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# TRAIL-FOLLOWING BEHAVIOR OF *Reticulitermes hesperus* BANKS (Isoptera: Rhinotermitidae)

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**Abstract**—The behavior of *Reticulitermes hesperus* Banks pseudergates (workers) was assessed on artificial trails containing different concentrations of sternal gland extract. On nongradient trails, more pseudergates were recruited to trails of greater pheromone concentration, they traveled a greater distance without pausing, and their rate of locomotion increased over that observed on trails of lesser concentration (positive orthokinesis). Of the individuals pausing before completing trails of high concentration, fewer left the trails or reversed direction (negative klinokinesis) than on trails of lower concentration. Termites walking down concentration gradients failed to complete these trails to the low-concentration termini. At a point representing an average decrease of slightly more than 10-fold in the original concentration of pheromone, individuals reversed their direction of travel and returned to the high-concentration terminus. Termites walking up pheromone gradients proceeded to the high-concentration termini without reversing direction. *R. hesperus* pseudergates are thus able to orient along a gradient of trail pheromone by longitudinal klinotaxis.

**Key Words**—Termite, *Reticulitermes hesperus*, Isoptera, Rhinotermitidae, pheromones, trail following, orientation.

## INTRODUCTION

*Reticulitermes* spp. (Isoptera: Rhinotermitidae) establish chemical trails with pheromones secreted by the sternal gland, an epidermal gland located in the anterior portion of the fifth abdominal segment beneath the fourth abdominal sternite (Mosconi-Bernardini and Vecchi, 1964; Smythe and Coppel, 1966;



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Quennedey, 1971; Liang et al., 1979). The trail pheromone of *Reticulitermes virginicus* Banks was identified by Matsumura et al. (1968) as *cis*-3,*cis*-6,*trans*-8-dodecatrien-1-ol. This compound is attractive to other *Reticulitermes* spp. (Matsumura et al., 1972) and has been implicated as the pheromone of *Reticulitermes lucifigus* var. *santonensis* (Feytaud) (Ritter and Coenen-Saraber, 1969) and *Reticulitermes speratus* Kolbe (Honda et al., 1975). It may also be closely related to that of other species (Howard et al., 1976) or represent one component in a multicomponent trail pheromone (Kaib et al., 1982; Prestwich et al., 1984).

Traniello (1982) and Hall and Traniello (1985) consider termite trail-following behavior to consist of both recruitment and orientation components. Additionally, there are two aspects to orientation: orientation to the lateral boundaries of the trail space (cf., Bossert and Wilson, 1963) and orientation along the longitudinal axis of the trail. With respect to longitudinal orientation, we describe here the concentration-dependent responses elicited by *Reticulitermes hesperus* Banks pseudergates by different concentrations of sternal gland extract on artificial trails. Orientation responses were evaluated both on trails drawn with single concentrations of sternal gland extract and on trails containing discrete (incremental) concentration gradients of extract.

The significance of evaluating trail orientation with respect to pheromone gradients lies in the intriguing possibility that differences in pheromone concentration could indicate directionality on trails. Although behavioral assays with *Hospitalitermes sharpi* (Holmgren) (Jander and Daumer, 1974), *R. flavipes* (Runcie, 1983), and *Trinervitermes trinervoides* (Sjöstedt) (Tschinkel and Close, 1973) indicated that these species could not distinguish the direction of their nest from the direction of foraging areas, these assays employed short segments of naturally laid trails. Any differences in trail pheromone concentration over a short distance might not be sufficient to elicit changes in termite orientation. Leuthold (1975) suggested, based on observations of grass-feeding termites, *Trinervitermes* spp., that orientation could be facilitated by the presence of a pheromone gradient over an extensive network of trails.

These studies emphasize mechanisms of individual termite orientation. Rather than attempting to draw conclusions about behavior to unknown stimuli on natural trails, we chose to define the behavioral responses to a series of known stimuli on artificial trails. Thus, with the individual behavioral parameters established, investigation of the unknown natural stimuli, in the appropriate social context, can follow.

