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## **Reproductive Conflict and Division of Labor in *Eutetramorium mocquerysi*, a Myrmicine Ant Without Morphologically Distinct Female Reproductives**

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### **Abstract**

The myrmicine ant *Eutetramorium mocquerysi* Emery from Madagascar exhibits a unique social organization. All female individuals are similar in size and appearance; female reproductives with a distinct external morphology do not exist. Based on ovarian anatomy, however, two major types of females can be distinguished: females with six ovarioles and a spermatheca, which can mate and produce diploid offspring, and females with only two ovarioles, which lack a spermatheca but can lay unfertilized eggs. Individuals with three to five ovarioles are rare. Anatomical differences are not correlated with different roles. Both types of females were observed foraging, tending brood, and laying eggs. However, only females with six ovarioles and a spermatheca were the reproductively and socially most dominant individuals. Nestmate antagonism, which for the first time is demonstrated for an ant species belonging neither to the Ponerinae nor the Formicolenini, consists of biting, antennation bouts, and ritualized dominance postures. In two colonies, removal of the dominant individual resulted in the destruction of all larvae and pupae.

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### **Introduction**

Colonies of social insects appear to be harmonious, smoothly functioning superorganisms. Most individuals surrender their own reproduction in order to raise sisters and brothers and their behavioral activities appear programmed to

increase the efficiency of the colony (Wilson 1971; Hölldobler & Wilson 1990; Seeley 1997). Only a minority of females per colony mate and lay fertilized eggs. Nevertheless, conflict among group members may exist about the allocation of resources and the partitioning of reproduction within the colony (Hamilton 1964, 1972; Ratnieks 1988; Seger 1991). For example, in most social Hymenoptera, unmated workers are capable of producing males from unfertilized eggs and because they are more closely related to their own sons than to those of other workers or the queens, they should prefer to rear males from their own eggs over males produced by their nestmates. Diverging interests concerning the origin of males may lead to open conflict among nestmates and result in the formation of social and reproductive hierarchies, in which only one or a few top-ranking individuals reproduce (Heinze et al. 1994a).

Dominance hierarchies have long been known to regulate reproduction in the societies of the more primitively eusocial Hymenoptera, such as paper wasps, sweat bees and bumble bees (Huber 1802; Pardi 1940; West-Eberhard 1967; Michener 1990). Here, queens and workers are morphologically very similar and probably equally efficient in the production of haploid eggs. In contrast, in many of the more advanced eusocial bees, and especially in ants, queens and workers usually show a high degree of morphological specialization and the ovaries of queens consist of many more ovarioles than those of workers. In those species, the queens are more efficient in laying haploid eggs and workers therefore refrain from reproducing (self-policing, Ratnieks 1988).

Overt conflict among nestmates in ants is known only from species with very small colony size and a strongly reduced caste dimorphism. In several genera of the subfamily Ponerinae, workers have a spermatheca; they can mate and produce diploid, female offspring (e.g. Peeters 1991a, 1993). In some species, fertile mated workers (gamergates), nonlaying mated workers, unmated workers and, if present, mated queens compete by overt aggression or ritualized dominance contests for their chances to lay eggs (Sommer & Hölldobler 1992; Sommer et al. 1994; Peeters & Hölldobler 1995). Reproductive conflict is similarly intense in several slave-making species of the myrmicine tribe Formicoxenini, where the ovaries of workers may have the same number of ovarioles as those of queens (but the workers do not mate; Franks & Scovell 1983; Bourke 1988; Heinze 1996a) and several nonparasitic Formicoxenini, where queen-worker dimorphism is also comparatively small (Cole 1981; Heinze et al. 1997).

Here, we describe the peculiar social organization and reproductive conflict in *Eutetramorium mocquerysi* Emery (1899) from Madagascar, a myrmicine ant, which also has very small colonies. Furthermore, it apparently lacks female reproductives with a distinctive external morphology, which renders the species unique among the morphologically more derived subfamilies of ants.

### Materials and Methods

In Nov. and Dec. of 1993, Nov. 1994, and Apr. 1996, a total of 14 colonies of *E. mocquerysi* were collected by one of us (G.A.) from their nests in decaying wood

in primary rainforest in the north-east of Madagascar (Marojejy National Reserve Integral 12, Anjanaharibe-Sud Reserve, Masoala National Park; for details see Alpert & Rabeson 1999). In the laboratory, ants were housed at 25°C in  $20 \times 20 \times 8.5$  cm<sup>3</sup> plastic boxes with a plaster floor. A  $5 \times 5 \times 1$  cm<sup>3</sup> cavity in the plaster, covered with a glass plate, served as nest. The ants were fed twice per week with diluted honey and pieces of cockroaches or other insects.

