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# Turbulence, trees and semiochemicals: wind-tunnel orientation of the predator, *Rhizophagus grandis*, to its barkbeetle prey, *Dendroctonus micans*

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**Abstract.** Insects apparently responding to the visual stimulus offered by a tree or other object may also be responding to the variety of physical effects caused by the obstruction, including turbulence, a reduction in wind velocity and eddies. The relative importance of prey odour associated with the bark beetle *Dendroctonus micans* (Kugelann) (Scolytidae), prey-host-tree odour, silhouette, and physical barrier to the wind, in the orientation and landing behaviour of a predatory beetle, *Rhizophagus grandis* Gyllenhall (Rhizophagidae), were investigated. *R.grandis* responded very positively to the frass of its prey when presented on a 'tree' but not to the same quantity of frass presented alone. Frass on black plastic uPVC pipes was significantly more attractive than frass on real host tree logs, suggesting that host-tree volatiles do not enhance the attractiveness of prey frass.

The beetles responded to 'transparent' Mylar three-dimensional cylinders in the same way as they did to the black plastic pipes. Beetles were also tested with frass and two-dimensional stimuli offering a visual stimulus or physical barrier alone or in combination. Frass with a two-dimensional visual stimulus was no better than frass alone: few insects landed. Significantly more insects were attracted to the same two-dimensional visual stimulus with a hidden upwind barrier producing turbulence, but a similar number were attracted to the frass in front of a hidden barrier alone. As many plants are similar in size to the logs used in these experiments, comparable effects might influence the behaviour of other phytophagous insects, their predators and parasitoids.

**Key words.** *Rhizophagus grandis*, *Dendroctonus micans*, Coleoptera, kairomone, flight, predator, Scolytidae, Rhizophagidae, visual orientation, *Picea abies*, olfactory, bark beetle.

## Introduction

Bark beetles have been shown to orientate to silhouettes and the visual stimuli of tree trunks in addition to aggregation pheromones and odour cues from the host tree (Borden, 1985; Borden *et al.*, 1986). Predators and parasitoids of bark beetles are likely to use similar cues because their prey are always associated with trees. In a comparable way, male gypsy moths (*Lymantria dispar*) locating females, which call on trees, orientate at close range by the 'interactions between pheromone and optical stimuli associated with ... vertical silhouettes' (Charlton & Cardé, 1990).

However, a further cue to orientation, not previously investigated, may be the effects of the tree as a physical obstacle to the wind. Insects apparently responding to the visual stimulus offered by a tree or, in an experiment,

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to a solid model, may also be responding to physical effects such as turbulence, a reduction in wind velocity and eddies. In this paper we present evidence which suggests that for some tree-associated insects this cue may be as or more important than visual cues for short-range orientation and landing.

We have investigated the relative importance of prey odour, prey-host-tree odour, silhouette and physical barrier to the wind in the orientation and landing behaviour of a predatory beetle, *Rhizophagus grandis* Gyllenhall, in a wind-tunnel.

*Rhizophagus grandis* is a highly specific predator of the bark beetle *Dendroctonus micans* (Kugelann) (Grégoire, 1988). It enters the brood system of the prey, and adults and larvae feed on the juvenile stages of *D.micans*. The specificity of *R.grandis* has been an important factor in its successful use in biological control programmes both in the Massif Central, France (Grégoire *et al.*, 1984, 1985) and in Shropshire and Wales, U.K. (King & Evans, 1984). *R.grandis* is strongly attracted in the field and laboratory by the frass of its prey and components of it which include monoterpenes and oxygenated monoterpenes (Tondeur & Grégoire, 1980; Grégoire *et al.*, 1992; Wainhouse *et al.*, 1991).

## Materials and Methods

*Wind-tunnel.* The wind-tunnel was 170 × 60 × 60 cm with sliding glass sides, glass floor and roof (Fig. 1). A sheet of fine white nylon screen delimited the upwind end of the wind tunnel. The air was filtered through a 60 × 60 × 2 cm layer of activated charcoal (BDH, granular activated for gas absorption, 10–18 mesh). The air temperature was 19–25°C and the windspeed 0.05 m/s. The wind-tunnel was lit from above by four 150 cm long 58 W white fluorescent tubes (Thorn Pluslux 3500), giving 3610 lux (32 W/m$^2$) 20 cm above the floor. During the experiment, white nylon net curtains were drawn over the sides of the wind-tunnel to reduce distraction from objects (including the observers) outside the wind-tunnel.

*Targets and stimuli.* The standard target was a 50 cm length of 70 mm diameter shiny black uPVC drainpipe, placed vertically and closed at the top with a polystyrene lid. Preliminary experiments showed that the rate of attempted landings on 70 mm diameter black uPVC 'trees' with frass was almost twice that to a 30 mm diameter black 'tree' baited in the same way. However, increasing the diameter to 110 mm did not increase the rate of landing.

