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TERMITE PREDATION BY *Megaponera foetens* (FAB.) (HYMENOPTERA: FORMICIDAE)

Coordination of Raids by Glandular Secretions

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Abstract—Termite predation by the ponerine ant, *Megaponera foetens*, is coordinated by chemicals from at least two glands. Columns of ants are guided to termite foraging areas by pheromones originating from the poison apparatus. On finding groups of termites, ants release alkyl sulfides (dimethyl disulfide and trisulfide) from their mandibular glands which attract sister workers who dig, into the termite galleries, in response to other unidentified mandibular gland pheromones.

Key Words—*Megaponera foetens*, Hymenoptera, Formicidae, chemical coordination, termite predation, dimethyl disulfide, mandibular gland.

INTRODUCTION

Megaponera foetens is a termitophagous ponerine ant with a specialized recruitment and foraging regime (Collart, 1927; Wheeler, 1936; Levieux, 1966; Fletcher, 1973; Longhurst and Howse, 1979). At Mokwa, Nigeria, the main prey items are the fungus-growing termites *Macrotermes bellicosus* and *Odontotermes* spp (Longhurst et al., 1978) which forage for food over an area of several square meters at some distance from their nests. The termites are sought by single major workers referred to as 'scout ants' (Fletcher, 1973). The scout ant, having located its prey from cues in the covering of soil sheeting constructed by foraging termites (Longhurst and Howse, 1978), returns to the nest where it recruits a column of sister workers to the foraging area of the termites. During the raid, the ants spread out and break open any soil sheeting constructed by the termites and then small groups of ants dig into

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the underlying termite galleries. After the raid has been completed, worker ants pick up termites in their mandibles and the column forms again and returns to the nest by the same route taken on the outward journey (Longhurst and Howse, 1979). In this paper the use of glandular secretions in coordinating this predation by *M. foetens* on termites is described. The *M. foetens* studies relate to observations made in the field which were followed by field and laboratory experiments.

Scout ants of *M. foetens* lay a scent trail which guides the ant column to the foraging termites, and the reinforcement of this trail then appears to guide ants back to the nest (Longhurst, 1978, Longhurst and Howse, 1979). The existence of such trails was demonstrated by Levieux (1966), who found that physical removal of the trails disorientated the ants and that the trail could be obliterated with kerosene but not with water. Levieux also indicated that, in the Ivory Coast where scout ants were not found in this species, the trails were very long-lasting, and he suggested that they were colony-specific. When the trails of two colonies crossed, a column followed only its own scent trail.

The source of recruitment trail pheromones in ponerine ants appears to differ in different species. The hind gut of *Termitopone laevigata* (Blum, 1966) and the poison gland of *Leptogenys* spp. (Fletcher, 1971) have been implicated as the source of trail pheromones in these two genera. In *Leptogenys ocellifera,* an Asiatic species which feeds mainly on earthworms and termites, the poison gland secretion also has orientating and recruiting effects (Maschwitz and Mühlenberg, 1975). In the related *Leptogenys chinensis,* Maschwitz and Schönegge (1977) have found an additional gland between the last and penultimate gastral segments. This dorsal gland has a secretion that is synergistic with that of the poison gland secretion in releasing orientation and recruitment and in coordinating nest moving. In this study the use of scent trails, their source in the ant, and longevity in both field and laboratory conditions were investigated in *M. foetens.*

A second possible use of chemical signals is during the attack of the recruited ants on the termites. After arriving at the termite foraging area, ants spread out over several square meters and break into the termites' foraging galleries which are protected by soil sheeting. Sister workers are attracted to points where single ants are digging and assist them; termites are usually retrieved from these excavations. In some cases the single ants are not in direct line-of-sight with the attracted ants, suggesting that nonvisual cues may be used. Pheromones which release attraction and digging behavior have been found in other ant species. In *Acanthomyops claviger n*-undecane and other *n*-alkanes from the Dufour's gland release attraction and "excitement" (Regnier and Wilson, 1968) and in *Oecophylla longinoda n*-hexanol, part of a multicomponent pheromone system, releases attraction (Bradshaw et al.,

1975). High concentrations of 4-methyl-3-heptanone from the mandibular glands of *Pogonomyrmex* spp. release alarm and digging (Wilson, 1958) and alkyl sulfides from the mandibular glands of *Paltothyreus tarsatus* release digging behavior when the pheromone source is buried (Crewe and Fletcher, 1974). In view of these findings studies were carried out on the role of pheromones in attraction and digging in *M. foetens*. The source and identity of pheromones was also investigated. To distinguish attraction and digging from alarm behavior seen in disturbed ant columns, the alarm-defense system of *M. foetens*, and some of the chemicals controlling this system, were investigated.

