THE EFFECT OF THE NUMBER OF QUEENS OF THE ANT *LASIUS FLAVUS* (FAB.) (HYM., FORMICIDAE) ON THEIR SURVIVAL AND ON THE RATE OF DEVELOPMENT OF THE FIRST BROOD

by

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INTRODUCTION

There is generally only one queen in well established nests of the underground yellow ant *Lasius flavus*, but nests with several queens are also recorded. DONISTHORPE (1927, p. 260) found two, three and nine queens in populous colonies and recently I have come across nests with more than one queen on two occasions. My observations on *Lasius flavus* have been made at the Imperial College Field Station at Sunninghill, Berkshire, where the soil is of light sandy—gravel with local mixtures of clay and the vegetation is of the acid—grassland type. There are extensive areas with the nests of *L. flavus*, with some of the larger less disturbed hills approximating to 30 cm. in height. In one of these areas a populous nest 15 cm. high was dug up on 26.IX.52 and 2 ♀ were found under a stone; another nest 15 cm. high and 45 cm. in diameter dug up on 6.VII.53 had several thousands of workers, many pupæ and sexual pupæ and three ♀ in a cell under a group of small stones 10 cm. below the surface. These queens had large distended gasters, appeared worn and were far less mobile than the young queens.

Although the occurrence of several queens in well established nests of *L. flavus* must be considered as rare, cooperation between groups in nest—foundation is not uncommon and numerous records have been listed by DONISTHORPE (1927), EIDMANN (1931) and WASMANN (1910). There appears to be no adult reproductive diapause, or if it exists it is of a very short duration, as the first batches of eggs in the artificial nests at room temperature were usually laid 2-4 weeks after flight and DONISTHORPE (1927, p. 259) states that the first eggs are laid in September. In 1952 and 1954 numerous queens were dug up in September, some with eggs larvæ or even workers. In 1952 the main flight was early, 19-20.VII., in 1954 it occurred in August, but the exact date is unknown. The numbers of queens dug up from their hibernation sites and the state of their brood are given in Table 1, where all the workers listed were micrergates. The queens were generally found in small bare patches of soil in grassland. They were 5-6 cm. below the surface, often in cells under stones 5 cm. or more in length. These hibernation sites which may become the sites of nest foundation are usually in fairly loose soil, in some instances overgrown by moss or small tufts of *Agrostis tenuis* or *Holcus* with straggling diffuse roots. At no time have the queens been found amongst dense root tufts, such as those of *Festuca rubra*.

After flight the dealated queens may enter the small crevices in the soil, or participate in active digging of new burrows. On 4.VIII.53 queens after flight were observed on a previously prepared square metre plot of bare soil on a sloping grassland. The

TABLE 1. — THE NUMBERS OF QUEENS HIBERNATING TOGETHER AND THE STATE OF THEIR BROOD.

DATE.	No. ♀♀ IN COMMON CELL.	No. CELLS FOUND.	STATE OF BROOD.
22-26.	1	17	1 ♀ with eggs; 1 ♀ 2☿; 1 ♀ 3 ☿; 1 ♀ 5 ☿; 1 ♀ eggs, 5 ☿.
IX. 52.	3	1	Eggs and ☿☿.
21-28.	1	24	2 ♀ with eggs.
IX. 54.	2	9	1 group with eggs.
	3	6	1 group with eggs, larvæ; 1 group eggs and 1 ☿.
	4	1	No brood.
	5	2	No brood.
	6	2	No brood.
	7	2	1 group with eggs, larvæ, 5 ☿.
	17	1	No brood.
	20	1	Eggs, larvæ, pupæ, 8 ☿.
	21	1	No brood.

dealated queens were digging six burrows on the lower part of the square metre and seven other queens were "exploring" the plot. The sites of these burrows were numbered. On site 1 five queens were removing soil particles and small pebbles up to 0.6 cm. in length and there appeared to be complete cooperation between them. Site 2 was only 1 cm. away from site 1 and another three queens were similarly digging in. After half an hour one of the exploring queens joined this group and all the four then dug together. On site 3 one queen was closing in a burrow; the burrow on site 4 was also dug by a single queen, while two females were digging together on site 5; site 6 was occupied by one queen who dug herself in and closed the exit hole in 40 minutes. At times, the digging queens left their burrows, but they returned to them. From 4-5.30 p. m. the queens who were searching the plot, frequently probed the soil, made small holes and then rejected them. It was thought that the somewhat moister soil in the lower part of the plot was more acceptable, and small patches of earth were then moistened with water on the upper half. The searching queens stopped and probed the moist soil, they were attracted to it, but they did not dig in. By the next morning, all but the queens in site 2 had dug themselves in. The 4 ♀ on this site were still digging and by then they had excavated a burrow 2.5 cm. in depth. These marked burrows were left until 29.IX.53, when the whole square metre was examined. Sites 4-6 were found empty, while sites 1 and 2 had coalesced into a large single burrow, with 17 ♀ and a brood of eggs, large larvæ and 6 minute ☿☿.

On the basis of the data in literature and the observations described above, varying numbers of queens were set up in artificial nests and the rate of development of the first brood and the survival of the queens was recorded.

