Chapter 18

Reproduction of *Varroa jacobsoni* Introduced into an *Apis cerana japonica* Colony

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Damage to *Apis mellifera* by *Varroa* mites is a serious problem for beekeepers; however, *A. cerana* suffers hardly any damage. The lack of damage may be because the sealed duration of *A. cerana* worker cells is too short to allow full development of *Varroa* offspring (Sasaki *et al.*, 1995). Drone production is limited to April–June (Sasaki, 1989). Workers rapidly remove mites by allo-grooming and kill them with their mandibles (Peng *et al.*, 1987). The ability of *A. cerana* to recognise and identify foreign material is high (Peng *et al.*, 1987). *Varroa* is rarely found naturally in *A. cerana japonica* colonies suggesting high resistance. When *Varroa* was introduced into an *A. cerana* colony, a few eggs and female deutonymphs were observed (Rath and Drescher, 1990; Tewarson *et al.*, 1992). The behaviour of *A. cerana japonica* workers and the reproduction of *Varroa* in an *A. cerana japonica* colony were investigated by introducing newly emerged *A. mellifera* drones each with one parasitic mite into an observation hive with two or three combs of *A. cerana japonica*.

Hygienic grooming behaviour induced by parasitic *Varroa* mites in *Apis cerana japonica*

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The Japanese honeybee (*Apis cerana japonica*) (Acj) and the European honeybee (*Apis mellifera*) (Am) share the same habitat in Japan. *Varroa jacobsoni* (Vj), originally an ectoparasitic mite of *A. cerana*, now heavily infests Am colonies. Experiments were conducted to compare the hygienic grooming behaviour of Acj and Am in response to *Varroa* parasitism. A semiochemical compound from cuticular extract of *Varroa* was identified using GC-MS, GC-EAD and behavioural bioassays. This compound induced typical hygienic allo-grooming behaviour in Acj worker bees. Am worker bees showed no response to mite or mite extract.

Materials and Methods

Wire net was used to separate combs and bottom board, which was covered with paper smeared with Vaseline. Mites that fell on the paper were counted. In the first experiment, 205 *Varroa* mites were introduced into an *A. cerana japonica* colony with worker cells over five days in May 1997. Nine days later, worker cells were opened and inspected for the presence of mother mites and offspring. In a second experiment, 543 *Varroa*

Predatory behaviour of *Vespa velutina* and *Vespa mandarina* attacking honeybee colonies

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Predatory wasps are serious enemies of honeybees in Himachal Pradesh, India. Ten species of wasps were observed preying on *Apis cerana* and *A. mellifera* colonies; of these, *Vespa velutina* and *V. mandarina* were of primary concern. Maximum attack by *V. velutina* was during August; September and October were peak periods of *V. mandarina* attack. Peak hours of attack varied for both species. Studies of behavioural traits revealed that *A. cerana* was more resistant to attack than *A. mellifera*. Some control measures to check the menace of these wasps in apiaries are suggested.

mites were introduced into an *A. cerana japonica* colony with drone and worker cells over eight days in June 1997. In this series, the queen died and worker cells disappeared. Fifteen days later, only drone cells were opened. The same procedure was also used for an observation hive of *A. mellifera* with drone and worker cells where 189 *Varroa* mites were introduced over eight days in July 1997.

Results

Introduction of *Varroa* mites into *A. cerana japonica* worker cells

Fallen mites were observed soon after introduction. The total number of fallen mites was 167 (81.5 %) (Table 1). There were 113 (67.7%) undamaged mites and 54 (32.3%) damaged. Thirty-six (66.6%) of the damaged 54 mites had damage on the legs, 15 (27.8%) on the legs and shield, and 3 (5.6%) on the shield. The other 38 mites apparently remained in the colony.

