

***Ropalidia rufoplagiata*: a polistine wasp society probably lacking permanent reproductive division of labour**

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Summary

Ropalidia rufoplagiata Cameron (Hymenoptera: Vespidae), a polistine species from peninsular India, appears to be unique among all known primitively eusocial wasps. A total of 33 out of 46 identified females from an observed colony were found to oviposit on 1–17 occasions. No single predominant egg-layer could be identified during the 45-day period. Of the 17 dissected egg-layers, 12 were mated. All egg-layers showed several oviposition-related behavioural patterns including systematic, but indiscriminate, cannibalism of eggs and larvae, cleaning of empty cells, and guarding of freshly-laid eggs. There was no correlation between the egg-laying activity of the females (whether mated or not), oophagy, and their position in the dominance hierarchy. All nest-maintenance activities were performed exclusively by the egg-layers, while the non-egg-layers were mainly involved in the extranidal task of foraging. No significant morphometric differences between egg-layers and foragers could be discerned. Almost all the older individuals in the colony were egg-layers, while foragers were mainly younger animals. Such a temporal differentiation in reproductive labour suggests the absence of a permanent reproductive caste in this species.

Introduction

Ropalidia is a large genus with about 136 known species confined to tropical and subtropical regions of the Old World, including Africa, southern Asia, Australia, and Okinawa (for a review, see Gadagkar 1991 a). This is the only genus that includes both independent-founding and swarm-founding species and consequently shows remarkable variation in adult social behaviour and colony organisation (Jeanne, 1980, 1991; Wenzel, 1987, 1992; Gadagkar, 1991 a). The independent-founding species of *Ropalidia* are characterised by a relatively primitive level of eusociality: they lack morphological caste differentiation and possess an essentially behavioural mechanism of queen control. A linear dominance hierarchy among founding females or among colony members results in the most dominant female being the principal or sole egg-layer (“queen”), rarely or never leaving the nest to forage, while the subordinate “workers” rarely or never lay eggs.

Dominance interactions and dominance displays shown by colony members usually reflect the extent of reproductive competition that these individuals might face at different phases of colony development. Several behavioural patterns involved in establishing reproductive dominance in multiple-foundress associations or in post-emergence colonies are common among polistine wasps. These include egg-guarding, differential oophagy and egg substitution, and even physical prevention of subordinates from laying eggs. Such behaviour, however, appears to be rare in *Ropalidia*. Cell-guarding, for example, has never been recorded for any species, while oophagy has only been demonstrated in *R. cyathiformis* (Gadagkar and Joshi, 1982) and *R. fasciata* (Itô, 1987b).

The nature and extent of reproductive competition, in turn, determines the number of functional egg-layers in a colony. Thus, even in species of *Ropalidia* where nests may be founded pleometrotically, monogyny is usually established in the early stages of colony development through a definite dominance hierarchy, e.g. in *R. revolutionalis* (Itô, 1986). Although typical of small colonies, monogyny may often give way to serial polygyny as a result of usurpation or queen succession, especially in species with indeterminate nesting cycles like *R. fasciata* (Turillazzi and Marucelli Turillazzi, 1985), *R. variegata jacobsoni* (Yamane, 1986), and *R. marginata* (Gadagkar et al., 1991). It must be stressed, however, that at least in the case of *R. marginata*, there is only a single egg-layer at any given time. A different situation, on the other hand, occurs in species like *R. cyathiformis* (Gadagkar and Joshi, 1982) or *R. fasciata* from Okinawa (Itô, 1987b). Here, the dominance hierarchy established within the nest is weak, especially in large post-emergence colonies, and simultaneous polygyny may prevail, with the majority of eggs being laid by the most dominant α -female and the remaining small fraction by the higher-ranked subordinates.

This paper reports the social behaviour and colony organisation of the independent-founding polygynous wasp, *R. rufoplagiata* Cameron (Hymenoptera: Vespidae) (see Das and Gupta, 1989). This species appears to be unique among all polistines described so far in that the majority of adults in the colony regularly lay eggs. There does not appear to be any one predominant egg-layer of higher dominance rank, and there is, in fact, no correlation between egg-laying activity and ranks in the dominance hierarchy. All the egg-laying individuals systematically guard cells and practise oophagy. Finally, there is some evidence of a temporal reproductive division of labour so that all colony members may ultimately lay eggs, an indication that this species may not have evolved permanent reproductive castes.

Materials and methods

Study colony

This study was conducted on a single post-emergence colony at the Indian Institute of Science, Bangalore (13°00'N and 77°32'E), between 27 February and 15 April 1991. A single multi-pedicellate nest, measuring approximately 12 cm × 6 cm, was found suspended in an inverted fashion from the portico of a house in the Malleswaram locality of the city and transferred to the vespiary at the Institute on 26 February.

There, the nest was maintained in an open cage such that the adults could forage freely, not only from natural sources but also from a supply of honey, *Corcyra cephalonica* larvae, and water provided within the vespiary.

At the time of collection, 47 adults were present on the nest consisting of 295 cells with 125 eggs, 118 larvae, 42 pupae, and only 10 empty cells. All the adults were marked for individual identification with spots of coloured paint; each day newly-eclosed wasps were marked. A total of 153 animals was marked during the entire observation period, of which 96 individuals had eclosed in the vespiary. A census of all animals was made several times randomly during each day and a map of the nest and its contents maintained regularly. Unfortunately, it was not possible to distinguish between the sexes initially; this could only be accomplished after the individuals had been collected and examined under a binocular microscope.

Sampling methods

Behavioural observations of the colony included *ad libitum* sampling, instantaneous scanning, and recording all occurrences of selected behavioural patterns, as described by Gadagkar and Joshi (1983).

A total of 25.5 h of observation was performed during a period of 7 days, between 9 and 15 April 1992. Initial *ad libitum* observations showed that the wasps were active mainly between 0800 and 1800 h. Observations were thus conducted during these hours with a random intermingling of instantaneous scans and sessions recording all occurrences of selected behavioural patterns.

All adult females, considered in the final behavioural analysis, were at least 4 days of age on the first day of observation (9 April 1991) and were also present for at least 20 h of the entire 30-h observation period.

Data on egg-laying and cannibalistic behaviour were collected during 46.5 h of observation distributed over two periods: from 21 to 29 March, and again from 9 to 15 April 1991. The 15 sessions, dedicated to recording every occurrence of oviposition and oophagy, ranged in duration from 1 to 6.75 h with a median of 3.5 h, and spanned 10 h of the day, from 0800 to 1800 hours.

