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Reproduction by queens and gamergates in the Oriental ponerine ant *Pachycondyla* (= *Ectomomyrmex*) *leeuwenhoeki* var. *sumatrensis* Forel

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Abstract Colony composition and behavioural characteristics of the Oriental ponerine ant *Pachycondyla* (= *Ectomomyrmex*) *leeuwenhoeki* var. *sumatrensis* Forel were investigated in Ulu Gombak, Peninsular Malaysia. The species is AQ+G type: sexual reproduction was performed by dealated queen (seven colonies) or mated worker (eight colonies). All colonies were monogynous with one mated egg-laying female. Colony size was very small with a handful of workers. In both the gamergate and queen colonies, dominance interactions were very rare but frequent aggressions were observed in orphan colonies. The most dominant worker in an orphan subcolony mated with a male. Alpha worker of the other orphan subcolony was intensively attacked by workers of the mother colony when the subcolony was merged to the mother colony.

Keywords *Pachycondyla leeuwenhoeki* – ponerine ant – reproduction – gamergates

INTRODUCTION

Ants in the subfamilies Amblyoponinae, Ectatomminae, and Ponerinae show remarkable diversity in reproductive structure of colonies [1-3]. Most species show sexual reproduction only by morphologically distinctive queens. However, some species reproduce by mated and egg-laying workers (gamergates) without queens. Furthermore, sexual reproduction by both queens and gamergates is also known in other species [1, 3]. In the species with gamergates, the mechanism regulating reproduction among workers varies greatly. However, the significance of the diversity of reproductive regulation is still unknown. We still need information on the biology and behaviour of several gamergate species.

The ponerine genus *Pachycondyla* has many gamergate species [1, 3]. In the Oriental tropics, the colony composition and regulation of reproduction has been demonstrated for only *Pachycondyla* (= *Bothroponera*) sp. [4, 5]. In this paper, we report colony composition and behavioral characteristics of *P.* (= *Ectomomyrmex*) *leeuwenhoeki* var. *sumatrensis* collected in Malaysia.

MATERIALS AND METHODS

All colonies were collected from decayed woods on the forest floor or under the soil in Ulu Gombak, Peninsular Malaysia (Table 1). Adult females of all but three colonies were dissected just after collection to determine the reproductive condition. Three colonies (FI92-MG515, FI99-282, AG07-25) were kept in a laboratory. All adult individuals were marked by enamel paints (Fig. 1). Their behaviour was observed for 20 to 100 hours.

Two subcolonies consisted of six virgin workers each, were created from one gamergate colony (FI99-282) when the colony consisted of 20 workers. The behaviour of workers was observed for one hour per day until the establishment of a dominance relationship. After that, such observation was carried out once or twice a week. Three weeks after orphaning, a male that emerged in the other orphan

Table 1. Colony composition of *P. leeuwenhoeki*. Incipient colonies were excluded. DQ: dealated queen; AQ: alate queen; mW: mated worker; vW: virgin worker; C: cocoon; L: larva; E: eggs; -: not counted; +: present but not counted. FI,YI, and AG mean colonies collected by F. Ito, Y. Ikeshita and A. Gotoh, respectively.

	Number of individuals						
Colony code	DQ	AQ	mW	vW	C	L	E
FI92-MG515	1(1)	0	0	2	-	-	-
YI04-97	1(1)	0	0	3	-	-	-
FI92-MG610	1(1)	0	0	10	-	-	-
AG07-25	1(1)	1	0	11	0	8	+
YI04-69	0	0	1(1)	1	0	0	0
FI92-MG567	0	0	1(1)	3	0	4	7
FI03-54	0	0	1(1)	3	-	-	-
YI04-86	0	0	1(1)	3	0	0	0
FI92-MG53	0	0	1(1)	5	3	0	1
FI96-547	0	0	1(1)	6	-	-	-
YI04-43	0	0	1(1)	8	-	-	-
FI99-282	0	0	1(1)	12	-	-	-

Figure 1. A colony of *Pachycondyla leeuwenhoeki* collected in Ulu Gombak. Individual was marked with enamel paints.

colony collected in the field was introduced into the subcolony A and subsequent behaviour of the workers was observed. Three weeks after the male was introduced, all workers in the subcolony were dissected. The other subcolony was merged with the mother colony after the establishment of a dominance hierarchy among the virgin workers, and behavioural interactions between workers of the mother colony and those of the subcolony was observed. After that all the workers of the subcolonies were dissected. Spermatheca width was measured under a light microscope for three randomly selected queens and 17 workers. Head width and width of the 2nd gaster segment of these individuals and other three queens and 15 workers were measured under binocular microscope.

