

CRUCIAL STEPS LEADING TO SOCIAL PARASITISM IN THE CAPE HONEY BEE, *APIS MELLIFERA CAPENSIS*

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ABSTRACT

Social parasitism of African honeybee colonies, Apis mellifera scutellata, by the Cape honeybee, A. m. capensis, has caused huge losses of African bee colonies in northern South Africa. Once inside African colonies, Cape workers start producing female bees parthenogenetically, which has led to pseudo-clones of the social parasites spreading in the African population. Crucial steps leading to parasitism are invasion into new colonies, development of ovaries without being killed by African workers and production of eggs that are accepted. In mixed apiaries of African and Cape bees invasion into new colonies seems to readily occur and in contrast to other bee races eggs from Cape laying workers are accepted by nest-mates. The interaction between Cape bees starting to develop their ovaries and African workers is crucial, however, in determining if a successful take-over of the colony follows.

Keywords: *social parasitism, capensis, scutellata Apis mellifera, capensis, scutellata, larval food, guarding, worker ovary development, policing*

INTRODUCTION

Social parasitism of African honeybee colonies, *Apis mellifera scutellata*, by the Cape honeybee, *A. m. capensis*, has caused huge losses amounting to tens of thousands of African honeybee colonies (Allsopp 1993) in the central and northern parts of South Africa. These losses, the 'Capensis Problem', are due to the 1990 migration by beekeepers of some Cape honeybee colonies to beyond their natural range (Allsopp 1993), and the peculiar nature of these bees in the presence of African bees.

Once inside African colonies, Cape workers are able to develop their ovaries and to start reproducing. Unlike workers of other *Apis* races, which lay haploid eggs, workers of the Cape honey bee produce diploid eggs by thelytoky: parthenogenetic reproduction giving rise to females. Nursed by African workers, Cape larvae receive more food compared to Cape larvae nursed by their own workers (Calis *et al.* 2002). This results in the production of intermediates between workers and queens. In general, such worker-queen intermediates have enlarged spermathecae and higher numbers of ovarioles compared to workers. They are also heavier than workers, develop faster and have reduced pollen combs (Beekman *et al.* 2000, Calis *et al.* 2002). Hence, the resulting worker-queen intermediates are physically more adapted to reproductive behaviour. Also, they may be unable to perform normal worker tasks,

illustrated by the lack of foraging behaviour in parasitic Cape bees (Martin *et al.* 2002b). Usually the African queen eventually disappears, and this results in either the death of the colony due to the increase of laying Cape bees (and a decrease in foraging and brood care), or in supersedure of laying workers by a worker-produced Cape queen . Since the parasitic bees replicate thelytokously, invasion into new African colonies has led to pseudo-clones of social parasites spreading in the African population (Kryger 2001).

In the process of social parasitism, a minimum of three crucial steps can be identified. First, Cape bees have to invade African bee colonies. Secondly, they have to activate their ovaries; and thirdly, they have to lay eggs that are not removed by the host workers. During all three steps interaction with African workers will occur (Boot *et al.* 2002). African guard bees may prevent invasion at the hive entrance, African workers may harass and kill Cape bees that develop their ovaries, and African workers may remove the eggs eventually laid by Cape bees.

INVASION INTO AFRICAN BEE COLONIES

The first step in *capensis* parasitism is for Cape bees to enter African colonies. In theory, the hive entrance is guarded by guard-bees and only nest mates can pass unhindered (Breed *et al.* 1992). In practice, however, drifting of bees in apiaries is extensive and continuous (Boylan-Pett *et al.* 1991). Naïve foragers drift especially easily into neighbouring colonies without interference of guard-bees (Pfeiffer & Crailsheim 1998). Hence, African guard-bees are expected to play only a minor role in preventing potential parasites to enter the hive. Similarly, only foraging Cape workers would be expected to gain easy access to African honey bee colonies.

