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Parasitic *Varroa destructor* mites influence flight duration and homing ability of infested *Apis mellifera* foragers*

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Abstract – This study confirmed that infestation by *Varroa destructor* is lower in foragers returning to the colony than in those leaving the colony and explored causes of mite loss. Video recordings of bees at the flight entrance revealed that some mites may get lost from foragers but also showed that infested bees stay outside the colony longer. Returning tests of foragers released at some distance of the hive confirmed that infested bees take longer time to return or do not return at all. The loss of foragers per flight was higher in a highly infested colony compared to a less infested colony. In a visual orientation test at the hive entrance infested bees scored lower, indicating impaired orientation abilities. The results show that infestation by *V. destructor* mite influences the flight behaviour of forager bees, to the effect that foragers might not return to the colony. This is interpreted as an adaptive behaviour of the bees to remove the parasites or pathogens from the colony.

Varroa destructor / forager / *Apis mellifera* / flight behavior / host-parasite relationship / homing

1. INTRODUCTION

Varroa destructor is a relatively recent parasite of the Western honey bee *Apis mellifera* in terms of co-evolution with its host. In contrast to the original host *A. cerana*, *A. mellifera* is not well adapted to the mite and infested colonies suffer severe damage (Oldroyd, 1999). Nevertheless, several mechanisms to decrease virulence of *V. destructor* have been well documented in *A. mellifera* (Harbo, 1992; Büchler, 1994; Boecking and Ritter, 1994; Fries et al., 1994; Calis et al., 1999). All of these mechanisms affect either the reproduction or cause death of the mites within the colonies. Some efficiently decrease the infestation within colonies and are used in breeding programs (Spivak, 1996; Spivak and Reuter, 1998, 2001; Boecking and Spivak, 1999; Harbo and Harris, 2003). During their

phoretic phase the mites are attached to the bees and may leave the colony with foragers. Defence mechanisms of foraging bees might also operate during this phase of the parasite's life cycle.

The population dynamic of the mite is affected during foraging flights in several ways. Some mite mortality is bound to occur due to the natural death of ageing foragers, according to the “conveyor belt” effect by which pathogens are removed from colonies during population turnover in social insects (Schmidt-Hempel, 1998). However, this effect is not likely to be strong but rather weak in honeybees (Fries et al., 1994; Fuchs and Kutschker, 2000). Further, bees may not return to the colony due to drifting into other colonies (Free, 1958) which is the main path enabling the parasite to spread between colonies (Hüttinger et al., 1981; Sakofski et al., 1990).

A primary finding which supported that other foraging-related processes might

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diminish a colony's mite load was a marked difference in the infestation of returning foragers compared to departing foragers demonstrated by Kutschker (1999). This showed that foragers lose considerable numbers of mites by unloading them either during foraging or while temporarily visiting other colonies. As a further possibility, Fuchs and Kutschker (2000) hypothesized that such a situation could be caused by enhanced non-returning of infested foragers. That parasitized workers might tend to leave the colonies is also supported by the observation that colony breakdown caused by *V. destructor* is characterised by a rapid loss of foragers until the colony remains with the queen and few workers (Martin, 1997). A change in flight behaviour is also indicated by enhanced drifting of bees from infested to uninfested colonies (Sakofski, 1990). Similarly, the effect of frequent non-returning to the colony was documented from honey bees parasitized by tracheal mites (*Acarapis woodi*, Harrison et al., 2001).

That parasitism may cause behavioural changes in insects is well documented. These changes may be in the interest of parasites to promote their dispersal to a new host, as in *Formica* ants which attach themselves to the tip of grass due to parasitism with the liver fluke *Dicrocoelium dendriticum* (Hohorst and Graefe, 1961; Peacock, 2004). It may, however, also be in the interest of the host. In bumble bees, parasitized workers may stay in the field overnight instead of returning to the nest, or they may spend more time in cold areas to retard parasitoid development and diminish its survival chance (Müller and Schmidt-Hempel, 1993). In its extreme the behavioural response might even result in the death of the host to increase its inclusive fitness (Smith-Trail, 1980).

