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DIVISION OF LABOUR WITHIN ONE AGE GROUP OF HONEYBEE WORKERS (*APIS MELLIFERA*)

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Résumé:
Division du travail chez des ouvrières d'*Apis mellifera* appartenant au même groupe d'âge

200 ouvrières, fraîchement écloses, furent marquées et introduites dans des ruches d'observation. Leur comportement fut étudié pendant les semaines qui suivirent. Une spécialisation s'établit parmi ces ouvrières pour: 1) la construction du rayon; 2) l'"allogrooming" (ce comportement n'est pas orienté vers un groupe particulier d'ouvrières); 3) le comportement agressif (ce comportement est nettement orienté vers un groupe spécial d'ouvrières). Il n' y a pas, en revanche, de spécialisation pour: 1)le comportement de cour et 2) l'approvisionnement des cellules royales.
Mots clés: *Apis mellifera*; comportement; division du travail; spécialisation.

Summary

200 individually tagged callow workers were introduced into observation hives after which their behaviour was studied during the following weeks. Within this age group there appeared to be specialists for: 1) construction of the comb; 2) allogrooming (this behaviour was not directed to a certain group of workers in particular); 3) aggressive behaviour (that was clearly directed to a special group of workers). No specialization seemed to take place for: 1) presence in the queen's court and 2) queen cell provisioning.
Key words: *Apis mellifera*; behaviour; division of labour; specialization.

INTRODUCTION

Division of labour in honeybee colonies has been subject of a great number of studies. Until now most of the attention was directed to age dependent factors of the workers under study (e.g. Winston & Punnett, 1982). Significant differences in the frequency with which behaviour is performed on average by workers of different age groups have led to well known schemes of age dependent division of labour (Michener, 1974). Lindauer (1952) stressed that there exists a considerable individual variation in the age at which certain behaviours are started.

However, for most behaviours of honeybees, it is still unknown whether within bees of the same age specialism takes place, or if there is only a difference in involvement based on coincidence. In this study, individual involvement in a number of behaviours in both the queen-right and the queen-less situation was

examined. These behaviours are:
(*queen-right* situation:)
- **queen attending** (i.e. presence in the queens' court);
- **construction of the comb** (i.e. manipulation of wax);
- **to allogroom** a nest mate; - **to be allogroomed** by a nest mate;
(*queen-less* situation:)
- **to provision a larva in a queen cell;**
- **to be aggressive**; - **to be treated aggressively.**

MATERIAL AND METHODS

Two-frame observation hives were installed with a small colony of about 1500 workers and a queen, but without any brood. After about a week we introduced 200 individually tagged teneral workers. Behaviour of these workers was observed daily during periods of 3 to 4 hours of continuous observation. After 8 to 10 days the queen was taken out of the hive in order to be able to study behaviours in queen-less conditions. At about the time when the first egg-laying workers appeared (about 14 days after the queen was removed), the whole colony was placed into a deep-freezer in order to be able to examine the ovarian development of all marked workers. These results are published elsewhere (Van der Blom, in prep. A). During the experiment we tried to keep count of the mortality of marked workers as accurate as possible.

The different behaviours were scored as follows:
- **Queen attending.** Workers at a position less than 1 cm away from the queen for at least 10 seconds, with their body axis in her direction, were considered to participate in the court. The marked bees performing this behaviour were scored every two minutes separately. The total number of two-minute-scores was considered as a measure of time; the number of different times a worker participated in the court was also taken as a measure for analysis (without regarding the amount of time spent there).
- **Construction of the comb.** All workers seen to manipulate wax for the construction of the comb or for cell capping were scored per 10 minute intervals. The number of these intervals during which the individuals were scored was used for analysis.
- **Allogrooming.** The hives were regularly scanned for the occurrence of allogrooming. All interactions in which marked workers were involved were noted.
- **Provisioning of queen cells.** All marked workers that were observed to enter into queen cells far enough to reach the larva at the bottom were scored if they stayed inside for at least 5 seconds. Workers were only counted again at least five minutes after a previous body insertion.
- **Aggression.** Agonistic interactions, which are frequent in the queen-less situation, were scored as often as possible as far as marked individuals were involved.