RESULTS

Colony composition

In all fifteen complete colonies were collected over a nine-year period in the Southeast Asian tropics, indicating that *P. leeuwenhoeki* is not a common species. Seven of the 15 colonies were reproduced by only one dealated queen. Of these three were incipient colonies with a dealate queen and a few immatures, indicating that *P. leeuwenhoeki* queens start their colonies independently. Two incipient colonies produced three worker pupae each. The remaining eight colonies were reproduced by gamergates. All colonies were monogyny with one mated worker as gamergates. Virgin workers had neither yolky oocytes nor yellow bodies. Colony size of both queen colonies and gamergate colonies was small (mean number of virgin workers in queen colonies 6.5 $\pm$ 4.7, gamergate colonies 5.1 $\pm$ 3.5, Mann Whitney U test, Z = –0.26, p = 0. 79).

Queen body size was significantly larger than workers (Head width, queens, 1.67 $\pm$ 0.08 mm, workers, 1.53 $\pm$ 0.06 mm, Mann Whitney U test, Z = –2.87, p = 0. 004; 2nd gaster segment, queens, 1.60 $\pm$ 0.11 mm, workers, 1.41 $\pm$ 0.06 mm, Z = –3.58, P = 0.0003). However, as in other AQ+G species, the difference was not conspicuous. Both queens and workers had three ovarioles per ovary. Spermatheca size of queens (131.7 $\pm$ 20.8 μm) was slightly larger than workers (119.7 $\pm$ 13.2μm), but not statistically significant (Mann Whitney U test, Z = 0.59321, p = 0.553).

Reproduction

Aggression was never observed in the gamergate colony (FI99-282), and the queen colonies (FI92-MG 515, AG07-25). In FI99-282, egg-laying was observed 10 times, all of which were performed by the gamergate. As indicated by dissection data, the virgin workers never laid eggs during the 100 hours observation.

In the orphan subcolonies, aggressive antennations and biting were frequently observed among the workers. Just after creating these subcolonies, mutual antennations were very frequent but it was difficult to know who was the most dominant. Mutual antennation often continued for a few minutes. The frequency of

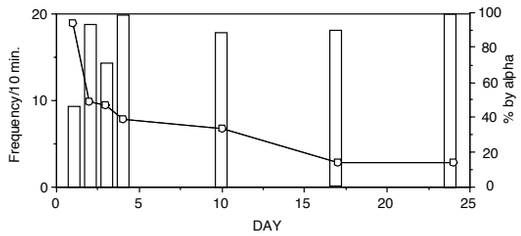

Figure 2. Successive change of frequnecy of aggressive behaviour/10 min (circle, left axis), and proportion of the behaviour performed by alpha workers (bar, right axis) in orphan subcolony A.

aggressions, however, drastically decreased on the 2nd to 4th day, then most aggressions were performed by one worker (Fig. 2). Once the dominance hierarchy was established, mutual aggression between alpha workers and the other workers became rare. The aggressed workers showed a typical subordinate behaviour against the alpha: they crouched and froze in response to the antennation.

In subcolony A, the first egg appeared 18 days after orphaning. Three weeks after orphaning, the alpha worker sometimes showed sexual calling behaviour inside the nest chamber: she bent her abdomen and extruded the sting. When the alpha worker showed such behaviour, we introduced a male produced in a different orphan colony that was collected in the field. The male quickly went to the nest chamber, rushed to the alpha worker and copulated with her (ca. 5 seconds). It repeated such copulation with her three times. He was never interested in the other workers. Insemination of the alpha worker was confirmed by dissection after the observation. Developing ovaries were found in the alpha worker only.

In subcolony B, egg laying by the alpha worker was confirmed on the 21st day after orphaning. The subcolony was merged with the parental colony on 30th day when five eggs were found in the subcolony. The alpha worker of subcolony B was intensively attacked by the resident workers in the parental colony. Subordinate workers of the subcolony were rarely attacked. Dissection after observation indicated that only the alpha worker had developed ovaries.

DISCUSSION

The present result of colony composition indicates that *Pachycondyla leeuwenhoeki* belongs to the AQ+G species [1, 3]. However, the data of colony composition is not enough for illustrating the life history of this species, because it is not common. Speculation of the life history, however, may be possible: single dealate queens start new colony independently, then, gamergate reproduction occurs after the death of the queen or after fission from queenright colonies. In seven of the eight AQ+G species so far reported, there is a rule that queen colonies are monogynous while gamergate colonies are polygynous (reproduction involving multiple gamergates) [6]. The remaining species, *Gnamptogenys striatula*, shows polygyny in both the queen and gamergate colonies [7]. In *P. leeuwenhoeki,* however, both the queen colonies and gamergate colonies are monogynous. One of the reasons why the species shows monogyny with gamergate may be their remarkably small colony size. The AQ+G species *Pachycondyla* (*Mesoponera*) sp. and *P.* (*Bothroponera*) sp., which also show monogyny in gamergate colonies also have small colony size with a handful of workers [4, unpub.]. In such small colony sized species, the presence of excess number of gamergates may result in high cost for colony productivity.

