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Pheromone-mediated optomotor anemotaxis and altitude control exhibited by male oriental fruit moths in the field

T. C. BAKER and K. F. HAYNES* Department of Entomology, Iowa State University, Ames, Iowa, and *Department of Entomology, University of Kentucky, Lexington, Kentucky, U.S.A.

Abstract. In the field over short grass, pheromone-stimulated oriental fruit moth males, *Grapholita molesta* (Busck), flying under high windspeeds tended to steer courses more into the wind and to increase their airspeeds compared with those flying in low windspeeds. Thus, optomotor anemotaxis enabled the males to steer relatively consistent upwind track angles and to maintain an upwind progress of between *c.* 50–100 cm/s despite variable wind velocities. Zigzagging flight tracks were observed at both 10 m and 3 m from the source, as were tracks with no apparent zigzags. Transitions from casting to upwind flight or vice-versa were observed. The durations of the intervals between reversals during both upwind zigzagging flight and casting were consistent with those observed in previous wind-tunnel experiments. The control of altitude was more precise during upwind zigzagging flight than during casting. In general, the side-to-side deviations in the tracks were greater than the up-and-down deviations, with both the side-to-side and vertical distances and their ratios being consistent with previous wind-tunnel studies of pheromone-mediated flight. One difference between the field and laboratory flight tracks was that males in the field exhibited much higher airspeeds than in the wind tunnel. Males occasionally were observed to progress downwind faster than the wind itself, and further analysis showed that they were steering a downwind course in pheromone-free air following exposure to pheromone, which is the first time this has been recorded in moths. We propose that such downwind flight may aid in the relocation of a pheromone plume that has been lost due to a wind-shift, by enabling the moth to catch up to the pheromone as it recedes straight downwind away from the source.

Keywords. *Grapholita molesta,* oriental fruit moth, pheromone, flight orientation, pheromone, counterturning, anemotaxis.

Introduction

Pheromones are powerful elicitors of controlled flight in insects. Flight control involves the actions and reactions with which freely flying insects steer and change their course, airspeed and lift in order to control their horizontal and vertical displacement in air, the medium that both supports and transports them. In locating pheromone sources such as conspecific females emitting the correct blend of components, a male moth must visually monitor its own progress and react to this feedback in order to displace over the ground against the wind (Kennedy & Marsh, 1974; Marsh *et al.*, 1978). At the same time it must react visually to regulate its altitude in order to maintain contact with the pheromone plume (David, 1982, 1985). An endogenous program called self-steered counterturning is a separate mechanism that is integrated with the anemotactic responses, and results in wide or narrow lateral movements across the wind-line depending upon how recently pheromone has been contacted (Baker & Kuenen, 1982; Kuenen & Baker, 1983; Kennedy *et al.*, 1980, 1981; Kennedy, 1983; Baker & Haynes, 1987; Vickers & Baker, 1994; Mafra-Neto & Cardé, 1994).

Studies of odour-mediated flight control have in the past focused on movements in the horizontal plane, since steering reactions in this plane will be what allows the moth to compensate for changes in wind direction and speed (Kennedy, 1940; Kennedy & Marsh, 1974; Marsh *et al.*, 1978, 1981; David & Birch, 1989; David & Kennedy, 1987; Preiss & Kramer, 1986). Analyses of flight tracks derived from laboratory flight-tunnel experiments have been valuable in ascertaining the shapes of the flight tracks and changes in the overall rate of progress toward the source

Correspondence: Dr Thomas C. Baker, Department of Entomology, Iowa State University, Ames, IA 50011, U.S.A.

(Cardé & Hagaman, 1979; Baker & Kuenen, 1982; Kuenen & Baker, 1982, 1983; Willis & Baker, 1984; Baker *et al.*, 1985; Baker & Haynes, 1987; Willis *et al.*, 1991; Willis & Arbas, 1991; Willis & Cardé, 1990). Use of the triangle of velocities analysis technique (Kennedy, 1940) with flight tracks has, importantly, enabled the actual behavioural reactions involved in anemotaxis to be determined, i.e. changes in the courses steered by males and their airspeeds at any instant, not just their resultant flight track angles and groundspeeds (Marsh *et al.*, 1978, 1981; Willis & Baker, 1987; Baker & Haynes, 1987; Witzgall & Priesner, 1984).

Very few analyses of flight tracks of pheromone-stimulated moths have been performed under field conditions (Willis *et al.*, 1991; Murlis & Bettany, 1977; Murlis *et al.*, 1982; David *et al.*, 1983; David & Birch, 1989) and fewer still at a level that would enable conclusions to be drawn concerning the behavioural reactions of males to different windspeeds and directions. In the field, David *et al.* (1983) recorded the overall upwind or crosswind progress of gypsy moth males, *Lymantria dispar*, under shifting wind conditions and pheromone loss and gain. Important information about the adaptive significance of displacement and successful source location was gained from these experiments. However, because the focus of the study was on the direction of the resultant movement, not the behavioural reactions to wind and pheromone that created it, no information on flight control was gained. Likewise, Murlis & Bettany (1977) and Murlis *et al.* (1982) recorded *Spodoptera littoralis* flight in response to pheromone in the field using two cameras and an anemometer, but did not analyse the flight in three dimensions. At higher windspeeds the males kept the same net upwind groundspeed (rate of progress) toward the source, meaning that they reacted to and compensated for the wind anemotactically, the first time this was shown in the field. However, the behavioural reactions, i.e. the degree to which the moths increased their airspeed or headed more into the wind, or both, was not determined. Willis *et al.* (1991) determined the changes in gypsy moth flight behaviour under semi-natural field conditions as they approached a pheromone source and experienced different concentrations of pheromone in the plume's filaments. The moths' behavioural responses to changes in windspeed and direction were not reported.

Most studies in a wind tunnel have either neglected the vertical deviations in the flight tracks that are related to altitude control, or else have examined them separately as descriptors of different pheromone blend qualities (Palaniswamy *et al.*, 1983), and thus they have not provided insight into flight control. An exception is the recent work performed in a wind tunnel by Vickers (1992) and Vickers & Baker (1995) that describes the three-dimensional (3-D) shapes of the surging and casting responses of *Heliothis virescens* males in response to single filaments of their sex pheromone. One earlier study performed in our laboratory began to examine the changes in flight tracks of *Grapholita molesta* in three dimensions both during and after pheromone contact, but the behavioural reactions (flight forces) remained undescribed (Von Keyserlingk, 1984; Baker, 1989a). Witzgall & Arn (1990) likewise performed 3-D analyses of flight tracks of male moths but did not describe the behaviour (airspeeds and course angles steered) that created these tracks.

Other analyses or descriptions of flight tracks in the field, although of importance in other ways, are of limited value for understanding flight control because of inaccuracies in measuring wind direction and speed due to using the prevailing average (Perry & Wall, 1984), or the often erroneous notion that the wind direction where the male is flying is the same as where an observer standing several metres away is assessing the direction according to pressure on his or her body. Windspeed and direction can vary significantly even over a few metres (David *et al.*, 1982, 1983). Other field observations, again of value on other levels, remain limited because tracks were not actually recorded, and neither was the windspeed and direction (Witzgall & Priesner, 1984; Baker & Cardé, 1979; Baker *et al.*, 1976; Rothschild & Minks, 1974; Baker & Roelofs, 1981). The reactions of males to wind and odour remain unclear.

