Original article

The deposition of anal excretions by *Melipona favosa* foragers (Apidae: Meliponinae): behavioural observations concerning the location of food sources

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Abstract – *Melipona favosa* consistently deposited anal excretions while foraging. Anal depositions were released more frequently and by more bees on artificial food sources at a greater distance from the nest. Our hypothesis that these deposits serve as scent marks is supported by experimental evidence regarding the choices made by foraging bees arriving at food sources either with or without anal excretions. The clearly reduced visitation rate in the experimental situation without depositions indicates the importance of these cues during visitation of the food source.

chemical communication / anal excretions / *Melipona favosa* / recruitment / stingless bees

1. INTRODUCTION

The communication systems of eusocial stingless bees (Apidae, Meliponini) (Michener et al., 1974) differ from those of honey bees. It is well known that *Apis mellifera* transmits information about direction and distance of the food source by performing dances on the vertical combs in the nest. This locomotive signal is accompanied by odour and sound signals (Kirchner and Dreller, 1993). Stingless bees do not have dances comparable to those of the honeybees.

Stingless bee communication systems involve sound production (Esch, 1967; Esch et al., 1965; Hncir et al., 2000; Michener, 1974; Nieh and Roubik, 1998), scent marking (Kerr, 1994; Lindauer and Kerr, 1960;

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Nieh, 1998; Wilson, 1971; Wille, 1983), and piloting (Esch, 1967; Esch et al., 1965). As in honeybees the odour of the food source plays an important role in recruitment: the impregnated odour on the body and mouth parts of foragers gives information to nestmates (Esch et al., 1965; Lindauer and Kerr, 1960; Michener, 1974). A combination of cues and signals allows at least some stingless bees to communicate the location of a food source in three dimensions (Lindauer and Kerr, 1960; Nieh and Roubik, 1995) compared to two-dimensional communication in honeybees (von Frisch, 1967; Lindauer and Kerr, 1960). In *Melipona panamica*, for example, the duration of sounds emitted during unloading is correlated with the height of the food source (Nieh and Roubik, 1998). In various *Melipona* species the duration of sounds emitted while dancing is positively correlated with the distance to the food source (Esch et al., 1965; Michener, 1974; Nieh and Roubik, 1998). The direction of the source is probably communicated outside the nest by means of the pilot system (Esch et al., 1965; Michener, 1974). In this system a group of bees follows a guide bee to the food source. In addition, scent marking along the route (Lindauer and Kerr, 1960) and a scent beacon deposited by the bees near a food source (Nieh, 1998) seem to guide newcomers to the indicated food source.

Communication about food source location by means of scent marks in stingless bees

Scent marks are used as a communication method in at least 24 stingless bee species. Not only do foragers deposit mandibular scent marks at the food source, they also place these marks on the ground or vegetation (Lindauer and Kerr, 1960). The distance between scent marks is reported to differ from species to species. The effect of such marks may last for 8 to 19 minutes

(Kerr et al., 1981). Most of these studies state that scent marks consist of secretions from mandibular glands (Johnson, 1987; Kerr 1969, 1994; Kerr et al., 1963, 1981; Lindauer and Kerr, 1960). In addition to mandibular secretions, foragers of different *Melipona* species leave anal excretions at or near the food source (Kerr, 1994; Nieh, 1998 and preliminary obs. by first author on *M. favosa*). The role of these anal droplets in food source communication is still unknown. Nieh (1998) states that foragers of *M. panamica* do deposit a scent beacon at the food source, but that neither mandibular nor anal secretions, nor the sucrose solution itself are the origin of it.

To increase our knowledge on the role of scent marks in communication we studied the deposition behaviour of anal excretions by *M. favosa* workers in relation to the distance from a food source. We also examined the effect of these excretions on the behaviour of other foragers.