Whether *V. destructor* affects the behaviour of foragers or whether such behavioural changes could be to the benefit of the bees by decreasing colony infestation is unknown. The focus of the study was to investigate whether *V. destructor* influences flight behaviour and returning frequency of foragers to the colony. We used several approaches. We first repeated the experiments of Fuchs and Kutschker (2000) with some methodological

changes to confirm that infestation is lower in returning foragers compared to departing foragers. Second, we investigated passages of infested and uninfested bees at the colony entrance using a video technique. Third, we investigated flight duration and returning time after release of infested and uninfested bees. Fourth, we investigated orientation of bees toward the nest entrance. Fifth, we investigated loss of foragers in relation to forager infestation.

2. METHODS

2.1. Infestation of foragers that leave and return to the colony

Departing and returning workers were sampled over 10 days in 5 commercial colonies from August 13 to September 13, 2001 to compare their infestation. To exclude sampling younger bees that guard the entrance, the hive entrance was extended into a wire net cage (40 cm × 30 cm × 10 cm) which led the bees into a removable bee trap. This trap consisted of a wooden frame (11 cm × 11 cm × 4 cm) blocked by a gauze which funnelled either incoming or departing bees into an attached plastic jar. In total, 54 pairs of samples of departing and returning bees were collected once per day between 11:00–15:00 h. Samples, each containing approximately 100 bees, were frozen and mites were washed from the bees with detergent water (Fuchs, 1985). Sample infestation was calculated as the number of mites per bee.

2.2. Video recordings of departing and returning workers at the flight entrance

Video recordings were conducted in June 2001 and 2002. The entrance of a nucleus colony containing approximately 2000 bees was extended into a tunnel made from Perspex 110 mm in length with internal dimensions of 20 × 6 mm. In the middle region the tunnel was narrowed over a length of 20 mm to 7 × 6 mm internal dimensions to allow only one bee to pass through at time. One video camera (VW-KS 152) and two light diodes were placed above and a second video camera with two light diodes was placed below the narrow part of the tunnel to record the bee from a ventral and dorsal

side. Both channels were combined using a video splitter (Tokina TW 104).

Hatched workers from *V. destructor* free colonies (healthy) were individually marked with numbered tags and introduced into the infested nucleus colony to ensure that bees were infested later in life and were not harmed by the mite in larval and pupal stages. The colour of the tags indicated the day of emergence and the number the individual bee. About 600 marked *V. destructor* free workers were introduced each year over a period of one month prior to recording. In the same period 400–500 mites were added into each colony by introducing infested newly-emerged workers or by direct placement of mites on the bee bodies. Exits and entries of bees were continuously recorded on tapes and analysed over two days from 8:30 h to 18:30 h, one in 2001 (29 July) and one in 2002 (19 July). Recordings over complete days were preferred over short-time observations collected over several days so that bee losses until evening could be determined. In total 914 passing tagged workers were investigated frame by frame for tag numbers and mite infestation and time and direction of passing was noted. The average age of the recorded bees in both years was 21 days. The accuracy of the method was tested with 113 infested and 87 uninfested bees, each passing in each direction. Results of the video recordings were later matched with infestation status. Only in 7 of the passages infestation status was misread, showing a tolerable error margin.

Passages of bees were listed as departing and returning, together with their infestation status. Time outside the nest was calculated as the time difference between departing and returning. To offset considerable variation from bee age, daytime and outside conditions, the time outside the colony was compared within pairs of bees of the same age departing closest in time.

2.3. Individual release of workers

The ability of infested and not-infested workers to find their home was investigated in 2 colonies in the year 2002 from 19 July to 1 September and in the year 2003 from 1 July to 15 August. The entrance of a nucleus colony was extended into a prolonged landing board of 10 cm length to make it easier to recognize returning workers. The hive entrance was narrowed by transparent plastic foil to an opening 1.5 cm long and 1 cm wide, which could be closed by a transparent plastic slider when a returning bee was expected. Mites had been added