Frass was presented on Blu-tak (proprietary inert solid adhesive, Bostik, Leicester) in the opening of a pipette tip (Gilson C20) placed in an angled hole 23 cm from the base so that it projected from the pipe upwards at an angle of 45°. The frass was collected from broods of *D.micans* in Norway Spruce, *Picea abies* (in Wales, U.K.) and stored at −13°C. In the experiments described in this paper 50 mg of frass was used, as response to frass plateaued between 10 and 100 mg (Wainhouse *et al.*, 1991, Fig. 3) (control was pipette tip with Blu-tak).

The target drainpipe 'tree' was placed 30 cm from the upwind end of the wind-tunnel. The take-off platform, a roughened sheet of Perspex 10 × 12.5 cm raised 1 cm off the floor on a Petri dish lid, was 65 cm downwind from the 'tree' (Fig. 1).

*Preconditioning and pretreatment of Rhizophagus.* Adult *R.grandis* were supplied (January 1988 to June 1989) by the Université Libre de Bruxelles (reared by the methods of Grégoire *et al.*, 1986). The insects had been collected directly on emergence and stored in damp bark powder in the dark at 4°C. Before the experiments, the beetles were floated out of the bark powder, placed on damp filter paper overnight under the lights of the wind-tunnel at room temperature, the optimum preconditioning for walking response to frass (Merlin *et al.*, 1984), and then sorted by sex. Frequent matings were observed during the overnight preconditioning but the beetles had not oviposited. Beetles were pre-tested in bulk prior to the experiments by selecting only those beetles which took off from the platform within 5 min (without tree or frass in the wind-tunnel). The pre-test was a good predictor of beetles that would fly in the experiment. Unless otherwise noted the insects were used once only.

Ten minutes before the experiment twenty or thirty beetles were placed on the take-off platform under a clear

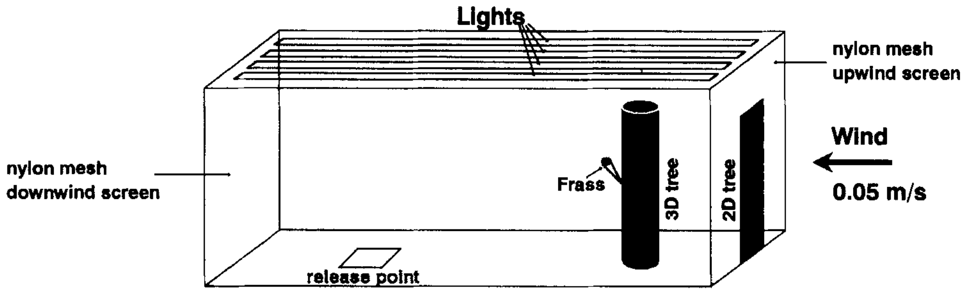

**Fig. 1.** Diagram of the wind-tunnel (not to scale) showing the position of three-dimensional or two-dimensional 'trees'. The three-dimensional 'trees' were placed 30 cm downwind from the upwind screen. The two-dimensional 'trees' were marked out with permanent black ink on a second, removable, upwind screen pinned to the first (for black silhouette), and a Mylar strip was placed immediately unwind to give an obstruction to the wind when required. The pipette tip, with or without frass, was placed on the 'tree' (three-dimensional (3D), two-dimensional (2D), or point) at the same height as on the three-dimensional 'tree' in the diagram.

lid which was gently removed at the start of the experiment. The percentage response to the stimulus in each batch was out of the number that actually took off in the experiment.

The order of experiments was randomized within each replicate of all treatments. Between each experiment the wind-tunnel floor and take-off platform were cleaned with alcohol and the 'trees' were soaked in 10% Decon 90 (Decon Laboratories, Hove, Sussex BN3 2ZZ, U.K.) cleaning agent for 12–18 h and rinsed. The low response to controls suggests these methods were effective at eliminating contamination.

*Response to prey frass and to host-tree odour.* uPVC pipes were compared to pieces of freshly cut *P.abies* logs 50 cm long, 100–150 mm in diameter. Pipes and logs were tested with or without 50 mg frass in a pipette tip. Pipes were also tested with a 2.5 $cm^2$ square of fresh *P.abies* bark in place of the frass to give host-tree odour plus visual stimulus from the pipe.

*Effect of visual and physical characteristics of targets.* The experiments tested three-dimensional or two-dimensional targets offering different combinations of visual characteristics and physical obstacles to the wind flow (Fig. 1). Each combination was tested with or without 50 mg frass as in the previous experiments. Separate screens were used for the frass and control to avoid any potential contamination. The order of experiments was randomized.