MATERIALS AND NESTS USED IN OBSERVATIONS ON THE DEVELOPMENT OF THE FIRST BROOD

The dealated queens were collected immediately after flight on 4. and on 10.VIII.53, on 15, 17 and 28.VIII.55 and on 13.IX.56. These queens were weighed, grouped together in different numbers and introduced into artificial nests. In 1953 the nests were of plaster of Paris of 20 cm. square. They were divided into three chambers,

the first with wet cotton wool, the middle forming the brood chamber and the outer, the foraging chamber. The brood chamber was sprinkled with a thin layer of sand from which the queens constructed the walls of their cells. These nests were covered with glass sheets and with black paper. In 1955 and in 1956 the ants were kept in small plastic nests, each 13 cm. long and 7-5 cm. wide, divided into 3 compartments, each compartment with a sliding transparent lid of its own. The compartment with the wet cotton wool communicated with the brood chamber by numerous perforations in the dividing wall made with a hot needle. The brood chamber was again sprinkled with sand and its lid perforated for aeration; it communicated with the third compartment in which food was placed after the appearance of the first workers.

The first brood was reared entirely on the reserves in the queen's fat body and on those derived from the break down of the wing muscles. After the hatching of the first workers, the foragers were given cut up insects (usually mealworms), 5 % sucrose and 5 % marmite solutions on cotton wool placed on a coverslip. In some nests large colonies of a 100-150 ☿☿ were eventually produced on this diet, but it cannot be considered as satisfactory as at no time were the workers bigger than the first brood of micrergates. This however does not affect the issue of these observations, which were essentially confined to the rearing of the first brood fed on the reserve substances of the queens themselves. The nests were kept in a laboratory where the temperature was fairly constant, fluctuating between 17°-20° C. and only on a few rare occasions falling to 15° and rising to 23° C. The nests were inspected twice and sometimes three times a week and the numbers of the brood in different stages were counted under a binocular. Obviously, daily observations would have been more accurate, but owing to the small size and the slow development of the first brood, it is considered that only a small proportion of the total may have been missed.

As the first brood was nourished by the reserve substances in the queens, the production of the first workers, which foraged for food, marked the turning point in the economy of the colony. This was taken as the first fixed point in the interpretation of the results. The second fixed point was the production of one ☿ per ♀ in the nests i.e. in the colonies with 4 ♀, the time of hatching of the first 4 ☿. This is probably a less important stage, as on the whole it closely follows on the first, but it provides a comparison of the nests with groups of queens with those with single queens, where the two stages are coincident. Omitting the records of preliminary observations made in 1952, the following results are based on 63 nests set up in 1953, 1955 and 1956. Observations on the nests in 1953 and 1955 were continued for a year, those set up in 1956 were terminated on 4.I.57, when the queens were dissected and the state of their ovaries and that of the fat body examined. Out of the 63 nests, 39 were with 1 ♀, 13 with 2 or 3 ♀, 9 with 4 or 5 ♀, one with 9 ♀ and one with 16 ♀.

RESULTS

1°. — ***Mortality of the queens*** within a period of a year after flight was much higher in the nests with single queens than in those with grouped

TABLE 2.—MORTALITY OF ♀♀ *L. flavus* THROUGHOUT THE PERIOD OF A YEAR AFTER FLIGHT.

No. ♀♀ PER NEST.	No. NESTS.	TOTAL NO. ♀♀.	% MORTALITY *Up to 1* ☿.	*Up to 1* ☿ *per* ♀
1	39	39	62	62
2 or 3	13	30	10	20
4 or 5	9	33	3	3
9 or 16	2	25	8	8

females (see Table 2). Death due to animosity and fighting between the queens of *L. flavus* appears to be rare, but at least one queen in a nest with fifteen others died mutilated, with her abdomen torn off. Three others, in the multi-female nests died from unknown causes, with large stores in the fat bodies, but at least four of the single queens also died with large reserves. Single queens take a much longer time to produce the first workers than do the queens in groups (see Tables 3 and 4) and the loss of weight accompanying the depletion of their reserves is considerable (Table 7). Dissections showed that many of the isolated queens died with exhausted fat bodies. This occurred before the hatching of the workers, i.e. before the reserves were supplemented by the foragers. Three of the queens in the multi-female nests also died with completely exhausted fat bodies, but this state followed by death was much rarer amongst the grouped individuals than amongst the solitary ones. Grouped queens were continually stroking one another with antennae, cleaning and licking one another and thus probably removing the fungi which settle on their bodies. None of them died from fungal attack, whereas five of the single females died covered with fungi.

2°. — ***The time taken to produce the first workers.*** — This interval was longer in the nests with solitary queens than in most of those with 2 or 3 queens, while the most rapid development of the first brood was in nests with 4 or more queens. In the nests with 1 ♀ the average time to the hatching of the first worker was 206 days and the limits 96-347, three of the surviving 15 ♀ failing to produce workers in 350 days. This average time in the nests with 2 or 3 queens was 122 days and varied between 72 and 195, while in the nests with 4 or more queens the average

TABLE 3.—THE NUMBER OF DAYS AFTER FLIGHT TO PRODUCTION OF THE FIRST ☿ AND TO 1 ☿/1 ♀.

To first ♀.	No. of days in intervals of 20.														
	51 — 70	90	110	130	150	170	190	210	230	250	270	290	310	330	> 350
1 ♀ per nest.	—	—	3	1	—	—	1	1	1	1	1	—	2	—	4
2-3 ♀ per nest.	—	2	1	3	3	—	—	1							
⩾ 4 ♀ per nest.	1	6	2	1											
To 1 ☿/1 ♀.															
1 ♀ per nest.	—	—	3	1	—	—	1	1	1	1	1	—	2	—	4
2-3 ♀ per nest.	—	—	1	1	3	—	—	—	—	—	2				
⩾ 4 ♀ per nest.	—	4	4	—	1	1									

was 84 days and the extremes 51 and 112. The same sequence also held true for the rate of development of 1 ☿ per ♀. These data are given in Table 3. The slow rate of development of the first brood in the single queen nests is also shown in Table 4, which gives the stage of development of the colonies with 1 ♀ and with 4 ♀, kept at room temperature for 110 days.