## METHODS AND MATERIALS

*Source of Insects and Gland Extracts.* In behavioral assays we used undifferentiated *R. hesperus* pseudergates (workers) older than the third instar (as

determined by size) from two colonies. One of these was removed from severely infested Douglas-fir (*Pseudotsuga menziesii* (Mirb.) Franco) wood substructure framing in a home in Alameda County, California (Oakland). The other colony was from a Douglas-fir soil-retaining board on the grounds of a University of California family housing complex in the same county (Smyth). After collection, termites were removed from the wood and placed in plastic trays containing a small block of the Douglas-fir, damp cotton, and several pieces of Whatman No. 1 filter paper as feeding substrates. These trays were maintained in a humidity chamber at 94 ± 5% relative humidity, 21–25°C (Grace, 1986). Individuals from the two colonies were kept separate from each other and assayed only with extracts of pseudergates from the same colony.

Stock sternal gland extracts containing trail pheromone (1× dilution) were prepared by removing the fourth and fifth sternites from 10 *R. hesperus* workers (immobilized by exposure to Dry Ice) and extracting these sternites in 1 ml dichloromethane (Baker analyzed) for 24 hr. Tenfold (0.1×), 100-fold, and several lesser dilutions (0.0625× and 0.03125×) were prepared from the stock solution. Artificial trails containing higher concentrations of trail pheromone than the stock solution were prepared by repeatedly overlaying trails with the stock solution.

*Assay Conditions.* Each artificial trail consisted of a straight 100, 150, or 200-mm line, 1–2 mm in width, drawn on Monroe No. 41 parchment tracing paper (5 × 25 cm) with a microliter syringe containing 1 μl of extract per 50 mm of trail. A thin, uniform line of solvent could be more readily applied to this particular paper than to others that were tested. Preliminary tests indicated that trails drawn on this paper with pure solvent and with extracts of the paper itself did not elicit any noticeable behavioral response (e.g., trail following, arrestment, repellence). A light pencil line on the underside of the paper served as a guide for extract application and as a reference during the assay. Our extract application rate of the stock glandular extract (1×) at 1 μl/50 mm of trail was equivalent to a concentration of $2 \times 10^{-4}$ glandular equivalents (GE) per millimeter of trail.

The papers were placed on a glass surface and uniformly illuminated by overhead fluorescent lighting (13.5–19.5 foot-candles). All assays were performed at ambient conditions (21–25°C, 35–45% relative humidity), which were standardized throughout each experiment by regulating the laboratory temperature. In each assay, an artificial trail was drawn with extract and the solvent allowed to evaporate for ca. 15 sec. A single *R. hesperus* pseudergate was gently deposited from a small glass vial onto one end of the trail. Timing of the assay and measurement of distance traveled on the trail was begun when the insect moved a distance greater than one body length in any direction. In experiments where distance traveled on the trail was the critical parameter, only the distance traveled on new (previously untraveled) portions of the trail was considered. The points of initiation and termination of locomotion along the trail

axis were marked lightly in pencil at the base of the paper for later measurement. In all of our experiments, a new trail was drawn for each individual assay, and each insect and each trail were used only once to prevent any effects from behavioral conditioning or trail reinforcement.

*Orientation on Nongradient Trails.* Two experiments were performed on nongradient artificial trails drawn with different dilutions of sternal gland extract. The first experiment was designed to determine whether a concentration-dependent relationship existed in the response to artificial trails and longitudinal orientation upon them. We recorded the distance traveled in 0.5, 1.0, 2.0, or 3.0 min by pseudergates from the Oakland colony on 200-mm artificial trails of either stock (1×), 0.1× or 0.01× dilute extract and control trails of dichloromethane. Any movement along the trail axis was scored as a positive response, with the short movements recorded along the control trails representing the background level of behavioral "noise" in the assay. Twenty-five individual assays were performed for each time interval with each of the three concentrations of glandular extract and the dichloromethane control, for a total of 16 independent treatments.

