

## IMPROVING AVOCADO POLLINATION WITH BUMBLEBEES: 3 SEASONS SUMMARY

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### **ABSTRACT**

Experiments in avocado pollination under field conditions using bumblebees were conducted at eight sites (replications) during the 1995-96, 1996-97 and 1997-98 seasons. The experiment was terminated at four of these sites in 1998, and is continuing in the other sites. At the onset of avocado bloom honey-bee colonies were placed in equal density in the treatment and in the control plots of each site, and 25 bumblebee hives (seven per ha) were added to the "bumblebee treatment" plot. At three sites, which were established in 1997, a "honey-bee treatment" plot was also included, in which the honeybee hive density was doubled. The experiment plots of each replication were located one to four km apart. A row of 'Ettinger' was selected in each plot, as well as a row of 'Hass' adjacent to 'Ettinger' ("near Hass"), and a row of 'Hass' at a distance of three to four rows away from the 'Ettinger' ("far Hass"). Bee density, flower density and initial fruit set of each cultivar was monitored throughout the blooming season. Yield data was collected prior to harvest from 20 sample trees per cultivar, and later the post harvest data of the entire plots was recorded. Pollination rates were examined at two sites, and the percent of 'Hass' fruits resulting from cross-pollination was determined at one site. Each replication was run for two consecutive years.

No significant differences in honey-bee activity were observed between the bumblebee treatment and the control, while in the honeybee treatment the honey-bee activity increased. The correlation between honey bee-activity and avocado flowering was low or negative, whereas the same correlation of bumblebee activity was positive. The pollination efficiency of the honey bees was low in the first two seasons, and higher during the 1997 season. Pollination rates were higher in the bumblebee treatment plots than in the honey-bee treatment and in the control plots. In the first two experimental years higher rates of surviving 'Hass' initial fruits were found in the bumblebee treatment, in comparison to the control, however no clear trend was observed in 1997. In the four replications that ended in 1998 the average yield of 'Ettinger' increased by 66% in the bumblebee treatment plots as compared to the control. The average 'Hass'

yield increased by only 14%, which was mainly due to the 34% increase in the "far Hass" yield. A significant increase in 'Hass' yield in the bumblebee treatment (+51%) was found in the organic orchard in Yodfat. At this stage, a limited use of bumblebees for 'Ettinger' pollination is recommended.

## **INTRODUCTION**

Insufficient pollination is an important limiting factor for avocado productivity in Israel. The honey bee is the only commercially available pollinator in Israel, and during bloom honey-bee hives are placed in most orchards. However, the efficiency of avocado pollination by honey bees is insufficient. They prefer citrus and wild flowers over the avocado flowers, which negatively affects the yield of the early-to-bloom cultivars ('Pinkerton', 'Fuerte', 'Ettinger' and 'Hass'). Also their efficiency in effecting cross-pollination at a distance greater than one to two rows away from the pollen donor is low, which reduces the yield of most cultivars, that require cross-pollination to achieve maximum production (see references: 2, 3,4, 6, 14,15, 18, 19).

In the Western Galilee, a wild bumblebee (*Bombus terrestris*) was observed visiting avocado flowers (Ish-Am, not published). This *Bombus* species is a social feral bee that is widespread from northern Europe to the Galilee and the Carmel in Israel. Today it is raised commercially for pollination purpose, primarily for greenhouse tomatoes (16). *Bombus terrestris* was tested under field conditions as a pollinator in Kiwifruit orchards in New Zealand (17) and in almond orchards in Israel (12). Although *Bombus terrestris* is not a natural pollinator of the avocado, it may pollinate avocado flowers better than the honey bee. The bumblebee is ten times more efficient than the honey bee in performing close pollination (within the culti-var) and at least 20 times more efficient effecting cross-pollination (13). The bumblebee behavior during foraging differs from the honey bee (8), which leads to the assumption that under a competitive environment of more attractive flowers, they would visit the avocado flowers at a relatively higher rate than the honey bees (13). Also, bumblebees are active under conditions during which honey bees do not leave the hive, including periods of low temperature, cloudy weather and light rain (1, 5, 7, 17).