half a month prior to the experiment and during the experimental period. In total each colony received about 400–500 mites. Approximately 700 workers from colonies with no or very low *V. destructor* infestation that emerged in the incubator were used for the experiment to ensure these bees had not been parasitized during their larval or pupal stages. They were individually marked to be recognized during the experiment and were introduced into the highly infested colony. For the experiments, we collected workers from the colony and placed them individually in 5 mL vials with perforated plastic lids for respiration, together with a small lump of candy sugar. The average age of collected bees was 22 ± 6 days to ensure that they had some flight experiences. For each infested bee we collected 1–2 uninfested bees of the same age. Bees in the batch were released one by one. The maximum time that workers spent in vials was 20 min. Bees were released from randomly chosen locations at different distances of 5 m, 10 m, and 50 m in the year 2002 and 20 m, 50 m and 400 m in the year 2003. Time of departure and arrival was noted for each bee. Different distances were chosen to find whether effects would depend on them.

2.3.1. Recording returning time

We recorded the time that workers took to return to the colony from randomly chosen locations at different distances. During experiments, 160 infested and 123 uninfested individually marked workers of the same age were released. To identify the workers, the entrance was temporarily closed to cause sufficient delay to record the bee's number, but not to hinder foraging. The returning time was measured for bees returning within a maximum time interval of 15 min in 2002 and of 30 min in 2003. To offset the influence of bee age, and daytime and outside conditions, we compared returning time of infested and uninfested workers of the same age released one after another at short time intervals. In total 127 pairs were compared, mostly composed of one infested and one uninfested worker. In some cases, 2–3 infested and 2–5 uninfested workers were released at the same time. In such cases medians for these groups were paired.

2.3.2. Recording returning rate until evening

Whether infested or uninfested workers return until evening was examined from 1 July until 15

August 2003. We included the 130 workers which had not returned within the observation period during the tests to measure returning time, and 35 additional bees released at a distance of 400 m. Colonies were checked in the evening between 19:00 and 20:00 h. Colonies were checked twice, each check lasted 15 min. The accuracy to recover marked individual workers in the evening was tested by recording 101 coloured marked workers which were additionally marked one hour before examination of the colony. From these, 6 workers were not found, indicating 6% rate of non-detection.

2.4. Orientation of bees toward the nest entrance

The accuracy of orientation of infested and uninfested workers toward the nest entrance was tested in a nucleus colony from 3 to 12 September, 2002 and from 8 to 16 August, 2003. The colony was infested by adding infested emerged workers as described above. The hive was connected to its entrance by a tunnel opening through a white wall in the middle of a circle of 16.5 cm in diameter. To the left and the right side of the nest entrance 2 further circles of the same size were drawn at a distance of 4 cm. The nest entrance was marked by a 10 × 20 cm blue-coloured square. During the experiments, an additional identical dummy blue square was presented alternately in one of the circles to the left or the right side of the entrance. Accuracy of nest entrance orientation was observed from a fixed position of 1.3 m in the front of the middle circle. In total, 241 bees (115 infested, 126 uninfested) were released from vials as described above. We scored whether bees returned to the entrance directly or before finding the entrance, or crossed either the circle containing the dummy or the empty circle. Marked infested and uninfested bees of the same age were released individually from a short distance of 3 m. The release of an infested worker was followed by a release of an uninfested worker of the same age or vice versa.

2.5. Forager infestation and daily loss of foragers from colonies

Infestation of forager bees and bee losses were investigated in a high and a low infested commercial colony, as determined by mite counts in colony debris beforehand. Thirteen samples of bees leaving the colony (six in high and 7 in low infested

colony) were taken in 2003 from 11 July to 3 August to determine infestation. Losses of forager bees were determined by an attached bee counter (Beescan, Lowland Electronic bvba), which counts departing and returning workers through 32 direction sensitive channels. Data from the bee counter were taken every 15 min. Daily bee loss was calculated from the difference between the numbers of departing and returning bees per day. Data were considered for the day either before or after taking the samples, when colonies were not disturbed.

3. RESULTS

3.1. Infestation of foragers that leave and return to the colony

In total 54 pairs of samples from departing and returning foragers were taken. These contained an average of 103 ± 41.6 and 99 ± 56.7 workers, respectively. Mite counts in the samples varied between 0 and 10 mites. The infestation levels were low, between 1–2%. The mean infestation of departing workers was 0.019 ± 0.018 mites per bee and was about twice as high as the mean infestation of returning workers (0.009 ± 0.018 mites per bee). The difference was highly significant ($z = -3.321$, $P < 0.001$, Wilcoxon matched pairs rank test).