On Day 9, the colony was inspected for the 38 unaccountable mites. 1203 *A. cerana japonica* workers and 68 (33.2 %) introduced *A. mellifera* drones were found. Sixteen of the 1203 workers were infested with mites, leaving 22 mites unaccounted for. Worker cells were investigated on Day 10. Eight (3.4 %) of 238 worker cells were infested. Eight mother mites (3.9 % of introduced mites) were recovered from worker cells, and offspring were found in two worker cells (25 %) with pupae 14 days old (Table 2). The remaining unaccounted for 14 mites might have disappeared with introduced *A. mellifera* drones driven out of the hive.

Table 1. Number of fallen mites introduced with *Apis mellifera* drones into *Apis cerana japonica* observation hive

Day	No. of mites introduced with *A. mellifera* drones*	No. of fallen mites		
		Undamaged	Damaged	Total
1	30	14	9	23
2	40	14	8	22
3	35	10	8	18
4	60	17	3	20
5	40	11	6	17
6	–	7	3	10
7	–	2	4	6
8	–	6	1	7
9	–	32	12	44
Total	205	113	54	167 (81.5 %)

Note: * One *Varroa* mite per newly emerged drone.

Introduction of *Varroa* mites into *A. cerana japonica* drone cells

Fallen mites were observed soon after introduction. Total number of fallen mites was 268 (49.4 %) (Table 3). There were 198 (73.9 %) undamaged and 70 (26.1 %) damaged mites. Forty-seven (67.2 %) of the 70 damaged mites had damage on the legs, 19 (27.1 %) on the legs and shield, and four (5.7%) on the shield. The other 275 mites apparently remained in the colony.

On Day 15, the colony was inspected for remaining mites. 1668 *A. cerana japonica* workers, three *A. cerana japonica* drones and 279 (51.4 %) introduced *A. mellifera* drones were found. Twenty-three of the 1668 workers were infested with mites, leaving 252 mites still unaccounted for. On Day 16, drone cells were opened. Eighty-two (54.7 %) of the 150 drone cells were infested. 141 introduced mother mites (26 % of introduced mites) were still in drone cells, 66 (80.5 %) of

Table 2. Survival and reproduction of *Varroa* mites artificially introduced into worker cells of *Apis cerana japonica* colony (n = 238)

Age of worker pupa	Stage	Composition of mites								
		Egg	Proto-nymph		Deuto- nymph			New adult	Mother mite	Total
			male	female	male	female	male female			
13	Dark brown-eye pupa (dbe)	0	0	0	0	0	0	0	1	1
14	Dbe, lightly-pigmented thorax pupa	0	0	1	1	2	0	0	1	5
14	Dbe, lightly-pigmented thorax pupa	1	0	1	1	2	0	0	1	6
15	Dbe, medium-coloured thorax pupa	0	0	0	0	0	0	0	1	1
15	Dbe, medium-coloured thorax pupa	0	0	0	0	0	0	0	1	1
16	Dbe, dark thorax pupa	0	0	0	0	0	0	0	1	1
16	Dbe, dark thorax pupa	0	0	0	0	0	0	0	1	1
16	Dbe, dark thorax pupa	0	0	0	0	0	0	0	1	1

which were filled with offspring. Sixteen cells (19.5 %) contained mother mites only with no offspring. The pupal period from pink-eye to dark thorax was parasitised at a high rate. New adult mites were found at the stage of dark brown-eye pupa. Some were at an unknown stage, meaning that some infested pupae were not at an exact stage when they died (Table 4).

Introduction of *Varroa* mites into *A. mellifera* worker and drone cells

Fallen mites were observed soon after the introduction. The total number of fallen mites was 120 (63.5 %) (Table 5). There were 86 (71.1 %) undamaged and 34 (28.3 %) damaged mites. Twenty-seven (79.4 %) of the 34 damaged mites had damage on the legs, five (14.7 %) on the legs and shield, and two (5.9 %) on the shield.

On Day 15 after the introduction, the colony was inspected. 2301 *A. mellifera* workers and 83 drones were living in the colony, 81 of which were infested with mites. The mite total was 201 or 13 mites more than the total introduced. Possibly there were some mite survivors in cells although the observation hive had been treated with acaricide. Differentiation of introduced and survivor mites was impossible.

