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RECRUITMENT TRAILS IN THE HARVESTER ANT *POGONOMYRMEX BADIUS*

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Introduction

Pogonomyrmex is the most abundant and specialized genus of harvester ants in North America. The workers mainly collect seeds for food, but they also carry dead insects into their nests. In view of the great abundance of these ants in many parts of the United States (Cole 1968), the ease with which they can be cultured in the laboratory, and their considerable economic importance, surprisingly little has been learned to the present time concerning their communicative behavior. Most attention has been focused on alarm behavior. Wilson (1958) discovered that alarm responses are released in workers of the Florida Harvester Ant (*P. badius*) by a pheromone produced in the mandibular gland of the ant. This behavior ranges, according to the stimulus intensity and duration, from mild attraction to attack and prolonged digging in the soil. In 1966 McGurk *et al.* identified the pheromone as 4-methyl-3-heptanone and also detected it in *P. barbatus, P. californicus, P. desertorum, P. occidentalis* and *P. rugosus.* Although in many ant species chemical trails laid down by worker ants are the essential signals for the initiation of mass foraging behavior in nestmates (see review by Wilson 1971), they have not yet been implicated (or even suspected) in *Pogonomyrmex.* This article reports the discovery and subsequent analysis of such a recruitment system in *Pogonomyrmex badius.*

Materials and Methods

Field work was conducted in Tampa, Florida, in an open field near the campus of the University of South Florida. Laboratory experiments at Harvard University utilized three large colonies of *Pogonomyrmex badius* housed separately in two sand-filled terraria

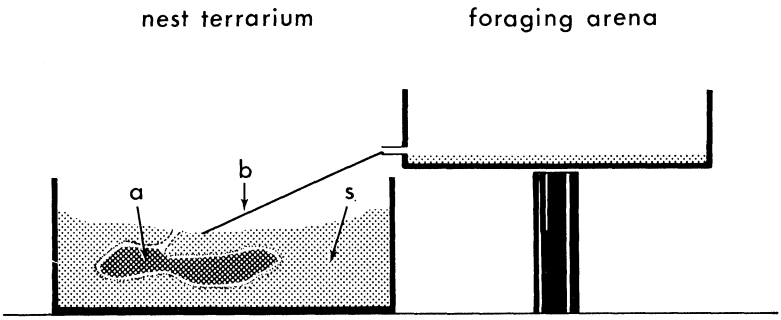

Fig. 1. Laboratory nest arrangement. *a* = nest chambers; *b* = bridge; *s* = sand.

(75 × 30 × 30 cm). These colonies had been kept in the insectary for three years, since the early spring of 1967. Their good health was attested by the fact that they continuously reared large quantities of brood, and each year produced winged males and females. Each nest terrarium was connected with a foraging arena 70 cm in diameter (Fig. 1). This combination provided an excellent arrangement for laboratory pheromone tests and orientation experiments that simulated natural conditions. Additional methodological details will be given with the description of the individual experiments.

Experiments and Results

Description of the food alarm behavior

We have noted that when individual workers of *P. badius* attack large, active insect prey in the vicinity of the nest, they discharge the alarm pheromone 4-methyl-3-heptanone from their mandibular glands. The pheromone both attracts and excites other workers within distances of ten centimeters or so, with the result that the prey is more quickly subdued. This observation led to the question: Do workers also employ a directed food alarm when they encounter a food source several meters distant from the nest entrance? The following laboratory experiments were designed to find an answer to this question.

In the center of the foraging arena (Fig. 1) a freshly killed cockroach (*Nauphoeta cinerea*) was offered. The first ant to discover the prey was marked; all other scouting ants were then removed. After several unsuccessful attempts to transport the cockroach, the scout ant typically ran toward the arena exit near the nest. At this time its locomotory behavior changed markedly. The abdomen was bent downward and the tip dragged over the ground with the sting fully extruded (Fig. 2). Almost invariably within seconds after the ant had entered the nest, a group of 10-15 nestmates ran out and took the precise course of the scouting ant over the bridge to the foraging arena (Fig. 3). It is noteworthy that the recruited group often included the big soldier caste, because this caste is seldom seen making individual foraging excursions. Before the first recruited group arrived at the food source, the original recruiting ant usually also returned to the food in the arena, following its own trail and reinforcing it with the extruded sting. At this time it was closely followed by other ants (Fig. 3). After a few more minutes a number of ants finally assembled around the prey (Fig. 4). Small dissected pieces were then carried

