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Effect of Different Concentrations of Oxalic Acid in the Fallen Mites Mean on the Hive Board During Spring and Early Summer Season

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Abstract. The results of the current study showed that the lowest mean of falling mite on the hive board was recorded in the 4.2% concentration treatment which amounted to 0.16 mite compared with the control treatment which recorded the highest mean which was amounted to 4.69 mite. As for the mean number of the expected mite inside the hive, the highest mean was recorded in the control treatment which amounted to 468.6 mite compared with the 4.2% concentration treatment in which the lowest mean was recorded, which amounted to 15.69 mite. As for the relative effectiveness, the highest average was recorded in the control treatment, which amounted to 66.67%, compared with the lowest mean of the 4.2% concentration treatment, which amounted to 17.75%. The above results were reflected in the treatment efficiency to regest the highest mean of efficiency in the 4.2% concentration treatment with a mean of 60.47% compared with the lowest average in the control treatment of 9.47%, knowing that the statistical analysis showed the moral superiority of all oxalic acid treatments over the control treatment.

Keywords: *Varroa* , Oxalic Acid , Mite, relative effectiveness, fall mite number, relative efficiency.

Introduction

The honey bee*, Apis mellifera* , like other animals, is exposed to a myriad of stress factors as it is exposed to infection with many pathogens (Brosi et al., 2017). *Varroa destructor* is the main pest of honey bees, as the bee population in *Varroa*-infested colonies significantly deteriorates (Figen et al., 2012). Among the factors, the use of pesticides is in general exposed to and experimented with the use of a group of organic acids including oxalic acid, whose effectiveness reached 93.7% to reduce mites on bee colonies (Goulson et al. 2015), Girişgin and Aydin (2010). Nanetti (2003) explained that using the injection trickling method of oxalic acid by mixing 42 gm of oxalic acid / liter of a sugar solution with a concentration of 60%, that is, at a concentration of 4.2% g / l and at a dose of 5 ml /, bee space between two frames with the highest concentration gave a killing rate of 96.8 compared with a killing rate of 89.5% for a concentration of 2.1. Aboushaara et al. (2017) showed that the effectiveness of oxalic acid ranged from 70.12% to 93.4% when oxalic acid was used in the form of solutions and at concentrations of 0.5%, 2.9%, 3% and 3.2%, where the concentration of 3.2% of the acid gave the highest killing rate of 95.81% while the killing rate of the two concentrations of 4.2% and 2.1% was 81% and 46%, respectively both of which were used by trickling, evaporation or immersion of cellulose strips in oxalic acid solution.

Materials and Methods

All experiments were designed according to the Randomized Complete Block Design, divided into four blocks. Each block had three replicates (colonies). A concentration of 60% sugar syrup was prepared in order to use a percentage of to dissolve the amounts of oxalic acid prescribed for each treatment. Then, it was divided into four groups; each group was 200 ml of sugar solution. The first group is the control group, the second group was the 3.0% concentration where 6 gm of oxalic acid was added to 200 ml, the third group was the 3.5% concentration where 7 gm was added to 200 ml while the fourth group was to prepare the 4.2% concentration where 8.4 gm was added to 200 ml. The number of fallen mites was daily calculated using the IPM/PUS board divided into white and black squares (spaces) manufactured by IPM INC sticky monitoring board at the UK where the study was photographed. The IPM monitoring board was designed with a Lancastroth hive board area as shown in (Fig.1). The plate was placed on a wooden slide under the screen bottom board for each replicate, and the

number of mites falling on the white squares (areas) was calculated and the result was multiplied by three to get the total number (Sammataro et al. 2000). The following equations were used to get the studied results and characteristics according to the method of Moretto and Mello, (2000):

1- The expected number of mites in the hive = natural fallen mean * 100.

2- Relative effectiveness = fallen after treatment – pretreatment natural fallen mean / fallen after treatment *100.

3-The treatment efficiency = the treatment dead mite / dead mite total (the fallen mite for each treatment + the fallen mite after the treatment).

