Behavior Studies of the Stingless Bees, with Special Reference to the Oviposition Process. I. *Melipona compressipes manaosensis* Schwarz[1)2)]

By

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(*With* 3 *Text-figures*)

In a previous paper, one of us (S.F.S.) reported the observations on the provisioning and oviposition process of a stingless bee, *Trigona* (*Scaptotrigona*) *postica* Latreille (Sakagami & Zucchi, 1963). The discovery of a quite complicated behavior sequence in this species necessitated further comparative observations using diverse species. The present study is the first outcome of such work and deals with the observations made on an Amazonian form, *Melipona compressipes manaosensis* Schwarz, together with some miscellaneous observations on the other aspects of behavior within the artificial hive. Some field observations on this subspecies (henceforth called "species" or simply *compressipes*) is reported elsewhere (Kerr *et al.* in press.). The colony used for observations was quite large when received in Manaus, Amazonas. But it dwindled considerably in the course of

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3) At the work, Contracted Professor of Faculdade de Filosofia, Ciências e Letras de Rio Claro, State of São Paulo and Research Fellow (Pr 3720/60) of Campanha Nacional de Aperfeiçoamento de Pessoal de N.vel Superior, Rio de Janeiro.

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transfer into the provisional wooden hive and subsequent transport by air to Rio claro. The population was approximately 120 individuals when introduced to the observation hive (*Cf.* Sakagami & Zucchi, 1963), on February 17, 1963 but decreased to 50 plus 20 introduced workers of *Melipona quadrifasciata anthidiodies* Lepeletier (henceforth abbreviated *quadrifasciata*) at the end of observations on March 14, 1963. Correspondingly the activities of colony were sluggish throughout the observation period.

I. Queen-worker Behavior in Non-oviposition Phase

During the interval between two successive ovipositions, the queen either stays quietly away from the developing comb, often prefering definite places such as below the combs or on hive floor, or walks slowly to and fro within the hive. In the resting stage, her antennae are moved with a moderate rhythm, accompanied by slight but uninterrupted movements of body and legs. The wings are regularly beaten, the time spent for 10 storkes was 5.5 to 7.1 sec. (Mean: 6.53 sec. in 17 measurements). Each beat is a simple stroke without vibration. After resting for an indefinite time, she begins to walk very slowly, continuing the shakings of antennae and beatings of wings. The rhythm of wing beats during walks appears to be slightly slower; time spent for 10 strokes was 6.0 to 10.5 sec. (Mean: 7.06 sec. in 26 measurements). The areas passed by the queen were confined to the nest area alone; that is, because the colony was very weak, combs and storage pots occupied only a small portion of the hive (approximately 30×17 cm^2, 36×32 cm^2 for the whole hive), and she never left the area occupied by the nest elements. The resting and patrolling phases alternate but without definite rhythm nor intervals.

When a worker, walking rather rapidly, encountered the resting queen, the subsequent reaction of the former is nearly invariably a distinct retreat and an escape by a rapid change of direction. But, if the queen continues to rest in a same place, a weak royal court is gradually formed. the number of attendants never exceeded six, but this may mostly be conditioned by the weak colony size. The behavior of attendants does not differ essentially from that found in the honeybee. Each attendant stretches her antennae toward the queen, trembling the body with repetition of rapid advances and retreats. These rhythmical movements are especially conspicuous in the workers standing in front of the queen. Such workers occasionally make short but violent dash to the queen as if attacking her. In such a royal court, the touching the queen with the antennae was always observed, but the licking and feeding of the queen were never noticed.

When the queen is walking within the hive, the attitudes of workers are variable according to the situation. When a worker suddenly encountered the walking queen, the subsequent response is nearly always escape by rapid running (Reaction A). When a worker noticed the approaching queen, one of the following reactions appears: (B) Escape at a slow pace, (C) Orientation to the queen with stretching and trembling antennae, accompanied by gradual retreat, (D) Raising the fore-body slightly, stretching antennae as well fore-legs, and opening mandibles, then giving a rapid dash to the queen as if attacking the latter, (E) Crouching, with antennae withdrawn and legs tigthly in contact to the sides of the body. Often the body is slightly inclined to one side. When such akinetic posture is taken by a worker, the queen often gives rapid antennal touchings to the worker's face (Fig. 1,).

The reactions mentioned above often appear in combinations such as CB, CA, DA, EA and EDA. It appears that Reactions D and E, which are more *elaborated* than B, are usually followed by A.

The queen often eagerly begged food from the workers, and the buccal contact between them was occasionally observed. But true feeding of the queen, as seen frequently in the honeybee colony, was observed only rarely. The duration of feeding was measured in three cases as follows: 5, 8 and 15 sec.

As in other congeneric species, the virgin queens in queenright colonies of this species seem to be killed by the workers soon after emergence. The attack on one virgin queen was observed on Febrary 18, 1963 as follows:

One queen emerged at 9: 20. One worker immediately began to attack her. Thereafter, the virgin queen was continuously attacked by several workers. The latter seized her legs and wings with their mandibles and dragged her in opposite directions. Occasionally the queen escaped and rapidly ran throughout the nest area but was again caught by other workers. At 10: 46 she was already nearly immobile. She had disappeared when the hive was inspected in the afternoon.

Fig. 1. Touching of the face of a crouching worker by the queen.

Throughout, the attacked virgin queen showed no sign of defensive behavior.

II. Behavior in Provisioning and Oviposition Process

In a previous paper, one of us (S.F.S.) divided the oviposition process of

Trigona postica (henceforth abbreviated *postica*) into five sub-processes: 1) Cell construction, 2) Queen Fixation, 3) Provisioning, 4) Oviposition, 5) Sealing or Operculation, and 6) Food intake by the queen. This division, though not always applicable to other stingless bees, can more or less be adopted in the case of *compressipes*.