Discussion

The average rate of falling mites was 65.5 % for *A. cerana japonica* and 63.5 % for *A. mellifera* (Fig. 1). Peng *et al.* (1987) reported that 73.8 % of fallen mites in an *A. cerana* colony were damaged; Fries *et al.* (1996) reported 30 % in *A. cerana* and 12.5 % in *A. mellifera* colonies. Lodesani *et al.* (1996) noted that 26.1 % of dead mites in an *A. mellifera* colony were damaged. In these studies, the mite damage rate in *A. cerana* colonies was higher than in *A. mellifera* colonies. However, in this

Table 3. Number of fallen mites introduced with *Apis mellifera* drones into *Apis cerana japonica* observation hive

Day	No. of mites introduced with *A. mellifera* drones*	No. of fallen mites		
		Undamaged	Damaged	Total
1	50	18	1	19
2	45	11	1	12
3	35	7	6	13
4	30	5	3	8
5	17	7	6	13
6	75	9	1	10
7	128	41	12	53
8	163	44	14	58
9	–	5	8	13
10	–	8	2	10
11	–	11	3	14
12	–	8	7	15
13	–	11	5	16
14	–	8	1	9
15	–	5	0	5
Total	543	198	70	268 (49.4 %)

Note: *One *Varroa* mite per newly emerged drone.

Table 4. Survival and reproduction in drone cells of *Varroa* mites introduced into *Apis cerana japonica* colony (n = 150)

Stage	No. of cells examined	No. of infested cells (%)	Eggs	Proto-nymph male	Proto-nymph female	Deuto-nymph male	Deuto-nymph female	New adult Male (a)	New adult female (a)	Mother mite(a)
Prepupa	9	6 (67)	0	0	0	0	0	0	0	100 (1.5)
Pink-eye pupa	7	6 (86)	83	83	83	0	33	0	0	100 (2.3)
Red brown-eye pupa	10	7 (70)	29	57	71	43	57	0	0	100 (2.0)
Dark brown-eye pupa (dbe)	16	12 (75)	67	33	58	67	67	8 (1.0)	8 (1.0)	100 (1.6)
Dbe, lightly-pigmented thorax puta	10	7 (70)	57	71	100	43	43	14 (1.0)	4 (3.0)	100 (2.4)
Dbe, medium-colored thorax pupa	67	27 (40)	30	0	41	70	89	30 (1.0)	5 (1.0)	100 (1.3)
Dbe, dark thorax pupa	22	8 (36)	50	0	0	50	75	25 (1.0)	38 (1.3)	100 (1.5)
Unknown stage	9	9 (100)	33	33	33	11	11	11 (2.0)	11 (2.0)	100 (2.3)

Note: (a) Average no. of mites per infested cell.

Table 5. Number of fallen mites introduced with *Apis mellifera* drones into *Apis mellifera* observation hive

Day	No. of mites introduced with *A. mellifera* drones*	Undamaged	Damaged	Total
1	25	2	1	3
2	20	5	4	9
3	4	6	7	13
4	20	5	4	9
5	25	9	2	11
6	40	11	0	11
7	30	6	4	10
8	25	8	2	10
9		5	4	9
10		6	2	8
11		12	1	13
12		6	3	9
13		5	0	5
Total	189	86	34	120 (63.5 %)

Note: *One *Varroa* mite per newly-emerged drone.

experiment there was little difference (Fig. 2). This may be because the ability of *A. mellifera* to recognise foreign material was stimulated as many mites were introduced into the colony at one time. Tewarson *et al.* (1992) reported that 25 % of mites were recovered when mites collected from an *A. cerana japonica* colony were introduced into worker cells but there were no new female adults; 3 % of the grafted mites laid eggs. Yoshida *et al.* (1995) reported parasitism and reproduction of *Varroa* mite in *A. cerana japonica*. In a survey of absconding *A. cerana japonica* colonies, a 1.9 % parasitisation rate was counted in worker cells. In drone cells of a worker-laying colony, it was 38.4 %, 97 % of