Fig 1. The IPM INC sticky monitoring board is designed with a Lancastroth board area on top of a wooden slide under the screen bottom board.

Results and discussion

The numbers of mites in spring and summer are closely related to the results obtained at the end of the fall. The results of Table (1) show a decrease in the numbers of mites when reading on 2/17 and in all treatment. As the lowest mean was recorded in the concentration treatment 4.2% was 0.33 mites; however, it did not significantly differ according to Duncan's test from the control treatment with a mean of 2.33 mites which did not significantly differ from the treatments concentrations of 3.0% and 3.5% with their mean of 0.67 mites respectively, for each of the above mean which were recorded at a mean of 7.4°C and70.65% for temperature and relative humidity respectively. The weekly table: The increase in the number of mites when reading on 3/3 to a mean of 7.33 mites in the comparison treatment, which significantly differed from all oxalic acid treatments, where the lowest mean was recorded in the 4.2% concentration treatment amounted to 0.33 mites, but it did not significantly differ according to Duncan's test for 3.0% concentrations and 3.5% with their mean of 2.33 and 1.0 mites. Thus, the results continued to fluctuate up and down with the progress of the season and with the change in external temperatures and relative humidity when read on 3/24 to record the highest average number of mites falling on the ground. The number of cells in the compared treatment during the season amounted to 7.67 mites which did not, according to Duncan's test, significantly differ from the 3.0% concentration treatment with a mean that rose to 4.67 mites. The mean of zero were continuous in the majority of readings during the season where the mean temperature at the above reading which was 15.70 °C and relative humidity 73.54. The results of the table show the continuation of recording the lowest mean of zero in the concentration treatments 3.5% and 4.2% with the progress of the season and up to reading 19/5; yet, it did not, according to Duncan's test, significantly differ from the average comparison treatment in which the mean number of mites decreased to 3.33 mites and the 3.0% concentration treatment was amounted to 0.33 mites. Perhaps, and because of the environmental conditions the wire, mesh base played a role in reducing the numbers of mites in the comparison and concentration treatment to score 3.0%, as the mean. The temperature was 17.80 ° C and relative humidity was 43.45%. The results of the abovementioned table show a slight increase in the number of mites in all treatment at the end of the season when read on 9/6/2021, and the highest average was 6.67 mites in the control treatment which significantly differed according to Duncan's test from oxalic acid treatments, and recorded the lowest mean in the 4.2% concentration treatment which was 0.33 mites. However, it did not significantly differ according to Duncan's test from the 3.0% and 3.5% concentration treatment with a mean of 1.33 mites respectively for each where the mean temperature at this reading was 29.25°C and the relative humidity was 29.0%. Accordingly, it can be said that the reason for this rise in all treatment was the great brood activity in all replicates, but the numbers of mites in oxalic acid treatment were considered acceptable and below the level of general equilibrium compared with the control treatment that was somewhat high or due to the reproductive cycles of mite continuously within the treated replicates, and that the falling numbers reflect the extent of mite activity and its development within the replicates depending on the natural death rates of mite and the healthy behavior of honey bee workers, which was originally associated with the exit of the mite females found in the wax cell with the newly hatched workers which was not affected by pesticides .

As for the general average of the numbers of mites falling on the base of the hive, the concentration

treatments 3.5% and 4.2% were significantly superior according to Duncan's test, which were the lowest mean amounted to 0.35 mites and 0.16 mites respectively compared with the 3.0% concentration treatment with an average of 1.17 mites which was also significantly superior according to the Duncan's test on the control treatment in which the highest average of 4.69 mites was recorded. This showed that the effect of oxalic acid in its concentrations affected mite activity and reduced its numbers to below the general equilibrium level.

Table 1. The effect of different concentration of oxalic acid on the fallen mite during spring and early summer season /2020.

