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Original article

Colony aggregations in *Apis mellifera* L

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Summary — Natural aggregations of *A mellifera* L have not been reported. However, in the related species *A dorsata*, aggregations of colonies are common. A survey of the spatial distribution of feral *A mellifera* colonies showed that they too can be markedly clumped, with up to 10 colonies/ha. For these heavily clumped colonies, we inferred queen genotype from worker samples for: 1) malate dehydrogenase; 2) a mitochondrial DNA polymorphism; and 3) a microsatellite locus. The aggregation examined was composed of colonies headed by potentially related (*ie* parent/offspring or sister) queens, and unrelated colonies. Thus, it is likely that existing colonies attract swarms and that swarms may not always travel far from the natal nest in an environment that is replete with nesting sites.

***Apis mellifera* / *Apis dorsata* / swarming / nest site / Nasonov pheromone / genetic relationships / microsatellite / mitochondrial DNA**

INTRODUCTION

This study was prompted by a casual observation in a caravan park in Devonport, Tasmania, Australia of 12 colonies of feral honey bees *(Apis mellifera)* all within 200 m of each other (Oldroyd *et al,* 1995). This seemed an extraordinary density of honey bees. Densities of feral colonies are typically estimated as 0.5–5 colonies/km^2 (reviewed by Ratnieks *et al,* 1991; Oldroyd *et al,* 1994). Aggregations are completely unexpected on the basis of Lindauer's (1961) comment that honey-bee swarms prefer distant nest sites to nearby ones, and the observation of Hubell and Johnson (1977) that stingless bee nests have a uniform rather than a random or clumped spatial distribution. We therefore surveyed a population of feral honey bees to determine how often aggregations of colonies occur in the species.

The distance travelled by swarms to natural nest sites can be inferred by measuring the dance tempo of the consensus dances just prior to the departure of the swarm for the new nest. (The dance tempo encodes the distance of the potential nest site from

the swarm (von Frisch, 1967).) On this basis, most natural swarms move 500–1 500 m from the natal nest (Lindauer, 1951, 1955; Seeley and Morse, 1977), although scouts have been reported to dance for nest sites over 10 km from the natal nest (Villa, 1993). Lindauer (1955) suggested that swarms aware of 2 nest sites of equal value select the more distant 1 to move to. However, this conclusion was questioned by Seeley and Morse (1977) and Jaycox and Parise (1980, 1981). They showed that under controlled conditions where swarms were offered nest cavities of equal merit at different distances from the cluster, swarms had a preference for the nearest nest site.

If, as Jaycox and Parise (1980, 1981) and Seeley and Morse (1977) suggest, swarm movement is small when there is an abundance of suitable cavities near the clustered swarm, then aggregations of related colonies in cavity-rich environments would result. Aggregations caused by limited swarm dispersal would comprise a family group. On the other hand, if swarms are attracted to existing colonies distant from the natal nest, then aggregations would comprise unrelated colonies.

In *A mellifera,* no aggregations of any kind have been reported. This implies that natural swarms have a greater tendency to disperse than Jaycox and Parise (1980, 1981) suggest, that suitable nest sites are rare in most environments, or simply that systematic searches for honey-bee nests have not revealed the phenomenon.

In the related species *A dorsata,* the giant honey bee of Asia, aggregations of colonies are common (Seeley *et al,* 1982; Seeley, 1985, p 150). Colonies tend to aggregate in groups of 2–60 under a single rock ledge or on a single tree (Koeniger and Koeniger, 1980). These aggregations are not due to limitations of available nest sites. Trees of identical form and species but with no *A dorsata* colonies are often found next to trees with large numbers of nests (personal observation, but see Seeley *et al,* 1982). It is not known if these aggregations are a family group or are completely unrelated.

In this study we report the spatial distribution of 28 feral *A mellifera* colonies found in a survey of seven 500 x 100 m plots in a mixed *Eucalyptus camaldulensis* (red gum) and *E largiflorens* (black box) woodland. An extremely dense natural aggregation of 10 colonies was selected and genetic markers were used to infer genetic relationships among these colonies to see if they comprised a family group or a random aggregation.