Therefore we analysed the tracks made by oriental fruit moth males, *G. molesta* (Busck), in the field under varying wind conditions and at two different distances from the odour source, to determine the behavioural reactions made by the males under these varying conditions. The oriental fruit moth presents a special challenge for such a study due to its small size and rapid flight, which make individuals very difficult to image in a large enough field of view such that an adequate sampling of behaviour can be observed in one individual. To record and analyse these small moths in three dimensions, we developed a 3-D recording and analysis system that pinpoints the position in space of both the flying moth and of soap bubbles (David *et al.*, 1982, 1983). In our case the bubbles served as anemometers for the air mass through which the male is moving. Accurate analysis of the reactions of male moths to wind, and hence the study of flight control in the field, is only possible if the wind is measured as closely as possible in space and time to the flying moth. The key components to the field analysis reported here are: the use of the triangle of velocities (Fig. 1) analysis of movements (Kennedy, 1940); high-resolution 3-D video imaging using rotary shutter cameras that pinpoint the positions of the moths and the bubbles; and a computer-based analysis system that integrates the windspeed and direction (using bubbles) near the moth's position in space with the moth's groundspeed and direction in order to calculate the moth's airspeed and course reactions. We also analysed the tempo of counterturning, a second element integrated with anemotaxis in pheromone-mediated flight (Kuenen & Baker, 1983; Kennedy, 1983, 1986; Baker, 1986, 1990).

Materials and Methods

Moths were reared on small green thinning apples; the pupae were separated according to sex, and the adult males were separated daily, according to age. All life stages were maintained at *c.* 25°C on a L:D 16:8 h photocycle. Adult males were held in an environmental chamber with positive air pressure to eliminate the possibility of exposure to female pheromone prior to the experiments and had an 8% sucrose solution available to them at all times. All males were 3–5 days old at the time of the experiments.

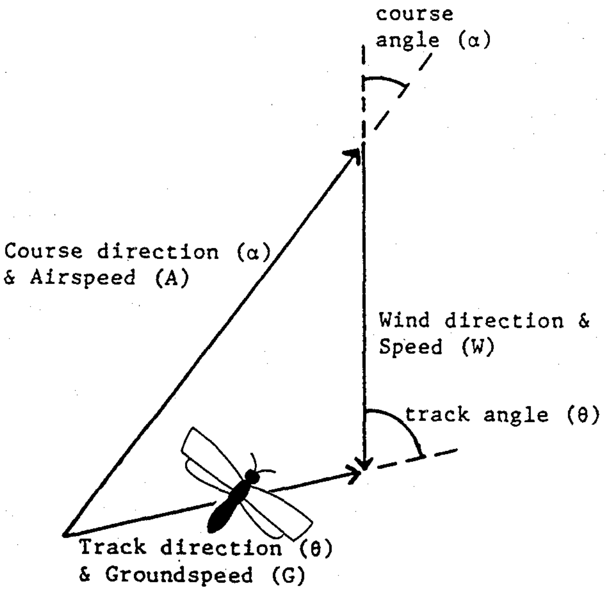

Fig. 1. The triangle of velocities, in which a moth's groundspeed and its track angle (flight path) with respect to wind of a known direction and speed can be used to calculate the behaviours (the airspeed and course angle, i.e. direction of thrust) that must have been used by the moth to result in that track angle and groundspeed.

A rubber septum dispenser containing 36 μg of the three-component pheromone blend, 5.9% (*E*)-8-dodecenyl acetate plus 3% (Z)-8-dodecenyl alcohol (Cardé *et al.*, 1979) in (Z)-8-dodecenyl acetate (Roelofs *et al.*, 1969), was used to evoke upwind flight in male *G.molesta* released downwind in the field. The septum was placed, with the wide, pheromone-loaded end facing upward, on a wire located 30 cm below a motor-driven bubble generator (David *et al.*, 1983), which itself was 1.5 m above the ground. The bubbles emerged from wire loops rotating through a detergent solution and were blown straight up for a few centimetres by a small motor-driven fan so that no extra velocity or directional bias was imparted to them before they were carried along above the pheromone plume by the wind. Thus, the bubbles served as tracers for the location of the pheromone plume whose origin was directly below the bubbles' origin. The bubbles also served as anemometers and wind-direction indicators during flight track analysis.

The experiments took place either from 08.00–10.00 or 17.00–19.00 hours due to the favourable light conditions for video recording the moths against the blue sky. At either time, the light cycle of the chamber in which males were held was such that they were in their optimal time of activity at the time of testing, which was 2–0 h before scotophase. Groups of about twenty males were released 5 m downwind of the recording cameras, which themselves were placed 3, 10 or 30 m downwind of the pheromone source. The experimental area was a flat, open park area of mowed grass with a few, small ornamental trees on the campus of the University of California at Riverside. An observer holding a netting cage containing the males would wait downwind of the cameras and when bubbles (indicating the presence of pheromone) were observed floating over the cameras and over the cage of moths, the top of the cage was removed and the males were allowed to fly upwind over the cameras toward the source. The tracks of the bubbles moving downwind served as anemometers for both the wind velocity and the direction of the wind in the air mass through which the moths were flying upwind. Thus the tracks of the moths could be placed in the context of the wind through which they were flying, by measuring the path length in time (the speed) and the direction of a bubble near the moth.

The speed of a bubble, i.e. its path length as measured during a specified time period, can only be determined in a video record having no other reference points if the bubble's height is known. Height measurements were therefore accomplished by recording the tracks of bubbles against the sky on two video cameras and then reconstructing the bubble track over a 0.5 s interval in 3-D by means of a computer program to obtain both its height and its length over the 0.5 s interval. Both the height and the length of 1/30th s vectors of moth tracks were also determined by using the same 3-D reconstruction program. Two Sony RSC 1050 rotary shutter cameras and recorders (SLO 340) were fixed to a moveable flatbed cart. One camera was pointed directly upward toward the sky (vertical), perpendicular to the plane of the ground. The second camera was also fixed to the cart and was angled toward the first camera at 25° from perpendicular. Its lens was 42 cm from the lens of the first camera. Care was taken not to point the cameras toward the sun, and so morning and evening recording periods were selected so that the sun would be near the horizon and away from the upward-pointing cameras. Neutral density filters were placed over the lenses to protect the vidicon tubes of the cameras.

The centre of the fields of view of the two cameras intersected at a height of 90 cm above the vertical camera. Thus there was substantial overlap in the fields of view of the two cameras. A computer program was written to transform the two-dimensional coordinates from the two cameras into a 3-D record. During calibration tests a metre rule stick was suspended over the cameras at various heights, angles and degrees of incline and served as a test of the accuracy of the program's ability to produce height, angle, length and incline readings that were accurate to within ± 5% when 0.5–1.5 m above the cameras and in the middle two-thirds of the fields of view of both cameras. The recordings from the two cameras were synchronized on playback from the two cassette tapes by means of observing the flashes of a randomly long- and short-flashing light-emitting diode (LED) which was affixed to the tip of a 2 mm-diameter, 40 cm-high metal rod at the edge of both cameras' fields of view. An audio record of the events as they happened in the field was created by the two observers speaking into a microphone, and the audio record served to roughly synchronize the two tapes before they were synchronized at the frame-by-frame level using the LED flashes that could be matched to a particular video frame on both tapes as a reference point.

The synchronized tracks of a single moth from the two recordings were played back, and at 1/30th s intervals the distance of the moth (in screen-units) from the centre of the screen of a Panasonic black-and-white video monitor (model WV-5470) was determined. In addition, the angular measure of deviation of each of these moth images was measured from a line running through the centre of the field of view, with 0° located at the top centre of the monitor and 90° located at the left. Thus the position of the moth was tabulated in modified polar coordinates on two recordings. The integration of the two perspectives led to a translation into standard X, Y and Z coordinates. A similar procedure was applied to determine the movement and height of a soap bubble through the field of view. A soap bubble was chosen that was as close to the moth as possible but no more than 2 m away either above, below or beside the moth. Since the wind vector is known (speed and angle) from the bubble track, and the moth's groundspeed and track angle relative to the wind are known each 1/30th s, the computer program calculated the airspeed and course angle using the formula of Kennedy (1940) and Marsh *et al.* (1978) which invokes the triangle of velocities to solve for the third, side of the triangle (airspeed and course angle) (Fig. 1). All bubble and moth tracks were plotted, and the track angles from the records of many moths checked by hand against the bubble vector to make sure that they were correct. These calculations were done by using only the two-dimensional coordinates of the tracks since only the vector indicating the moth's speed relative to the ground and track angle relative to the bubble vector are needed.