3.2. Video recordings of departing and returning workers at the flight entrance

Examination of the videos taken over two days, one in 2001 and one in 2002, showed a total of 914 passages of individually marked bees. From these 750 returned, of which 117 were infested and 633 uninfested.

3.2.1. Mite loss from and with bees

Numbers of departing, returning and non-returning workers, infestation and loss of mites from bees registered by video technique are presented in Table I. The infestation of workers was higher in departing than in returning workers. In total from 914 departing workers,

Table I. The number of infested and uninfested workers departing or returning to the colony as recorded by video at the colony entrance. The percentage of returning and not returning workers was calculated in relation to the number of departing workers.

Year	Leaving (N)	Infested			Leaving (N)	Uninfested		
		Returning		Not returning		Returning		Not returning
		With Varroa	Without Varroa			With Varroa	Without Varroa	
2001	109	63	13	33	469	2	367	100
N, %		57.8	11.9	30.3		0.4	78.3	21.3
2002	70	41	23	6	266	11	230	25
N, %		58.6	32.8	8.6		4.1	86.4	9.4
Total	179	104	36	39	735	13	597	125
N, %		58.1	20.1	21.8		1.8	81.2	17.0

179 and from 750 returning workers 117 were infested. The infestation of workers departing the colony was significantly higher (19.6%) compared to the infestation of returning workers (15.6%, $\text{Chi}^2 = 3.95$, $\text{df} = 1$, $P < 0.05$). Results did not differ between the two years. The infestation of departing workers in 2001 and 2002 amounted to 18.9% (109 from 578) and 20.8% (70 from 336), respectively, and the infestation of returning workers amounted to 14.6% (65 from 445) and 17.0% (52 from 305), respectively.

Some infested workers returned without the mite. Thirteen of 109 mites (11.9%) did not return on the bees in 2001, and 23 of 70 mites (32.9%) did not return in 2002. A significantly higher proportion of mites were lost in the year 2002 compared to 2001 ($\text{Chi}^2 = 11.62$, $\text{df} = 1$, $P < 0.001$). Some bees which were uninfested when they departed returned with a mite (0.4% in 2001 and 4.1% in 2002), but mites were lost significantly more often than were gained in both years ($\text{Chi}^2 = 95.46$, $\text{df} = 1$, $P < 0.0005$).

Workers which were infested when they left the colony did not return more often compared to uninfested workers in the year 2001 showing pronounced loss among infested bees ($\text{Chi}^2 = 4.00$, $\text{df} = 1$, $P < 0.05$). Bee loss was significantly higher in 2001 compared to 2002. It was 30.3% and 8.6% in the infested group, respectively ($\text{Chi}^2 = 11.78$, $\text{df} = 1$, $P < 0.001$) and 21.3% and 9.4% in the uninfested group, respectively ($\text{Chi}^2 = 17.09$, $\text{df} = 1$, $P < 0.0005$).

3.2.2. Flight duration of workers

The duration of time bees spent outside the colony was calculated as the difference between departing and returning time. Data included 127 pairs of infested and uninfested workers which were of the same age and were departing next in time. The time difference between departing infested and uninfested same-age workers ranged from few second to 7 min in maximum. The median time outside the colony was 1.7 times higher in the infested workers compared to the uninfested workers (214 s and 128 s respectively), the difference was significant (Fig. 1, $z = -4.617$, $P < 0.0005$, Wilcoxon matched pairs rank test).

