Turkish Journal of Veterinary & Animal Sciences

Volume 33 | Number 4 Article 4

1-1-2009

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TÜBİTAK

Research Article

Turk. J. Vet. Anim. Sci.
2009; 33(4): 285-288
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doi:10.3906/vet-0712-16

Use of oxalic acid to control *Varroa destructor* in honeybee (*Apis mellifera* L.) colonies

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Received: 13.12.2007

Abstract: This study was carried out to determine the effects of oxalic acid (OA) on reducing *Varroa* mite (*Varroa destructor*) populations in honeybee (*Apis mellifera* L.) colonies in the fall. Twenty honeybee colonies, in wooden Langstroth hives, were used in this experiment. Average *Varroa* infestation levels (%) of the OA and control groups were 25.87% and 24.57% on adult workers before the treatments. The OA treatments were applied twice, on 3 November and 13 November 2006. Average *Varroa* infestation levels were 5.24% and 31.43% after the first application and 2.87% and 41.74% after the second application in the OA and control groups, respectively. Average efficacy of OA was 93.40%. No queens were lost, and there was no brood or adult honeybee mortality in any of the colonies during the experiment.

Key words: Honeybee, *Varroa destructor*, oxalic acid, control, diagnosis

Bal arısı (*Apis mellifera* L.) kolonilerinde Varroa (*Varroa destructor*) kontrolünde okzalik asit kullnımı

Özet: Bu çalışma bal arısı (*Apis mellifera* L.) kolonilerinde *Varroa* paraziti (*Varroa destructor*) zararını azaltmak için sonbahar sezonunda kullanılan okzalik asit'in etkinliğini belirlemek üzere yapılmıştır. Çalışmada standart ahşap Langstroth kovanlar'a yerleştirilmiş 20 adet bal arısı kolonisi kullanılmıştır. İlaç uygulamasından önce okzalik asit ve kontrol grubunda, işçi arılar üzerindeki ortalama *Varroa* bulaşıklık seviyesi sırayla % 25,87 ve % 24,57 olarak belirlenmiştir. İlaç uygulaması 3 Kasım ve 13 Kasım 2006 tarihlerinde 10 gün ara ile iki defa yapılmıştır. Ortalama *Varroa* bulaşıklık seviyesi okzalik asit ve kontrol gruplarında birinci uygulamadan sonra sırayla % 5,24 ve % 31,43, ikinci uygulamadan sonra ise % 2,87 ve % 41,74 olarak bulunmuştur. Okzalik asit'in ortalama etkinliği % 93,40 olarak belirlenmiştir. Deneme süresince hiç bir kolonide ana arı kaybı, yavru ve ergin arı ölümü gözlenmemiştir.

Anahtar sözcükler: Bal arısı, *Varroa destructor*, okzalik asit, kontrol, teşhis

Introduction

The pests, diseases, and parasites of honeybees (*Apis mellifera* L.) are the most important factors that influence the productivity of colonies (1,2). *Varroa destructor* is the most common and devastating parasite of honeybees and it is the number one problem in beekeeping worldwide (3). Honeybee colonies have to be checked regularly especially in the

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spring and fall and *Varroa* control measures have to be applied to keep the *Varroa* population level under the economic threshold. If the colonies are left untreated they normally die within a few years (4).

Many chemicals have been used to reduce or eliminate the damage caused by the *Varroa* mite throughout the world (5,6). Excess or incorrect use of the chemicals results in the contamination of hives and bee products (7-9).

Frequent use of the same chemicals such as fluvalinate and coumaphos over a period of time resulted in the development of more resistant mites and decreased treatment efficacy (2,8,10,11). Because of residue accumulation in beeswax and honey less toxic and more natural organic acids such as formic acid, oxalic acid (OA), and lactic acid (12) and essential oils such as thymol, eucalyptus oil, and citrus oil (13) have been used for the control of the mite (2,14,15).

OA is an organic acid; it is a natural constituent of honey and very effective against the *Varroa* mite. Uses of OA for the control of *Varroa* have been increasing in recent years (16). OA is safe to use, has no residue problems and a 5% dose of it can be tolerated by bees (11,17). It has been used by the beekeepers in the USA and Europe and the EU regulations permit its use in biological beekeeping (EU Council Regulation, No. 1804/1999). It is simple to use, cheap, and safe for beekeepers; no case of honeybee toxicity has been reported and it is very effective especially in broodless colonies (11,18). Milani (19) reported that OA was nearly 100% effective on mites in laboratory conditions. However, OA is not recommended by Imdorf et al. (8) or Donders and Cornelissen (9) during the honey flow period because it does not affect the mites in brood cells and may have low efficacy on mites.