Two types of three-dimensional 'trees' were tested: 70 mm diameter black uPVC pipes presenting a strong visual silhouette together with 'mechanical turbulence caused by air flowing round the [obstacle]' (Prokopy, 1986), or transparent 70 mm diameter tubes made from Mylar plastic presenting a physical obstacle to the wind flow only. Circular zones were marked at 5 and 10 cm from the base of the tree on the floor. The three-dimensional 'trees' were placed at the standard position 30 cm downwind from the upwind end.

The two-dimensional 'trees' offered three combinations of presence or absence of silhouette, marked with black felt tip pen on a second, removable, screen pinned to the downwind side of the upwind screen (Fig. 1), and physical obstacle to the wind, provided by a 70 mm wide vertical Mylar strip hidden immediately upwind of the screen: silhouette and no Mylar; Mylar and no silhouette; silhouette and Mylar. Fine monofilament fishing line 0.06 mm in diameter was stretched from floor to ceiling of the wind-tunnel to mark the edges of the 'tree' and zones 5 cm and 10 cm to each side. The monofilament was almost invisible to human observers. Hemicircular zones were marked at 5 and 10 cm from the base of the 'tree' on floor.

In addition, for comparison with all the two-dimensional and three-dimensional experiments, the series included replicates of (a) a point source of frass odour affixed on the screen without a silhouette or physical obstacle and (b) a control with no frass or target (Fig. 1).

The spectral reflectivities of the materials were measured using a Pye Unicam SP8-100 spectrophotometer fitted with an integrating sphere, against a barium sulphate standard (Green, 1988). The black uPVC pipe had a nearly constant 3% reflectivity across the spectrum from 300 to 700 nm. A sample of Norway Spruce bark showed a similar reflectivity in the uv and higher frequencies but reflected more in the 500–700 nm spectrum, having 25% reflectivity at 700 nm. The white netting of the upwind screen placed over a black Perspex standard increased the reflectivity from 1.5% to 23% above 360 nm. The 'transparent' Mylar had less than 10% absorbance above 400 nm, but the absorbance increased rapidly below 400 nm (in the uv) (CH Green, pers. comm.). In the absence of a strong ultraviolet component to the illumination, it seems unlikely that the visibility of the Mylar would have been appreciably greater in ultraviolet wavelengths, even if the beetles were more sensitive to such wavelengths.

In this experiment females were used for four replications of each treatment because in this particular batch almost all the beetles that flew in the preflight test were females. In other experiments no differences in response were observed. One additional replication of males to the frass treatments was carried out.

The number of beetles landing on or within 10 cm of the target was recorded as were the 'glancing' hits on the target made by beetles which did not land.

*Description of take-off and flight. Rhizophagus grandis* were flown individually from the platform in the wind-tunnel and their behaviour recorded in detail until they landed. The target was either the 'tree' baited with 50 mg *D.micans* frass or the 'tree' plus pipette tip alone as a control. The *R.grandis* were placed on the take-off platform under a clear plastic lid for 1–3 min before the experiment. Beetles not taking-off within 3 min were not included in the analysis. Only those taking-off in a preflight test without frass were used for the experiment.

*Statistics.* Data expressed as proportions were arcsine transformed before statistical analysis to reduce the dependence of the variance on the mean (Sokal & Rohlf, 1981). Means and SE presented in text and figures were untransformed after analysis.

## Results

### *Response to D.micans frass and to host-tree odour*

Beetles were strongly attracted to land on a log or pipe with frass (Fig. 2). In contrast, the responses to the pipe plus bark square, and fresh spruce log were low and were not significantly different from the responses to the control pipe. Significantly more *R.grandis* were attracted to land on the pipe with frass than on the log with frass [one-way Anova, and Neuman-Keuls multiple range test ($a = 0.05$)]. Response was not influenced by sex of the beetles (two-way Anova; targets: $F = 16.18$, d.f. = 4, $P < 0.0001$; sex: $F = 0.96$, d.f. = 1, $P = 0.351$; interaction: $F = 2.12$, d.f. = 4, $P = 0.152$).