3.3. Individual release of workers

3.3.1. Returning time

A total of 283 workers were released from different distances in pairs of groups containing 1–3 infested or 1–5 uninfested bees of the same age group, thus omitting the influence of age, daytime and distance. The maximum observation time was 30 min. Group medians for the resulting 127 pairs were compared. The median returning time of the infested workers (122 s) was 2.3 times longer than that of uninfested workers (52 s, $z = -4.694$, $P < 0.005$, Wilcoxon matched pairs rank test). Bees released from longer distance needed more time to return, and there was no apparent difference

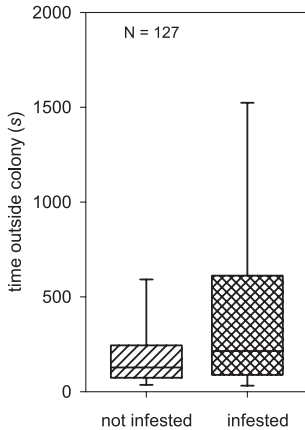


Figure 1. Times spent outside the colony for 127 pairs of infested and uninfested workers registered by video recordings. The pairs include bees of the same age that left close in time. The chart indicates medians, inter quartile ranges, 10% and 90% percentiles.

in relation to infestation (uninfested: $r = 0.574$, $P < 0.0005$, $n = 126$; infested: $r = 0.412$, $P < 0.0005$, $n = 127$, Spearman rank correlation).

3.3.2. Returning rate

The returning rate until evening was determined from 318 released workers, which were comprised of the 283 bees released for measuring returning time and an additional 35 bees released at 400 m distance without measuring returning time. From 137 infested workers 58 (42.3%) did not return to the colony until evening, compared to 52 from 181 uninfested workers (28.7%), which is a significant 1.5 fold increase ($\text{Chi}^2 = 6.38$, $\text{df} = 1$, $P < 0.012$). The returning rate was also higher within the observation period of 30 min. Most of the bees (153 from 283, 54.1%) returned within 30 min and the proportion of returning bees was higher in the uninfested group ($\text{Chi}^2 = 4.18$, $\text{df} = 1$, $P < 0.05$).

3.4. Orientation of bees toward the nest entrance

In the orientation experiment, 115 infested and 126 uninfested bees were released at 1.5 m

distance from the nest entrance, of which 225 returned to the colony. More than two thirds of the infested workers (70.9%, 73 from 103, Tab. II) crossed once or several times the circle, which contained the dummy entrance marking before entering the colony through the true entrance. In contrast, this was the case in only about one third of the uninfested workers (35.2%, 43 from 122, Tab. II, $\text{Chi}^2 = 28.38$, $\text{df} = 1$, $P < 0.0005$). The dummy entrance was significantly more often crossed by the infested than by the uninfested workers ($z = -5.918$, $P < 0.0005$, Mann Whitney U test). Only few workers crossed the empty circle.

3.5. Forager infestation and daily loss of foragers from colonies

The two full-size colonies differed in infestation with an average of 60.4 ± 22.56 and 5.2 ± 9.71 dead mites per day registered on the bottom inserts. Samples of departing bees containing an average of 130.1 ± 49.7 bees were collected from each colony. Infestation of departing bees in the high infested colony (0.015 mites per bee) was 7.7. higher than the infestation in the low infested colony (0.002 mites per bee, $z = -3.169$, $P < 0.002$, Mann Whitney U test). Average flight activity the day before or after sampling was 24184 ± 5607 and 56400 ± 21894 in the high and low infested colony, respectively. Loss of foragers per day was 512 ± 83 in the high and 576.6 ± 149 in the low infested colony, amounting to a relative loss of 2.20 ± 0.832 and 1.00 ± 0.289 per flight per day respectively. Thus bee loss was 2.2 times higher in the high infested colony compared to the low infested colony ($z = -2.877$, $P < 0.004$, Mann Whitney U test). In particular, a significant correlation was found between the proportion of bee loss and the infestation of departing workers ($r = 0.736$, $P < 0.006$, $n = 13$, Spearman rank correlation).

4. DISCUSSION

The basic observation of a substantial decrease in the infestation of the honeybee

Table II. Orientation to the colony entrance of infested and uninfested workers that returned directly (nest entrance) or crossed the circle containing the dummy or the empty circle before entering the nest entrance for both years 2002 and 2003. Percentages are calculated in relation to the released bees.