Results

We were able to record on the two cameras thirty-six flight tracks of male *G.molesta* flying upwind to pheromone – eighteen at 3 m and eighteen at 10 m from the source – in which there was also a bubble traversing the field of view less than 2 m from the moth and no more than 1 s before or after the moth passed through. We also obtained one track at 30 m from the source. The paucity of tracks at this distance stems from the difficulty in getting the pheromone plume (and bubbles) to pass over the cameras at that distance, and in timing the release of males to coincide with the bubbles passing over the cameras. There were nearly as many additional tracks recorded at the 3 and 10 m distances on only one of the two cameras, but they were not usable because information on the height of the moths, their vertical movement, and their horizontal deviations, was unobtainable. The usable tracks amounted to a 0.5–2 s long

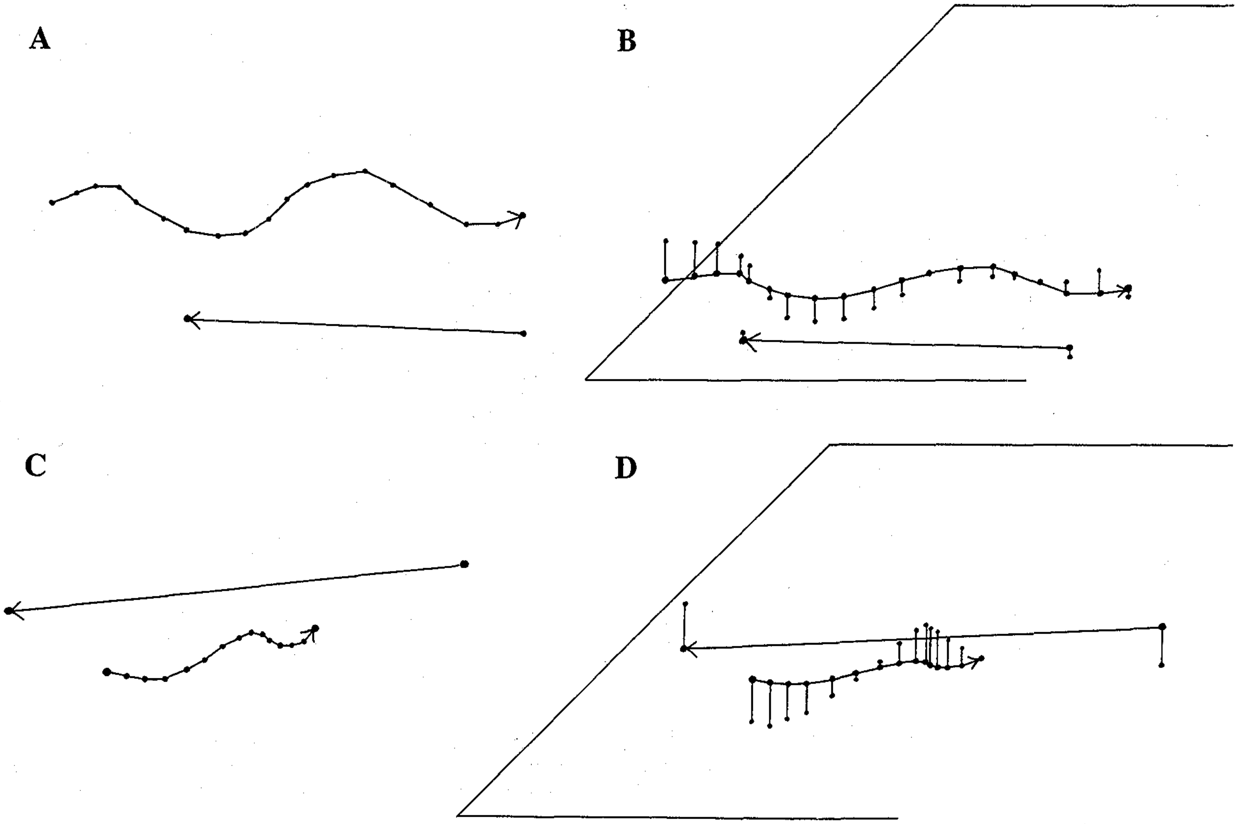

Fig. 2. Flight tracks of two *G.molesta* males that exhibited zigzagging-type flight in response to pheromone 10 m away from the pheromone source. The tracks are shown in two (A and C) and three dimensions (B and D) with moths' positions shown every 1/30th s and the bubbles' tracks (straight lines) shown for a 0.5 s interval. In B and D, the projection of the tracks onto a two-dimensional plane is shown with the actual vertical deviation of the moth from that plane indicated by vertical lines projecting upward or downward to the moth's location every 1/30th s. The moth's track in A is 71 cm in length, and the track in C is 33 cm in length.

