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Original article

Determination of residues in honey after treatments with formic and oxalic acid under field conditions

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Abstract – Formic acid and oxalic acid field trials for control of *Varroa destructor* were carried out in autumn according to the Swiss prescriptions during three successive years in different apiaries in Switzerland. The following parameters were determined in honey that was harvested the year after treatment: formic acid, oxalic acid and free acidity. The following range of values were found in honeys of untreated colonies: formic acid, from 17 to 284 mg/kg, n = 34; oxalic acid, from 11 to 119 mg/kg, n = 33. There was a small, but unproblematic increase in formic acid levels in comparison to the levels in the controls; average: 46 mg/kg, maximum: 139 mg/kg. No increase in formic acid was found with increasing number of treatment years. If emergency formic acid treatments were carried out in spring, the residue levels were much higher: average increase of 193 mg/kg, maximum 417 mg/kg. The oxalic acid content remained unchanged, even after two successive treatments during the same autumn. No rise of free acidity was encountered after a combined treatment with formic and oxalic acid during the three trial years.

honey/residue/formic acid/oxalic acid/free acidity/*Varroa destructor*

1. INTRODUCTION

Presently synthetic acaricides are used regularly for the control of *Varroa destructor* Anderson and Truman, (formerly *Varroa Jacobsoni* Oudemans). However, due to their lipophilic and persistent nature, they accumulate in wax, and to a smaller extent in honey (Bogdanov et al., 1998b; Wallner, 1999). Recently, acaricide resistant mites have appeared in many countries of the world (Milani, 1999). Because of these problems, natural, non-toxic substances such as different organic acids (Liebig, 1997; Imdorf and Charrière, 1998) and essential oils (Imdorf et al., 1999) were developed for the control of *V. destructor*. These substances are used increasingly throughout Europe.

Oxalic and formic acids are natural constituents of honey. They are allowed for use in biological beekeeping (EU Council Regulation, 1999). According to the EU residue regulation (1995) formic acid has a GRAS status (Generally Recognised As Safe) and the establishment of a MRL (Maximum Residue Limit) is not necessary. On the other hand, oxalic acid is not mentioned in the EU residue prescriptions. Oxalic acid is not registered in most West European countries, although it is widely used because of its high efficiency in the absence of brood (Nanetti and Stradi, 1997). Thus, there is an urgent need to supply residue data in order to enable the registration of oxalic acid products for the control *V. destructor.*

Until now residue studies have been carried out under controlled experimental conditions

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after single treatments with formic acid (Stoya et al., 1986; Hansen and Guldborg, 1988; Krämer, 1994; Barbattini et al., 1994; Capolongo et al., 1996; Radke and Hedtke, 1998). There are several short communications on residues after single treatments with oxalic acid (Radezki, 1994; Mutinelli et al., 1997; Del Nozal et al., 2000; Bernardini and Gardi, 2001), but they do not allow for a detailed assessment of eventual residue problems. To date, no long-term field trials have been carried out measuring the possible residues after repeated use of formic and oxalic acid in practical beekeeping. Indeed, such studies are necessary for the assessment of possible long-term effects of acaricide residues on the quality of bee products. Field trials on the elucidation of long-term effects of the repeated application of thymol (Bogdanov et al., 1998a) and synthetic acaricides (Bogdanov et al., 1998b) on honey and wax quality have been conducted. As organic acids are hydrophilic substances which do not accumulate in wax, only honey studies are necessary. The purpose of the present study was to determine honey residue levels following formic and oxalic acid treatments under practical Swiss beekeeping conditions.

2. MATERIALS AND METHODS

2.1. Treatments with organic acids

Three trials were carried out in apiaries, situated in various parts of Switzerland during 1996, 1997 and 1998. The number of apiaries involved in the trials each year varied over the three trial years (see Tabs. III to VI). The average number of hives per apiary was 12 (minimum 9). In these apiaries the beekeepers followed a definite treatment protocol. Two formic acid treatments were carried out in August–September, followed by one oxalic acid treatment during the broodless late autumn period between November and December. Honey samples from neighbouring apiaries, treated with Apistan were taken as controls (see also Sect. 2.2).