METHODS AND MATERIALS

Field Observations

Field observations were carried out in primary savanna woodland, located approximately 17 km north of Mokwa, Nigeria (9°18′ N, 5°5′ E; Wood et al., 1977; Collins, 1977).

Culture Conditions

Colonies of *M. foetens*, obtained from primary savanna woodland, were kept in large Plexiglas nest boxes with plaster of Paris bases (Longhurst, 1978). At Southampton University the relative humidity was 65-85%, temperature 24-29°C and a 12-hr light-12 hr dark cycle was maintained. At Mokwa experiments were carried out at ambient temperature and humidity under a natural light cycle of approximately 13 hr light-11 hr dark. The ants had access from their nest boxes to foraging arenas, 1×0.35 m at Southampton and 1.5 m^2 at Mokwa. The foraging arenas were covered with clean sand (Southampton) or sterilized topsoil (Mokwa). Bioassays were carried out at the beginning and end of the light regime when ants were foraging for termites (*Macrotermes bellicosus* or *Reticulitermes* spp.) supplied in petri dishes.

Coordination of Attacks on Termites

Exploring scout ants recruited sister workers to the termites. Because the distance between the petri dishes of termites and the nest entrance was less than 1 meter, a single discrete column of ants (Wheeler, 1936; Fletcher, 1973, Longhurst and Howse, 1979) was not observed, but a continuous column of ants formed, moving in both directions. Some ants would leave this two-way column and forage in the arena. It was these ants which encountered the experimental stimuli. Major workers were observed as they approached a

stimulus and their behavior noted. Distances were judged in relation to 1-mm-diameter metal stakes pushed into the soil (Mokwa) or marks in the sand (Southampton).

In order to study the effects of the ants' own secretions on sister workers, body components and excised glands of ants were crushed onto filter papers (13 mm diameter). Ants for assay were killed by plunging them into a bed of dry-ice and breaking off the head and gaster from the thorax. The body sections were then crushed with a clean spatula onto filter papers, and presented by placing the paper approximately 200 mm from the edge of the foraging ant column. Further tests were carried out on extracts of active glands which had been fractionated by micropreparative gas chromatography (Baker et al., 1976). Pure chemicals were also presented to the ants in a similar manner after allowing the dichloromethane solvent to evaporate. Both fractions and pure chemicals were presented at a level of 1 gland-equivalent (GE) of the component on the filter paper.

Pure chemicals were also presented to ants foraging naturally in primary savanna at Mokwa. Columns of ants were followed on their outward journey to sites where they attacked termites, and tubes containing the chemicals, or control tubes, were sunk into the soil about 200 mm to the side of the foraging trails. The reactions of the ants were then observed on their return journey. The chemicals for assay were injected into melting-point tubes (1 mm OD × 30 mm long, sealed at one end). When dimethyl disulfide and dimethyl trisulfide were tested, 5 μl was injected into each tube. For benzyl-methyl sulfide a solvent-free preparative technique was used (Longhurst 1978). The compound was eluted from the gas chromatograph (GC) into an open-ended tube (1 mm OD) surrounded by dry ice. The tube was sealed at both ends until needed for use, when one end was removed to leave a tube 30 mm long containing about 200 μg of the synthetic compound.

Alarm Behavior

Field and laboratory observations were made in order to distinguish alarm behavior from other chemically mediated behavior. Two states of alarm behavior were defined, preceded by an alerting stage. An alerted ant was in a state of arrest, with its antennae porrect, usually oriented towards the stimulus. Low-intensity (directional) alarm behavior was defined as that in which the ant stood with its antennae porrect and mandibles open. The ant sometimes approached the stimulus with its antennae and mandibles held in these positions. High-intensity (undirectional) alarm was characterized by the ants moving rapidly in all directions, often colliding with sister workers. Bursts of stridulation (Markl 1973) were often heard during this type of alarm. To investigate intraspecific alarm communication, ants were presented

with crushed gasters, Dufour's glands and poison glands, crushed on 13-mm-diameter filter papers.