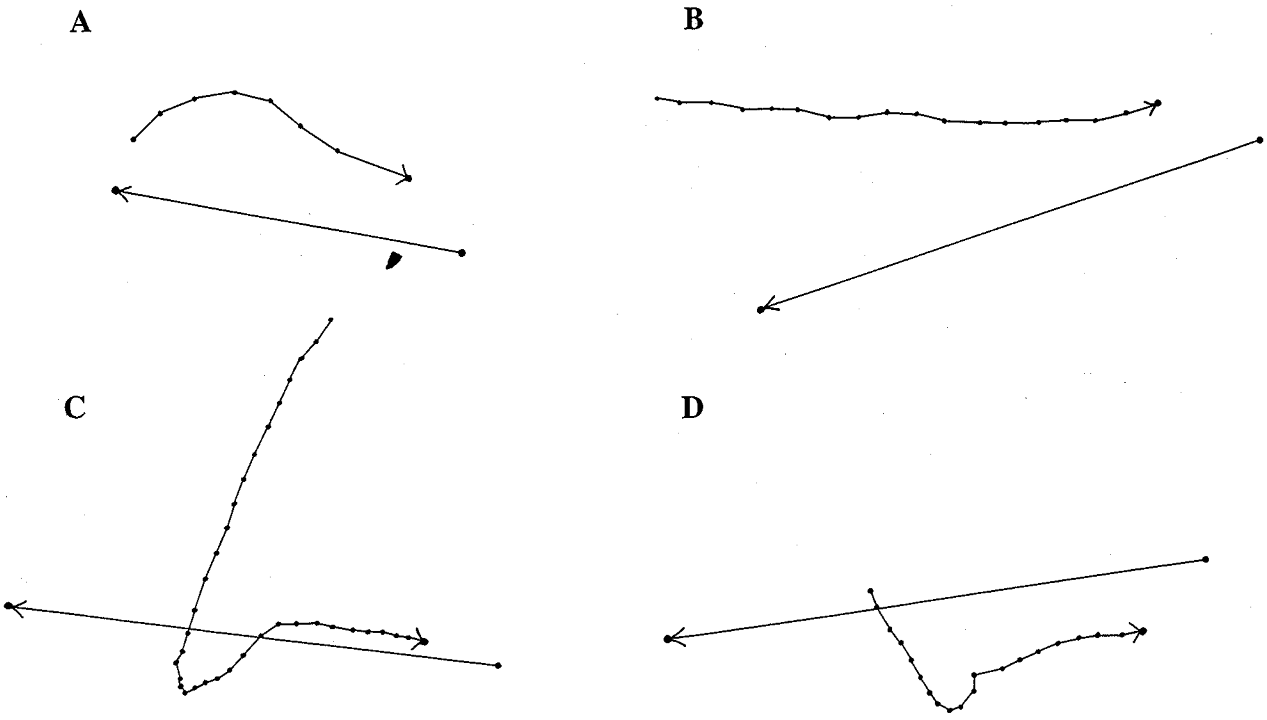

Fig. 3

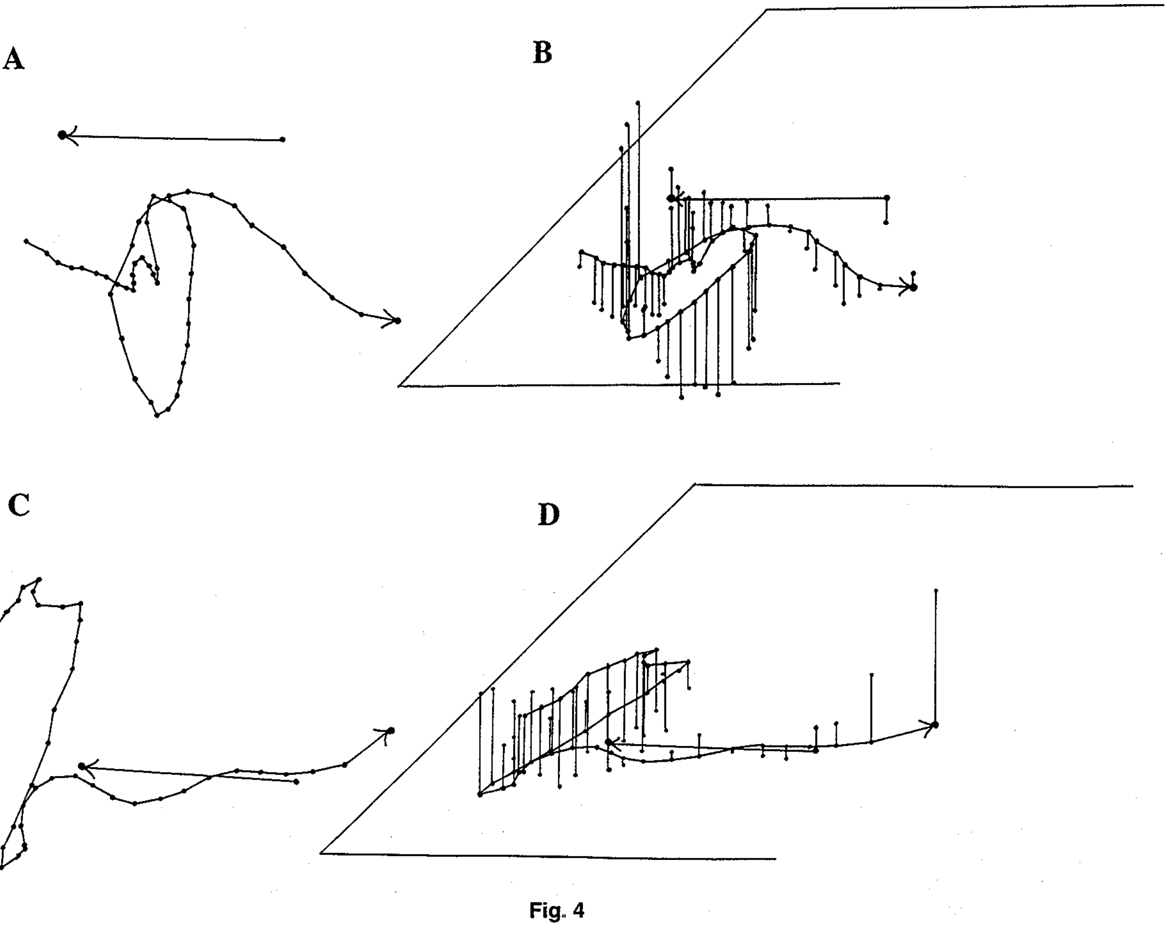

Fig. 4

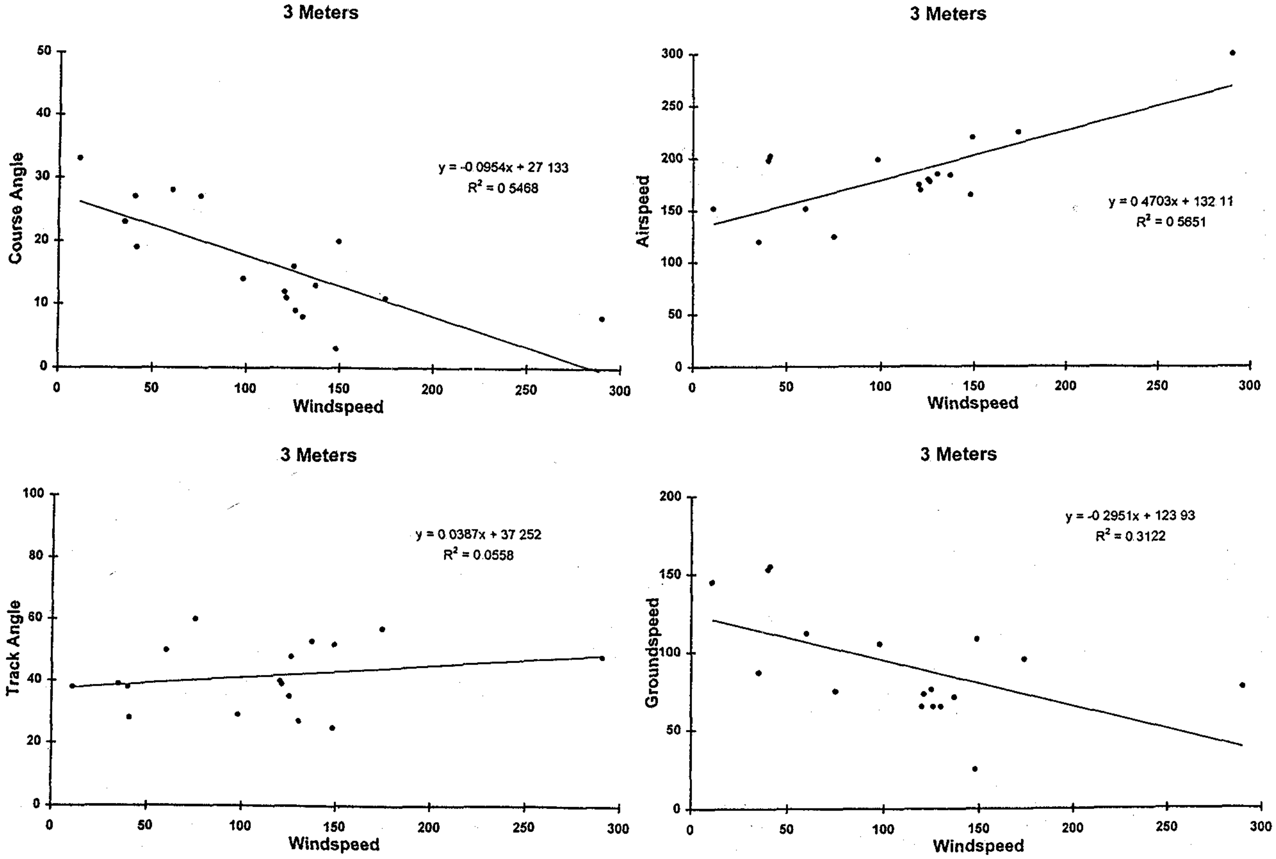

Fig. 5. The effect of windspeed on the behaviours (course angles steered, upper left; and airspeeds, upper right) performed by male oriental fruit moths as they flew upwind in response to pheromone at a distance of 3 m downwind of the pheromone source. The resulting track angles and groundspeeds related to windspeed are also shown, at the lower left and lower right, respectively. Each data point represents the average of the values for one moth calculated at 1/30th s intervals during its entire track through the field of view of the cameras. Regression lines for these data points are shown in each graph. Course angles, airspeeds and groundspeeds ($r = 0.684$, 0.752 and 0.559, respectively) were significantly affected ($P \leq 0.05$; 16 df) by windspeed, whereas track angles did not vary with windspeed ($P \geq 0.05$; 16 df; $r = 0.236$).

sequence of a male's progress toward the source, usually with an overall upwind displacement through the fields of view of the camera.