2.1.1. Normal treatments with formic acid in autumn

Formic acid treatments were applied twice in autumn, usually in August and September by different application devices, mostly by the Liebefeld dispenser (Charrière et al., 1998a). Two applications with 130 ml 70% formic acid per colony were applied by evaporation over seven days.

2.1.2. Emergency treatments with formic acid in spring

Emergency treatments with formic acid in spring were carried out during 1997 and 1999 in 7 different apiaries which did not take part in the trials indicated above (Tab. IV). The emergency treatments had to be carried out because of high mite infestation pressure that endangered colony survival. One treatment with formic acid was carried out just after the spring honey harvest during one week, in the period between the end of May and the beginning of June, as described above for the normal treatment. The honey supers were taken off before the treatment.

2.1.3. Oxalic acid treatments

Oxalic acid treatments by spraying were carried out during the broodless period between beginning of November and end of December. Spraying was carried out by applying a solution of 30 g oxalic acid dihydrate/L water according to Charrière et al. (1998b).

In 1998, trickling was carried out in some apiaries before spraying in separate trials to test the efficacy of different methods of application of oxalic acid. In these cases, 50 ml of 60 g oxalic acid dihydrate/L water:sucrose (1:1) solution was trickled per bee colony according to Charrière et al. (1998b) before the spray treatment.

2.2. Honey sampling

Most trials samples were taken from the first honey harvested in spring. However under some circumstances (e.g. lack of a sufficient spring honey flow) beekeepers in Switzerland harvest the first honey in summer. For that reason we had to include summer honeys in the trial.

Honey yields were not measured, as we wanted to test the residues under practical conditions independent of the honey yield. The honey yields are taken from the official regional harvest reports of the Swiss Beekeeping Associations (averages and range of variation):

1996: 4.1, 1.7–7.3 1997: 6.9, 2.0–12.6
1998: 12.9, 7.9–15.6 1999: 9.9–12.4.

As the control and trial samples were taken from neighbouring apiaries, the yields of the control and trial honeys were similar.

Control and treatment samples were divided into two groups according to their electrical conductivity (see Sect. 2.3). The hypothesis that the electrical conductivity for a given date did not differ among samples from untreated and treated colonies was tested by the Student's *t*-Test (Microsoft Excel 1997).

2.2.1. Trial honey samples

After the autumn treatments, honey samples were collected from the first honey harvest in spring or in summer of the following year. After the spring treatments, the samples were collected from the summer harvest of the same year. In each apiary three honey samples were collected during the filling of honey from the honey tank at the beginning, middle and the end of the filling process. The average of the measurements of these three samples was used for the trial evaluation.

2.2.2. Control honey samples

Control honey samples were collected in the same way as the trial samples from apiaries that were in the neighbourhood of the treated apiaries to guarantee that the control honeys were as similar as possible to the trial honeys. The control apiaries were treated with Apistan.

2.3. Honey analysis

The honey samples were stored at room temperature until analysis within 6 months after the honey harvest. Formic and oxalic acid were assayed by the Böhringer oxalic acid kit, by which both acids are determined in the same sample (Böhringer Mannheim, 1997).

The stability of formic and oxalic acids was tested by measuring these acids in two honeys, stored at room temperature (20–25 °C) for six months. The honeys had the following initial concentrations:

honey 1: formic acid 71.6 mg/kg and oxalic acid 45.0 mg/kg;
honey 2: formic acid 224.0 mg/kg and oxalic acid 89.0 mg/kg.

Measurements after 3 and 6 months showed that the acid concentrations remained at their initial level (all measurements fell within the precision limits of the method, see Tab. I).