The significance of co-occurrence of queens and gamergates in the same species is still not well understood. In the Indian ponerine ant *Harpegnathos saltator,* it has been proposed that colonial fission by workers is absent because workers retain the very complex underground nests which are valuable for them [8]. Thus, alate queens are necessary for dispersal in this species. In *Gnamptogenys menadensis*, dispersal by alate queens is crucial, because they are arboreal ants [9]. Nests of *P. leeuwenhoeki* are just rotten wood on the forest floor, and it is unlikely that they inherit such fragile nest substrates. In other AQ+G species in the Oriental tropics, most of them nest in dead wood fallen on the ground [unpublished]. Thus, social structure of AQ+G species is not always associated with the existence of very valuable nests. In general, the retention of the dispersal form like alate queens of ants is adaptive even under stable environment [10]. Thus, if colony foundation by single alate queens is possible, alate queens may be produced even in species with gamergates.

In some other *Pachycondyla* species with gamergates – e.g. *P.* (*Bothroponera*) *sublaevis*, *P.* (*Bothroponera*) sp. – a dominance hierarchy based on aggressive dominant-subordinated behaviour is frequently observed among the workers, and

gamergate is the top-ranked individual [4, 11]. In contrast, such aggressive behaviour was rarely observed in *P. leeuwenhoeki* colonies with reproductive females. It is noteworthy why some species make a dominance hierarchy based on aggressive behaviour while the others do not show such aggression in the presence of reproductive females, even though all three species mentioned above have similar colony size. A knowledge of many other ponerine species with gamergates is needed for the answer. As in other ponerine ants [e.g. 11-13], workers of *P. leeuwenhoeki* showed aggressions in the absence of reproductive females. The frequency of aggressions was drastically decreased in a few days after orphaning, and the alpha worker subsequently laid eggs. Furthermore, virgin workers under gamergate-present condition showed typical policing behaviour: they attacked the workers with developed ovaries. These observations indicate that workers recognized the ovary development of nestmate as shown in the gamergate species *Dinoponera quadriceps* [14], *Gnamptogenys menadensis* [15], and *Diacamma* sp. [16]. The recognition mechanism is another important topic for the study on the biology of this ant species.

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REFERENCES

1. Ito F. and Ohkawara K. (1994) Spermatheca size differentiation between queens and workers in primitive ants: relationship with reproductive structure of colonies. *Naturwissenschaften* **81**: 138-140.
2. Peeters C. (1997) Morphologically "primitive" ants: comparative review of social characters, and the importance of queen-worker dimorphism. *In* Choe J. and Crespi B. (eds) *The Evolution of Social Behavior in Insects and Arachnids,* pp 372-391. Cambridge Univ. Press.
3. Peeters C. and Ito F. (2001) Colony dispersal and the evolution of queen morphology in social Hymenoptera. *Annual Review of Entomology* **46**: 601-630.
4. Ito F. (1993) Functional monogyny and dominance hierarchy in the queenless ponerine ant *Pachycondyla* sp. in West Java, Indonesia. *Ethology* **95**: 126-40.
5. Ito F. (1999) Male behavior and regulation of worker mating in the ponerine ant *Pachycondyla (Bothroponera)* sp. (Hymenoptera : Formicidae). *Journal of Insect Behavior* **12**: 193-198.
6. Monnin T and Peeters C. (2007) How many gamergates is an ant queen worth? *Naturwissenschaften* (in press).
7. Blatrix R. and Jaisson P. (2000) Optimal gamergates in the queenright ponerine ant *Gnamptogenys striatula* Mayr. *Insectes Sociaux* **47**: 193-197.
8. Peeters C. and Hölldobler B. (1995) Reproductive cooperation between queens and their mated workers: the complex life history of an ant with a valuable nest. *Proceedings of the National Academy of Sciences of the United states of America* **92**: 10977-10979.
9. Gobin B., Peeters C. and Billen J. (1998) Colony reproduction and arboreal life in the ponerine ant *Gnamptogenys menadensis* (Hymenoptera: Formicidae). *Netherland Journal of Zoology* **48: 53-63.**
10. Hamilton W.D. and May R.M. (1977) Dispersal in stable habitats. *Nature* **269:** 578-81.
11. Ito F. and Higashi S. (1991) A linear dominance hierarchy regulating reproduction and polyethism in the queenless ponerine ant *Pachycondyla sublaevis*. *Naturwissenschaften* **78**: 80-82.
12. Gobin B, Peeters C. and Billen J. (2001) Dominance interactions regulate worker mating in the polygynous ponerine ant *Gnamptogenys menadensis. Ethology* **107**: 495-508.
13. van Walsum E., Gobin B., Ito F. and Billen J. (1998) Worker reproduction in the ponerine ant *Odontomachus simillimus*. *Sociobiology* **32**: 427-440.
14. Monnin T and Peeters C. (1999) Dominance hierarchy and reproductive conflicts among subordinates in a monogynous queenless ant. *Behavioral Ecology* **10**: 323-332.
15. Gobin B., Billen J. and Peeters C. (1999) Policing behaviour towards virgin egg-layers in a polygynous ponerine ant. *Animal Behaviour* **58**: 1117-1122.
16. Tsuji K., Egashira K. and Hölldobler B. (1999). Regulation of worker reproduction by direct physical contacts in the ant *Diacamma* sp. from Japan. *Animal Behavior* **58:** 337-343.