

Morphological differences between worker-laid eggs from a queenright colony and a queenless colony of *Melipona rufiventris paraensis* (Hymenoptera: Apidae)

by

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ABSTRACT. — Worker eggs laid in a queenless colony of *Melipona rufiventris* were similar to eggs laid by queens. These worker eggs developed into males. The worker eggs laid in a queenright colony had an incomplete reticulate chorion pattern and micropyle. These queenright worker eggs, which had a high tendency to topple over on the surface of the liquid larval food in the brood cell, were always eaten by the queen.

Introduction

The stingless bees (Apidae, Meliponinae) from the tropics are, like the honeybees (Apidae, Apinae), highly social insects. Their social behavior is, however, rather different. One basic difference is the way in which they feed their larvae. Like in solitary and in primitive social bees, brood cells in stingless bee colonies are mass-provisioned before the egg-laying. The provisioning of these brood cells occurs in a peculiar and very characteristic behavioral sequence, in which a small number of workers and the queen cooperate. In quick succession, these workers regurgitate the contents of their honey stomach into the cell for its provisioning with a liquid mixture of honey, pollen and glandular secretions. It is filled to 2/3 of its capacity. The elongated egg is now deposited on the surface of this liquid, on which it typically stands erect. Only the base end of the egg is in contact with the liquid. The cell is sealed immediately after the oviposition, and after 5-7 days the egg hatches. The fact that workers in queenright colonies of stingless bees can lay eggs would appear to be in sharp contrast to the mechanism by which the reproductive dominance of honeybee queens is maintained. The worker eggs (WE) may serve as trophic worker eggs (TWE) that are eaten by the queen or give rise to males (reproductive worker eggs, RWE). Some authors report slight morphological differences in these two types of WE's.

Within the Meliponinae, comprising about 300 species subdivided over the two tribes, Trigoniini and Meliponini, there is a considerable diversity of the phenomenon of worker oviposition (WO) behavior. It is generally accepted that this variation especially occurs among the Trigoni-