The Dufour's gland was examined by GC (Figure 2) and some of the components were identified. *n*-Undecane and *n*-tridecane, two of the major components, were presented both to ants in culture and to foraging columns of ants using the methods described previously.

Trail-Following Behavior

Field Studies on Duration of Trails. In order to establish the duration of recruitment trails in the field, columns of ants foraging in the morning (Longhurst and Howse, 1979) were observed and the trails marked with wooden stakes (1 mm diameter) at 0.2-m intervals. Major workers were captured from the returning column of ants by trapping them separately in specimen tubes. At recorded times the ants were released onto the trail at least 15 m from the nest. The tube was inverted over the trail and left for at least 60 sec before the ant was released. It was then followed for at least 10 meters if it followed the trail or 300 sec if it lost the trail and entered the surrounding vegetation. A number of ants were also released 2 meters to the side of the trail and observed.

Laboratory Studies on Source of Trail Substances. Twenty major and twenty minor workers from cultures at Southampton were placed in traps made from 100 × 50-mm polythene containers, with a sliding door at one end. Extracts of various parts of major workers, killed by plunging in dry ice, were made in purified hexane at the following concentrations: 1 head + thorax in 500 μl hexane; 1 gaster in 500 μl; 1 Dufour's + poison gland on sting apparatus in 2 ml; 1 hind gut, removed by dissection in 1 ml; 1 poison gland in 2 ml; and 1 Dufour's gland in 1 ml.

The Dufour's and poison glands were separated by dissection under distilled water; any ruptured glands were rejected. The hind gut was taken as a piece of gut approximately 5 mm long, starting from a point as close to the anus as possible.

The extract (100 μl) to be tested was taken up in a Drummond microcapillary tube and streaked along a convoluted pencil line, about 250 mm long, on cartridge paper (300 × 200 mm). Presentations to sister workers were made so that any ant had to make a choice between two potential trails. Presentations were paired as follows: head + thorax with gaster; Dufour's + poison gland with solvent blank; hind gut with pencil blank; poison gland with hind gut and Dufour's gland with poison gland.

The start of the trail was made flush with the sliding door of the trap. Ants were released from the trap and scored as responding positively if they followed a trail for 200 mm from the door. A fresh piece of cartridge paper was

used for each ant, as observations suggested that ants could be laying trails of their own on the paper.

To compare trails laid by foraging ants with poison gland extract from sister workers a different experimental situation was used. Petri dishes of termites were placed under the traps used above and ants were allowed to forage to them and establish trails approximately 500 mm long. When the ants had finished foraging, 20 major and 20 minor workers from the same colony were placed in the trap which was replaced in its original position in the foraging arena. The natural trail was fairly wide (about 60 mm), but narrowed where it entered the trap, and was in a straight line between the trap and the nest entrance. The artificial trail was laid from the trap to the nest at one side of the natural trail, and was slightly longer than the latter. The ants were released from the trap and scored according to which trail they followed. No attempt was made to provide a new trail for each ant, but ten trials were made with one natural trail against 0.1 or 1.0 GE of poison gland extract. Statistical analysis, using the binomial test (Siegel, 1956), was carried out to compare the two trail stimuli presented.

Identification of Mandibular and Dufour's Gland Components

Identification was carried out by gas chromatography on 5% Carbowax 20M (A), 5% OV101 (B), 5% PPGA (C) and 10% E30 (D) 2-mm ID × 3-m all-glass columns with nitrogen carrier gas. Excised glands were examined using solid-sample gas chromatography (SSGC) (Morgan and Wadhams, 1973). Whole ant heads were examined using a modified Morgan-Wadhams solid sampler which allowed the use of biological material with a maximum diameter greater than 1 mm (Longhurst, 1978). Extracts of heads and excised Dufour's glands were made in purified dichloromethane which was reduced in volume under oxygen-free nitrogen for analytical work. GC-mass spectra were obtained on a MS 12 with a VG Digispec 16 data system calibrated with perfluorokerosene.

