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Animal Biology

Control of varroosis with oxalic acid trickling under conditions in the Czech Republic

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Abstract: Oxalic acid is a powerful acaricide commonly used for the control of varroosis. The winter trickling of a bee colony constitutes an easy and effective treatment method leaving no harmful residues in bee products. Due to some scepticism in the Czech Republic about using oxalic acid for the winter treatment of a colony the object of the study was to verify this application under the Czech conditions and to test an alternative way of the oxalic acid application potentially reducing the winter disturbation of a colony. The results prove the high effectiveness (> 95%) of the trickling also under conditions in the Czech Republic including for newly proposed way of treatment not requiring a super unfolding. The results are discussed also in relation to the Czech beekeeping practice.

Key Words: oxalic acid, trickling, *Apis mellifera*, *Varroa* destructor, varroosis

INTRODUCTION

Oxalic acid (OA) has been used as acaricide against mites (*Varroa destructor*) since late nineties and showed several advantages compared to earlier used formic acid (Kunzler et al. 1979, Takeuchi and Harada 1983). Recently, oxalic acid is a commonly used medicament with positive results (Rademacher and Harz 2006). In the Czech Republic there have still been doubts about OA's eligibility for the control of varroosis. Therefore, this form of treatment has not been spread in the Czech Republic.

The object of this contribution is to verify efficiency of OA trickling application under conditions in the Czech Republic with the possibility of reducing the colony disturbance.

MATERIAL AND METHODS

The experiment was made in winter 2014/2015 at 3 apiaries. The apiaries were under the same varroosis treatment during summer.

All 66 experimental colonies genetically originated in Vigor®. All hives were protected against ant access by Formistop® stand accessories (Klíma 2009). The therapeutic mite fall was monitored at the varroa diagnostic hive bottom (Přidal and Svoboda 2012a).

The treatment solution was prepared as a mixture of 42 g oxalic acid dihydrate, 600 g sucrose (sugar-beet sugar) and water up to one liter of the total volume. The solution contained 4.2% of OA dihydrate and 60% sucrose (all in w/v). The treatment solution was trickled in gaps among frames ("interframe gaps") where the bee cluster was located. Five millilitres of OA solution per an interframe gap were applicated (Rademacher and Harz 2006).

Ways of OA trickling

A) Direct application – OA trickling only directly on the bee cluster, i.e. only in the occupied interframe gap where the position of a bee cluster was visible. The hives with the lower position of a bee cluster were unfolded for the direct OA application in the lower occupied super (apiaries: Černá Pole and Markvartice),

B) Indirect application – OA trickling only via the top super into every interframe gap regardless the horizontal or vertical position of the cluster (apiary: Příbram na Moravě).

Effectivity of the treatment by OA trickling

To prove effectivity of the OA trickling the control treatment by Varidol FUM with active substance amitraz was used for fumigation of colonies after the 1st experimental oxalic acid treatment in the control group. The therapeutic mite fall was monitored in the period between the first (November/December 2014) and the second (February 2015) treatment and then again 1 month after the second treatment.

The effectivity of trickling or fumigation was expressed as a percentage of the therapeutic mite fall related to the total mite fall.

Statistics

Average values are presented with their standard errors (S.E.). The statistical significance of differences of the mean values was analysed by t-test and the difference with p-value above 0.05 ($p>0.05$) was considered statistically insignificant.

RESULTS

The therapeutic mite fall after the 1st OA application for each apiary and all apiaries together are displayed in Table 1. The fall intensity differed in both groups but all differences were statistically insignificant ($p>0.05$). The total averages from all apiaries are different between groups by only three mites. The groups were infested equally.

Table 1 The first therapeutic mite fall after OA treatment

Apiary	Date	1st therapeutic mite fall (only OA)		p-value
		Control group	Experimental group	
Černá Pole (n = 30)	20. 11. 2014	108 ± 43	110 ± 20	0.959
Markvartice (n = 16)	23. 11. 2014	410 ± 83	319 ± 79	0.472
Příbram na Moravě (n = 20)	6. 12. 2014	47 ± 13	115 ± 57	0.298
altogether (n = 66)	November/December 2014	161 ± 37 [3–746]	164 ± 32 [2–661]	0.951

Legend: n = number of colonies, [min–max]

In Table 2, the therapeutic fall after the February fumigation treatment and the 2nd OA trickling are displayed regarding each apiary alone and altogether. The mite fall differences are minimal and statistical insignificant ($p>0.05$).