At the end of the observations, on 15 April 1991, all adult wasps (with a minimum age of 9 days) were collected. The females were assessed in terms of their body size, ovarian condition (as measured by oocyte number), and mating status, and finally dried at 60 °C for 15 days to record their dry weight.

Data analysis

1. Behavioural repertoire, time-activity budgets, and rates of selected behavioural patterns: The behavioural repertoire of this species was classified into distinct categories that were similar to those of *R. marginata* and *R. cyathiformis* (Gadagkar and Joshi, 1983, 1984).

Time-activity budgets, or the proportion of time spent in different behavioural patterns by different animals, were estimated using data from instantaneous scans.

The hourly frequencies with which certain selected behavioural patterns were performed were calculated from the sessions recording all occurrences of such behavioural events, as described earlier (Gadagkar and Joshi, 1984).

2. *Principal component analysis*: Time-activity budgets, constructed from the proportions of time spent in six common behavioural patterns, were subjected to principal component analysis (Frey and Pimentel, 1978). The resulting clusters were identified as described earlier by Gadagkar and Joshi (1983).

3. *Index of body size*: An index of body size was computed as the amplitude along the first principal component of 27 input morphometric measures, namely, length and width of the head, interocular distance, oculo-ocellar distance (left and right), interantennal socket distance, length and width of the frontal segment of the antennae (left and right), length and width of the clypeus, length and width of the mesoscutellum, alitrunk length, length of left and right wings, length of the first median wing cell (left and right), number of hamuli (left and right), and the length, width, and height of the first and second gastral segments. Due to the wide variation in the absolute values of the different variables, a correlation matrix, rather than a covariance matrix, was used to perform the principal component analysis.

4. *Dominance hierarchy*: A dominance hierarchy was constructed for all the colony members using an index of dominance described by Premnath et al. (1990). The index was computed for each animal as:

$$\frac{\sum_{i=1}^n B_i + \sum_{j=1}^m \sum_{i=1}^n b_{ji} + 1}{\sum_{i=1}^n L_i + \sum_{j=1}^p \sum_{i=1}^n l_{ji} + 1}$$

where $\sum B_i$ is the rate at which the subject shows dominant behaviour towards other colony members and $\sum b_{ji}$ is the sum of the rates at which all animals, dominated by the subject, in turn, show dominant behaviour towards other individuals. Similarly, $\sum L_i$ is the rate at which the subject shows subordinate behaviour towards colony members and $\sum l_{ji}$ is the sum of the rates at which the latter, in turn, show subordinate behaviour towards other individuals.

5. *All statistical tests were two-tailed and performed according to Sokal and Rohlf (1981).*

Results

Identification of behavioural groups

The most striking feature of the wasps in the study colony was their egg-laying behaviour. Approximately 70% (33 out of 46 females observed for a minimum period of 14 days and 19 out of 27 females for whom quantitative behavioural data were available) of the colony members were seen to lay an egg at least once. Furthermore, many of these individuals oviposited quite regularly and frequently.

There were two distinct behavioural groups in the colony. Of the 27 females whose behaviour was analysed in detail, 17 laid eggs at least once but were never seen to forage, while 6 brought food or liquid to the nest at least once but were never observed to lay an egg. There were, however, two egg-layers that brought food and liquid on one occasion each, but also two females that neither laid eggs nor foraged (Tab. 1). Ignoring the latter and considering the former as egg-layers, two behavioural groups were considered in the subsequent analysis, namely egg-layers and foragers. Three additional arguments justifying such a behavioural classification are given below.

1. *Clusters identified by principal component analysis.* In an independent approach to examine other behavioural criteria on the basis of which the wasps could be clustered, time-activity budgets were constructed for 27 individually identified members of the colony. It was found that the wasps spent 81–100% (mean \pm s.d., 90.94 ± 0.04) of their time in six common behavioural patterns, namely sit and groom, sit with raised antennae, absent from nest, walk, in cells, and guard cells (in which the wasp sits with her head inside a cell for a variable period of time). Individuals allocated their time differently to each of these behavioural patterns. Accordingly, in order to discern behavioural patterns within the colony, these time-activity budgets were subjected to principal component analysis. The results show that the first two principal components together account for 98.24% of the total variance and may thus be considered adequate to describe the behaviour of these individuals (Tab. 2). The representation of each wasp as a point in the coordinate space of the first two principal components results in two distinct clusters, further confirmed by the nearest centroid criterion (Gadagkar and Joshi, 1983) (Fig. 1).

An examination of the behavioural profile of the individuals in the two clusters revealed that there was a significantly greater proportion of egg-layers in Cluster I than in Cluster II (Tab. 3, G-test of independence, $p < 0.01$). The proportion of foragers, in contrast, was significantly greater in Cluster II than in Cluster I (Tab. 3,

Table 1. Task-allocation between the colony members with respect to egg-laying and foraging. The distribution was examined with the G-test of independence

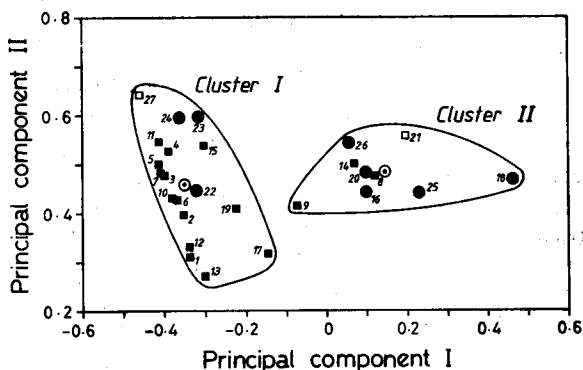
| Behavioural groups | Foragers | Non-foragers | G | p |
|--------------------|----------|--------------|--------|--------|
| Egg-layers | 2 | 17 | 11.032 | <0.002 |
| Non-egg-layers | 6 | 2 | | |

Table 2. Principal component analysis of time-activity budgets of colony members

| Behaviour | Principal components | |
|-----------------------------------|----------------------|---------|
| | I | II |
| Sit and groom | -0.5577 | 0.7409 |
| Absent from nest | 0.8204 | 0.4285 |
| Raise antennae | 0.0149 | -0.0181 |
| Guard cells | -0.0551 | -0.3403 |
| Walk | -0.0676 | -0.1830 |
| In cells | -0.0901 | -0.3433 |
| Eigen value | 0.0624 | 0.0080 |
| Percentage of variance | 87.12 | 11.12 |
| Cumulative percentage of variance | | 98.24 |