TABLE 4. — THE STAGE OF DEVELOPMENT OF THE FIRST BROOD ON 1. I. 57 IN NESTS WITH 1 ♀ AND 4 ♀. THE ♀♀ WERE COLLECTED AFTER FLIGHT ON 13.IX.56.

No. ♀♀ per nest.	No. nests.	Total ♀♀.	Mortality ♀, to 1. I. 57.	Nests with ☿☿.	Brood in nests per surviving ♀			
					Eggs.	*Larvæ.*	*Pupæ.*	☿☿.
1	20	20	25 %	15 %	3.1	5.1	0.3	0.4
4	5	20	0 %	100 %	6.1	5.9	2.3	2.5

The slow development of the first brood in the nests with 1 ♀ is probably due to two causes. Firstly, the queens in all the nests frequently ate up several of the initial small egg batches which they had laid, but in the multi-female nests some of the original communal heap remained untouched and developed into larvae. Secondly, the larval stage of the first brood in the single-female nests was very long, for instance in 9 of the nests the period between the appearance of the first larva and that of the first pupa was 49, 60, 77, 84, 107, 116, and 126 days. In nests with 4 or more queens this period lasted between 13 and 38 days. Considering that the larger groups of queens have fewer larvae per head to feed than do the solitary ones (see Table 5), and that the food received by the growing larvae is derived from the reserves in the females, communal rearing of the first brood appears to be more efficient.

3°.—***The total number of eggs laid and the resulting brood produced up to the time of hatching of the workers.*** — In the interval of time preceding the hatching of the first worker, single queens lay a higher total of eggs than do the queens in groups. This laying, however, is much more protracted and desultory and the small batches of eggs are often eaten up before any larvae result from them. The absolute number of eggs in nests with grouped queens was of course much greater than in those with single queens, although the number per queen was lower (see Table 5). Moreover, the rate of oviposition of the grouped queens was more rapid than of the solitary females. Each ant in the groups of 4 or more queens, laid one egg every 2.5 days, and every 3.7 days when the groups were of 2-3 females. The solitary females laid one egg every 4.3 days. The eggs were laid in small batches and this rate of oviposition is given only for the sake of comparison. The eggs were readily eaten, and the frequent disappearance of the larvae was also assumed to be due to cannibalism. As can be seen in Table 5, the total mortality of the brood was slightly higher in the single—than in the multi-queens nests.

TABLE 5.—SUM TOTAL BROOD PER ♀ (INCLUDING THOSE EATEN) UP TO THE DAY OF THE FIRST ☿ AND UP TO THE TIME OF HATCHING OF 1 ☿ PER ♀.

	No. ♀ per nest.	No. nests.	Total.			No. surviving.		No. eaten.		Brood eaten as % of (*Total eggs-surviving eggs*).
			Eggs laid.	*Larvæ Produced.*	*Pupae.*	*as Eggs.*	*as Larvæ.*	*Eggs.*	*Larvæ.*	
To day of hatching of 1st. ☿.	1	12	48	19	3	4	5	25	11	82
	2-3	10	33	13	2	4	5	16	6	76
	4-5	8	37	10	3	11	4	16	3	73
	9-16	2	17	9	1	3	6	5	2	50
To hatching of 1 ☿/1 ♀.	1	12	48	19	3	4	5	25	11	82
	2-3	8	41	15	3	7	4	19	8	79
	4-5	8	39	11	4	8	3	20	4	77
	9-16	2	25	13	3	5	2	7	8	75

It may also be noted that the initial broods were small. Subsequently, when the number of workers increased the rate of oviposition was much greater.

4°.—***The loss of weight of the ♀♀ before the production of the workers.*** — The weights of *L. flavus* queens after flight were variable, but mostly they ranged between 15 and 20 mg. Exceptionally light queens of 7 and 8 mg. have also been taken, but they did not survive for any lenght of time. Heavy females, weighing up to 30 mg. have also been encountered, but these were old queens dug up from populous nests. The distribution of the weights of the ♀♀ collected after flight, in 1953, 1955, and in 1956 are given in Table 6.

TABLE 6.—THE DISTRIBUTION OF WEIGHTS IN mg. OF ♀♀ *L. flavus* COLLECTED AFTER FLIGHT.

Year.	Weight in mg.															
	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22
1953	1	2	—	1	2	1	2	—	2	4	15	7	2	4	2	1
1955.....	—	—	—	—	—	—	—	1	6	11	11	21	7	8	1	1
1956.....	—	—	—	—	1	5	2	3	9	5	9	11	2	1	2	—

A detailed account of the variations of size in the genus *Lasius* is given by Wilson (1955). The variations in the weights of the queens of *L. flavus* produced in the Silwood Park populations probably depend

on two factors, i. e. on the variations in the sizes of the queens and on the variations of the amounts of the reserves accumulated in their fat bodies.