### *Odour plumes on and from trees*

The presence of a 'tree', whether a pipe or real log,

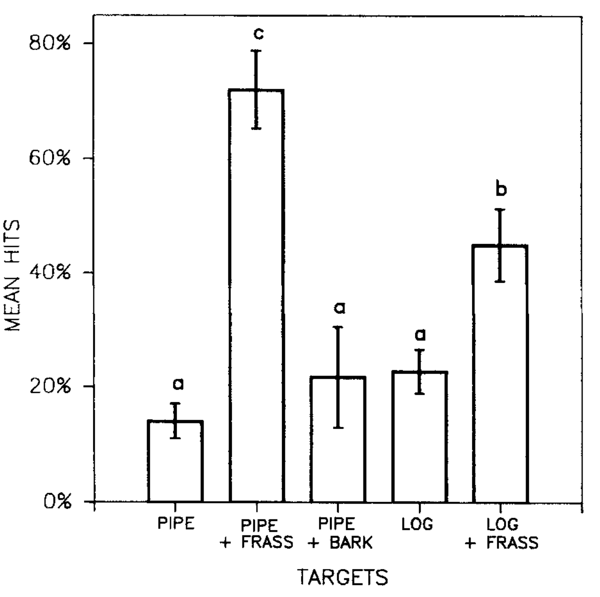

**Fig. 2.** Prey and prey-host-tree odour. Landing response of adult *Rhizophagus grandis* to a log ('Log') of the prey-host-tree, *Picea abies*, or black uPVC pipe ('Pipe'), with or without 50 mg frass of the bark beetle prey, *Dendroctonus micans*. 'Pipe + Bark': pipe with $2.5\,\mathrm{cm}^2$ square of fresh *P.abies* bark in place of the frass to give host-tree odour plus visual stimulus from the pipe. Means ±1 SE of four replicates for each test; two replicates with males, two with females; twenty beetles per replicate. Different letters mark significant differences in landing response (Newman-Keuls multiple range test, $\alpha = 0.05$).

made a big difference to the pattern of plumes in the wind-tunnel. With air currents made visible with ammonium hydroxide and concentrated hydrochloric acid on cotton wicks placed at the position of the frass on the tree or pipe, the plume often flowed either down or up the 'tree' before breaking away. This appears to be the product of a strong boundary effect.

Once landed on the 'tree' the beetles would then walk towards the frass, showing typical 'wig-wagging' movements (Kennedy, 1986). In one experiment when the 'smoke'-producing cotton wick was replaced with frass in the same spot on the log after showing the human observers the path of the plume, it appeared that the beetles were walking up the plume; having landed near the base of the log, they followed the same meandering path made visible shortly before.

After breaking away from the boundary layer on the 'trees', the plume tended to flow turbulently downwind along a vertical plane.

### *Effect of visual and physical characteristics of targets*

The landing response of adult *R.grandis* was the same to both the black pipe 'tree' and the transparent (to human eyes) Mylar pipe 'tree' of the same size (Fig. 3, two-way Anova on arcsine transformed data; $F = 0.101$, d.f. $= 1$; $P = 0.76$); for both targets, the responses to frass were significantly different from the responses to control 'trees', black or transparent ($F = 43.8$, d.f. $= 1$, $P < 0.0001$). The