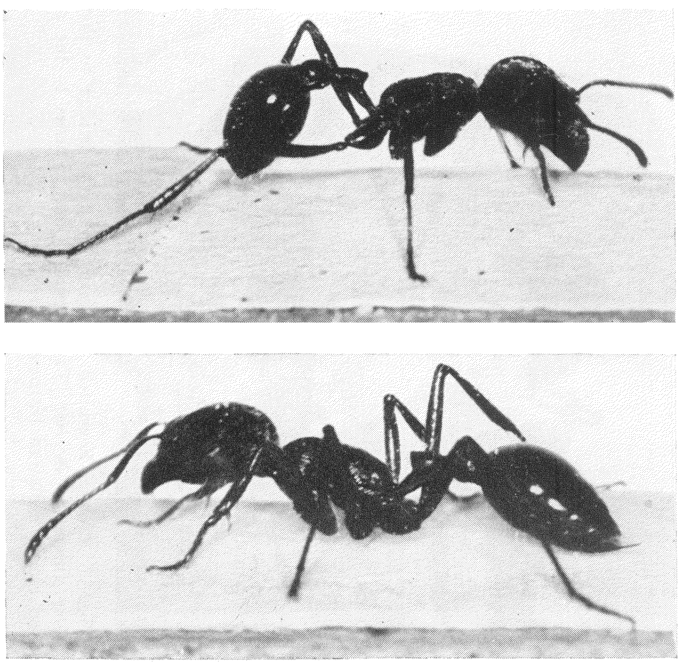

Fig. 2. *Above:* A foraging *Pogonomyrmex badius* worker passes over the bridge to the arena. During this searching phase the ant only occasionally touches the surface with the tip of its abdomen. *Below:* After discovering a source of food, the scouting ant returns to the nest, dragging the tip of the abdomen over the surface with sting extruded.

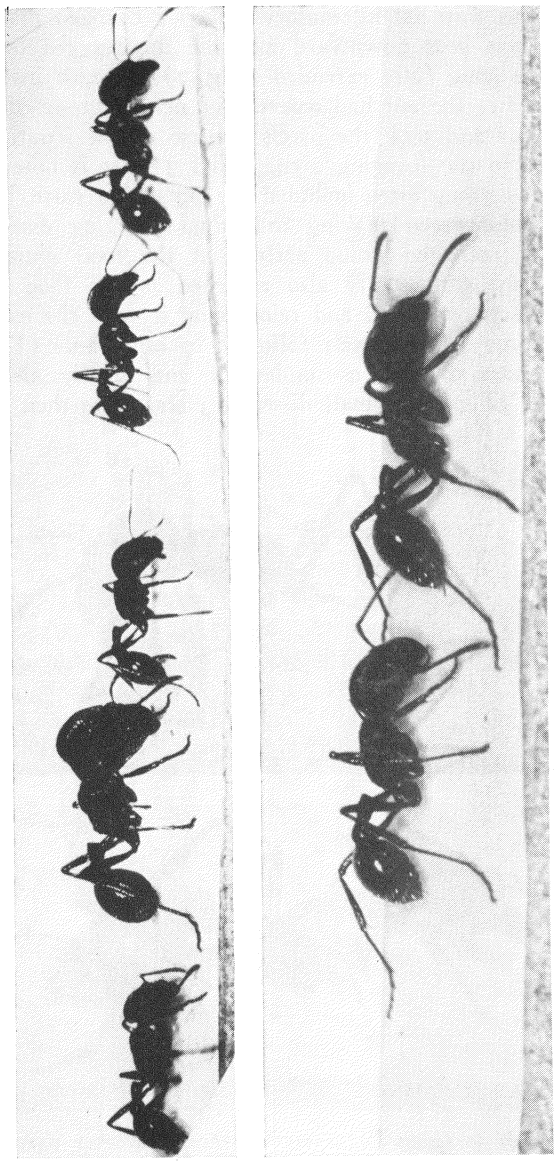

Fig. 3. *Above:* Recruited ants pass over the bridge to the arena while carefully scanning the surface with their antennae. *Below:* The recruiting ant returns to the arena, reinforcing its trail with its extruded sting and closely followed by nestmates.