Table 3. Proportion of egg-layers and foragers in the two behavioural clusters obtained by principal component analysis of time-activity budgets. The distributions were examined with the *G*-test of independence

| Behavioural group | Clusters | | G | p |
|-------------------|----------|----|--------|-------|
| | I | II | | |
| Egg-layers | 16 | 3 | 8.8004 | <0.01 |
| Non-egg-layers | 2 | 6 | | |
| Foragers | 2 | 5 | 5.9800 | <0.05 |
| Non-foragers | 16 | 4 | | |

**Figure 1.** Behavioural clusters of *R. rufoplagiata*. Time-activity budgets of 27 wasps are shown as points in the coordinate space of the first two principal components. The points fall into two clusters using the criterion of nearest centroid. The centroids are represented by circled points, the egg-layers by solid squares, the foragers by solid circles, the individuals showing both egg-laying and foraging by circled squares, and those showing neither by open squares

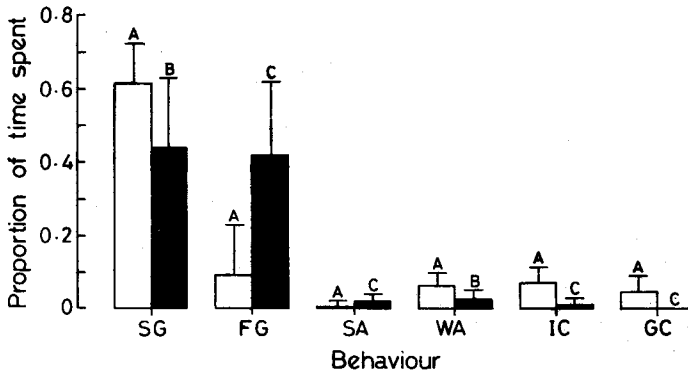


Figure 2. Mean time-activity budgets (mean + s.d.) for egg-layers (open bars) and foragers (solid bars) in the study colony. The mean proportion of time spent by a wasp in the six common behavioural patterns has been compared for the two groups using the Mann-Whitney U-test ($n_1 = 19$ egg-layers, $n_2 = 6$ foragers). Significant differences between the groups are indicated as AB ($0.05 > p > 0.01$) and AC ($p < 0.01$). SG: Sit and groom; FG: Absent from nest; SA: Sit with raised antennae; WA: Walk; IC: In cells; GC: Guard cell

G-test of independence, $p < 0.05$). One forager (Wasp 24), that collected food from within the vespiary and therefore did not spend much time away from the nest, belonged to Cluster I. Three egg-layers (Wasps 8, 9 and 14), on the other hand, were allocated to Cluster II by virtue of the significant periods of time they spent away from the nest, although never returning with food or liquid. Two individuals (Wasps 21 and 27), one from each cluster, were never seen to oviposit or to forage. It may be emphasised here that the two clusters emerged as a result of choosing, for analysis, those six behavioural patterns that had the highest ranks in the proportion of time the wasps devoted to them, and not taking into consideration egg-laying behaviour. These behavioural clusters, nevertheless, appear to be principally represented by egg-layers and foragers, respectively.

2. Correlates of the behavioural groups. In order to examine more closely the differences between egg-layers and foragers, the mean behavioural profiles of the two groups for select behavioural patterns were examined. When compared with the foragers, egg-layers invested a significantly greater proportion of their time in sitting and grooming on the nest (Mann-Whitney U-test, $p = 0.05$), while the former spent comparatively more time absent from the nest ($p < 0.01$) (Fig. 2). It may be recalled that these are also the behavioural patterns that had the maximum weight on the first two principal components that permitted the earlier clustering of the two behavioural groups.

A remarkably distinct pattern of task allocation between egg-layers and foragers becomes evident on examining other behavioural variables not used in deriving the clusters (Fig. 3). For example, egg-layers were significantly more responsible for nest-maintenance activities, such as breaking walls of unused cells (Mann-Whitney U-test, $p < 0.002$), extending old cell walls ($p < 0.05$), or cleaning empty cells ($p < 0.002$). In contrast, foragers brought food more often to the nest ($p < 0.002$) and fed larvae more frequently ($p < 0.05$) than did the egg-layers.

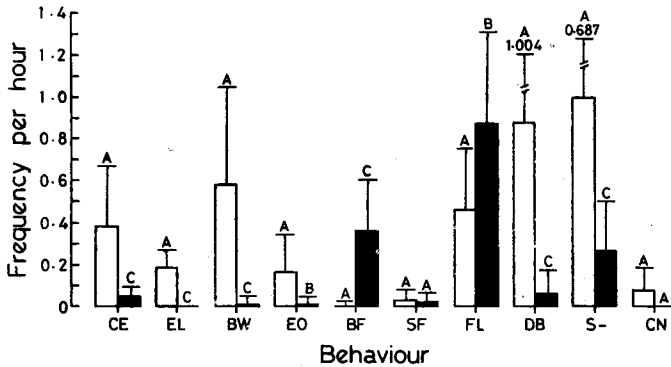


Figure 3. Mean frequency of selected behavioural patterns (mean + s.d.) performed by egg-layers (open bars) and foragers (closed bars) in the study colony. The mean frequency of occurrence of each behaviour per hour per animal was compared for the two groups using the Mann-Whitney U-test ($n_1 = 19$ egg-layers, $n_2 = 6$ foragers). The differences between the two groups are shown as AA (not significant at $p > 0.05$), AB ($0.05 > p > 0.01$), and AC ($p < 0.01$). CE: Clean empty cell; EL: Lay egg; BW: Break wall of cell; EO: Extend old wall of cell; BF: Bring food; SF: Snatch food; FL: Feed larva; DB: Dominance behaviour; S-: Subordinate behaviour; CN: Cannibalism

Dominance interactions on the nest were mainly restricted to the egg-layers and they thus showed significantly higher levels of both dominant and subordinate behaviour than did the foragers.

3. Clustering of behavioural patterns by correlation analysis. In a third approach to discern individual behavioural patterns, especially with reference to task allocation, Kendall's rank correlation coefficients were computed for the proportion of time spent by the wasps in certain behavioural states, the frequency of occurrence of select behavioural events, and oocyte number. Egg-laying was found to be strongly correlated with guarding cells, cleaning empty cells, breaking cell walls, extending old cell walls (for all behaviours, $p < 0.05$, $n = 27$), and oocyte number ($p < 0.05$). Bringing food and liquid was positively correlated with being absent from the nest and feeding larvae, but negatively correlated with egg-laying and many of its associated behaviours as well as oocyte number (for all correlations, $p < 0.05$). In order to visualise the association of different behavioural patterns as an indication of possible task allocation, the Kendall's rank correlation coefficients were used as indices of similarity between the behavioural patterns and a hierarchical cluster analysis was performed with the single linkage algorithm (De Gheff, 1978) (Fig. 4). The clustering of behavioural patterns, obtained from this analysis, parallels closely the patterns of task distribution already evident from the behavioural profiles of the egg-layers and the foragers.