In contrast to workers, strange queens are recognised through their pheromonal bouquet and may be attacked to prevent usurpation of the colony (Pettis *et al.* 1998). This may also hold for developed Cape workers because they have a relatively queen-like pheromonal bouquet (Velthuis *et al.* 1990, Wossler 2002). If worker-queen intermediates try to invade, their bouquet is expected to be even more queen-like. Under commercial conditions (Allsopp 1993, Martin *et al.* 2002) and in experimental apiaries (Allsopp 1998, Neumann *et al.* 2001), however, large numbers of Cape bees have been quickly found in African colonies in mixed apiaries. Moreover, Beekman *et al.* (2002) recently showed that guard-bees behave similarly to parasitic Cape bees and African bees from another colony. Hence, in mixed apiaries invasion of African colonies by Cape workers seems to be relatively easy and predictable.

DEVELOPMENT OF OVARIES

Worker ovary activation is normally rare in queen-right honey bee colonies (), and this also holds for Cape bees (,). Workers that do develop their ovaries are usually recognised by their nest-mates (Sakagami 1954, Blom 1991) which may react by harassing the particular workers in order to stop the development, defined as one aspect of worker 'policing' by Ratnieks (1988).

Since Cape bees behave as parasites in African colonies, it is clear some do develop their ovaries. We recently found, however, that 'policing' behaviour by host bees may still play an important role. In four queen-right African colonies we found both successful take-over and failure to establish as parasites (Table 1). After introduction, just emerged (0-24h) Cape bees originating from the parasitic pseudo-clone in Pretoria were killed and removed in three of the four colonies. Colony 1 was most effective and removed all 'parasitic' bees within two days, whereas colony 2 took three days to remove them all. Colony 3 removed about 40% of the introduced bees during the first three days, after which no further bees were removed. Twelve of the remaining 'parasitic' bees were sampled after 12 days and two bees showed some ovary development, although no fully developed eggs were found. In colony 4, no 'parasitic' bees were removed and 8 out of 10 sampled bees showed fully developed ovaries after 9 days, and 6 out of 10 after 12 days. Apparently some feature of the colony plays a role in determining whether Cape bees are able to remain in an African colony, and secondly, in determining whether or not the Cape bees develop their ovaries in the colony, crucial for their establishment as parasites.

The extreme variety in response to Cape invader bees found in these 4 African honey bee colonies, all originating from the same apiary, argues for more research into what defines the outcome of interactions between African bees and their Cape invaders. We used African colonies from Kenhardt in the Northern Cape, a region not infested by Cape parasites. Possibly these bees, the "Kenhardt blend", react more aggressively to Cape bees that start developing their ovaries than do the African bees that currently are suffering from social parasitism, since effective removal of Cape parasites has not been reported before. Instead, whole apiaries are lost within several months after Cape parasites have been found (Allsopp 1993). Alternatively, the state of the colony may be critically important. For instance, in queenless African colonies from the same population, Cape bees readily develop their ovaries and start laying (unpublished results). Our results with queen-right colonies suggest that here the state of the colony is important as well. In colonies 1 and 2, the density of bees on the combs was high, whereas in colonies 3 and 4 the density was low. This suggests that relatively more bees were nursing the brood or working on related tasks in colonies 3 and 4, and fewer bees may have been present to do 'policing' tasks, which could allow Cape bees to develop their ovaries.

Table 1. Removal from the colony and ovary development following the introduction of 'parasitic' Cape bees from the pseudo-clone in Pretoria into queen-right colonies of the African honey bee. Just emerged (0-24h) Cape bees were marked with plastic tags glued to the top of their thorax and subsequently introduced into the colonies by letting them walk through a small hole in the top board of the hives.

Colony	Number of bees tested	Number of frames	Density of bees on combs	Removal of 'parasitic' bees	Fully developed ovaries after 12 days
1	20	3	high	All within 3 days	-
2	20	3	high	All within 2 days	-
3	150	8	low	Partly removed	0%
4	150	8	low	Not removed	60%