The purpose of this study was to examine the possibility of improving pollination and yield of avocado with the help of bumblebees, by using commercially produced *Bombus terrestris* colonies. The research hypotheses were: 1.) The addition of bumblebees will increase the rate of total pollination of the avocado, which, in turn, will increase fruit set and yield during seasons and in plots where a low activity of honey bees is recognized, especially in the early-to-bloom cultivars; and 2.) The addition of bumblebees will also increase the cross-pollination rate, which will, in turn, increase the rate of cross-pollinated fruits, lower the rate of fruit drop and would ultimately increase yield. This effect would be most noticeable in trees of avocado cultivars that prefer cross-pollination, which are more than two rows away from the pollen donor trees.

## **METHODS**

The experiment was conducted at eight sites, which constitute eight replications (Table 1).

**Table 1. The experimental sites**

Site	Seasons	Treatments	Cultivars	Honey bee Hives per ha	Comments
Macker	1995/6	Bumblebee (Macker)	Hass	3.3	Pilot experiment 1995/96
	1996/7	Control (Beit Ha'emek)	Ettinger		
Kabri	1996/7	Bumblebee	Hass	3.3	
	1997/8	Control	Ettinger		
Matzuba	1996/7	Bumblebee	Hass	3.3	Terminated due to tree pruning
		Control	Ettinger		
Yodfat	1996/7	Bumblebee	Hass	5.0	Organic orchard 1996: No follow-up in season 1997: Bee and initial fruit counts only
	1997/8	Control	Ettinger		
Zikim	1996/7	Bumblebee (Zikim)	Hass	10.0	
	1997/8	Control (Yad Mordechai)	Ettinger		
Eilon	1997/8	Bumblebee	Hass	2.5	
	1998/9	Honey bee Control	Ettinger		
Ra'anana District	1997/8	Bumblebee (Tel Yitzhak)	Hass	3.3	
	1998/9	Honey bee (Ga'ash) Control (Shefa'im)	Ettinger		
Upper Galilee	1997/8	Bumblebee (Kefar Giladi)	Pinkerton	3.3	Doesn't include control plot
	1998/9	Honey bee (Ma'ayan Baruch)	Ettinger		

In each site two to three similar plots 01 about 3.3 hectares each (8.65 acres), one to four km apart, were selected. Each plot consisted of solid rows of 'Hass' (or 'Pinkerton') next to 'Ettinger'. In each one a row of 'Ettinger', a row of 'Hass' next to the 'Ettinger' ("near Hass") and a row of 'Hass' at a distance of three to four rows away from the 'Ettinger' ("far Hass") were selected. In each of the selected rows 20 trees were marked for yield evaluation and 5 trees for bee density determination. For the purpose of flower and fruit counts, two branches per tree on two trees per cultivar were marked, with a north-facing and a south-facing branch on each tree. The selected trees were healthy, had no canopy pruning in the previous two years and were not located at the field perimeter. Due to the alternate bearing behavior of the avocado, data was collected for two consecutive years from each replication.

At the onset of bloom, honey-bee hives were placed in equal density in the "control" and in the "bumblebee treatment" plots of each site (Table 1). Additionally, 25 bumblebee hives were placed along the central axis of each of the "bumblebee treatment" plots (about seven hives per ha), and in the "honey-bee treatment" plots the honeybee hive density was doubled. The bumblebee hives were supplied by 'Pollination Services' of Yad Mordechai and by the 'Bio Bee' plant of Sedeh Eliyahu. Honey bees were supplied by local beekeepers at each site.

Throughout the bloom season, and up to the first week of June, two weekly observations were conducted, simultaneously in all the site's plots, in which honey bees and bumblebees per tree were counted, as well as open flowers and initial fruits per branch. The time for the observation was set the previous evening, according to the forecasted temperatures for the observation day at the given site, so that it will take place during the 'Hass' (or 'Pinkerton') daily peak of female bloom and the 'Ettinger' pollen shedding (10,11). At that time, also, the morning peak of bee activity on the avocados is occurring, and the majority of cross-pollination of A type cultivars is performed (11).