Our second experiment was conducted to examine concentration-mediated differences in orientation behavior following initiation of trail following. Pseudergates from the Oakland and Smyth colonies were deposited at one end of 150-mm trails containing glandular extract (from their respective colony-mates) in either 10×, 1×, or 0.1× concentration. Trails of the 0.01× concentration and solvent controls were not included in this experiment since these elicited very little or no trail following in the first experiment. We recorded the distance traveled on each trail without pausing or leaving the trail, the time taken to travel that distance (rate of locomotion), and the behavior of those that did not immediately complete the 150-mm trail (i.e., continuing forward after pausing on the trail, leaving the trail, or reversing direction). Thirty individuals from each colony were tested with each of the three concentrations of glandular extract.

*Orientation on Gradient Trails.* Three separate experiments were performed on artificial trails consisting of a concentration gradient of sternal gland extract. In the first experiment, a pseudergate from the Oakland colony was deposited at either the high or the low concentration end of a 100-mm artificial trail containing a 20-fold arithmetic gradient of either the stock 1× sternal gland extract or its 10-fold dilution. These 0.1×–2× and 1×–20× arithmetic gradients were created by repeatedly overlaying successive 5-mm increments of the trail with either the stock 1× extract or the 0.1× dilution. Thus, individuals deposited at one end of the trail were initially either walking up or down a steep gradient of trail pheromone. Under these conditions we recorded the number of termites completing trails in a 3-min period, the time taken to complete trails, and the number reversing their direction of travel before reaching the end of the trail. Behavior of pseudergates walking up or down a pheromone gradient was

compared to behavior on nongradient trails containing only the 1× extract or the 0.1× dilution. Twenty-five individual assays each were performed walking up the gradient, down the gradient, and on nongradient trails of the 1× stock extract, while 20 individual assays in each of these three treatments were performed with the 0.1× dilution.

A second experiment was performed to supplement observations made during the first experiment with respect to the distance traveled by individuals walking down a pheromone gradient before reversing their direction of travel. Twenty-five pseudergates from the Oakland colony were individually tested walking down 20-fold gradients (100 mm in length) and on nongradient artificial trails drawn with 0.0625× and 0.03125× dilutions of the stock extract. These dilutions were based upon the earlier observation that a 0.01× dilution of the stock extract elicited very little trail following. The proportions of individuals completing trails and reversing direction without completing trails were compared on gradient and nongradient trails at each dilution.

Our third experiment measured the behavior of 30 pseudergates each from the Oakland and Smyth colonies walking either up or down a 10×–1×–0.1× incremental logarithmic pheromone gradient in individual assays on 150-mm artificial trails. We compared the distances traveled without reversing direction and the proportion of pseudergates in each treatment completing trails without reversing direction.

*Analyses.* Statistical analyses employed two-tailed $t$ tests and analysis of variance (ANOVA) with the rank transformation (SAS Institute, 1982). This ANOVA is equivalent to the Kruskal-Wallis test with the $F$ approximation (Conover and Iman, 1981; Quade, 1966). Comparison of means was performed with the Tukey-Kramer method (Kramer, 1956) or the Ryan-Einot-Gabriel-Welsch (REGW) multiple $F$ test ($\alpha \leqslant 0.05$). Proportions were compared with multiple $Z$ tests using Bonferroni's inequality to maintain $\alpha \leqslant 0.05$ (Dixon and Massey, 1983).

## RESULTS

*Orientation on Nongradient Trails.* In the first experiment on nongradient trails, the proportion of pseudergates initiating trail following increased significantly as the concentration of glandular extracts increased (Table 1). With the 23% overall response recorded to the dichloromethane control trails representing random movements, almost twice as many individuals (41%) responded to the 100-fold (0.01×) dilution of glandular extract. The number of respondents increased significantly (90% and 99%) at the 0.1× and 1× concentrations. Conversely, there was no apparent relationship between the length of the exposure time and the number of individuals responding to trails (Table 1).

