# Test results

I run a large beekeeping operation with around 1,000 hives in Sardinia and Germany, and have conceived the general concept of influencing the natural frequency of bodies with high-frequency sound. In April 2014 we started to investigate the frequency to which bees and mites respond. To do that, I had to identify the sound frequencies of bees and mites. The treatment does not harm bees at all: neither the brood and queen nor bee behaviour are affected.

We placed bees in a viewing box and exposed them to various frequencies. Staff sat in front of this box and ran through all the frequencies, observing the behaviour of bees and varroa mites. In our investigation we found that bees responded at 2,000 to 8,000 hertz. Bees do not respond to frequencies above 9,000 hertz at all. Mites react at 12,000 to 17,000 hertz. The ideal frequency is 14,000 to 15,000 hertz. We also found that wave form is critical. A square wave is much more effective than a sinus wave. We need a sound frequency of at least 35-40 decibels within the brood. At that level the juvenile mites (white mites) die within 2 days (reproduction is interrupted). Old mites (adults) need at least 80 decibels, then they die within 2-8 days. This means that even when the adult mites are in the brood they are in a protected space, and so all aspects of treatment, especially the time of application, need to be taken into consideration.

The device is placed directly on top of the combs. An empty frame and then the lid are placed on top of the device. In practice, the treatment is carried out at least twice a year. Because of reinfection, all the hives in the apiary need to be treated at the same time. The ideal time is when there are no more than two frames in the hive, so that the sound pressure reaches the bottom frame as well. In winter, efficacy is around 95% or more, and around 90% from February to April and from July to October, from a single treatment. The device can also be used in the case of severe varroa infestation in summer when the honey chambers are also in the hives, because ultrasound does not harm the honey. However, under these conditions the efficiency is only around 70%, because the decibel output falls the further the device is from the lowest brood frame. In this situation we recommend reapplying the sound device again immediately (after recharging the battery). That raises the overall efficacy rate to +/- 95%. It is also important that all the hives in the apiary are treated at the same time, so each hive needs its own device. Beekeepers with several apiaries can use one device for two apiaries.

The device is fitted with a rechargeable dry battery. One battery charge is enough for the total treatment duration of 40-45 days.

In October 2016, all the test series and sample tests had been completed, our Varroa-Killer-Sound was ready for mass production and I had applied for a patent. Starting in November/December 2016 tests of the Varroa-Killer-Sound were carried out independently by myself, a large-scale beekeeper, H. J. Morhart from Bad Hindelang, and the senior health officer Mr. Dörler from Vorarlberg. The tests were carried out over the winter (December/January). The results were identical in all groups. The bees remained in their winter cluster underneath the sound device. They were perfectly calm and did not consume more feed than normal or move to another location.

Results obtained by Mr. Dörler, Vorarlberg senior health officer:

# Varroa mite infestation rates

٦,

Treatment with Varroa Killer-Sound

Day	Hive 1	Hive 2		Hive 4		Hive 6	Hive 7	Hive 8		
-1	0	0	0	0	0	0	2	0	1	0
-2	0	1	1	0	0	0	0	1	0	1
-3	0	0	0	0	0	0	0	0	0	0
1st total	0	1	1	0	0	0	2	1	1	1
1	0	1	1	о	1	0	0	o	0	0
2	1	2	0	0	0	1	0	0	2	0
3	0	1	0	0	0	0	1	1	3	0
4	0	0	0	0	0	0	0	1	0	1
5	0	0	0	0	0	0	1	3	0	0
6	0	0	0	0	0	0	0	2	2	0
7	0	1	0	0	0	0	0	3	1	0
8	0	1	1	0	0	1	1	0	0	0
9	0	0	0	0	0	0	1	1	2	0
10	0	0	5	0	6	1	2	2	3	2
11	0	0	0	0	0	0	0	0	0	1
12	0	0	0	0	0	0	0	2	0	0
13	0	0	0	0	0	0	0	0	0	1
14	0	0	2	0	0	1	1	0	0	0
15	0	0	2	0	2	0	1	0	1	0
16	0	2	0	0	0	0	0	0	0	0
17	0	0	0	0	0	0	1	0	0	0
18	0	0	0	0	2	0	3	0	0	0
19	0	0	0	0	0	1	0	1	0	0
20	0	0	0	0	0	0	2	1	0	0
21	. 0	0	2	0	0	0	1	0	1	0
22	0	0	2	0	0	0	0	0	0	0
23	0	0	1	0	1	0	0	0	0	0
24	0	1	0	0	0	0	0	0	0	0
25	0	0	1	0	0	0	1	0	0	0
2nd total	1	9	17	0	12	5	16	17	15	5
26	0	0	0	0	0	0	0	0	0	0
27	0	0	0	0	1	0	0	1	0	· 0
28	0	1	0	0	0	0	0	0	0	· 0
29	0 0	0	0	0	0	0	0	1	0	0
30	0 0	0	0	0	0	0	1	5	0	0
Total	0	1	0	0	1	0	1	7	0	0

Mr. Dörler stated that just beforehand he had conducted a very good winter mite elimination in his hives and was surprised to find so many mites despite this. He thought that he had no or very few mites. The results for hive 8 are particularly interesting. He was very satisfied with the result, and had not noticed anything unusual about the hives.