2. 1. *Cell construction*

Compressipes is a comb maker like all species of the genus *Melipona*. Development of combs is successive, not spiral, and that of each comb is concentric. No systematic observation was carried out on the cell construction, because it is fundamentally the same among all stingless bees. The description given in the previous paper can be applied here without much modification. But the body posture of the bee working in a half-built cell will briefly be described, because the posture is same as the body insertion of workers observed in the queen fixation phase later referred to. Usually the fore-body is at first inserted deeply. The mesosoma is completely immersed in the cell, while the metasoma as well as mid- and hind-legs are vertically oriented. Probably this posture signifies the inspection behavior. Then the worker slightly withdraws the body, which is now supported by mid- and hind-legs, the posterior half of mesosoma is visible and the metasoma is bent down. In this posture, she works with mandibles and fore-legs, often rotating the body. The duration of such insertion is very variable from instantaneous to more than one minute, occasionally mixed with quick deep insertions in the latter case.

The time required for the completion of one cell was not accurately measured but approximately varied from 2 to 4 hours. As in *postica*, each complete cell has a collar, which is quite high, being 1 mm or more above the comb surface, and the upper margin is very thin. *Compressipes* also constricts the cell opening as in *postica* but not so conspicuously as in the latter, retaining a more cylindrical form. There is another more important difference from *postica*. There are always only one or two cells ready to be provisioned. This depends not only on the small colony size but relates to the fundamental difference in the oviposition processes of *postica* and *compressipes* as described subsequently.

2. 2. *Queen fixation*

The provisioning of the brood cell is preceded by the fixation of the queen to the cell, which typically appears as follows: When one or two cells are morphologically ready to be provisioned, a concentration of excited workers begins. They encircle the cell and one of them inserts the fore-body to it. The body insertion is made in the same manner as that in cell-building. But each insertion becomes shorter and is soon followed by another, so that the number of insertions per unit time increases.

Meanwhile, the queen arrives at the comb and inspects the cell by inserting her head, but leaves the cell if the situation, probably the worker excitation, is still not sufficient. After several repetitions of such arrival, inspection and departure, she finally fixes at the side of cell, either sitting on the comb surface or clinging on the outer wall of the cell. She fixes in this position until the beginning of provisioning, or, occasionally leaves the place and walks around on the comb surface or the hive floor nearby but returns and refixes. Her antennae are extended to the cell and alternately touch the cell margins. Occasionally she inspects the cell by the slow insertion of her head. The excitation of the workers around the queen and the cell gradually increases. Their continuous advance-retreat responses to the queen carry an excited tone of rapid rocking movements. The bees run away from the cell after body insertion, though returning soon after. The queen touches these inserting bees violently with her antennae. Finally, the first droplet of larval food is discharged by one of the workers around the cell.

But this sequence, which we called *typical* because it is approximately the same as that found in a moderately strong colony of *quadrifasciata* by one of us (S.F.S.), appeared rather rarely in the colony of *compressipes*, indubitably because of the small colony size. In many cases, the queen fixed on the cell in spite of the lack of the excited alternate body insertions of workers, which developed only later or were not ultimately be evoked. The queen either fixed until the provisioning was complete (*final fixation*) or abandoned the cell to return later. These differences are seen in Fig. 2, which shows the temporal sequence of the provisioning-ovipositing process, with the observational errors of about $\pm$ 0.5 sec. in short term behavior such as each discharge of food and $\pm$ 1.0 sec. in long term behavior. The behavior of queen is shown above, while of the workers below the base line.

In the first case (A, March 6) the fixation was relatively brief; the first provisioning occurred about 55 sec. after the arrival of the queen at 14:30. In that time the gradual increase of rapid alternations of body insertions was recognized. In case B, the queen fixed, after the oviposition, to the second cell, which was morphologically ready to be provisioned, about 8 min. 45 sec. after the beginning of the first fixation. But, in spite of relatively frequent insertions by workers, the general excitement seemed to be insufficient to release the food discharge. After four departures from, and immediate returns to the cell, the queen abandoned the fixation at approximately 7 min. 35 sec. after the beginning of the fixation to the cell. Thereafter, this cell remained without provisioning. At 13: 25, the queen again fixed this cell (C in Fig. 2). (It is uncertain whether or not fixation occurred between 12: 00 and 13: 00 because no observation was made during this interval). The duration between the arrival of queen and the first provisioning was 17 min. 37 sec., with a short interruption from 7 min. 52 sec. to 8 min. 4 sec. In this case, too, the gradual increase of worker insertions was recognized.

Consequently, it is not always easy to give the mean duration of the queen fixation. The relatively well observed cases are summarized in the following table, in which each numeral signifies the duration of fixation (in minutes) and, in parentheses, that between departure from and return to the cell to which the queen