All individuals in four colonies, two collected in Nov. 1994 (94A with 38 individuals and 94B with 15 individuals—the latter colony had contained 22 individuals when collected) and two in Apr. 1996 (96A with 22 individuals, 96B with 52 individuals), were individually marked with Edding paint markers. Behavior was recorded either directly or from videotape (using a Panasonic AG-6730 time-lapse video recorder and a F15 HS camera) by ad libitum sampling. We attempted to take note of all cases of nestmate antagonism, egg laying, or egg eating. In addition, the behavioral status of each individual was noted every 2 min during observation sessions of 30 min each (scan sampling, Altmann 1974). For the analysis of dominance relationships, all aggressive interactions and for the investigation of the division of labor all recorded activities were evaluated. Behavior was sorted in the following categories, which for ants in general are described and defined in Wilson (1971) or Hölldobler & Wilson (1990) (see Appendix 1): 'foraging' (including all activities outside of the nest), 'nest guarding' (i.e. sitting in the nest entrance), 'cleaning the nest' (licking the plaster floor of the nest cavity, picking up and handling refuse particles and pieces of prey), feeding on solid food (pieces of prey, but also eggs and dead pupae), 'self-grooming', 'allogrooming' (active and passive), 'trophallaxis' (active and passive exchange of fluid food), 'mutual antennation', 'aggressive interactions' (attacks, i.e. rapid antennation bouts, biting, grasping an individual's head, pulling on head, legs, or antennae) and 'interactions with brood' (mostly grooming and carrying pupae, larvae or eggs; excludes the eating of eggs and dead pupae). Additional types of behavior were only rarely observed and therefore not included in the analysis (egg laying, defecation, etc.).

In two colonies (96A, 96B), the socially dominant individual died and aggression among nestmates increased. To investigate the effects of the removal of the dominant individual in greater detail, we gently displaced the glass plate covering the nest, removed the socially dominant individual from two colonies with forceps (in 96A we removed the female which had replaced the formerly dominant individual after its death) and subsequently kept them in isolation. After the nest was covered again, the ants remaining in the nest continued to be disturbed by the manipulation for  $\approx 15$  min, after which period the pattern of activity was normal again, with the exception of an increase in aggressive interactions. After  $\approx 8$  and 24 h, respectively, the dominant individual was gently placed near the entrance with forceps, from where it returned to the nest. The experiment was repeated in both colonies after four and 6 wks, respectively; at this time, and in a third colony (94B), however, the dominant individual was not returned. To control for the impact of the manipulation itself we similarly removed a randomly chosen individual from colony 96A; this did not result in an increase in aggressive interactions. Behavior was observed both before and after removal and before and after replacement for at least 30 min.

Individual size was measured with an eyepiece micrometer at  $\times 40$  magnification. For dissections, the ants were killed by freezing and their subgenital plates were pulled out of the gaster (the terminal major body part of ants) with a pair of forceps under a dissection microscope. Not all ants were available for dissection or measurement directly after the observation, as some were used in other experiments, others had lost their paint mark or died and had decayed before they could be dissected or measured. Statistical analyses were performed with STATISTICA 4.5.

## Results

### Field Data

Of the 14 colonies collected in Madagascar, 12 contained brood and, on average,  $44 \pm 23$  SD adult females (range 19–102), one colony contained  $\approx 50$  adult individuals (not counted directly after collection), and one colony consisted of a single adult individual, two eggs and 13 larvae. Surprisingly, queens with a distinct external morphology were not found. The dissection of all individuals in three colonies collected in 1993 and 1994 revealed that in each colony only a single female had strongly developed ovaries. The ant found solitarily in a small nest with eggs and larvae also had well-developed ovaries. Ovarian anatomy, however, was not studied in detail in these colonies.

### Ovarian Anatomy, Morphology and Reproduction

Dissection of a total of 76 individuals from four other colonies collected in 1994 and 1996 revealed enormous variation in the anatomy of the ovaries of females. In 39 individuals, the ovaries consisted of two ovarioles, one individual each had three and five ovarioles, respectively and 33 females had six ovarioles and a spermatheca. In two individuals with spermathecae, the number of ovarioles could not be determined. The bimodal distribution of ovariole numbers suggested the existence of two distinct types of females. Subsamples of females drawn at random from the dissected individuals did not reveal differences in external morphology or size correlated with the number of ovarioles. For the total sample, 14 individuals with six ovarioles, 12 individuals with two ovarioles (Mann–Whitney U-test): Weber's thorax length,  $U = 74$ ,  $p = 0.61$ ; thorax width,  $U = 78.5$ ,  $p = 0.78$ ; head width,  $U = 79.5$ ,  $p = 0.82$ . Intracolony comparisons were as follows: Colony 96A, four individuals with six ovarioles, five individuals with two or three ovarioles (Mann–Whitney U-test): thorax length,  $U = 7$ ,  $P = 0.46$ ; thorax width,  $U = 5$ ,  $p = 0.22$ ; head width,  $U = 6$ ,  $p = 0.33$ . Colony 94A, nine individuals with six ovarioles, six individuals with two ovarioles (Mann–Whitney U-test): thorax length,  $U = 19$ ,  $p = 0.35$ ; thorax width,  $U = 11$ ,  $p = 0.06$ ; head width,  $U = 15$ ,  $p = 0.18$ .

Members of both groups engaged in foraging (e.g. individuals RR with two ovarioles, GO with six ovarioles in colony 94A; Appendix 1, Fig. 1) as well as in nursing activities (K with two ovarioles, G with six ovarioles; see below).