For determination of recovery, honey was spiked with 150 and 300 mg formic acid/kg and with 50 and 100 mg oxalic acid/kg (n = 8 for each concentration level). The recoveries for the two concentration levels were the same, thus, the recovery is presented here as an average of both concentration levels:

formic acid: 93% (sd = 4%, n = 16) for honey spiked with 150 and 300 mg/kg;
oxalic acid: 84% (sd = 24%, n =16) for honey spiked with 50 and 100 mg/kg.

The precision of the measurement, defined by the coefficient of variation CV %, was determined for three different concentration levels (Tab. I). One of the honeys measured was an "unspiked" blank, the others were the same sample of honey, spiked with 150 and 300 mg/kg formic acid and 50 and 100 mg/kg oxalic acid. The precision of the formic acid measurement was somewhat better than that of the oxalic acid determination. The limit of detection of the two enzymatic methods fell between 3 to 5 mg/kg.

Free acidity (in milliequivalents (meq) per kg) and electrical conductivity (in millisiemens (mS) per cm) were determined according to the harmonised methods of the International Honey Commission (Bogdanov et al., 1997). Honeys with less than 0.8 mS/cm belonged to the group consisting of floral honeys and blends of floral and honeydew honey, while those having more than 0.8 mS were assigned to the honeydew type (Bogdanov et al., 1999b). As the content of formic and oxalic acid depends on the honey type (Tab. II), the trial and the control honeys belonged to the same honey group, which, in all but one trial (see Sect. 3.1) belonged to the group consisting of blossom and blends of blossom and honeydew honeys. There was no statistically significant difference between the electrical conductivities of the trial and the control honeys. No unifloral honeys were present in the control and

Table I. Precision and repeatability of the enzymatic determination of formic and oxalic acid. Values in mg/kg.

	Formic acid			Oxalic acid		
average	74.1	214.9	353.9	47.9	86.7	134.1
st. deviation	3.3	7.7	10.3	6.4	16.0	22.7
CV %	4.4	3.6	2.9	13.4	18.4	16.9
n (samples)	11	8	10	9	8	12

Table II. Formic acid, oxalic acid and free acidity in untreated honeys. Data expressed as: conductivity (Co) in mS/cm; formic acid (FA) and oxalic acid (OA) in mg/kg; free acidity (FAc) in meq/kg.

	blossom, blossom-honeydew blends				honeydew honeys			
	Co	FA	OA	FAc	Co	FA	OA	FAc
average	0.41	41	25	17	0.98	93	70	27
st. deviation	0.14	17	11	5	0.14	68	25	6
min-max	0.15–0.69	17–85	8–51	8–26	0.85–1.3	42–284	38–119	19–37
n (samples)	23	24	23	21	10	10	10	8
P (*t*-Test)					0.000	0.002	0.000	0.000

the treatment honey groups as tested by sensorial analysis.

3. RESULTS

3.1. Natural content of formic acid, oxalic acid and free acidity

Table II summarises the measurements of total acidity, and formic and oxalic acid in all control honeys during the three trial years. The natural formic acid content varied between 17 and 284 mg/kg, while the oxalic acid content varied between 8 and 119 mg/kg. The average content of both acids and of the free acidity in honeydew honey was significantly higher than that of the group consisting of blossom honey and blends of blossom and honeydew honey. On average, honeydew honeys contained twice the amount of formic acid, almost three times more oxalic acid, and 60% more free acidity.

3.2. Effects of the treatments on formic and oxalic acid content and on free acidity

3.2.1. Normal treatments with formic acid in autumn

The effects of the formic acid treatments on honey residues over in three consecutive years are summarised in Table III. There was a significant increase in formic acid in the honeys from apiaries which had been treated during the preceding autumn. The overall average increase of formic acid content during the three year trial was 46 mg/kg, range 0–139 mg/kg.