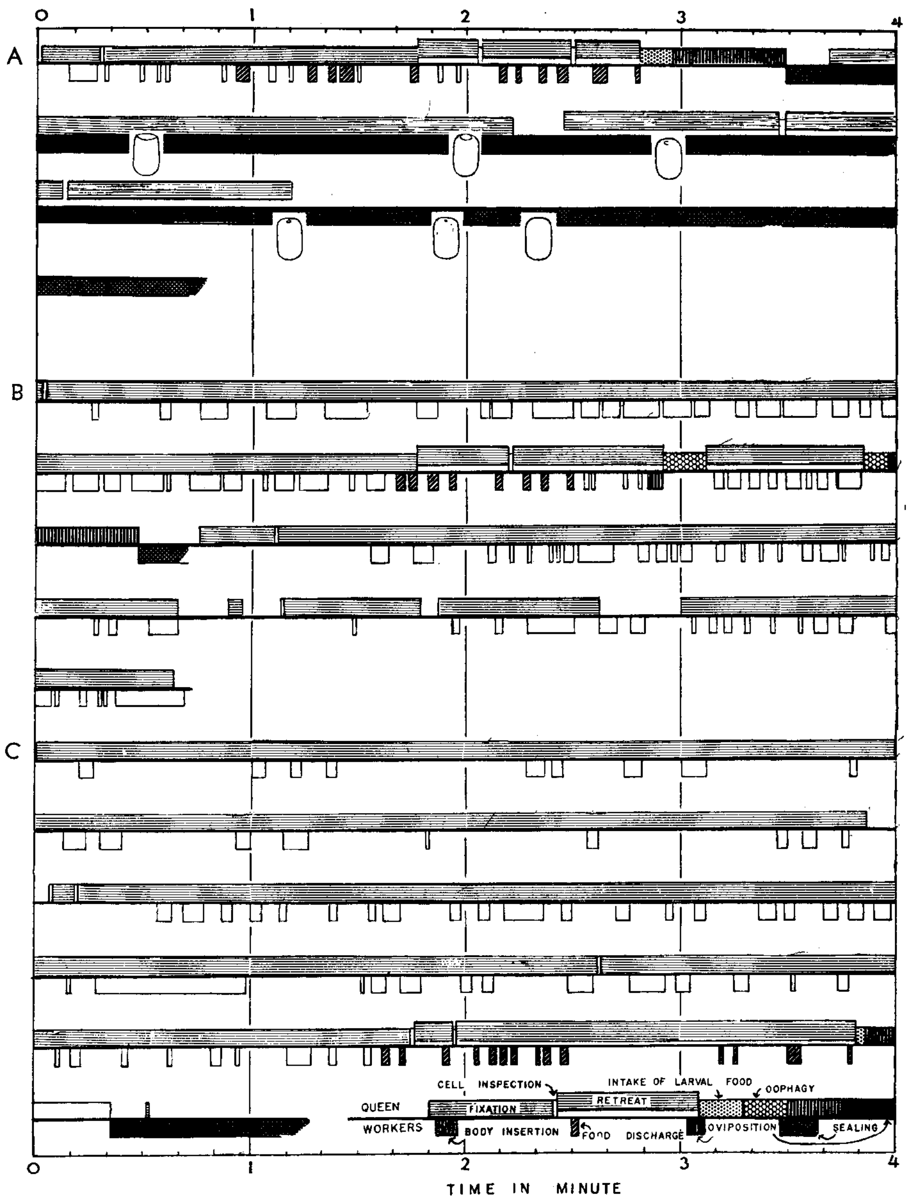


Fig. 2. Temporal sequence in the three cases of provisioning-oviposition process. A. March 6, 14: 30, B. March 4, 11: 40, C. March 4, 13: 25. In each case, the behavior of the queen is expressed above, while that of workers below the horizontal base line. The complete observation of sealing was made only in A, accompanied by diagrams showing gradual reduction of cell orifice. Other explanations are given at the bottom of the figure.

fixed. The figures, except for those for final fixations, are rounded to the nearest minute. The letter X signifies durations of less than one minute and P the beginning of provisioning.

No.	Date	Hour of beginning of observation	Minutes of fixation, and, in parentheses, of absence from fixation	Duration of final fixation	Minutes of total fixation (rounded to 0.5')
1	Feb. 20	15:30	7-(3)-11-(25)................	1'35"P	19.5'
2	〃 21	11:10	0............................	7'10"P	7.0'
3	〃 〃	14:45	X-(78)-3-(53)-13-(X)-2-(9)..	ca.30"P	18.5'
4	〃 22	8:56	X-(3)-X-(X)-X-(X)-X-(5)-10 -(13)-X-(X)................	ca.30"P	10.5'
5	March 3	15:20	11-(7)-1-(X)-1-(4)-3-(2)-14-(20)..	13'P	43.0'
6	〃 4	(Fig.1,B) 10:45	1-(14)-1-(6)-X-(X)-1-(5)-X-(27)..	5'40"P	6.0'
7	〃 〃	(Fig.1,C) 13:25	...... 8-(X)	9'37"P	17.5'+
8	〃 6	12:25	0..............................	14'P	14.0'
9	〃 〃	(Fig.1,A) 14:30	0..............................	55"P	1.0,

The table shows nothing other than that net duration necessary for the fixation varies greatly from case to case, though it seldom exceeds 30 minutes.

The number of body insertions by workers during the fixation is quite variable, but it is more frequent and each act is briefer than during cell building. The frequency distribution per 30 sec. was gathered from all data more or less precisely obtained during the fixation. The results are as follows; O signifies the lack of insertion, X that the insertions continued more than 30 sec.

Number of insertion per 30"	X	0	1	2	3	4	5	6
Frequency observed	17	7	54	53	36	9	3	2

Thus, the most frequent cases were 1–3 insertions per half minute. But these data have no precise significance, because the frequency varies according to the duration of each insertion, which was accurately measured only in certain cases. In these observations, including those shown in Fig. 1, the time spent for each insertion and interval between two insertions in the final fixation phase were:

Time in second

	〈1	1	2	3	4	5	6	7	8	9	10	11-15	16≤
Duration of insertion	3	14	13	19	8	11	4	3	5	3	2	5	1
Interval btw. insertions.	2	1	10	12	8	7	4	3	5	2	3	16	3

From these figures, it can be seen that the duration of each insertion and of the interval between two insertions are relatively short, that is, mostly 1/3 sec. and 2/5 sec. respectively in the final phase of queen fixation.