home by individual ants, some of which extruded their stings and touched them to the surface (Fig. 5). Simultaneously, another group of ants, mostly belonging to the smaller caste, ran with extruded stings back and forth between the nest and the prey, apparently reinforcing the recruitment trail. A freshly killed cockroach usually caused a much stronger recruitment alarm than oat flakes. Moreover, when all of the oat flakes were offered at one spot, the recruitment activity was more intense than if the same number of flakes were scattered over the whole arena. Fig. 6 gives the quantitative data from representative experiments in which ants were placed in the three feeding circumstances.

Our observations on the recruitment behavior strongly suggest that chemical signals are involved. The next series of experiments tested this hypothesis:

1) In one series the ants were counted as they passed over the bridge after a recruiting ant had returned on it from the arena to the nest. In a second series the old bridge was replaced with a new one immediately after the recruiting ant had passed over it, and again the number of ants running to the arena was recorded. The number of ants passing over the bridge in the second series proved to be far smaller than in the first series (Table 1).

	Number of ants that passed over the bridge after a recruiting ant returned to the nest.	Number of ants that passed over a new bridge, which replaced the old one after a recruiting ant returned to the nest.
M ± sd	17.8 ± 5.3	1.7 ± 1.9
Range	12 — 28	0 — 5

Table 1. During a 5 minute period all ants that passed over the bridge to the arena were counted (see Fig. 1). The data are derived from 8 replications of each of the two experiments.

2) If the arena was rotated after the recruiting ant had left the arena, thus removing the trail from the arena entrance, or if the surface was covered with a new layer of sand, the recruited ants no longer oriented accurately to the source of food.

The results of these experiments left little doubt that *P. badius* utilizes special chemical recruitment and orientation signals. We next conducted a search for the anatomical source of the recruitment pheromone.