Synthesis

Benzylmethyl Sulfide. Sodium hydrogen sulfide reagent was made by the method of Khormandaryan and Brodovich (1930). Benzyl chloride was refluxed with a 50% ethanolic solution of the reagent (50 ml ethanol, 50 ml of aqueous NaSH solution) for 4 hr. The reaction mixture (75 ml) was added to saturated sodium hydroxide solution (100 ml). Dimethyl disulfide (50 ml, Koch Light) was added and the solution refluxed for three hours. The products were taken up in ether, and this extract was dried with magnesium sulfate. The benzylmethyl sulfide was purified by GC (column A) and the product trapped in cooled capillary tubes (Longhurst, 1978).

Dimethyl Trisulfide. Dimethyl disulfide (10g) was refluxed with sodium hydrogen sulfide solution (30 ml) in ethanol (30 ml) with the addition of sulfur (5 g) and ethylamine (5 ml). The reaction mixture was refluxed overnight, the products taken up in ether, and the extract dried with magnesium sulfide; the product was purified as above.

RESULTS

Identification of Exocrine Secretions

Mandibular Glands. Modified SSGC studies showed the presence of at least 13 compounds (Figure 1) and that the castes did not differ in the proportion or patterns of compounds. In the major workers, the most abundant component (B, Figure 1) was present at the level of only 20 ng/ant. SSGC-MS and GC-MS studies with solvent extracts produced the following information about the major mandibular gland components.

Component B was identified as dimethyl disulfide by comparison of its

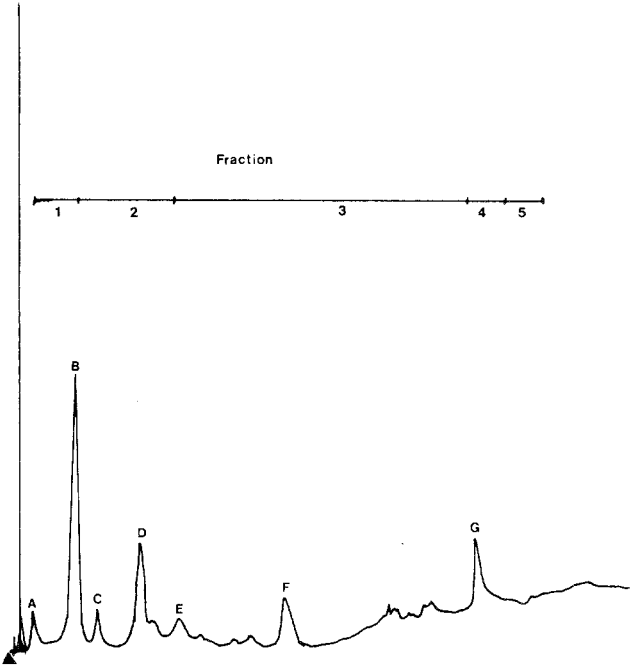

FIG. 1. Chromatogram of the mandibular gland secretion of a major worker of *Megaponera foetens*. 5% Carbowax 20M; 80–180°C at 6°/min.

mass spectrum with published spectra (Crewe and Fletcher, 1974) and co-elution with an authentic sample on GC columns A, C, and D.

The mass spectrum of component C revealed a base peak at m/e 108 (100%) which was also the molecular ion and large fragments at 80 (89), 45 (49), 47 (33), 76 (31), 94 (28), 79 (26). Fragments at m/e 109 (4) and 110 (11) suggested a sulfur-containing compound and the large fragment at 80 (89%) is probably from the loss of C_2H_4 from the molecule. No published mass spectra corresponding to the natural product could be found. The evidence points to a compound with the empirical formula $C_3H_8S_2$; the loss of the ethyl radical suggests $C_2H_5S_2CH_2$, ethylmethyl disulfide, as a possible structure.

The mass spectrum of component D corresponded to the published spectrum of dimethyl trisulfide (Crewe and Fletcher, 1974). A synthetic sample gave a mass spectrum congruent with, and that coeluted with, the natural product on GC columns A, C, and D.

Component F exhibited a molecular ion at m/e 138 (89%) with additional fragments at 139 (2%) and 140 (8%), characteristic of the isotopic ratio of sulfur. The base peak at m/e 91 ($C_7H_7^+$) is characteristic of an aromatic ring with a substituted methyl side chain ($C_6H_5CH_2X$). The loss of 47 from the molecular ion to form the base peak (91, 100%) suggests the loss of CH_3O_2 or CH_3S, the latter being most likely because the presence of sulfur was indicated by the isotopic ratios. A synthetic sample of benzyl-methyl sulfide gave a mass spectrum congruent with the natural product and coeluted on GC columns A, B, and D.