During the 1996 season two pollination tests were conducted in the Macker replication, to analyze the rate of cross-pollination and total pollination in "near Hass" and "far Hass" (9, 11). Similar examinations were conducted in 1997 at the Eilon site, three with the 'Hass' and two with the 'Ettinger' cultivars. The results for the 'Hass' were compared in a Three Way Anova (treatment x distance x days), and for the 'Ettinger' by a  $\chi^2$  test. The male parent of 'Hass' fruits was determined at Macker, in the 1996 season, using isozyme analysis. At harvest, in both the bumblebee-treatment and the control plots, 50 fruits were sampled from the "near Hass" and from the "far Hass" rows, and the male parent of the fruits was determined according to Degani *et. al.* (3). Yield analyses were performed in two steps. First, prior to harvest (September-October) counts of fruits on the 20 marked trees of each cultivar and treatment (fruits per tree) were done. This data was then used to compare "near Hass" and "far Hass" yields within the plot, and as a measure for the between-tree variability. Secondly, after-harvest data for the total yield of each cultivar in each plot (tons per ha) and fruit-size distribution was obtained from the packinghouse.

## RESULTS

### Honey bee and bumblebee density (bees per tree)

Previous research has demonstrated that at least five honey bees per medium size avocado tree in full bloom is required to achieve a significant initial fruit set (14). According to the visitation rate of bumblebees to the flowers and the quantity of pollen on their bodies we had postulated that about 0.5 bumblebee per tree is needed to achieve a similar result. The values of maximum honey-bee density at the experimental sites (Table 2) did not reach the threshold value (5 per tree) on most observation days, and was lower than that in all the days at 65% and 13% of the 'Hass' and 'Ettinger' blocks, respectively. Honey-bee density was high mainly in the honey-bee treatment plots (Table 1), and also in Zikim site, where 10 hives per ha were set, and in the Macker replication. Therefore, we assume that honey-bee density was a limiting factor in avocado pollination at most sites and on most days. On the other hand, the maximum bumblebee density surpassed the required value (0.5 per tree) at all the bumblebee treatment plots and in a significant portion of the bloom days (Table 2), and was able therefore to improve pollination since initial fruit set was observed in the control plots, where the threshold value for honey-bee density was not recorded (Table 2). This bee density may have occurred in between the two weekly observation days, or during periods outside of the observation hours.

Honey-bee density on 'Ettinger' in the bumblebee treatment plots was found to be nine to fourteen times greater than that of the bumblebees (highly significant), while on 'Hass' and 'Pinkerton' it was only three to six times greater (not significant). The number of available honey bees in these plots was approximately 100 times higher than the available bumblebees. Therefore, the bumblebees efficiency as an avocado visitor was twenty times higher than that of the honey bees in the less attractive 'A' type cultivars 'Hass' and 'Pinkerton', and only eight times higher in the 'Ettinger'. In other words, the honey bees preferred 'Ettinger' over the 'A' cultivars by a factor of 6.4 to 32 (highly significant), while the bumblebees preferred 'Ettinger' by a factor of only 1.4 to 2.5 (not significant). Thus, the bumblebees were five to ten times less influenced by the attractiveness differences of the avocado cultivars than the honey bees, and visited the

unattractive cultivars almost as much as the attractive 'Ettinger'.

**Table 2: Bee density and its correlation with avocado flower density**

Average bee density (bees per tree) and the Maximum observed density (in parenthesis) are presented in the upper part of the table. In the lower part the correlation coefficient  $\text{\textcircled{r}}$  between bee density and flower density during the season is given. The closer the  $\text{\textcircled{r}}$  value approaches (+1) the greater the efficiency of the bee as an avocado pollinator. An empty cell indicates a treatment that did not exist at the site, and a line indicates that the correlation coefficient could not be calculated for lack of data