The source of the recruitment pheromone

In preliminary experiments we learned that *P. badius* workers

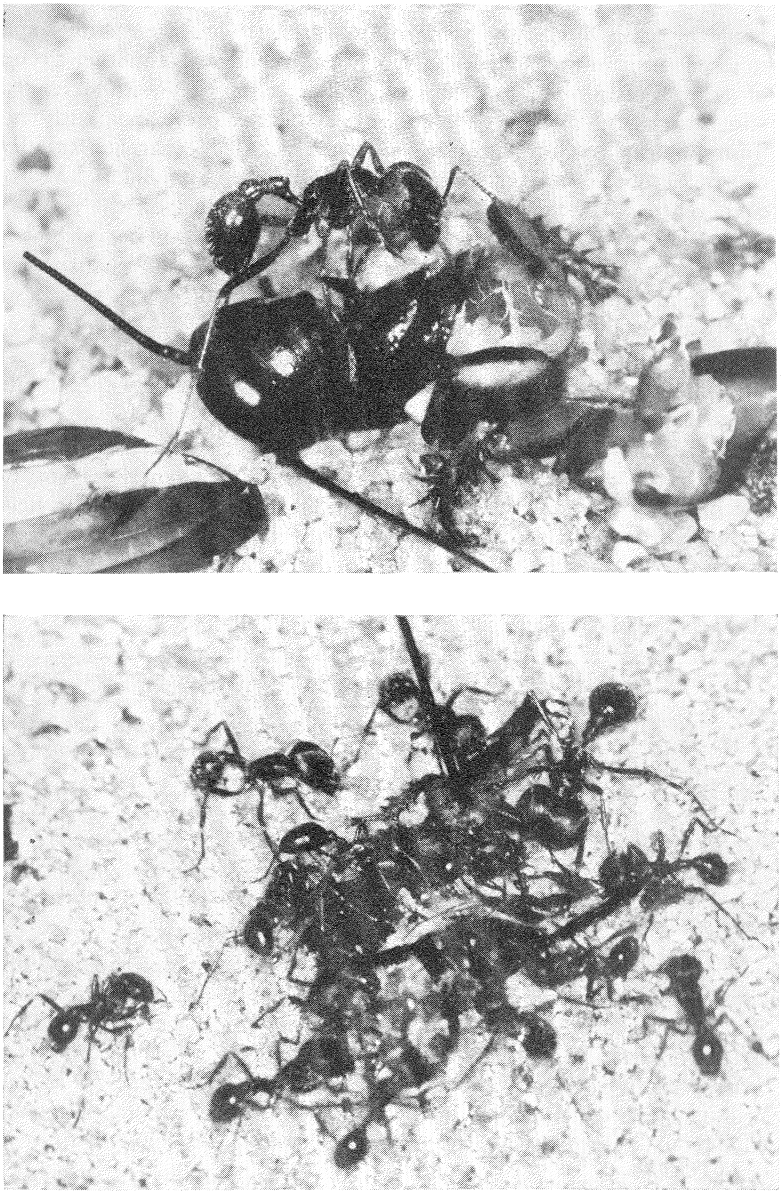

Fig. 4. *Above:* A scouting ant discovers a cockroach in the arena. *Below:* About 10 minutes later she has recruited 10-15 ants, which are now assembled around the prey.

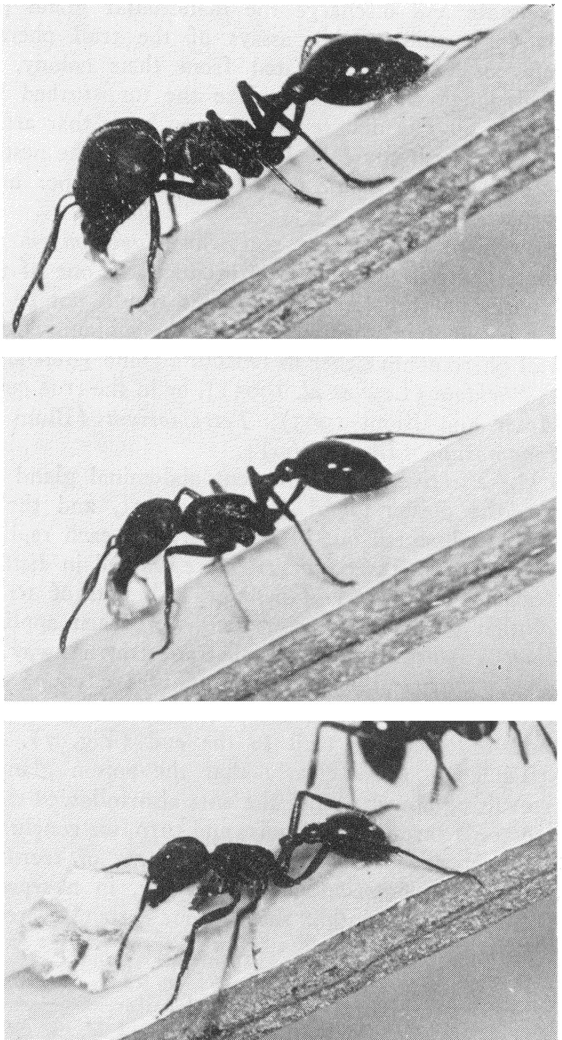

Fig. 5. Recruited foragers return to the nest transporting food. *Above,* "soldier" (very large worker) with extruded sting and carrying cockroach fragment; *middle,* smaller worker with extruded sting, carrying cockroach fragment; *below,* smaller worker with oat flake, no sting extruded.

are too excitable and discharge the mandibular gland pheromone too readily to permit reliable assays of the trail pheromone on small groups of workers separated from their colony. For this reason we designed the test to utilize the undisturbed laboratory nest arrangement. The nest terraria are so large that artificial test trails 50 cm in length could be laid beginning at the nest entrance. Furthermore such trails could be laid over the paper bridge, and into the arena.