The mean distance traveled on the 200-mm trails by responding pseuder-

TABLE 1. NUMBER RECRUITED AND MEAN DISTANCE TRAVELED IN DIFFERENT TIME INTERVALS BY *Reticulitermes hesperus* PSEUDERGATES ON 200-mm ARTIFICIAL TRAILS OF STERNAL GLAND EXTRACT

| Trail concentration[a] | Total percent recruited[d] | Number recruited ($N$) and mean distance ± SEM(mm)[c] | | | |
|---|---|---|---|---|---|
| | | 0.5 min | 1.0 min | 2.0 min | 3.0 min |
| 1× | 99a | (25) 181.72 ± 7.92a | (25) 186.60 ± 8.96a | (25) 191.92 ± 6.03a | (24) 196.33 ± 3.67a |
| 0.1× | 90b | (23) 94.22 ± 13.20b | (22) 126.73 ± 14.99b | (23) 124.70 ± 14.70b | (22) 149.00 ± 13.81b |
| 0.01× | 41c | (7) 26.86 ± 9.09c | (12) 23.08 ± 3.92c | (12) 25.08 ± 4.53c | (10) 37.30 ± 13.09c |
| Control | 23d | (6) 10.33 ± 2.22c | (6) 13.00 ± 1.91c | (6) 13.50 ± 2.87c | (5) 17.20 ± 3.51c |

[a] 1× = 10 sternal glands per milliliter dichloromethane, applied at the rate of 1 μl per 50 mm of trail. Control = dichloromethane.

[b] Total percent recruited in all four time periods ($N$ = 100). Different letters indicate that each proportion is significantly greater than the one appearing immediately below it in the column (one-tail $Z$ test of proportions, $\alpha \leq 0.05$).

[c] $N$ = number of pseudergates (Oakland colony) recruited to trails in 25 individual assays with each concentration of extract in each time interval. SEM = standard error of the mean. Means in the same column followed by different letters are significantly different (ANOVA of ranks, Tukey-Kramer test, $\alpha \leq 0.05$).

gates also increased with increasing concentration of glandular extract (Table 1). Of the 100 total termites tested with each dilution of glandular extract, 88 completed trails drawn from the stock 1× concentration, 32 completed trails at the 0.1× concentration, and none completed trails of 0.01× concentration or the dichloromethane alone. There was no statistically significant relationship (ANOVA, $\alpha \leqslant 0.05$) between the distance traveled and the length of the exposure period. This can be attributed to the relatively short length of the trail (200 mm) and the rapidity with which trail following was initiated under our assay conditions.

Results from our second experiment (Table 2) indicate that differential movement along trails of different pheromone concentration is due, at least in part, to concentration-dependent differences in rates of locomotion. Individuals from both the Oakland and Smyth colonies walked faster on trails of 1× and 10× concentrations of glandular extract than on trails of 0.1× concentration. Oakland pseudergates also walked faster on 10× than on 1× trails.

The distance traveled without pausing, reversing direction, or deviating from the 150-mm trail was also greater at higher concentrations of glandular extract (Table 2). The mean distances traveled by pseudergates from both colonies at the 1× and 10× concentrations differed significantly from the distances traveled on trails of 0.1× concentration.