2. 3. Provisioning and food intake by queen

The posture taken by the workers at each discharge of larval food is similar to that in body insertions during the fixation phase. But, after the deep insertion of the fore-body, the metasoma is contracted cephalocaudally, which is, in typical instances, very distinct because of the narrowing of the distances between each tergal fascia. Thereafter, the worker shows a distinct avoidance reaction, *escaping* from the cell, though not in such an exaggerated manner as in *postica.* The first discharge is sooner or later followed by the second and third ones. After several discharges, the cell is filled about up to 2/3 of its capacity. Then, often short body insertions without metasomal contraction appear. Some of them, especially shorter ones, are apparently mere inspections, while some others might involve the discharges of glandular secretion. For the time being, the segregation of these two possible actions is impossible.

Meanwhile, the queen examines the cell 2 or 3 times, the number being variable according to the length of time from the first discharge to the queen oviposition. Further, there is a trait characteristic of this species, or probably of all species of the genus *Melipona.* When the cell is filled with larval food, the queen, which had hitherto fixed at the margin of cell opening, retreats 0.5 to 1.0 cm from the cell, as if she has a *fear* of the discharged liquid food. The time of retreat is variable in *compressipes.* Sometimes, the queen retreats immediately after the first discharge of larval food, but sometimes later. The retreat can occur after a short inspection of the cell or without such.

In Fig. 2, the retreat is shown by the higher position of the transversly striped bar, which means the fixation of the queen. In case A, the retreat occurred 49 sec. after the first discharge. Before the retreat, five discharges and three body insertions without contraction of metasoma were performed by the workers. In case B, the retreat occurred 49 sec. after, and in case C, 8 sec. after the beginning of food discharge. In both latter cases, the retreats occurred after two discharges, and in case C, after a brief inspection of the cell.

Once retreated, the queen maintains the distance until her oviposition. When she occasionally inspects the cell or takes food from the cell as described below, she advances, performs the act and immediately retreats, again maintaining a definite distance from the cell. Throughout this process of provisioning, the queen continues her regular wing beats. The workers around her and the cell which is being provisioned maintain the excited attending behavior previously described.

The food intake from the larval cell, resported previously in *postica* (Sakagami & Zucchi) was confirmed in *compressipes.* As to the ingestion of larval food, our data are still meagre. As described above, the queen occasionally inspects the cell, inserting her head entirely. This inspection is usually brief. But among 7 cases out of 13, in which the provisioning-oviposition process was more or less continuously observed, the distinct delay of such inspection was observed one in each case. The time spent for these delayed 'inspections' was measured only in four instances as follows: 4, 4, 3 (Fig. 2, C) and 7 sec. (Fig. 2, A). The first three cases are still doubtfully ingestion of larval food. But the last case indubitably involved food intake as shown not only by the delay but also by the movement of queen's mouthparts after she withdrew her head from the cell. Such probable intake occurred in 6 cases immediately before her oviposition, but in once case at the middle of provisioning.

Another, more remarkable event, the oviposition of workers in the cell and subsequent oophagy by the queen, were more precisely observed in 6 out of 13 cases. The results are summarized as follows:

No.	Date	Duration of oviposition and, after hyphens, subsequent oophagy (in sec.)	Total number of eggs laid by workers
1	Feb. 20	?-20, ?-19, 4.5-?, 2.9-25, 4,5-?	5
2	〃 21	5-36, 5-23, 5-30, ?-?, ?-49, 7-23,	6
3	〃 22	4-?, 4-23	2
4	March 4	(Fig2, B) 4-12, 6-8	2
5	〃 6	4-52	1
6	〃 9	7-31, 32-?	2

As seen above, up to 6 workers successively oviposited in one cell and all these eggs were devoured by the queen. In each oviposition, the worker makes a rapid inspection of the cell, then slightly advances and inserts the posterior half of her metasoma. During the oviposition she neither beats the wings nor moves appendages. After oviposition she shows a slight excitation but never so exaggerated as in laying workers in orphan honeybee colonies. Among the cases cited above, one case on March 6, in which the oviposition required 32 sec., was abnormal. The worker attempted to lay, but could not do it. Consequently, she made rotative movements, fixing her metasomal tip in the cell. After two repetitions, each lasting about 10 sec., she succeeded in ovipositing on the final attempt. Excluding this case, the mean duration of worker ovipsition was 4.77 sec. and of queen oophagy 27.0 sec.

The worker-laid eggs are slightly smaller than the queen eggs but of similar form, and are placed, like the queen eggs, vertically on the centre of the surface of larval food except for one instance, in which an egg was accidentally laid on the upper margin of a cell, because it remained at the metasomal tip after the oviposition. Worker-laid eggs were usually devoured immediately by the queen. In some instances, however, there was a short interval lasting to 20 sec. between the worker oviposition and the inspection by the queen. In such cases, the workers encircling the cell often inspected the eggs, but immediately showed an avoidance reaction. Consequently, no worker-laid egg was eaten by the workers before the arrival of queen.