In some of the upwind tracks males exhibited zigzagging (Figs 2A–D), whereas in others there were few or no obvious horizontal oscillations (Figs 3A–D). Several males initiated casting flight while in the field of view of the cameras, and then regained pheromone contact and flew upwind before disappearing from the screen (Figs 4A–D). In addition to the eighteen upwind- or upwind-plus-casting tracks, we recorded a few tracks of males dropping back downwind while casting widely, after having approached close to the source or landing on it, as described on the audio record. In these downwind tracks the males often flew very low to the ground, and thus their images were usually captured on only one camera.

In flying upwind to pheromone, males performed optomotor anemotaxis, that is, they compensated for higher windspeeds by flying with higher airspeeds, often between 200 and 300 cm/s, and steering more into the wind (smaller course angles) (Figs 5 and 6, top) (see figure legends for r values; $P \leq 0.01$

Fig. 3. Flight tracks in two dimensions of four *G.molesta* males that were placed in the category of non-zigzagging upwind flight in response to pheromone at 3 m (A and C) and 10 m (B and D) from the source. Details as in Fig. 1. The males in C and D had apparently been casting when they came into the field of view of the cameras, and then they began surging upwind with a fairly straight upwind track. The males' tracks are 32 cm, 77 cm, 90 cm and 48 cm in length in A–D, respectively.

Fig. 4. Flight tracks of two *G.molesta* males that exhibited a combination of casting flight and upwind flight while in view of the cameras 3 m away from the pheromone source. The tracks are shown in two (A and C) and three dimensions (B and D); details as in Fig. 1. The moth in A (and B) showed first upwind flight with a transition into casting, followed by a resumption of upwind flight. The moth in C (and D) entered the field of view while casting, and then made an upwind zigzagging track as it recontacted pheromone. In both cases one should note the greater vertical deviation during casting than during upwind flight. The moth's track in A (and B) is 139 cm in length, and the track in C (and D) is 132 cm in length.

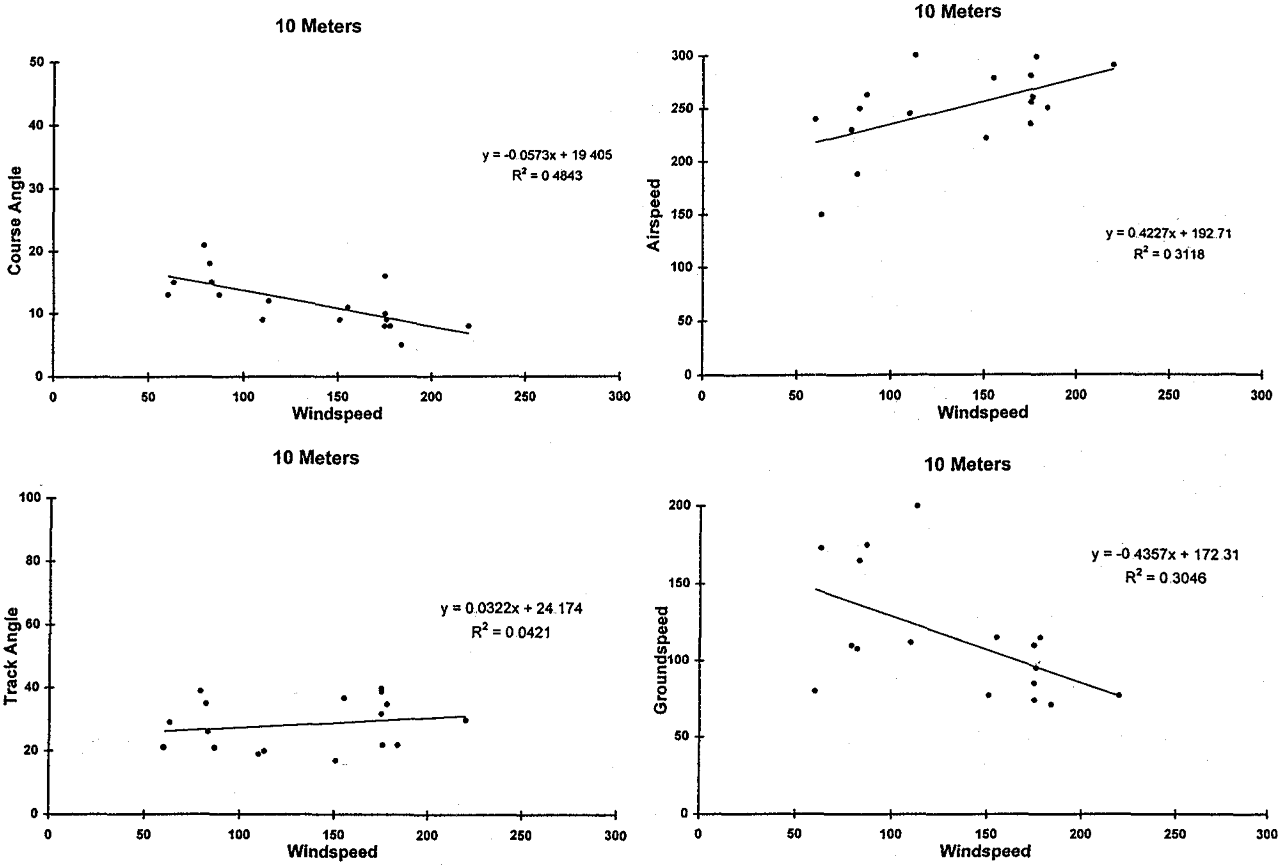

Fig. 6. The effect of windspeed on the behaviour (upper) and resultant tracks (lower) of male oriental fruit moths flying upwind in response to pheromone at a distance of 10 m downwind of the pheromone source. Course angles, airspeeds and groundspeeds (r = 0.696, 0.558 and 0.552, respectively) were significantly affected by windspeed ($P \leq 0.05$; 16 df), whereas track angles did not vary with windspeed ($P \geq 0.05$; 16 df; $r = 0.205$).

for all course angles and airspeeds correlated with windspeed). The result of the males' behavioural compensations for windspeed and direction was a fairly stable track angle upwind of between 20° and 60° regardless of windspeed (Figs 5 and 6, bottom left) ($r = 0.236$, NS, and 0.205, NS, for track angles correlated with windspeeds at 3 m and 10 m, respectively). Males did not appear to control their groundspeed as rigidly as their track angles, due mainly to a tendency for the males to maintain a high airspeed at low windspeeds (below 75 cm/s). Groundspeeds at higher windspeeds appeared to be more stable at between 50 and 100 cm/s (Figs 5 and 6, bottom right).

Males passed over the cameras at an average height of 69.5 cm (±14.62 SD) at 3 m from the source and 75.9 cm (±26.4 SD) at 10 m from the source. Thus, the somewhat higher groundspeeds at 10 m than at 3 m cannot be explained by a greater height of flight at 10 m compared to 3 m, wherein faster flight at a higher altitude would be needed to achieve the same velocity of image movement as at a lower height. In this limited sample of tracks it appeared that males at 10 m flying in similar windspeeds to those males at 3 m (between 50 and 200 cm/s) steered more into the wind and flew with higher airspeeds than those at 3 m (Figs 5 and 6, top). Whether this tendency, if real, would be due to a difference in time course of exposure to the more attenuated filaments at 10 m or to shallower peak-to-trough filament concentrations (Murlis *et al.*, 1990; Baker & Haynes, 1989) is not known. The highest average height at which a moth was observed to fly upwind in pheromone over the cameras was 126 cm, whereas the lowest was 40 cm. We know that even lower-flying males passed upwind through the field of view (due to their extremely large image on one of the cameras), but their behaviour could not be analysed due to being recorded on only one of the two cameras.