Individuals that did not immediately complete the 150-mm trails evidenced three behaviors: (1) continuing forward on the trail after pausing, (2) leaving the trail, or (3) turning around and reversing their direction of travel on the trail. Combining the results from both colonies (Figure 1), 28% of the pseu-

TABLE 2. MEAN RATE OF LOCOMOTION AND MEAN DISTANCES TRAVELED WITHOUT VARIATION IN BEHAVIOR BY *Reticulitermes hesperus* PSEUDERGATES ON 150-mm ARTIFICIAL TRAILS DRAWN WITH STERNAL GLAND EXTRACT[a]

| Colony | Concentration[b] | Rate ± SEM (mm/sec)[c] | Distance ± SEM (mm)[c] |
|---|---|---|---|
| Oakland | 0.1× | 5.30 ± 0.52c | 122.97 ± 7.73b |
| | 1× | 7.33 ± 0.54b | 143.00 ± 3.93a |
| | 10× | 10.11 ± 0.74a | 146.90 ± 3.10a |
| Smyth | 0.1× | 6.22 ± 0.48b | 87.33 ± 9.69b |
| | 1× | 8.32 ± 0.35a | 131.37 ± 6.37a |
| | 10× | 8.64 ± 0.65a | 120.70 ± 7.88a |

[a] Treatment $N = 30$.
[b] 1× = 10 sternal glands per milliliter dichloromethane, applied at the rate of 1 μl per 50 mm of trail.
[c] Means in each column within each colony followed by different letters are significantly different (ANOVA of ranks, REGW multiple $F$ test, $\alpha \leqslant 0.05$). SEM = standard error of the mean.

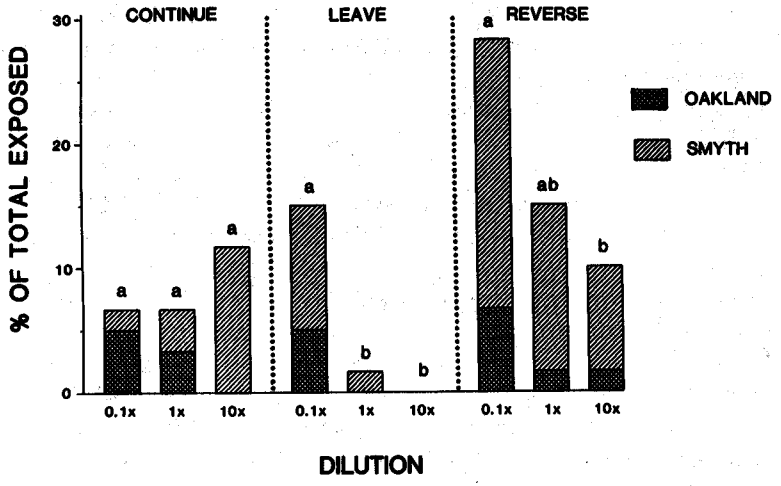


FIG. 1. Behavior of *Reticulitermes hesperus* pseudergates after pausing on 150-mm artificial trails drawn with three different dilutions of sternal gland extract. Thirty individuals from two colonies (Oakland and Smyth) were tested with each dilution ($N = 60$). Different letters over bars in each category indicate that proportions are significantly different (two-tail $Z$ test of proportions, $\alpha \leqslant 0.05$).

dergates on trails of low 0.1× concentration of glandular extract reversed their direction of travel, in contrast to 15% of those on trails of 1× concentration and 10% of those on trails of 10× concentration. Significant differences occurred between the 0.1× and 10× concentrations. Although fewer termites left the trail in each treatment than reversed direction, the relationship to pheromone concentration was similar to that observed for directional reversals. However, the proportion leaving the trail at both the 1× and 10× concentrations was significantly different from the 0.1× concentration. More individuals continued forward after pausing on trails of the 10× concentration than was the case with either of the lower two concentrations, although these differences were not statistically significant.