Site	Season	Bumblebee treatment				Honey bee treatment		Control	
		Hass/Pinkerton Honey Bees	Bumble Bees	Ettinger Honey Bees	Bumble Bees	Hass/P Honey Bees	Ettinger Honey Bees	Hass/P Honey Bees	Ettinger Honey Bees
Seasonal average of bee density (and the maximum observed), bees per tree									
Macker	1995	0.62 (4)	0.11 (3)	3.7 (16)	0.49 (3)			0.36 (4)	2.6 (8)
	1996	10 (24)	0.55 (2)	15 (40)	1.4 (7)			6.4 (23)	19 (47)
Matzuba	1996	0.17 (3)	0.07 (2)	1.06 (8)	0.17 (2)			0.35 (3)	4.1 (20)
Kabri	1996	0.34 (2)	0.11 (2)	0.80 (5)	0.32 (2)			0.30 (3)	1.4 (10)
	1997	0.15 (2)	0.08 (2)	0.34 (3)	0.15 (2)			0.03 (1)	0.54 (3)
Zikim	1996	0.76 (6)	0.34 (3)	10 (49)	0.68 (4)			0.13 (1)	7.3 (42)
	1997	1.1 (7)	0.14 (1)	8.3 (37)	0.12 (2)			4.9 (26)	12 (62)
Yodfat	1997	0.16 (6)	0.40 (2)	2.4 (10)	0.38 (2)			0.04 (1)	2.8 (12)
Eilon	1997	0.73 (5)	0.20 (1)	0.62 (6)	0.24 (3)	0.30 (2)	3.5 (20)	0.20 (1)	1.0 (14)
Ra'anana	1997	0.49 (5)	0.42 (4)	4.3 (14)	0.81 (6)	0.09 (2)	5.9 (24)	0.31 (9)	6.4 (22)
Upper Galilee	1997	0.20 (2)	0.28 (2)	0.71 (3)	0.40 (3)	0.16 (4)	2.7 (14)		
Seasonal correlation coefficient (r) between bee density and flower density									
Macker	1995	-0.01	+0.28	+0.06	+0.05			-0.10	-0.31
	1996	-	-	+0.19	+0.22			+0.73	+0.37
Matzuba	1996	-	-	+0.03	+0.35			-0.08	+0.34
Kabri	1996	-0.11	+0.69	-0.10	+0.27			-0.13	+0.48
	1997	-0.18	+0.09	+0.58	+0.46			+0.57	+0.19
Zikim	1996	-0.25	+0.60	+0.23	+0.32			-0.20	-0.17
	1997	+0.23	+0.22	+0.33	+0.40			+0.29	+0.51
Yodfat	1997	-	-	-	-			-	-
Eilon	1997	-0.11	+0.28	+0.41	+0.57	+0.29	+0.22	+0.53	-0.13
Ra'anana	1997	-0.18	+0.36	+0.31	+0.68	+0.45	+0.66	+0.04	+0.32
Upper Galilee	1997	+0.31	+0.47	+0.08	+0.16	+0.07	-0.22		

The density of honey bees in the honey-bee treatments, and also in Zikim, relative to the other treatments and replications, was higher on 'Ettinger' but similar on 'Hass' and 'Pinkerton' (Table 2). It appears that increasing hive density increased honey-bee activity mainly on the more attractive cultivar 'Ettinger'. Other than this, honey-bee density was similar between the treatments within a replication. Thus one may not attribute yield differences between treatments to different honey-bee activity.

The correlation between honey-bee density and avocado bloom (Table 2) was mostly low (negative or positive), while the correlation of the bumblebee density was always positive, close to significant in 'Hass' and highly significant in 'Ettinger'. Thus, it appears that honey-bee activity was more influenced by the competitive bloom than that of the bumblebees. A highly positive correlation between honey-bee density and avocado bloom was mainly found in plots of high honey-bee hive density, namely in the honey-bee treatment at Ra'anana and at the Zikim site. This correlation was low in the 1995 and 1996 seasons, and increased in 1997, which may indicate a higher efficiency of

avocado pollination by honey bees in the 1997 season.

### Pollination rates

The percent of pollinated flowers was higher in the bumblebee treatment plots than in the honey-bee treatments and in the control (significant in 'Hass'), but was not statistically different between the honey-bee treatment and the control (Table 3). As expected, pollination rates were higher in the "near Hass" comparing to the "far Hass", and also the ratio of cross-pollination to the total daily pollination was higher in the bumblebee treatment plots than in the other plots (not significant). However, contrary to expectations, this ratio was similar in both "near Hass" and "far Hass".

**Table 3: Average pollination rates**

The percent of pollinated flowers was determined by sampling 50 stigmas per treatment. Samples were collected in 1996 at Macker site and in 1997 at Eilon site. Number of samples: cross-pollination in 'Hass' - 5, total daily pollination in 'Hass' - 3, total daily pollination in 'Ettinger' -2 (insufficient to conduct a statistical analysis)

	Bumblebee treatment			Honey bee treatment			Control		
	near Hass	far Hass	Ettinger	near Hass	far Hass	Ettinger	near Hass	far Hass	Ettinger
Cross Pollination	15.6	10.8	-	3.6	1.4	-	6.2	5.2	-
Total daily Pollination	33.2	18.7	39.0	18.8	8.0	31.0	16.5	13.0	26.9
All Measurements	22.2	13.8	39.0	7.4	3.0	31.0	10.0	8.1	26.9
Significance difference compared to bumblebee treatment	Cross pollination			0.017	0.071	-	0.0044	0.13	-
	All measurements			0.0047	0.073	-	0.0007	0.11	-

