NOTES AND COMMENTS



A scientific note on small hive beetle (Aethina tumida) oviposition and behaviour during European (Apis mellifera) honey bee clustering and absconding events

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Small hive beetles (*Aethina tumida*) were discovered in the southeastern USA in 1998 (Elzen *et al.*, 1999) where they have subsequently caused severe damage to colonies of European-derived subspecies of *Apis mellifera*. However, in their native range of sub-Saharan Africa (Hepburn & Radloff, 1998), they are considered only minor honey bee pests (Lundie, 1940; Pettis & Shimanuki, 2000); therefore small hive beetle behaviour is little studied and less understood. Here, we report observations on beetle behaviour during European honey bee clustering and absconding events and on beetle oviposition.

Experiments were conducted in Warren County, Georgia, USA, during August–September 2002. Six, 3-frame nucleus colonies were created from standard Langstroth-style hives; each colony had bees on all three combs, one comb with honey, two combs with brood, and a laying queen. One hundred small hive beetle adults were introduced into each of the colonies every day at dusk for 14 consecutive days. Two colonies absconded and beetle behaviour was monitored during the absconding events. On day 17 of the experiment, the remaining four colonies were put into a cool storage room (7° C) for one day (which caused honey bee clustering behaviour), after which they were frozen for two weeks. The colonies were thawed and the honey bee clusters examined for small hive beetles. The bees were then removed from the comb in order to examine female beetle oviposition.

The first of two colonies absconded on day 8. During absconding, 5–10 small hive beetles were seen leaving the colony with the bees. The queen and the swarm settled on the ground c. 15 m from the hive. The following day, an empty hive box was placed beside the swarm. A single small hive beetle entered the box with the cluster of bees indicating that the beetle had been present in the cluster. It is possible, however, that the beetle entered the cluster while the bees were on the ground. With the second colony, beetles were also seen leaving with the absconding bees; however this swarm was not captured and the presence of beetles in its cluster could not be confirmed.

We found 253–905 beetles in the four non-absconding colonies. In every case, most of beetles were found inside the bee cluster. Of the few beetles found outside the cluster (always < 50), most (> 75%) were within 5 cm of the cluster perimeter. Clustering bees often enter cells head first in order to form a more contiguous cluster. All such bees were pulled from their cells to facilitate individual cell examination. We found > 50 cells per colony containing beetles (some having > 5 beetles per cell). It is likely that the bees were clustering naturally and beetles infiltrated the cluster to keep warm. Our findings support earlier



FIG. 1. Evidence for small hive beetle ovipositing behaviour. Photo A shows a brood cell capping with an arrow pointing to a small puncture. After the capping of this cell was removed (photo B), one can see more than 15 small hive beetle eggs oviposited on or around the honey bee prepupa.

observations (Eischen, 1999; Pettis & Shimanuki, 2000) of small hive beetles in bee clusters.

Data were also collected on beetle oviposition. Others (Lundie, 1940; Schmolke, 1974) have shown that female beetles often oviposit on pollen reserves and in cracks of hives. However, there were no pollen reserves in any of the non-absconding colonies. Upon examination, we observed many puncture marks in the brood cell cappings (fig. 1a). When the cappings to these cells were pulled back or removed, we observed small hive beetle eggs (fig. 1b). In most instances, there were > 10 beetle eggs per cell; eggs were laid directly on or around the honey bee pupae. Approximately one-third of the remaining capped brood in all non-absconding colonies was affected in this way. Beetles may be able to bite holes in the brood cappings and insert their long, flexible ovipositors (Schmolke, 1974) to lay eggs on the immature bee. Further, we noted that all four colonies had begun aborting brood by the end of the experiment, as indicated by the presence of cannibalised larvae and pupae outside the hive entrances. Bees may have detected and removed the beetleinfested brood.

Analyses of four beetle-infested colonies (1500 A. tumida each) of Cape honey bees (A. m. capensis) in South Africa, established identically (food stores, number of bees, etc.) to the European colonies, showed no punctured brood cappings in any colony. However, punctured cappings appeared in the brood cells two days after small hive beetle females were given brood comb (4 cm²) in the laboratory (free from adult bees). Our data possibly

suggest that in their native range small hive beetles do oviposit in brood combs but only in the absence of adult honey bees, which contrasts with the results we found for European honey bees in the USA. Therefore, there may exist fundamental differences between European and Cape honey bee behaviours toward small hive beetles that help explain the comparative tolerance exhibited by *A. m. capensis*. Further, a reproductive threshold for small hive beetles in European honey bee colonies may exist, above which small hive beetle females are free-running in colonies and able to oviposit in unprotected brood combs.

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Fagara budrunga fruit extract as an attractant for Apis cerana





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Attractant formulations based on chemical constituents of Nasonov gland pheromone have been reported for *Apis mellifera* (Free et al., 1984), *A. cerana* (Naik et al., 1989) and *A. florea* (Naik et al., 2001). Screening of other chemicals for their attractant properties may offer a broader range of compounds. Plant extracts are a potential source of such molecules as they contain a variety of secondary metabolites such as volatile terpenoids.

Melissa officinalis, Lavandula latifolia and Euodia hupehensis can selectively attract swarms or foragers (Banhatti, 2002). 2-Keto tridecan-1-yl acetate and the corresponding alcohol are reported from *E. hupehensis* (Rutaceae). In our programme on medicinal plants, fruits of *Fagara budrunga* (Rutaceae), commonly known as mullilam, were extracted. The crude extract contains an aliphatic, long-chain ester (*Z*) 1'-propylbutyl 3-octadecenoate along with β -terpeniol, sabinene, phellandrene, oleic acid and linoleic acid (Naik *et al.*, 1999). These similarities with *E. hupehensis* prompted us to examine the hitherto unknown attractant properties of the extract of *F. budrunga* fruits.

Fruits of *F. budrunga* were collected from Konkan area of Maharashtra. The fruit had a sweetish pungent spicy odour. Fruits were shade dried, then coarsely powdered (2.7 kg) and extracted with ethanol (3×2 litre) at room temperature. The combined alcohol extracts were concentrated under reduced pressure to yield a dark-coloured, viscous liquid (60 g, 2.2 %). This extract was tested for its bioactivity on honey bees, *A. cerana*.

Bioassays of the extract were carried out in the campus of Agharkar Research Institute. We used a rotating table with a circular (diameter 60 cm), removable glass top mounted on an electric motor. The table-top was rotated once per three minutes while the tests were in progress and the top was changed after each test. Test formulations were prepared by mixing extracts of *F. budrunga* in liquid paraffin at 10 concentrations (50–1000 mg extract/ ml paraffin) (fig. 1). 30 µl of a formulation was placed on each of the eight pre-dried cylindrical blocks of plaster of Paris (1 \times 1 cm). These blocks were arranged symmetrically in a petri dish (8 cm diameter). Another petri dish with eight sim-



• Visiting the dish containing formulation of Fagara budrunga fruit extract

FIG. 1. Graph of the average number of bees visiting the dish of (\spadesuit) formulation of *Fagara budrunga* fruit extract (\blacktriangle) liquid paraffin, control and the attractiveness (\bullet) against concentration of the formulation.

ilar blocks loaded with 30 μl of liquid paraffin was prepared as control.

The test table was located 10 m from a colony of *A. cerana*. Before beginning the bioassay, bees were trained to feed on sugar syrup. After about 100 bees were trained, the top of the table was replaced with another top on which the dishes with the extract and the control dishes were kept diametrically opposite each other. Each petri dish was covered with a clean, dry wire

Attractiveness (A)