In each colony, several females, both with two and six ovarioles, had elongated

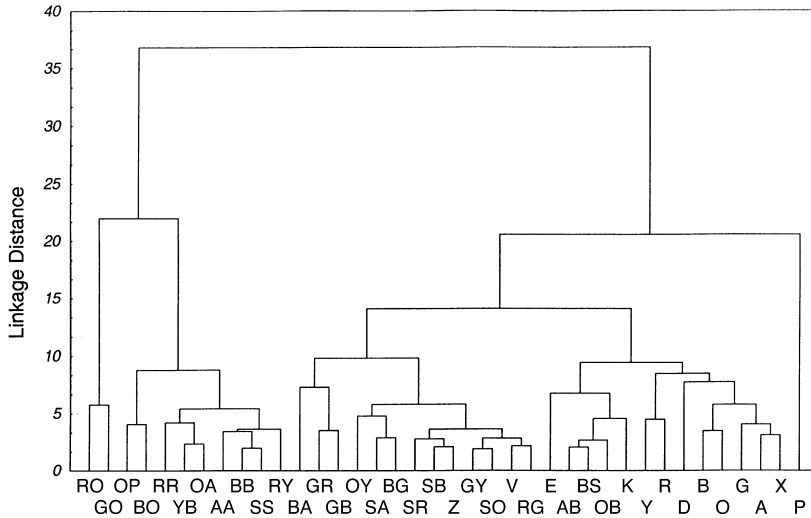


Fig. 1: Hierarchical cluster analysis of the frequency of different behaviors exhibited by individuals in colony 94A of the ant *Eutetramorium mocquerysi* (270 scans during 12 d). The letters indicate individuals shown in Appendix 1 and Table 2. Where available, the number of ovarioles is given in Appendix 1. On average, both types of females are indistinguishable in their behavioral repertoire. Individuals in the cluster on the left can be considered as foragers, whereas the remaining individuals mostly engaged in activities inside the nest. The individual on the far right, P, was the socially dominant egg-layer with six strongly elongated ovarioles and numerous eggs in development. The linkage distance gives an estimate of how similar the behavioral profiles of individuals are

ovarioles with maturing eggs and/or yellow bodies (the remnants of previously laid eggs) and/or were observed laying eggs during the observations (Table 1). The ovaries of most of the dissected egg layers contained only one, two or three maturing eggs ( $n = 15$ ,  $\bar{x} = 1$ ). However, as in the colonies dissected in the field, in each colony a single major egg layer was present, who had strongly elongated ovarioles, up to eight developing eggs, and numerous yellow bodies. Two of these individuals (one each in colonies 96A and 96B) were inseminated, but none of the 33 other dissected females with spermatheca had sperm in their spermathecae. This includes the two major egg layers in 94A and 94B, although both colonies had contained female brood when collected. Eggs laid in a group of nine uninseminated females, isolated from their nest directly after eclosion, developed into male pupae within  $\approx 7$  mo. Hence, there is no evidence for the production of females from unfertilized eggs by thelytokous parthenogenesis and we suggest that the mated individuals in colonies 94A and 94B died or were accidentally lost before we could start our observations.

The number of eggs or larvae present on the egg pile increased only slowly, suggesting that not all eggs produced developed into larvae. Indeed, egg eating by socially dominant individuals and larvae was occasionally observed (colony 94A, six cases during 16 h; colony 96A, one case during 21 h).

Table 1: Ovarian development and egg laying by females in colonies of the ant *Eutetrarmorium mocquersyi*. In each colony, several individuals laid eggs, although the dominant egg layer always had six ovarioles and a spermatheca. Those individuals that were observed laying eggs and were later dissected, are included both in the columns ‘Dissections’ and ‘Observation of egg laying’. A number of individuals that were observed laying eggs could not be dissected. They are listed in the column ‘Not dissected’. Numbers in brackets indicate individuals with a rare number of ovarioles (three and five, respectively)

Colony	Dissections				Observations of egg laying		
	Females with two (three) ovarioles		Females with six (five) ovarioles		Females with two (three) ovarioles	Females with six ovarioles	Not dissected
	dissected	ovary developed	dissected	ovary developed			
94A	8	4	16	2	1	3 <sup>1</sup>	3
94B	3						
	2						
	1	5	2				
96A	6 (1)	4 (1)	8	3	(1)		1
96B	9	1	5 (1)	2			

<sup>1</sup> All three eggs were laid by a single individual, P, during a total observation time of 16 h.

Division of Labor

The behavioral repertoire of females of *E. mocquersyi* consisted basically of the same types of behavioral acts as in other ants. In addition to common behaviors, such as foraging, brood care, self-grooming and allogrooming, trophallaxis, mutual antennation and resting, we occasionally observed the handling of refuse particles and pieces of prey, and the eating of eggs and of pupae. Dominance interactions were especially frequent and conspicuous (see below). Observations in three colonies suggested a simple division of labor typical of other ant species between foragers and individuals engaging in activities inside the nest. Cluster analysis of behavioral data collected in colony 94 A by scan sampling (270 scans during 12 d; the observation period ended when several young workers and males eclosed; see Appendix 1) revealed the presence of two major clusters (Fig. 1). One consisted of 11 individuals that were significantly more frequently observed in the arena than the 27 individuals in the other major cluster and are therefore referred to as ‘foragers’ (observed in the arena: foragers, median 76 of a total of 270 scans, range 58–185; nonforagers, median 1, range 0–33; Mann–Whitney U-test,  $U = 0$ ,  $p < 0.001$ ). Foragers engaged significantly less frequently in brood care than other individuals (grooming pupae: foragers, median 2, range 0–14; nonforagers, median 25, range 3–60;  $U = 5$ ,  $p < 0.001$ ). The cluster of nonforagers also contained the major egg-layer, P, somewhat set apart from the other nonforagers. P differed from all other nestmates in its higher frequencies of exhibiting aggressive behavior (see below, Table 2) and receiv-