The extrusion of the sting by recruiting *Pogonomyrmex* workers suggests that the trail pheromone is produced in one of the glands associated with the sting apparatus. This would not be surprising in view of the fact that other members of the subfamily Myrmicinae produce trail pheromones either in Dufour's gland [*Solenopsis* (Wilson 1959), *Pheidole* (Law *et al.* 1965)], or in the true poison gland [*Atta* (Moser and Blum 1963), *Tetramorium* (Blum and Ross 1965), *Monomorium* (Blum 1966)].

To test the effectiveness of different abdominal gland secretions, the hindgut, the poison gland (with vesicles), and the Dufour's gland were first dissected out of workers. For each replication the organs of a kind from three workers were washed in distilled water and then crushed in 0.5 ml of benzine. Aliquots of 10 μl of the benzine solution of the gland secretions were then applied with a syringe along a 50-cm-long artificial trail drawn away from the nest entrance. During the next five minutes a count was taken of all the ants — except those carrying sand — that left the nest entrance and followed the trail to the end (Fig. 7). The data obtained (Table 2) show clearly that the poison gland has the strongest recruiting power. Since the ants also followed the artificial poison gland trails through each twist and turn, we conclude that the poison gland secretion serves both as a releaser of recruitment behavior and as an orientation cue. However, in a separate study Hölldobler (1971) found that the Dufour's gland secretion of *P.*

	Poison Gland			Dufour's Gland			Hindgut		
	N	M	Range	N	M	Range	N	M	Range
Colony I	6	77.3	39-120	9	11.3	5-27	9	5.1	1-12
Colony II	8	97.9	38-187	10	8.2	3-18	8	5.1	0-17
Colony III	7	81.6	32-157	9	15.5	5-29	9	6.1	0-12

Table 2. Artificial trails composed of benzine solutions of different abdominal gland secretions were drawn for distances of 50 cm from the nest entrance. During the next 5 minutes all the ants that followed the trails to the end were counted. The number of replications (*N*) and the mean and range of the number of responding workers are given.

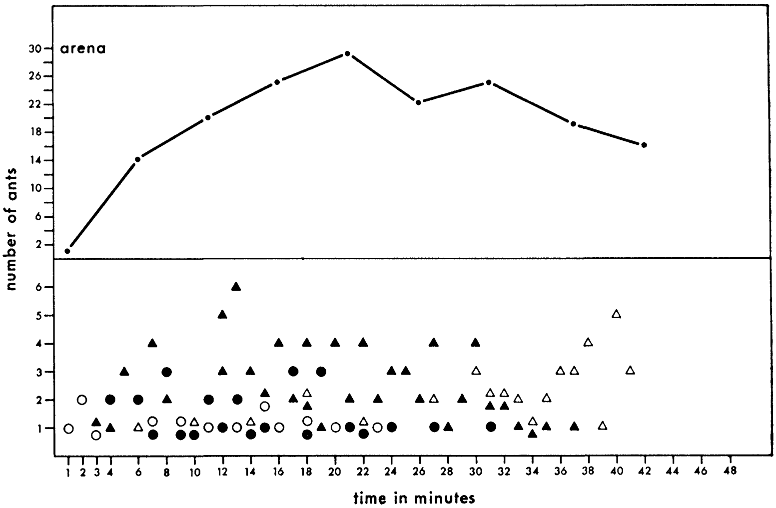

Fig. 6a. The progress of recruitment and foraging activity at insect prey (the cockroach *Nauphoeta cinerea*). *Above:* Total number of ants in the arena. *Below:* Ants returning to the nest. O = sting extruded, no food; ● = sting extruded, with food; solid black triangle = sting not extruded, with food; △ = sting not extruded, no food.