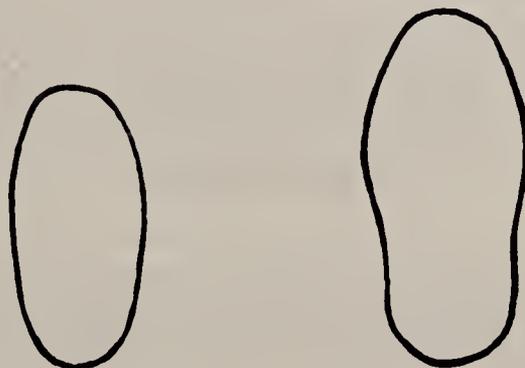
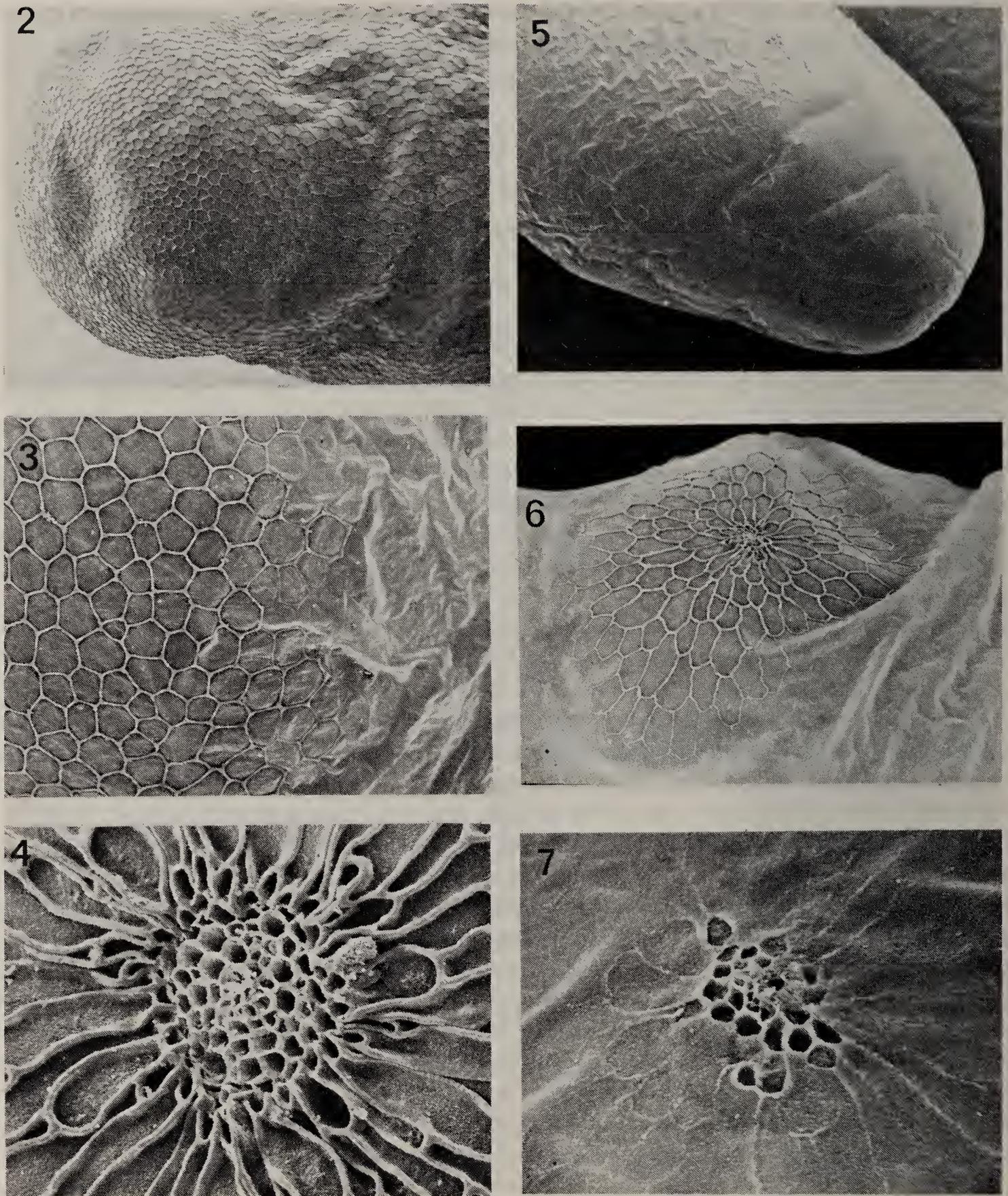


Fig. 1. Typical shape of queen egg (QE), at the right, and smaller queenright worker egg (WE), at the left, of *Melipona rufiventris paraensis*.



Figs 2-7. SEM photographs of eggs of *Melipona rufiventris paraensis*. 2-4. Queen eggs; 2. net-like chorion pattern and the micropyle at the tip of the egg; 3. transition from marked pattern into smooth bottom part; 4. micropyle. 5-7. Queenright worker eggs; 5. absence of distinct chorion marking; 6. reduced network pattern, only on very top, around the reduced micropyle; 7. much more reduced micropyle and hardly any network pattern on chorion. The ruffled appearance of the egg surface, especially of the smooth parts, results from the fixation and dehydration process.

ni, although the data are few and mainly pertain to *Trigona (Scaptotrigona) postica*, which is common in southern Brasil. In this species and other Trigonini, typical TWE's, which are reported to be anucleate and of different form, occur next to RWE's. TWE's are often deposited on the cell margin or even outside the cell. In *Scaptotrigona postica* they are laid before the queen oviposits, while RWE's are here laid after the queen's oviposition and next to the



Fig. 8. A typical brood nest of a *Melipona* stingless bee (*Melipona favosa*). Three horizontal combs are partly surrounded by a waxen „involucrum” on the left. The upper comb has two ready-built brood cells, protruding with their collar above the comb surface. Three large food storage pots, one closed and two open, are partly visible left bottom. The distinct heteromorphic queen is interacting with a worker on the lowest comb.

queen's egg in the same cell (Beig, 1972). In the Meliponini (*Melipona* spp.) WE's are always laid in the same manner as the queen eggs (QE's), i.e. standing erect on the surface of the larval food. The WE's laid before the queen's oviposition are very common in this group; these eggs are always ingested by the queen. It is assumed but not well demonstrated, that in *Melipona* spp. WO also may occur after the queen has laid, just prior to operculation. These WE's would then just as in the Trigonini, result into males. That *Melipona* workers are able to produce males is evident from the occurrence of this form of reproduction in queenless colonies. Since *Melipona* WE's are all deposited in the same way and since it is assumed that they resemble QE's, it has generally been accepted that in contrast to the Trigonini, WE's of *Melipona* are all the same. So WE's laid before the queen oviposits (but these are always eaten by the queen) and those laid during cell closure, as well as those laid in queenless colonies have been assumed to be capable of producing males (Sakagami, 1982).