Table 2: Number of aggressive interactions (antennation bouts, biting, pulling, etc.) among pairs of nestmates of *Eutetramorium mocquerysi* in colony 94A during a total observation time of 8 h 40 min (after this period, new workers and males eclosed and colony composition changed considerably). Individuals are ordered according to the total number of aggressive interactions in which they were dominant over another individual. P is clearly dominant over all other individuals, whereas rank relations among the others are often not resolved. Only females that were involved in 15 or more aggressive interactions are listed individually

Attacks	P	B	O	R	Is attacked								26		Total
					Y	A	G	D	X	E	Z	K	Others		
P	–	1	2	3	6	6	4	8	2	3	1	3	37	76	
B		6	9	7						3	2	2	11	40	
O		11	–	1		2		2	1				9	26	
R				–	19				1	2			4	26	
Y			5		–			3	2	10	1	1	4	26	
A	2		3		1	–	5		2	3		1	8	25	
G			5	3	2	2	–		1			1	4	18	
D					2			–	1	4	1		10	18	
X		2	2	2	2	1		1	–		2		3	15	
E				1				6	1	–		1	4	13	
Z				4			2	4			–		1	11	
K		1		1	1			2				–	1	6	
26 Others		1	1	3	2	2	1	2	1	8	21				
Total	2	16	24	27	42	13	12	26	11	27	8	9	104	321	

ing grooming from its nestmates (P, 58 observations; other individuals, median 4, range 0–16). The bipartition of the remaining cluster appears uncorrelated with conspicuous differences in a single behavior, although individuals in the right half of this cluster tended to be more active than those in the left part. A similar, simple division of labor was observed in the other two studied colonies (94B, 96A), where, however, only one individual each was the major forager and formed a cluster separate from the other individuals, including the major egg-layers.

Only in colony 94A were males present during a part (8 h) of the observation period. During most of this time, the four males were inactive or moved about the nest. No mating attempts were observed. Only once in 8 h did a male attempt to leave the nest. In two instances we observed a male opening its mandibles, extruding its labium and offering food to a female and another male. During at least the male–male interaction, food was transferred over  $\approx$  30 s, during which the recipient showed stridulatory movements with its gaster. Once a male was observed assisting the eclosion of a young female by biting the pupal cuticle.

Dominance Behavior

In all four colonies studied in the laboratory, aggressive interactions among individuals occurred at least during a part of the observation time. In colony 94A,

aggressive interactions were noted during the first observation sessions (Table 2), in the other three colonies fighting started only after the major reproductive individual died or had been removed. In the following text, individuals initiating aggressive interactions are referred to as attackers and all cases of aggressive interactions initiated by an individual, regardless of their strength, are referred to as 'attacks' (i.e. antennation bouts, biting, grasping an individual's head and pulling on head, antennae or legs).

The mildest type of recognizable attack consisted of short bouts of rapid antennations towards the head and thorax of another ant. Antennal movements during these aggressive antennations were much faster than during mutual, inspective antennations (Fig. 2a). During direct observation it appeared that the duration of antennation bouts differed between individuals. Therefore, a total of 43 videotaped antennation bouts of six individuals, which were sufficiently in focus for a detailed analysis in slow motion, were examined more closely. Attacks in this sample were directed towards an average of 3.8 ( $\pm 1.7$  SD) recipients. The duration of antennation bouts, which began when the ant turned its head towards the opponent, ranged from 0.3 to 1.8 s (median 0.8), during which from 6 to 34 beats were made (median 14). Individuals differed significantly in the duration of antennation bouts (Kruskal–Wallis ANOVA,  $df = 5$ ,  $\chi^2 = 24.62$ ,  $p < 0.001$ ) as well as in the number of beats they made during an individual bout ( $\chi^2 = 20.79$ ,  $p < 0.001$ ). However, the sample size of the data was not sufficiently large to control for the recipient of the antennations. Therefore, the relevance of these differences is not yet understood.

A more overtly aggressive interaction consisted of the attacker grasping another individual's head with its mandibles and pulling it along through the nest cavity for several centimeters while pointing the tip of its gaster towards the other ant and occasionally touching it with the gaster tip (Fig. 2b). Females responded to the attacks by crouching, turning away from the attacker, slowly retreating, or attempted to antennate or bite the attacker.