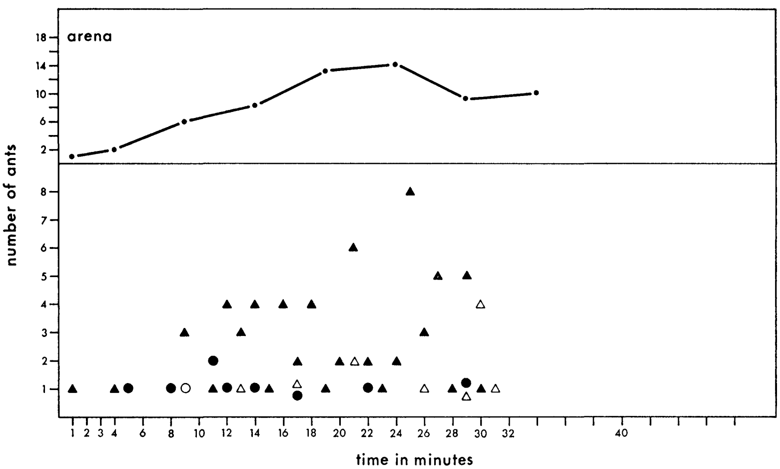

Fig. 6b. The progress of recruitment and foraging activity at oak flakes at one spot. Details as in caption of fig. 6a.

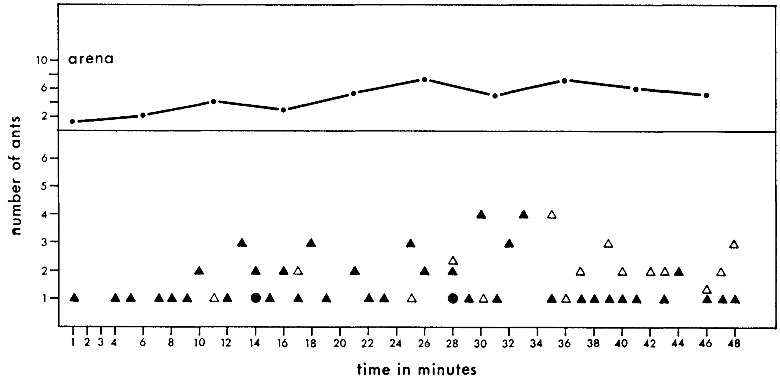

Fig. 6c. The progress of recruitment and foraging activity at oak flakes widely dispersed. Details as in caption of fig. 6a.

badius attracts homing ants. His orientation experiments indicated that the Dufour's gland secretion serves as a special chemical homing signal, since homing harvester ants closely follow artificial Dufour's gland trails. These results raise the following question: Is the Dufour's gland secretion used as an orientation cue for recruited ants as a supplement to the poison gland material? The following experiments were devised in an attempt to disentangle these two functions.

An artificial poison gland trail was laid from the nest entrance over the bridge to the opening of the arena. In the arena the artificial poison gland trail was continued while an artificial trail made from the Dufour's gland substance was simultaneously drawn at an angle of about 45° to the poison gland trail. In a second series the artificial poison gland trail was stopped at the arena entrance and then continued in one direction with a Dufour's gland trail and in another

	Poison Gland	Dufour's Gland	Dufour's Gland	Hindgut
N	5	5	5	5
M ± sd	70.8 ± 24.4	20 ± 10.4	46 ± 11.3	6 ± 3.5
Range	46-109	9-36	31-60	2-10

Table 3. In the arena a poison gland trail and a Dufour's gland trail (*left*) or a Dufour's gland trail and a hindgut trail (*right*) were offered simultaneously. During the next 5 minutes the ants which followed the trails were noted. The number of replications (N) as well as the mean and range of numbers of responding workers are given. The differences in response to the two trails are significant at the following levels: *left*, $p < 0.005$; *right*, $p < 0.002$.