In three different apiaries, formic acid treatments were carried out over three successive years. No increase of the formic acid content of honey after the repeated treatments was observed (Fig. 1). The lack of increase stems from the large natural variation in formic acid, so that small differences between the honeys

Table III. Influence of a formic acid treatment in autumn, carried out over three consecutive years, on the formic acid content of honey during the following year in different Swiss apiaries. Values in mg/kg. The honeys were of blossom and mixed blossom-honeydew type (see Materials and methods).

	1996		1997		1998	
	control	treatment	control	treatment	control	treatment
average	45	94	31	91	41	71
st. deviation	18	49	6	27	21	33
min-max	20–80	17–157	20–40	66–133	17–85	42–156
n (apiaries)	10	10	5	6	9	11
P (*t*-Test)		0.008		0.001		0.030

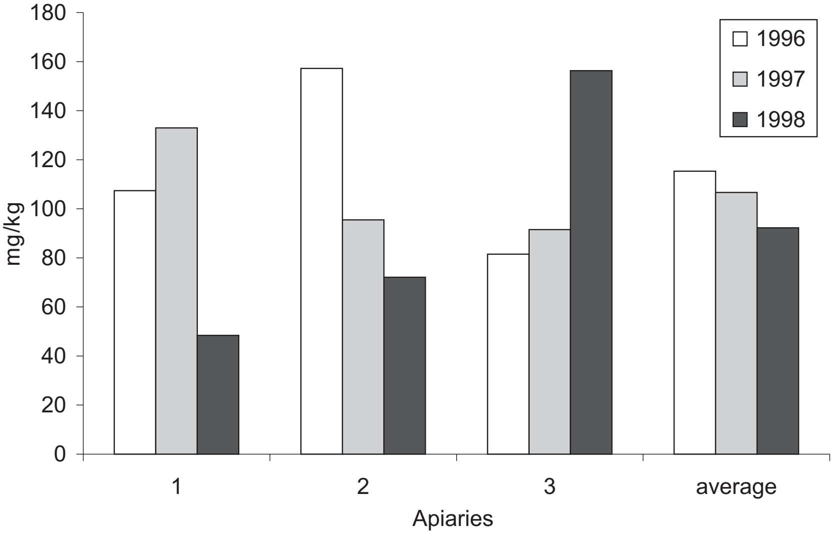

Figure 1. Residues in honey after repeated long-term treatment with formic acid in autumn during three consecutive years. The trials were carried out in three Swiss apiaries. The honeys were of blossom and mixed blossom-honeydew type. (Further details, see Materials and methods.)

Table IV. Influence of a formic acid treatment, carried out in spring, on the formic acid content and free acidity of honey, harvested during the summer of the same year. Formic acid (FA) values in mg/kg, free acidity (FAc) in meq/kg, Ap.: apiary number.

Ap.	Trial year, honey type	Control FA (FAc)	Treatment FA (FAc)	Increase of FA (FAc)
	1997			
1	honeydew	127 (27)	403 (30)	276 (3)
2	honeydew	89 (18)	506 (30)	417 (12)
	1999			
3	blossom, blossom-honeydew blends	20 (14)	58 (13)	38 (-)
4	blossom, blossom-honeydew blends	81 (21)	285 (22)	204 (1)
5	blossom and blossom-honeydew blends	42 (23)	261 (26)	219 (3)
6	blossom and blossom-honeydew blends	23 (25)	103 (26)	80 (1)
7	honeydew	47 (28)	166 (26)	119 (-)
	average	61 (22)	254 (25)	193 (3)

harvested in the apiaries during the three years could not be detected. Originally we had planned to carry out formic acid accumulation experiments in seven apiaries. However, in four of them honeydew harvest was encountered in one of the trial years. As only blossom and blossom-honeydew honeys were considered for our study, the results from these four apiaries could not be taken into account.

3.2.2. Emergency treatments with formic acid in spring

As described in Section 2.1.2, emergency treatments due to high *Varroa* infestation rate had to be applied in seven apiaries in Switzerland. The results of the trials are summarised in Table IV. The average increase in the formic acid content was 193, (range 38 mg/kg–417 mg/kg). In some cases the increase of formic acid could changed of in honey taste (see Sect. 4.5.2). On the other hand, these urgent treatments caused only a slight, statistically insignificant increase in free acidity; the 40 meq/kg limit was never exceeded.