In each colony, a single individual appeared to be despotically dominant over all other nestmates. Rank relations among the other aggressive individuals were

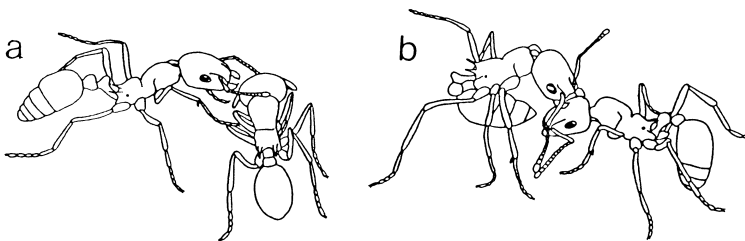


Fig. 2: Nestmate antagonism observed in colonies of the ant *Eutetramorium mocquersyi*. a. The attacker violently antennates head and thorax of a submissively crouching individual. b. The attacker attempts to pull the opponent backwards, with the gaster pointing towards the opponent. Both drawings are based on photographs



not always clear, making it difficult to construct linear dominance hierarchies from the data (Table 2). Among several aggressive individuals, the direction of attacks changed from day to day and only after the removal of the top-ranking individual did the individual who would in the end replace the former dominant become evident.

According to observations and/or dissections, the top-ranking female was also the major egg layer—its ovaries always consisted of a spermatheca and six ovarioles, which were elongated and contained numerous developing eggs (see above). Death or removal of this individual resulted in a dramatic and almost immediate increase of aggressive interactions. For example, in colony 96A, with 22 individuals, not a single aggressive interaction was observed during 5.5 h of observation in the presence of the inseminated female OR. Shortly after OR died and was removed by us from the nest, female RG attacked its nestmates 118 times during 2.25 h (52.4 attacks/h), and two other individuals (GO and BB) launched one attack each. During and between attacks, RG walked on stilted, i.e. fully extended, legs and had a strongly ‘inflated’ gaster. This posture was also exhibited by the individual with the highest frequency of aggression each in 94B and 96B and the two most aggressive individuals in colony 94A after removal of the major egg layer.

RG continued to be highly aggressive over the next 2 d (26 and 17 attacks/h), but the frequency of attacks decreased to 3.5 and 6 attacks/h on day 3 and 4, respectively. On days 5–12, after the death of OR, only 0 or 1 attack/h was observed (total observation time since removal of OR, 590 min). When RG was removed 12 d after the death of OR, aggression rate strongly increased again, and involved seven or more of a total of 20 colony members and reached a maximum of more than 180 antennation bouts in 30 min after  $\approx 7$  h (Fig. 3; total observation time since removal of RG, 370 min; only the females with the highest frequency of aggression are shown). When RG was returned to the colony after 24 h, the frequency of attacks had again decreased and RG resumed its dominant status after violently attacking both previously aggressive and passive individuals for approximately 1 h (Fig. 3). During this time, RG again exhibited the stilting posture.

A similar increase in aggression was observed when RG was removed from the nest a second time 4 wks later (from 0 to 54 attacks/h within 3 h after removal) and also when we removed the major egg layers from colonies 94A (increase from 16 to 325 attacks/h within 3 h after removal; this experiment was repeated 6 wks later and within 45 min after the removal a young worker, which had eclosed in the meantime, exhibited a stilting posture and almost continuously attacked its nestmates; the frequency of attacks was not determined) and 94B (frequency of attacks not determined). In contrast, removal of a randomly chosen individual from 96A 3 wks after we first removed RG did not result in an increase in the frequency of attacks (no attacks were observed in 30 min intervals both before and 2 h after the removal).

RG could not be dissected and the condition of its ovaries therefore is unknown. However, among those individuals from 96A and other colonies that exhibited high frequencies of aggression and could be dissected, most had six

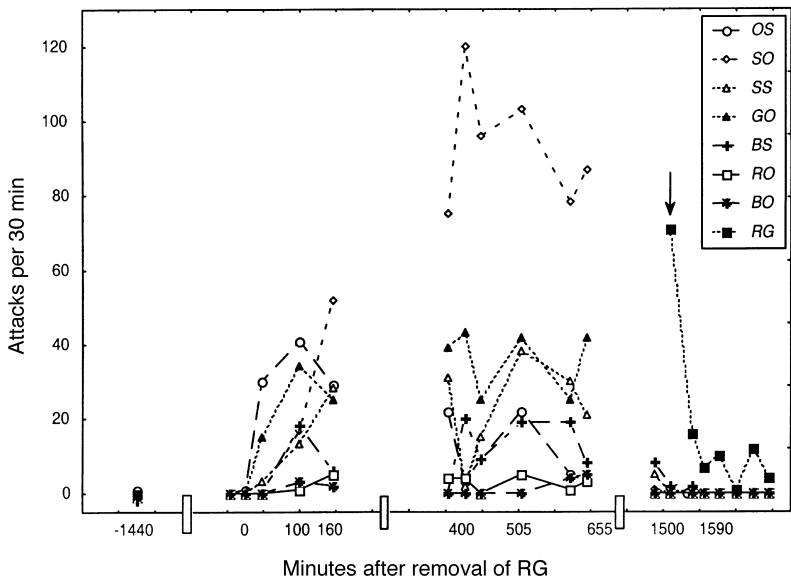


Fig. 3: Frequency of attacks exhibited during observation periods of 30 min by individuals of *Eutetramorium mocquerysi* before and after removal and return of the top-ranking female RG from colony 96A. RG was removed at time 0 and returned after 24 h (arrow). Only females that exhibited a high frequency of aggression are included

ovarioles (G, D, E, Z in 94A, Table 2; GO, SS, OS in 96A, Fig. 3). Nevertheless, several females with two ovarioles also actively engaged in dominance interactions, although at a lesser frequency (K in 94A, Table 2; SO, RO, BO in 96A, Fig. 3)

In both colonies, where the inseminated female died (96A, 96B), all brood was subsequently destroyed and over the course of 4 wks replaced by eggs laid by the new dominant.