2. MATERIALS AND METHODS

2.1. Bee species and study site

Two queenright colonies of *Melipona favosa* (about 275 bees at the end of September) from Trinidad and Tobago (West Indies) were housed in wooden observation hives and installed in a large tropical greenhouse (1.5 ha) at Burgers' Zoo, Arnhem, The Netherlands. We collected data on foraging behaviour from July until October 1995. During the study the minimum day temperature was 21 °C and the maximum temperature was 32 °C. The relative humidity was between 65% and 95%.

2.2. Training, foraging on food sources at different distances and anal excretions

Bees of *M. favosa* were individually marked with paper tags with two-letter

codes. We trained the bees to visit a feeder with 2 M honey solution. Once they had started visiting the feeder, we gradually moved it farther away to selected distances from the hive: 1 m, 9 m, 21 m, 36 m and 49 m.

We recorded data about the behaviour and the arrival time of individual bees landing at the feeder. Observation periods lasted one hour for each distance. Particular attention was given to the deposition of excretions at the feeder or on plants in the surroundings. We measured the volumes of the anal excretions using calibrated capillary tubes (Drummond 5 µl). We collected fresh drops immediately after deposition and analysed the carbohydrate (Carroll et al., 1956) and the protein contents (Schacterle and Pollack, 1973).

2.3. Effect of food concentration on the frequency of anal excretions

To test whether sugar concentration influenced the frequency of anal excretions, we used three concentrations (0.5 M, 1.0 M and 2 M honey-water, using a 80% honey, weighed with a Hand Refractrometer ATAGO Type 500). We trained the bees to visit a feeder 9 m from the hive. We offered a series of honey solutions (2, 0.5, 2, 1, 2 M) for 30 min. We performed an analysis for a total of 18×30 minutes. Each sequence was presented on a single day, around midday. Data about the frequency of anal excretions was collected using the "one-zero sampling" method (Lehner, 1979).

2.4. Measuring the effect of anal excretions on bee behaviour

We fed the foragers using a small container placed on a removable glass plate (20×20 cm). Instead of honey we now used 2 M sugar water solution to prevent influence of the odour of the food itself.

2.4.1. Effect on the visitation rate

We studied the effect of the depositions on bees flying to the food source 17 m away from the hive. We trained the bees for at least 15 min at a feeder 10 m from the hive, then the feeder was moved to a new distance 17 m from the hive. With the feeder at 17 m we started the experimental situation (1) in which we removed all depositions. We compared the flight frequency and frequency of excretion with a control situation (0) where excretions were left in place. We replaced the glass plate each time an excretion was deposited during the experimental situation and cleaned all of the plates with 70% alcohol at the end of the day. One observation period consisted of observations of 2×30 minutes (30 min: experimental setup and 30 min: control setup). There was a 15 min break between the two half hours. We noted the time of arrival of individual bees upon every flight to the feeder. The time of arrival was taken to be the moment a bee started to drink for the first time.

2.4.2. Effect on how bees locate and arrive at the feeder

We studied the effect of depositions on the timing and behaviour of the first arrival at a feeder that was moved to a new place. After training the bees for at least 15 minutes at a feeder 10 m from the hive, it was moved to a new place with different distance and direction. Once we placed the feeder in the new position the period of 30 min observations started. The experimental setup (1) consisted of 30 min observation using a clean plate which was replaced by a clean one before the observation started. In the control setup (0) the excretions were left in place.

We defined the time when the feeder was located as the time a bee came circling within one metre of the feeder. The difference between the time when feeder was located and the actual landing at the new

feeder was called the "orientation time". Furthermore, we recorded the total number of flights carried out and how many times the bees deposited excretions on the feeder.