During upwind progress, side-to-side deviations (between left and right reversals) were greater than vertical deviations (Fig. 2). For moths exhibiting zigzagging flight upwind, the mean side-to-side deviation was 7.2 cm (±4.8 SD) and the mean ratio of side-to-side to vertical deviation was 1.44 (±1.02 SD). For males flying upwind with no discernible oscillations, the mean curvature of the track from the time they entered the field of view to the time they left was 13.5 cm (±7.6 SD; $n = 10$ males), whereas the mean vertical change was 11.0 cm (±6.7 SD, $n = 10$ males), with an overall mean ratio of side-to-side to vertical motion of 1.45 (± 0.90 SD; $n = 10$ males).

Five males exhibited casting tracks amongst their upwind tracks (Fig. 4), and these were analysed separately. The mean side-to-side deviation during casting flight was 17.0 cm (±2.2 SD; $n = 5$

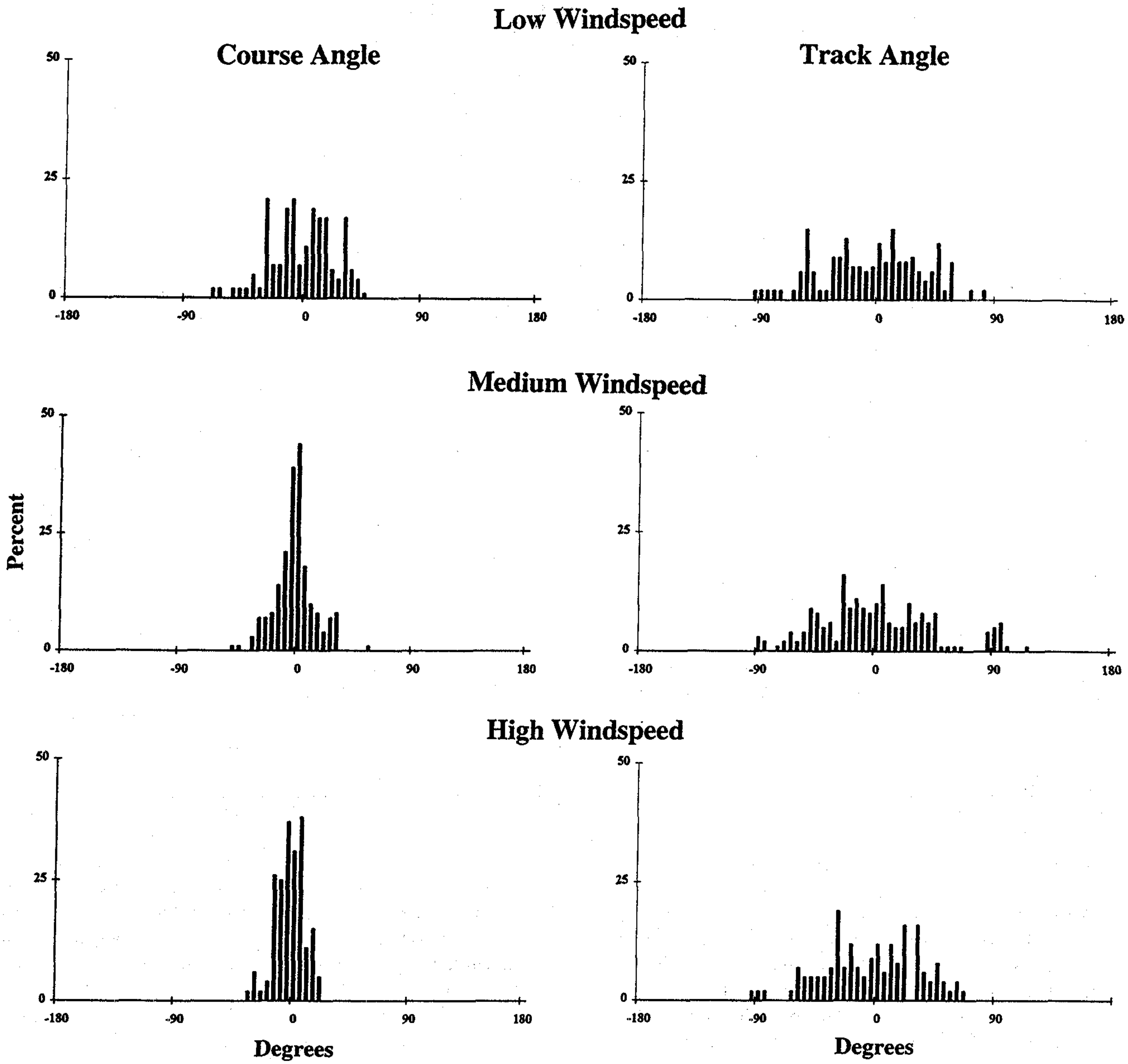

Fig. 7. The distribution of course and track angles, taken in 1/30th s vector intervals, of *G. molesta* males flying upwind to pheromone either with a zigzagging or non-zigzagging track, at low (0–75 cm/s), medium (76–150 cm/s), or high windspeeds (>150 cm/s).

males), whereas the mean vertical deviation during these same casting tracks was 15.6 cm (±4.2 SD); $n = 5$ males). The overall mean ratio of side-to-side to vertical displacement during casting was 1.2 (±0.4 SD; $n = 5$ males).

During casting, the mean inter-reversal duration of the casting track legs was 0.27 s (±0.03 SD; $n = 5$ males at 10 and 3 m combined), whereas during upwind zigzagging males exhibited an inter-reversal duration of 0.15 s (±0.05 SD; $n = 18$ males at 10 and 3 m combined). There was no significant difference between the inter-reversal duration during zigzagging upwind flight at 10 m (0.15 s) compared to 3 m (0.16 s) (±0.05 SD; $n = 10$, and ±0.04 SD; $n = 8$, respectively, t-test; $P \geq 0.05$) or at different windspeeds.

For all upwind tracks at 3 and 10 m of both the zigzagging and non-zigzagging types, the distributions of course and track angles measured for each 1/30th s vector were centred about 0°, regardless of the wind velocity (Fig. 7). There was little or no indication of a bimodal distribution of either course angles or track angles. Both course and track angles exhibited a bimodal distribution during casting flight (Fig. 8).

During the several periods in which many males regressed downwind, away from the source, the height of flight was described on the audio recordings as being at about the same height or lower than that at which males had flown upwind. Only a few small portions of these tracks were successfully recorded on both cameras with bubbles present as well, to

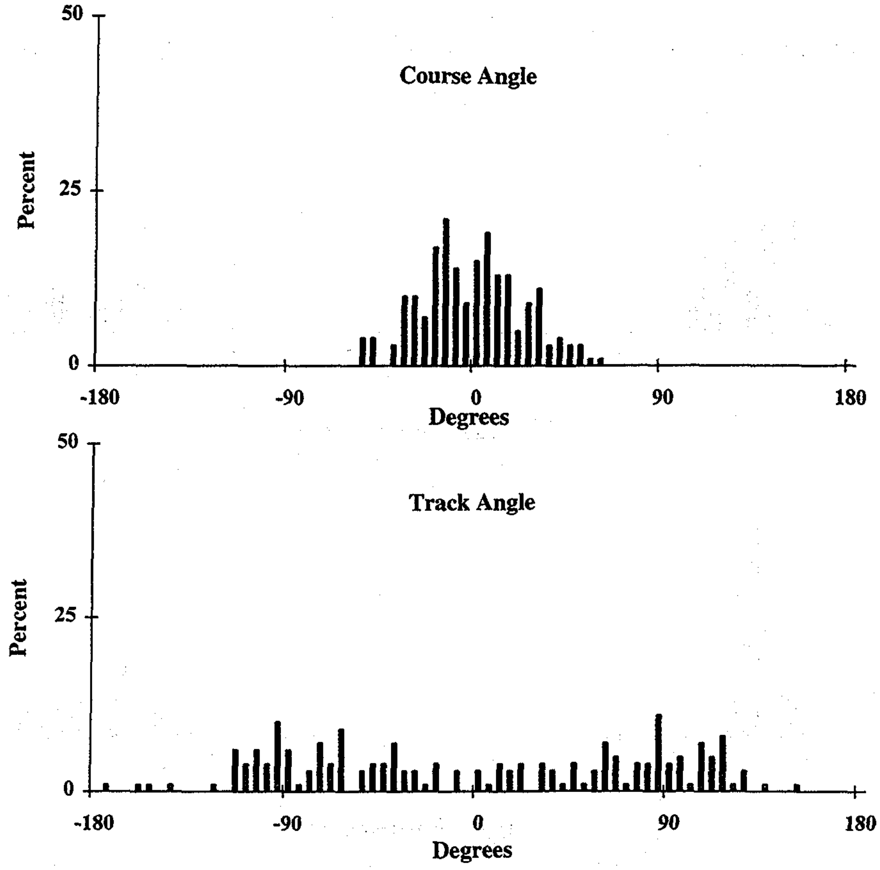

Fig. 8. The distribution of course and track angles, taken in 1/30th s vector intervals, of *G. molesta* males during casting flight.

