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Different methods for the management of Varroa mite (*Varroa destructor*) in honey bee colony

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Abstract
Varroa mite (Varroa destructor) is the major challenge for beekeeping worldwide. Therefore, various methods and materials for varroa control have been suggested: plant extracts, essential oils, biological Agents, mechanical methods and some chemicals. The presented review highlights that still more comparative studies among varroa control options under different ecological conditions are strongly required. Basically the performed researches have been concentrated on testing chemical materials and essential oils while relatively few studies have been done on the other control options. It is concluded that the varroa mite should be continuously (monthly) managed within honey bee colonies using mechanical methods or treatment with essential oils (mainly thymol) and others also. In severe cases, and especially during the fall and not during honey seasons, the use of chemical materials can be done with preference to oxalic or formic acid.

Keywords: Varroa mite, methods (mechanical control, biological control, chemical control

1. Introduction
Honey bee colonies are subject to infestation by insects, mites and diseases. The ectoparasitic mite Varroa destructor (Anderson and Trueman, 2000) [9] is considered as one of the most serious pests of beehives, causing huge losses to honey bees (*Apis mellifera* L) and great economic loss to the beekeeping industry (Abbadi and Nazar, 2003) [6]. Ectoparasitic mite of honeybee Varroa destructor was first described by Oudemans (1904) [30] from Java on *Apis cerana*. In 1962-63, the mite was found on *Apis mellifera* in Hong Kong and Philippines (Delfinado, 1963) and spread rapidly from there. The Varroa mite parasitizes only honeybees. Parasitism can result in a loss of up to 25% of adult weight, severe deformations of the wings and reduced longevity of worker and drone honey bees (De Jong *et al.* 1982; Kanga and James, 2002) [16, 25]. Its specialized mouthparts enable the mite to feed on bee brood and adult honeybees. The mite brood development is closely synchronized with bee brood development and colonies heavily infected by Varroa produce little or no honey (Ritter, 1981) [33]. Colonies infested with Varroa destructor have significantly reduced worker bee populations and eventually die if left without controlling. The development of infested brood is also affected because emerged bees have a low weight and shorter life span (De Jong *et al.* 1982) [16]. Beekeepers can use an integrated pest management (IPM) approach in which they use several different mite control methods in combination or in rotation throughout the year. A combination of various treatment protocols is effective and it reduces the likelihood that resistance to chemicals will develop, as happens when only one treatment method is used repeatedly.

2. Integrated pest management (IPM)
2.1 Mechanical method
Controlling Varroa mite populations via manipulations of the colony or hive can be effective, especially if several of the methods are used in conjunction. Mechanical controls include screened bottom boards, drone brood removal, and powdered sugar dusting.

2.2 Mite trapping/Drone comb
Drone brood removal takes advantage of the mites' preference for drone brood for reproduction, using them as a trap. Varroa mites have higher reproductive success in drone brood than in worker brood due to the post-capping period allowing mites to produce only 1.3-1.4 offspring per attempt in worker cells, but 2.2-2.6 offspring in drone cells.

In addition, the period of attractiveness of drone brood is 40-50 hours, as opposed to only 15-30 hours in worker brood. Together, these reproductive advantages of drone brood manifest as a 6-fold increase in mites found under the cappings of drone cells than under worker cells. Adding drone comb to a colony, encourages drone production that acts as a trap for mites. Removing that comb prior to drone emergence effectively removes the varroa mites reproducing in the cells. The drone brood can then be frozen and returned to the colony or scraped off of the frame. This practice reduces mite reproduction, which prolongs the length of time before the population reaches the threshold. However, it may not effective enough to act as the only means for controlling varroa mites. This technique depends on the preference of Varroa mite to drone cells with large size than worker cells with small size. However, combs with small cells cannot reduce Varroa infestation (Berry *et al.* 2010) [11]. It worth to mention that queen caging to reduce brood area is another method for controlling Varroa (Kotwal and Abrol 2013) [26].