In artificial nests the queens have been known to lose as much as 40 per cent of their original weight in the rearing of the first brood (i.e. the weights dropped from 20 to 12 mg). In natural nests, the queens of *L. flavus* emerge from the pupae with fat bodies, which though already large increase in size and fill up the whole haemocoele by the time of flight. Some alate queens, which emerged in the field nests on 19-20.VII.53 and queens collected after flight on 4.VIII.53, i. e. a fortnight later, were dissected and it was seen that the fat bodies had increased in size considerably in that interval of time. Since it is the reserves derived from the break-down of the wing muscles and from the fat body which contribute to the development of the eggs and to the feeding of the first brood of larvae, the rate of production of the foraging workers must be of great importance. In the laboratory nests, the queens which had not produced foragers by January generally lost about 25 per cent of their original weight (see Fig. 1 on the next page). Some of the queens in the nests with 1 ♀ and with 2 ♀ were still without workers by the middle of March and, by then, they had lost up to 35 per cent of their original weight. There were 15 of these females in all; 5 of them produced workers later, one died covered with fungus, 4 died with completely exhausted fat bodies and the rest lived for over a year, but failed to produce a brood. It may thus be considered that the loss of about 35 per cent of the original weight indicates a dangerous stage of depletion.

In 1953, 5 of the lightest queens collected after flight (7, 8, 8, 10, and 11 mg) were put together in a common nest, but after laying small batches of eggs which they ate up, all died without rearing a brood. It is supposed that their initial reserves were small and were rapidly used up.

TABLE 7.—LOSS OF WEIGHT BY ♀♀ *L. flavus* BEFORE PRODUCTION OF THE FIRST ☿. WEIGHT IN mg. NO. NESTS IN BRACKETS.

No. ♀ per nest.	Year.	♀ Weight after flight.	January wt. of ♀ with no. ☿.	% Loss.	March wt. of ♀ with no. ☿.	% Loss.
—	—	—	—	—	—	—
1 (5)	1953	17.8	13.6	24	11.8	34
1 (16)	1955	17.4	13.0	25	11.4	35
1 (15)	1956	15.4	11.3	27	—	—
2 (4)	1953	16.0	13.5	16	11.0	31
2 or 3 (9)	1955	17.4	13.1	25		
4 (9)	1955	18.6	14.5	22		

5°.—***Condition of the ovaries and the fat body in the solitary females and in the ♀♀ kept in groups.*** — The queens collected after flight on 13.IX.56 were weighed and introduced into small plastic nests. Twenty-four females were reared in isolation and another 24 in 6 groups each of 4 ♀♀. As has been said, at this stage the haemocoele was filled with

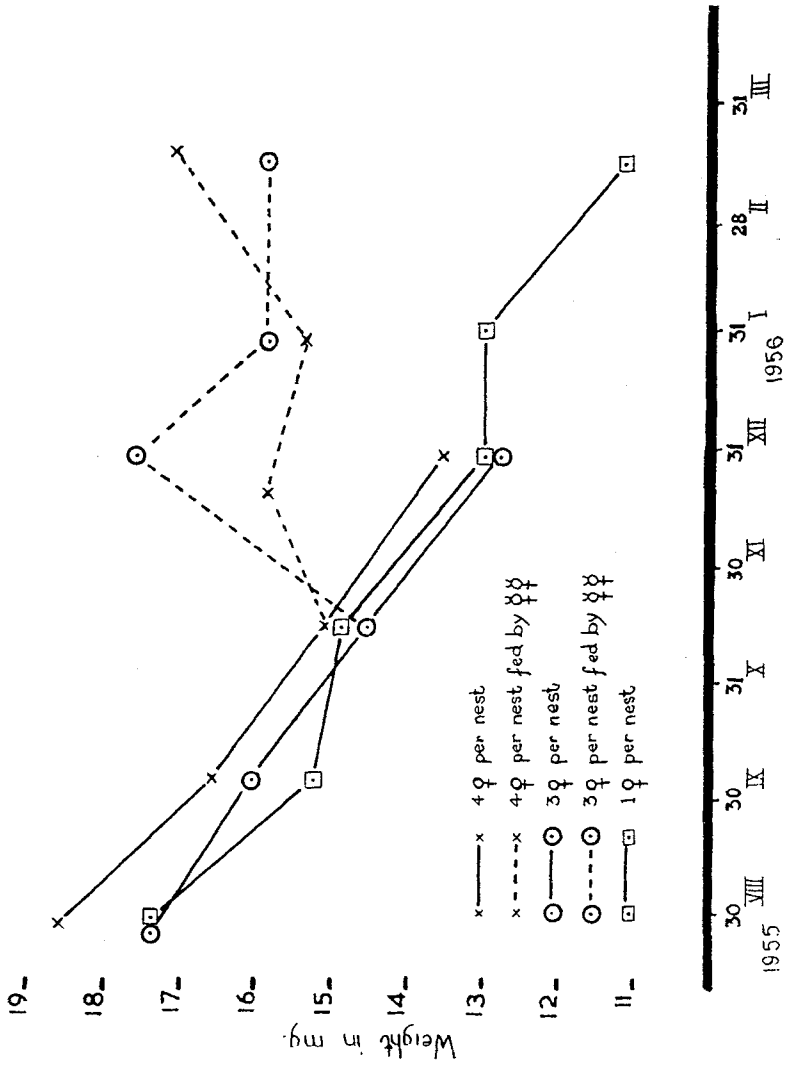

Fig. 1. — Loss of weight by the ♀♀ *Lasius flavus* before the hatching of the first ♀♀ and the subsequent gain by the females fed by the foragers.

an enormous fat body, but the ovaries were minute and threadlike and although the proximal egg (egg 1) was differentiated it was barely wider than the rest of the ovariole. On 1.XI.56, 4 solitary queens and one group of 4 were weighed and dissected, while all the remaining surviving queens were killed for dissection on 4.I.57. By that time all the colonies with 4 ♀ had produced workers (up to 1 ☿ per ♀) and not one of these queens had died. Five of the 20 solitary females died, and 3 of the remaining 15 had colonies with workers, the rest had broods in early stages (see Table 4).