Returning to the provisioning, the frequency of discharges and of body insertions by workers during provisioning, and the total duration of provisioning process, that is, time spent from the beginning of first discharge to queen oviposition, were measured as follows:

No. Date	Number of discharges	Number of insertions	Total duration of provisioning	Duration of provisioning excl. time spent by worker oviposition and queen food intake
1 Feb. 20	7	2	6′49″	ca. 5′14″
2 〃 21	10	0	8′37″	ca. 3′44″
3 〃 〃	6	12	4′53″	ca. 2′03″
4 March 4 (Fig.2,B)	8	11	2′18″	1′50″
5 〃 〃	11	2	1′47″	1′47″
6 〃 〃 (Fig.2,C)	14	0	2′15″	2′13″
7 〃 6	15	0	5′00″	4′30″
8 〃 〃 (Fig.2,A)	11	5	2′02″	1′53″
9 〃 9	10	3	3′37″	ca. 2′07″
Mean	10.2	3.8	6′26″	3′02″

As seen above, all values fluctate considerably from case to case. The relation between the number of food discharges and of mere body insertions, which might be include, as already mentioned, possible discharges of glandular secretion, is very variable. But this is partly due to some observational errors. The metasomal contraction at food discharge is very distinct in typical cases, but in some cases the distinction from the mere body insertion is very subtle. Hence, some insertions counted as discharges could be mere body insertions without metasomal contraction. Generally, discharges concentrate in the earlier half of the provisioning while the body insertions in the latter half, of which the typical appearance is seen in Fig. 2, B. But both can be intermingled throughout the course of provisioning and the very typical discharges can occur near the end as seen in the penultimate discharge in Fig. 2, C.

The durations of discharges and mere body insertions were measured. The interval between two successive discharges is variable. As seen in Fig. 2, there is a tendency for the initial discharges to occur in rather rapid succession, whereas the latter ones often have prolonged intervals, but the tendency is not always definite.

	Time in sec.					
	≦1	≦2	≦3	≦4	≦5	Mean
Frequency of Discharge	16	28	4	4		1.94
Insertion	8	2	4	1	1	2.06

2. 4. Queen oviposition and sealing of cell

The queen oviposition may occur immediately after her food intake or not. If the latter is the case, she rapidly inspects the cell by inserting her head, then crosses over the cell and inserts her metasomal tip in it. The time spent for the oviposition was measured in various cases as follows (in sec.):

19, 20, 26, 26, 30, 35, 38, 39, 41, 50.5, 52, 59 and with two abnormal instances, 105–42 and 32–56. Excluding these two instances, the mean duration is 36.3 sec., that is, distinctly longer than in worker oviposition.

In the two abnormal cases mentioned above, the queen took the laying posture but left the cell without laying. Later she attempted once more and finished the act. The numerals in the data show the duration of the inicial effort and the second successful oviposition respectively. The last instance is interesting and the record is cited herewith. After the beginning of the final fxation at 17: 32 (February 21), the sequence of the process in each 30 sec. was recorded as follows (A: Food discharge, I: Body insertion by workers, o: Worker oviposition, D: Intake of larval food by the queen, E: Queen oophagy, O: Queen oviposition, O': Incomplete queen oviposition):

Time sequence in minute (taken at intervals of 30 sec.)

	0	-	1	-	2	-	3	-	4	-	5	-	6
Behavior sequence	IAA	AO′	AAoEAAA	A	o	—	E	A	DAI	AAAA	AAAAA	ABO	—

The fact that a considerable number of food discharges occurred after the first attempt at queen oviposition suggests the discrepancy between the necessary amount of food and its perception by the queen, having an interesting implication for further analysis of the provisioning process.

The behavior of sealing or operculation of the oviposited cells is essentially the same as that in *postica*. The description given in the previous paper is applied here without modification: The sealing is at first made by a single worker sitting and rotating on the cell. Her mandibles and fore-legs work on the outer surface of the cell, while the metasomal tip is inside (Rotation phase). After the opening is considerably reduced, she changes her position and works at sides of cell opening. In that phase (Side work phase) often another worker momentarily participates in the work. The bee which has worked singly since the beginning of sealing is frequently replaced by another when the opening is completely closed. Thereafter, the work continues for a short time.

As to the duration of sealing, a precise record is reproduced in Fig. 2, A, which shows the gradual reduction of the hole and final closing. The other measurements are given as follows (Time from the beginning of the sealing to the closing of cell opening, plus time from the closing to the departure of the worker) 2′53″+30″, 3′18″+59″, 5′12″+24″, 5′08″+49″, 6′51″+2′22″, 8′08″+1′24″, 9′42″+1′14″ (Mean: 5′48″+1′31″). A quite large variation in the time spent for sealing is partly due to the interference of the queen. In this species, the queen often stays for a whole after oviposition and occasionally touches the sealing workers with her antennae. In contrast to *postica*, the workers of *compressipes* often abandon the sealing work when the queen approaches and return after the latter leaves the cell. Further the sealing does not always begin immediately after queen oviposition but frequently only after a short pause up to 7 sec.

III. Miscellaneous Observations on Behavior Within the Hive

Although systematic observation was not attempted, some miscellaneous data were obtained concerning the behavior other than the provisioning-oviposition process. These fragmentary results are added here, as basic data to the preparation of comparative ethograms of stingless bees in the future.

3. 1. Food intake and related responses

As the colony was very weak, diluted honeybee honey was provided in a small petri dish to freely be consumed by the workers. The feeding behavior differs but little from that of honeybees, obvious movements being extension of the glossa and rhythmic contraction of the metasoma. The time spent for intake varied from 30 to 84 sec. Thereafter, the worker ran to and fro in an excited manner, and either regurgitated the food to her nest-mates or, more frequently, deposited it in a honey pot. In regurgitation, both feeder and recipient usually slightly twist their heads, or incline their bodies in opposite directions. This posture is also observed occasionally in honeybees but is not so conspicuous, while it is seen in all stingless bees observed by one of us (S.F.S.) and can be regarded as a posture characteristic of Meliponini. The deposition of ingested honey in the pot is made by means of 3–4 strong contractions of the metasoma. The deposition of pollen is made in the mode similar to that of the honeybees. The pollen forager examines briefly the pot by inserting the head, then changing her direction, perches on the upper margin of pot, directing her head exteriorly. The pollen masses are brushed off from the corbicular surfaces by middle-legs.