Absence of morphological differences between egg-layers and foragers

The index of body size and dry weight did not differ significantly between egg-layers and foragers (data not shown). A comparison of all the evaluated morphometric

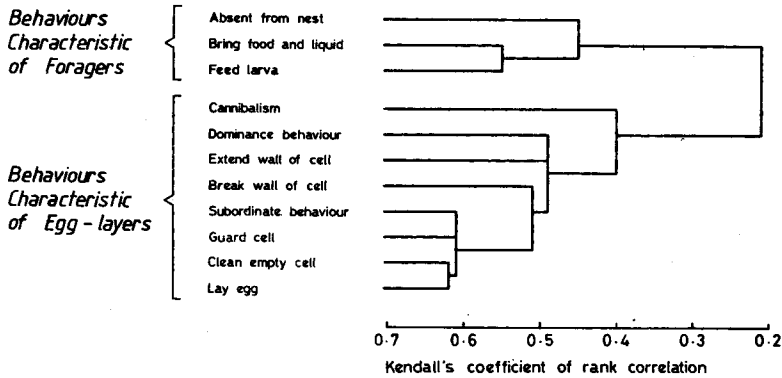


Figure 4. Hierarchical cluster analysis of the behavioural patterns shown by *R. rufoplagiata*. Time-activity budgets and the frequency of occurrence of select behavioural events for 27 colony members have been used in the analysis. The similarity between individuals has been shown in terms of Kendall's coefficients of rank correlation between these variables. The method of single linkage algorithm was used for the clustering

parameters also failed to discern any significant difference between the two behavioural groups. As might be expected, however, the oocyte number of egg-layers (mean \pm s.d., 17.4 ± 6.9) was significantly higher than that of foragers (0.8 ± 1.6) (Mann-Whitney U-test, $p < 0.001$, $N = 17$ egg-layers, 5 foragers).

Age structure of egg-layers and foragers

Of the 27 individuals included in the final behavioural analysis, 17 were females originally present on the nest when it had been collected. The exact ages of these "old" wasps were thus not known, although they were at least 25 days old at the beginning of the observation sessions. The ages of the other 10 "young" wasps, that had eclosed in the vespiary after transplantation of the nest, ranged from 6 to 21 days, with a mean of 14 ± 4.6 days.

The old wasps were overwhelmingly represented by egg-layers, while the young wasps formed a greater proportion of foragers (Table 4, G-test of independence,

Table 4. Proportion of egg-layers and foragers in the two age classes of colony members. The distribution was examined with the G-test of independence

| Behavioural group | Age class | | G | p |
|-------------------|-----------|-------|--------|--------|
| | Old | Young | | |
| Egg-layers | 16 | 3 | 12.992 | <0.002 |
| Non-egg-layers | 1 | 7 | | |
| Foragers | 1 | 7 | 12.992 | <0.002 |
| Non-foragers | 16 | 3 | | |

$p < 0.002$). Two of the three egg-layers in the young group were also foragers and were observed bringing food and liquid to the nest, although, as mentioned earlier, only on a single occasion each. None of the 16 egg-layers in the old group, however, were ever seen to forage for either food or liquid. Assuming the older wasps to be only 25 days old, the mean age of the egg-layers in the study colony was 23.6 ± 3.5 days and that of the foragers significantly different at 15.3 ± 5.6 days (Mann-Whitney U-test, $p < 0.01$, $n = 19$ egg-layers, 6 foragers).

Analysis of egg-laying behaviour

Data from 46.5 h of observation, exclusively devoted to oviposition, yielded an average rate of 4 eggs laid per hour by a total of 28 individuals, with about 3 wasps laying eggs every hour. The frequency of oviposition per hour for each of the 28 egg-layers ranged from 0.06 to 0.55, with a mean of 0.19 ± 0.12 . This is probably the most prolific rate of egg-laying by the highest proportion of colony members (70%) ever to be reported for a polistine wasp, including the swarm-founding species (see Jeanne, 1991; Spradbery, 1991). There did not appear to be any significant difference in the rate of oviposition at different times of the day (data not shown). Notwithstanding the variation in the individual rates of egg-laying shown by the wasps, no single principal egg-layer was evident during this period.

Of the 28 individuals analysed for their egg-laying behaviour, 17 were dissected and found to have mature developed ovarioles with oocytes. No correlation could be established between the frequency of oviposition and the number of oocytes present in an individual. The most remarkable finding, however, was that 12 of the 17 dissected egg-layers were mated (the spermatheca being lost during dissection for one wasp). Two individuals that had eclosed in the vespiary after the nest had been collected were also mated. This species is very rare; indeed this is the only colony we have ever seen. This suggests that, at least in this instance, mating had possibly been accomplished with males from the natal nest.

Dominance, cannibalism, and egg-laying behaviour

A nearly universal feature of primitively eusocial wasp colonies is the presence of a dominance hierarchy among the adult females and the positive correlation of egg-laying ability of the individuals with their dominance rank. This species, however, proved to be an exception. Although dominance interactions appeared to be restricted to the egg-layers alone, with very little interaction between the egg-layers and the foragers, and none whatsoever among the foragers (Fig. 5), no apparent relationship was evident between egg-laying by an individual and its dominance profile, as measured by its index of dominance. Effective oviposition by an individual also seemed to be unrelated to its mating status (Tab. 5). A negative correlation, in fact, existed between the frequency of oviposition by an individual and its index of dominance when only egg-layers were considered in the analysis (Fig. 6; Kendall's rank correlation coefficient, $\tau = -0.4201$, $p < 0.01$, $n = 19$). When the two outlying individuals with the highest values for dominance index were removed, the negative