Year	Workers	Returned bees (N)	Nest entrance direct: (N, %)	Dummy (N, %)	Empty circle (N, %)
2002	Uninfested	58	31 53.4	26 44.8	1 1.7
	Infested	54	9 16.7	40 74	5 9.2
2003	Uninfested	64	45 70.3	17 26.6	2 3.1
	Infested	49	13 26.5	33 67.3	3 6.1
Total	Uninfested	122	76 62.3	43 35.2	3 2.5
	Infested	103	22 21.3	73 70.9	8 7.8

workers that return from foraging flights (Fuchs and Kutschker, 2000) was fully confirmed by the present study. Measurements of infestation showed almost double the infestation in departing workers compared to returning workers. By collecting bees with minimal disturbance a distance from the hive entrance we excluded sampling younger and possibly more highly infested guard bees together with the departing foragers. A lower infestation of returning bees was also confirmed by analyzing video recordings over two complete days, in which the difference was less marked but still significant.

Both results indicated an enhanced loss of mites, but the observations using a video technique gave more insight into possible causes of mite loss. Approximately one fifth of the mites were missing from returning workers that were infested at departure, which would account for about half of the registered mite loss. The missing mites could have been removed actively by workers or could have accidentally fallen from the bees during foraging. However, we suspect that the mites also could have changed host within the entrance tunnel, which was particularly crowded at the narrowed part of the tunnel where bees pushed their way in or out. Such a change of host is supported by marked bees that gained mites at

their return, although they were not likely to pick them up from other nearby colonies. Losing mites from foragers thus might be less frequent in a more natural situation.

A loss of mites from foragers would also be consistent with the possibility that mites change host during temporary visits of other colonies. Even more interesting, it would be consistent with a change of host on flowers. Whether *V. destructor* can use such a pathway for spread similarly to the bumble bee parasite *Crithidia bombi* (Schmid-Hempel, 1998) has never been demonstrated conclusively, nor has it been completely ruled out. The ability to survive on flowers and infest new bees over several days (Hartwig and Jedruszuk, 1987; De Guzman et al., 1993) and the occasional detection on flowers shipped to USA (Kevan et al., 1990; Pettis et al., 2003) may support such a pathway of spread, while the absence of mites on flowers in the vicinity of infested colonies (Pettis et al., 2003) suggests this to be uncommon.

As an additional main source of mite loss, the video recordings showed that mites disappeared from the colony together with bees. Though natural mortality of foragers can be expected to contribute to mite mortality (Fries et al., 1994; Fuchs and Kutschker, 2000), the proportions of infested workers that did not

return to the colony in the study exceeded by an order of magnitude the approximate average mortality of 2.3% per trip, as estimated by Visscher and Dukas (1997).

This high loss of infested foragers suggested that the mite infestation itself could have influenced the returning rate of the bees. At least in one of the two years (2001) the proportion of non-returning bees increased by about one third in the infested over the uninfested bees. In addition, examination of the times spent outside the colony showed that these were 1.7 times longer in the infested bees compared to uninfested and the difference was significant in both years. Similar results were obtained by less laborious testing of infested and uninfested workers released at some distance from their colonies. In these tests, infested workers needed about 2 times longer to return than the uninfested workers, and the proportion of bees that were not found in the colonies by late evening was 1.5 fold higher in the infested group.

Actual parasitization of a forager bee may conceivably influence the behaviour outside the colony, causing longer times of absence or even prevent its return. That the loss of foragers is higher in infested colonies was also supported by correlating forager infestation and bee loss in two colonies. Bee loss, determined by a bee counter, was about 2 fold higher in the colony where forager infestation was about 8 fold higher. The common observation in colonies collapsing due to Varroosis is a pronounced loss of forager bees (Martin, 1997) which corresponds well to the observed behavioural changes, though a shortened life span of workers emerging from cells infested by reproducing mites (Schneider and Drescher, 1987; Kovac and Crailsheim, 1988) and virus infections transferred by the mite (Bowen-Walker et al., 1999; Martin, 2001; Chen et al., 2004; Sumpter and Martin, 2004) might also contribute.

The orientation experiment further pointed to the interpretation that the extended returning times or lack of return of infested foragers might be more of an inability than a change of motivation. The infested bees were less likely to find the correct entrance without previously approaching a similar dummy en-

trance marking close by. In accordance with the use of visual cues to find the colony (von Frisch, 1967), only few workers crossed the unmarked circle. As bees were already aiming toward the colony, they were likely to be equally motivated for return, and the observed behavioural changes thus may reflect a deficiency in orientation ability. Impaired orientation might also contribute to enhance drifting between colonies, which was shown to increase with the level of *V. destructor* infestation (Sakofski, 1990).