2.4.3. Effect on feeder preference

To analyse whether bees discriminated between a cup (block of plexiglass $2 \times 2 \times 0.5$ cm) with and a cup without anal excretions, cups were placed only 5 cm apart and on the same feeder. For this, we first trained the bees at the feeder placed at a distance of 10 m from the hive and with only one cup. We allowed anal excretions to be deposited during a period of 15 min. Thereafter we put the control cup without anal excretions 5 cm apart from the cup with anal excretions. Control and experimental cups were shifted randomly during the observations. We repeated this experiment 60 times and between the repetitions we put the marked feeder back in the middle of the short distance between experimental and control cup.

2.5. Statistical analysis

We tested the association between the frequency of anal excretions and distance using the stratified χ^2-test for trends (EGRET programme). We tested for the effect of different sugar concentrations using the Chi-square test. The effect of anal excretions on visitation rate, arrival time and preference were tested using the t-test.

3. RESULTS

3.1. Flight pattern and behaviour at feeder

Three minutes after we had started training by offering food at the entrance to the nest, a forager made its first flights, but not in a straight line to the feeder. These flights were in a zigzag pattern or around the feeder. The area covered by the first flights was larger than that during the following flights when the feeder was gradually located by zig-zag movements. Bees spent some time resting somewhere near the feeder, where they performed standing, pumping abdomen and body grooming activities.

Bees at the feeder never had direct social interactions with other bees. Most common activities were: landing, walking, resting, pumping the abdomen, depositing anal excretion and grooming. Less common activities during standing were moving mandibles (opening and closing), trembling and extending the proboscis. After one and a half hour of foraging the bees started rubbing mandibles on the edges of leaves of plants near the feeder. This behaviour resembles the behaviour described by Kerr (1994). We observed bees rubbing mandibles on the edge and horizontal surface of the feeder, and the edges of plant leaves near the feeder. They also dragged their venter over the substrate.

3.2. Deposition of anal excretion

After food uptake bees sometimes released a droplet from the distal part of their abdomen on the substrate. While depositing, they often moved the abdomen to the left and right in a zigzag manner, scattering the droplet over the substrate. Occasionally they released an excretion with a small squirt. Sometimes two drops were deposited, the first being bigger than the second. Foragers also deposited anal excretions over an area of 14×14 cm on the feeder plate very close to the small food container (87%, N = 141), (total feeder table area is about 20×20 cm). Autogrooming or standing behaviour sometimes preceded the anal deposition behaviour. After the deposition the bees flew off; sometimes they left excrement at the feeder table. The excrement consisted of a viscous orange liquid, which smelled like pollen from the storage pots. We never saw bees ingesting the anal excretions. Deposited excretions always originated from the anus.

3.3. Nature and volume of the anal excretions

The anal liquid was transparent and only very light yellow. A chemical analysis of the samples indicated that it contained very small quantities of carbohydrates (12–16 µg/µl) and proteins (2.0–6.7 µg/µl). The bees deposited on average 0.5 µl of liquid (S.D = 0.3, N = 88) at 9 m and 0.2 µl (S.D. = 0.3, N = 11) at 49 m.

3.4. Effect of distance on frequency of anal excretions

There was a significant positive association between the distance of the food source and frequency of the excretions ($\chi^2_{[1]} = 210.87$, $p < 0.001$) (Fig. 1). Anal excretions also occurred before food uptake. We found that the release of excretions before feeding was more frequent over the medium distances of our experiment; in parenthesis the total number of excretions before uptake, two repetitions at each distance: 1 m: 0%, 9 m: 2.34% (N= 14); 21 m: 4.3% (N = 21); 36 m: 11.6% (N = 42) and 49 m: 1.27% (N = 2).

Bees visiting the feeder deposited anal excretions at least once. The percentage of bees depositing peaked at 70 to 80% when feeders were more than 21 m from the nest (Fig. 2).

When the distance between the hive and the feeder was more than two metres the bees deposited excretions not only on the feeder but also on plants around it. Bees did not deposit anal excretions while taking up food at 1 m. Foraging bees (N = 8) were seen depositing an anal excretion during their first flight on the artificial feeder. During training the bees deposited anal excretions on leaves near the old position immediately after the feeder was moved out to the selected distance.