The structure of the ovaries in the genus *Lasius* has been described by Bickford (1895), who gives the number of ovarioles in *L. flavus* as 24. The ovaries are polytrophic with groups of fairly large nurse cells between the oocytes. In the present dissections the number of differentiated oocytes in an ovarioles was 1-3, the proximal growing egg 1 being much larger than the rest. Only rarely was a large number of ovarioles functioning simultaneously i.e. egg 1 was rarely of the same size in many of the ovarioles. The recorded numbers of the proximal growing eggs in the R. and L. ovaries were 3 + 6, 4 + 5, 6 + 2, 6 + 6, 5 + 8, and 8 + 8. Several of the queens had ripe eggs, but never more than 2 in one individual; these could be distinguished from the growing ones by their size and by the shiny chorion. The lengths of 30 eggs laid by these queens were measured and they ranged between 0.63 and 0.94 mm (7 being 0.6-0.7 mm; 6, 0.7-0.8; 13, 0.8-0.9 and 4, 0.9-0.94 mm), while the lengths of the longest proximal eggs in the ovarioles ranged from 0.10 to 0.94 mm. The results of these dissections are given in Table 9. The size of the fat body was arbitrarily subdivided into 5 categories, 5 indicating a large one filling the haemocoele and 1 a state of almost complete depletion. The dissections on 1.XI.56 mainly indicated that the 4 solitary queens and the 4 queens kept as a group, all had functioning ovaries with growing proximal eggs and very large fat bodies. By 4.I.57 there were females with completely threadlike ovaries, both amongst the single and the grouped queens and much of the fat body had been used up. Amongst the 13 dissected solitary queens, there were 5 with threadlike ovaries, but 3 still had ample reserves in their fat bodies (the indices being 3, 3 and 4), while 2 appeared truly spent as not only were the ovaries threadlike, but the fat bodies were near to depletion (assigned the values of 1 and 2). Queens with appearently non-functioning ovaries were found in 3 of the nests with grouped queens. In each of two of these nests there was one queen which appeared to be "spent" i.e. all the ovarioles were threadlike and the fat body almost completely used up; in the third nest there were 2 ♀ with threadlike ovarioles, but with ample reserves in the fat body (values of 3 and 3). As there was little difference in the state of the ovaries of the grouped and single queens, no evidence was found for mutual inhibition of the queens in groups, at the time of the rearing of the first brood. Possibly the grouped queens were at a slight advantage, as the threadlike ovaries occurred in 4 out of their total

TABLE 8.—WEIGHT, STATE OF FAT BODY AND OF THE OVARIES OF ♀ *L. flavus*. (LARGEST FAT BODIES = 5, SMALLEST=1; LENGTHS OF RIPE EGGS 0.63—0.94 mm.).

DATE DISSECTED.	No. ♀ IN NEST.	No. NESTS.	No. WITH GROWING EGG 1.	No. WITH THREAD-LIKE OVARIES.	♀♀ WITH FUNCTIONING OVARIES.					♀♀ WITH THREADLIKE OVARIES.			
					Weights in mg.		*State of fat body.*	*Largest ovariole eggs 1* (mm).	*No. ripe eggs.*	*Weight in mg.*		*State of fat body.*	*Largest ovariole eggs 1* (mm).
					After flight.	On dissection.				After flight.	On dissection.		
1.XI.56	1	4	4	0	16.8	14.8	5.0	.32—.92	(1 in 1 ♀)	—	—	—	—
	4	1	4	0	16.8	14.8	5.0	.39—.64	0	—	—	—	—
4.I.57	1	13	8	5	16.6	12.3	3.6	.41—.94	5 (1 in 5 ♀)	14.1	9.5	2.6	.10—.21
	4	1	3	1	17.0	11.0	3.0	.44—.85	(1 in 1 ♀)	16.0	9.5	1.0	.14
	4	1	3	1	15.7	11.3	3.3	.46—.78	4 (2 in 2 ♀)	13.0	8.0	1.0	.17
	4	1	2	2	17.5	12.8	3.0	.49—.77	(1 in 1 ♀)	12.0	8.8	3.0	.14—.20
	4	1	4	0	18.0	11.5	3.0	.47—.70	0	—	—	—	—
	4	1	4	0	16.0	11.3	3.0	.34—.85	(1 in 1 ♀)	—	—	—	—

of 20, whereas in those kept singly they were found in 5 out of the 13 dissected females.

A connection between the weights of the queens after flight and the subsequent rate of depletion of the fat bodies was indicated by these dissections. In other words, the fat bodies in the queens with the lower original weights were the more exhausted by the time of dissection, i.e. by 4-I-57 (see Table 8).

6°. — ***Behaviour of the grouped queens of L. flavus.*** — No animosity or fighting was seen between the queens of *L. flavus* which were grouped soon after flight. A mutilated queen was found only on one occasion, eight months after flight, when the multi-queen colony had produced a large number of workers. Prior to egg laying, the grouped, as well as the solitary queens, usually constructed small cells of sand in one of the corners of the nest. The eggs were piled together in one central heap in these cells, the queens forming a ring and facing the eggs. They continuously stroked the eggs with their antennae and licked them. They also frequently stroked one another and at times bunched up in two tiers around the eggs. There appeared to be complete cooperation between the females while the brood was in the egg stage, but the groups tended to split later, generally at the time of the appearance of the first pupae. The pupae were usually, though not invariably, carried away to another part of the nest. This splitting up of the groups was most clearly seen in five of the nests, where the queens carried away the pupae and some of the larvae, remaining with them and establishing new, independent units. The later pupae were then tended by the remaining queens. Thus in one of the nests with 4 ♀ one of the queens carried away the first six pupae and established an independent unit; in another nest with 4 ♀, two queens carried away some of the larvae and all the early pupae; this occurred in another nest with 4 ♀ i.e. it split up into two units with two females in each after the appearance of the first pupae; in the nest with 9 ♀, two queens carried away the earlier pupae and remained with them; in the 16 ♀ nest four queens tended the earlier pupae in a separate corner of the nest. In carrying away and tending the pupae, the queens reproduced the behaviour of the workers in the more advanced colonies.

