

Flight of *Heliothis virescens* males in the field in response to sex pheromone

NEIL J. VICKERS* and THOMAS C. BAKER

Department of Entomology, Iowa State University, Ames, Iowa, U.S.A.

Abstract. The behaviour of *Heliothis virescens* males flying upwind in the field in a sex pheromone plume was videorecorded and analysed. Males flew faster and straighter, with less counterturning, and heading more directly into the wind when they were 9–11 m away from the odour source than when they were 1–3 m away. Regardless of their distance from the source or the windspeed, they maintained an average groundspeed of c. 200 cm s^{-1} , except when they arrived within 1 m of the source, when their groundspeed slowed significantly. Two or more males flying in the plume at the same instant often exhibited either extremely straight and directly upwind tracks or else zigzagging tracks with significant counterturning (as did males flying through the field of view of the cameras at slightly different times). The males' position, either in the centre of the plume's axis or along one side, might explain these differences in track straightness, which previous studies with *H. virescens* have shown to be caused by higher frequencies of contact with plume filaments. When a significant shift in wind direction occurred, males tended to make an initial movement in the direction of the shift, perhaps due to latencies of response in both the olfactory and visual systems associated with flying into clean air. The males' behaviour in the field overall was similar to that observed in the wind tunnel, except that their airspeeds and groundspeeds were significantly higher than those observed in the laboratory. The fact that they flew faster in the field can be explained both by the significantly higher windspeeds that males need to compensate for in the field to attain a preferred velocity of image motion, as well as by a higher height of flight over the ground in the field causing a slower apparent motion of images at a given groundspeed compared with the laboratory.

Key words. Sex pheromone, *Heliothis virescens*, flight behaviour, field behaviour, pheromone plume, windspeed.

Introduction

Over the past 20 years or so the mechanisms used by male and female moths to locate sources of pheromone or host odour have been revealed by studies by many research groups using sex pheromones to evoke controlled flight in male moths flying in wind tunnels. 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. However, 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 allow conclusions to be drawn concerning the behavioural reactions of males to different wind speeds and directions.

Heliothis virescens, the tobacco budworm moth, has been the focus of a series of experiments in the wind tunnel designed to understand the mechanisms involved in males' in-flight location of sex pheromone sources. The results of these experiments have provided some key insights into how males surge upwind in response to contact with pheromone filaments in a typical finely structured plume, and how they lapse into sustained casting behaviour in pockets of clean air between filaments (Vickers & Baker, 1992, 1994, 1996, 1997). The quality of the odour blend in the filaments determines the strength of each surge upwind (Vickers & Baker, 1997).

In the current study we sought to understand how *H. virescens* males behave under field wind conditions, where the wind

* Present address: Division of Neurobiology, Arizona Research Laboratories, University of Arizona, Tucson, AZ 85721, U.S.A.

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

velocity and direction can change significantly from moment to moment, and also where the males are able to be observed flying from greater distances from the source than can be observed in a wind tunnel. We then sought to try to relate the male behaviour analysed in the field to that which we had observed in the wind tunnel. The key components to this field analysis were the use of the triangle of velocities analysis of movements (Kennedy, 1940), and a computer-based analysis system that integrates the windspeed and direction (by using bubbles) near the moth's position in space with the moth's groundspeed and direction 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 *et al.*, 1984; Baker, 1986).

Methods and Materials

Moths. Male *H. virescens* were reared on a modified pinto bean diet (Shorey & Hale, 1965). Males were isolated from the females at the pupal stage and placed in a separate environmental chamber on a LD 14:10h cycle with an 8% sugar water solution. Males were utilized between the fifth and eighth hours of their fourth to eighth scotophase. On the day that they were to be released in the field they were placed in small cylindrical wire mesh cages (6 cm high × 6 cm diameter). Four or five males were placed in each cage. The cages were then placed on a plastic retaining tray (twenty cages per tray) and returned to the environmental chamber. As darkness fell the moths were removed from the environmental chamber, taking care not to expose them to any stray light. The trays were wrapped in a thick black cloth during transportation to the field. Once it was dark at the field site the trays were uncovered to allow the moths at least 30 min of acclimation to the ambient conditions.