3.2.3. Treatments with oxalic acid

The effects of the oxalic acid treatments on honey residues, over three consecutive years are summarised in Table V. There was no increase of oxalic acid in the honeys from apiaries which had been treated during the preceding autumn; both control and trial honeys had similar values. A combined treatment with spraying and trickling of oxalic acid in

Table V. Influence of an oxalic acid spray treatment in autumn, carried out over three consecutive years, on the oxalic acid content of honey, harvested during the following year in different Swiss apiaries. In 1998 trickling was carried out in addition to spraying. Values in mg/kg. The honeys were of blossom and mixed blossom-honeydew type (see Materials and methods).
s: spraying; s+t: spraying+trickling.

	1996		1997		1998		
	control	treatment	control	treatment	control	treatment s	treatment s+t
average	41	33	22	18	19	19	26
st. deviation	12	18	9	9	5	10	12
min–max	18–79	16–51	8–30	6–27	10–30	9–36	15–44
n (apiaries)	9	9	5	6	9	4	7
P (*t*-Test)		0.149		0.152		0.969	0.145

Table VI. Influence of consecutive treatments with formic acid (long-term) and oxalic acid (spraying), carried out in autumn in different Swiss apiaries and harvested during the following year, on free acidity of honey. Values in meq/kg honey. The honeys were of blossom and mixed blossom-honeydew type (see Materials and methods).

	1996		1997		1998	
	control	treatment	control	treatment	control	treatment
average	22	24	17	18	13	14
st. deviation	5	9	1	5	3	8
min–max	13–26	13–46	16–18	14–28	8–17	6–34
n (apiaries)	11	7	5	6	9	10
P (*t*-Test)		0.671		0.525		0.694

autumn also did not lead to a significant increase of the oxalic acid concentration in honey.

3.2.4. Influence of a combined treatment with formic and oxalic acid on honey acidity

In most apiaries both formic and oxalic acid were applied during the three trial years. The effect of these combined treatments on honey acidity is summarised in Table VI. The free acidity of the control and treatment samples was practically the same. Only one honey, with 46 meq/kg slightly exceeded the present limit of 40 meq/kg for free acidity.

4. DISCUSSION

4.1. Determination methods

Enzymatic assays for the determination of organic acids in honey have been used mostly as they are very specific and do not need expensive instrumentation. Our results showed that the measurements with the Böhringer oxalic acid kit has acceptable precision and recovery. In this test, both oxalic and formic acid can be determined in the same sample solution, which is very convenient. Alternatively, the Sigma company oxalic acid determination kit can be used (Mutinelli et al., 1997). HPLC (Del Nozal et al., 1998, 2000) and GC-MS (Unterweger et al., 2001) are valuable alternatives, that have been used for organic acid determination in honey, especially when more then one organic acid is determined. The comparability of the enzymatic and the HPLC measurements has not been strictly established, but the values found by both methods are similar. Indeed, Del Nozal et al. (1998 and 2000) measured formic and oxalic acid by HPLC and found formic and oxalic acid amounts, similar to the findings of all other researchers, who used enzymatic measurement (see Sect. 4.2).

4.2. Natural content of organic acids

Formic and oxalic acid are natural honey constituents. It is not clear to what extent they originate from bees or from the nectar, but it has been reported that oxalic acid is added to honey by the bees (Echigo and Takenaka, 1974). The formic acid content of honey has been determined by many researchers. Values from 9 to 1229 mg/kg have been found, the highest values being those of chestnut honey (Stoya et al., 1986; Kary, 1987; Talpay, 1989; Sabatini et al., 1994; Capolongo et al., 1996; Del Nozal et al., 1998). Weakly aromatic honey like acacia, rhododendron, and citrus have lower values than strongly flavoured ones like erica, honeydew, and chestnut honeys (Talpay, 1989; Sabatini et al., 1994; Capolongo et al., 1996), which coincides with our results.

