

RESEARCH ARTICLE

Efficiency of natural spruce extract against varroaosis in organic beekeeping

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Varoatosis is the most dangerous disease of bees. A number of synthetic preparations are used for its treatment. In organic beekeeping they are forbidden; and the search for new effective acaricides, which are not toxic to bees and do not contaminate bee products, is highly relevant. Therefore, our purpose was to investigate the effectiveness of using natural spruce extract to fight against varroaosis at an organic apiary. In this study, we compared the effectiveness of using natural spruce extract and formic acid extract to combat varroaosis in 24 bee families (12 each the group). The preparations were administered according to the guidelines once in the spring and once in the autumn. In the course of the study, a number of indicators were evaluated such as spring and autumn mite infestation, the strengthening and weakening of families, honey and wax productivity, the percentage of families that died (lost their queen bee) during the winter. The results of our investigation show that natural spruce extract effectively controls the varroa mite. This is confirmed by the significant difference in mite infestation between the groups treated with formic acid and spruce extract. This indicator was lower in bee families treated with spruce extract. After the spring treatment, the difference was 3 times ($p < 0.05$), before autumn and spring next year it was 1.2 ($p < 0.01$). When spruce extract was applied, the bee families had by 1.2 times higher honey productivity and by 1.4 higher wax productivity. Therefore, it is advisable to use this anti-varroaosis preparation in organic beekeeping. We recommend administering preparations in the spring after the first flying rout and at the end of August, adding 15 ml per 1 liter of sugar syrup (90 ml/family). In the future, we plan to investigate the effectiveness of the use of eleutherococcus tincture in organic beekeeping.

Keywords: organic beekeeping; varroaosis; natural spruce extract

Introduction

According to the report from the Research Institute of Organic Agriculture (Willer & Lernoud, 2019), global organic farming is developing rapidly. In 1999, there were 200,000 organic producers; in 2017 their number was 2.9 million. Currently, there are 112.3 million hectares of organic land in the world, of which 70 (1.4%) are agricultural. Organic beekeeping is also developing dynamically. In 2017, organic farms had more than 3.2 million bee families, accounting for almost 3.5%. They are mainly concentrated in Latin America (45%) and Europe (30%).

The number of organic apiaries in Ukraine is also increasing. According to the national certification body in the field of organic certification "Organic Standard" (organicstandard.ua/), in mid-2019 there were about 50 of them, and in early 2020, already 75. According to experts (Willer & Lernoud, 2019), the main obstacle to the development of organic beekeeping is varroaosis. This problem remains one of the main issues in traditional beekeeping. It causes periodic large losses of bee families in Europe and the USA (Rosenkranz et al., 2010).

However, in organic beekeeping, it is particularly acute, since its technology requires only plants, herbal preparations and certain organic acids to be used to treat this disease (Commission Regulation (EC), 2007, 2008). Along with that, they also have a number of disadvantages that limit their use. It is possible to apply not artificial, but only natural acids, which are much more expensive (Regulation). Formic acid has low efficiency under ambient temperature range. According to Calderone (2010), a preparation based on this acid does not always provide an adequate level of control of varroa tick. Even when the temperature during the application falls within the recommended range specified in the instructions. Organic acids do not affect ticks in a closed brood (Bolli H. K. et al., 1993). The specificity of their use involves careful individual dose selection and increases the likelihood of overdose (Charriere & Imdorf, 2002). Formic acid has a neurotoxic effect on a bee brood. According to Gregorc & Planinc (2001), larvae that have intense metabolism are very sensitive to its action. There are reports (Giovenazzo & Dubreuil, 2011; Underwood & Currie, 2007) that the use of organic acids such as oxalic and formic can lead to the death of queen bees as well as workers. This slows down the development of bee families and, consequently, their productivity. However, most importantly, organic acids, like all chemical acaricides, contaminate bee products (Bogdanov et al., 2002; Moosbeckhofer et al., 2003). In addition, it is necessary to work with these acids in respiratory protective equipment and rubber gloves. Therefore, persons with special training are allowed to work with them.

Plants and herbal preparations are an alternative to chemical methods against varroaosis. At present, there are studies about the high effectiveness of thymol (Imdorf, 1999), menthol (Westcott & Winston, 1999), a mixture of thymol, eucalyptus oil, menthol and camphor (Calderone & Spivak, 1995), marjoram essential oil (Romo-Chacon et al., 2016; Sabahi et al., 2017), thyme and mint (Ariana et al., 2002) and other plants. The search for new effective acaricides, which are non-toxic to bees and do not contaminate bee products, is ongoing (Rosenkranz et al., 2010).

However, thymol, menthol and essential oils are very expensive for the Ukrainian beekeepers, so there is a need to find cheaper, but effective anti-varroaosisherbal preparations. One of such preparations is the natural spruce extract (Oriichuk, 2013; Petrovskiy, 2015), anti-varroaosis effect of which has not been studied sufficiently. It is a 50% aqueous extract containing 99.5 g of water extract of pine needles (spruce) and 0.5 of spruce essential oil (pine). In Ukraine, it is produced by the "Manevytske State Forestry", which is subordinated to the Volyn Regional Forestry and Hunting Administration.