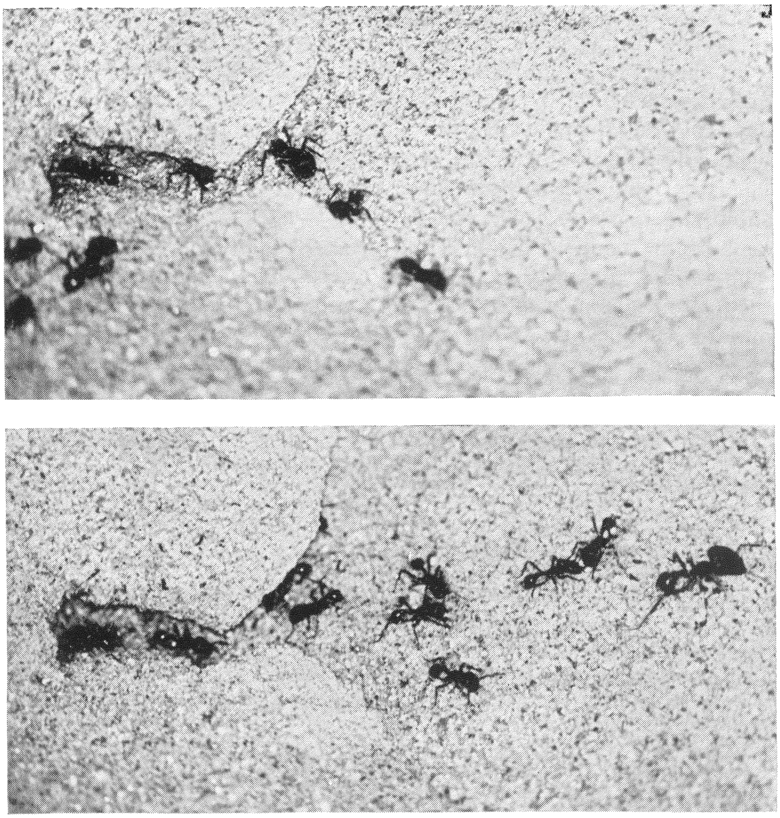

Fig. 7. *Above:* Activity at the nest entrance before a poison gland trail was laid. *Below:* Immediately after a poison gland trail was laid from left to right.

direction (45°) with a hindgut trail. In both series all ants which entered the arena during the next five minutes and which followed one or the other of the two trails were counted. The results (Table 3) indicate that the poison gland trail is much more attractive to recruited foragers than the Dufour's gland trails. However, if the poison gland trail substance is discontinued and replaced by a Dufour's gland trail, the recruited ants tend to follow the latter. Therefore it may well be that the Dufour's gland secretion also serves as an additional orientation cue in recruitment trails. This problem draws additional interest from the fact, demonstrated by the experiments to be described next, that the Dufour's gland trails are longer lasting than the poison gland trails.

In the arena a Dufour's gland trail and a poison gland trail were drawn simultaneously, beginning at the entrance to the bridge and deviating at an angle of 45°. Then after ten minutes in the first series and 25 minutes in the second series, a poison gland trail was laid from the nest entrance over the bridge to the arena entrance, stimulating ants to run immediately into the arena. A record was kept of the trail chosen by each ant as it continued on its course. The results, presented in Table 4, reveal that after 10-15 minutes the poison gland trail was still more attractive, but after 25-30 minutes significantly more ants followed the Dufour's gland trail. We conclude that the poison gland evaporates more rapidly relative to its threshold concentration than does the Dufour's gland secretion.

	10-15 Minute Interval		25-30 Minute Interval	
	Poison Gland	Dufour's Gland	Poison Gland	Dufour's Gland
	$p < 0.3$		$p < 0.002$	
N	6	6	8	8
M ± sd	39.2 ± 12.8	12 ± 6.5	7.4 ± 3.2	26.5 ± 6.07
Range	26-58	8-19	3-12	19-38

Table 4. A poison gland trail and a Dufour's gland trail were offered simultaneously in the arena. After 10 minutes (*left*) and after 25 minutes (*right*) the ants were induced to approach the two trails, and their choices were recorded during a period of 5 minutes. The number of replications (*N*) and the mean and range of numbers of responding ants are given.

*Recruitment trails in other Pogonomyrmex species**

Subsequent field and laboratory studies have revealed that recruitment behavior of the kind just described for *Pogonomyrmex badius* also occurs in other members of the genus (*P. occidentalis, P. rugosus, P. barbatus, P. maricopa, P. californicus*). In *P. maricopa* and *P. californicus* the change in the locomotory behavior of trail laying ants is particularly clear. In ordinary locomotion workers of these species hold their abdomen upwards (Fig. 8a). But when entering a new area (for example a new bridge in Fig. 1) they repeatedly touch the surface with the abdominal tip (Fig. 8b). A successful scouting ant on the other hand returns to the nest with the abdomen completely lowered and dragging the extruded sting over the surface (Fig. 8c). With the nest arrangement illustrated in Fig. 1 we were able to show that in the first case the ants apparently set orientation marks, whereas in the second case they lay

*Note added in proof.