There are few measurements of the natural content of oxalic acid in honey (Kary, 1987; Mutinelli et al., 1997; Del Nozal et al., 2000; Bernardini and Garda, 2001). Values from 8 to 300 mg/kg have been found in different honeys. Light honeys, such as rosemary and lavender, have a lower oxalic acid content than strong flavoured dark ones like honeydew and heather (Del Nozal et al., 2000), which also coincides with our results.

4.3. Residues after treatment with formic acid

In other publications, the formic acid residues after a long-term, 7-day formic acid treatments were measured in the sugar feed (Stoya et al., 1986; Krämer, 1994; Capolongo et al., 1996). In two studies (Stoya et al., 1986; Capolongo et al., 1996) a kinetic follow-up of residues after the treatment in autumn was carried out. The residues were measured before and after the treatment and were followed until spring. After a strong initial rise of formic acid in autumn, the levels in spring diminished to reach the values before treatment. In all of these studies it was not tested if the formic acid content of honey was significantly influenced. Our results show, that a 7 day treatment with formic acid leads to a small, but significant increase of formic acid in honey. This rise is not problematic, as it does not influence the quality of the honey (see Sect. 4.5). The increases in formic acid content were similar during all trial years, although the honey yields during these years were different. We think that that formic acid that remains in the sugar feed in the brood combs is the main reason for these residues. Such feed remains are common in Swiss type hives. Under beekeeping conditions in magazine hives, as used in other countries, smaller residues are expected.

In emergency cases with high mite infestation rate in spring, a treatment with formic is often necessary. Our results show that a treatment with formic acid in spring might lead to an increase of formic acid in summer honey that is near the taste threshold of this acid in honey (see Sect. 4.5.2). Thus, under moderate climate conditions, formic acid should be used generally during the period outside the honey flow, mostly just after the honey harvest in late summer.

4.4. Residues after treatments with oxalic acid

Other researchers (Radezki, 1994; Mutinelli et al., 1997; Del Nozal et al., 2000; Bernardini and Gardi, 2001) did residue studies after single oxalic acid treatments and could not detect residues in the sugar feed, compared to the levels found before the treatments. Our studies, carried out with honey, lead to the same conclusions. Even two oxalic acid treatments in autumn did not cause an increase of the oxalic acid concentration in honey of the next year.

4.5. Residues and honey regulations

4.5.1. Acidity

According to the present EU honey regulation (2002) and also according to the Codex Alimentarius draft for a new honey standard (2000), there is a maximum limit for free acidity of honey of 50 milliequivalents per kg honey.

Our results, as those of Stoya et al. (1986), show that long-term formic acid treatment in autumn according to the prescriptions will not increase honey acidity above the required limit.

4.5.2. Taste

According to the existing honey standards no substances are allowed to be added to honey to change its natural taste. The taste threshold for formic acid added to honey was determined for mildy flavoured blossom honeys and was found to be around 300 mg/kg (Capolongo et al., 1996; Bogdanov et al., 1999a). For stronger flavoured honeys like honeydew and chestnut honey it falls between 600 and 800 mg/kg (Capolongo et al., 1996; Bogdanov et al., 1999a). The residues after normal formic acid treatments in autumn are much lower than these thresholds, hence there is no risk for a change of honey taste due to an increase of formic acid concentration. However, according to our results, when emergency treatments with formic acid are carried out in spring, the formic acid residues in the summer honey might be close to the taste threshold of this acid. Therefore, this type of treatment should be avoided. It should be taken into consideration, that the honey yields in Switzerland are relatively modest (see Sect. 2.2) and in the Swiss type honey hive, there are often some small remains of autumn sugar feeds. In countries with greater honey harvests and different honey types the residues of formic acid should be theoretically smaller.