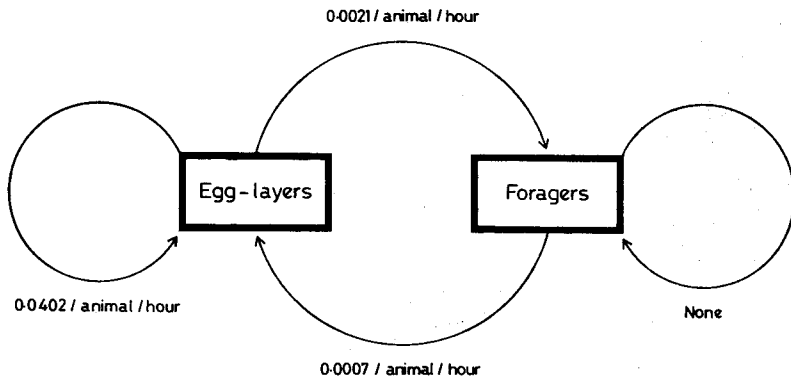


Figure 5. Mean frequency of dominant behaviour within and between egg-layers and foragers in the study colony

Table 5. Hierarchical dominance relationships between colony members

| Rank | Mating status | Oviposition | Oophagy | Foraging |
|------|---------------|-------------|---------|----------|
| 1 | + | + | + | - |
| 2 | + | + | + | + |
| 3 | ? | - | - | - |
| 4 | + | + | + | - |
| 5 | - | + | + | - |
| 6 | + | + | + | - |
| 7 | - | + | + | - |
| 8 | - | + | + | - |
| 9 | ? | + | - | + |
| 10 | - | - | - | + |
| 11 | - | - | - | + |
| 12 | + | + | + | - |
| 13 | ? | + | + | - |
| 14 | + | + | - | - |
| 15 | ? | - | - | - |
| 16 | - | - | - | + |
| 17 | + | + | + | - |
| 18 | + | + | + | - |
| 19 | + | + | + | - |
| 20 | + | + | + | - |
| 21 | + | + | - | - |
| 22 | ? | - | - | + |
| 23 | - | - | - | + |
| 24 | ? | + | + | - |
| 25 | + | + | + | - |
| 26 | - | + | + | - |

+: Mated individuals; oviposition, oophagy, and/or foraging observed

?: Mating status unknown

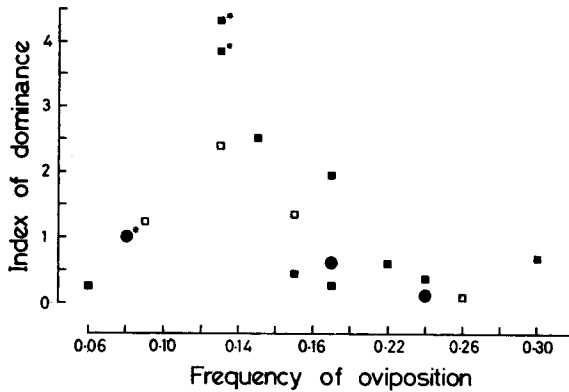


Figure 6. Scatter plot showing the dominance index of an egg-layer as a function of her frequency of oviposition per hour. Mated individuals are represented by solid squares, unmated individuals by open squares, and those of unknown mating status by solid circles. Asterisks indicate individuals that eclosed in the vespiary. Data plotted on the two axes are negatively correlated (Kendall's coefficient of rank correlation, $\tau = -0.4201$, $z = -2.5135$, $n = 19$)

correlation was still significant (Kendall's rank correlation coefficient, $\tau = -0.3866$, $p < 0.05$, $n = 17$).

From the very earliest observations *ad libitum*, it was noticed that oophagy was a regular feature of the colony. A stereotypic sequence of events, shown only by the egg-layers, consisted of cannibalism of an egg, cleaning of the now-empty cell, oviposition, and finally, guarding of the cell for variable periods of time. The number of empty cells in the nest at any time probably constrained oviposition by the majority of the colony members constituting the egg-layers. Oophagy, and less commonly, the destruction of larvae and pupae, could thus be a reflection of reproductive competition in the nest.

The observation sessions dedicated to oviposition and oophagy showed that of the 186 eggs laid during this period, 54 (29%) were eaten. Nest-map records indicated that of the 355 larvae already present on the nest when it was collected as well as those that had developed since, 186 (52%) were cannibalised. Similarly, 27 (22%) of a total of 123 pupae were destroyed or cannibalised.

The observation sessions on oviposition also provided an estimate of how long a freshly-laid egg survived prior to being eaten. A total of 17 eggs were observed to be cannibalised during periods of continuous observation. The time of survival of these eggs ranged from 6 to 223 min, with a median of 40 min. A detailed analysis of successive instantaneous scans and "all-occurrence" sessions also made it possible to examine the length of time an average egg was guarded by the egg-layer immediately following oviposition. Data from 85 instances of cell guarding showed that its duration ranged from 3 to 72 min, with a median of 18 min. Although an estimate of the effectiveness of guarding cannot be obtained from this data, it may be informative to note that of the 17 eggs seen to be eaten, only 3 (17%) were cannibalised within 18 min of being laid.

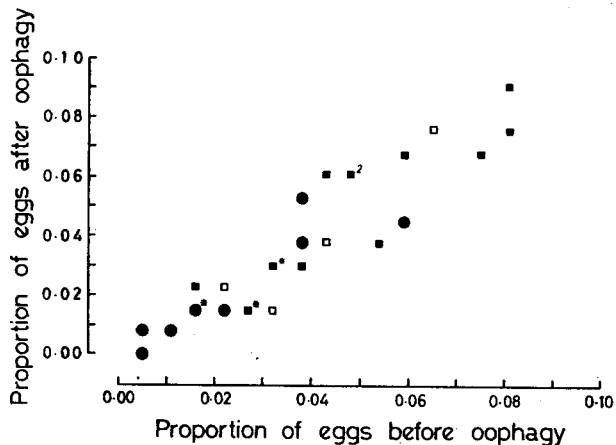


Figure 7. Scatter plot showing the proportion of eggs contributed to the final brood (after oophagy) by an egg-layer as a function of the proportion of eggs originally contributed by her (before oophagy). Mated individuals are represented by solid squares, unmated individuals by open squares, and those of unknown mating status by solid circles. Animals that had eclosed in the vespiary are indicated by asterisks. Two overlapping points, both represented by mated individuals, are indicated as "2". Data plotted on the two axes are positively correlated (Kendall's coefficient of rank correlation, $\tau = 0.8090$, $z = 6.0419$, $n = 28$)

Were eggs of only select individuals being cannibalised? The 54 eggs observed to be eaten during the sessions belonged to 22 of the 28 egg-layers seen to oviposit during this period. In 4 instances, the egg-layer even ate her own egg! During these observations, the number of eggs contributed by the 28 egg-layers (to a total of 186 eggs), before cannibalism, ranged from 1 to 15, with a mean of 6.6 ± 4.3 . After oophagy, the contributions (to the remaining 132 eggs) varied from 0 to 12, with a mean of 4.7 ± 3.4 . Two matrilines, consisting of only a single egg each, were entirely lost. The pattern of contribution to the brood by the colony egg-layers under the two conditions (before and after oophagy) are very similar (Kolmogorov-Smirnov two-sample test, $p > 0.95$); an indication of the absence of selective oophagy. This is further reinforced by the strong positive correlation between the number of eggs contributed by each egg-layer before and after cannibalism (Figure 7; Kendall's rank correlation coefficient, $\tau = 0.8986$, $p < 0.001$, $n = 28$). A positive correlation was also evident between the number of eggs eaten by an individual and the total number of eggs she laid (Kendall's rank correlation coefficient, $\tau = 0.5174$, $p < 0.001$, $n = 22$). This is not surprising since, as already mentioned earlier, the number of empty cells was a possible constraint on the frequency of oviposition by an individual and wasps invariably had to cannibalise eggs in order to lay their own.

