STUDIES TO ASSESS THE EFFICACY OF BACILLUS CBB AND APIGUARD® AGAINST *ASCOSPHAERA APIS* UNDER FIELD CONDITIONS

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Introduction

The bacillus CBB, developed by Dr Yacobson in Israel, has been found to inhibit the growth of *Ascosphaera apis* (causative agent of Chalk brood) in vitro. It was also found to be harmless to bees. Another environmentally friendly product, Apiguard® (Vita (Europe) Ltd) containing thymol crystals and applied against Varroa mites, was also found to inhibit the growth of this fungus. Thymol has been used by researchers in the past for the control of Chalk brood (Elde *et al* 1961, Barthel 1971).

To assess the efficacy of CBB, alone or in combination with Apiguard®, against Chalk brood in field conditions, three experiments were conducted during spring and summer of 2003, 2004, and 2005 in the area of Thessaloniki, North Greece.

Methodology

In 2003, 30 colonies of *Apis mellifera* macedonica, artificially infested by *A. apis*, were divided in three batches for the trials. Artificial infestation was applied according to a protocol by Martha Gilliam (Gilliam 1986). The division was made in such a way that all batches had about the same level of infestation and colonies of equal strength.

In batch 1, four applications of 1 litre sucrose syrup (60%) containing CBB were used. Batch 2 consisted of four applications, at weekly intervals, of 1 litre syrup (60%) containing CBB plus one application of 25g Apiguard®. The control (batch 3) was four applications, at weekly intervals, of 1 litre syrup (60%).

To prepare the suspension of CBB bacillus, vials containing the product were removed from the refrigerator and allowed to come to room temperature. Sterile distilled water (1 ml) was then injected into each vial. The vials were shaken very well until the complete dissolution of the pellet. The suspension was diluted in 10 litres of 60% sugar solution. One litre of the solution was transferred into a plastic bag, which was then sealed.

Each plastic bag was placed on the top of brood frames of every colony in Batches 1 and 2. The bags were perforated with a needle in about 20 places. In Batch 2, 25g of Apiguard® spread on a dosing card was placed next to the plastic bags. In Batch 3, (control colonies) plastic bags containing 1 litre of 60% sucrose syrup were placed in each colony.

The experiments in 2004 followed the same methodology as 2003, with the exception that 10 more colonies were used, allowing a forth batch. Batch 4 was treated with 25g of Apiguard®, spread on a dosing card, and placed on top of brood frames.

In order to reduce the impact of genetic variation to Chalk brood infection, all colonies were headed by one-year old sister queens.

In order to estimate efficacy under real conditions, a third experiment, (using the same treatments as in 2004) was conducted in the spring of 2005 at five different apiaries, naturally infected by *A. apis*. Each apiary consisted of eight colonies, and the 8 colonies were divided into four batches of two colonies. Each batch was treated following the 2004 methodology.

Results-Discussion

During the 2003 treatments, the decrease of the infection in batch 1 was estimated at 77.2%, in batch 2 at 83.9% and in batch 3 (control) at 7.8%, significantly different from batch 1 and 2 (see figure 1). No significant differences were observed between batches 1 and 2. Furthermore, the growth of colonies (number of bees and brood frames) was significantly better in colonies of batches 1 and 2 compared with control.

During the 2004 treatments, the efficacy of the bacillus was lower than in 2003. The decrease of the infection level in batch 1 was estimated at 58.9%, in batch 2 at 61.1% and in batch 4 (control) at 9.1%. In batch 3, where only Apiguard® was applied, the decrease of the infection was estimated at 53.9% (see figure 2). Furthermore, the growth of colonies (number of bees and brood frames) was significantly better in colonies of batches 1, 2, and 3 compared with control.

During the 2005 treatments, the efficacy of the bacillus was similar to that found in 2004. In batch 1, the decrease of the infection was estimated at 53.6%, in batch 2 at 62.6% and in batch 4 (control) at 13.8%. In batch 3 where only Apiguard® was applied, the decrease of the infection was estimated at 54.9% (see figure 3). The growth of colonies (number of bees and brood frames) demonstrated no significant differences.