The dehydration of honey (or nectar) was also frequently observed. Its manner is similar to that of the honeybees. The ripening bees stay quietly on the hive floor or walls and their glossa, which is bent below the head, holds a droplet of food liquid and is rhythmically moved. Corresponding to the rhythmic movements, the droplet expands and diminishes. The time required for 10 repetitions of each movement was 4.0 to 6.5 sec. (Mean: 5.8 sec.). This process is often continued more than 30 minutes without interruption.

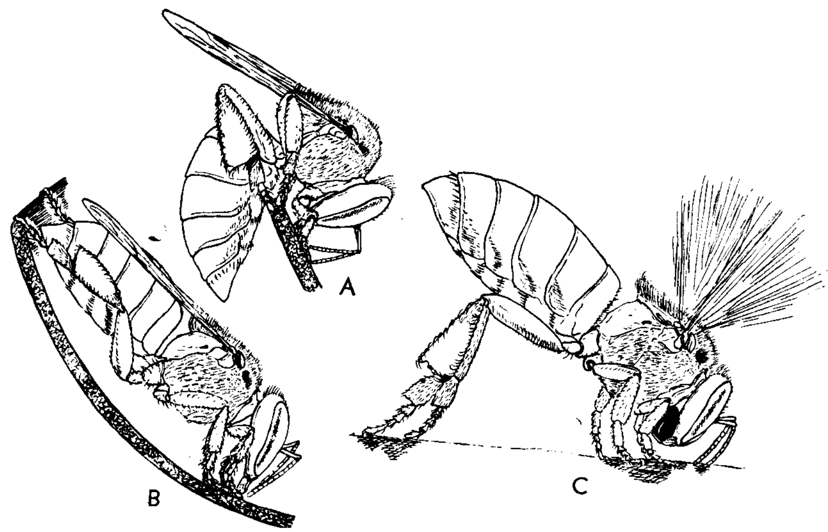
An abnormal aspect of feeding is the ingestion of brood cell contents. The cells already oviposited and sealed are perforated and the contents are ingested. In such a case, the workers show marked attentions to the cells. Several workers encircle the perforation and alternately ingest the contents. Thus, the whole contents are consumed during up to 20 min. This event is observed more or less in diverse groups of stingless bees and becomes frequent when the colonies are weak or food deficient, but it seems to especially be frequent in the genus *Melipona*. Therefore, the brood combs of weak colonies of this genus often show a mosaic appearance of intact and destroyed cells. It was recorded that, one brood cell, being constructed nearly to the level of the comb surface, received a deposit of nectar. In this case further increase of cell height ceased, so that formation of the collar was not observed.

3. 2. Other miscellaneous behavior

Ventilation behavior was frequently observed in the corridor and the area nearby. As in all other species of stingless bees observed by one of us (S.F.S., unpublished), the head is directed, as in *Apis cerana*, toward, not as in *Apis mellifera*, away from the entrance (Sakagami, 1960). The head is lowered while the metasoma is distinctly raised and not markedly bent as in *quadrifasciata*. Thus, the body forms an angle of approximately 30° or more to the substratum. The antennae hang downwards, the fore-legs flex while mid- and hind-legs are stretched. The wings are very rapidly vibrated. Often a dozen of workers stand in a row in corridor, all taking the same posture and continuing the ventilation often more than 40 min. without interruption. At such, many workers also are simultaneously engaged in dehydration (Fig. 3, C).

The manner of pot construction does not differ much from that in other stingless bees. The bees may work on the inner walls, or, in later stages, on the outer walls of the pots. But the most characteristic posture is seen when they work at the pot margins. Often 2–5 workers are seen in perching on the upper margins of a growing pot. They support their bodies with mid- and hind-legs and dash the fore-bodies to the inside. Often they extend the fore-bodies to the centre of pot, holding the bodies only with hind-legs, then retreat to the inicial posture (Fig. 3, A and B). During this retreat, the mandibles and fore-legs participate in the manipulation of the material. The repetition of this behavior is somewhat similar to the rocking movement seen in *Apis*.

Occasionally, *grooming of nest mates* was observed. One worker cleans the face of another with her mandibles, or occasionally crawls over the latter and cleans the mesosoma and wing-bases of the latter as is seen in Apis, both in *A.*



Figs. 3. A and B. Two phases in pot-building movement. C, Fanning posture, accompanied by honey ripening.

mellifera and *A. cerana* (Cf. Sakagami, 1960). In such instances, the bee being cleaned bends her metasoma and shows a weak akinetic tendency as seen in *Apis*, too.

3. 3. Behavior in relation to cohabitants

During the first serveral days of observation, a large pseudoscorpion lived in the hive. It moved freely to and fro within the hive, but often stayed in an empty pot. No worker attacked it. Workers sometimes inspected it with their antennae but did not disturb it.