*Orientation on Gradient Trails. R. hesperus* pseudergates on artificial trails containing a 20-fold gradient of pheromone exhibited significant differences in behavior when compared to pseudergates on trails of constant pheromone concentration (Table 3). In the 3-min assay period, fewer individuals walking down a pheromone gradient completed the 100-mm trails than was the case with those walking up a gradient or on nongradient trails of 1×, 0.1× or 0.0625× pheromone concentration. With the more dilute sternal gland extract (0.1×), more individuals walking up a 20-fold gradient completed the trails than did those walking on nongradient pheromone trails. This significant difference did not

TABLE 3. RESPONSES OF *Reticulitermes hesperus* PSEUDERGATES ON 100-mm ARITHMETIC GRADIENT AND NONGRADIENT TRAILS[a]

| Concentration at low end of gradient[b] | Trail gradient | Percent completing trail[c] | Time to complete trail (sec ± SEM)[d] | Percent reversing direction[c] |
|---|---|---|---|---|
| 1×[e] | Down | 76a | 36 ± 6a | 52a |
| | No gradient | 96b | 27 ± 4a | 16b |
| | Up | 96b | 20 ± 2a | 8b |
| 0.1×[e] | Down | 30a | 48 ± 7a | 85a |
| | No gradient | 70b | 31 ± 5a | 40b |
| | Up | 100c | 41 ± 8a | 25b |
| 0.0625×[f] | Down | 56a | 24 ± 2a | 44a |
| | No gradient | 76b | 20 ± 1a | 16b |
| 0.03125×[f] | Down | 56a | 21 ± 2a | 44a |
| | No gradient | 60a | 18 ± 1a | 20b |

[a] Termites (Oakland colony) were tested walking up or down 20-fold (20–1) arithmetic gradients and on nongradient trails corresponding to the pheromone concentration found at the low end of the gradient trails.

[b] 1× = 10 sternal glands per milliliter dichloromethane, applied at the rate of 1 μl per 50 mm of trail.

[c] Proportions within each concentration followed by different letters are significantly different ($Z$ test of proportions, $\alpha \leqslant 0.05$).

[d] Means within each concentration followed by the same letter are not significantly different (ANOVA of ranks with Tukey-Kramer test, or $t$ test of ranks, $\alpha \leqslant 0.05$). SEM = standard error of the mean.

[e] $N = 25$ (1×), or $N = 20$ (0.1×). Termites were observed for 3 min. Those reversing direction could return to complete the trail.

[f] $N = 25$. Termites were observed until they either completed the trail, reversed direction, or left the trail.

appear in tests with the 1× extract, suggesting the presence of an upper limit for pheromone perception and response under our assay conditions. With those termites completing trails, the time taken to do so did not differ significantly with respect to the trail gradient.

More termites walking down a 20-fold pheromone gradient reversed their direction of travel before (or without) completing the trail than did those walking up a gradient or on nongradient trails (Table 3). This difference was significant at every dilution of gland extract assayed. Thus, the response to a negative gradient was not to wander off the trail as the pheromone concentration decreased, but to reverse direction and walk in the direction of increasing concentration. The proportion of pseudergates completing nongradient trails declined to 60% with the 0.03125× dilution (Table 1). However, half of these

individuals (five) did not respond to the low concentration of pheromone at all, while the 11 termites not completing the gradient trails all reversed direction after initially responding to the trails and walked back to the high concentration termini.

The similarity in the distances traveled by termites down gradient trails of different pheromone concentration before reversing direction (Table 4) indicates that this is not a static klinokinetic response to an absolute threshold level of trail pheromone. Rather, the turning response appears to be initiated by a particular proportional decrease in the original concentration of pheromone detected. However, the relatively small sample size and high degree of variability in individual responses indicate that further trials are necessary to substantiate this hypothesis.