3.5. Effect of sugar concentration

There was no difference between the number of anal excretions deposited by bees feeding from different sugar concentrations (Chi-square test: χ^2:1.40, $p = 0.50$). Bees that imbibed food with the lowest concentration did not deposit significantly more anal excretions than bees that imbibed food with the highest concentration (35.36% and 26.1% respectively, Chi-square test: χ^2: 1.38, $p = 0.24$).

3.6. Do anal excretions influence bee behaviour?

3.6.1. *Visitation rate*

The visitation rate (number of flights each individual performed in 30 minutes) was
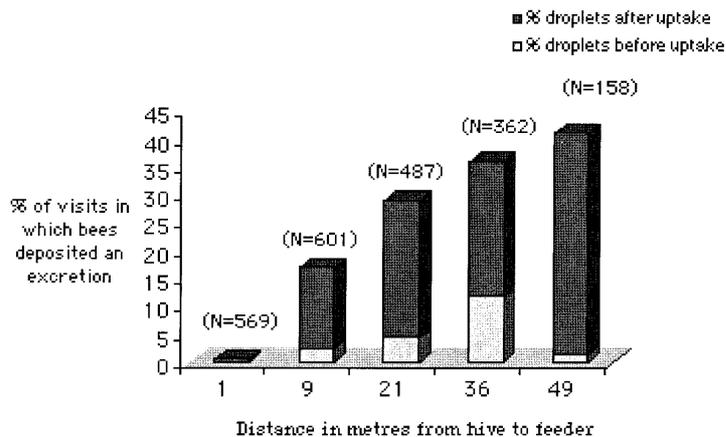

Figure 1. Increase of the average percentage of excretions in relation to distance. (N = number of visits to feeder.)
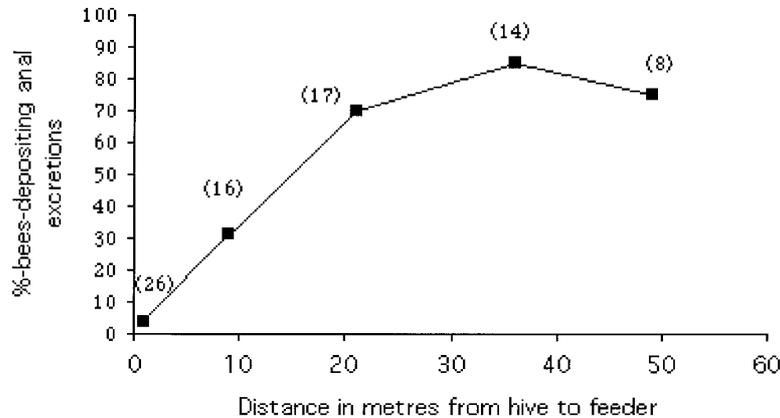
Figure 2. Variation of the percentage of depositing bees at different distances from the food source. Number of visitors is given in brackets.

significantly lower in the experimental situation where anal excretions were systematically removed upon being deposited, than in the control situation (average 18.0 and 20.4 respectively; t-test, df = 176, $p = 0.0006$; Fig. 3). The number of depositions was similar in both situations (average 4.7 and 4.8 respectively; t-test, df =105, $p = 0.9$, Fig. 3).

The arrival time (the time a bee starts drinking for the first time) was twice as long in the experimental observations than in the control situation (136.1 s $\pm$ 286.2 and 72.0 $\pm$ 155.0 respectively; N = 89 bees); the difference was significant (t-test, df = 176, $p = 0.008$).