The honey taste will be changed only if about 400 mg/kg of oxalic acid is added to blossom honey or if 900 mg/kg is added to honeydew honey (Bogdanov et al., 1999a). Treatments with oxalic acid do not cause oxalic acid residues, therefore there is absolutely no danger for a change of honey taste due to oxalic acid treatments.

4.5.3. Maximum residue limits

In an EU regulation, formic acid and components of essential oils like thymol and menthol are defined as GRAS (Generally Recognised As Safe) substances, therefore it is not necessary to fix a MRL (EU Regulation 2796, 1995). Oxalic acid is a natural constituent of most vegetables and its content lies between 300 and 17 000 mg/kg, the highest content being that of parsley (Agricultural Handbook, 1984). Thus, most vegetables contain much higher amounts of oxalic acid than honey. Considering the small daily intake of honey, its contribution to the total daily intake of oxalic acid is negligible. From a nutritional point of view, oxalic acid, like formic acid, also should have a GRAS status. Moreover, no significant residues are expected after oxalic acid treatments.

4.5.4. Registration of oxalic acid for the control of V. destructor

To our knowledge spraying and trickling of oxalic acid is accepted for use against *V. destructor* only in two Western European countries (Switzerland and Finland) but is widely applied by beekeepers throughout Europe as the treatments are very efficient. The main registration problem of oxalic acid treatments seems to be the toxicity of oxalic acid for the person applying the treatment. Provided the necessary precautions are met, there is no risk for the user when spraying and trickling of this acid (Knuti, 1996). On the other hand, the user toxicity of the recently developed oxalic acid sublimation treatment (Radetzki and Bärmann, 2001) has not been evaluated.

No oxalic acid residues are to be expected after repeated field spraying and trickling use of this acid, therefore there are no objective arguments against the registration of these treatment modes for the control of *V. destructor.*

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Résumé – Détermination des résidus dans le miel après traitements aux acides formique et oxalique en conditions de plein champ. Jusqu'à présent les études de résidus ont été faites en conditions expérimentales contrôlées après une seule application d'acide formique (AF) et d'acide oxalique (AO). Ces acides sont maintenant régulièrement utilisés dans la lutte à long terme contre l'acarien *Varroa destructor.* C'est pourquoi nous avons fait des essais en plein champ avec l'AF et l'AO dans divers ruchers de Suisse au cours des années 1996, 1997 et 1998. L'AF et l'AO ainsi que l'acidité libre ont été déterminés dans les miels issus des expériences et dans des miels témoins de l'année suivante.

En fonction de leur conductibilité électrique, les miels ont été classés en deux groupes qui différaient par leur teneur naturelle en acide :

– groupe 1 : miels de fleurs, mélanges de miels de fleurs et de miels de miellat. Acide formique : moyenne 41, min. 17, max. 85 mg/kg. Acide oxalique : moyenne 25, min. 8, max. 51 mg/kg (Tab. II),

– groupe 2 : miels de miellat. Acide formique : moyenne 93, min. 42, max. 284 mg/kg. Acide oxalique : moyenne 70, min. 38, max. 119 mg/kg (Tab. II).

Deux traitements à long terme à l'AF ont été faits entre début septembre et fin octobre. Les augmentations de la concentration en AF qui s'ensuivirent ont été mesurées durant les trois années d'expérimentation : moyenne 46, min. 0, max. 139 mg/kg (Tab. III). Cette augmentation n'est pas problématique car le goût du miel ne change qu'à partir de 300 mg d'AF par kg de miel. Il n'y a pas eu d'augmentation de la teneur en AF du miel au fur et à mesure des traitements (Fig. 1).

Lorsque des traitements d'urgence ont été effectués à l'AF au printemps, l'augmentation de la concentration en AF dans les miels d'été de la même année était beaucoup plus forte : moyenne 193, min. 38, max. 417 mg/kg (Tab. IV). Des résidus d'un tel niveau peuvent modifier le goût du miel, ce que les règlements internationaux sur le miel n'autorisent pas.