### Initial fruit set rate and the percent of surviving fruits in early June

The rate of the initial-fruit set, which is the ratio between number of initial fruits and number of flowers on a branch, was examined at one site in 1995, at three sites in 1996 and at six sites in 1997. The number of initial fruits per branch increased during the bloom period, reached a maximum value and later decreased, following the early fruit drop. Maximum fruit-set values did not differ significantly among treatments, but on June, following the May fruit drop, a difference between the treatments was noticed. This difference became more significant when the initial-fruit set in June had transformed to "percent of surviving fruits", which is the ratio between a brunch fruit-set rate in June and its seasonal maximum fruit-set rate, multiplied by 100 (Tables 4,5).

In the first two seasons (Table 4) the percent of surviving fruits in the bumblebee treatment plots was higher than in the control in the "far Hass" (highly significant) and in the "near Hass" (not significant), though similar in the 'Ettinger'. In the control plots this percent was higher in the "near Hass" over the "far Hass", but was similar in the bumblebee treatment plots (highly significant difference). Nevertheless, in the 3<sup>rd</sup>

season (Table 5) no clear tendency was observed in comparing surviving fruit percent between treatments and 'Hass' distances, except in the Zikim replication, where the surviving fruit percent was higher in the bumblebee treatment over the control.

**Table 4. Percent of surviving initial fruits, 1995 and 1996 season**

"Percent of surviving fruits" is the ratio between a brunch fruit-set rate in June and the brunch seasonal maximum fruit-set rate, multiplied by 100 (see text). The treatments were compared in a homogeneous test of  $X^2$ . The sites' significance values were added using a  $X^1$  significance addition procedure. In case where the result matched the project assumptions (treatment higher than control, "near Hass" higher than "far Hass") its significance was noted with a "+", and a significance of a result which opposed these assumptions was marked with a "-".

Treatment	Cultivar	Site and Year				Average
		Macker 1995	Macker 1996	Matzuba 1996	Zikim 1996	
Bumblebee	near Hass	22	86	100	69	69
	far Hass	64	88	43	68	66
	Ettinger	13	65	17	48	36
Control	near Hass	39	34	80	17	43
	far Hass	10	43	27	15	24
	Ettinger	25	27	43	45	35
Probability of difference (p) between bumblebee treatment and the control	Ettinger	-0.29	+0.079	-0.48	+0.98	1.00
	near Hass	-0.15	+0.071	+0.80	+0.068	+0.51
	far Hass	+1.7E-08	+0.17	+0.68	+0.17	+7.0E-07
	All Hass	+2.0E-06	+0.067	+0.87	+0.064	+9.7E-06
Probability of difference (p) between "near Hass and "far Hass"	Bumblebee	-3.9E-04	-0.98	+0.43	+0.99	-0.081
	Control	+9.3E-04	-0.60	+0.19	+0.92	+0.036
Probability of difference (p) of the ratio nH/fH between treatment and control		+2.5E-09	-0.41	+0.53	+0.77	+3.7E-06

### Rate of out-cross fruits of 'Hass'

The rate of 'Ettinger' progenies (in Macker 1996) was found to be 100% in the bumblebee treatment and 56% in the control for the "near Hass" fruits ( $p=0.0082$ , homogeneity test of  $X^2$ ), and 64% and 20% for the "far Hass", respectively ( $p=0.0069$ ). As was expected, this rate was higher in the "near Hass" than in the "far Hass" ( $p=0.005$ ), and also the rate of decrease with distance from 'Ettinger' was smaller in the bumblebee treatment compared to the control ( $p=0.055$ ). It is possible to relate the increase in the rate of out-cross 'Hass' fruits ('Ettinger' progenies) in the bumblebee treatment to the presence of the high mobility bumblebees.

**Table 5: Percent of surviving initial fruits, 1997 season**

See the explanation in Table 4. A line indicates a treatment that did not exist at the site. A "0% surviving initial fruits" occurred when a too small branch was selected for the observations. An empty cell indicates a treatment that did not exist at the site.