3.6.2. The effect of anal excretions on the behaviour upon arrival at the feeder

We recorded the behaviour of the bees in 14 observational periods with the feeder placed in a new position (different direction or distance). Twenty-five bees participated in the control observations (feeder with anal depositions; six sessions) and in the experimental observations (new feeder without depositions; eight sessions). Generally the individual bees arrived alone and took time to orient themselves. There is no effect of the anal excretions on the timing and behaviour of the first arrival at the artificial food source

Figure 3. The effect of the anal excretions on the average visitation and deposition rate $\pm$ S.D. within 30 min (t-test, p-value between brackets). Control setup with depositions (0) and the experimental setup from which depositions had been removed (1).
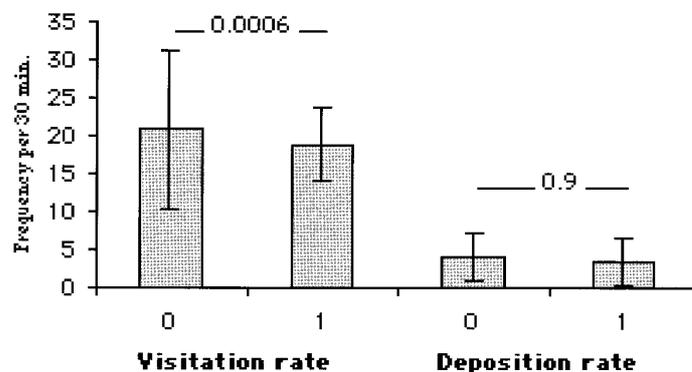
(Tab. I). We saw again a tendency of the visitation rate to be lower when a feeder without depositions was used. Moreover, the visitation rate was not significantly lower on the feeder without anal excretions (*t*-test, df = 37, *p* = 0.3).

3.6.3. Preference between food sources

In this experiment with 60 repetitions 293 separate flights took place. We recorded whether a bee opted for the clean feeder or a feeder with depositions. Bees had a very high preference for the feeder with depositions (68.3%, *t*-test, df =118, *p* = 0.0001).

4. DISCUSSION

4.1. Frequency of anal excretions

There have been earlier reports that *Melipona* species deposit scent marks in order to recruit newcomers to a specific place (Kerr and Ferreira, 1963; Kerr et al., 1981). However, they do not give clear details of the deposition behaviour or its function and solely imply that the scent marks are mandibular gland secretions (Kerr and Ferreira, 1963; Kerr et al., 1981; Lindauer and Kerr, 1960) or urine (Kerr and Rocha, 1988). On the other hand, the results of our study show that *Melipona favosa* systematically deposits anal excretions on substrates during foraging at artificial feeders.

There was a positive relationship between the frequency of excretions and distance up to 49 m from the colony.

Several explanations may explain the function of this behaviour. First, the deposition of anal excretions may be used for self-communication. The more frequent release of anal substances at larger distances could be a technique used by bees who find it more difficult to find the food source and thus have more need to mark that location. This strategy seems reasonable because of the environment they inhabit, tropical forest, where the role of the landmarks in their communication systems is still unknown. The second explanation could be that anal excretions are used as a recruitment strategy. It is possible that the information transferred inside the nest gives only partial recruitment information and experienced foragers need to add more cues near the food source. This explanation is supported by Jarau et al. (2000), they found that communication of distance in *M. scutellaris* and *M. quadrifasciata* is not accurate. These species communicate distance only up to 30 m and 40 m from their hives respectively. Also, this explanation could be valid particularly when it was found that the newcomers (*M. panamica*) obtained directional information outside the nest by a mechanism yet undiscovered, while height and distance were communicated inside apparently through the pulsed sounds produced

Table I. The effect of depositions on the behaviour of the bees upon arrival at the feeder after transfer of the feeder to a new spot. Control setup with depositions (0) and experimental setup without depositions (1). The mean and S.D. are given in seconds or minutes. The probabilities of the *t*-test are given in the lower row.