Dufour's gland. The identity of the compounds found in the Dufour's gland (Figure 2) are presented in Table 1. Chromatograms from *M. foetens* workers of all sizes exhibited similar proportions of components.

Poison Gland. Although crushed poison apparatus had a strong ester-like odor, only 10 minor components (1–2 ng, major workers) could be resolved by GC. Attempts at identification were not successful.

Alarm Behavior

Alarm behavior was defined in the Methods and Materials section. High-intensity (undirectional) alarm behavior was released by crushed gasters, crushed Dufour's glands, as well as two of the major components of this gland: *n*-undecane and *n*-tridecane.

Coordination of Attacks on Termites

Worker ants, when presented with crushed heads, turned towards them and then moved rapidly towards the stimulus. The ants surrounded the odor source, making biting movements with their mandibles into the sand and the filter papers, and digging into the sand with their forelegs. Two major be-

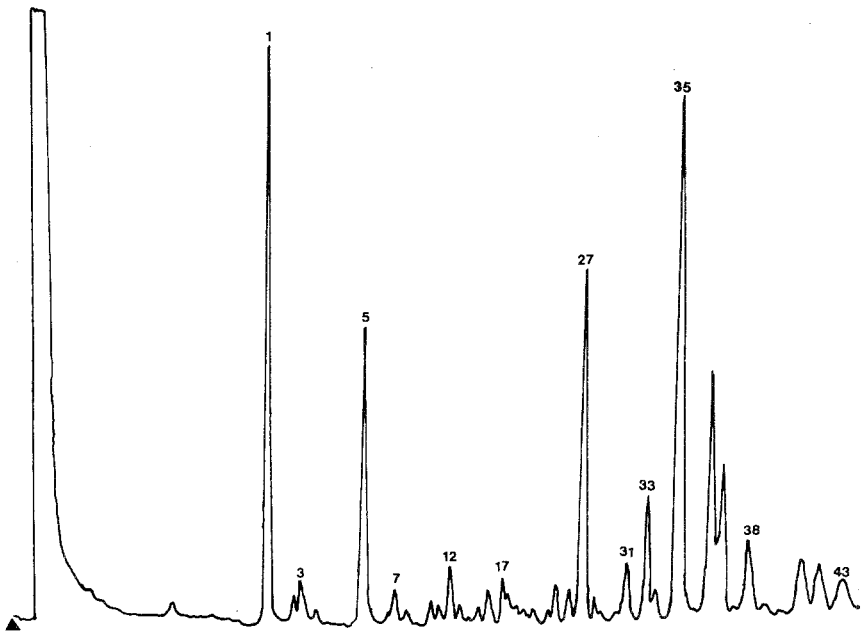

FIG. 2. Chromatogram of the Dufour's gland secretion of a major worker of *Megaponera foetens*. 5% OV101; 70-250°C at 8°/min.

havioral components were thus distinguishable—attraction and digging. Excised, crushed mandibular glands elicited the same reaction as crushed whole heads (Table 2).

Presentation of crushed thoraces elicited no reaction. Ants presented with crushed gasters were initially alerted, and then showed low-intensity alarm. A period of high-intensity alarm then followed. As this high-intensity alarm subsided (30–50 sec after presentation), a number of ants approached the odor source slowly and then stood around the crushed gaster, biting at it. Some of the ants remained with their mandibles fixed in the remains of the gaster or the filter paper for up to 60 sec. The rapid biting and stinging seen in reaction to some species of crushed termite soldiers (Longhurst, 1978) was not observed. The overall reaction to crushed gasters was a complex one, probably because the contents of more than one gland were released on crushing.