2.3 Screened bottom board

Mites naturally fall off of bees as a result of movement within the colony and honey bee grooming behavior. If a screened bottom board, rather than solid wood one is used, mites fall onto the ground and are less likely to climb back onto the bees. Screened bottom boards decrease mite invasion into brood cells, resulting in a lower percent of the population being found in the brood reproducing. Mite loads still reach economic thresholds in hives with screened bottom boards, so this physical method to control varroa must be used in combination with other control techniques.

2.4 Powdered sugar

Sprinkling or applying powdered sugar on bees can serve as a method for mite control as this stimulates grooming behavior, resulting in more mites collected on bottom boards. Its use can be effective on bees removed from the hive equipment, but this is labor intensive, so beekeepers should weigh the costs and benefits when considering this practice. This treatment will not likely control the mite population on its own, but it can be used to increase mite drop in combination with screened bottom boards. Dusting with powdered sugar could be considered as a weak tool for Varroa control (Berry *et al.* 2012) while honey bee sprinkling with sugar syrup increased mite drop 2.5–2.7 times compared with the natural mite drop (Pileckas *et al.* 2012) [31].

3. Biological methods

3.1 Entomopathogenic fungi

Various entomopathogenic fungi were tested as possible control agents to Varroa. Rodríguez *et al.* (2009) study, *Metarhizium anisopliae* had high pathogenic capacity against Varroa with mortality rate of 85% and with good ability to tolerate beehive temperature. Similarly, Ahmed and Abd-Elhady (2013) [4] found good results to *M. anisopliae*.

3.2 Mite predator

The use of mite predator (i.e. pseudo scorpions; Arachnida: Pseudoscorpionida) was tested. Fagan *et al*. (2012) [18] found possibility of using pseudo scorpions as Varroa predator with no harmful impacts on honey bee larvae. For a single beehive, they expect about 25 Varroa predators can manage Varroa population.

3.3 Chemical control

Varroa mite reproduction throughout the spring and summer often leads to a large population in the fall. If the economic threshold is reached, one will have the best overwintering success if a chemical Miticide is applied prior to the production of the winter bees. In an IPM system, soft chemicals are used when possible.

3.4 Soft chemicals

Organic acids, essential oils, and hop beta acids are considered soft chemicals because they are naturally derived. These treatments are effective without leaving chemical residues in hive products, such as wax. If chemicals are used in the hive, it is recommended to apply soft chemicals first prior to considering the use of hard chemicals. In addition, colonies should be treated only after monitoring efforts have indicated that they are needed.

3.5 Formic acid

Formic acid occurs naturally in the venom of honey bees and is a natural component of honey. This chemical is commonly used because, at high concentrations, this organic acid penetrates the wax cappings and effectively kills reproducing mites. One limitation is that the use of formic acid is temperature dependent and can cause damage to the colony if used at ambient temperatures higher than 85F because it can increase brood mortality and the potential for queen loss. When used below 50F, formic acid results in low efficacy. Formic acid can kill some of the mites in the sealed brood cells. It is recommended that the formic acid be allowed to evaporate in colonies with sealed brood for at least two to three weeks. In this way, mites emerging from the brood will also be killed. The formic acid should be introduced into the colony only in the late afternoon to avoid damage to bees and brood. In addition, physiological tolerance is improved if the entrance hole is wide open. An easier way to introduce formic acid is to use a sponge or a similarly absorbent material. A solution of 50 ml of 60 percent formic acid is applied onto the sponge tissue per comb (Langstroh size). The quantity must be reduced accordingly for smaller comb sizes. The application can be repeated two times at intervals of ten days. The effectiveness of formic acid is well known for sealed and unsealed brood, but its results are impacted by temperature (Czirják and Monica 2013) [15]. Under field conditions, Lupo and Gerling (1990) [27] found good results for formic acid treatment against Varroa in June and August. Bahreini *et al.* (2001) [10] concluded that formic acid had significant effectiveness against Varroa for honey bee colonies in Iran.