Brødsgaard et al. (20) reported that many environmental factors such as colony condition, time of the year, presence or absence of a sealed brood, and application methods may influence the action of OA. In order to obtain the best effect from oxalic acid, the temperature should not be lower than 0 °C, the weather should be cool and dry, and the hives should have few or no sealed broods (11,20). The efficacy of OA was reported to be between 89.6% and 99.5% during broodless periods by Radetzki (21), Nanetti et al. (22), Imdorf et al. (23), Nanetti and Stradi (24), Rademacher and Harz (11), and Brødsgaard et al. (20). Gregorc and Planinc (4,25,26) reported that the effectiveness of OA was 52% and 39% in August and September in brooded colonies and 99.4%, 97%, and 88.7% in broodless colonies in November. Gregorc and Poklular (27) reported a 77.9% efficacy in colonies with broods in September and 98.6% in broodless colonies in December. Mutinelli et al. (18) achieved 95% efficacy after 3 treatments of a 5% OA solution when the capped brood was present.

The aim of this study was to determine the efficacy of 3% OA solution against *Varroa destructor* in honeybee colonies, during the autumn in Turkey.

Materials and methods

The experiment was carried out on 20 honeybee colonies (*Apis mellifera* L.) headed by sister queens in Central Anatolia between 3 and 30 December 2006. Colony strength (number of combs covered with bees), brood areas, and amount of food were equal. The colonies were randomly divided into 2 groups. The first group was treated with OA, and the second group colonies were used as the control. OA solutions were prepared by dissolving 30 g of 99% pure OA dihydrate into 1 L of sugar syrup (1:1 sugar/water (v/w)) as described by Nanetti and Stradi (24).

Varroa infestation levels of the colonies were determined at the beginning and 10 days after the OA application by using the wash and roll technique described by De Jong et al. (28). Five milliliters of OA solution was dropped on adult worker bees between 2 combs with a syringe as recommended by Imdorf et al. (23) and Brødsgaard et al. (20). After 10 days, the same amount of OA was given again. We checked all the colonies for dead worker bees and queens after both treatments. The efficacy of the OA treatments was calculated by using Henderson-Tilton's formula and the data were analyzed by randomized plot design (ANOVA). The Levene statistic was used for testing homogeneity among the dependent variables. Logarithmic transformation was performed to stabilize the *Varroa* infestation rates after the OA applications. Antilog transformation was performed for group means and the 95% confidence intervals of

the means were calculated. Group comparisons among the means were done with Duncan's multiple range test (29). SPSS Ver. 15.0 was used for all statistical analyses.

Results

Varroa infestation levels before and after the first and the second drug applications are summarized in the Figure. Average level of *Varroa* infestation (%) of the OA group and control group was 25.58 ± 1.70% and 24.57 ± 1.54%, respectively, before the treatments. There was no significant difference ($P > 0.05$) between the group means in terms of *Varroa* contamination at the beginning of the experiment but there were significant differences ($P < 0.01$) between the group means after both the first and second treatments.

The average *Varroa* infestation of adult worker bees was 5.24 ± 0.33% and 31.43 ± 1.55% after the first treatment and 2.87 ± 0.17% and 41.74 ± 1.81% after the second treatment, respectively, in the OA and control groups. The efficacies of the first and final treatment of OA were 84.49% and 93.41%, respectively. No negative effect of either OA treatment was observed in worker bees or queens.

Discussion

It was observed that one application of OA can reduce the *Varroa* population by over 84%. One application may bring the mite population below the economic threshold but 2 applications are more effective to control the mites. Our results are in accordance with those reported by Brødsgaard et al. (20), Rademacher and Harz (11), Mutinelli et al. (18), Nanetti and Stradi (24), Marinelli et al. (30), Gregorc and Planinc (26), Radetzki (21), Nanetti et al. (22), Imdorf et al. (23), Gregorc and Planinc (4,25), and Gregorc and Poklular (27), showing that OA is very effective against *Varroa destructor.*

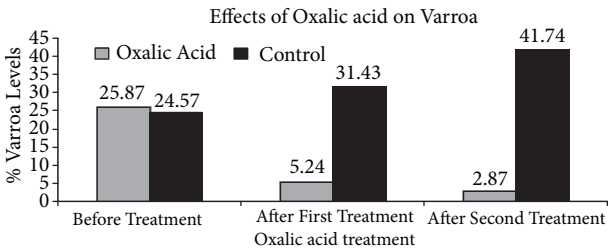

Figure. Effect of OA on Varroa destructor.

OA is a very promising candidate chemical for the control of *Varroa* mite. It has many advantages, namely it is easy to use, cheap, safe for beekeepers, and presents low variability between colonies in its final efficacy. Moreover, it causes very low or no honeybee toxicity, and there is no record of loss of queens, or brood or adult honeybee mortality. OA can be used effectively to safely reduce the damage done by *Varroa* mites in bee colonies especially during the broodless seasons.

Acknowledgement

Financial support from TÜBİTAK-TOVAG (Project 105O437) is gratefully acknowledged.

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