Of the reactions to crushed body sections, only the behavior elicited by crushed heads was similar to that seen in columns of ants after they spread out after locating the termite foraging area and commenced their attack on groups of termites. Ants were attracted to a sister worker digging into the soil at the entrance to the termite galleries, and then also began to dig. In view

TABLE 1. IDENTIFICATION OF SOME COMPONENTS IN DUFOUR'S GLAND OF *Megaponera foetens*[a]

Component No.	Identity	Method of identification[a]
A	*n*-Decane	LP,RT
1	*n*-Undecane	MS,LP,RT
4	*n*-Dodecane	LP,RT
5	*n*-Tridecane	MS,LP,RT
8	*n*-Tetradecene	LP
9	*n*-Tetradecane	MS,LP
12	*n*-Pentadecene	MS,LP
13	*n*-Pentadecane	MS,LP,RT
17	*n*-Hexadecene	MS,LP
18	*n*-Hexadecane	LP,RT
24	*n*-Heptadecene	LP
27	*n*-Heptadecane	MS,LP,RT
31	*n*-Nonadecene	MS,LP
33	*n*-Nonadecane	MS,LP
38	*n*-Heneicosene	LP
39	*n*-Heneicosane	MS,LP
41	*n*-Docosane	MS,LP
43	*n*-Tricosane	LP

[a]MS, mass spectrometry; LP, log plot; RT, relative retention time with standard.

of these observations the reactions of ants to crushed excised mandibular glands, and fractions prepared by GC (Figure 1) for these glands, were observed.

Crushed mandibular glands released the same behavior as did crushed heads. When fractions 1 and 2 (Figure 1) were presented worker ants orientated towards the odor source and then rapidly moved towards it. They did not stop at the odor source, but usually overshot by 30-50 mm, and then turned about and reorientated towards it. The ants eventually explored the odor source, moved away, but were usually attracted back towards it until 60-120 sec after their initial orientation to it. No reaction was observed to either fractions 3 or 5 (Figure 1). Worker ants encountering the treated filter papers showed no difference to ants encountering solvent-treated controls.

Ants encountering a filter paper, treated with fraction 4 (Figure 1), in the course of their explorations usually bit at the odor source and dug into the surrounding sand with their forelegs, in a similar manner to that observed in response to crushed heads. When fractions 1 and 4 were presented together, both attraction to and digging at the odor source were observed; this was the same as behavior observed to crushed heads.

Three of the known constituents of the mandibular glands were then bioassayed. Dimethyl disulfide and dimethyl trisulfide were present as the main components in active fractions 1 and 2, respectively, and benzylmethyl sulfide was present in inactive fraction 3. The reactions of ants to dimethyl disulfide and trisulfide, at a level of 100 ng/presentation, was the same as the reactions of ants to fractions 1 and 2, namely attraction to the odor source. Ants did not respond to benzylmethyl sulfide at a level of 50 ng/presentation. Dimethyl disulfide (20 ng), presented with 1 GE of fraction 4, released the same behavioral repertoire as crushed heads—attraction to the odor source followed by digging at and around the source. If dimethyl disulfide (on filter paper) was buried in the sand of the foraging arena, ants

TABLE 2. REACTIONS OF *Megaponera foetens* TO CRUSHED BODY SEGMENTS AND MANDIBULAR GLAND FRACTIONS AND COMPOUNDS FROM SISTER WORKERS.

Presentation	Concentration	No. of presentations	Behavior observed	No. of times behavior observed[a]
Crushed heads	1	10	Approach, digging	10
Crushed thoraces	1	10	No reaction	
Crushed gasters	1	10	Undirected alarm, followed by approach, arrested at source	10
Mandibular glands	1 pair	10	Approach, digging	10
Fraction[b]				
1	1 GE[f]	10	Approach	10
2	1 GE	10	Approach	8
3	1 GE	5	No reaction	
5	1 GE	5	No reaction	
4	1 GE	15	Digging	10
DMDS[c]	100 ng	10	Approach	9
DMTS[d]	100 ng	5	Approach	5
BMS[e]	50 ng	5	No reaction	
DMDS + fraction 4	20 ng + 1/2 GE	10	Approach, digging	7
DMDS buried in sand	100 ng	5	Approach	5

[a]At least one ant responding.
[b]Mandibular gland fractions (Figure 1).
[c]Dimethyl disulfide.
[d]Dimethyl trisulfide.
[e]Benzylmethyl sulfide.
[f]Gland equivalent.

responded in the same manner as they did to the compound presented on the surface. These results show that burying this alkyl sulfide does not release digging behavior in *M. foetens*, as it does in *Paltothyreus tarsatus* (Crewe and Fletcher, 1974). The reactions of *M. foetens* workers to mandibular gland contents are summarized in Table 2.