The nests which I have been using were small and unsuitable for the study of the results of permanent splitting up of groups of queens and sometimes when the workers had hatched and were foraging they would bring food to all the queens, irrespective of their position in the nests and then secondary amalgamations of the colonies would occur. It is realised that spatial separations of the colonies need now to be observed in different, much larger types of nests and if possible under natural conditions as well. However, even in these small nests there was further evidence for WASMANN'S hypothesis (1910) that pleometrosis in *L. flavus* is only of a temporary nature.

The behaviour of the queens in the nests also indicated that there may be other ways in which the splitting of the groups may occur. Fighting, between the queens, although rare did occur at least once. Also, after the hatching of the workers i.e. after food was introduced into the foraging chamber, some of the queens were seen to forage and exchange food with the other queens. It is thus possible that these active queens may sometimes leave the original hibernation sites and found new colonies near a source of food. Secondly, a curious behaviour by the workers was seen on two occasions; the micrergates after foraging and feeding the queen were seen to guide i.e. push her, towards the source of food. Under natural conditions this behaviour may lead to the splitting up of a group. However it must be remembered that although rareby multiple queen colonies of *L. flavus* do persist.

7°.— ***Nests of Lasius niger (L.) with single and with grouped queens.*** — The distribution of the weights of *L. niger* queens after flight may be of interest and are given in Table 9. The dealated queens were collected

TABLE 9.—WEIGHTS IN mg. OF THE DEALATED *L. niger* QUEENS AFTER FLIGHT ON 13.IX.56.

WEIGHTS IN mg.	20	21	22	23	24	25	26	27	28	29	30	31	32	33
NO. ♀♀.	1	3	6	4	9	14	16	10	13	1	3	1	0	1

at Silwood Park, Sunninghill, Berkshire' on 13.XI.56 weighed and introduced into the nests in different numbers. In 1956, five nests with 5 or 6 ♀, five with 3 or 4 ♀, five with 2 ♀ and 21 with 1 ♀ were set up. Some observations on these nests and on a few others set up in 1953, provide a comparison with *L. flavus*. At room temperature the surviving queens laid eggs a little later than did *L. flavus* i.e. within 4-5 weeks after flight. The stage of development of the brood at the beginning of February is given in Table 10 (cp. Table 4 for *L. flavus*).

TABLE 10.—STAGE OF THE BROOD BY THE BEGINNING OF FEBRUARY, 1957 IN THE LABORATORY NESTS OF *L. niger* COLLECTED AFTER FLIGHT ON 13.IX.56.

INITIAL No. ♀ PER NEST.	No. NESTS.	INITIAL TOTAL ♀♀.	No. DEAD.	% DEAD.	BROOD PER SURVIVING ♀ II. 57. *Eggs.*	*Larvæ.*	*Pupæ.*	☿☿.	No. NESTS WITH ☿.
5 or 6	5	29	15	52	9.1	2.0	0.8	0.1	1
3 or 4	5	18	8	44	15.1	0.9	0.1	0	0
2	5	10	0	0	9.0	3.0	0.1	0	0
1	21	21	6	29	8.1	6.4	0.9	0.2	3

Prior to egg laying the queens of *L. niger* sometimes behaved differently to *L. flavus*, i.e. they formed close bunches and palpated the abdomen of one of their number with their antennae, much as workers do to the laying queens.

As has been noted repeatedly in literature, there is a great difference in the degree of tolerance between queens of their own species in *L. flavus* and in *L. niger*. Queens of *L. niger* were often hostile to each other, but they did not begin fighting immediately after flight, only when egg laying had commenced were many mutilated bodies found. Fighting began again in some nests later, when laying was renewed. The mutilations after fighting included gashes through the abdomen and thorax, or even complete tearing off of the head or abdomen. The tolerance which the queens in pairs have shown to one another is surprising and not understood. Not all the mortality of the queens in multiple colonies was due to fighting, but in the nests with 3-6 ♀, though twelve females died of mutilations, eleven others died from unknown causes. These eleven, constitute 23 per cent of the total of 47 ♀, which is not very different from the mortality (29 per cent) in the nests with single queens. Of the six single queens one died covered with fungus, while the cause of mortality of the other 5 ♀ is unknown; they died with ample reserves in the fat body as did the eleven queens in the nests with grouped females. Most of the fighting occurred when the initial batches of eggs were laid, but after this members of the groups became more tolerant to each other.

The initial number of queens in the 3-6 ♀ nests in IX.56 was 47, by the end of X.56 thirteen of them were dead, mostly due to mutilation, one died in XI.56, six throughout XII.56 and three by I.57. Agressiveness between the queens appeared to be even more developed in the females kept in isolation for a period of three and a half months after flight. On 1.I.57 some of the queens of *L. niger* and of *L. flavus* were marked with small spots of aluminium paint and introduced into the nests of their own species. Three queens of *L. niger*, previously kept singly were introduced into 3 nests containing solitary *L. niger* females; there was immediate fighting between them and they had to be separated rapidly. A fourth *L. niger* queen was introduced into a nest with 2 ♀; there was no fighting, but she remained in a separate corner of the nest for an hour and was then removed. Simultaneously, 5 marked single queens of *L. flavus* were introduced into five other nests with single queens of their own species. There was no fighting and the queens were then left in the nests for three days. On 4.I.57 all the introduced queens were alive and undamaged, three of them in corners away from the host queens, the two others tending the brood together with the host queens. However, in spite of the aggressiveness of *L. niger* queens, the decrease in their numbers throughout the time of nest foundations was not only achieved by fighting to death. The apparently "peaceful" splitting of the colonies after the appearance of the earliest pupae, such as has been seen in the multi-female nests of *L. flavus*, occurred repeatedly in the *L. niger* nests as well.