	Localization		Arrival		Orientation		Visitation rate		Deposition rate	
	s	S.D.	s	S.D.	s	S.D.	min	S.D.	min	S.D.
0	479.4	398.7	502.6	394.9	23.3	53.0	20.8	10.6	4.1	3.2
1	499.6	381.8	546.5	412.8	68.9	228.8	18.9	4.9	3.4	3.1
P	0.9		0.7		0.4		0.3		1	

by recruiting foragers (Nieh and Roubik, 1998).

On the other hand, recruits of *M. panamica* can orient to a scent beacon only when they are 6–12 m from the food source (Nieh and Roubik, 1995). Nieh and Roubik (1998) found that after replacement of the feeder by a clean feeder and removal of trained *M. panamica* bees, significantly more newcomers still arrived at the training feeder. Their result shows that foragers can communicate distance inside the hive; nevertheless, they did not remove the anal excretions left on the vegetation, around the feeder or elsewhere between hive and feeder. The clean feeder substitution did not eliminate food odor and anal excretions deposited by foragers in the region from hive to feeder station. That could also be an additional part of the information for newcomers to reach that place.

The third possible explanation would be that anal excretions are used to excrete excess of water which allows for larger nectar volume to be carried and/or lowers bee mass which reduces flight (Nieh and Roubik, 1998). Our approach indicates that anal excretions are not merely water excess. We expected an increase in the frequency of anal excretions as food source becomes more diluted (e.g. in *M. panamica*) (Nieh, 1998), nevertheless, we observed that bees that imbibed food with the lowest sugar concentration did not deposit significantly more anal excretions than bees that imbibed food with the highest concentration. Consequently, we can hardly say that anal excretions are deposited by stingless bees to reduce their weight or the flight cost. In *Apis mellifera* the excretion of a large volume of water has a high metabolic cost (Visscher et al., 1996). This could explain how it can be that *M. favosa* foragers deposited a larger volume of anal excretion at a shorter distance from the hive. Furthermore, study of different glucose concentrations carried during flight by *A. mellifera carnica* demonstrated that only a minimum quantity of

water reached the rectum (Crailsheim, 1988).

What remains unclear in our study is that as a function of foraging in *M. favosa* the percentage of anal excretions before uptake increased with increasing distance up to 36 m, but a decrease at the 49 m station was observed. We may consider that other osmoregulatory problems could be the cause of water production during flight, for instance, this is the method by way of water foraging honeybees gather small amounts of sugar (Visscher et al., 1996) or as happens to *Xylocopa capitata*, where water excretions are the result of the low concentrations of Na and K in the nectar they ingested (Nicolson, 1990). Therefore, we may think about why for some stingless bees (e.g. the small *Plebeia frontalis* [personal observation] or the large *Melipona favosa* and *M. beecheii*) excretions of metabolic water would be an advantageous proceeding.

4.2. The anal depositions in the communication system of *M. favosa*

On the basis of our findings in this enclosed environment, we hypothesise that the release of anal excretions on and around the feeder may serve as communication during foraging for *M. favosa*. We clearly observed that bees preferred the food source with the anal excretions, thus such deposits could serve to localise the food source. The clearly reduced visitation rate in our experimental situation without depositions ($p = 0.0006$) indicates that experienced bees are attracted to their own odour (perhaps the anal excretions' odour). As we worked mostly with trained bees these results may support the self-communication hypothesis.

Further, there is a less-defined group of scents used by social insects which may attract other individuals, for example, the food odour, the bee odour, the colony odour including other odoriferous substances

produced by exocrine glands (Butler, 1970). For instance, the sting apparatus is absent in the stingless bees but there is a vestigial sting poison sac in some species (e.g. *M. quadrifasciata*) (Kerr et al., 1962), as well as, it seems they have a single gland opened at the end of the abdomen which has been considered a vestige of Dufour's gland (Lello, 1976). Unfortunately, information on the function of single glands of stingless bees is not available, nor their behavioural significance (Duffield et al., 1984; Lello, 1976). The fact that anal depositions were found to contain minor quantities of carbohydrates and protein indicates that such glandular secretions may be released in the anal excretions. Also, research by Jarau et al. (2000) indicates that recruited bees of *M. scutellaris* search for a food source smelling like a sample previously taken inside the hive. This approach suggests that the odour of the food is one of the most important factors in communication in this species.