After the presentation of dimethyl disulfide to foraging ants, workers left the foraging column, oriented towards the stimulus, and then moved rapidly towards it. If the ants overshot the stimulus, they usually reoriented towards it. This behavior continued for up to 150 sec, after which the ants either located the trail again and rejoined the remainder of the column, or moved into the surrounding vegetation. On some occasions, especially when large numbers of ants were moving towards the odor source, high-intensity alarm occurred, with bursts of stridulation. The overall reaction to the disulfide was attraction, but the large numbers of ants involved in the interaction appears to have released alarm. In similar conditions, neither benzyl-methyl sulfide nor control tubes elicited any reaction.

TABLE 3. ABILITY OF MAJOR WORKERS OF *Megaponera foetens* TO FOLLOW NATURAL RECRUITMENT TRAILS LAID BY COLUMNS OF SISTER WORKERS DURING MORNING RAIDS ON TERMITES[a]

Hours elapsed since trail was laid	Number of major workers (out of 10) following the trail for at least 10 meters	Number of major workers (out of 5) which, after being released 2 meters to the side of the trail, moved in the direction of the trail for 10 m
0.25	9	0
0.5	8	
0.75	6	
1.0	7	1[b]
1.5	6	
2.0	5	
2.5	5	
3.0	1	0
3.5	0	
4.0	0	
4.0	0	
7.0	0	0

[a]Trails followed for at least 10 meters.
[b]Trail relocated and followed to the nest.

TABLE 4. TRAIL-FOLLOWING RESPONSES OF *Megaponera foetens* MAJOR AND MINOR WORKERS TO EXTRACTS OF PARTS OF THE BODY OF SISTER WORKERS

Extract[a]	No. of major workers (of 20)	No. of minor workers (of 20)
Head/thorax	4	2
Gaster	14	6
No trail followed	2	12
Dufour's/poison gland	13	6
Solvent blank	1	0
No trail followed	6	14
Hind gut	4	3
Pencil blank	0	0
No trail followed	16	7
Poison gland	17	10
Hind gut	2	4
No trail followed	1	6
Dufour's gland	3	6
Poison gland	15	11
No trail followed	2	3
Natural trail	12	12
0.1 GE poison gland	7	5
No trail followed	1	3
Natural trail	3	
1 GE poison gland	16	
No trail followed	1	

[a]For extract concentrations see Methods and Materials.

Trail-Following Behavior

Duration of Trail in Primary Savanna. The results of releasing major workers along existing trails of their own colony, at different time intervals after its origin, are presented in Table 3. Trails showed some activity for up to 3 hr, but their efficiency was greatly reduced after $1\frac{1}{2}$ hr.

Source of Trail Substance. Fourteen major workers followed the gaster extracts and four the head + thorax extracts (Table 4). Although these differences were not statistically significant ($P = 0.09$), further studies were carried out on gaster extracts because it was believed that the presence of alkyl sulfides in the head extracts, which released attraction, may have caused the ants to follow the head + thorax extract trails.

The Dufour's + poison gland complex proved to be a more powerful releaser of trail-following behavior in major workers than the hind gut ($P =$

0.01), and separate assays on these two glands showed that the poison gland was more active than the Dufour's gland ($P = 0.04$), suggesting that the former gland is the major source of trail pheromones in *M. foetens*. Comparison of the poison gland extract with the natural trail showed that at a level of 0.1 GE, 12 of 20 major workers followed the natural trail and 7 of 20 the poison gland extract ($P = 0.08$). At the higher presentation level of 1 GE, 16 ants followed the extract and 3 the natural trail ($P = 0.01$).

Minor workers were less competent at following artificial trails than major workers (Table 4). When the head + thorax extract was compared with the gaster, only 6 ants followed the latter, 2 ants followed the head + thorax extract, and 12 ants did not follow any trail at all. Comparison of different glands in the gaster did show that major worker poison gland extract was more potent than that of the Dufour's gland or hind gut. The results were not always statistically significant because of the large number of ants not following any trail at all.

DISCUSSION

The obligate termite predator, *M. foetens*, shows at least two refinements of its chemical communication system to allow it to act efficiently as a predator of foraging termites. Prevous authors (Levieux, 1966; Fletcher, 1973) have demonstrated the use of trails by *Megaponera*, and in this study the trail substance has been shown to originate in the poison gland. No attempt was made to identify the trail pheromone(s), although SSGC of excised poison glands did reveal the presence of more than 10 compounds, all at a level of a few nanograms per major worker.