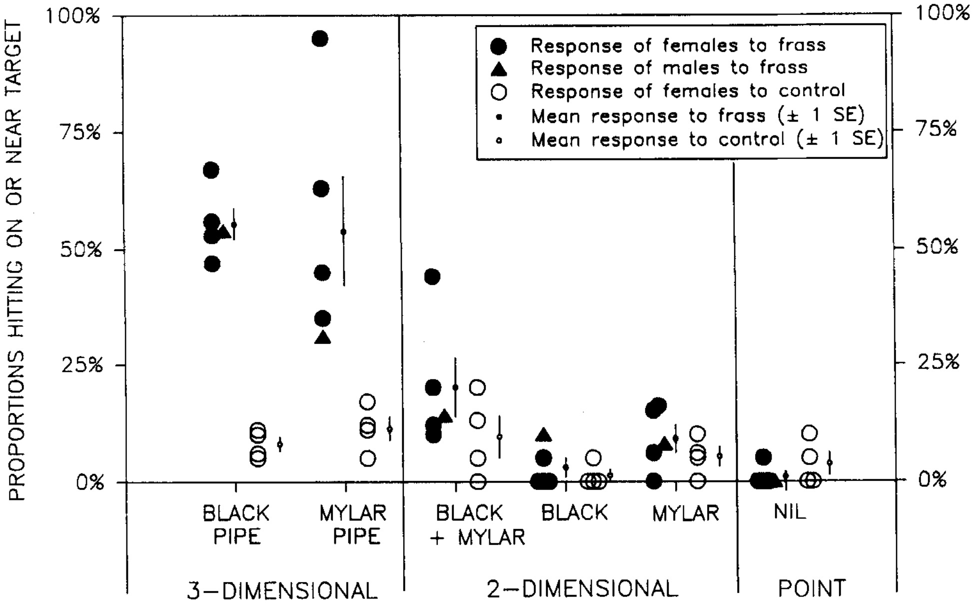

**Fig. 3.** Separation of visual and physical effects of targets. Landing response of *Rhizophagus grandis* to three-dimensional (3D) or two-dimensional (2D) targets offering different combinations of visual characteristics and physical obstacles to the wind flow. Each was tested with or without 50 mg prey frass. Individual percentages per batch of twenty insects, and mean percentages per treatment, including males (means ±1 SE). Three-dimensional: 70 mm diameter 3D 'trees': 'Black Pipe', black uPVC, giving visual silhouette and obstacle to wind; 'Mylar Pipe', 'transparent' obstacle to wind. Two-dimensional: 'Black + Mylar', black silhouette and obstacle to wind; 'Black', silhouette but no obstacle to wind; 'Mylar', obstacle to wind but no silhouette. Point: Nil, no visual target or upwind obstruction (pipette tip only).

interaction between target type and frass odour was not significant ($F = 0.075$, d.f. = 1, $P = 0.79$) nor was sex important ($F = 1.556$, d.f. = 1, $P = 0.23$). The number of glancing blows by insects in flight, which may have been attempting to land, also did not differ between the black and clear trees (two-way Anova: glances: females only: frass *v* control $F = 13.1$, d.f. = 1, $P = 0.0035$; black tree *v* clear tree: $F = 1.6$, d.f. = 1, $P = 0.23$; interaction, $F = 0.001$, d.f. = 1, $P = 0.97$). Almost 4 times as many beetles made glancing blows on trees of both types bearing frass, than on control trees (mean ±SE: glances black, frass 13.8 ± 5.0; Mylar, frass 18.0 ± 3.9; black, control 2.0 ± 0.6; Mylar, control 6.0 ± 1.6).

It is noticeable that the variance in the results for the three-dimensional Mylar with frass is much greater than for the black three-dimensional with frass; for almost the same mean, the SE is 4 times greater (Fig. 3). A comparison of variances including all the results in Fig. 3 (Bartlett test, SPSS) gives $F = 4.85$ ($P < 0.001$); excluding three-dimensional-Mylar, $F = 2.75$, ($P = 0.029$) (NS at $\alpha = 0.01$). This might be due to the incomplete transparency of the Mylar to the beetles.

There was little or no landing response to the point source of frass alone placed in front of the screen (Fig. 3). Frass with a two-dimensional visual stimulus alone was not better; few insects landed. Significantly more insects were attracted to the same two-dimensional visual stimulus combined with a hidden upwind barrier producing turbulence than to the visual stimulus along (*t*-test; $t = 3.8$, d.f. = 7.6, $P < 0.01$). The number of beetles landing to frass in front of a hidden barrier alone was significantly higher than to the point source of frass ($F = 6.3$, d.f. = 1, 8, $P = 0.036$) but not significantly different from the response to the combined visual stimulus and hidden upwind barrier (*t*-test; $t = 1.99$, d.f. = 7.6; $P > 0.05$) (analysis including males (see Fig. 3), variances not assumed equal)).

### *Description of flight*

After a characteristic spreading of their wings, elytra first then hindwings, the beetles took-off vertically. Initially they flew up, bobbing against the glass roof of the wind-tunnel before descending in the air column to about mid-height. Observations of individual beetles were made to gain a detailed description of the flight behaviour and to compare the behaviour of individuals in the presence and absence of frass. In bobbing flight the beetle flew upwards, hit the glass ceiling, fell, and then flew up again. Bobbing flight appeared very erratic and the fall could be as much as 20 cm. Some beetles flew from one end of the wind-tunnel to the other whilst bobbing; individual bouts of bobbing behaviour could last for as long as a minute. The greatest number of bobs was 134 during a flight of 109 s. In the absence of frass in the air stream, the beetles flew throughout the wind-tunnel, up to and around the target 'tree' but did not attempt to land. When frass was presented on the 'tree' the beetles characteristically hovered downwind of the 'tree' before attempting to land on it. Hovering flight was not punctuated by bobs and was usually at mid height in the wind-tunnel. This type of flight was slower and could last for several minutes.