Discussion

The most distinguishing feature of *R. rufoplagiata* is that a large number of individuals in the study colony were mated egg-layers. Although several studies have reported polygyny in the independent-founding polistines, most are based on indirect

evidence obtained from the ovarian condition of the individuals (*Ropalidia*: Yoshikawa et al., 1969; Itô, 1985, 1987a, submitted; Yamane, 1986; Itô and Higashi, 1987; Spradbery and Kojima, 1989; *Mischocyttarus*: Litte, 1977, 1979, 1981; Itô, 1984; *Polistes*: Miyano, 1980; Reed et al., 1988). Even in the few cases where egg-laying was observed, it is not clear whether more than one laying female was inseminated (*Ropalidia*: Gadagkar and Joshi, 1982; Yamane, 1986; Itô, 1987b; *Polistes*: Hoshikawa, 1979; Kasuya, 1981). A notable exception, however, is *Belonogaster grisea* where several inseminated females were observed to oviposit (Pardi and Marino Piccioli, 1970).

Several lines of evidence suggest the presence of two distinct behavioural groups of females in *R. rufoplagiata*: egg-layers and foragers. The two groups could be clearly distinguished on the basis of behavioural patterns other than oviposition and foraging. Principal component analysis of time-activity budgets produced two clusters significantly represented by egg-layers and foragers, respectively. The two classes of individuals were, however, morphometrically indistinguishable. Pardi and Marino Piccioli (1981) observed a similar behavioural differentiation in *B. grisea*. Females of one class were more frequently dominant, oophagous, nest-builders, and egg-layers. The other class rarely laid eggs, were never oophagous, and were usually subordinates and foragers. There was, however, a statistically significant difference in the size of the thorax in the two classes of females, that of the egg-layers being larger.

A most interesting finding in *R. rufoplagiata* is the correlation between behavioural differentiation and the age structure of the colony. Younger individuals were usually foragers, while oviposition was confined to most of the older individuals. The only two egg-layers observed to forage as well belonged to the younger group of wasps. Do these two individuals represent the transition stage between the two behavioural clusters? The evidence for a temporal division of labour in this species is remarkably reminiscent of the classic observations of Roubaud (1916) who proposed a similar temporal differentiation of castes in *Belonogaster juncea*, although this has never been subsequently substantiated. In the queenless ant *Pristomyrmex pungens* young adults oviposit, while the older ones forage (Tsuji, 1990). Similarly, younger workers (10 days from eclosion) of the wasp *Polistes jadvigae* perform intranidal work, while middle-aged workers (11–25 days old) are foragers (Tsuchida, 1991). Counterintuitive as it may seem, the involvement of younger animals in the risky task of foraging is not entirely without precedent. Miyano (1980) reported several long-lived workers in a colony of *Polistes chinensis antennalis* that emerged earlier, performed extranidal tasks for about 30 days, then stopped foraging and took charge of intranidal tasks. Although it was not known whether these workers were mated or not, they laid eggs at a rate not very different from that of the queen.

In almost all primitively eusocial species described so far, dominance hierarchies have been considered, and often shown, to have a fundamental role in the reproductive success and division of labour within the colony. In most species, there is a dominant queen at the top of the hierarchy that usually monopolises oviposition and actively prevents all the subordinate females from laying eggs through aggressive interactions (for a review, see Spradbery, 1991). Although there may be situations

when the dominant female loses her position in the hierarchy as a result of the eclosion of a large number of individuals during the late post-emergence phase when several females begin to lay eggs, this is often temporary (Yamane, 1986; Itô, 1987b). Orphaned colonies of some species may also have several egg-laying "workers" (Miyano, 1986; Suzuki, 1987), but a dominance hierarchy again develops with the most dominant workers monopolising oviposition. Even in species where the dominant-subordinate relationships are mild or absent and several females lay eggs (Hoshikawa, 1979; Kasuya, 1981; Itô, 1986), a single female usually has a higher frequency of oviposition than all other colony members. This is also true for the Stenogastrinae where even in the species with multiple egg-layers, there is a clear correlation between dominance status and reproductive capability (Turillazzi, 1991).

R. rufoplagiata presents a completely contrasting picture to this generalised scenario. There is no correlation between the rate at which individuals lay eggs and their position in the dominance hierarchy. Dominance-subordinance interactions were mostly restricted to the egg-layers, the foragers being attacked only rarely. This is in contrast to *B. grisea* where all the oophagous egg-laying females had higher dominance ranks than all the foragers (Pardi and Marino Piccioli, 1970). Several authors have also speculated on the role of mating in determining the dominant status of an individual (Pardi and Marino Piccioli, 1981; Kasuya, 1983). Although most of the egg-layers in *R. rufoplagiata* were mated, this did not influence their rank in the dominance hierarchy.

Differential oophagy has long been recognised a common manifestation of dominance in primitively eusocial wasps with the dominant female(s) eating the eggs of the subordinates (Pardi, 1942; West-Eberhard, 1969; Gamboa et al., 1978; Miyano, 1980; Pardi and Marino Piccioli, 1981; Suzuki, 1987). *R. rufoplagiata* again proves to be an exception in that there was no correlation between oophagy by an individual and her position in the dominance hierarchy. Wasps with higher rates of oviposition, however, ate more eggs, and as far as could be discerned, indiscriminately so. Egg-guarding in this species, never reported earlier for any *Ropalidia*, appears to be a countermeasure against widespread oophagy (see West-Eberhard, 1981). Although its effectiveness in decreasing the probability of an egg being eaten cannot be readily estimated, its importance is evident in that it invariably forms part of the egg-layer's repertoire of oviposition-associated behaviours.