MATERIALS AND METHODS

Study sites and colony location

This study was conducted in Wyperfeld National park in north-west Victoria, Australia. Beekeeping is prohibited in the park, but Oldroyd *et al* (1994) estimated that there are 77 feral colonies/km^2, in the 0.5–1 km wide band of *E camaldulensis* / *E largiflorens* woodland that borders the dry creek bed that passes through the park. This is an extremely high density of feral bee colonies (Otis, 1990; Ratnieks *et al,* 1991), and reflects the very favourable environment for honey bees. The distribution of mallee *(Eucalyptus* spp) and banksia *(Banksia ornata)* species provide an exceptionally rich and varied nectar and pollen resource (Oldroyd *et al,* 1994). Oldroyd *et al* (1994) estimated that there are up to 11 000 hollows/km^2 in the black box and red gum trees that line the creek, and that many of these are suitable cavities for honey-bee nests. Despite occupation by native fauna, it seems very unlikely that hollows are a limiting resource for bees.

Seven 500 x 100 m sites were pegged out with metal fence posts (see Oldroyd *et al,* 1994, for a full description). Sites were separated by at least 0.5 km and up to 6 km (fig 1). Sites were each divided into five 10 000 m^2 sectors, aligned north/south.

In March and April 1993, each tree in the plots was carefully examined for the presence of honey-bee colonies. Only 2 species of tree exist in the

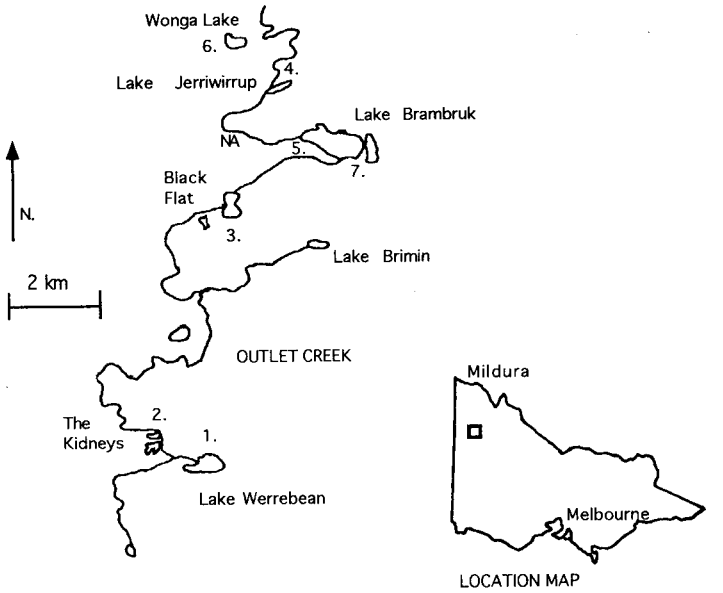

Fig 1. Map of Wyperfeld National Park in Victoria, Australia, and Outlet Creek which flows through the park (see Oldroyd *et al*, 1994, for details). Numbers are 50 000 m^2 sites, generally adjacent to a dry lake bed. NA is a natural aggregation of 9 colonies all within a 10 000 m^2 area.

plots examined (red gum and black box). Most trees were 10–20 m high, with the occasional 30 m individual. Trees are quite sparsely distributed. Thus bee colonies are relatively easy to locate. Trees were examined in fine weather when the temperature exceeded 18°C, and known colonies were foraging freely. Each tree was carefully examined from 4 compass points by pairs of observers. To ensure careful observation, a sketch was made of each of the 1 982 trees within the plots. The number of hollows (defined as a hole that might potentially lead to a dry cavity of > 4 l volume) was also recorded. When colonies were found, their location was recorded. All trees were carefully re-examined in September 1993 by a second pair of observers. The location of overwintered colonies was checked, and a few colonies missed in the first survey were located at that time. Sketch maps of the location of all colonies were then made.

Colony aggregations

Bees from a natural aggregation were obtained for detailed genetic analysis, to determine if the aggregation comprised of related colonies. Sample bees were caught either by aspiration of bees at the entrance, or by the use of ladders and long-handled insect nets. Bees were frozen in liquid nitrogen, and stored at –70°C until required.

Protein and DNA extraction

Frozen thoraces from 8–24 (usually 12) bees per colony were each ground in 100 µl distilled water in a 1.5 ml microcentrifuge tube. Ten microlitres of supernatant was removed and reserved for protein electrophoresis. The remaining material was used for extraction of total nucleic acid by the method outlined in Oldroyd *et al* (1995). DNA was resuspended in 50 ml of 1 x Tris-EDTA buffer.