Therefore, the purpose of this study was to evaluate the effectiveness of using natural spruce extract to combat varroaosis in organic beekeeping.

Materials and Methods

In order to accomplish the purpose, we set the task to conduct a scientific and economic experiment. During the experiment, we compared the mite infestation and indicators of viability and productivity of bee families, where different anti-varroaosis remedies were applied, in particular phytotherapeutic – natural spruce extract and chemical – formic acid, which is permitted in organic beekeeping.

The scientific and economic experiment was conducted at an organic apiary in the village Kumari (47°54'39" N, 30°39'12" E) in the Vradiiv raion of Mykolaiv region during 2017-2018. For this purpose, during the autumn inspection in 2016, two groups of bee families of Ukrainian breed – experimental and control with 12 families in each group – were formed by the method of pair-analogues (identical in strength, quantity of brood, feed, honeycombs, age and origin of the queen bee) (Brovarskyi et al., 2017). Bee families were kept in multiple-storey hives with frames 235×230 mm in size.

Natural spruce extract was used against varroaosis in the experimental group, while formic acid was used in the control group. In the experiment, there was no absolute control group, i.e. the group without treatment against mites, as it was found out that the absence of anti-varroaosis treatment for more than two years leads to the absconding or death of bee families (Le Conte et al., 2010).

The preparations were administered according to the instructions twice: in March immediately after the first flying rout and at the end of August. Glass dosage devices with formic acid (50 ml per family) were placed in the bee families twice in the spring every 12 days for 5 days, and in August once for 5 days. The devices were suspended from the upper bar of the empty frame on the side of the brood nest. Natural spruce extract was added to sugar syrup at a dose of 15 ml per 1 kg of feed; it was poured into a feeder above the brood nest. 90 ml of extract per family was used throughout the cycle.

In the course of the scientific and economic experiment, mite infestation was observed in bee families (Polishchuk, 2011; Rut et al., 1993). For this purpose, at least 50 bees were selected from the frames in the center of the nest. A glass jar was brought to vertically arranged honeycombs; and bees were collected with the help of careful movements from the bottom to the top, making sure that the queen bee does not get into the jar. The jar with bees was closed with a lid. About 400 ml of cold water was poured into the pot and was heated. The bees were gathered at the bottom of the jar by shaking; the lid was opened and quickly turned into a vessel with water, making circular motions to wet the bees. When all the bees became wet, the jar was taken out. The water gradually warms up and the mites leave the bees, falling to the bottom. When the water started boiling, the pot was put aside; the bees were removed with a tweezer, rinsed thoroughly with water, and counted. After that, the water was carefully poured out of the pot so that there was a residue with mites at the bottom, which was then poured into a vessel with the white bottom and the mites were calculated. The mite infestation was calculated as the ratio of mites to bees multiplied by 100.

Also, according to conventional methods (Brovarskyi et al., 2017), indicators that characterize the viability of bee families were determined, including strength and weakness, feed costs, percentage of families who died, percentage of families who lost their queen bee, and also honey and wax productivity. The strength of families was determined by the number of bee ways. A bee way is the space between two adjacent nest honeycombs which is completely covered by bees. The weakness of families was calculated as the difference between the number of bee ways as of autumn and the spring inspection.

Feed costs were calculated per one bee way. For this purpose, the honey in the nest was weighed and divided by the number of bee ways. The weight of honey was determined using a 5×5 cm square grid frame containing approximately 40 g of honey. The frame was set to the honeycombs where there is feed; the squares were counted and multiplied by 40 g. The share of families that died (lost their queen bee) was defined as the ratio of the number of families that died (lost their queen bee) during wintering to the number of families during the autumn inspection multiplied by 100. Wax and honey productivity were calculated at the end of the honey flow using the weight of the extracted honey and melted wax.

The significance of difference was determined by Student's t-test.

Results and Discussion

It was found out that at the beginning of the study prior to spring treatment in 2017, bees of the experimental and control families were not significantly different in terms of mite infestation. This indicator ranged from 3 to 5% and indicated that they needed treatment measures for varroaosis (Table 1).

After the spring anti-varroaosis treatment, the mite infestation of families treated with spruce extract was 0.25%, which is by 3 times less significant than the control one ($t_{22}=2.128$, $P<0.05$). This difference is explained by the fact that during the treatment the air temperature dropped below +14°C at night, which led to a decrease in the effectiveness of formic acid.