Treatment	Cultivar	Site (and 'A' type cultivar)						Average
		Eilon (Hass)	Kabri (Hass)	Yodfat (Hass)	Upper Galilee (P)	Ra'anana (Hass)	Zikim (Hass)	
Bumblebee	near Hass (or Pinkerton)	8.3	23	5.3	54	63	46	33.3
	far Hass (or Pinkerton)	13	3.7	0.0	0.0	96	45	26.3
	Ettinger	6.7	3.8	24	79	0.0	8.8	20.4
Honey Bee	near Hass (or Pinkerton)	11			7.4	62		26.8
	far Hass (or Pinkerton)	0.0			14	0.0		4.7
	Ettinger	8.2			91	14		37.7
Control	near Hass (or Pinkerton)	31	23	4.3		32	4.8	19.0
	far Hass (or Pinkerton)	33	5.2	20		9.5	8.7	15.3
	Ettinger	8.3	7.1	19		4.1	4.1	8.5

## Yield

The yield data from the 4 replications which were terminated in 1997-98 is presented in Table 6, and the corresponding fruit counts of the 20 sample trees of "near Hass" and "far Hass" are given in Table 7. Upon investigation it was found that the control plot at Kabri is located on an archeological hill and is one of the best plots at the site. The bumblebee treatment plot, on the other hand, is located in a low place on heavy soil, with weak 'Hass' trees that bear a low annual yield. We believe, therefore, that one should look at the 'Hass' yield result without the Kabri data.

### 'Ettinger' yield (Table 6)

The 'Ettinger' yield in the bumblebee treatment plots was consistently higher than that in the control, during both seasons and at all sites, except at Yodfat in season #1. The average yield increase in the bumblebee treatments was 7.8 ton/ha/yr (6,960 Lbs/acre/yr, +66%,  $p=0.042$ ). It is worth noting the high 'Ettinger' yield in the bumblebee treatment at the Macker site, that averaged 28.5 ton/ha/yr (25,430 Lbs/acre/yr), which was higher than the yield in the control by 17.4 ton/ha/yr (15,524 Lbs/acre/yr more, 157% increase). On the other hand, at Yodfat site the 'Ettinger' yield in the bumblebee treatment averaged 0.65 ton/ha/yr less than in the control, which constituted a small decrease in yield of 6%.

**'Hass' yield (Table 6):** The results with the 'Hass' yield differed between the sites. The yield in the bumblebee treatment plot was constantly higher than that in the control at Yodfat and Macker sites, the treatment and the control plots averaged the same at the Zikim site, and the treatment was consistently lower than the control at Kabri. The average for the three sites (ignoring the Kabri site) reveals a yield increase by 1.81 ton/ha/yr (1,615 lbs/acre/yr, +14%, not significant).



**Table 6: 'Hass' and 'Ettinger' total yield (kg/ha)**

Total yield of the experimental plots in 4 replications, 2 consecutive years per replication, based on post harvest packing house report. Within and between sites' yields were compared with a t-test. Average yields were tested with and without the Kabri data, since this replication did not meet the experimental conditions (see text).

Site	Treatment	Ettinger			Hass		
		Season 1	Season 2	Average	Season 1	Season 2	Average
Macker	Bumblebee	25,510	31,400	28,455	18,420	5,770	12,095
	Control	7,510	14,660	11,085	11,120	10,280	10,700
Zikim	Bumblebee	20,160	22,500	21,330	15,150	15,660	15,405
	Control	10,000	17,350	13,675	9,840	24,050	16,945
Yodfat	Bumblebee	12,400	15,700	14,050	25,000	8,230	16,615
	Control	15,500	13,900	14,700	20,000	2,060	11,030
Average without Kabri	Bumblebee	19,357	23,200	21,278	19,523	9,887	14,705
	Control	11,003	15,303	13,153	13,653	12,130	12,892
Treatment to control ratio		1.76	1.52	1.62	1.43	0.82	1.14
Probability of difference (p)		0.068	0.083	0.066	0.0074	0.33	0.24
Kabri	Bumblebee	13,810	15,000	14,405	9,410	9,530	9,470
	Control	8,020	7,180	7,600	24,000	13,000	18,500
Average with Kabri	Bumblebee	17,970	21,150	19,560	16,995	9,798	13,396
	Control	10,258	13,273	11,765	16,240	12,348	14,294
Treatment to control ratio		1.75	1.59	1.66	1.05	0.79	0.94
Probability of difference (p)		0.036	0.061	0.042	0.44	0.31	0.37