Discussion

Colonies of the endemic Malagasy ant *E. mocquerysi* are characterized by a remarkable social organization and unique ways of reproduction, which differ from those of all other ant species (Bourke & Franks 1995; Heinze & Tsuji 1995). Phylogenetically, *Eutetramorium* belongs to the Myrmicini, a tribe which, among others, contains *Pogonomyrmex* harvester ants and *Myrmica* red ants, both of which have a pronounced caste dimorphism and an elaborate division of labor. Yet, in the structure of its colonies *E. mocquerysi* is much more similar to bumble bees, paper wasps and several queenless ponerine ants. There is no caste of female reproductives with specialized external morphology in *E. mocquerysi* (see also Alpert & Rabeson 1999) and the division of labor is rather simple, as is typical for colonies with small worker numbers (Hölldobler & Wilson 1990). Although our conclusions are based on only a small number of colonies, the composition of

additional societies collected in the field (Alpert & Rabeson 1999) was similar to that of our observation colonies, suggesting that the majority of colonies in Madagascar will show the same social organization.

*E. mocquerysi* females can be separated into two distinct categories based on the anatomy of their ovaries, which either consist of two ovarioles or of six ovarioles and a spermatheca. Only two out of a total of 76 dissected females had three and five ovarioles, respectively. Hence, ovariole numbers show a bimodal distribution. Such a pronounced dimorphism in ovary structure without additional morphological differences has previously not been reported from ants. The two different types of females also did not differ in their behavioral repertoires: females with six and two ovarioles likewise took part in foraging, brood care, etc., although reproductively and socially dominant females in the societies always had six ovarioles. Both types of females were seen laying eggs and the ovaries of several individuals belonging to both categories were found to be developed. In several cases we observed that eggs were eaten by a high-ranking individual or fed to larvae, but because of the very low number of observed ovipositions and egg eating it is not yet clear whether all eggs laid by females with two ovarioles are eaten. Of 35 dissected individuals with a spermatheca, in only two was sperm found. Other socially or reproductively dominant individuals were uninseminated. Since there is no evidence for thelytokous parthenogenesis in *E. mocquerysi*, the presence of female brood in the two colonies without inseminated female suggests they had died or were accidentally lost in the weeks between collection and the start of detailed observations in the laboratory. No mating attempts were observed in one colony containing males.

Too little is known about the ontogeny of the two female classes to decide whether they are all to be recognized as workers, as in some ponerine ants without morphologically distinct queens (Peeters 1991a), or whether, instead, individuals with six ovarioles and a spermatheca are morphologically extremely derived worker-like queens (e.g. 'ergatoid queens' sensu Peeters 1991b). Large variation in the anatomy of ovaries, including the number of ovarioles, has been reported from several ant species, including social parasites (Heinze 1996b) and bulldog ants (Ito et al. 1994), although the bimodal distribution of ovariole numbers in *E. mocquerysi* is unique. If all individuals were considered to be workers, the inseminated egg laying females found in two colonies would then be the first gamergates outside the subfamily Ponerinae. Alternatively, evolution might have favored the loss of all morphological queen characters other than ovarian anatomy in female reproductives, resulting in ergatoid queens externally indistinguishable from workers. In other myrmicine genera, such as *Megalomyrmex* and *Monomorium*, some female reproductives similarly have lost most of the features characteristic of a queen caste specialized for dispersal and mating flights, such as wings, ocelli and a complex thorax structure, but differ from the workers in size or in having at least a much larger gaster (Bolton 1986; Brandão 1990; Buschinger & Heinze 1992). In *Ocymyrmex*, queens can only be distinguished from workers in the anatomy of their ovaries and differences in head sculpture, antennal scapes and frontal lobes (Bolton 1981; Forder & Marsh 1989).

In any case, it is surprising that the majority of *E. mocquerysi* females with six ovaries and spermatheca take over the duties of regular workers, including foraging. To solve the question of whether reproductive females of *E. mocquerysi* are derived ontogenetically from the worker or queen caste it might help to examine worker ovaries in other species of *Eutetramorium* which have morphologically distinct queens (e.g. *E. monticellii*, Alpert & Rabeson 1999). No such data are currently available.