During the honey flow, the mite infestation increased in both groups. However, in bee families treated with formic acid, it was almost 6%, which is by 1.2 times more significant than in the experimental one ($t_{22}=2.887$, $P<0.01$). In our opinion, this was a consequence of the higher mite infestation of bee families of the control group after the spring treatment. Medical treatments in the autumn with different remedies were equally effective, since there was no significant difference between the groups. During the autumn treatment, the air temperature was in the optimum range for the action of formic acid. The final mite infestation of bee families was less than 0.08%. That is, the natural spruce extract ensures the destruction of 99.92% of mites, provided it is used in spring and autumn.

Before the first spring inspection, the mite infestation in both groups did not exceed 5%, but in the experimental group it was by 1.2 times significantly lower than in the control one ($t_{22}=2.303$, $P<0.01$). The same mite infestation of the control and experimental group of bee families is explained by studies that characterize the viability and productivity of bee families (Table 2).

It was found out that at the beginning of the study (before the spring inspection in 2017) bee families had the same strength. However, before the autumn inspection in 2017, families of the control group had by 1.9 bee way significantly less strength than families in the experimental group ($t_{22}=9.319$, $P<0.001$). In our opinion, this is due to the fact that the greater mite infestation of

the families in the control group after the spring treatment slowed down their development during the honey flow. As they became weaker during the winter, they appeared to be by 1.8 bee way weaker after winter ($t_{22}=7.607$, $P<0,001$), although before the autumn treatment the mite infestation of the groups was the same.

Table 1. Mite infestation of bee families, % (n=12).

Mite infestation	Anti-varroaosis remedy			
	spruce extract		spruce extract	
	$\bar{x} \pm s_{\bar{x}}$	min-max	$\bar{x} \pm s_{\bar{x}}$	min-max
before spring treatment in 2017	4.33±0.23	3-5	4.50±0.19	3-5
after spring treatment in 2017	0.25±1.13*	0-1	0.83±0.24	0-2
before autumn treatment in 2017	4.83±0.27**	3-6	5.92±0.26	4-7
after autumn treatment in 2017	0.08±0.08	0-1	0.17±0.11	0-1
before spring treatment in 2018	4.00±0.25**	3-5	4.92±0.31	4-7

* $P \leq 0.05$, ** $P \leq 0.01$ compared to the control group.

The indicators that determine winter resistance depend on the strength of families. Thus, the weakening of bee families of the control group during winter was by 1.4 times significantly higher than in the experimental one ($t_{22}=2.419$, $P<0.05$). The difference in the amount of feed consumed was 1.2 times ($t_{22}=4.485$, $P<0.001$). However, these indicators were not critical and all 100% of families survived during the winter and did not lose their queen bee. According to the literature data (Gregorc & Planinc, 2001; Giovenazzo & Dubreuil, 2011; Underwood & Currie, 2007), organic acids, along with the therapeutic benefit of varroaosis have a side effect. They negatively affect the larvae, queen bees, workers, which affects the development of bee families and their productivity.

Table 2. Indicators of viability and productivity of bee families (n=12).

Indicator	Anti-varroaosis remedy			
	spruce extract		formic acid	
	$\bar{x} \pm s_{\bar{x}}$	min-max	$\bar{x} \pm s_{\bar{x}}$	min-max
Strength of families before spring inspection in 2017, bee ways	7.75±0.13	7-8	7.67±0.14	7-8
Strength of families before autumn inspection in 2017, bee ways	10.58±0.15***	10-11	8.67±0.14	8-9
Strength of families before spring inspection in 2018, bee ways	9.50±0.19***	8-10	7.67±0.14	7-8
Weakness of families, bee ways	1.08±0.08*	1-2	1.50±0.15	1-2
Amount of consumed feed,kg/bee way	1.35±0.04***	1.1-1.6	1.62±0.04	1.4-1.8
Honey productivity, kg	44.67±1.98**	25-51	37.42±0.79	32-40
Wax productivity, kg	0.56±0.02***	0.43-0.65	0.40±0.02	0.29-0.50

* $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$ compared to the control group.

The decline in the viability of bee families affected their productivity when formic acid was used. However, the productivity of families where natural spruce extract was used was better. It was found out that the bee families in the experimental group had by 1.2 times higher honey productivity ($t_{22}=3.407$, $p<0.01$) and by 1.4 times higher wax productivity ($t_{22}=5.943$, $p<0.001$).

Conclusion

Natural spruce extract is an effective alternative method to fight against varroaosis in organic beekeeping. It provides a final mite infestation of 0.08%, which contributes to the full development of bee families and increase in honey productivity by 1.2 times and in wax productivity by 1.4 times. These indicators are better than those for formic acid. We recommend using natural spruce extract for the treatment of varroaosis in organic beekeeping twice: in the spring immediately after the first flying rout and at the end of August, adding 15 ml per 1 kg of feed to sugar syrup (90 ml per family for the whole treatment cycle).

Research and development of effective natural methods of combating varroaosis is a prerequisite for the sustainable development of organic beekeeping. Therefore, in the future we plan to investigate the effectiveness of the anti-varroaosis action of eleutherococcus tincture in the context of an organic apiary.

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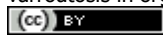
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