Day	Hive 1	Hive 2	Hive 3	Hive 4	Hive 5	Hive 6	Hive 7	Hive 8	Hive 9	Hive 10
23.12.	1	2	3	4	8	9	11	12	17	18
24.12.	2	3	1	4	9	8	4	3	0	1
26.12.	4	3	2	5	6	5	5	5	2	2
27.12.	2	1	3	4	9	3	8	3	3	4
29.12.	0	1	1	4	5	1	2	2	2	2
30.12.	2	0	0	6	7	2	4	3	4	3
01.01.	1	1	2	5	13	3	11	4	2	1
04.01.	0	2	2	4	8	2	6	4	3	3
07.01.	1	0	1	4	7	1	5	3	2	3
10.01.	0	1	2	3	3	1	3	3	1	3
12.01.	0	0	1	4	1	0	1	3	2	1
15.01.	0	0	0	0	0	0	0	1	0	1
16.01.	0	0	0	0	0	0	0	0	0	0
20.01.	0	0	0	1	2	0	0	1	0	0
Total	13	14	18	48	78	35	60	47	38	42

Results obtained by Mr. H.J. Morhart, large-scale beekeeper from Bad Hindelang:

Oxalic acid applied on 20.01.2017 to eliminate remaining mites. Mite numbers decreased after a total of 4 days.

Natural overall decrease in mit numbers within 5 days before treatment of the same hives.

Mites	Hive no.
1	1
1	2
2	3
2	4
2	5
2	6
3	7
2	8
3	9
1	10

54

This shows that mite elimination in winter is very effective. In addition, the hives did not produce any more broods, and so the mites were unable to hide there. The bees remained perfectly calm underneath the sound devic. On 01.04.2017 we set up 164 Varroa-Killer-Sound devices in Sardinia. Before setting up the devices we tested the hives with the icing sugar method, which is not very accurate but does demonstrate efficacy. The hives had an average of 5-7 mites per sample.

On removal on 28/29 April, the hives were tested again 122 samples with 0 mites 42 samples with 1 -2 mites

Baseline figures on 01.04.2017

Average bee weight approx. 2 kg bees per hive: 50 gr. Sample = 40 x avg. 6 mites = 240 mites 240 mites + 20% margin of error = 288 mites – start (with no mites in the brood)

Figure on 29 April 2017 - only samples with mites

Average bee weight approx. 2 kg bees per hive: 50 gr. sample = 40 x avg. 1.5 mites = 60 mites + 20% margin of error = 72 mites – end 288 = 100% residual mites = **75% efficacy rate** 

When applied twice:216 mites (first run = 75% efficacy rate) + 54 mites (second run = 75%efficacy rate)

Total: 270 mites Residue approx. 18 mites = 270: 2.88 = approx. 94%

### efficacy rate. Sample of all hives together:

Average bee weight approx. 2 kg. bees per hive 50 g sample = 40 x avg. 0.3 mites = 12 mites + 20% margin of error = 14.5 mites – end 288 = 100% residual mites = **95% efficacy rate when applied once** 

It must also be borne in mind that approximately 50% of the bees in our apiary are foraging bees. Varroa mites stop feeding when exposed to our sound emissions. This weakens them considerably and they lose their grip on bees during honey flow and when the bees are flying. We should assume that many of them are lost because of this, though it is not possible to measure the loss. It is also important to bear in mind that during the treatment period the number of mites in the hive would have doubled each month (without treatment).

30 control hives which were not treated and had the same baseline values had between 20 and 25 mites per sample on 29 April 2017.

#### Then 2 series of tests were carried out to determine efficacy:

Use of 10 Varroa-Sound devices in Maierhöfen from 23.05.2017 to 01 July 2017 (40 days), to 01 July 201

Natural daily die-off before treatment: 3-4 mites per day on average.

The hives were heavily engaged in collecting nectar from forests, and had 8 - 10 brood combs. No effect on bee behaviour or the brood was detected. In some cases, the bees placed wax on the sound-emitting devices. This had no impact on efficacy. The hives were using 3 - 4 frames while exposed to sound (1 brood chamber and 2-3 honey chambers). The batteries were replaced on 21.06.2017.