allow them to be subsequently analysed and reveal the males' behaviour. Some of the tracks appear to be those of males casting downwind regressively while maintaining an upwind heading (course angle) (Fig. 9B), as revealed by track analysis that calculates the course angles. Other tracks (six in total) occurred that included periods during which the males were calculated to be steering courses greater than 90° relative to the wind (i.e. in the downwind direction). In other words, these moths were flying downwind, not merely displacing downwind, as during regressive casting. In one of these moths the body orientation was recorded clearly, and showed the heading (course angle) of the moth in the downwind direction (Fig. 9A). The mean course angles steered by each of the six males during these downwind flights were calculated to be greater than 90° relative to the wind (101.2°, 103.8°, 114.9°, 167.8°, 161.4° and 103.3°, respectively). This created downwind track angles for the track of each male of 136°, 143°, 142.6°, 171.3°, 146.4° and 143°, respectively). The overall mean course angle of the group of six moths was 130.2° (±31.8° SD) (downwind), which created a mean downwind track angle of 147.1° (± 12.4° SD). We were not able to observe whether these males then progressed upwind once more from their location downwind of the cameras.

The groundspeed of each of six males exhibiting such downwind flights was greater than the windspeed in each case, indicating a downwind heading. The mean groundspeed was 142.1 cm/s (±15.0 SD; $n = 6$), which was greater than the mean windspeed of 112 cm/s. The airspeed in each case was lower than the groundspeed and also was lower than the typical levels observed during upwind flight (between 150 and 300 cm/s) with a mean airspeed for the six downwind-flying moths being 120.0 cm/s (±16.0 SD).

Discussion

The flight tracks of *G. molesta* recorded in the field revealed that oriental fruit moth males flying upwind in response to pheromone compensated for the wind by flying faster and steering more into the wind at higher windspeeds, resulting in progress over the ground at a fairly constant resultant track angle and groundspeed. This anemotactic response of changing both airspeed and course angle is similar to that reported for *Plodia interpunctella* in the wind tunnel (Marsh *et al.*, 1978, 1981). *Spodoptera exigua* were observed to fly with higher net airspeeds in the field under higher compared to lower wind velocities in order to maintain a rather consistent net groundspeed (Murlis & Bettany, 1977), showing some kind of anemotactic compensation. However, the details of how changes in course, airspeed and counterturn duration were integrated to produce the change in net airspeed were not reported. A recent, highly detailed study of *L. dispar* male

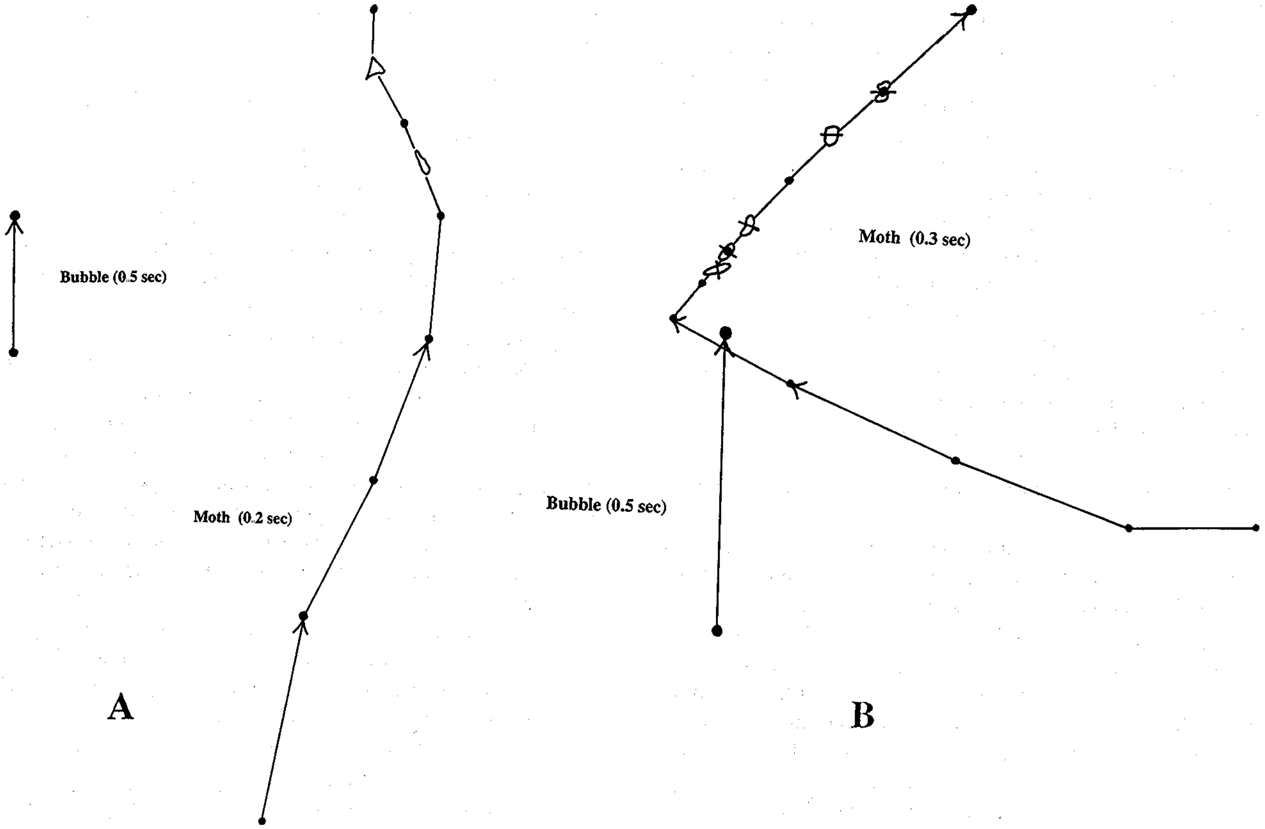

Fig. 9. The flight tracks (plus outlines of the bodies and wings where visible) of two *G.molesta* males that exhibited downwind movement during the recordings. In (A) the male progressed downwind faster than the wind by steering a downwind course and adding its airspeed to the windspeed to create a groundspeed that was greater than the windspeed. In (B) the male exhibited regressive casting while moving through the camera's field of view and the longitudinal body axis position has been augmented by the addition of straight lines.

flight behaviour in the field did not analyse the anemotactic response of males but reported important information on the changes in manoeuvres with distance from the source, and therefore with changes in plume structure and concentration (see below) (Willis *et al.*, 1991).

