essential oils for the Control of *Varroa jacobsoni* in honeybee colonies. Apidologie, 30(2-3):209–228.
- Ismail, A.M.; Ghonieny, A. H. and Awayss, A. A. (2006): Combatting honey bee. Varroa mites by plant oils alone or in are IPM program. the 2nd Conference of the Farm Integrated pest Management, 16-18 Jana Fac. Agric., Fayoum Univ., 172-185.
- Korneili, A.B. (1998): the impact of the varroa a mite on Iranian commercial bee keeping- Amer-Beej.,128:423-424.

- Mutinelli, F.; Baggio, A.; Capolongo, F.; Piro, R.; Prandin, L. and Biasson, L. (1997): A scientific note on oxalic acid by topical application for the control of varroosis. Apidologie, 28: 461-462.
- May-Itza, W.J.; Medina, L. A. and Marrufo Olivares, J.C. (2007): Effectiveness of thymol based jells for the control of *Varroa jestructor* mite that infests *Apis mellifera* honeybee colonies, under tropical condition in Yucatan. Mexico Vet. Mex., 38(1):1-8.
- Plettner, E.; Eliash, N.; Singh, N.K.; Pinnelli, G.R. and Soroker, V. (2017): The chemical ecology of host- parasite interaction as a target of *Varroa destructor* control agents. Apidologie, 48:78-92.
- Rashid, M.; Wagchoure, E.S.; Mohsin, A. U.; Raja, S. and Sarwar, G. (2012): Control of ectoparasitic mite Varroa destructor in honeybee colonies (*Apis mellifera* L.) by using

different concentrations of oxalic acid. J. of Animal and Plant Sciences, 22(1):72-76.

- Rosenkranz, P.; Aumeier, P.; Ziegelmann, B. (2010): Biology and control of *Varroa destructor*. J. Invert. Pathol. 103 Suppl 1: S96-119.
- Sammataro, D.; Gerson, U. and Needham, G. (2000): parasitic mites of honey bees: life history, implications, and impact Annu. Rev. Entomol., 45:519 -548.
- SAS Institute (1998): SAS users guide statistics (SAS Institute, Cay, NC. USA). Soroker,V.; Singh, **N.K.;** Eliash, N. and Plettner, E. (2019): Olfaction as a target for control of honey bee parasite mite Varroa destructor. In olfactory concepts of insect control-alternative to insecticides (1st Edition). J-F. Picimbon. Edited by Springer International Publishing, Heidelberg, Germany, 117-134.