PRODUCTION OF EGGS THAT ARE ACCEPTED BY THE WORKER BEES

Even when workers do lay eggs in queenright colonies, and approximately 0.01% of the workers have activated ovaries in non-Cape honey bees (), these eggs are rapidly removed by other workers (), which is defined as another aspect of 'worker policing' (Ratnieks, 1988). Martin *et al.* (2002a) recently showed that parasitic Cape bees evade egg-policing, emphasising that this is different from normal Cape workers, and hence the key to the 'unique parasitism' of the parasitic Cape pseudo-clones. The larvae of these parasitic Cape pseudo-clones are necessarily nursed by African workers, which results in intermediates between the worker and queen castes (Beekman *et al.* 2000, Calis *et al.* 2002). Therefore, we wished to investigate whether the eggs of intermediates (Cape bees originally reared in African honey bee colonies, and equivalent to the pseudo-clones) were less well 'policed' than were normal Cape worker eggs. Using both Cape and African bees as discriminator colonies we found that eggs of African laying workers were quickly removed whereas eggs of Cape worker bees were mainly untouched, for eggs both from worker-queen intermediates and normal Cape workers (Fig. 1), as were queen-laid eggs. This indicates that evasion of egg-policing is a general trait in Cape bees and not restricted to the parasitic pseudo-clones of the current 'Capensis Problem'.

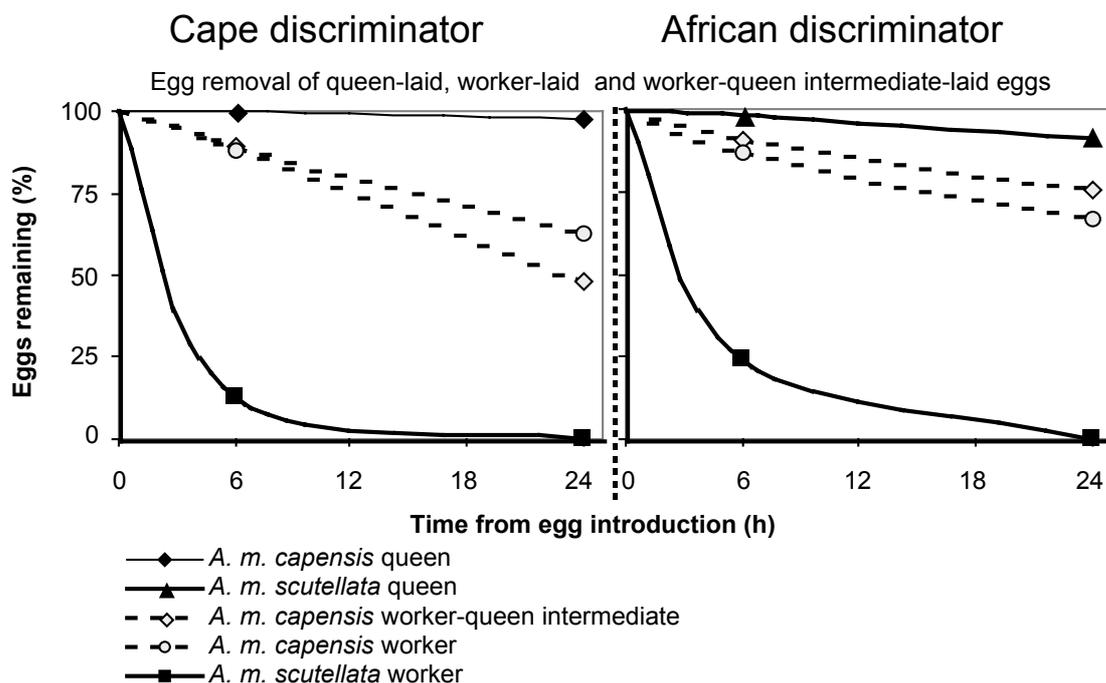


Figure 1. Removal rates of eggs of Cape and African bees. We transferred pieces of comb containing *A. m. capensis* queen-laid eggs, *A. m. scutellata* queen-laid eggs, eggs laid by *A. m. capensis* worker-queen intermediates, *A. m. capensis* worker-laid eggs and *A. m. scutellata* worker-laid eggs into a test comb. Test combs were placed into unrelated discriminator colonies of both races above a queen excluder, and sandwiched between two brood combs. Egg removal was quantified by inspecting cells after 6 and 24 hours. We considered only cells containing one egg (average 47 cells per egg source per discriminator; range 22-87).

CONCLUSION

Because invasion of African colonies by Cape bees seems to be easy, and because evasion of 'egg-policing' is common in Cape bees, the ability of Cape bees to escape both removal once in an African honey bee colony and reproductive inhibition in the colony, are critical and potentially limiting factors in the establishment of *capensis* parasitism.

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