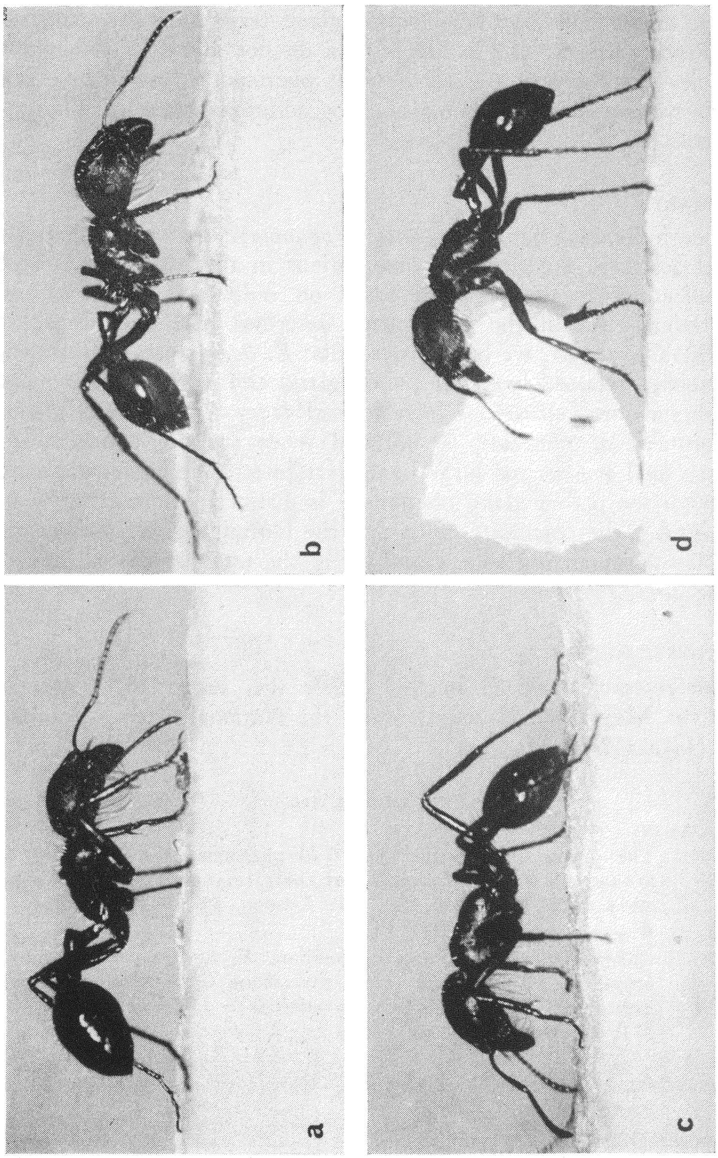

Fig. 8. *a*. *Pogonomyrmex californicus* with normal position of the abdomen. *b*. Marking a new bridge. *c*. Laying a recruitment trail. *d*. Carrying an oak flake on "tip-toes."

a recruitment trail. Artificial poison gland trails have the strongest recruitment effect. The available data do not allow us to suggest a species specificity of the recruitment pheromone. Nevertheless, a specificity may well be accomplished by additional chemical orientation cues.

Summary

Since individual harvesting ants *(Pogonomyrmex badius)* usually collect scattered seeds, casual observations in the past have seemed to indicate a foraging system based on individual initiative and orientation. But in the experiments described here and elsewhere (Hölldobler 1971), we have proved that *P. badius* uses recruitment pheromones released from the poison gland and orientation or homing pheromones released at least in part from the Dufour's gland. Recruitment of nestmates is initiated when scouting workers encounter food sources too large to retrieve in a single homeward trip. Although the poison gland pheromone is distinctly more effective in recruitment, the possibility exists that the Dufour's gland pheromone can play a supporting role, especially in the establisment of longer-lasting foraging trails.

Acknowledgements

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