The males in the current study behaved similarly to *G.molesta* males in laboratory flight tunnels in response to pheromone. The amplitudes of side-to-side zigzagging and casting were about the same as reported in flighttunnel studies (Kuenen & Baker, 1983; Willis & Baker, 1987; Baker & Haynes, 1987). This is not to say that even wider casting tracks do not exist in the field for males that have lost pheromone for a longer period of time than we apparently recorded here. Casting tracks become progressively wider over the course of several seconds after pheromone loss in the laboratory (Baker & Haynes, 1987), and we only sampled casting during 1 or 2 s, at most, following apparent pheromone loss in these field tracks. If many seconds had elapsed after pheromone loss, our small field of view would make it unlikely that we would record the very wide and infrequent reversals at the apices of the long straight legs that are typical of casting flight. Even if one or two straight legs angled across the wind-line occurred in the field of view of the recordings it would be difficult to declare the tracks as casting. The recordings in three dimensions support previous descriptions of 'zigzagging' flight in *G.molesta* males flying upwind to pheromone in the field (Rothschild & Minks, 1974; Cardé *et al.*, 1975; Baker & Cardé, 1979). The side-to-side deviations were greater than the vertical during upwind flight, supporting the impression of predominantly 'zigzagging' (side-to-side) movement in the laboratory as well (Von Keyserlingk, 1984; Baker, 1989a, b; T. C. Baker, M. A. Willis and K. F. Haynes, unpublished observations). In addition, even during casting, the dimensions and ratios of the side-to-side compared to vertical oscillations were virtually identical to those measured in the laboratory (Von Keyserlingk, 1984). Course angles and track angles of males here were also similar to those determined in laboratory experiments (Baker & Haynes, 1987; Willis & Baker, 1987), as were the counterturning frequencies (durations of the intervals between counterturns) during both zigzagging and casting flight (Baker & Haynes, 1987).

One major difference between these field tracks and those from the laboratory was that the airspeeds developed by males were far greater than those in the wind tunnel. One reason for this difference is that for many of the males the windspeeds they encountered in the field were up to 4–5 times as great as the usual laboratory velocities of 50–70 cm/s, and the anemotactic compensation that the males exhibited would require whatever airspeed is needed to maintain the same groundspeed (Fig. 4). Another factor is that in the field males flew over the ground at higher altitudes (73 cm, on average, from the closest visible ground pattern, the cameras' lenses) than in the wind tunnel, where the plume was kept at an average of 15 cm from the floor and *c.* 30 cm from the spotted ground pattern. Raising the plume height and forcing males to fly higher above the ground does cause male *G.molesta* to increase their net upwind groundspeed and their net airspeed, even in the wind tunnel (Kuenen & Baker, 1982). At 10 m from the source, it remains to be seen whether or not a lower overall concentration or lower fluctuation of concentration in attenuated pheromone filaments additionally contributes to the higher airspeeds. This is the first time that behaviours (and tracks) have been recorded and analysed in *G.molesta* males at such a great distance from a pheromone source.

The fairly unimodal distribution of course angles steered by males under different windspeeds is not unexpected, since very few of the recorded tracks involved much casting flight or casting legs. Baker (1990) hypothesized, and Vickers & Baker (1994) and Mafra-Neto & Cardé (1994) have shown, that in moths a bimodal distribution of course angles on either side of the wind-line is indicative of more bouts of casting flight due to insufficiently frequent contacts with pheromone filaments of optimal quantity and quality. A unimodal distribution would result from reiterative and sufficiently frequent contact with filaments such that casting is rarely elicited. Additionally, the high airspeeds developed under the conditions of these studies require that for the given resultant track angles the course angles needing to be steered must be very low, with little room for significant oscillation of steering off of the wind-line.

In another field study (Baker & Haynes, 1989), at windspeeds similar to or slightly lower than those experienced during these behavioural recordings, EAG depolarizations caused by pheromone emitted from the same rubber septum dispensers placed in the same position on the bubble generator occurred at an average of 1.1 s^{-1} at 3 m and 0.9 s^{-1} at 10 m from the source, during 10 s sample intervals. Depolarizations occurred more frequently (2–3 s^{-1}) during recurrent short intervals of 1 or 2 s. If the contact with filaments due to the moth's increased airspeed of 2–3 m s^{-1} is factored in, then filament contact during upwind flight should occur at an average frequency of *c.* 2–3 s^{-1} at both distances during upwind flight along the plume, with recurrent periods of between 4 and 9 contacts s^{-1} under the conditions in these studies. Counterturning frequency was 6.7 turns s^{-1} during upwind flight in these moths, which is as high as it ever gets during sustained upwind flight, even under optimally unvarying wind and plume conditions (Kuenen & Baker, 1983; Willis & Baker, 1987; Baker & Haynes, 1987). Thus in this study there was very little opportunity to observe any long bouts of casting flight in males because they passed rapidly through the field of view and apparently experienced very little clean air.

Witzgall & Arn (1990) observed a bimodal distribution of course angles in males of *Lobesia botrana* flying upwind in response to synthetic sex pheromone but not to female pheromone gland extract. They viewed zigzagging flight tracks (and the bimodal distribution of course angles) as an artefact of improper stimulation of the moths, and implied that zigzagging should not occur in moths if the correct blend of synthetic components (or the naturally extracted or emitted blend) is used. Their overall response level (source contact) using the synthetic 'pheromone' was quite low (25%) compared with the natural extract (67%), and so their conclusion that their synthetic blend was suboptimal and perhaps the cause of suboptimal steering is justified. However, response levels of *G.molesta* to the blend and concentration of synthetic components used in the present study usually exceeds 80–90% (Baker & Cardé, 1979; Baker *et al.*, 1981; Linn *et al.* 1986, 1987). Therefore the quality of the synthetic blend should not be as big a factor in this species as are physical factors such as the fine structure of the plume, windspeed, airspeed, frequency of filament contact and the way these factors interact with the innate reaction time of the moths to pheromone onset and offset (Baker, 1990; Vickers & Baker, 1994). The result, varying mixtures of casting and upwind surging, explains the zigzagging shape of the tracks.

The downwind, regressive casting of males leaving the pheromone source observed in a few recorded tracks had not been reported before from the field for *G.molesta*, nor had the downwind headings taken up by males that performed the most rapid downwind displacement after leaving the source. Wind-tunnel recordings from our laboratory had shown regressive casting with downwind displacement to be commonplace (Von Keyserlingk, 1984; Willis & Baker, 1987, 1988; Baker & Haynes, 1987), including instances of males flying upwind then becoming arrested in the plume due to high concentrations, following which their displacement became downwind during regressive casting (Willis & Baker, 1987, 1988). Such arrestment is known to be associated with the adaptation of antennal receptors (Baker *et al.*, 1988). There were no instances of *G.molesta* males actively flying downwind in the wind tunnel, however. Gibson *et al.* (1991) observed tsetse flies, *Glossina morsitans* and *Glossina pallipides*, flying downwind in the field in odour-free air, but such flight was not known to have occurred after contact with attractive odour, as was the case for the oriental fruit moth males in our current field study. Presumably the males we observed leaving the source had become adapted to the concentrations of pheromone there and had therefore 'lost' the plume.

The significance of actively flying downwind following pheromone contact, with rapid displacement faster than the wind, can only be speculated upon. Keeping station and scanning more widely side-to-side by casting does aid in relocating a recently lost plume (Baker & Haynes, 1987), but doing so for a long time after losing pheromone is no guarantee that the wind will shift the plume back over the moth. Such relocation may be ensured, however, by the male abandoning casting flight and flying actively downwind, due to the fact that in a shifting wind-field over relatively open ground, pheromone generally follows straight-line trajectories away from the source (David *et al.*, 1982, 1983). Downwind displacement faster than the air mass itself moments after losing the plume thus should always lead to relocating the lost pheromone, albeit at a much greater distance from the source than where it was lost. The male, having caught up with the receding, pheromone-bearing air, could then begin its usual reiterations of pheromone-mediated upwind flight and casting flight. This is the first time that odour-experienced moths have been shown to actively steer downwind, although these were very short snippets of the larger tracks, capturing only fractions of seconds of this behaviour. It remains to be seen for how long such males maintain this downwind heading, and whether they do eventually catch up with and respond to the 'lost' pheromone.

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