### *Influence of frass on flight patterns*

There was no difference in the proportion taking-off with (22/32) and without (24/32) frass in the wind-tunnel (pooled across the sexes). Similarly, the time before take-off was not influenced by the presence or absence of *D.micans* frass (take-off time in seconds: mean ±SE frass 80 ± 12.6, control 65 ± 9.7; $F = 0.89$, d.f. = 1, $P = 0.37$). There was no difference between the males and females ($F = 0.92$, d.f. = 1, $P = 0.36$) and no significant interaction between sex and stimulus ($P = 0.5$). There was no difference in the duration of flight comparing frass and control ($F = 0.09$, d.f. = 1, $P = 0.77$); nor by sex ($F = 1.719$, d.f. = 1, $P = 0.21$) and the interaction between sex and stimulus was also not significant ($F = 0.002$, d.f. = 1, $P = 0.96$). The mean flight to frass was 50 ± 13.9 s ($n = 13$) and to the control 46 ± 12.2 ($n = 8$). There was a marked effect of frass on the type of flight, with much more hovering flight when frass was present. This was shown quantitatively by the proportion of hovering flight out of the total flight time: median, with frass 100% (lower and upper quartile; 64–100%, $n = 13$) with control 26.5% (8–54%, $n = 8$); Mann-Whitney U test, $Z = -2.87$, $P = 0.004$. There was no difference between frass and control in relation to whether the beetles ended their flight upwind or downwind of the take-off platform ($\text{Chi-square}_{(\text{corr})} = 2.9$, d.f. = 1, $P > 0.05$) (frass, up:down: 19:3; control 11:8). However, this records the point of landing not the total movement up and down the length of the wind-tunnel during the flight.

The results of observations of single beetles were supported by analysis of other experiments involving flights of twenty or thirty beetles from the platform. The take-off of these beetles seemed independent of the presence of frass in the wind-tunnel (mean number ±1 SE beetles taken off in first 5 min (out of twenty beetles): with frass 18.4 ± 0.46 ($n$ = eight batches of twenty beetles); control, without frass 18.8 ± 0.37 ($n = 8$); $F = 0.406$, d.f. = 1, $P = 0.54$).

The flight of *R.grandis* is weak. In preliminary experiments with windspeeds of about 0.5 m/s the beetles made little headway but at windspeeds in the neighbourhood of 0.05 m/s the beetles flew well.

Males and females were tested separately in the experiments but there were no differences in their response to the frass (for example, mean proportions ±1 SE: males 71.6% ± 0.3 ($n$ = four batches of twenty beetles), females 68.7% ± 0.1 ($n$ = four batches of twenty beetles) one-way Anova: $F = 0.273$, d.f. = 1, $P = 0.63$).

## Discussion

For *Rhizophagus grandis*, the operating rule might be 'no tree–no prey' so far as landing is concerned. In the wind-tunnel *R.grandis* responds very positively to the frass of its prey when presented on a 'tree' but not to the same quantity of frass presented alone. Frass on black plastic uPVC pipes was significantly more attractive than frass on real host tree logs which suggests that host-tree volatiles do not enhance the attractiveness of prey frass. Prey frass does in any case contain host-tree monoterpenes (Grégoire *et al.*, 1991; Wainhouse *et al.*, 1991).

If a uPVC pipe can substitute for a log, what features of the 'tree' are important in the landing response to frass? Past work would suggest a response to the visual silhouette (e.g. Borden *et al.*, 1986), a feature offered equally well by logs and black pipes. An alternative explanation is that the beetles are responding to aspects of the disturbance of the air flow caused by the physical barrier formed by the 'tree'.

Our results suggest that physical effects were as or more important than the visual stimulus presented by the tree. The physical effects might include, for example, the sudden loss of wind velocity, changes in turbulence, and eddies immediately downwind of the obstacles. However, visual stimuli from the 'trees' cannot be totally ruled out, as Mylar used to make the transparent 'trees' showed about 50% reflectivity between 300 and 400 nm in the uv range. However, for the two-dimentional 'trees', the strip of Mylar was hidden behind the upwind screen of the wind-tunnel, which made it even less conspicuous. Arguments for caution are well summarized in Prokopy (1986).

The almost complete lack of response by *Rhizophagus grandis* to a point source of prey frass in the absence of a physical 'tree' is quite different from the behaviour shown by many other insects in a wind-tunnel or in the field. For example, the woodworm beetle, *Anobium punctatum*, and the drug store beetle, *Stegobium paniceum*, both orientated to point sources of female pheromone (Birch & White, 1988). However, in the case of the gypsy moth, *Lymantria dispar*, males orientate to 'tree' silhouettes with pheromone (Charlton & Cardé, 1990) but will not land if no pheromone is present.

Wind-tunnel experiments by Mizell *et al.* (1984) on the bark beetle predator, *Thanasimus dubius*, presented the beetles with a point source of prey or host-tree volatiles on the upwind screen. As they observed no clear pattern in the position of the beetles at the end of the test, they used initial upwind flights as their criterion. The present study suggests that if the experiments were repeated with the compounds presented on a 'tree' as reported here, a clear landing response might be found to the most attractive compounds.