3.6 Oxalic acid

Oxalic acid is a naturally-occurring compound found in plants, such as rhubarb, kale, beets, and spinach. As a chemical for mite control, oxalic acid can be used in two formulations: vapor and dribble. Because it does not penetrate the cappings, oxalic acid is most effective during brood less periods making it a useful component to an integrated varroa control program as a winter or early spring method. However, it should not be used as a stand-alone treatment. If overused or used at high dosages, oxalic acid can harm bees by crystalizing in the midgut of larvae, increasing larval mortality and reducing brood area. Overuse of this treatment can also decrease the activity and longevity of worker. Thirty two grams of crystal oxalic acid (dehydrate) is thinned in one

litre of sugar water (1:1). When handling crystal acid special precautions must be taken because of the health risks. Protective spectacles and acid-proof gloves must be work together with an adequate mouth protector. Depending on the size of the colony 20 to 30 ml of the suspension per chamber are dropped into the bee-ways. A repetition of the treatment can lead to damage to the bees. Applicators are available by which the acid can be evaporated. Higes *et al.* (1999) [23] found the efficacy of oxalic acid was 94 % and 73% in autumn and spring, respectively with losing three queens within the treated colonies. Gregorc and Planinc (2004) [21] found significant increase in mite mortality after the treatment with Oxalic acid (2.9%). Akyol and Yeninar (2009) [5] found efficacy of oxalic acid with mean 93.40% in the fall with no harmful impacts on queens, brood or bees. A water solution of 0.5% sprayed oxalic acid gave effective control of the mite (Toomemaa *et al*. 2010) [36]. The treatment with oxalic acid also reduced Varroa mite infestation in adult bees by 87.4% (Castagnino and Orsi 2012) [12]. Cornelissen and Gerritsen (2006) [14] found more effectiveness of 3% oxalic acid sprayed on the bees by 1% than formic acid against varroa in combination with swarm prevention technique (hive splitting into two) in Spring. The effect of the oxalic acid treatment was higher than that of the formic acid with less harmful impacts on honey bees (Pileckas *et al.* 2012) [31]. In line with this finding, Mahmood *et al.* (2012) [28] found 3.2% oxalic acid treatment recorded higher effectiveness in controlling Varroa than formic acid and flumethrin strip (Bayvarol).

3.7 Lactic acid

It is clearly better tolerated by bees and does not cause problems in warmer climatic zones. The disadvantage is that every single comb must be extracted to spray the bees with the acid. The dosage applied per comb side is 8 ml of 15 percent acid.

3.8 Plant extracts and essential oils

Essential oils and plant extracts are natural compounds distilled from plants (Table-1)

Table 1: Comparative studies on plant extracts and essential oils.

Studied plant extracts and essential oils / experiment type	Author (year)
Heterotheca latifolia and Tagetes minuta essential oils / Laboratory	Ruffinengo *et al.* (2001) [34].
Spearmint, eucalyptus, thyme, marjoram, basil, cumin, garlic, orange, geranium, and eugenol menthol / Laboratory and Field	Ismail *et al*. (2006) [24].
Black cumin oil, Nigella sativa, in comparison with a mixture of black cumin oil, lemon oil and jasmine oil, *Jasminum grandiflorum* / Field	Allam and Zakaria (2009) [7].
Marjoram, coriander, peppermint and black cumin / Field	Omran *et al.* (2011) [29].
Basil oils, camphor (*Eucalyptus* spp.),apiguard (thymol gel) and thyme (*Thymus vulgaris*) / Field	Refaei (2011) [32].
Neem seed, *Azadirachta indica* / Laboratory	Gonzalez-Gomez *et al.* (2012) [19].
Lemon grass, *Cymbopogon flexuosus*; thyme, *Thymus vulgaris*; cinnamon, *Cinnamomum zeylanicum*; anise, *Pimpinella anisum* / Field	Abd El-Wahab *et al.* (2012) [1].
Oils of common rue, eucalyptus, mint and thymol / Field	Castagnino and Orsi (2012) [12].
Plant extract of fenugreek, Santonica, mixture of fenugreek and santonica, and thyme / Field	Al-Zarog and El-Bassiouny (2013) [8].
Oils of garlic, turmeric, Tulsi, ajwain, clove and cinnamon / Field	Goswami and Khan (2013) [20].
Eucalyptus, cinnamon and menthol oils in combination with other control methods / Field	Kotwal and Abrol (2013) [26].
Thymol powder / Field	Ahmad *et al.* (2013) [2].