Table 2 The second therapeutic mite fall after treatment by amitraz (A) and oxalic acid (OA)

Apiary	Date	2nd therapeutic mite fall		p-value
		Control group (amitraz)	Experimental group (OA)	
Černá Pole (n = 16 and 14)	23. 2. 2015	3.4 ± 1.30	4.9 ± 1.25	0.432
Markvartice (n = 8 and 8)	21. 2. 2015	4.0 ± 1.72	3.9 ± 1.57	0.961
Příbram na Moravě (n = 10 and 10)	21. 2. 2015	0.3 ± 0.28	0.9 ± 0.48	0.323
Altogether (n = 34 and 32)	February 2015	2.6 ± 0.79 [0–22]	3.4 ± 0.76 [0–16]	0.495

Legend: n = number of colonies, [min–max]

The average effectivity achieved in the control bee colonies (amitraz treatment) at all the apiaries 96.4% effectivity and in the experimental colonies treated by OA for the 2nd time 94.9% effectivity. The difference is statistically insignificant (p = 0.483). The effectivity of OA at each apiary ranged from 92.1 to 99.0 (Table 3).

Table 3 Oxalic acid treatment effectivity

Apiary	**Effectivity of OA treatment**			**p-value**
	Control group (A)	Experimental group (OA)	Both groups together	
Černá Pole	92.9 ± 2.2%	91.2 ± 3.4%	92.1 ± 2.0%	0.697
Markvartice	99.2 ± 0.3%	98.9 ± 0.3%	99.0 ± 0 .2%	0.620
Příbram na Moravě	99.7 ± 0.3%	96.7 ± 2.0%	98.2 ± 1.1%	0.202
Altogether	96.4 ± 1.2%	94.9 ± 1.7%	95.6 ± 1.0% [76–100%]	0.483

Legend: [min–max]

The impact of the direct and the indirect application of OA was observed at apiary Příbram na Moravě (Table 4). The therapeutic mite fall was higher in the group of colonies with the lower bee cluster position and thus trickled with OA indirectly via the top hive super. However, the difference of the mite fall between the both ways of OA trickling was statistically insignificant (p = 0.251). The effectivity of the indirect treatment was lower about 3.1% in average in comparison with the direct treatment. The difference is statistically insignificant (Table 4).

Table 4 The impact of direct and indirect OA trickling on the winter bee cluster

Date of treatment	**Therapeutic mite fall after:**		**p-value**
	Indirect OA trickling on a bee cluster via interframes gaps of the top super (bee cluster in a lower super)	**Direct OA trickling** on a bee cluster (bee cluster in the top super)	
6. 12. 2014 (OA)	77 ± 26	41 ± 12	0.251
21. 2. 2015 (OA/A)	1 ± 0.48	0.1 ± 0.1%	0.113
Effectivity	96.8 ± 0.019% [80.8–100%]	99.9 ± 0.001% [99.1-100%]	0.146

Legend: [min–max]

DISCUSSION

Varroa mites reduction effectiveness of the winter oxalic acid trickling into interframe gaps was in average higher than 95% that is in accordance with previous studies (Nanetti et al. 2003, Bahreini 2003, Gregorc et al. 2004). Therefore, OA treatment is reliable and therefore should be highly recommended for routine usage also in the Czech Republic. The high effectivity of the OA trickling was fully comparable with the effectivity of the amitraz fumigation so far widely used in the Czech Republic. There were no apparent differences in the colony condition (colony strength, overwintering, brood area etc.) among bee colonies treated twice and only once by OA. It is necessary to use the modern approaches in the control of varroosis especially when the risk of use (Gregorc et al. 2004, Hatjina and Haristos 2005, Schneider et al. 2012, Staroň and Staroňová 2013) is difficult to track in practice

(Toomemaa et al. 2010) especially in the broodless period e.g. in winter (Higes et al. 1999). Hence, the winter OA treatment is recommended for the wide Czech beekeeping practice.

As the results confirmed if a bee cluster is not located at the top of the hive the top supers don't have to be unfolded for the OA trickling application. Thus, the solution can be applicated also indirectly on a bee cluster via the top super interframe gaps. This way of the application reduces laboriousness, stress of a colony in the wintertime period and the risks of worker or even the queen damaging. Considering the extreme minimal value of 80.8% a certain worry about lower effectivity of the indirect trickling with OA can exists. Therefore, for more reliable results further tests of the indirect trickling are needed. The higher mite fall after the indirect OA trickling can be caused by the lower capturing of mites in the hive. The height of the unoccupied combs under the winter cluster is dependent on its position. Under the bee cluster with a lower position the area for the mite catching is shorter unlike the cluster located at the top of the hive as it was analogically argued by Přidal and Svoboda (2012b).

CONCLUSION

OA trickling into interframe gaps as the treatment for the control of varroosis in the wintertime was in average higher than 95%. It was not proved that OA trickling would have any lower effectivity compared to effectivity of the fumigation by amitraz. Hence, OA trickling is recommended as an effective treatment in the winter broodless period also under Czech conditions.

It was not proved that the indirect application of OA solution via the top hive super onto the bee cluster sitting in a lower super leads to any reduction in efficiency. Therefore, the method is highly recommended due to reducing of colony disturbation during wintertime. Regarding high effectiveness of OA treatment with minimal side effects, no harmful residues in bee products and easy application is desirable to keep recommending this effective treatment in the official Czech beekeeping practice.

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