Protein electrophoresis

The 10 µl crude extracts were electrophoresed for 20 min on cellulose-acetate gels (Helena Laboratories, Beaumont, TX, USA) at 200 V in a 0.08 M Tris-EDTA-maleic acid running buffer (pH 8.2). Malate dehydrogenase isozymes (Sylvester, 1976; Cornuet, 1979) were visualised by coating gels with 8 ml 1% molten agar to which 0.6 ml 0.1 M Tris-HCl (pH 8.0), 0.4 ml each of 0.04 M nicotinamide adenine dinucleotide and 1 M di-

sodium malate (pH 7.0), and 0.2 ml each of 0.145 M methylthiazoyle blue and 0.065 M phenazine methosulphate had been added. Three alleles were scored as S, M and F according to the amount of anodal migration. Where genotypes were ambiguous, they were re-run against known standards on the same gel.

Microsatellites

One primer of the microsatellite locus A107 (Estoup *et al*, 1994) was radioactively end-labelled. In a total reaction volume of 10 μl, the γ-phosphate from ^{33}P-dATP (Dupont) was transferred to the 5'-terminus primer-2, using T4 polynucleotide kinase (Promega). The reaction contained 70 mM Tris-HCl, 10 mM $MgCl_2$, 2 μM primer, 5 μl ^{33}P-dATP, and 4 units of polynucleotide kinase. The reaction was incubated for 30 min at 37°C and was stopped by heating to 90°C for 2 min.

Polymerase chain reactions (PCRs) were performed in 10 μl of a mixture containing 1 μl of undiluted sample DNA, 0.167 mM of each dNTP, 1 μg BSA, 0.4 μM unlabelled primer, 0.02 μM labelled primer, 1 x Promega reaction buffer and 0.4 units of Promega *Taq* polymerase. PCR temperature profiles for microsatellite A107 were given in Estoup *et al* (1994). PCR products were run on standard 6% polyacrylamide sequencing gels with M13 control DNA sequencing reactions run on the same gel as size standards. Microsatellite alleles were scored as fragment lengths in base pairs.

Mitochondrial DNA

The region between the NDII-COII genes of the honey-bee mitochondrial genome contains a *Hinc*II restriction site in some individuals but not in others. PCRs were performed in a Perkin-Elmer 480 thermocycler using the following primers designed from Crozier and Crozier (1993):

5'TCCCACAAATAAAACCCCAAGATT 3'
5' CCACAAATTTCTGAACATTGACC 3'

which flank the region between NDII and COII.

The reaction mixture (total volume 50 μl) contained 1 μl of undiluted sample DNA, 25 pmol of each primer, 10 nmol of each dNTP, 0.8 units of *Taq* polymerase (Promega) and 1.5 mM $MgCl_2$. A ramped temperature profile was performed with the following steps: 92°C 1 min, 45°C 1 min, ramp to 64°C over 2 min, hold 1.5 min (10 cycles); ramp to 92°C over 1 min, hold 0.5 min, 45°C 1 min, ramp to 64°C over 2 min, hold 3 min (5 cycles); ramp to 92°C over 1 min, hold 0.5 min, 45°C 1 min, ramp to 70°C over 3 min, hold 2 min (15 cycles).

Five microlitres of PCR product were digested with 5 units of *Hinc*II (Promega) in a total volume of 25 μl according to the manufacturer's directions. Digests were electrophoresed in 1% agarose gels for 2 h at 70 V, and stained with ethidium bromide. Two patterns were discernible, 1 with 2 bands (scored as type A), and 1 with a single band (type B).

Analysis of genetic data

Each queen genotype was inferred from her worker genotypes for MDH and A107 by the following rules: i) a homozygous worker infers that the queen carries that allele; ii) two workers homozygous for 2 different alleles means that the queen is heterozygous for those 2 alleles; iii) if an allele is present in all workers the queen is probably homozygous for that allele; and iv) if all workers carry one of 2 alleles, then the queen is likely to be heterozygous for those 2 alleles. In combination, these rules can almost always resolve cases which are ambiguous by a single rule. With the data set used, queen genotype could be unambiguously inferred for all queens.