Dissections of some of the grouped females were made at the beginning of February 1957 and they clearly indicated that all the surviving members

of the groups were contributing towards the establishment and rearing of the first brood. The females of *L. niger* had large fat bodies, not one of them was exhausted or "spent" as were some of the *L. flavus* females. Also, in none were the ovaries threadlike and in all the 16 dissected ants they were functional with either growing or with fully grown proximal eggs. The numbers of ovarioles in *L. niger* is 30-40 (BICKFORD, 1895) and only some of them were functioning simultaneously, but 13-20 ovarioles with growing proximal eggs have been seen. The lengths of 20 eggs laid by these females ranged between 0.79-0.94 mm, while the lenghts of egg 1 are given in Table 11. Examination of the table will indicate

TABLE 11.—DISSECTIONS OF ♀♀ *L. niger* ON 8.II.57.
WEIGHTS IN mg; LENGTHS OF EGGS IN mm.

ORIGINAL NO. ♀ IN NEST.	NO. ALIVE *8.II.57.*	AVERAGE WEIGHT *13.IX.56.*	AVERAGE WEIGHT *8.II.57.*	% LOSS.	STATE OF FAT BODY *(0—5).*	LARGEST EGG 1.
6	4	25	19	24	4.3	.51—.70
6	3	27	17	37	3.3	.61—.65
4	3	28	18	36	3.0	.65—.86
4	2	27	19	30	3.5	.77—.83
2	2	27	21	22	3.5	.52—.67
2	2	26	20	23	4.0	.68—.81
1 (6)	1 (6)	26	18	31	3.9	.50—.88

that at least at this stage of brood development there is no apparent inhibition of ovarial growth in some of the queens, such as exists in other Hymenoptera.

DISCUSSION

It is well known that the established nests of *Lasius flavus* are usually haplometrotic, that is they are characterised by the presence of a single fertilised queen, yet groups of queens sharing the same hibernation sites, which ultimately may become the sites of nest foundation, are not uncommon.

Observations on the single and multi-female nests in the laboratory have indicated that, at the time of the rearing of the first brood, the queens in groups are at an advantage over the solitary ones, as not only is their mortality greatly reduced, but the first workers are produced by them more rapidly. The appearance of the first foraging workers marks the turning point in the economy of the incipient colonies, as the rearing of the brood is then no longer dependent on the reserves derived from the break-down of the wing muscles and, no doubt, from the substances stored within the enormous fat bodies of the queens. Although the rate of egg laying by the grouped queens was higher than by the solitary

ones, the rate of development of the brood was also higher and fewer eggs per queen were laid before the hatching of the workers. Thus more reserves were available to the larvae and it is probable that they grew and metamorphosed more rapidly in the multi-female nests because they received more food. The queens of *L. flavus* lose much weight before the hatching of the workers and this loss is accompanied by a considerable exhaustion of the fat body. Some of the solitary females in which brood development was slow appeared completely spent before they were able to produce the foraging workers to supplement their reserves. Thus it is considered that the exhaustion of the fat body, or some cause associated with it, accounts for the greater mortality amongst the solitary females. It may also be recalled that some of the single queens died covered with fungi, whereas none amongst the groups died from this cause and it is probable that the frequent stroking of one another with their antennae by the grouped females helps to remove the fungi which settle on the surface of their bodies. Pleometrosis in *L. flavus* is temporary, but it is interesting to compare the founding of colonies in this species with that of the permanently pleometrotic *Monomorium pharaonis* L., as PEACOCK (1950) found great difficulty in starting new units with single females and no difficulty when the units were multi-female. Although Brian's analysis (1953 a) of brood rearing in relation to the worker number of *Myrmica* concerns a much later stage of development of an ant colony, it was nevertheless interesting to see that the broods which were tended by the larger numbers of workers metamorphosed more rapidly.

In contrast to the aggressiveness of their workers, the young queens of *L. flavus* show no animosity to one another. If this tolerance is due to habituation, as is suggested by THORPE (1956), then it is probably acquired soon after flight, at the time of digging of burrows by groups of females. This mutual toleration persists for a long time and only once was a mutilated queen found in the multi-female nests of *L. flavus*. The queens of *L. niger* show more hostility to one another and their initial tolerance seems to be frequently upset by the new stimuli presented by an ovipositing female and deaths due to mauling usually coincided with the appearances of new egg batches. Certain parallels may be drawn between the behaviour of the females of *L. niger* and those of *Pollistes gallicus* L. in N. Italy (PARDI, 1946), but in the wasps the most aggressive female of a group established herself as the dominant queen and the ovaries of the other queens began to degenerate. Dissections of the surviving members of the groups of *L. niger* have shown that all the queen-founders had functional ovaries and were thus contributing to the production and the rearing of the first brood. There was also no essential difference in the states of the ovaries of the queens of *L. flavus* kept singly and in groups. It would seem that haplometrosis is attained by different means in *Lasius* to other groups of social Hymenop-

tera i.e. not by the inhibition of the ovarial development in some members of a group (PARDI, loc. cit., BUTLER, 1954). This is witnessed not only by the presence of functional ovaries in the groups of queen-founders, but also by the presence of the laying workers in the colonies of the genus *Lasius* (BICKFORD, 1895; BERNARD, 1951). The production of eggs by workers may even be an integral part in the economy of the colony, as has been shown in the pleometrotic colonies of the ant *Myrmica rubra* L. by BRIAN (1953 b).