	Fallen mite number in the hive																	
date treatment	2/17	2/24	3/3	3/10	3/17	3/24	3/31	4/7	4/14	4/21	4/28	5/5	5/12	5/19	5/26	6/2	6/9	total
Control	2.33 e-k	5.67 a-e	7.33 a-b	5.33 a-f	3.67 a	7.67 a	3.00 e-k	5.33 a-f	7.00 a-c	4.00 c-i	3.33 e-k	4.33 b-h	2.33 e-k	3.33 e-k	3.67 d-j	5.00 a-f	6.67 a-d	4.69 a
3.0% Concentration	0.67 i-k	4.33 b-h	2.33 e-k	0.00 k	0.33 jk	4.67 a-g	2.00 f-k	2.00 f-k	3.00 e-k	4.67 a-g	0.33 jk	0.33 jk	0.33 jk	0.33 jk	1.33 g-k	1 gh	1.33 g-k	1.71 b
3.5% Concentration	0.67 i-k	1.00 h-k	1.00 h-k	0.67 i-k	0.00 k	0.00 k	0.00 k	0.00 k	0.00 k	0.00 k	0.00 k	0.00 k	0.00 k	0.00 k	0.67 i-k	0.67 i-k	1.33 g-k	0.35 c
4.2% Concentrati	0.33 i-k	0.67 j-k	0.33 jk	0.33 j-k	0.00 k	0.00 k	0.00 k	0.00 k	0.00 k	0.00 k	0.00 k	0.00 k	0.00 k	0.00 k	0.00 k	0.67 i-k	0.33 jk	0.16 c
total	2.58 a-c	1.92 a-f	2.67 ab	1.58 a-f	1.00 c-f	3.08 a	1.25 b-f	1.83 a-f	2.50 a-d	2.17 a-f	0.92 d-f	0.67 f	0.67 f	0.83 e-f	1.42 b-f	1.83 a-f	2.42 a-e	

As for the number of expected mites inside the hive which was a reflection of the previous results, the results of the Table (2) shows that the highest average of the numbers of expected mites inside the cell was recorded in the control treatment when read on 3/24/2021 which amounted to 767 mites. This did not significantly differ according to Duncan's test from the concentration treatment 30 in which the mean ability of 467 mites was recorded while the concentration treatments 3.5% and 4.2% were significantly superior according to Duncan's test to the comparison treatment continuously during the season and until the reading on 5/26 where the mites reappeared in the concentration treatment 3.5% and recorded an average capacity of 67 mites which did not significantly differ from the two treatments of concentration 3.0% with a mean of 133 mites and the comparison treatment in which the highest mean of 367 mites was recorded where the mean temperature was 19.65 °C and a relative humidity of 40.16%. Continuous oxalic acid after applying the treatment on 17/2/2021 when read on 9/6/2021 where the lowest mean was recorded in the concentration treatment 4.2% which was 33.0 mites did not significantly differ according to Duncan's test from the 3.0% and 3.5% concentration treatments. With mean adults of 133.0 mites for each of them respectively, all of them outperformed the comparison treatment in which the highest average was recorded which was 667.0 mites where the mean temperature was 29.25°C and the relative humidity was 29.0%.

As for the general mean of the treatments according to Duncan's test, concentrations 3.5% and 4.2% with their mean of 35.29 and 15.69 mites were significantly superior to the control treatments and the 3.0% concentration, in which the highest mean of 468.6 and 170.6 mites were recorded respectively. 3.5% and 4.2% reduced the number of mites below the general equilibrium level in the treated sects.

Table 2. The effect of different concentrations of oxalic acid in the expected number of mites in the hive during the early spring and summer / 2021.