Mites fallen during sound emission:

Hive no.	Number of mites falling	Number of mites falling in	Estimated remaining	Total mites	Efficacy rate
	during sound	5 days by oxalic			
	emission		mites not falling		
		emission	as a result of		
			oxalic acid		
1	578	304	50	932	62%
2	645	365	50	1060	61%
3	572	270	50	892	64%
4	516	264	50	830	62%
5	659	320	50	1099	60%
6	759	390	50	1029	73%
7	524	255	50	1199	44%
8	622	322	50	999	62%
9	497	264	50	811	61%
10	594	295	50	939	63%

10 control hives with the same baseline values not exposed to sound emission. The following mite fall figures were observed during oxalic acid treatment:

Because of the large number of mites, the figures were counted to within +/- 50 mites.

Hive	Mites
1	1650
2	1550
3	1210
4	1580
5	1450
6	1250
7	1230
8	1350
9	1460
10	1280

Following the test period, control hives which had the same starting conditions were found to have between 1,200 and 1,500 mites per hive at the time of residual mite elimination (plus mites in the brood)

# A beekeeper from the Chiemgau region published the following series of tests:

Numbers of mites fallen in the 5 days immediately before the use of the Varroa-Sound device were 23 (avg. 4.6 mites/day). Sound emission started on 29 April. Hive size: 1 brood chamber and 2 honey chambers

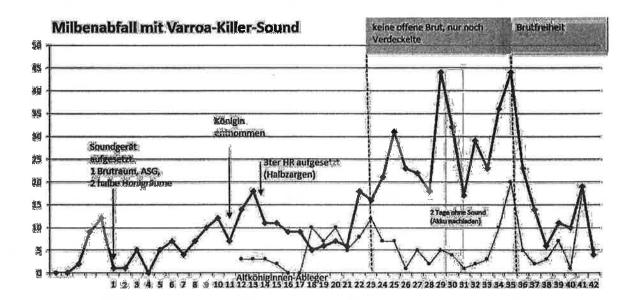
# Effects on bees

No unusual bee behaviour was observed during the test period.

# Effects on mites

The effects on mites were only observed by means of the "sticky board" method.

Mite fall remained low in the first 3 days, but then rose slowly each day. On day 4 we started to coat the discs with oil, as ants and other creatures were taking material from the debris (which is probably why there were no mites on day 4).



Milbenabfall mit Varroa-Killer-Sound	Mites fallen with Varroa-Killer-Sound		
Keine offene Brut, nur noch Verdeckelte	No open broods, now only capped broods		
Brutfreiheit	No broods		
Soundgerät aufgesetzt. 1 Brutraum, ASG, 2 halbe Honigräume	Sound device in place. 1 brood chamber, queen excluder, 2 half honey chambers		
Königin entnommen	Queen removed		
3ter HR aufgesetzt (Halbzargen)	3rd honey chamber inserted (half-frames)		
Altköniginnen-Ableger	New hive with old queen		
2 Tage ohne Sound (Akku nachladen)	2 days without sound (battery recharge)		

# During the test

During the 42 days, the loudspeaker frequently fell from the battery and came to rest directly on the top supports. This had no noticeable effect on fall statistics.

On day 11 the old queen was removed and a separate chamber for the old queen was set up. Counts were performed every day on this separate chamber as well until the end of the test, and the results are shown in red in the remainder of the graph. This chamber was not exposed to sound.

An additional half-sized honey chamber was inserted on day 13. The mite count fell dramatically after this! It is clear that additional space significantly reduces the effectiveness of the sound.

On day 20 a young queen emerged but was lost during the mating flight.

The removal of the old queen on day 11 meant that there was no longer any open brood present from day 22 onwards, and the mites could no longer hide from the sound. As a result, the incidences of varroa mites falling rose again sharply.

The sharp decline on days 30/31 was caused by a 2-day interruption of sound emission. During this time the battery was being recharged.

Once the hive was brood-free from day 36 the number of fallen mites dropped significantly, and stood at only 4 mites on day 42. The test was concluded at that point.

Over the span of 42 days a total of 616 mites fell from the hive exposed to sound.

# After the test - degree of efficacy

A total of **371 mites** fell in the 5 days after the oxalic acid treatment. Even when no brood is present, a single application of oxalic acid is not sufficient to eliminate all mites from the hive. We therefore assumed that at that stage there could still be about 50 mites in the hive.

This means that the Varroa Sound device	has the following efficacy results:
Mites during the sound phase	616
Mites after the test by oxalic acid	371
Estimated residual mite numbers	50
Total mites	1,037
Level of efficacy: 616 x 100 / 1,037	59% (around 83% if used twice)

It is true that a simple count is not completely accurate because the mites continued to breed during the long period, but it should be accurate enough for an assessment.