Conclusions

The Results of these three experiments indicate that both CBB and Apiguard®, alone or in combination, demonstrate fungistatic action against A. apis.

The efficacy was satisfactory during the 2003 trials (artificial infected colonies) but was lower in 2004 and 2005 (naturally infected colonies). Furthermore, the variability among the colonies receiving the same treatment, was high, a factor that requires further investigation.

There is no difference in efficacy between lightly and heavily infected colonies.

Bacillus CBB appears to be completely tolerated by bees and gives no negative effect on a colony's strength (population and brood area).

The application of CBB in its present form requires special knowledge and skills from the user and, as a result, application by beekeepers is complicated. Further research is needed to develop a simpler mode of application.

References

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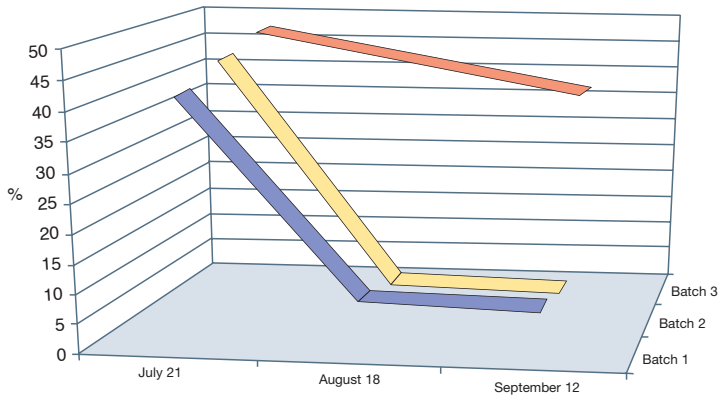
Elde, H., Weide, W. 1961. Die Bekampfung von Pericistis apis Maas, in Labor und Praxis. Wissenschafliche Zeitschrift Universitat Halle, Mahtematisch-Naturwissenschaftliche Reihe, 10: 83-86

Gilliam, M. 1986. Infectivity and survival of the chalkbrood pathogen, Ascosphaera apis, in colonies of honeybee, Apis mellifera. Apidologie. 17 (2): 93-100
Figure 1.



Chalk brood infection during 2003 trials

Batch 1
Batch 2
Batch 3

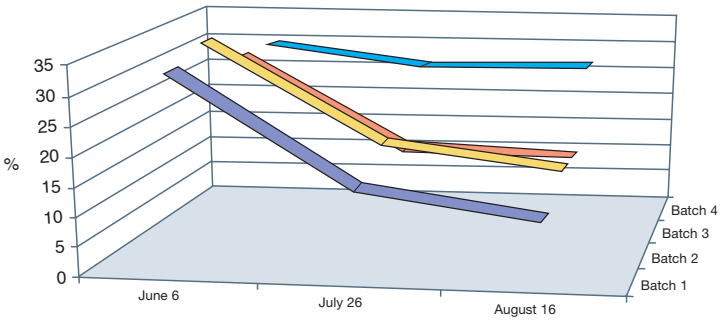


Figure 2.
Chalk brood infection during 2004 trials

Batch 1
Batch 2
Batch 3
Batch 4

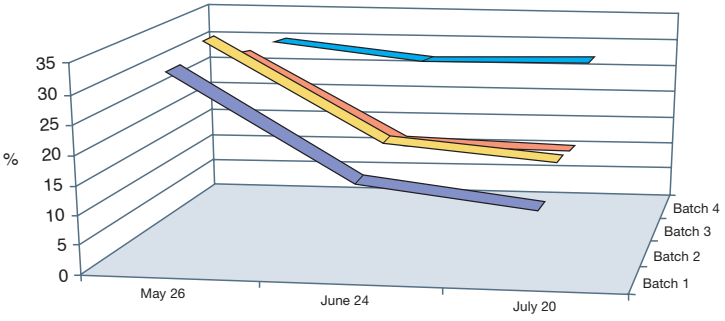


Figure 3.
Chalk brood infection during 2005 trials

Batch 1
Batch 2
Batch 3
Batch 4

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