	The expected number of mites in the hive																	
date treatment	2/17	2/24	3/3	3/10	3/17	3/24	3/31	4/7	4/14	4/21	4/28	5/5	5/12	5/19	5/26	6/2	6/9	total
Control	233 e-k	567 a-e	733 a-b	533 a-f	367 a	767 a	300 e-k	533 a-f	700 a-c	400 c-i	333 e-k	433 b-h	233 e-k	333 e-k	367 d-j	500 a-f	667 a-d	468.6 a
3.0% Concentration	67 i-k	433 b-h	233 e-k	0.00 k	33 jk	467 a-g	200 f-k	200 f-k	300 e-k	467 a-g	33 jk	33 jk	33 jk	33 jk	133 g-k	1 gh	133 g-k	170.6 b
3.5% Concentration	67 i-k	100 h-k	100 h-k	67 i-k	0.00 k	0.00 k	0.00 k	0.00 k	0.00 k	0.00 k	0.00 k	0.00 k	0.00 k	0.00 k	67 i-k	67 i-k	133 g-k	35.29 c
4.2% Concentrati	33 i-k	67 j-k	33 jk	33 j-k	0.00 k	0.00 k	0.00 k	0.00 k	0.00 k	0.00 k	0.00 k	0.00 k	0.00 k	0.00 k	0.00 k	67 i-k	33 jk	15.69 c
total	258.3	191.7	266.7	158.3	100	308	125.0	183.3	250.0	216.7	91.7	66.7	66.7	83.3	141.7	183.3	241.7	

	a-c	a-f	ab	a-f	c-f	a	b-f	a-f	a-d	a-f	d-f	f	f	e-f	b-f	a-f	a-e

The results of the Table (3) regard the relative effectiveness of the control materials which are directly proportional to the number of mites, and show that the highest mean of the relative effectiveness was recorded in the 3.0% concentration treatment amounted to 86.0%, and when read on 24/2/2021, that is after applying the treatment with oxalic acid. It significantly outperformed the rest of the treatments to record the lowest average in the concentration treatment 3.5% amounted to 59.88%, compared with 71.25% and 67.0% in the comparison treatment and the 3.5% concentration treatment. 17/3/2021 until 19/5/2021 where it decreased to zero% as a result of the absence of mites in the above two treatments during the previous period, compared with the control and concentration treatments 3.0% where different percentages of relative effectiveness were directly recorded which were related to the number of mites in the sects and where the percentage of which reached 56.60% and 3.0% in both the control treatments and the 3.0% concentration treatment to significantly outperformed the other treatments. The highest mean in the control treatment amounted to 68.49% significantly differed from the rest of the mean of the other treatments, and recorded the lowest mean of 50% in the 3.5% concentration treatment where the above means were recorded at an average temperature of 29.25% C and a relative humidity of 29.00%.

As for the general mean of the treatments, the control treatment significantly differed, according to the Duncan test, by recording the highest mean relative effectiveness of 66.07% compared with the lowest mean of 17.75% which in turn was recorded in the 4.2% concentration treatment and which also significantly differed according to a test on the 3.0% and 3.5% concentration treatments with their mean of 54.68% and 21.02% respectively. These previous results were directly related to the number of fallen mites present within the different transaction denominations.

Table 3. A relative effectiveness of different concentrations of oxalic acid during the early spring and summer / 2021.

	Relative effectiveness																
date treatment	2/17	2/24	3/3	3/10	3/17	3/24	3/31	4/7	4/14	4/21	4/28	5/5	5/12	5/19	5/26	6/2	6/9	total
Control	-	71.3 j	76.04 f	69.74 k	61.67 r	77.0 d	57.67 u	69.74 k	75.27 g	63.49 q	58.93 t	65.15 o	50.00 x	56.60 v	61.67 r	68.49 l	74.44 i	66.44 a
3.0% Concentration	-	86.0 b	76.67 e	0.0 z	30 y	53.7 w	74.91 h	74.91 h	81.74 c	87.04 a	30.00 y	30.00 y	30.00 y	30.00 y	65.00 p	59.88 s	65.00 p	54.68 b
3.5% Concentration	-	59.88 s	59.88 s	50.0 x	0 z	0 z	0 z	0 z	0 z	0 z	0 z	0 z	0 z	0 z	50 x	50 x	66.50 n	21.02 c
4.2% Concentrati	-	67.0 m	50.0 x	50.0 x	0 z	0 z	0 z	0 z	0 z	0 z	0 z	0 z	0 z	0 z	0 z	67 m	50 x	17.75 d
total		71.03 a	65.65 b	42.44 f	22.92 m	32.68 k	33.15 j	36.16 i	39.25 g	37.63 h	22.23 n	23.79 l	20.00 p	21.65 o	44.17 e	61.34 d	63.99 c	