In conclusion, the results show that a main factor contributing to the lower infestation of returning forager bees is an influence of the parasite on the host to the effect that the bees fly longer or may not return at all, possibly due to impaired orientation. It needs to be noted that due to frequent host changes of the mites between workers it can be assumed that the workers of the uninfested control groups had likely also been infested some time before. The true effects of the parasites thus are probably underestimated and might be more pronounced in workers from completely parasite-free colonies. High general parasite levels might also explain the unusually high rate of non-returning workers in the video recordings, even in the uninfested workers. This was especially pronounced in 2001, where the colony broke down in late August.

The frequent host change of the mites also suggests that the effects of the parasite might operate within comparatively short time after a mite has entered a bee. If being parasitized influences the nervous system, as indicated by the orientation experiment, it would be interesting to know whether general systems such as the immune and endocrinological system are involved because these might affect learning abilities (Mallon et al., 2003; Maleszka and Helliwell, 2001) and respond to stress (Lin et al., 2004).

The changed flight and orientation behaviour may be interpreted as an adaptive response. Direct physical or physiological effects lead to similar effects from parasitization by tracheal mites (*Acarapis woodi*, Harrison et al., 2001). However, in respect to the small size and weight of the *Varroa* mite and its low hemolymph uptake such physiological effects

are less likely to be a cause. The behavioural changes could be in the interest of the parasite to enhance spread into other colonies, a point supported by increased drifting of foragers from infested colonies (Sakofski, 1990). The behavioural changes might also be for the benefit of the bees. Rather than being an advantage to diseased foragers with low life expectancy to take higher risks to increase food collection, as proposed by Woyciechowski and Kozłowski (1998) for bees infested by *Nosema apis*, we suggest there could be a colony benefit from not returning to the colony by the removal of the pathogens. Smith-Trail (1980) discussed the possibility for an adaptive sacrifice of one's life for the benefit of kin in response to parasites ("suicide hypothesis"), and some examples have since been described in butterflies (Shapiro, 1976) and aphids (McAllister et al., 1990). In social insects, bumble bees infested by conopid flies respond by behavioural changes costly to the individual but beneficial to the colony (Müller and Schmid-Hempel, 1993), but up to now there are no clear examples that parasitized individuals might leave the colonies in order to remove pathogens (Schmid-Hempel, 1998).

Though we cannot conclude whether the behavioural changes in this study are more beneficial to the parasite or to the host, the cost-benefit ratio in honey bees infested by *V. destructor* would presumably be in favour of suicidal behaviour. Colonies could greatly benefit from timely removal of *V. destructor* mites which threaten colony survival at numbers of some thousands, while the loss of similar numbers of workers may be tolerable to strong colonies. Furthermore, to sacrifice their life for the benefit of the colony is common in honey bee workers during defence, where they lose their lives after fixing their sting apparatus in the skin of a mammal intruder. As *V. destructor* is a relatively recent pest to the western honey bee *A. mellifera* and specific adaptations cannot be expected, we hypothesize that not returning to the colony could be a more general response to diseases, and might be a trait which could be enhanced in breeding programs to strengthen the behavioural defence against *V. destructor* and, possibly, to other honey bee diseases.