Individuals with higher rates of oviposition appeared to be successful in attaining greater reproductive success since they tended to leave behind more eggs. They had, however, lower ranks in the dominance hierarchy when compared with other egg-layers. The ability of an individual with low rates of oviposition to dominate another with a much higher potential to lay eggs could provide an alternative reproductive strategy for the former in their attempts to prevent the latter from laying eggs. The more active egg-layers, in contrast, possibly maximise their fitness by spending a greater proportion of time in cannibalising eggs and larvae, oviposition, and guarding freshly-laid eggs.

What are the implications of this unique social organisation of *R. rufoplagiata* for the evolution of sociality in wasps? West-Eberhard (1978), in her "polygynous family" hypothesis, envisaged a "rudimentary-caste-containing stage", an evolutionary intermediate between casteless group-living and a stage with a regularly-

occurring worker caste. Although a temporal division of labour was considered one of the possible forms of a rudimentary caste, no example of this had ever been found among the polistine taxa (Carpenter, 1991). *R. rufoplagiata* could represent such a polygynous, rudimentary-caste-containing species. With such extensive polygyny, however, intracolony genetic relatedness would be rather low. Nevertheless, this should not really pose a serious problem for the origin of eusociality. One of the authors (Gadagkar, 1990, 1991 a) has postulated that a "gambling stage", without high relatedness, kin recognition, or manipulation, but where an average member of the group has greater fitness than a solitary individual, may serve as the starting point for the evolution of eusociality. *R. rufoplagiata* appears to be an excellent example of a species at such an initial "gambling stage" of social evolution. This species also illustrates quite clearly the advantage of mixed reproductive strategies, namely working and reproducing within an individual's lifetime, in the early stages in the evolution of eusociality (Gadagkar, 1991 b).

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References

- Carpenter, J. M., 1991. Phylogenetic relationships and the origin of social behavior in the Vespidae. In: *The Social Biology of Wasps* (K. G. Ross and R. W. Matthews, Eds.), Comstock Publishing Associates of Cornell University Press, Ithaca, New York, pp. 7–32.
- Das, B. P. and V. K. Gupta, 1989. *The Social Wasps of India and the Adjacent Countries*. Oriental Insects Monograph, No. 11. Association for the Study of Oriental Insects, Gainesville, 292 pp.
- De Gheff, V. J., 1978. Hierarchical cluster analysis. In: *Quantitative Ethology* (P. W. Colgan, Ed.), John Wiley and Sons, New York, pp. 115–144.
- Frey, D. F. and R. A. Pimentel, 1978. Principal component analysis and factor analysis. In: *Quantitative Ethology* (P. W. Colgan, Ed.), John Wiley and Sons, New York, pp. 219–245.
- Gadagkar, R., 1990. Origin and evolution of eusociality: a perspective from studying primitively eusocial wasps. *J. Genet.* 69:113–125.
- Gadagkar, R., 1991 a. *Belonogaster, Mischocyttarus, Parapolybia* and independent-founding *Ropalidia*. In: *The Social Biology of Wasps* (K. G. Ross and R. W. Matthews, Eds.), Comstock Publishing Associates of Cornell University Press, Ithaca, New York, pp. 149–190.
- Gadagkar, R., 1991 b. Demographic predisposition to the evolution of eusociality: a hierarchy of models. *Proc. Natl. Acad. Sci. USA* 88:10993–10997.
- Gadagkar, R. and N. V. Joshi, 1982. Behaviour of the Indian social wasp *Ropalidia cyathiformis* on a nest of separate combs (Hymenoptera: Vespidae). *J. Zool., Lond.* 198:27–37.
- Gadagkar, R. and N. V. Joshi, 1983. Quantitative ethology of social wasps: time-activity budgets and caste differentiation in *Ropalidia marginata* (Lep.) (Hymenoptera: Vespidae). *Anim. Behav.* 31:26–31.
- Gadagkar, R. and N. V. Joshi, 1984. Social organisation in the Indian wasp *Ropalidia cyathiformis* (Fab.) (Hymenoptera: Vespidae). *Z. Tierpsychol.* 64:15–32.
- Gadagkar, R., K. Chandrashekara, S. Chandran and S. Bhagavan, 1991. Worker-brood genetic relatedness in a primitively eusocial wasp – a pedigree analysis. *Naturwissenschaften* 78:523–526.