In order to know the interspecific tolerance, a piece of the comb of *quadrifasciata* was given to the hive on February 21. On the next day, five cells were seen destroyed but some workers of *quadrifasciata* emerged. In such a way, a considerable portion of the cells were destroyed but about 30 workers and some queens and males emerged. The males, queens and many workers of *quadrifaciata* were attacked, killed and eliminated from the hive by the workers of *compressipes* immediately after emergence. Some workers of *quadrifasciata*, however, passed this period and survived within the hive until the end of observations (The oldest ones were 26 days old after emergence). The hostile attitude of *compressipes* to *quadrifasciata* showed a marked individual difference. In one instance, it was observed that a *compressipes* worker helped the emergence of a *quadrifascitata* worker from the cell. On the other hand, it was occasionally observed that some *quadrifasicata* workers, which lived more than 10 days within the hive in safety, received sudden attacks of some *compressipes* workers after being treated rather *friendly* by several *compressipes* workers. The manner of attack of *compressipes* does not differ from that which they use to attack their own virgin queens. In any case, the attacked *quadrifasciata* showed no sign of defense, bending their metasoma slightly and demonstrating an akinetic posture. After escape, they usually ran around the hive in an excited state. The attitude is therefore similar to that shown by attacked *Apis* (Sakagami, 1959), though the tongue stropping, characteristic of the latter genus, was never observed.

Regurgitative food transfer between the workers of the two species was occasionally observed in both directions. Further, the feeding of the *compressipes* queen by a *quadrifasciata* worker was also observed on March 4. In such instances, there was no behavioral difference from the homospecific cases.

The participation of the adopted *quadrifasciata* in the colony activities was seldom observed, limiting our attempt to reinforce the *compressipes* colony by them. Even sufficiently aged individuals of *quadrifasciata* with completely black mesosomal hairs spent most of their time on the combs, involucrum or hive floor in sluggish resting state. The only activities they participated in were the construction and closing of pots and the improvement of pillars and involucrum, which were done with a very *lazy* manner.

Occasionally, a few *quadrifasciata* workers stayed on the comb during the oviposition process. Most of them were indifferent to the event or only passively moved because of the excited traffic of *compressipes* workers, nevertheless all of them sufficiently aged to act as brood-carers (10–20 days old, *Cf.* Kerr & Santos Neto, 1956). Only in one instance was the body insertion of a *quadrifasciata* worker into the cell observed immediately before the provisioning in front of the *compressipes* queen.

Remarks

As the present paper is the first report of serial works on the behavior of stingless bees, some general remarks applicable to all subsequent papers are given here.

The discovery of an elaborated behavioral sequence in the oviposition process of *postica* stimulated one of us (S.F.S.) to further observations of diverse species. These observations revealed that the behavioral sequence in the process varied remarkably among species, or, probably, among supra-specific categories. In this and subsequent works, attention was focused on recording the probable behavior characteristics in each species, especially those concerning the oviposition process. Thus, the observations on the functional aspects of the colony dynamics were not systematically attempted.

In our serial publications, the results will be compared only to those already published by one of us (S.F.S.) and his collaborators. Furthermore, comparisons will mention only important differences. Sometimes, comparison might, if necessary, be made using still unpublished data. But the synoptic perspective will be given only after all data have been published. There are some contributions by earlier authors on the behavior of stingless bees within hive, with special reference to the oviposition process and queen-worker relationship (*Cf.* Sakagami & Zucchi). The comparion between our own data and these previous records will also be given at the end of our serial works, mainly to avoid a complication due to troublesome cross-reference.

A remark must be made as to the stability of the observed behavior characteristics. In any behavioral sequence, we can notice some aspects that are relatively stable in each species, and these that are quite variable according to the conditions, both external and internal. In stingless bess, the colony size, quantity of food storage, presence or not of queen, weather contiditons and co-habitants, especially the phorid flies, could be counted the chief controling factors. It is not always easy to separate constant and variable components. Occasionally, a careful comparison among data, using many colonies and observing under diverse conditions, are required to obtain accurate results. Unfortunately, such closer studies using more than one colony could not be made because of the lack of material and time. Nevertheless, we have a conviction that some, probably, many characteristics discovered by us are really of specific nature for the following reasons: 1) In some closely studied species such as *postica* and *quadrifasciata*, some characteristics are variable according to the situations, but others are well fixed irrespective of environmental and colony conditions. Such difference could be applied to other species which were not observed in detail. 2) Some characteritics appear among allied species unmodified or only slightly modified. In such cases, we can apply the principles of homology, though in a careful manner (*Cf.* Baerends, 1958).

But the probability of misinterpretation may always be higher than zero, and be quite high in some characteristics, especially in quantitative ones. We shall indicate when there are some doubts on the stability of observed differences. But further critical observations are always necessary. The purpose of our comparative work is merely to set a small path in a promising but hitherto poorly explored field.

Returning to *compressipes*, the aspects common to this species and *postica*, and those remarkably different between them are summarized below. The differences 2), 3), 5), 6a), 7) 8) and 12) are of particular importance:

1) The behavior patterns of the queens in the resting or patrolling state are similar in both species.

2) The attitude of workers to the queen is in general the same. However, royal attendants of *compressipes* do not show the dashing movements so exaggeratedly as in *postica*. Especially, wing beats are never observed. The touching of the face of crouching workers by the queen does not occur in *postica*.

3) Virgin queens of *postica* are not killed by the workers so soon as in

compressipes after emergence.

4) In both species, the cells ready to be provisioned have distinct collars. The opening is narrowed, but more conspicuously in *postica*.

5) The provisioning process begins in *postica* when 10–20 cells become ready to be worked, whereas in *compressipes* each process deals with one cell alone. Correspondingly, *postica* has two peculiarities which are logically unexpected in *compressipes*; a) The queen of *postica* often changes the cell to which she fixes. b) In the ealirer phase of the process, one or two cells receive a long fixation of the queen, together with the prolonged agitation by workers before and even after the food discharge. On the other hand, in the later phase, each cell receives nearly simultaneously the food discharge and the oviposition proceeds very rapidly.