Termites exposed to logarithmic gradients on the artificial trails behaved similarly to those on arithmetic gradients (Table 5). Very few of the pseudergates walking down the 10×–1×–0.1× gradient completed the trails to the low-concentration termini. Rather, they reversed direction and walked back up the pheromone gradient. Although the mean distances traveled down the gradient trails at which these reversals occurred differed significantly between the

TABLE 4. DISTANCES TRAVELED BY *Reticulitermes hesperus* PSEUDERGATES BEFORE REVERSING DIRECTION ON 100-mm ARITHMETIC GRADIENT AND NONGRADIENT TRAILS[a]

| Concentration at low end of gradient[b] | Mean distance ± SEM (mm)[c] | | |
|---|---|---|---|
| | No Gradient[d] | Down gradient[d] | Up gradient[e] |
| 1× | 45 ± 10a ($N$ = 4) | 72 ± 6a ($N$ = 13) | 60 ± 20a ($N$ = 2) |
| 0.1× | 34 ± 10a ($N$ = 8) | 60 ± 7a ($N$ = 17) | 55 ± 16a ($N$ = 5) |
| 0.0625× | 23 ± 4a ($N$ = 4) | 65 ± 8a ($N$ = 11) | |
| 0.03125× | 40 ± 11a ($N$ = 5) | 62 ± 9a ($N$ = 11) | |

[a] Termites (Oakland colony) were tested walking up or down 20-fold (20–1) arithmetic gradients and on nongradient trails corresponding to the pheromone concentration found at the low end of the gradient trails.

[b] 1× = 10 sternal glands per milliliter dichloromethane, applied at the rate of 1 μl per 50 mm of trail.

[c] SEM = standard error of the mean.

[d] Means in the same column followed by the same letter are not significantly different (ANOVA of ranks, Tukey-Kramer test, $\alpha \leq 0.05$).

[e] Means are not significantly different ($t$ test of ranks, $\alpha \leq 0.05$).

TABLE 5. RESPONSES OF *Reticulitermes hesperus* PSEUDERGATES ON 150-mm LOGARITHMIC GRADIENT TRAILS[a]

| Colony | Trail gradient | Percent completing trail[b] | Time to complete trail (sec ± SEM)[c] | Percent reversing direction[b] |
|---|---|---|---|---|
| Oakland | Down | 17a | 29 ± 2a | 83a |
| | Up | 90b | 24 ± 2a | 10b |
| Smyth | Down | 0a | — | 100a |
| | Up | 73b | 32 ± 3 | 27b |

[a] 30 pseudergates from each colony were tested walking either up or down 150-mm trails containing three 10-fold dilutions of sternal gland extract in successive 50 mm increments (0.1×–1×–10×). Termites were observed until they either completed the trail, reversed direction, or left the trail.
[b] Proportions from the same colony in each column followed by different letters are significantly different ($Z$ test of proportions, $\alpha \leqslant 0.05$).
[c] Means for the Oakland colony are not significantly different ($t$ test of ranks, $\alpha \leqslant 0.05$). SEM = standard error of the mean.

Oakland and Smyth colonies (Table 6), identical concentrations of pheromone (e.g., 1×) were associated with both of these mean distance values. With respect to the gradient, this 1× concentration represents a 10-fold decrease in the amount of pheromone to which the pseudergates were initially exposed. Summing the results from the Oakland and Smyth colonies, slightly more termites starting at the 10× terminus reversed direction when they encountered the 1× portion (29 workers) of the trail than the 0.1× portion (22 workers). This variation in individual response agrees with our observations on arithmetic gradients

TABLE 6. DISTANCES TRAVELED BY *Reticulitermes hesperus* PSEUDERGATES BEFORE REVERSING DIRECTION ON 150-mm LOGARITHMIC GRADIENT TRAILS[a]

| Colony | Mean distance ± SEM (mm)[b] | |
|---|---|---|
| | Down gradient | Up gradient |
| Oakland | 86 ± 7a ($N$ = 25) | 25 ± 6b ($N$ = 3) |
| Smyth | 65 ± 5b ($N$ = 30) | 48 ± 11b ($N$ = 8) |

[a] Termites were tested walking up or down 150-mm trails containing three 10-fold dilutions of sternal gland extract in successive 50-mm increments (.1×–1×–10×).
[b] Means in the same column followed by different letters are significantly different ($t$ test of ranks, $\alpha \leqslant 0.05$). SEM = standard error of the mean.