De l'AO a été vaporisé durant la période sans couvain entre début novembre et fin décembre. Il n'y a pas eu d'augmentation de la concentration en AO, même lorsqu'un second traitement par dégouttement était effectué au cours de la même période (Tab. V).

Les traitements successifs à l'acide formique et à l'acide oxalique indiqués ci-dessus n'ont pas augmenté l'acidité libre du miel récolté l'année suivante (Tab. VI).

miel / résidu / acide formique / acide oxalique / acidité libre / ***Varroa destructor***

Zusammenfassung – Bestimmung der Rückstände in Honig nach wiederholten Behandlungen von Bienenvölkern mit Ameisen- und Oxalsäure. Die Rückstände von Ameisen- und Oxalsäure in Honigen wurden bisher nach lediglich einzelnen Anwendungen dieser Substanzen gegen Varroamilben unter kontrollierten Bedingungen bestimmt. Heute werden diese Säuren in der imkerlichen Praxis in der Regel alljährlich zur Bekämpfung der Varroamilben eingesetzt. Deshalb wurden zwischen 1996 und 1998 auf mehreren Bienenständen in der Schweiz Rückstandsmessungen nach wiederholten Anwendungen von Ameisen- und Oxalsäure durchgeführt. Folgende Parameter wurden in Versuchs- und Kontrollhonigen des nächsten Jahres bestimmt: Ameisensäure, Oxalsäure und freie Säure. Aufgrund der Messung der elektrischen Leitfähigkeit wurden die Honige in zwei Gruppen aufgeteilt, welche sich im natürlichen Gehalt von Ameisensäure und Oxalsäure unterscheiden:

Gruppe 1, Blütenhonige und Mischhonige aus Blüten- und Waldtracht:

Ameisensäure: Durchschnitt 41, Min.17, Max. 85 mg/kg, und Oxalsäure: 25, Min. 8, Max. 51 mg/kg, (Tab. II).

Gruppe 2, Honigtauhonige:
Ameisensäure: Durchschnitt 93, Min. 42, Max. 284 mg/kg, und Oxalsäure 70, Min. 38, Max. 119 mg/kg (Tab. II).
Zwei Langzeit-Ameisensäurebehandlungen wurden jeweils in der Zeitspanne September-Oktober durchgeführt. Folgende Zunahmen der Ameisensäurekonzentration wurden während der dreijährigen Versuchsperiode gemessen: Durchschnitt 46 mg/kg, Min. 0, Max. 139 (Tab. III). Dieser Zuwachs ist unproblematisch, weil die Wahrnehmungsschwelle von Ameisensäure im Honig bei rund 300 mg/kg liegt und nicht erreicht wurde. Mit zunehmender Anzahl der Behandlungsjahre gab es keine Zunahme der Ameisensäuregehalte (Abb. 1).
Bei Notbehandlungen im Frühjahr gab es hingegen beträchtlich grössere Zunahmen der Ameisensäurekonzentrationen: Durchschnitt 193 mg/kg, Min. 38, Max. 417 (Tab. IV). Diese grossen Zunahmen können dazu führen, dass die Wahrnehmungsschwelle überschritten und der natürliche Honiggeschmack verändert wird. Dies ist nach der internationalen Honiggesetzgebung nicht zulässig.
Oxalsäurebehandlungen (Sprühen) wurde im brutlosen Zustand zwischen November und Dezember während 3 Jahren durchgeführt. Es gab keine Zunahme der Oxalsäuregehalte im Honig, auch wenn eine zweite Behandlung durch Träufeln während der gleichen Periode durchgeführt wurde (Tab. V). Nacheinander folgende Behandlungen mit Ameisen- und Oxalsäure im Herbst erhöhen den Säuregehalt des Honigs des nächsten Jahres nicht (Tab. VI).

Honig / Rückstände / Ameisensäure / Oxalsäure / freie Säure / *Varroa destructor*

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