

Communication and recruitment for the collection of food in stingless bees: a behavioral approach

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The central point of my research has been the communication mechanisms and recruitment abilities concerning foraging activities of the stingless bees (Apidae: Meliponini) *Plebeia tica*, *Trigona corvina*, *Melipona favosa*, *Melipona costaricensis* and *Tetragonisca angustula*.

We first analyzed the recruitment and communication of food source location in *Plebeia tica*, *Trigona corvina* and *Tetragonisca angustula*. We showed that both distance and direction of the food source were communicated very accurately in *T. corvina*. In *P. tica* and *T. angustula* only the direction of the food source was communicated. Recruitment intensity was high in *T. corvina* (3.12 ± 2.5 recruits per scout flight) and much lower in *P. tica* (0.11 ± 0.17) and *T. angustula* (0.07 ± 0.04). Our results suggest that the mechanisms for indication of distance and direction in *T. corvina* involve experienced bees (trained bees) that guide new bees by means of pilot flights; significantly more recruits landed accompanied by the trained bee. Thereafter, piloting explained the arrival of one or two recruits with the experienced forager in *P. tica*. However, it could not explain the arrival of large groups of recruits in *T. corvina*. The behavior of the experienced bees away from the feeder suggested that a scent trail might be used by *T. corvina*.

In addition, we reported on the cues and signals that are used to locate the position of the food source and its value in the recruitment of new bees in *Plebeia tica* and *T. corvina*.

The role of olfactory information, especially that of food odor and scent-marks, on direct attraction to food sources in *P. tica* was investigated. We found that at close distance,

recruits and experienced bees use the odor of the food source to localize the source. In *P.tica* the odor of the food source is used for accurate orientation of the recruits. Likewise, recruits use scent-marks as location communication mechanisms at the food source.

Our choice experiments, in which we eliminated scent-marks at the control feeder (used without food odor), demonstrated the use of attractant scent-marks at the experimental food source. Within-nest recruitment behavior was also studied. The influx of sugar solution at the nest entrance caused *P. tica* foragers to make irregular runs inside the nest and increased trophallaxis activity. However, we found that the influx of sugar solution does not change the buzzing sound production of foragers.

We concluded that the elements used by *P. tica* in food source location communication make up a multimodal recruitment system with general alarming by means of weak sounds and random running performed by returning foragers inside the nest. Outside the nest, recruits use the odor of the food source and orientate with the scent-marks deposited by forager bees at the food source.

In *T. corvina*, recruitment was significantly more effective during experiments where the trained bee was present in comparison with experiments where the trained bee was removed. Moreover, we found that the odor of the food is important for the trained bee to relocate the food source (she is not guided nor does she use a trail of scent-marks).

Higher recruitment intensity during trials with scented food indicated that odor played a role in the recruitment process. Our observations support the hypothesis that both the trail of scent-marks and the trained bee are of crucial importance in the field. However, the recruits also have the trail of scent-marks to rely on and may not be too dependent on food odor in the field. During the experiments where a scented control feeder was placed in front of the scented training feeder, no recruits arrived at the control feeder. We concluded that food odor was not a crucial cue, although of some help, for recruits of *T. corvina*. We assume that for the recruits, food odor might be more important inside the nest, where the trained bee is communicating her find.

The deposition of anal excretions by *Melipona favosa* foragers and behavioral observations concerning the location of food sources were studied. *Melipona favosa* consistently deposited anal excretions while foraging. Anal depositions were deposited 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. We trained the bees to visit a feeder and, once they had started visiting the feeder, we gradually moved it further away to the selected distances. The total frequency of depositions is significantly higher at long distances from the hive.

The bees strongly preferred the feeder with depositions to the one without depositions, even if these feeders were only a few centimeters apart and their positions were frequently and randomly changed.

We also report on the analysis of the effect of sugar concentration and nectar source distance on the sound signals emitted by *Melipona costaricensis*. The principal finding presented is that *Melipona costaricensis* produces sounds that vary significantly in relation to varying sugar concentrations. Different sugar concentrations were used to determine the effect of the quality of a food source on sounds produced by the stingless bee *Melipona costaricensis* inside the hive. Sound signals may contribute to the recruitment of nest mates to rich food supplies.

Finally, the combination of intranidal and extranidal cues and signals allows the accurate food source location communication in the stingless bees. Within the nest, the information concerning the food source found can be transferred to the nest mates (potential recruits or experienced foragers) by a behavior that alerts (e.g. running, turning), informs quantitatively or qualitatively (e.g. sounds) and stimulates (e.g. food samples). These intranidal sources of information, if individually considered by the bees, would not provide adequate information

to stimulate or to bring recruits (naïve bees) to the specific food source locations for adequate and efficient exploitation. The incorporation of alternative cues and signals outside the nest probably depend on their suitability for either orientation or localization of the food source. The odor of the food source is one of the most common sources of information and it is used to communicate that "foraging is rewarding" but also indicates what type of food is rewarding. Some species are only orientated to the odor of the food source in their random search, others only use it as a back up system when one or more other recruitment cues are absent. The importance of the food odor as a cue practically disappears when food sources are located further away. In that situation, food odor is only important in the detection of the final location of the food source. Thus, communication of profitable food sources using the odor from that particular source is a mechanism that is used in several species, including bumblebees, stingless bees and honey bees.

The available information shows that 61% of the non-Melipona species from the total 46 already studied use scent-marks. It is clear that species indicating more precisely food source location use more sophisticated communication methods (e.g. sound signals and scent-marks). Scent marking turns out to be one of the most widespread and sophisticated methods used by stingless bees, together with the use of odor of the food source. Only about 12 % of the approximately 500 species of stingless bees in the world have been studied or observed in relation to the communication methods used for localization of food sources. Unfortunately, in many of these species the information has not been clearly documented.

Table 1: Summary of communication of location with cues and signals used by five species of stingless bees.

X*: Sounds correlated with the distance from food source and quality. X**: Experienced foragers use the odor of the food. ?: No information available. A: Odor of the food source. B: Buzzing. C: Zigzag running. D: Zigzag running + sharp turns. E: Audible sounds. F: Odor from mandibular glands. G: scent-marks. H: pilot flights.

Species	Location communication		Systems							
	Distance	direction	A	B	C	D	E	F	G	H
<i>T. (Tetragonisca) angustula</i>	No	Yes	x	x	x	x	–	?	?	?
<i>Plebeia tica</i>	No	Yes	x	x	x	–	–	?	x	x
<i>Melipona favosa</i>	?	?	?	–	–	x	x	?	x	?
<i>M. costaricensis</i>	?	?	?	–	–	x	x*	?	?	–
<i>Trigona corvina</i>	Yes	Yes	x**	?	?	?	?	?	x	x