The results of Table (4) show the efficiency of the treatment which is inversely related to the number of mites and moral superiority according to the Duncan's test for the two concentration treatments 3.5% and 4.2%, by recording the treatment efficiency of 100%, and the reading on 3/17/2021;viz. about a month after the application of the treatment with oxalic acid up to the reading on May 12, 2021 for the 4.2% concentration treatment compared with the lowest mean in the control and 3.0% concentration treatments, to record the lowest mean of efficiency when read on 9/6/2021 amounted to 4% in the 4.2% concentration treatment. This significantly differed from the 3.5% concentration treatment with a mean of 3% and that of the control and 3.0% concentration treatments with their means of 9.19% and 14.5% respectively.

As for the general mean, the results of the above table show the efficiency and effectiveness of the different treatments. The 4.2% concentration treatment according to Duncan's test at the probability level of 5% and its mean of 60.47% outperformed all other treatments. In addition, the lowest mean was recorded in the control treatment amounted to 9.47% which significantly differed from the two concentration treatments 3.0%. and 3.5% with their means of 19.38% and 55.22% respectively. The previous results were consistent with what Nanetti (2003) lower oxalic acid concentrations usually yielded worse and/or more variable efficacy. However, 3.2% repeatedly demonstrated to be an acceptable alternative to 4.2% OA, that might be preferred when poor tolerability is recorded with the last concentration. Maggi et al.(2016) reported that final efficacy of the oxalic acid treatment had significant

differences. All trials had a low variability in the final efficacy (range between 85.9 ± 98.8 %). The highest mortality for the three assays was recorded during the first 22 days with an average partial efficacy of 74.4 %.

Table 4. Treatment efficiency of different concentrations of oxalic acid during the early spring and summer / 2021

	Treatment efficiency																	
date treatment	2/17	2/24	3/3	3/10	3/17	3/24	3/31	4/7	4/14	4/21	4/28	5/5	5/12	5/19	5/26	6/2	6/9	total
Control	10.32 l	6.36 y	6.53 x	9.19 o	7.29 t	7.75 r	9.92 m	6.70 w	7.52 s	11.27 j	10.78 k	12.40 i	15.50 e	13.78 h	9.54 n	7.09 v	9.19 o	9.47 d
3.0% Concentration	5.8 z	4.35 b*	12.43 h	87 b	5.8 z	4.14 c*	7.25 u	5.80 z	3.78 e*	5.80 z	43.50 c	43.5 c	43.5 c	17.43 d	12.43 h	12.43 h	14.5 f	19.38 c
3.5% Concentration	3.6 f	3 g*	3.6 f*	9 p	100 a	100 a	100 a	100 a	100 a	100 a	100 a	100 a	100 a	9 p	4.5 a*	3 g*	3 g*	55.22 b
4.2% Concentrati	2.67 h*	2.67 h*	4 d*	8 q	100 a	100 a	100 a	100 a	100 a	100 a	100 a	100 a	100 a	100 a	4 d*	2.67 h*	4 d*	60.47 a
total	5.59 p	4.10 q	6.65 n	28.30 k	53.27 f	52.97 h	54.29 d	53.13 g	52.83 i	54.27 e	63.57 c	63.98 b	64.75 a	35.05 j	7.62 m	6.30 o	7.67 l	

Conclusion

From the previous results, it is clear that the concentration 4.2% came first in terms of efficiency, then focus 3.5%, and finally focus 3.0% in terms of applying transactions and its impact on reducing the number of mites and their efficiency in limiting the Varroa growth and reproduction in reducing their numbers within the colonies to a level below the general equilibrium level so as not to harm the bees compared with the comparison treatment that showed high numbers of Varroa infection on the honeybee colonies and harming them.

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