**Yield ratio between "near Hass" and "far Hass" (Table 7):** In both seasons the decline of 'Hass' yield with the growing distance from 'Ettinger' was smaller in the bumblebee treatment plots than in the control. This difference was large and statistically significant at the Macker, Zikim and Yodfat sites, whereas small and not significant at the Kabri site. Eliminating the Kabri data the ratio of "far Hass" to "near Hass" yield was higher in the bumblebee treatment than in the control plots by 24% ( $p=0.019$ ,  $n=3$ ), and for the four sites it was higher by 21% ( $p=0.013$ ,  $n=4$ ). The bumblebee treatment average yield of the three sites was higher than that of the control by 34.4% in the "far Hass" ( $p=0.005$ ), and by only 8.2% in the "near Hass" (not significant). The "Hass" yield increase at the bumblebee treatments was mostly gained in the first test year ("far Hass" to "near Hass" ratio higher by 51%,  $p=0.072$ ), whereas only a small increase was observed in the second year (+3.8%, not significant).

**Table 7: "Near Hass" and "far Hass" yield (fruits/ha) and yields' ratio**

Yield of 20 sample trees per cultivar in 4 replications, 2 consecutive years per replication, based on pre harvest counting. Average yields were tested with and without the Kabri data, since this replication did not meet the experimental conditions (see text). The between treatment comparison of the yield ratio "far Hass"/"near Hass" was performed employing t-test on arcsine transformation of the ratios.

Site	Season	Bumblebee treatment			Control			Treatment/ Control ratio of fH/nH
		near Hass	far Hass	fH/nH	near Hass	far Hass	fH/nH	
Macker	1995	82,730	73,330	0.886	58,880	27,680	0.470	1.885
	1996	32,650	28,150	0.862	46,930	43,410	0.925	0.932
	2 Year Average	57,690	50,740	0.880	52,915	35,545	0.672	1.309
Zikim	1996	33,980	32,460	0.955	27,230	8,310	0.305	3.130
	1997	65,810	63,560	0.966	85,050	70,500	0.829	1.165
	2 Year Average	49,895	48,010	0.962	56,140	39,405	0.702	1.371
Yodfat	1996	100,810	79,000	0.748	95,740	66,170	0.691	1.134
	1997	39,100	33,670	0.861	14,500	14,760	1.018	0.846
	2 Year Average	69,955	56,335	0.805	55,120	40,465	0.734	1.097
Average without Kabri	Year 1	72,507	61,597	0.850	60,617	34,953	0.562	1.512
	Year 2	45,853	41,793	0.911	48,827	42,890	0.878	1.038
	2 Year Average	59,180	51,695	0.874	54,722	38,472	0.703	1.242
Kabri	1996	8,350	11,060	1.325	69,180	57,010	0.824	1.607
	1997	32,360	21,960	0.679	29,820	19,620	0.658	1.031
	2 Year Average	20,355	16,510	0.811	49,500	38,315	0.774	1.048
Average with Kabri	Year 1	56,468	48,963	0.867	62,758	39,793	0.634	1.368
	Year 2	42,480	36,835	0.867	44,075	37,073	0.841	1.031
	2 Year Average	49,474	42,899	0.867	53,416	38,433	0.719	1.205

## DISCUSSION

Each site of the experiment represents a replication, or block, which was investigated over two consecutive years. A high number of replications are needed, due to significant differences between seasons and replications, and between plots within the replication. The four completed replications give answer to the research questions, though often without sufficient certainty.

A consistent advantage of the bumblebees over the honey bees as avocado pollinators has revealed, from both aspects of the relative "on-tree" density and of the correlation between bee and flowering density (Table 2). The maximum honey-bee density values did not reach the threshold value of five bees per tree on most observation days, while the maximum bumblebee density surpassed the required value of 0.5 per tree in all the bumblebee treatment plots (Table 2). Since no significant difference between treatment and control regarding honey-bee activity was noted, it is likely that the above advantage is from the ground for the higher pollination rates in the bumblebee treatment plots, as compared to the other plots. The increase in both fruit surviving percent and 'Hass' out-cross fruit rate may also be attributed to this advantage, and, in turn, constitute the reason for the increase in productivity.