6) The fixation of the queen of *compressipes* differs from that of *postica* as follows: a) Once fixed, the queen does not move to and fro in front of the cell, b) Her mandibles are usually closed, c) Workers around the cell and the queen do not exhibit much exaggerated *ceremony*.

7) After the discharge of larval food, the queen of *postica* does not retreat from the cell as in *compressipes*. On the other hand, the excitation of workers during the provisioning is far more intense in *postica*. Some very impressive attitudes in the provisioning phase of *postica*, such as the violent running around of many workers on the comb surface and the occupation of the cell by one worker which gives a dashing movement to the queen as if threating the latter, never appear in *compressipes*. The *escape* from the cell after discharge of food is seen in both species, but in a more exaggerated form in *postica*.

8) The interval between the first food discharge and queen oviposition in *postica* is probably longer (the measurement are still not well put in order) in the earlier ovipositions, but distinctly shorter in the later ovipositions than in *compressipes*.

9) The queen oviposition of *compressipes* is more prolonged than in *postica*.

10) The sealing of oviposited cells is made in the same manner in both species. The work is done in *postica* with a greater consistency.

11) The ingestion of larval food is seen in both species, though the data are still insufficient in *compressipes*.

12) Worker oviposition and subsequent queen oophagy are seen in both species. In *postica*, the worker eggs are usually larger and more rounded than the queen ones, and are placed on the upper margins of the cells. In *compressipes*, however, the worker eggs are slightly smaller than, and similar in form to those of the queen. Further they are laid vertically on the centre of larval food like those of the queen.

The comparison of *compressipes* to the other observed species of the genus *Melipona* will later be given in another paper. But a brief comment on the differences between *compressipes* and *quadrifasciata* is here presented, based upon the unpublished data obtained by one of us (S.F.S.). The behavior patterns of both species are very similar. The important differences are synoptically given as follows:

	quadrifasciata	*compressipes*
1) Wing beats in queen	absent	present
2) Touching of crouching worker by queen	Frequent, directed to occiput of worker making a bow	Relatively rare, mostly directed to face of worker which is not making a bow
3) Excitement of worker before queen fixation	Constant	Inconstant, often queen fixing without excitation
4) Queen fixation	Uninterrupted and relatively short	Often interrupted and usually prolonged in total duration
5) Food discharge	Successive	Often intermittent, mixed with mere body insertions
6) Duration of queen oviposition	long	very long
7) Delay of sealing after oviposition	very often	occasionally
8) Number of worker oviposition to each cell	0–1, very rarely 2	0–6
9) Interval between two queen ovipositions	long but occasionally short	long

Among these differences, items 1) and 2) seem to be specific to each. Item 7) is still difficult to give a precise decision. On the other hand, all other items are probably caused by the different conditions of colonies observed. Perbaps the colony size of *compressipes* was too small to evoke a sufficient amount of excitation of workers after the cells became morphologically ready to be oviposited (Item 3). Consequently, the queen often interrupted the fixation, which resulted in the prolongation of total duration (Item 4). The poorly organized food discharges (Item 5) and the absence of successive oviposition by the queen (Item 9) are also explained in the same way. The limited cell construction might produce excessive egg formation in worker ovaries, which resulted in the numerous worker ovipositions into one cell (Item 8). As already described the miscarried ovipositions by the queen were twice noticed. Therefore, it is very probable that the prolonged queen oviposition (Item 6) was caused by some functional defect in the ovipositing apparatus, though it is certain that the qeen oviposition in the genus is much more prolonged than in other stingless bees.

A few words must be added as to Item 9. In *quadrifasciata*, two queen ovipositions occasionally occurred in succession with an interval of 5–10 minutes

(Exact data are still not put in order). But this does not signify that the oviposition process of this species approaches the type of *postica*, because each of successive oviposition in *quadrifasciata* must be repeated in the same manner, without the synchronization of provisioning or the abbreviation of prolonged queen fixation as in *postica*.

Summary

In the present paper, some behavior characteristics of *Melipona compressipes manaosensis* Schwarz were described and compared to those of *Trigona* (*Scaptotrigona*) *postica* Latreille already reported upon by one of the writers. The main differences of *compressipes* from *postica* are: 1) In each oviposition, only one cell is worked out, 2) The fixation of the queen and excitation of the workers are less exaggerated, 3) The queen shows a peculiar retreat reaction after the food discharge to the larval cell by the workers. 4) The worker oviposition in the cells is also seen, but the eggs are smaller than those of queens and laid in the normal position. A preliminary comparison of the behavioral differences between *compressipes* and *Melipona quadrifasciata anthidioides* Lepeletier and some general remarks on this series of papers were also given.

References

Baerends, G.P., 1958. Comparative methods and the concept of homology in the study of behaviour. Arch. Néederland. Zool., **13**: 401–417.

Kerr, W.E. & G. dos Santos Neto, 1956. Contribuição para o conhecimento da bionomia dos Meliponini, V. Divisão de trabalho entre operárias de *Melipona quadrifasciata quadrifasciata* Lep. Insectes Sociaux, 3: 423–430.

Sakagami, Sh, F., 1959. Some interspecific relations between Japanese and European honeybees. Studies on the Japanese honeybee, *Apis cerana cerana* Fabr., VI. J. anim. Ecol., **28**: 51–68.

——— 1960. Preliminary report on the specific difference of behaviour and other ecological characters between European and Japanese honeybees. Studies on the Japanese honeybee, *Apis cerana cerana* Fabr., V., Acta Hymenopterologica, **1**: 171–198.

——— & R. Zucchi, 1963. The oviposition process in a stingless bee, *Trigona* (*Scaptotrigona*) *postica* Latreille. Studia Entom., Petrópolis, 6: 497∼510.