From queen genotype, some assessment of the relatedness of colonies can be made. Offspring queens must share the same mt-DNA type and at least 1 allele at both the MDH and microsatellite loci as the parent queen. Supersister queens will also share mt-DNA type and at least 1 allele at each nuclear locus. Half-sister queens or grand-daughter queens may not share alleles at both loci, but will have the same mt-DNA type. Unrelated queens may or may not share nuclear loci and mt-DNA type.

RESULTS

A total of 27 colonies were located in the survey area, a mean (± se) of 0.77 ± 1.21

colonies per 10 000 m^2 plot. A mean of 111.6 ± 50.0 (range 29–242) hollows were present in each plot and 56.6 ± 19.2 (range 24–92) trees. There were non-significant correlations between the number of bees and the number of hollows ($r = 0.26$; $P > 0.1$) and the number of trees ($r = 0.30$; $P > 0.05$) per plot. Thus variance in numbers of trees or hollows per plot is insufficient to explain any variation in colony numbers.

The location of each colony within the 7 plots is given in figure 2. By inspection, there appears to be a non-random distribution of colonies, with a particularly strong aggregation in sector 4 of plot 7 (fig 2). An additional cluster of 9 colonies was located outside the plots by fortuitous discovery (fig 1).

The usual way to determine if a population of organisms is spread randomly or in aggregations is to compare the distribution of organisms per sampling unit to the Poisson distribution (Ludwig and Reynolds, 1988). The distribution of organisms is expected to follow the Poisson distribution if the organisms are scattered randomly in the environment, and the negative binomial distribution if the organisms are aggregated. The observed distribution of colonies and the expected distributions under the 2 theoretical models are given in table I. They show that the observed distribution differs significantly from the Poisson distribution ($\chi_5^2 = 187.7$, $P < 0.001$) but not from the negative binomial distribution ($\chi_4^2 = 5.83$, $P > 0.25$). These results indicate that the colonies were strongly aggregated in the environment, but should be treated with some caution. Because the expected values in the tails of the distributions are very low, inflated values of χ^2 are expected (Sokal and Rohlf, 1981). If classes with small expected value are pooled so the minimum expected value is around 3, there is no significant difference between the observed frequency distribution and those expected under the Poisson ($\chi_1^2 = 1.0$, $P > 0.5$) or negative binomial distributions ($\chi_1^2 = 0.21$, $P > 0.75$).

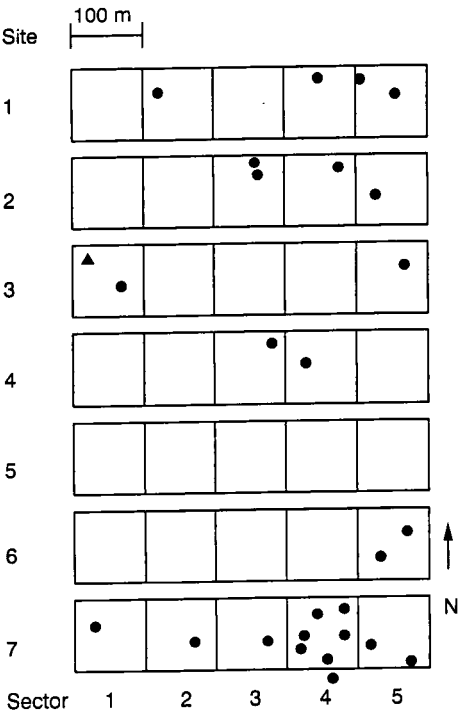

Fig 2. Schematic location of 28 colonies within the 7 sites of figure 1. Filled circles indicate 1 colony, the triangle 2 colonies in 1 tree.

Ludwig and Reynolds (1988) recommend that these distribution tests should not be used if the number of sampling units is < 40, as in this case. For smaller number of sampling units, they recommend the index of dispersion, ID, which is equal to s^2/x where s^2 and x are the variance and mean of colony number between the 35 100 x 100 m plots. From the data in table I, ID has the value 1.9, which is larger than unity, and again suggests that the colonies are strongly clumped. Ludwig and Reynolds (1988) suggest that ID values computed from < 30 sampling units are Poisson distributed and, if this is so, the ID from these data is significantly greater than unity, again suggesting a strongly aggregated distribution of colonies

Table I. Observed number of feral bee colonies per plot, and the expected number under the Poisson and negative binomial distributions.