Experiments by Salom & McLean (1990) on the behaviour of *Trypodendron lineatum* (Coleoptera: Scolytidae) used drain-pipe traps similar to our uPVC 'trees' as the upwind targets. Their beetles hovered and then landed on the 'trees' in a similar way to the *R.grandis* in this study. However, their study did not investigate features of the targets promoting landing. *T.lineatum* would be a good species with which to test the ideas proposed here. It should be noted that neither species is found on standing timber only: *R.grandis* also colonizes *D.micans* broods on stumps and *T.lineatum* attacks stumps and fallen timber rather more than standing timber. This may correspond to the suggestion that, for *R.grandis*, turbulence has at least as much importance as visual cues.

Unless wind speed in our experiments was as low as 0.05 m/s, the beetles were not able to fly upwind. In field release–recapture experiments, *R.grandis* did not fly when there was any detectable wind at all (D. Couillien, pers. comm.). In a wind-tunnel study of *T.lineatum* greater upwind flight occurred at low wind speeds in the range 0.0–0.9 m/s, with the greatest capture of beetles in still air (Salom & McLean, 1991). Similarly, Lanier *et al.* (1976) reported that *Scolytus multistriatus* would not emerge from elm trees and fly off in windy conditions. This last species, however, successfully flew in a wind-tunnel at a windspeed of 0.25 m/s (Choudhury & Kennedy, 1980).

The prey searching behaviour of predatory beetles that specialize on bark beetles is still largely unknown, in part because long-range orientation occurs in flight and little research has been carried out on the flight of beetles of any kind. The bulk of information available so far is provided by field studies focused on arrival at traps in response to specific attractants, and on temporal sequences of arrival for natural enemies of bark beetles (and associates of unknown status) as a response to prey (host) odours. However, other circumstances associated with prey-finding are often unknown, such as sex, age, physiological state of the predators, daily flight periods, wind, visual/physical stimuli, L/D cycles. In this context, wind-tunnel observations under standardized conditions are extremely useful. A good bioassay has been identified here, for an insect which, due to the possibility of mass-rearing, could easily be used in a model system for a better understanding of a species-specific predator–prey relationship.

The response to the physical effects of targets may be more general, applying to other insects, such as other bark beetles and their predators. In a wider context, other plants, for example maize, are on a very similar scale to the logs used in these experiments and similar physical effects might be important for other phytophagous insects and their predators and parasitoids. Three topics in particular need further investigation: first, the flow of air currents around solid obstacles at the low wind speeds at which beetles and other insects are most likely to be flying and their effects on odour plumes, second, the behaviour of near-target insects as they approach to land, and third, a more critical investigation of the effects, and visibility, of 'transparent' targets.

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## References

Birch, M.C. & White, P.R. (1988) Responses of flying male *Anobium punctatum* (Coleoptera, Anobiidae) to female sex pheromone in a wind tunnel. *Journal of Insect Behavior*, **1**, 111–115.

Borden, J.H. (1985) Aggregation pheromones. *Comprehensive Insect Physiology, Biochemistry and Pharmacology* (ed. by G. A. Kerkut and L. I. Gilbert) Vol. 9, pp. 257–285. Pergamon Press, Oxford.

Borden, J.H., Hunt, D.W.A., Miller, D.R. & Slessor, K.N. (1986) Orientation in forest Coleoptera: an uncertain outcome of responses by individual beetles to variable stimuli. *Mechanisms in Insect Olfaction* (ed. by T. L. Payne, M. C. Birch and C. E. J. Kennedy), pp. 97–109. Clarendon Press, Oxford.

Charlton, R.E. & Cardé, R.T. (1990) Orientation of male gypsy moths, *Lymantria dispar* (L.), to pheromone sources: the role of olfactory and visual cues. *Journal of Insect Behavior*, **3**, 443–469.

Choudhury, J.H. & Kennedy, J.S. (1980) Light versus pheromone-bearing wind in the control of flight direction by bark beetles, *Scolytus multistriatus*. *Physiological Entomology*, **5**, 207–214.

Green, C.H. (1988) The effect of colour on trap- and screen-oriented responses in *Glossina palpalis palpalis* (Robineau-Desvoidy) (Diptera: Glossinidae). *Bulletin of Entomological Research*, **78**, 591–604.

Grégoire, J.-C. (1988) The Greater European Spruce Beetle. *Dynamics of Forest Insect Populations* (ed. by A. A. Berryman), pp. 455–478. Plenum Press, New York.

Grégoire, J.-C., Merlin, J., Pasteels, J.M., Jaffuel, R., Vouland, G. & Schvester, D. (1984) Mass-rearings and releases of *Rhizophagus grandis. Proceedings of the EEC Seminar on the Biological Control of Bark Beetles (Dendroctonus micans), Brussels, 3–4/10/1984* (ed. by J.-C. Grégoire and J. M. Pasteels), pp. 122–128. Brussels.