Since we collected data for all individuals, we could make frequency distributions for all behaviours that were studied. Each of these was compared with a Poisson distribution, calculated on the basis of the average value for all workers and the number of workers present (Fig. 1). The Kolmogorov-Smirnov test was applied to compare the two distributions. This indicates whether the observed variation in involvement can be explained by ran-

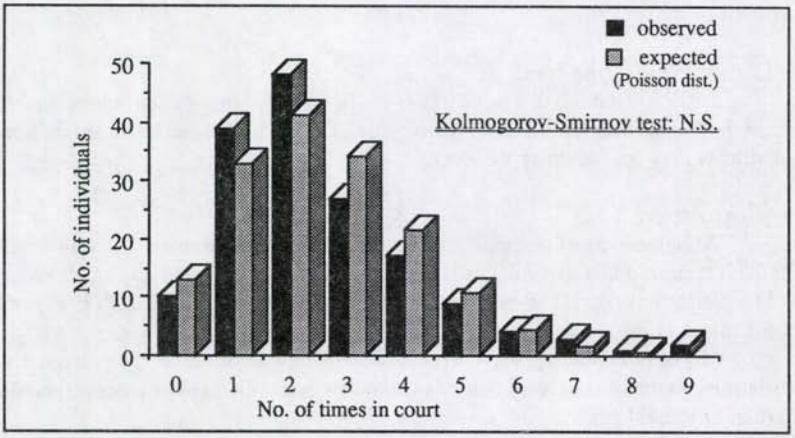

Figure 1. *Example of an observed frequency distribution (Queen attending, Hive 5) and the calculated Poisson distribution*
Figure 1. *Exemple de fréquence de distribution (Soins à la reine, ruche 5) et calcul de la distribution de Poisson.*

dom factors only (no significant deviation from the Poisson distribution), or if specialization takes place (significant difference, i.e. a limited number of individuals perform a certain behaviour more often than can be expected purely by chance. It is known that this test is very conservative for this analysis, so we find less significant differences (specialization) than with other, e.g. Chi-square, tests).

RESULTS AND DISCUSSION

- Queen attending

The observed frequency distributions of the number of times that the individuals participated in the court does not significantly deviate from the calculated Poisson distributions (Fig. 1; Table 1). These results are consistent over all four observation hives in which this behaviour was studied. This means that there seem to be no workers who specialize in this behaviour. We thus agree with Seeley (1979) who had similar results on a more limited scale. A more detailed analysis on this behaviour is in preparation (Van der Blom, in prep. B).

Usually, workers were observed in the court only as long as the queen remained still on one spot. The composition of the court changed as soon as she started to move. Workers hardly followed her (as was also mentioned by Butler et al., 1973). Time spent in the court thus seems to reflect the activity pattern of the queen rather than characters of individual workers. Since workers in the court are supposed to play an important role in the dispersal of the queens' pheromones (Seeley, 1979; Velthuis, 1972), also this seems to depend largely on the queens'

pattern of movement.

- Construction of the comb

Sufficient data on this behaviour could be collected in only one colony. In this case, both distributions differed significantly (Table 1), so it can be stated that individuals may specialize as 'builders'.

-Allogrooming

Allogrooming of nest mates was examined in three colonies (Table 1). In all three it appeared that grooming was performed mainly by a relatively small group of specialized workers. This was described earlier by Frumhoff & Baker (1988) and by Kolmes (1989).

'To be groomed' appeared to occur completely according to the Poisson distribution, so that it is evident that this behaviour is not directed towards a specific group of individuals.

Table 1.

Queen attending (No. of times in court)

Hive	av./worker	|Dmax|	P	Sign.
1	2.30	.0969	> .2	N.S.
2	1.69	.0627	> .2	N.S.
4	1.86	.0431	> .2	N.S.
5	2.50	.0624	> .2	N.S.
Comb construction (No. of 10 min. scores)				
4	1.99	.2660	< .01	**
Grooming nest-mates (No. of interactions)				
1	1.04	.1949	< .01	**
2	1.82	.2523	< .01	**
5	.775	.2080	< .01	**
Being groomed (No. of interactions)				
1	.518	.0231	> .2	N.S.
2	1.13	.0454	> .2	N.S.
5	1.12	.0641	> .2	N.S.

- Provisioning of queen cells.

As Lindauer (1952) pointed out for worker larvae, a very great number of workers is involved in the nourishment of queen larvae. Two out of three queen cells present in colony 5 were kept under constant observation during 8 hours while they contained larvae. During this time more than 50% of the marked workers entered into both of the cells,

Table 1: Queen-right situation
Results of the Kolmogorov-Smirnov (one sample) test:
Hive = No. of the observation hive; Av./worker = Average No. of observed behavioural acts per marked worker; |D max| = K-S test statistic
Tableau 1: Situation en présence de reine. *Résultats du test de Kolmogorov-Smirnov (un échantillon):*
Hive = numéro de la ruche d'observation; Av./worker = nombre moyen d'actes comportementaux observés par ouvrière marquée; |D max| = test statistique K-S.
*Significances: P >.05 = N.S.; .05 > P > .01 = *; P < .01 = ***

whereas the marked workers on the whole represented only about 30% of all workers that entered into the cells. This means that we can estimate that the cells were visited by at least 300 different individuals during the observation hours only.