- Gamboa, G. J., B. D. Heacock and S. L. Wiltjer, 1978. Division of labor and subordinate longevity in foundress associations of the paper wasp, *Polistes metricus* (Hymenoptera: Vespidae). *J. Kansas Entomol. Soc.* 51:343–352.
- Hoshikawa, T., 1979. Observations on the polygynous nests of *Polistes chinensis antennalis* Pérez (Hymenoptera: Vespidae). *Kontyû* 47:239–243.
- Itô, Y., 1984. Social behaviour and social structure of neotropical paper wasps, *Mischocyttarus angulatus* Richards and *M. basimacula* (Cameron). *J. Ethol.* 2:17–29.
- Itô, Y., 1985. Colony development and social structure in a subtropical paper wasp, *Ropalidia fasciata* (F.) (Hymenoptera: Vespidae). *Res. Popul. Ecol.* 27:333–349.
- Itô, Y., 1986. On the pleometrotic route of social evolution in the Vespidae. *Monit. Zool. Ital. (N.S.)* 20:241–262.
- Itô, Y., 1987a. Social behaviour of the Australian paper wasp, *Ropalidia revolutionalis* (de Saussure) (Hymenoptera: Vespidae). *J. Ethol.* 5:115–124.
- Itô, Y., 1987b. Role of pleometrosis in the evolution of eusociality in wasps. In: *Animal Societies: Theories and Facts* (Y. Itô, J. L. Brown and J. Kikkawa, Eds.), Japan Scientific Societies Press, Tokyo, pp. 17–34.
- Itô, Y. Long-term and short-term polygyny in primitively eusocial polistine wasps: lessons from Australian, Southeast Asian and Japanese *Ropalidia*. Submitted.
- Itô, Y. and S. Higashi, 1987. Spring behaviour of *Ropalidia plebeiana* (Hymenoptera: Vespidae) within a huge aggregation of nests. *Appl. Entomol. Zool.* 22:519–527.
- Jeanne, R. L., 1980. Evolution of social behaviour in the Vespidae. *Ann. Rev. Entomol.* 25:371–396.
- Jeanne, R. L., 1991. The swarm-founding Polistinae. In: *The Social Biology of Wasps* (K. G. Ross and R. W. Matthews, Eds.), Comstock Publishing Associates of Cornell University Press, Ithaca, New York, pp. 191–231.
- Kasuya, E., 1981. Polygyny in the Japanese paper wasp, *Polistes jadwigae* Dalla Torre (Hymenoptera, Vespidae). *Kontyû* 49:306–313.
- Kasuya, E., 1983. Social behaviour of early emerging males of a Japanese paper wasp, *Polistes chinensis antennalis* (Hymenoptera: Vespidae). *Res. Popul. Ecol.* 25:143–149.
- Litte, M., 1977. Behavioural ecology of the social wasp, *Mischocyttarus mexicanus*. *Behav. Ecol. Sociobiol.* 2:229–246.
- Litte, M., 1979. *Mischocyttarus flavitarsis* in Arizona: social and nesting biology of a polistine wasp. *Z. Tierpsychol.* 50:282–312.
- Litte, M., 1981. Social biology of the polistine wasp *Mischocyttarus labiatus*: survival in a Colombian rain forest. *Smithsonian Contr. Zool.* 327:1–27.
- Miyano, S., 1980. Life tables of colonies and workers in a paper wasp, *Polistes chinensis antennalis* (Hymenoptera: Vespidae). *Res. Popul. Ecol.* 22:69–88.
- Miyano, S., 1986. Colony development, worker behavior and male production in orphan colonies of a Japanese paper wasp, *Polistes chinensis antennalis* Pérez (Hymenoptera: Vespidae). *Res. Popul. Ecol.* 28:347–361.
- Pardi, L., 1942. Ricerche sui Polistini. V. La poliginia iniziale di *Polistes gallicus* (L.). *Boll. Ist. Entomol. Univ. Bologna* 14:1–106.
- Pardi, L. and M. T. Marino Piccioli, 1970. Studi sulla biologia di *Belonogaster* (Hymenoptera, Vespidae). 2. Differenziamento castale incipiente i *B. griseus* (Fab.) [sic]. *Monit. Zool. Ital. (N.S.)* 3:235–265 (suppl.).
- Pardi, L. and M. T. Marino Piccioli, 1981. Studies on the biology of *Belonogaster* (Hymenoptera: Vespidae). 4. On caste differences in *Belonogaster griseus* (Fab.) [sic] and the position of this genus among social wasps. *Monit. Zool. Ital. (N.S.)* 14:131–146 (suppl.).
- Premnath, S., K. Chandrashekar, S. Chandran and R. Gadagkar, 1990. Constructing dominance hierarchies in a primitively eusocial wasp. In: *Social Insects and the Environment* (G. K. Veeresh, B. Mallik and C. A. Viraktamath, Eds.), Proc. XI Congr. Int. Union Study Social Insects, Oxford and IBH, Bangalore, p. 80.
- Reed, H. C., J. Gallego and J. Nelson, 1988. Morphological evidence for polygyny in post-emergence colonies of the red paper wasp, *Polistes perplexus* Cresson (Hymenoptera: Vespidae). *J. Kansas Entomol. Soc.* 61:453–463.

- Roubaud, E., 1916. Recherches biologiques sur les guêpes solitaires et sociales d'Afrique. La genèse de la vie sociale et l'évolution de l'instinct maternel chez les vespides. *Ann. Sci. Nat. Zool. (Sér. 10)* 1:1-160.
- Sokal, R. R. and F. J. Rohlf, 1981. *Biometry*. W. H. Freeman and Company, New York, 859 pp.
- Spradbery, J. P., 1991. Evolution of queen number and queen control. In: *The Social Biology of Wasps* (K. G. Ross and R. W. Matthews, Eds.), Comstock Publishing Associates of Cornell University Press, Ithaca, New York, pp. 336-388.
- Spradbery, J. P. and J. Kojima, 1989. Nest descriptions and colony populations of eleven species of *Ropalidia* (Hymenoptera: Vespidae) in New Guinea. *Jpn. J. Entomol.* 57:632-653.
- Suzuki, T., 1987. Egg-producers in the colonies of a polistine wasp, *Polistes snelleni* (Hymenoptera: Vespidae), in central Japan. *Ecol. Res.* 2:185-189.
- Tsuchida, K., 1991. Temporal behavioral variation and division of labour among workers in the primitively eusocial wasp, *Polistes jadvigae* Dalla Torre. *J. Ethol.* 9:129-134.
- Tsuji, K., 1990. Reproductive division of labour related to age in the Japanese queenless ant, *Pristomyrmex pungens*. *Anim. Behav.* 39:843-849.
- Turillazzi, S. and C. Marucelli Turillazzi, 1985. Notes on the social behaviour of *Ropalidia fasciata* (F.) in West Java (Hymenoptera: Vespidae). *Monit. Zool. Ital. (N.S.)* 19:219-230.
- Turillazzi, S., 1991. The Stenogastrinae. In: *The Social Biology of Wasps* (K. G. Ross and R. W. Matthews, Eds.), Comstock Publishing Associates of Cornell University Press, Ithaca, New York, pp. 74-98.
- Wenzel, J. W., 1987. *Ropalidia formosa*, a nearly solitary paper wasp from Madagascar (Hymenoptera: Vespidae). *J. Kansas Entomol. Soc.* 60:549-556.
- Wenzel, J. W., 1992. Extreme queen-worker dimorphism in *Ropalidia ignobilis*, a small-colony wasp (Hymenoptera: Vespidae). *Ins. Soc.* 39:31-43.
- West-Eberhard, M. J., 1969. The social biology of polistine wasps. *Misc. Publ. Mus. Zool. Univ. Michigan* 140:1-101.
- West-Eberhard, M. J., 1978. Polygyny and the evolution of social behavior in wasps. *J. Kansas Entomol. Soc.* 51:832-856.
- West-Eberhard, M. J., 1981. Intragroup selection and the evolution of insect societies. In: *Natural Selection and Social Behavior: Recent Research and New Theory* (R. D. Alexander and D. W. Tinkle, Eds.), Chiron Press, New York, pp. 3-17.
- Yamane, S., 1986. The colony cycle of the Sumatran paper wasp *Ropalidia (Icariola) variegata jacobsoni* (Buysson), with reference to the possible occurrence of serial polygyny (Hymenoptera: Vespidae). *Monit. Zool. Ital. (N.S.)* 20:135-161.
- Yoshikawa, K., R. Ohgushi and S. F. Sakagami, 1969. Preliminary report on entomology of the Osaka City University 5th Scientific Expedition to Southeast Asia 1966 - With descriptions of two new genera of stenogasterine [*sic*] wasps by J. van der Vecht. *Nature Life Southeast Asia* 6:153-182.

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