Number of colonies per plot	*Observed frequency*	*Expected frequency*	
		Poisson	*Negative binomial*
0	19	16.182	19.00
1	10	12.483	9.347
2	4	4.815	3.992
3	1	1.238	1.619
4	0	0.238	0.639
5	0	0.0368	0.248
6	1	0.00531	0.153

in the field ($\chi^2_{34} = 65.0$, $P < 0.01$). If ID values are computed from more than 30 sampling units, Ludwig and Reynolds suggest that ID can be tested for deviation from unity with the d statistic:

$$d = \sqrt{2(\mathrm{ID})(N-1)} - \sqrt{(2(N-1)-1)}$$

which tends to the normal distribution. Again, this statistic strongly suggests a clumped distribution ($d = 3.21$, $P < 0.01$).

Although all tests suggest that the colonies observed were aggregated, we recommend cautious interpretation. The validity of tests of the significance of ID values rests on assumptions about their statistical distributions, which may not be valid in this instance. Further, the conclusion of aggregation rests solely on the very dense aggregation of colonies in sector 4 of site 7 (fig 2). If this sector is excluded from the analysis then ID = 1.08, which is not significantly different from unity ($\chi^2_{33} = 35.7$, $P > 0.05$).

The genetic relationships among parental queens in the aggregation at site 7 is given in table II. The presence of 2 mitochondrial types demonstrates that there were at least 2 lineages present. Only queens heading colonies 60 and 61, 56 and 62, and 61 and 62 may be highly related as parent-offspring or super-sister queens. All other colonies are more distantly related.

DISCUSSION

This study is the first to report that a wild population of *A mellifera* colonies can form dense natural aggregations. Taber (1979) mapped the location of 21 colonies along the Verde river in Arizona. Inspection of his figure 1 also suggests a non-random distribution of colonies. Further, we have found an additional aggregation of 9 colonies at Wyperfeld (fig 1), and an aggregation of 12 colonies at Devonport in Tasmania (Oldroyd *et al,* 1995). We have learned that similar dense aggregations of honey-bee colonies occur in Louisiana (TE Rinderer personal communication). These observations suggest that wild colonies of *A mellifera* may often be found in aggregations. This phenomenon is common in *A dorsata* (Koeniger and Koeniger, 1980; Seeley *et al,* 1982), and may occur in other Apis species where data are lacking, although clumping in *A cerana* and *A florea* was not observed by Seeley *et al* (1982). If *Apis* colonies often form aggregations, then this is the complete reverse of the situation in stingless bees, which have a uniform distribution of colonies, presumably to reduce competition (Hubell and Johnson, 1977). Note also that Wenner's (1989) map of the spatial distribution of feral honey-bee colonies on Santa Cruz

Table II. Genetic relationships among queens heading aggregated honey-bee colonies.

Colony	*56*	*57*	*58*	*59*	*60*	*61*	*62*	*63*
56		Δ	❑	❑	❑	Δ	♥	❑
57			❑	❑	Δ	Δ	Δ	❑
58				Δ	❑	❑	❑	Δ
59					❑	❑	❑	Δ
60						♥	Δ	❑
61							♥	❑
62								❑

Δ Queens of these colonies are not highly related (parent/offspring or super-sisters). They may be half-sisters or unrelated. ❑ Queens of these colonies are unrelated as they do not share the same mitotype. ♥ Queens of these colonies may be as closely related as parent offspring, but may also be unrelated colonies that share alleles and mitotypes by chance alone.

Island, California, shows no hint of aggregation. This may be because floral or nest site resources are limited there (Wenner, 1992).

Why should honey-bee nests sometimes form aggregations? Several hypotheses are plausible.

Aggregation is a result of preference for swarms to travel a short distance from the natal nest

Jaycox and Parise (1980, 1981) suggested that swarms have a preference for the nearest of 2 otherwise equal nest sites, while Lindauer (1955) suggested that swarms would take the more distant site. Our study shows that the aggregated colonies at Wyperfeld comprised some completely unrelated ones (fig 3). Therefore, while short dispersal distances by swarms may explain some aggregations, this factor is unlikely as an explanation of the aggregation reported here.

Short dispersal distance cannot explain aggregations of *A dorsata* colonies. *A dorsata* regularly undertake long-distance migration which may exceed 200 km (Koeniger and Koeniger, 1980; Dyer and Seeley, 1994). When a migrating swarm arrives at a new location, it has the choice to join an existing aggregation, or to settle distantly from existing colonies (provided that there are sufficient nest sites available, see below).