(Table 4) and suggests that the mean proportional decrease in trail pheromone needed to trigger a turning response is slightly in excess of a 10-fold change.

## DISCUSSION

Although termite trail pheromones may consist of multiple chemical components, as suggested by Kaib et al. (1982), Traniello (1982), Traniello and Busher (1985), Runcie (1983) and Prestwich et al. (1984), certain aspects of trail-following behavior appear to be modulated quantitatively. The behavior of *R. hesperus* on nongradient trails of sternal gland extract indicates that initiation of trail following, rate of movement (orthokinesis), and distance traveled without any variation in behavior are directly related to the concentration of the compound(s) eliciting trail following. Reversing direction on the trail (klinokinetic behavior), and propensity to leave the trail, on the other hand, are negatively related to pheromone concentration. The positive orthokinesis we observed is in agreement with the suggestion by Prestwich et al. (1984) that the speed of termite trail-following may be related to discrimination of pheromone. Van Vorhis Key et al. (1981) also suggested from their work with *Iridomyrmex humilis* that speed of locomotion might be a relevant factor in trail-following assays; they subsequently (Van Vorhis Key and Baker, 1982) incorporated locomotory rate into their index of anemotaxis induced in *I. humilis* by trail pheromone.

Our laboratory assays were designed to illuminate individual orientation in response to specific chemical stimuli. Initiation of trail following in response to sternal gland extracts is one component of, but not necessarily analogous to, termite recruitment to trails under field conditions. Other social and environmental stimuli are likely of great importance in affecting their behavior. However, concentration-dependent responses in initiating trail following, rate of locomotion, distance traveled without leaving the trail, and a reduced klinokinetic response would all be advantageous to *R. hesperus* in locating the nest after a disturbance or in enhancing foraging efficiency. More individuals could be recruited to well-traveled trails, presumably containing a higher concentration of pheromone, and would be able to reach the nest or forage more rapidly. Individuals encountering trails of low pheromone concentration would tend to leave these trails or reverse direction on them, increasing their probability of encountering a well-traveled trail and minimizing the time spent following old and possibly unprofitable trails.

Detection of a change in pheromone concentration by *R. hesperus* pseudergates is dependent upon successive longitudinal sampling of trail increments and temporal processing of this information. This orientation mechanism has been referred to as longitudinal klinotaxis (Ewer and Burrell, 1950; Kennedy,

1978) to distinguish it from lateral klinotaxis, or side-to-side sampling. Orientation to the lateral boundaries (edges) of the trail-space may involve either lateral klinotaxis or tropotaxis, independent of orientation along the longitudinal axis of the trail. Kennedy (1986) recently proposed the alternative label schemakinesis to emphasize the internally programmed, self-steered basis of this response to longitudinal gradients.

The ability to detect and respond to changes in trail pheromone concentration suggests the possibility that differential deposition of pheromone could indicate directionality on natural trails. Such a mechanism does not necessarily require that individuals have the ability to vary the amount of pheromone deposited. As Leuthold (1975) has suggested for grass-harvesting termites, concentration of termite activity in particular areas of a gallery network could passively polarize the system in an additive fashion. Although behavioral studies to date have not documented the existence of pheromone gradients on termite trails, chemical analyses of trail increments may prove useful in this regard. Identification of the trail pheromone(s) is crucial to such investigations.

Concentration-dependent responses to crude glandular extract do not imply that a single component is responsible for eliciting the multiple behaviors comprising the trail-following response. Traniello (1982) and Traniello and Busher (1985) suggested that both an ephemeral recruitment component and a very persistent orientation component were present in glandular extracts from the termite *Nasutitermes costalis* (Holmgren). Similarly, additional components in the pheromone blend may excite locomotion or suppress klinokinesis. Researchers must develop assays designed to measure these single behaviors rather than a general trail-following response if such semiochemicals are to be isolated and identified.

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