Experimental

We tried to study the fertility of WE's laid in a queenright colony of *M. rufiventris paraensis*. Although rearing meliponids from the egg stage is in general quite simple (one has only to isolate the sealed brood cells) we were not very successful with worker-laid eggs from our queenright colony. We ascribed our failure to the fact that these WE's failed to remain in the vertical position. They apparently did not possess the surface characteristics of QE's. Therefore, scanning electron micrographs (SEM) were prepared from these eggs, which were compared to those of QE's. Fortunately, we also had available a queenless colony of the same species, where male-producing WE's were laid by the orphaned workers. Such queenless WE's could be studied as well.

Although only a small number of worker eggs were collected for SEM, a distinct variation

between the different WE's was revealed. The queenless WE's were very similar to the QE's. The length of a QE was 2.8 mm whereas two measured queenless WE's had an average length of 2.5 mm. Both were typically skittle-shaped, and stood erect on the surface of the larval food, with only their flattened base in contact with the liquid (fig. 1). Their vertical position appeared very stable. They wobbled when the comb was shaken but always remained in an upright position. The chorion of these eggs had a very distinct reticulate marking (figs 2 and 3). The pattern of this network clearly indicates, like a plaster cast, the former position of the follicle cells, which produced this chorion material in the first place. This prominent network is absent from the flattened basal part of the egg (fig. 3). So only the part of the egg that rests on the liquid larval food has a smooth surface. None of the WE's from the queenright colony had this net-like pattern in full. Some of these eggs only had this marking at the top, while on other queenright WE's this pattern extended further downwards, but never as far as observed in the queenless WE's and the QE's. Some queenright WE's even lacked this pattern completely. The smooth part of the queenright WE's varied, but was always present to a considerable degree. In the eggs we examined the relative absence of the pattern was attended with a less distinct skittle shape. These eggs were then ovoid or subspherical, and had an average length of 2.2 mm. These eggs, although they had been placed in the regular vertical position, were very unstable and tended to fall over on the food surface (when we separated the cell from the comb).

The micropyle of both the QE and the queenless WE also appeared to be very similar. This is a structure of about 60 small openings together forming the tip of the egg (fig. 4). This structure is slightly sunken into the surface of the surrounding chorion. A micropyle was not always present on the queenright WE's. Whenever there was considerable network pattern on the chorion of these eggs the micropyle was quite distinct, but smaller than in the QE's: only up to 20 openings were counted. Eggs with a weak or absent chorion network had a similar weak or absent micropyle (figs 6 and 7).

Discussion

These preliminary results indicate that the WE's laid in the queenless colony, being very similar to QE's and producing males, could be considered as RWE's. The WE's from the queenright colony, showing varying deficiencies in comparison to QE's, could be considered as TWE's. As mentioned above, these eggs are normally eaten by the queen. It is very improbable that these defective TWE's could serve for the production of males, even supposing that they were not all to be eaten by the queen. Their high tendency to topple over on the surface of the larval food makes development very unlikely. However, the degree of the observed morphological differences appeared variable. It is possible that WE's with only minor chorion pattern deviations could have the ability to develop.

We may conclude that the queen of this *Melipona* species exercises control over the reproductive output of her workers by means of influencing the quality of the worker-laid eggs. The precise mechanism of this form of queen dominance is not yet fully understood. The deficient WE may be a result of partial inhibition or deregulation of oogenesis in queenright workers. It is also possible that the queen stimulates the release of immature worker eggs. This latter type of queen control has also been described for the ant *Myrmica rubra* (Brian & Rigby, 1978). Most likely the phenomenon of a gradual differentiation among WE's also occurs in other congeneric species of *Melipona*.