Chemical control of *Vespa* spp. attacking *Apis* spp. in Pakistan

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The hornet (*Vespa* spp.) attacks *Apis* spp. during July–October in Pakistan. *Vespa velutina* caused heavy mortality of *A. mellifera* and *A. cerana* followed by *V. tropica* and *V. basalis*. *Apis cerana* copes with hornets better than *A. mellifera*. Earlier methods of hornet control are either expensive or have practical limitations. Poisoned baits using strychnine hydrochloride and zinc phosphide mixed with honey (1:20) were tested against hornets. Some 30–50 *velutina* adults, captured from *A. cerana* and *A. mellifera* apiaries, were fed on sugar solution, honey and minced meat. Before sunset, one drop of bait was poured on to the thorax of each hornet. Some 200–300 adults treated with poisoned baits were released at 9 sites in Islamabad/Rawalpindi. Hornet mortality was noted by daily counts of adults dead on ground under nests. After 3–4 days, nests were opened and remaining adult hornets/brood counted. Overall hornet mortality was 76.2–86.7% in treatment with strychnine hydrochloric, 71.6–80.3% with zinc phosphide and 0.9–2.2% in control. This technique seems to be useful provided the operation is extended over a long period. It can be practiced by beekeepers on a large scale.

which had offspring. In this experiment, the parasitisation rate of introduced mites in worker cells of *A. cerana japonica* was 3.9 % and offspring were found at a rate of 0.8 %. Eggs and nymphs were observed. On the other hand, new female adults were not seen, possibly because the timing was too early. It has been reported that the reproduction rate in worker cells is higher than in drone cells (Martin, 1995): drone cells of *A. cerana japonica* had 26 % parasitisation and 80.5 % had offspring. In this *A. cerana japonica* colony, the reproduction rate in drone cells was also higher than in worker cells.

Biological control of wax moth in Yunnan

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Wax moth (*Galleria mellonella* and *Achroia grisella*) cause serious damage to *Apis cerana* every year in Yunnan. Traditional insecticide treatment causes pollution of bee products and is not economical. After observing the life cycle of wax moth in apiaries in Mongzi county and Yaon county from 1995–96, two key periods (March and October) were found for treatment by cleaning the hive, exchanging old combs with new and melting old combs. Tests showed that wax moth harm could be reduced by 84% in this way.

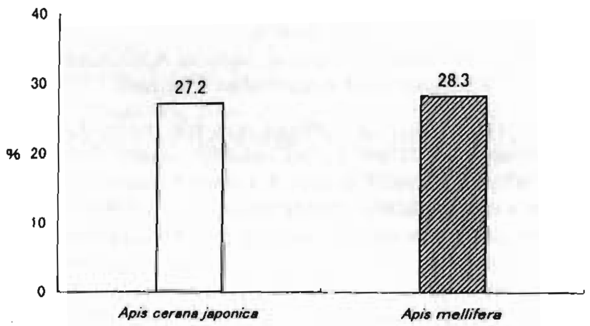

Fig. 1. Percentage of damaged mites in total mites fallen to bottom of *Apis cerana japonica* and *A. mellifera* colonies

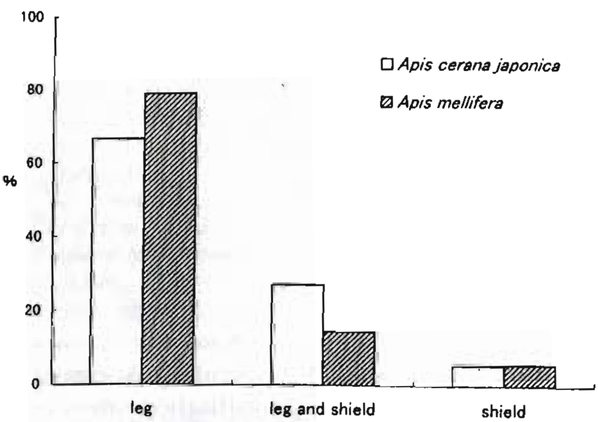

Fig. 2. Type of damaged to mites in *Apis cerana japonica* and *A. mellifera* colonies

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