It was WASMANN (1910) who originally described the splitting up of the pleometrotic founder—groups of *L. flavus*. He found 4 ♀ *L. flavus* in a hibernation site and transferred them to an artificial nest. His queens remained together until the larvae were produced and then split into two units, with two females in each. In the present observations the splitting up of the multi-female colonies was seen in several nests, but at a later stage i.e. at the time of the production of at least the first pupa. Some of the queens then removed the early pupae and some larvae to another part of the nest and remained with them, forming new and independent units. This is probably the most usual way in which the multi-female founder-colonies of *L. flavus* split up. EIDMANN (1931) once observed a queen of *L. flavus* wandering about on a sunny day as late as April and it is possible that some of the queens, which are still capable of foraging several months after flight, may leave their original hibernation sites and found new colonies elsewhere. Peculiar behaviour of the workers, which on two occasions pushed the queen towards a source of food also indicates that some females may be lead away by the foragers to other sites, for instance those with root aphids. Fighting, resulting in death of the queens of *L. flavus* cannot altogether be ruled out, but it is probably rare. As a contrast, fighting and mauling to death is probably the most usual method of attaining haplometrosis in the colonies of *L. niger*, although splitting up of the multi-female colonies at the time of the appearance of the earliest pupae was also repeatedly seen in these nests. EIDMANN (1926) and DONISTHORPE (1927) give lists of records of several queens of *L. niger* combining in starting a colony, but DONISTHORPE points out that only rarely are populous nests found with more than one queen. He cites a number of experiments in which the artificially grouped queens combined in the rearing of the first brood, but the colonies ended with single queens after fighting and the death of the others. BERNARD (1951) points out that the great majority of ant societies are haplometrotic, and it would seem that unless there were mechanisms which would inhibit the ovarial growth in some members of a group of queens, the development of aggressive behaviour is an efficient method of attaining this state. In many of the species where the colonies remain permanently pleometrotic the queens retain high mobility, and BERNARD (loc. cit.) says that « il est remarquable que la plupart des espèces nuisibles aux cultures (*Tapinoma*, *Iridomyrnex*...) soient polygynes,

les nombreuses reines, rendant difficile la destruction de la société ». The queens of another common pest, *Monomorium pharaonis* also retain high mobility and are known to be relatively short lived (PEACOCK et al., 1950). The question of the advantages of high mobility and potential oviposition rate of the pleometrotic colonies such as those of *Polybia*, are discussed by RICHARDS and RICHARDS (1951) in their study of the South American wasps. They point out that these factors are probably the best defence against certain types of enemies, such as the ants.

The old queens of *Lasius flavus* which were dug up from large nests had distended gasters and were very slow moving. It may be that colony-survival is more likely if such queens receive maximum dispersal than if several are concentrated in one nest. However, an answer to this would only be obtained through a study of the economy of the nests in different climatic zones. WILSON (1955) says that *Lasius flavus* has a very wide range of distribution throughout Eurasia and N. America, which is exceeded within the genus only by *L. alienus* (Först). Though rarely, populous pleometrotic colonies have been found in Great Britain and it would be of great interest to known of their incidence in climates where the rearing of the first brood is not interrupted by hibernation during the cold spell.

Summary.

Populous colonies of *Lasius flavus* F. are generally haplometrotic, yet not infrequently several queens combine in founding nests. This aggregation after flight is not always accidental, but may arise from cooperation between the females in the digging of new burrows. Probably the queens become habituated to one another at this stage. In the laboratory the multi-female colonies were at an advantage over the single female ones, as the mortality of the queens was reduced and the first brood of workers was reared more rapidly. Grouping of the queens of *L. flavus* and of those of *L. niger* did not appear to influence the ovarial development of the survivors, and they all contributed to the establishment and the rearing of the first brood. This pleometrosis was temporary and the multi-queen colonies began to split into smaller units at the time of the appearance of the earliest pupae. In *L. niger*, fighting to death between the queens, at times of repeated ovipositions, also contributed towards the final state of haplometrosis.

Résumé.

En général, les nids populeux de *Lasius flavus* F. montrent de l'haplométrose, mais, quelquefois, plusieurs reines s'unissent pour fonder des nids. Ce rassemblement après le vol n'est pas toujours fortuit ; il peut résulter de la coopération entre les reines pendant les excavations de

nouveaux trous dans la terre. Il est probable qu'à ce moment les reines s'habituent les unes aux autres. Au laboratoire les colonies avec plusieurs reines étaient plus favorisées, car leur mortalité était assez réduite et les premières ouvrières furent produites plus rapidement que dans les nids à reines solitaires. Les ovaires des reines de *L. flavus* et de *L. niger* rassemblées en groupes, restèrent fonctionnels, et toutes les reines qui survécurent contribuèrent à l'établissement et à la nourriture de la première couvée. Cette pléométrose était temporaire et les colonies avec plusieurs reines commencèrent à se diviser à l'instant où parurent les premiers cocons. Plusieurs reines de *L. niger* s'engagèrent en combat jusqu'à la mort au moment des pontes, ce qui contribua également à l'état d'haplométrose.

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