Aggregation is a consequence of an uneven environment

Honey bees often make foraging trips exceeding 2 km (Knaffl, 1953; Beutler, 1954; Wenner, 1992) and over 6 km when forage is scarce (Visscher and Seeley, 1982). Therefore all colonies in this study had flying ranges that potentially overlapped those of colonies at other sites. There was no nest site, floral or water resource at or near site 7 that was not within flying range of sites 2 and 5 (fig 1). Site 5 in particular is less than 300 m from site 7 (fig 1), and the aggregation of 9 colonies found outside the plots (fig 1) is less than 2 km from site 5. No colonies were found at site 5. Twelve colonies were found at site 7. Sites were specifically selected for their similarity. The number of hollows suitable for nesting bees was similar and universally high among the

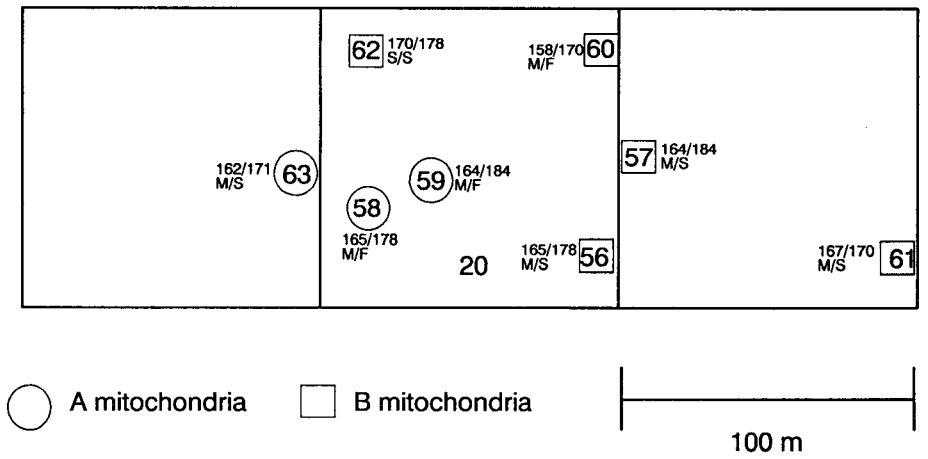

Fig 3. Relationships among a natural aggregation of 8 *A mellifera* colonies at site 7. Colonies represented by circles have a different mitochondrial lineage (A mitochondria) to those represented by squares (B mitochondria). Numbers within symbols are colony identification numbers. Numbers adjacent to symbols are the queen's genotype at the A107 microsatellite locus of Estoup *et al* (1994), in base pairs. Letters adjacent to symbols are the queen's genotype at the malate dehydrogenase locus.

plots (Oldroyd *et al*, 1994). We found no significant correlation between the number of colonies per plot, and the number of trees or hollows. (Note however, that some hollows that were too small to be favoured by bee colonies were included in these data.) It is therefore extremely unlikely that some unique resource was available at site 7 that was not available to colonies at other sites.

Seeley *et al* (1982) suggested that aggregations of many *A dorsata* colonies on particular trees is a consequence of a shortage of suitable nesting sites. In support of this claim, they reported that at 4 urban sites, the average number of colonies per aggregation was 10.0, while in undisturbed forest there were 1.7 colonies ($n = 11$). However, their conclusion presupposes that there is a shortage of sites in cities. Our own observations (unpublished) are that *A dorsata* successfully nests on many human structures such as tall buildings and water towers. Further, we have seen aggregations in undisturbed forest which is replete with trees suitable for nesting.

Aggregation provides increased nest defence

Although colonies respond to the alarm pheromones of other colonies (*eg*, Stort and Gonçalves, 1991), disturbance of 1 hive usually causes other colonies to become alerted rather than to attack. Seeley *et al* (1982) disturbed an *A dorsata* nest in order to determine the response of nearby colonies. They observed that nearby colonies did not attack the intruder, although they did become alerted. *A mellifera* and *A dorsata* colonies have very few enemies, and single colonies are well equipped to defend themselves without assistance.