In the related ponerines, *Leptogenys attenuata* and *L. nitida*, Fletcher (1971) has demonstrated that the source of the trail pheromone is also in the poison gland. In both *Leptogenys* species the trail pheromone is used in the coordination of emigration to new nests and in *L. nitida* is also used in the recruitment of ants to their isopod prey. In the termitophagous ponerine, *Termitopone laevigata*, the glandular source of the trail pheromone is the hind gut (Blum, 1966).

To allow efficient utilization of the foraging termite party, the column of ants must arrive as a group. An odor trail achieves the efficient guiding of the ants to the foraging termites where other chemical cues can complete the coordination of predation. The scent trails at Mokwa did not last more than 3 hr; which in an area where foraging occurs only in the morning and evening (Longhurst and Howse, 1979) may be of use in preventing confusion with the colonies' trails from previous raids. In the Ivory Coast, where the details of recruitment differ from Mokwa and a scout ant is not used, Levieux (1966) claims that the trails last for up to 24 hrs. It is possible that the different foraging pattern observed by Levieux has led to a lower threshold of detection

of the trail odors. The low level of response of minor workers to artificial trails may reflect the fact that this caste never needs to follow trails in the absence of major workers (Longhurst and Howse, 1979) and may stay with the foraging column by visual or tactile means.

Mandibular gland pheromones from ponerine ants have been found to be very diverse. In *Gnamtogenys pleurodon* methyl-6-methylsalicylate acts as an alarm pheromone (Duffield and Blum, 1975), as do 4-methyl-3-heptanone in *Neoponera villosa* (Duffield and Blum, 1973) and alkyl pyrazines in *Odontomachus*, *Hypoponera*, and *Ponera* species (Wheeler and Blum, 1973; Duffield et al., 1975; Longhurst et al., 1979). Dimethyl disulfide and trisulfide have been found previously in the ponerine *Paltothyreus tarsatus* (Crewe and Fletcher, 1974) where, like 4-methyl-3-heptanone in *Pogonomyrmex* spp. (Wilson, 1958) they act as releasers of digging behavior. By contrast, in *M. foetens* these two components are attractants, acting in conjunction with other unidentified mandibular gland components which release digging, as part of a multicomponent pheromone system. Field observations and tests support the interpretation of these alkyl sulfides as part of the predation system, and not just alarm attractants. After an ant has broken open the entrance to a termite foraging gallery, other ants move towards it from all directions. They then distribute themselves around the gallery entrance and commence to dig before any minor workers enter the galleries and predate the termites. This regular spacing does suggest that other pheromones—perhaps the digging pheromone in fraction 4—may be repellent at high concentrations or that other unknown stimuli may be involved.

Although the trail pheromone leads the ants to the termite foraging area, it does not locate ants at all the points were termites are foraging in the area. A second chemical system (attraction and digging) coordinates the final attack within the overall foraging area and allows a more efficient utilization of the prey. If each ant had to search out its prey, or if groups of ants had to be led to foraging termites by new recruitment trails, many termites might escape. If a number of ants can be concentrated at the entrance to the termite galleries, more prey can be secured if the retreating termites can be cut off before they reach the gallery entrance.

Some comment must be made on the different roles of dimethyl disulfide in *M. foetens* and *Paltothyreus tarsatus*. In the latter species Crewe and Fletcher (1974) found that the workers were ". . . indifferent to crushed worker heads and filter papers impregnated with [1 μl of] dimethyl disulphide," but that buried whole ants and buried filter papers treated with 0.1 μl of the disulfide elicited digging and rescue of ants or filter paper. The lack of response to unburied compound is difficult to explain; Crewe and Fletcher suggest that as *P. tarsatus*, unlike *M. foetens*, lacks stridulatory organs (Markl, 1973), the response is needed only to release sister workers trapped in nest falls. In *M. foetens* dimethyl disulfide has a completely dif-

ferent function, as an attractant in a multicomponent system with another less volatile component (fraction 4) that releases digging. The amounts of dimethyl disulfide and dimethyl trisulfide also differ markedly in each species. *P. tarsatus* contains 4.6 μg of the disulfide and 23.3 μg of the trisulfide, whereas *M. foetens* major workers contain 28 and 10 ng, respectively, of the two compounds.

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