Thus, the anal excretions may play a role as a cue to forager bees along the route to the food source as a side effect that it can be used as food source information. This is similar to the fecal pellets of the termite *Nasutitermes corniger* and the ant *Myrmica ruginodis* that serve to lay a scent trail (Butler, 1970). Anal excretions can also be used (e.g. *X. capitata*) in defence of the nest (Nicolson, 1990). The assumption that the anal excretions may emphasise the trajectory to the food source could explain why newcomers arrived equally at both feeders (opposite direction from hive), during remotion of experienced *M. panamica* foragers from potential newcomers (Nieh and Roubik, 1998), they were not allowed to lay the anal excretions. Nieh (1998) concluded that *M. panamica* does not need a scent trail to recruit over a watersurface. Furthermore, the absence of a scent trail does not mean that the bees do not use scent at all. Thus, it remains to be tested whether these anal excretions (*M. favosa and M. panamica*)

can be important only for experienced bees or for others.

We found that the scent of the depositions stimulates the bees to reduce the time they need for a round trip. However, an important aspect in this particular experiment has to be mentioned: in most of the observational periods, the bees were familiar with the replacement from the training place at 10 m to 17 m at the start of the observation. Thus this experiment is not sufficient in finding a clear effect of the depositions on arrival. More observations have to be made with similar experiments.

In conclusion, it is possible that scent of the anal excretions should have an effect on large distances, although it could not be observed clearly in this study. Much more research is needed in order to know if anal excretions have a communicative function, basically because it is difficult to analyse a complex mixture of substances which might be present in very small quantities. The anal excretions may contain a scent beacon deposited during deposition of the anal excretions, its own odour and probably the food odour. It is less feasible that the latter could play a role in communication in *M. panamica* because they always landed at the training feeder whether or not they were food-scented (Nieh, 1998). Finally, the physiological causes of the anal excretions and their relation to stingless bee metabolism is an important aspect yet to be taken into account for future research.

Résumé – Le dépôt d'excrétions anales par les butineuses de *Melipona favosa* (Apidae, Meliponinae) : observations comportementales sur la localisation des sources de nourriture. Les marques odorantes sont utilisées comme moyen de communication chez au moins 24 espèces d'abeilles sans aiguillon. La plupart des études affirment que les marques odorantes sont constituées de sécrétions des glandes mandibulaires, mais les butineuses de diverses espèces de *Melipona* déposent des sécrétions anales près de la source de nourriture. L'étude porte sur le dépôt systématique d'excrétions anales par des butineuses de *M. favosa,* chez laquelle nous avons mis en évidence une relation entre la présence de dépôts et la distance des sources de nourriture, situées entre 1 m et 50 m du nid. Nous avons entraîné les abeilles à visiter un nourrisseur comportant une solution de miel à 2 M. Lorsque le nourrisseur commençait à être visité, il était éloigné graduellement à diverses distances. La fréquence totale des dépôts a été significativement plus élevée lorsque le nourrisseur était loin de la ruche. Une analyse chimique des échantillons a montré que les dépôts contenaient de très petites quantités d'hydrates de carbone (entre 12 et 16 µg/µl) et de petites quantités de protéines (environ 2,0 à 6,7 µg/µl).