Seeley *et al* (1982) suggested that aggregation may reduce the probability of being attacked by reducing the probability of becoming the predator's victim (Brock and Riffenburgh, 1960). This seems a very difficult hypothesis to test. Furthermore, the major predators of honey bees are large mammals including humans and bears

(Caron, 1990). Aggregation would appear to increase the chances of detection and the likelihood of predation, so we do not favour this hypothesis.

Aggregation improves mating efficiency

Honey bees mate on the wing, a potentially hazardous activity (Moritz, 1985). Aggregation could reduce the time required for virgin queens to locate a drone congregation area (Ruttner and Ruttner, 1972; Koeniger *et al,* 1994), mate and return to the nest. Because the fitness of a colony is severely reduced if its queen does not return from a mating flight, there may be strong selective reasons for aggregating. More importantly, aggregations may reduce the probability of queens mating with brothers, provided that aggregations are not closely related groups. Sex in honey bees is determined by heterozygosity at the sex locus (Crozier, 1975). Diploid individuals that are homozygous at the sex locus are male, but are eaten by workers at the first larval instar (Woyke, 1963). Mating with brothers reduces brood viability by causing an increase in homozygosity at the sex locus. Shaskolsky (1976), Page (1980), Page and Metcalf (1982), Crozier and Page (1985) and Ratnieks (1990) all argue that reduction of brood viability caused by the presence of diploid drones could lead to the evolution of polyandry. Other mechanisms, such as the aggregation of unrelated colonies, may help to avoid inbreeding and the genetic load imposed by diploid males. In *A dorsata* particularly, the duration of the mating flight is brief (< 10 min, Rinderer *et al,* 1993; Koeniger *et al,* 1994) and proximity to a drone congregation area containing a diverse population of drones may be very important.

Our genetic data reveal that the colonies at site 7 were not all related, and the aggregation was comprised of at least 2 maternal lineages, and quite possibly more. It is known that scout bees searching for nest sites are attracted by Nasonov pheromones (Free *et al,* 1981a, 1984; Kigatiira *et al,* 1986; Witherell and Lewis, 1986; Schmidt and Thoenes, 1987, 1992; Schmidt *et al,* 1989; Villa, 1993; Winston *et al,* 1993; Schmidt, 1994). It is usually assumed that scouts are attracted because Nasonov pheromones guide swarms and induce clustering (Avitabile *et al,* 1975; Free *et al,* 1981a, b). We suggest that in fact, scouts from natural swarms are attracted to existing colonies by the presence of Nasonov pheromones, and seek cavities in that area. In evolutionary terms, the presence of existing colonies might indicate that: i) the local environment can support bees; and ii) the swarm's future queens would have nearby unrelated mates. Nearby colonies might also provide some additional colony defence. This attraction of scout bees by Nasonov pheromones may therefore be adaptive, since it may promote colony aggregation. The large foraging range of *A dorsata* and *A mellifera* probably ensures that aggregation does not impair survival due to intercolonial competition. Although our present data support the conjecture that aggregated colonies can be unrelated, confirmation requires a more detailed genetic analysis of the relationships between new swarms found near existing colonies.

Finally, it should be noted that *A mellifera* colonies are found singly (fig 2) as well as in aggregations. This does not detract from our general argument of a tendency to aggregation. Founder colonies have a low survival rate, and established colonies have a low reproductive rate (Seeley, 1978). Therefore daughter colonies may not always be found near established colonies. In environments where nest sites are rare, aggregation may not be possible.