Removal of the major egg layer resulted in an almost immediate increase of aggressive interactions among the remaining individuals. Nestmate antagonism resembled aggressive interactions observed in other ant species. Compared with, e.g. *Leptothorax* colonies (Cole 1981; Heinze et al. 1997), fights occurred at an extraordinarily high rate (see Table 2) and were very intense, although they never led to injuries. Ants whipped the head and thorax of an opponent with their antennae or seized its head with their mandibles and pulled the opponent over a distance of several centimeters. This latter type of antagonism has previously not been reported from ants. During pulling, the tip of the gaster typically pointed towards the other ant, resembling the dominance displays in *Harpagoxenus sublaevis* (Bourke 1988; Heinze et al. 1994b) and *Dinoponera quadriceps* (T. Monnin, pers. comm.). From this position, chemicals might be sprayed or secreted from a gland towards the opponent. Pygidial and sternal glands are indeed well developed in *E. mocquerysi*, but bioassays with secretions from these glands did not elicit submissive behavior in the ants. Instead, it is likely that at least one of the two glands produces a trail pheromone (B.H., unpubl. obs.). Alternatively, it might be mechanically easier to move rapidly backwards with the gaster bent underneath the alitrunk. Duration and number of strokes during antennation bouts differed significantly between individuals but the significance of this variance is not yet understood.

Highly aggressive individuals exhibited a stiling posture and inflated their gasters. This behavior is reminiscent of the ritualized display of workers of *Myrmecocystus* honey ants during territorial contests, which has been interpreted as an attempt by individuals to appear larger (Hölldobler 1981, 1986; Lumsden & Hölldobler 1983). Worker size in *Myrmecocystus* increases with colony size, and because larger colonies have an advantage in territorial contests it was suggested that individual workers attempt to appear larger to feign larger colony size. In an intracolony context, size is probably an important determinant of dominance rank in paper wasps and bumble bees (e.g. Sullivan & Strassmann 1984; van Doorn 1989). If this was also the case in *E. mocquerysi*, workers might stilt to feign larger body size during dominance interactions. However, there is no evidence for top-ranking individuals in *E. mocquerysi* being larger than lower-ranking individuals (J. Heinze, unpubl. obs.), hence in this species the meaning of stiling in intracolony contests is unclear.

In each colony, the top-ranking individual had more strongly developed ovaries than low-ranking individuals, suggesting that fighting serves to establish reproductive hierarchies. Interestingly, males that were present in one colony were fiercely attacked by the dominant individual (J. Heinze, unpubl. obs.). Whether

this aggression was misplaced or was intended to prevent males from attempting to mate with other females is not known. Males remained in the nest near the brood pile and did not show sexual behavior. It is noteworthy, however, that males exhibited social behavior, i.e. active trophallaxis and aiding young nestmates in pupal eclosion. Such behavior is rare in male ants and has been studied in detail only in some *Camponotus* species, where males have a relatively long life span (Hölldobler 1965, 1966).

Our study on *Eutetramorium* demonstrates for the first time the occurrence of nestmate antagonism among mated and unmated nestmates in an ant taxon other than the Formicoxenini or Ponerinae and supports the idea that reproduction may be regulated by physical aggression only in species of social insects with small colony size (Heinze et al. 1994a). Only in societies consisting of less than a few dozen individuals, be it wasps, bumble bees or ants, is it possible for a dominant individual to maintain its reproductive monopoly by physically controlling all nestmates with antennation bouts or biting. Furthermore, in small societies the mated reproductives do not lay large numbers of eggs per day and, consequently, the caste dimorphism is not very pronounced. Workers may potentially be as efficient egg layers as the reproductives and, instead of refraining from reproduction (self-policing), they may compete with the reproductive for egg-laying.

The queens of monogynous species with large colony size also show adaptations to dispersal and solitary founding, such as strong flight muscles and large fat reserves. The surprising discovery of a founding colony of *E. mocquerysi* consisting of a single individual and brood (Alpert & Rabeson 1999) in this species suggests that mated individuals may be capable of independently founding a new society and sheds doubt on the common assumption that mated females less well endowed with reserves are generally incapable of solitary founding. It is nevertheless likely that new colonies of *E. mocquerysi* may also be founded by budding or fission, as is typically the case in ant species with worker-like reproductives (e.g. Buschinger & Heinze 1992).

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Appendix 1

Percentage of scans during which a certain category of behavior was performed by individuals (named by capital letters) in colony 94A of the ant *Eutetramorium mocquerysi* (total number of scans: 270 over 12 d). The percentages do not add up to 100 per individual because resting is not included. The data in this appendix were used to construct the tree in Fig. 1