While comparing the bumblebee treatment plots to the control we found an increase of total pollination of the two cultivars in the former, as well as of cross-pollination of 'Hass' (Table 3). However, since an increase of the fruit survival rate was observed only in the 'Hass' (Tables 4, 5), and since the increase of 'Hass' yield in the bumblebee treatment plots was mainly noticed in the 'Tar Hass' (Table 7), it appears that the bumblebees contributed to the 'Hass' yield mainly by increasing cross-pollination rate and, in turn, decreasing fruit drop. On the other hand, they helped the 'Ettinger' yield mainly by increasing the overall pollination and, in turn, increasing fruit set rate.

The increase of the 'Ettinger' yield in the bumblebee treatment plots (+66%) appears to be significant and promising (Table 6). However, more studies are needed to measure its average effect and variability. Therefore, a small-scale commercial introduction of bumblebees (in addition to honey bees) in blocks where 'Ettinger' is planted can be recommended. The average increase in 'Hass' yield in the bumblebee treatment plots was small and not significant (+14%, Table 6). It was mainly manifested in the "far Hass" (+34%, Table 7), which constituted about one third of the 'Hass' in the experimental plots. It is not yet possible to recommend the commercial introduction of bumblebees with this cultivar, though its productivity may benefit when it is planted with 'Ettinger' and bumblebees are introduced to the plot. Nevertheless, the large increase in 'Hass' yield in the bumblebee treatment at Yodfat biological orchard (+5.58 ton/ha/yr, +51%, Table 6) indicates that the contribution of the bumblebees to the 'Hass' yield may be more significant in an organic orchard, where plant growth regulators are not used. We will continue to examine the possibility of replacing growth regulators' spray in 'Hass' with bumblebee pollination.

The yield increase in the bumblebee treatment plots against the control was achieved mainly in the first year and less so in the second one (Tables 6 and 7). It is possible to relate it to the higher honeybee pollination efficiency in 1997 as compared to the previous two years, which provided the bumblebees a significant advantage over the honey bees in 1995 and 1996, and less so in 1997.

Alternate bearing appeared to be minimized in the bumblebee treatment plots. For example, at Yodfat in the season 1995/96, prior to the experiment commencement, the bumblebee treatment plot's 'Hass' carried 19 ton/ha (16,952 Lbs/acre), a heavy crop for an organic orchard, while the control plot was in an off-year and bore only 5 ton/ha (4,461 Lbs/acre). Nevertheless, in the 1996/97 season (Table 6) the treatment plot produced a bumper crop of 25 ton/ha (22,305 Lbs/acre) and the control only 20 ton/ha (17,844 Lbs/acre). In the next season (1997/98) the control yield was down to 2.06 ton/ha (1,838 Lbs/acre) while the treatment plot decreased to only 8.23 ton/ ha (7,343 Lbs/acre). This trend was also observed in 'Hass' at Zikim and Kabri sites and in 'Ettinger' at Zikim and Macker. It could be a result of a higher pollination efficiency of bumblebees comparing to honey bees in off-season trees.

At the Zikim site, with high honey-bee density of 10 bee hives per ha, we found an increase in both bee activity and the correlation between it and the avocado flowering (Table 2). A similar increase was observed in the honey bee treatment plots. Indeed, at the Zikim site higher yields were recorded, comparing to the other experimental sites (except the 'Ettinger' yield in the bumblebee treatment plot at Macker). It appears, therefore, that a significant increase in yield may be achieved by adding honey-bee hives to these orchards. Nevertheless, even at the Zikim site we found a significant yield increase in 'Ettinger' in the bumblebee treatment, similarly to the other sites (Table 6). It is important to note that till now we have only examined a use of bumblebees in a density of seven hives per ha, in addition to honey bees in a range of densities. In continuation, we shall examine different combinations and densities of honey bees and bumblebees, including the use of bumblebees alone.

## ACKNOWLEDGMENTS

We wish to thank the orchard owners, the work managers and the technicians, who allowed and helped us throughout the experiment implementation. Dr. Hillary Foot helped with the statistical analyses and Reuben Hofshi with Mary Lu Arpaia helped with the English translation. The research was funded by the Israeli Fruit Council and the Western Galilee R&D, and was implemented with the support of the bumblebee producers 'Pollination Services' of Yad Mordechai and 'Bio Bee' of Sedeh Eliyahu.

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