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Résumé — Rassemblements de colonies chez *Apis mellifera* L. On n'a pas mentionné jusqu'à présent de rassemblements naturels de colonies d'*Apis mellifera.* Pourtant ces rassemblements sont courants chez l'espèce voisine *Apis dorsata.* Nous avons étudié la répartition spatiale des colonies sauvages d'*A mellifera* dans le parc national de Wyperfeld dans l'État de Victoria en Australie (fig 1) pour déterminer si les colonies d'*A mellifera* pouvaient aussi former des rassemblements. Le parc possède d'excellentes ressources en nourriture et de nombreux sites de nidification (Oldroyd *et al,* 1994). L'étude a porté sur 7 sites de 500 x 100 m chacun. La présence de nids d'abeilles a été examinée attentivement pour chaque arbre (N = 1982) de ces parcelles. L'étude montre que, dans ce milieu favorable, les colonies d'*A mellifera* se sont nettement rassemblées avec une densité pouvant atteindre 10 colonies à l'ha (fig 2). L'indice de dispersion *ID* (Ludwig et Reynolds, 1988) atteint 1,91. C'est significativement supérieur à l'unité (χ^2_{34} = 65,0 ; P < 0,01), ce qui indique un écart significatif de la distribution de Poisson et une répartition spatiale en agrégats. Nous voudrions pourtant faire remarquer que ce résultat dépend du rassemblement particulièrement dense de colonies sur le site n° 7. Pour ce rassemblement remarquable nous avons déduit le génotype de la reine en étudiant, sur des échantillons d'ouvrières, le polymorphisme de la malate deshydrogénase et de l'ADN mitochondrial et un locus microsatellite. Le rassemblement étudié était composé de colonies dont les reines étaient potentiellement apparentées (*ie* parent/descendance ou sœur) et de colonies non apparentées (fig 3 ; tableau I). Une explication vraisemblable pourrait être que, d'une part, des colonies présentes attirent les essaims et que, d'autre part, dans un milieu riche en sites de nidification les essaims ne s'éloignent pas toujours très loin du nid parental. Nous pensons que des essaims peuvent évaluer des sites de nidification proches de colonies existantes prospères comme susceptibles d'accroître leur survie en assurant aux futures reines de l'essaim un accouplement plus rapide et en leur indiquant que le milieu convient bien aux abeilles.

***Apis mellifera* / essaimage / ADN mitochondrial / microsatellite / degré parenté**

Zusammenfassung — Völkeransammlungen bei *Apis mellifera* L. Über natürliche Ansammlungen von *A mellifera* Völkern liegen bisher keine Berichte vor. Bei der verwandten Art *A dorsata* gibt es viele Beobachtungen über Häufungen von Völkern an einem Standort. Um zu bestimmen, ob Völker von *A mellifera* ebenfalls in Häufungen vorkommen, untersuchten wir die räumliche Verteilung von wildlebenden *A mellifera* Völkern im Wyperfield Nationalpark, Victoria, Australien. Der Park bietet hervorragende Nahrungs- und Nistbedingungen für die Bienen (Oldroyd *et al,* 1994). Die Untersuchung umfasste 4 Standorte mit einer Fläche von jeweils 500 x 100 m, die in Bezirke von 100 x 100 m unterteilt wurden. Jeder einzelne Baum (N = 1982) innerhalb dieser Bezirke wurde sorgfältig auf die Besiedlung durch Bienen untersucht. Es zeigte sich eine deutlich aggregierte Ver-

teilung der Bienenvölker mit bis zu 10 Kolonien innerhalb von 10.000 m^2 (Abb 2). Der Dispersionsindex nach Ludwig and Reynolds (1988) betrug ID = 1.91. Dies ist signifikant größer als 1 ($\chi^2_{34} = 65,0$, $P < 0,01$). Dies deutet auf eine signifikante Abweichung von einer Poissonverteilung und eine aggregierte räumliche Verteilung hin. Wir möchten aber anmerken, daß dieses Ergebnis auf die besonders dichte Aggregation von Völkern am Standort 7 beruht. Für diese bemerkenswerte Völkeransammlung ermittelten wir den Genotypus der Königinnen aus der Untersuchung von Arbeiterinnenproben aus Polymorphismen von Malatdehydrogenase und mitochondrialer DNA, sowie einem Mikrosatelliten-Locus. Die untersuchte Ansammlung setzte sich aus Kolonien mit möglicherweise verwandten (*ie* Eltern/Nachkommen oder Schwestern) und nichtverwandten Königinnen zusammen (Abb 3 und Tabelle I). Als wahrscheinliche Erklärung schlagen wir vor, daß einerseits bestehende Völker für Schwärme attraktiv sein könnten, und daß andererseits Schwärme sich in einer Umgebung mit vielen Nistmöglichkeiten nicht unbedingt sehr weit von ihrer Mutterkolonie entfernen. Wir halten es für möglich, daß Schwärme Nistplätze nahe von erfolgreich bestehenden Völkern als förderlich für das Überleben einschätzen, da dies eine raschere Paarung der zukünftigen Königin gewährleistet und die Eignung des Standortes für Bienenvölker anzeigt.

***Apis mellifera* / Schwärmen / Nistbedingungen / Verwandtschaft / mitochondriale DNA / Mikrosatellite**

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