	Ovarioles	Foraging	Guards			Feeds			Grooming		Trophallaxis		Aggressive interactions		Interaction with brood	
			nest	cleans	on solid food	self	active	passive	active	passive	mutual	attacks	Pupae	Larvae	Eggs	
P	6	+	0.37	1.31	0.19	0.75	5.79	0.00	10.84	0.00	0.19	0.37	14.21	0.37	0.56	0.19
X	—	—	0.19	0.00	0.00	0.19	1.12	0.75	0.56	0.56	0.00	0.37	2.62	0.37	5.23	0.19
A	—	—	0.00	0.00	0.00	0.00	3.36	1.31	1.87	0.00	0.19	0.19	4.49	2.43	5.05	0.93
G	6	—	0.00	0.00	0.00	0.00	0.75	0.75	0.56	0.00	0.00	0.19	2.43	2.24	5.98	2.43
RG	2	—	0.56	0.00	0.37	0.56	0.56	0.75	1.12	0.75	0.93	0.00	0.37	0.75	3.36	1.68
O	—	—	0.00	0.00	0.19	0.00	0.56	1.87	1.31	0.00	0.37	0.37	4.86	4.49	3.55	0.56
V	—	—	0.00	0.00	0.19	0.00	0.93	0.56	0.19	0.00	0.93	0.00	0.19	1.49	4.48	0.93
B	—	—	0.37	0.00	0.00	0.00	0.56	1.31	0.75	0.00	0.19	0.37	7.48	3.18	5.05	0.93
K	2	—	0.00	0.00	0.00	1.68	1.68	2.24	0.37	0.00	0.19	0.00	1.12	1.68	11.21	0.75
Z	6	—	0.00	0.00	0.00	0.00	0.19	0.75	0.75	0.00	0.00	0.00	0.37	1.31	2.24	0.37
R	—	—	0.00	0.00	0.00	0.00	0.935	1.87	2.62	0.00	0.37	0.56	4.86	5.23	11.21	0.56
Y	—	—	0.37	0.00	0.00	0.00	1.31	2.06	1.68	0.56	0.75	0.00	4.86	7.85	7.85	0.56
OB	—	—	0.00	0.00	0.00	0.00	1.12	0.93	0.75	0.00	0.19	0.00	0.93	0.37	8.04	1.87
E	6	—	0.37	0.00	0.00	0.00	1.49	2.80	2.06	0.00	0.19	0.00	2.43	5.05	6.73	0.37
D	6	—	5.47	0.00	0.00	0.00	2.95	0.42	0.63	0.00	0.00	0.21	3.79	5.47	5.89	0.42
RY	6	—	11.78	3.36	0.00	1.87	2.62	0.37	0.37	0.00	0.75	0.00	0.19	0.37	0.00	0.19
GB	6	—	6.17	0.37	0.37	2.43	5.05	1.68	0.00	0.56	0.00	0.00	0.00	0.19	2.24	0.00
SS	—	—	11.18	1.68	0.00	1.87	2.06	1.68	0.00	0.00	0.19	0.00	0.00	0.19	1.12	0.00
BB	—	—	10.84	0.37	0.00	2.06	2.06	1.12	1.31	0.19	0.37	0.00	0.00	0.19	0.93	0.00
GO	6	—	32.71	1.49	0.93	1.31	1.68	0.75	0.37	0.00	0.00	0.00	0.75	0.87	0.37	0.00



Appendix (continued)

	Ovarioles	Foraging	Guards		Feeds		Grooming		Trophallaxis		Aggressive interactions		Interaction with brood	
			nest	cleans nest	on solid food	self	active	passive	active	passive	mutual	attacks is attacked	Pupae	Larvae Eggs
GR	6	5.05	0.75	0.00	1.12	5.23	1.49	1.87	1.49	0.00	0.00	0.00	4.30	0.00 0.00
BO	6	18.13	2.43	0.00	2.24	3.92	0.93	0.93	1.68	0.00	0.00	0.56	0.19	0.00 0.00
BG	6	0.37	0.00	0.00	1.31	4.30	2.21	1.31	0.19	0.75	0.00	0.00	3.18	0.00 0.00
BS	2	0.00	0.00	0.00	0.00	1.68	0.00	0.93	0.00	0.37	0.00	0.00	8.60	0.37 0.00
OY	6	0.00	0.00	0.00	0.19	2.43	2.43	0.56	0.00	3.92	0.19	0.00	5.42	0.19 0.19
BA	2	1.31	0.00	0.00	5.42	6.92	4.30	1.31	0.00	1.12	0.00	0.00	4.67	0.00 0.00
SO	2	0.93	0.19	0.00	1.68	1.68	0.93	0.19	0.00	0.19	0.00	0.00	4.11	0.19 0.00
AB	2	0.00	0.00	0.00	0.56	2.80	0.93	1.49	0.00	0.56	0.19	0.00	7.85	0.37 0.56
SA	—	0.00	0.19	0.00	0.00	5.05	1.12	0.00	0.00	0.56	0.00	0.00	3.92	0.37 0.00
RO	6	27.85	3.74	0.00	0.75	0.37	0.56	0.56	0.56	0.00	0.00	0.00	1.31	0.00 0.00
OA	—	14.21	1.12	0.37	0.19	2.80	0.56	0.56	1.12	0.00	0.00	0.00	0.75	0.00 0.00
RR	2	15.70	1.68	0.00	1.12	1.31	2.43	0.37	0.00	0.37	0.19	0.00	1.12	0.00 0.00
GY	—	0.00	0.19	0.00	0.75	1.31	2.06	0.37	0.00	0.37	0.19	0.19	3.92	0.19 0.00
OP	—	17.57	5.23	0.00	1.31	1.49	0.19	0.75	2.62	0.00	0.00	0.19	0.19	0.00 0.00
AA	6	11.78	1.12	0.37	1.87	2.99	0.93	2.99	0.00	0.00	0.00	0.00	0.37	0.00 0.00
YB	6	12.90	0.75	0.19	1.31	2.80	1.31	1.68	1.68	0.00	0.00	0.00	0.19	0.00 0.00
SR	2	0.00	0.00	0.00	0.56	1.31	0.93	1.68	0.00	0.56	0.00	0.00	1.49	0.75 0.19
SB	6	0.19	0.00	1.12	0.00	1.12	1.12	0.19	1.49	0.19	1.31	2.80	0.37	0.00