3.9 Thymol

The most popular essential oil for varroa mite control is thymol (from a thyme plant). While thymol treatment can effectively control mites on adult bees, it cannot penetrate the cell cappings, so does not control mites in brood cells. Efficacy of thymol is dependent on colony strength as well as ambient conditions. During treatment, the workers react by emptying cells near the product so this treatment can reduce the overall area of brood in colonies when applied in the spring. In addition, thymol treatment can induce robbing behavior and increase aggressiveness of colonies. Efficacy of thymol treatment can be low so it should be combined with other treatment methods.

3.10 Hops beta acids

Potassium salts of hops beta acids are derived from the hops plant and it is safe for use any time of the year, even during the honey flow. However, it is more effective as a mite control treatment when there is less brood because it does not go through the cell cappings. Use during brood rearing requires multiple applications. Ambient temperature does not impact Hop guard treatment. Efficacy varies, but it is generally not as high as other soft chemical treatments.

3.11 Hard Chemicals

Chemical control of varroa mites can be achieved through the use of various acaricides/miticides. Synthetic miticides are generally effective, killing up to 95% of the mite population. Historically, fluvalinate and coumaphos have been the most widely used mite treatments, but mites have developed resistance to these chemicals and residues persist and accumulate in wax. While these two hard chemicals are still legal to apply. Miticide residue in wax can harm bees directly, and makes bees more susceptible to Nosema disease. In addition, these residues can be found in bee products, which makes them less desirable to consumers. Synthetic chemicals should be a last resort for beekeepers practicing IPM. Amitraz-The most popular synthetic acaricide is amitraz (sold as Apivar). Amitraz does not, in its original form, persist as a contaminant of honey or wax. However, some metabolites of amitraz have been found to persist and there is a synergistic effect of amitraz and viruses that has been linked to increased bee mortality. In addition, resistance to amitraz has been documented, so its efficacy must be monitored closely. The results of Gregorc and Planinc (2012) [22] showed oxalic acid, thymovar, apiguard or amitraz fumigations were of limited use during the brood periods. Amitraz efficacy did not change

over time although the problem of mite resistance to miticides (Semkiw *et al.* 2013) [35]. Great variability in chemical efficacy is found and it could be said that amitraz is good option.

4. Conclusion

The Varroa mite can be considered as a continuous problem for beekeepers especially since there are various dispersal methods for the Varroa, beekeepers cannot easily control Varroa mite by 100% in their colonies. It is concluded that the required task is the continuous management of Varroa mite within colonies. From the studies, beekeepers can monthly do one practice to manage Varroa mite in their colonies using mechanical methods (e.g. sprinkling honey bees with sugar syrup) or treatment with essential oils (mainly thymol). In severe cases, and especially during the fall and not during honey seasons, the use of chemical materials can be done with preference to oxalic or formic acid. It could be expected that the continuous management of Varroa will reduce its damages greatly and will save honey bee colonies from losing. In an IPM approach, beekeepers should do cultural and mechanical practices for mite control before using soft or hard chemicals. Mite monitoring and rotation of treatments is critical for effective management and reduction of resistance to chemicals in these pests.

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