## THYMOL, A SUBSTANCE WITH A WIDE SPECTRUM OF ACTION

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One of the most important sequences in the pathogenesis of *Varroa* disease is often represented by additional infections in bee colonies. It was demonstrated that the *Varroa jacobsoni* mite is an active carrier of the pathogens causing American foulbrood and acute paralysis, and of *Escherichia coli*. In a *Varroa* diseased colony, a higher death rate due to *Nosema* disease is recorded, and very severe and sharp symptoms of acute paralysis are recorded, which previously could be obtained under laboratory conditions alone.

In this particular sequence, preparations with a wide spectrum of action on the different pathogen agents are available to be used. One of them is thymol.

The acaricide effect of thymol was demonstrated by KOSTECKI, in the control of *Acarine* disease (*Acarapis woodi*). 10 g of thymol, in a Petri dish, were placed on the bottom board in April; after feeding of bees in September, another 15 g were placed into the hive and left there during winter. The author showed that thymol has an inhibitory action thus preventing infestation of bees with *Acarapis woodi*. During our experiments, we found that thymol is efficiently active also against *Varroa* mites.

When administered, during field experiments, in powder form — 0.25 g/beeway, or as retard preparation — 10—15 g/hive, the preparation was found to have an efficiency of  $73-97^{\circ}/_{0}$ . Under very strict experimental conditions, a 76% efficiency of thymol was recorded. The mites begin to fall from adult bees 48 hours after administration, their fall lasting for 4—7 days. The adult Varroa females are deprived of the capability of fixation on the host's body. The preparation causes an increased death rate of the mites inside brood cells, and decline of the egglaying of Varroa females. The preparation may be administered in any period of the year, being efficient at temperatures ranging from 8 to 42°C. With serious infestation — over  $20^{\circ}/_{0}$  of bees infested, thymol was found to be efficient by administration 3-4 times, every 4 days. With slight infestation, 2—3 applications, every 7 days, are enough. Curative treatment are made in spring, in summer after honey extraction, and in autumn. Treatment with thymol is more efficient when reiterated: first it is applied as powder, in spring; then, during the season, 10-15 g in a bag. Thymol may also be administered together with other substances (phenothyazine) or with other treatment methods (heat). The preparation is 1.5 times more active than phenothyazine and requires 4.7—9 times less work for application. When thymol is administered during winter, the death rate of parasites is 2 times higher than in control colonies.

Thymol was found to also be efficient in controlling other acarine mites in the hives. When administered in winter,  $823.9\pm43.8$  Acaridiae and Gamasoidae (in addition to Varroa mites) were killed, as compared to  $364.4\pm172.04$  found dead in the control hives.

According to different authors thymol is also efficient in the control of  $Braula\ coeca$ ; doses of  $1{-}100\ g$  are recommended to be administered, or to fumigate 3.5 g for 12 frames. The drug is also efficient against waxmoth. By keeping combs in thymol vapours spread of the above pests is prevented.

The study of the effect of thymol on bees showed that LD 50 for bees, by ingestion, is recorded when its concentration in sugar syrup is  $0.89^{0}$ <sub>0</sub>. A  $0.2^{0}$ <sub>0</sub> content in sugar syrup is not toxic to 5—10-day old bees. Administration of the drug in these concentrations in syrup, and treatment of bees, with doses and reiterations required to control Varroa disease, have not changed the content of glucose and of total lipids in bees' haemolymph. Administration of thymol in Varroa diseased colonies was found to increase the foraging activity and the amount of honey obtained, to have no influence on the sizes of ventriculus, and to increase the activity of bees in removing the thymol crystals and the dead and infested larvae and pupae. The sealed drone cells were opened by bees. Introduction of thymol into colonies intensified their activity, and the bags with thymol were covered with propolis. Following the intensification of activity, the average length of life of healthy bees increased by 3-70/0 and the infested colonies resisted for a longer time. When  $10-15^{\circ}/_{0}$  retard thymol was administered into colonies, at temperatures higher than 27°C, queens would discontinue egglaying, and in one apiary  $6^{\circ}/_{0}$  queens died. Thymol was also found to slow down the building of drone cells. By placing the drug in the upper section of the brood nest, the egglaying area would move 6-7 cm downward. Administration of thymol in weak colonies (less than 3 combs) causes their death. Thymol remains in honey for 48-72 hours.

The effect of thymol against protozoans was demonstrated by Tomito KADSO. The author used thymol in treating Nosema disease: 1 g of thymol dissolved in 2.3 l boiled water of which 142 ml were added to 4.5 kgs of 1:1 sugar syrup.

During our experiments in apiaries with *Nosema* diseased colonies, with administration of thymol in summer, no symptom of disease would be recorded in the next year. No spores were found to exist in colonies. Administration of sugar syrup, prepared as indicated above, resulted in a lower number of parasite spores on infested bees under laboratory conditions.

Antimicrobial action Administration of thymol was found to reduce 15 times the number of bacteria in the intestine of bees during winter as compared to the controls. In spring, bacteria were found in the haemolymph of all untreated bees, in all individuals. No bacteria was found in the colonies treated with thymol; in the colonies treated with thymol in summer, the haemolpmph seedings showed that bacteria existed in  $25^{0}/_{0}$  of bees in the treated colonies as compared to  $50^{0}/_{0}$  in the controls. In the intestine of bees in the colonies treated with thymol, the number of bacteria was considerably smaller than in controls.

This shows that thymol is one of the most efficient drugs in controlling *Varroa* disease in bees, and the results of our experiments recommend it to be used together with the other measures to control the disease. The drug has a wide spectrum of activity against the agents of different

bee diseases and pests, and is easy to administer.