The frequency distributions of this behaviour do not significantly deviate from the Poisson distribution (Table 2), so we may conclude that there is no specialization for the raising of new queens.

However, we have to be very careful while considering these data. Inside the queen cell, workers may do all kind of different things apart from depositing food. They may just inspect the contents of the cell, work on the construction (Lindauer 1952) or maybe even eat from the food present in the cell. We could not discriminate between these behaviours during our observations.

Aggression.

Aggressive behaviour may cause mutilations, or even the death of the victim. It is performed by a very small percentage of the workers. Within the marked age group usually only very few individuals were responsible for all agonistic interactions observed. As a result, the frequency distribution very significantly differs from the calculated Poisson distribution in all four of the colonies from which data on this behaviour were collected (Table 2).

The outcome of the observations on the workers that receive aggression seems to be less consistent over the four colonies (Table 2). In three of these, we found significant deviations from the Poisson distribution (although in one of these the significance was weak), while in one, colony 2, we did not.

This may be explained by the results of the fourth colony (Hive No. 6). The time during which aggression was scored was divided into three different periods, and the normal procedure was followed for these periods separately (Table 3).

The results from the first week during which aggression was seen do not show a deviation from the Poisson distribution. Thus, in the first week aggression does not seem to be

Table 2.

Provisioning of queen cells (insertions)

Hive	av./worker	|Dmax|	P	Sign.
4	0.90	.0490	> .2	N.S.
5	2.41	.1139	< .1	N.S.
Being aggressive (No. of interactions)				
2	.383	.2781	< .01	**
3	.578	.3390	< .01	**
5	4.06	.8441	< .01	**
6	2.19	.2875	< .01	**
'Victim' of aggression (No. of interactions)				
2	.584	.1062	< .1	N.S.
3	1.13	.2779	< .01	**
5	5.00	.1275	< .05	*
6	2.63	.2417	< .01	**

Table 2: Queen-less situation
(legend as in Table 1)
Tableau 2: Situation sans reine
(légende comme dans leTableau 1)

Table 3. 'Victim' of aggression

First week

Hive	av./worker	|Dmax|	P	Sign.
6	.216	.0188	> .2	N.S.
Second week				
6	.662	.1604	< .01	**
Last two days				
6	1.331	.3460	< .01	**

Table 3. Victims of aggression, Hive 6 (legend as in Table 1)
N.B. Since during this period a number of the marked individuals died, the sum of the averages per worker from this table does not equal the average No. per worker over the whole period as indicated in Table 2.

***Tableau 3. Victimes de l'agression, Ruche 6** (légende comme dans le Tableaux 1). N.B. Comme durant cette période, plusieurs individus marqués sont morts, la somme des moyennes, par ouvrière, dans ce tableau n'égale pas le nombre moyen pendant la période totale (indiquée sur le Tableau 2).*

directed towards a certain group of individuals. However, in the second week this changes and we find an even more significant deviation during the last two days.

This can be understood if we take the development of laying workers into consideration. It is known that aggression is directed mainly towards individuals with a high degree of ovary activation (Velthuis, 1976). In the first queen-less period, there will not yet be a very clear differentiation between the workers, because there are no individuals with ovaries in highly advanced stages of development. In the later stages, closer towards the first moment of egg laying by workers, there will be a small group of workers with nearly mature eggs inside their ovaries. They will receive almost all the aggression in the colony. In our colonies we found that the workers that were actually killed by their nest mates were always workers with a high degree of ovary activation.

In colonies 2 and 5 we finally found that ovaries of the workers were activated to a lesser extent in colonies 3 and 6. This means that the period during which data were collected in the colonies 2 and 5 can probably be compared to the first period in colony 6 (Table 3), where we did not find a significant deviation from the Poisson distribution.

GENERAL REMARKS.

It is obvious that for some behaviours we found a clear kind of specialization and for others we did not. However, if we did not find specialization for a behaviour with this method, this does not necessarily mean that there are no differences between the workers. Although the involvement in a behaviour shows a random fluctuation, it may be that the cause of this is to be found in stochastic intrinsic characters of the workers (=intrinsic differences). This may lead to a certain level of performance of the behaviour, which is relatively independent of external factors. (In fact, we should say that in this case there is a kind of specialisation, based on intrinsic

stochastic characters. An example of this can be found in Abraham et al. 1984).

Furthermore, it may be that workers who find themselves at one of the extremes of a distribution become different from the rest because of that. E.g. it is possible that workers who are in the presence of the queen for a long time receive more pheromones than other workers. This may change the position of such workers in the group, although the process on which this is based is random. A further analysis on this is reported elsewhere (Van der Blom, in prep. B).

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