Grégoire, J.-C., Merlin, J., Pasteels, J.M., Jaffuel, R., Vouland, G. & Schvester, D. (1985) Biocontrol of *Dendroctonus micans by Rhizophagus grandis* in the Massif Central (France). *Zeitschrift für Angewandte Entomologie*, **99**, 182–190.

Grégoire, J.-C., Merlin, J., Jaffuel, R., Denis, Ph., Lafont, P. & Schvester, D. (1986) Elevage à petite et moyenne échelle du prédateur *Rhizophagus grandis* Gyll. en vue de la lutte biologique contre *Dendroctonus micans* Kug. *Revue Forestière Française*, **38**, 457–464.

Grégoire, J.-C., Baisier, M., Drumont, A., Dahlsten, D.L., Meyer, H. & Francke, W. (1991) Volatile compounds in the larval frass of *Dendroctonus valens* and *Dendroctonus micans* (Coleoptera: Scolytidae) *in* relation to oviposition by the predator, *Rhizophagus grandis* (Coleoptera: Rhizophagidae). *Journal of Chemical Ecology*, **17**, 2003–2019.

Grégoire, J.-C., Couillien, D., Krebber, R., König, W.A., Meyer, H. & Francke, W. (1992) Orientation of *Rhizophagus grandis* (Coleoptera: Rhizophagidae) to oxygenated monoterpenes in a species specific predator–prey relationship. *Chemoecology*, **3**, 14–18.

Kennedy, J.S. (1986) Some current issues in orientation to odour sources. *Mechanisms in Insect Olfaction* (ed. by T. L. Payne, M. C. Birch and C. E. J. Kennedy), pp. 11–25. Clarendon Press, Oxford.

King, C.J. & Evans, H.F. (1984) The rearing and release of *R.grandis* against *D.micans* in the United Kingdom. *Proceedings of the EEC Seminar on the Biological Control of Bark Beetles (Dendroctonus micans), Brussels, 3–4/10/1984* (ed. by J.-C. Grégoire and J. M. Pasteels), pp. 87–97. Brussels.

Lanier, G.N., Silverstein, R.M. & Peacock, J.W. (1976) Attractant pheromone of the european elm bark beetle (*Scolytus multistriatus*): isolation, identification, synthesis, and utilisation studies. *Perspectives in Forest Entomology* (ed. by J. F. Anderson and H. K. Kaya), pp. 149–175. Academic Press, New York.

Merlin, J., Grégoire, J.-C., Baisier, M. & Pasteels, J.M. (1984) Some new data on the biology of *Rhizophagus grandis. Proceedings of the EEC Seminar on the Biological Control of Bark Beetles (Dendroctonus micans), Brussels, 3–4/10/1984* (ed. by J.-C. Grégoire and J. M. Pasteels), pp. 107–121. Brussels.

Mizell, R.F., III, Frazier, J.L. & Nebeker, T.E. (1984) Response of the clerid predator *Thanasimus dubius* (F.) to bark beetle pheromones and tree volatiles in a wind tunnel. *Journal of Chemical Ecology*, **10**, 177–187.

Prokopy, R.J. (1986) Visual and olfactory stimulus interaction in resource finding by insects. *Mechanisms in Insect Olfaction* (ed. by T. L. Payne, M. C. Birch and C. E. J. Kennedy), pp. 81–89. Clarendon Press, Oxford.

Salom, S.M. & McLean, J.A. (1990) Flight and landing behavior of *Trypodendron lineatum* (Coleoptera: Scolytidae) in response to different semiochemicals. *Journal of Chemical Ecology*, **16**, 2589–2604.

Salom, S.M. & McLean, J.A. (1991) Flight behavior of scolytid beetle in response to semiochemicals at different wind speeds. *Journal of Chemical Ecology*, **17**, 647–661

Sokal, R.R. & Rohlf, F.J. (1981) *Biometry*, 2nd edn. W. H. Freeman, New York.

Tondeur, A. & Grégoire, J.-C. (1980) Chemical orientation of *Rhizophagus grandis* towards mates and towards prey: *Dendroctonus micans. Animals and Environmental Fitness* (ed. by R. Gilles), pp. 93–94. Pergamon Press, Oxford.

Wainhouse, D., Wyatt, T.D., Phillips, A., Kelly, D.R., Barghian, M., Beech-Garwood, P., Cross, D. & Howell, R.S. (1991) Response of the predator *Rhizophagus grandis* to host plant derived chemicals in *Dendroctonus micans* larval frass in wind tunnel experiments (Coleoptera: Rhizophagidae, Scolytidae). *Chemoecology*, **2**, 55–63.

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