Nous avons enregistré le comportement et l'heure à laquelle les abeilles se posaient sur le nourrisseur. Nous avons étudié l'effet des dépôts sur les abeilles qui volaient vers la source de nourriture. Nous avons comparé la fréquence de vol et le fréquence de dépôts entre une situation test, où nous avons éliminé les sécrétions anales après leur dépôt par les abeilles, et une situation témoin où elles étaient laissées sur place. Le taux de visites (nombre de vols effectués par chaque abeille) en 30 min était significativement plus faible dans la situation test ($p = 0{,}0006$). Le temps d'arrivée, défini par la première prise de nourriture au nourrisseur, était deux fois plus long dans la situation test. Le temps de localisation et le temps d'arrivée étaient tous deux plus longs dans la situation test

dépourvue de marquage. Néanmoins les temps d'orientation, durée entre la localisation et l'atterrissage, n'étaient pas significativement différents. Le fait que des nourrisseurs nouvellement installés, marqués par des sécrétions anales, aient attiré les butineuses confirme notre hypothèse : les abeilles préféraient nettement les nourrisseurs avec dépôts de sécrétions anales, même s'il n'étaient éloignés les uns des autres que de quelques centimètres et qu'ils étaient déplacés fréquemment et au hasard.

Melipona favosa / abeille sans aiguillon / communication chimique / recrutement / excrétion anale

Zusammenfassung – Absetzen analer Exkrete durch Sammlerinnen von *Melipona favosa* (Apidae: Meliponinae): Verhaltensbeobachtungen bei der Auffindung von Futterquellen. Ein Absetzen analer Exkrete von sammelnden Stachellosen Bienen an Futterquellen kommt regelmäßig vor. Dieses Verhalten wurde bei *Melipona favosa* untersucht, weil bei diesen Bienen das Absetzen der Exkrete in Abhängigkeit von der Entfernung zwischen 1 bis 50 m von der Futterquelle auftritt. Wir dressierten Bienen auf eine Futterstelle mit 2 M Honiglösung. Hatten die Besuche einmal begonnen, verschoben wir die Futterstelle nach und nach auf die gewählte Entfernung.
Die Gesamtfrequenz der Exkretabsetzungen war bei größerer Entfernung vom Stock signifikant höher (Abb. 1 und 2). Eine chemische Analyse von Proben ergab, dass sie sehr kleine Mengen von Kohlenhydraten (zwischen 12–16 µg/µl) und geringe Menge Eiweiß (etwa 2,0–6,7 µg/µl) enthielten.
Wir erhoben Daten über das Verhalten und die Ankunftszeit von einzelnen Bienen, die am Futter landeten. Wir untersuchten die Wirkung der abgesetzten Exkrete auf die zur Futterquelle fliegenden Bienen. Wir entfernten bei einem Versuch alle Exkrete und verglichen die Flugfrequenz und die Frequenz

vom Absetzen des Exkrets mit einer Kontrolle, bei der die Exkrete am Futter belassen wurden. Die Besuchsrate (Anzahl der Flüge von einzelnen Bienen innerhalb von 30 Minuten) war unter den Versuchsbedingungen signifikant geringer ($p = 0,0006$) als bei der Kontrolle (Abb. 3). Die Ankunftszeit (die Zeit bis zur ersten Futteraufnahme) war am Versuchsfutterstand doppelt so lang wie bei der Kontrolle. Beides, die mittlere Zeit zur Auffindung des Futters und zur Landung, waren unter den Versuchsbedingungen ohne Markierung länger (Tab. I). Die Orientierungszeit (Zeit zwischen Auffinden und Landung) unterschied sich nicht signifikant. Neu eingerichtete, mit analen Exkreten markierte Futterstellen lockten Sammlerinnen an. Diese Beobachtung unterstützt unsere Hypothese, dass die Bienen in starkem Maβe Futterstellen mit abgesetzten Exkreten denen ohne Exkrete bevorzugen. Die Bevorzugung trat auch dann auf, wenn die Futterstellen nur ein paar Zentimeter von einander entfernt waren und ihre Positionen häufig und zufällig gewechselt wurden.

chemische Kommunikation / anale Aussonderungen / *Melipona favosa* / Rekrutierung / Stachellose Bienen

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