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REFERENCES

- Beig, D., 1972. The production of males in queenright colonies of *Trigona* (*Scaptotrigona*) *postica*. — *J. Apicult. Res.* 11: 33-39.
- Brian, M. V. & C. Rigby, 1978. The trophic eggs of *Myrmica rubra* L. — *Insectes Sociaux* 25 (1): 89-110.
- Sakagami, S. F., 1982. Stingless Bees. — *Social Insects* (H. R. Hermann, ed.) 3: 361-423. Acad. Press, N.Y.

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BALOGH, J. & S. MAHUNKA, 1983. THE SOIL MITES OF THE WORLD, vol. 1. Primitive Oribatids of the Palaearctic Region, 372 pp., 2 krt., 33 tekstfig., 133 pl. (bibliografie 11 pp., index 16 kolommen). — Elsevier Science Publishers B.V., Amsterdam (voor de U.S.A. en Canada: Elsevier Science Publishing Co. Inc., New York), ISBN 0-444-99655-9. Prijs (gebonden) Hfl. 250,— (excl. BTW).

Onze kennis van de Oribatiden (mosmijten) van het Palaearktische gebied is, sinds het verschijnen van de klassieke determineerwerken van Sellnick (1928; supplement 1960) en Willmann (1931) in resp. *Die Tierwelt Mitteleuropas* en *Die Tierwelt Deutschlands*, aanzienlijk toegenomen. De talrijke nieuwe soorten en genera, de geheel nieuwe classificaties, en de zeer uitgebreide literatuur maakten de studie echter, van jaar tot jaar, vooral voor beginners, moeilijker en tijdrovender. Weliswaar publiceerden Kunst (1971) en Ghilarov & Krivolutsky (1975) bewerkingen van de fauna van resp. Tsjechoslowakije (in het Tsjechisch) en de U.S.S.R. (in het Russisch), maar deze boeken waren, ondanks de talrijke illustraties, slechts beperkt toegankelijk, en behandelden bovendien slechts een beperkt faunagebied. Ook met het overzicht van de Oribatiden-genera van de wereld, dat Balogh in 1972 publiceerde, kan men geen soorten determineren.

Met het hier besproken boek van Balogh & Mahunka wordt nu een belangrijke leemte gevuld. Het behandelt, uit het Palaearktische gebied, alle beschreven soorten van primitieve Oribatiden (er volgen nog twee delen, waarin de hogere Oribatiden behandeld zullen worden). Het boek bevat een morfologische inleiding, determineertabellen, diagnoses van supraspecifieke taxa, korte karakteristieken van de soorten (meestal vergezeld van afbeeldingen), literatuurverwijzingen, en opgaven van de verspreidingsgebieden (ook de type-localiteiten worden daarbij genoemd).

De classificatie volgt, in grote lijnen, de moderne inzichten op dit gebied, en hetzelfde geldt voor de gebruikte terminologie. Het boek bevat geen beschrijvingen van de onvolwassen vormen, hoewel deze (bijv. voor de bodemzoöloog) even belangrijk zijn als de adulten. Bij vele primitieve groepen lijken juvenielen en adulten inderdaad veel op elkaar, en levert identificatie geen moeilijkheden op; dit geldt echter niet voor groepen als *Mesoplophoroidea*, *Euptyctima* en *Holosomata*, waar de onvolwassen vormen soms sterk van de adulten verschillen. Hoewel niet van alle soorten de juvenielen bekend zijn (dit geldt vooral voor de *Euptyctima*), had men toch voor iedere familie het nymfen-type kunnen afbeelden. Vooral voor de delen 2 en 3, die de hogere Oribatiden zullen behandelen, zal dit een onmisbaar gegeven zijn; misschien kunnen de auteurs, wanneer zij mijn suggestie volgen, in die delen alsnog een aanvullende plaat met de nymfen-typen van deel 1 opnemen.

Het boek bevat, in beknopte vorm, zeer veel informatie; de uitgave is verzorgd, en de illustraties (merendeels ontleend aan de literatuur) zijn uitstekend. Ongetwijfeld is het een onmisbaar handboek voor ieder die met de determinatie van Oribatiden te maken heeft. — L. van der Hammen.