#### Beekeeper's conclusion

The sound certainly does have a fatal effect on the mites. Our findings therefore clearly run counter to tests conducted by other beekeepers who found the sound to be ineffective. The effect does exist - but is not sufficiently high (in this test!). As indicated on the manufacturer's home page, the device becomes much less effective the further away from the brood it is placed (because of honey chambers). This means that it may not be as effective when used while bees are collecting nectar.

# **OVERALL CONCLUSIONS:**

The ideal time for use was clearly found to be from February to mid-April and from mid-July to October. Between mid-April and mid-July the volume of nectar brought back to the hive and the size of the population meant that efficacy was markedly reduced, but nevertheless still sufficient to reduce the level of mite infestation in the hives. Use during the brood-free period (winter) in November and December is also recommended because of very high efficacy rates.

A single use produces a satisfactory effect.

Two uses in succession produce a very good effect.

#### This is an alternative form of treatment that has now been proven to be effective.

For information: The devices are currently undergoing neutral and independent testing at the Bavarian Regional Institute for Wine Production and Horticulture in Veitshöchheim and at Berlin Technical University's Bee Research Centre.

Results will probably be available and published in August / September 2017.

# Evaluation and assessment of this set of tests

These results are almost identical to the first initial results obtained by the Bee Centre of the Bavarian Regional Institution for Viticulture and Horticulture in Veitshöchheim, which differed in that only one device has been used so far. The final results from Veitshöchheim with 12 devices will not be available until mid-August, we will publish the results once they are sent to us. In general, the effect of the device is regarded as good. When the device is used twice in succession, the efficacy rate is around 90%. This can be achieved without difficulty as the device does not harm the bees in any way, and this produces a very good effect. It must also be borne in mind that approximately 50% of the bees in our apiary are foraging bees. Our treatment stops varroa mites from feeding. This weakens them and causes them to lose their hold on bees while they are foraging and flying. It must therefore be assumed that many varroa mites on foraging bees are lost outside the hive. It is difficult to measure the extent of this loss, but it would increase the efficacy rate. The diagram for example shows very clearly that without sound application the rate of varroa mite fall decreased dramatically on days 30-31. This is strong confirmation of the efficacy of Varroa-Killer-Sound. Furthermore, removal of the queen (and allowing the brood to hatch) makes it very easy to calculate the total number of mites in the hive in order to ascertain efficacy. However, in everyday beekeeping this is not necessary. It must also be noted that in hives with 3 or 4 frames, the sound no longer penetrates sufficiently into the lower brood frame, and that in the capped brood we no longer have the necessary sound level of at least 35 decibels which we require for the juvenile mites in the brood. This means that in these 3- or 4frame hives mites continue to reproduce at quite a high rate. Nevertheless, the overall effect is good. But this also means that hive demiting should ideally be carried out on 1- to 2-frame hives. Another point is that if the hives had not been treated the number of mites would have doubled in each treatment cycle. At the moment, we are using approximately 500 devices in 2 treatment cycles of 30 days each.

We performed a test using the icing sugar methods (which is not very accurate, but which records efficacy) on 164 hives on 1 April before introducing the Varroa-Killer-Sound, and had 5-7 mites per sample. When the devices were removed on 28/29 April, we had 122 samples with no mites, and the other samples contained 1 or 2 mites each in the case of 2-frame hives. This means that the efficacy rate is over 90%. We have strong, healthy hives with few or no mites. Normally in April / May we would be suffering very heavy varroa infestation with a large number of crippled bees and total hive loss even at this early stage. It should be borne in mind that our hives are in Sardinia, and that we have 4-7 brood combs in December and 3-4 brood combs in January, which means that we have very high varroa reproduction. This is no longer the case.

Beekeepers are currently using approximately 10,000 devices with very good results and feedback.

This device is and remains a revolution in the treatment of varroa, and is still the alternative method of varroa control.

Sound measurements with these apps are very inaccurate. The decibel level at a distance of approximately 2 cm is +/- 100 decibels for all devices, but at least 95 decibels.

The ideal treatment time is still February/March to April and from July/August to September/October. Success rates in winter are also very good. At this period, the efficacy rate is approximately 90% per treatment cycle. It is true that the efficacy rate falls with each additional frame that is inserted. This is why 1-2 frames is ideal, although the number of mites killed is higher in hives with more frames. With best wishes

121

e#

Master beekeeper Rainer Krüger

Bienen-Varroa-Sound GmbH Untersteig 3 D-88167 Maierhöfen Tel.: +49 (0)8383 9292968 info@varroa-sound.de www.varroa-sound.de