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Résumé – L'acarien parasite *Varroa destructor* influence la durée de vol et la capacité de retour au nid des butineuses d'*Apis mellifera*. *Varroa destructor*, ectoparasite des colonies d'abeilles domestiques, effectue son cycle de développement à l'intérieur des colonies, mais quitte aussi les colonies accroché aux dos des butineuses. Les études sur la dynamique des populations et les mécanismes de résistance ont porté principalement sur les paramètres internes à la colonie. Nous avons étudié la perte des acariens au cours du butinage et l'influence de *V. destructor* sur le comportement de vol des ouvrières. Conformément aux études antérieures, les butineuses qui rentraient à la colonie étaient moins infestées que celles qui la quittaient. Un enregistrement vidéo des butineuses quittant la colonie ou y rentrant a montré que soit les acariens étaient perdus par les butineuses qui rentraient soit les butineuses infestées ne rentraient pas (Tab. I). Chez les ouvrières infestées, l'intervalle de temps entre le départ et le retour était presque doublé (Fig. 1). Des ouvrières infestées et non infestées ont été relâchées à quelque distance de la colonie pour mesurer l'heure du retour. Les ouvrières infestées mettaient 2,3 fois plus de temps pour rentrer à la colonie et la proportion d'abeilles infestées qui ne rentraient pas le soir était 1,5 fois plus élevée. La perte de butineuses par vol a été mesurée dans une colonie fortement infestée (C+) et une colonie faiblement infestée (C-). L'infestation des butineuses a été plus forte dans la C+ et la perte de butineuses par vol plus élevée. Lors d'un test d'orientation visuelle, les abeilles infestées ont été mieux notées pour leur précision à trouver l'entrée de la colonie (Tab. II). Les résultats montrent que l'infestation par *V. destructor* influence le comportement de vol des butineuses de sorte qu'elles peuvent ne pas rentrer à la colonie. Notre interprétation est qu'il s'agit d'un comportement d'adaptation des abeilles pour éliminer les parasites ou les pathogènes de la colonie.

***Varroa destructor* / *Apis mellifera* / butineuses / comportement de vol / retour au nid / relation hôte-parasite**

Zusammenfassung – Die parasitische Milbe *Varroa destructor* beeinflusst die Flugdauer und die Fähigkeit zur Heimkehr von Bienenarbeiterinnen (*Apis mellifera*). Der Lebenszyklus der parasitischen Bienenmilbe *Varroa destructor* findet innerhalb der Bienenvölker statt, allerdings kann diese die Völker mit den Sammlerinnen auch verlassen. Die meisten Untersuchungen zur Populationsdynamik der Milben und zu Resistenzfaktoren der Bienen beziehen sich auf die Situation innerhalb der Bienenvölker. Diese Untersuchung befasst sich mit dem Verlust an Milben während des Sammelns und dem Einfluss der Milben auf das Flugverhalten der Bienen. In Übereinstimmung mit früheren Untersuchungen wiesen die zurückkehrenden Sammlerinnen einen niedrigeren Befall auf als die ausfliegenden Sammlerinnen. Durch Videoaufnahmen am Flugloch konnte gezeigt werden, dass die Milben entweder die Arbeiterinnen verlassen hatten, oder dass die befallenen Arbeiterinnen nicht zurückgekehrt waren (Tab. I). Bei den befallenen Arbeiterinnen war die Gesamtflugzeit (Zeit zwischen dem Verlassen des Volkes und der Rückkehr) fast doppelt so lang wie bei Unbefallenen (Abb. 1). Außerdem wurde die zur Rückkehr benötigte Zeit gemessen, indem befallene und nicht befallene Arbeiterinnen in einigem Abstand vom Bienenvolk aufgelassen wurden. Die Rückkehrzeit war bei den befallenen Arbeiterinnen etwa 2,3 mal länger, und der Anteil der bis zum Abend nicht zurückgekehrten Bienen war um das 1,5fache erhöht. Bei dem Vergleich des Bienenverlustes zwischen einem hochbefallenen und einem niedrig befallenen Volk wiesen die höher befallenen Sammlerinnen eine höhere Verlustrate pro Flug auf. In einem Test zur visuellen Orientierung schnitten die befallenen Bienen beim Auffinden des Nesteingangs schlechter ab als die unbefallenen (Tab. II). Die Ergebnisse zeigen, dass der Befall mit *V. destructor* das Flugverhalten der Arbeiterinnen beeinflusst mit der Folge, dass diese mit erhöhter Häufigkeit nicht in ihr Volk zurückkehren. Dies wird als adaptives Verhalten der Bienen interpretiert, um Parasiten oder Pathogene aus den Völkern zu entfernen.

***Varroa destructor* / Sammlerinnen / *Apis mellifera* / Flugverhalten / Wirt-Parasitbeziehung / Heimkehrverhalten**

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