Field recording. The field site used was at the Agricultural Operations facility of the University of California, Riverside. A single Sony 1050 rotary shutter video camera was located on top of a 15 m-high weather tower. The tower had a platform allowing an operator and the necessary equipment to be placed alongside the camera. The camera was directed straight down at the ground and was fixed to a metal bar slightly away from the tower. Moth flights were recorded on a Sony SLO 340 video cassette recorder. The signal from the camera was first processed by a FOR. A time/date generator that imprinted the tape recording with the date and a stopwatch. Lighting was provided by two powerful architects' lamps (DR 3000-1, Underwriters Laboratories) fitted with quartz bulbs. Each lamp was fitted with a Rosco deep red filter (no. 27) that transmitted wavelengths only >600 nm. Despite the fact that the light was likely not completely invisible to the moth eye (as infra-red would be), no moths were observed to fly even slightly toward the lights, and there was no observable bias in the flight tracks toward one side of the windline or the other.

The moths were flown over bare sandy ground with rocks and various bits of debris scattered about randomly. The pheromone source (and bubble generator, see below) were stationed variously at 2, 5 and 9 m from the centre of the camera's 4.5 × 3 m field of view, to record flight tracks of males flying at different distances from the source. Recordings were made during September and October 1991.

Bubble generator. In order to measure accurately the wind velocity and proximity of pheromone to the flight path taken by the moths, a bubble generator was used (as per David *et al.*, 1983). This device produces long-lasting bubbles at the point of origin of the pheromone plume (Baker & Haynes, 1989). The bubbles did not appear to interfere with the flight of the moths. The bubble generator, secured to a large tripod 1.5 m above the ground, was positioned according to prevailing wind conditions.

Pheromone. A stock solution of 10 µg/µl of a six-component mixture containing Z11-16:Ald, Z9-14:Ald, 16:Ald, 14:Ald, Z9-16:Ald, and Z7-16:Ald was maintained. The mixture contained the components of the pheromone in the ratio 100:2.5:50:5:1:1, respectively, in hexane. This mixture elicits high levels of upwind flight by *H. virescens* males in wind tunnels and was formulated to mimic very closely the blend ratios emitted from the extruded pheromone glands of calling *H. virescens* females (Vetter & Baker, 1983). Individual components of this mixture were >99% pure as confirmed by gas chromatography. 10 µl of the stock hexane solution were loaded onto a rubber septum (A. H. Thomas and Co., No. 8753 D22, sleeve type, 5 × 9 mm) giving a final loading of 100 µg of the most abundant component, Z11-16:Ald. During field recordings the septum was suspended directly beneath the bubble generator at a height of 1.5 m, such that the pheromone plume and the bubbles would be aligned as closely as possible. When not in use the septum was stored at -20°C.

Flight track analysis. Video tapes were replayed frame by frame onto a black and white Panasonic monitor via a Sony S VM 1010 motion analyser. Tracks of moths progressing across the screen were transcribed onto Mylar® sheets. The position of the moth was recorded every 1/30 s. Bubbles appearing on the screen at the same time as the moth also were transcribed, the nearest bubble to the moth being taken. A 1 m ruler on the ground provided a scale for the speed of the bubbles and the moths over the ground. For this study, both the bubbles and the moths were considered to be at a height of 0.75 m in order to determine the windspeed. Observers stationed on the ground during the recordings confirmed that most of the males (and the bubbles) flying through the field of view did so at between 0.5 and 0.75 m. Once windspeed and direction had been obtained, the tracks of the moths were digitized from the Mylar® sheets by using a Radio-Shack computer and X/Y digitizer. Input of the digitized tracks into the computer allowed a program to calculate the males' course angles and airspeeds each 1/30 s, by using their track angles and groundspeeds, plus the windspeed and direction measured from the track of the bubble.

Results

An overall picture of the tracks generated by moths flying through the field of view of the cameras indicated that there were two main types of upwind track. Some moths flew upwind with a slow zigzag flight track (Fig. 1A), whereas others flew relatively fast and straight upwind, deviating little from the windline (Fig. 1B). This variation in track shape did not appear to be a function of differences in wind speed or plume structure, because sometimes two moths simultaneously flying through the field of view exhibited the two types of upwind flight track (Fig. 1C).

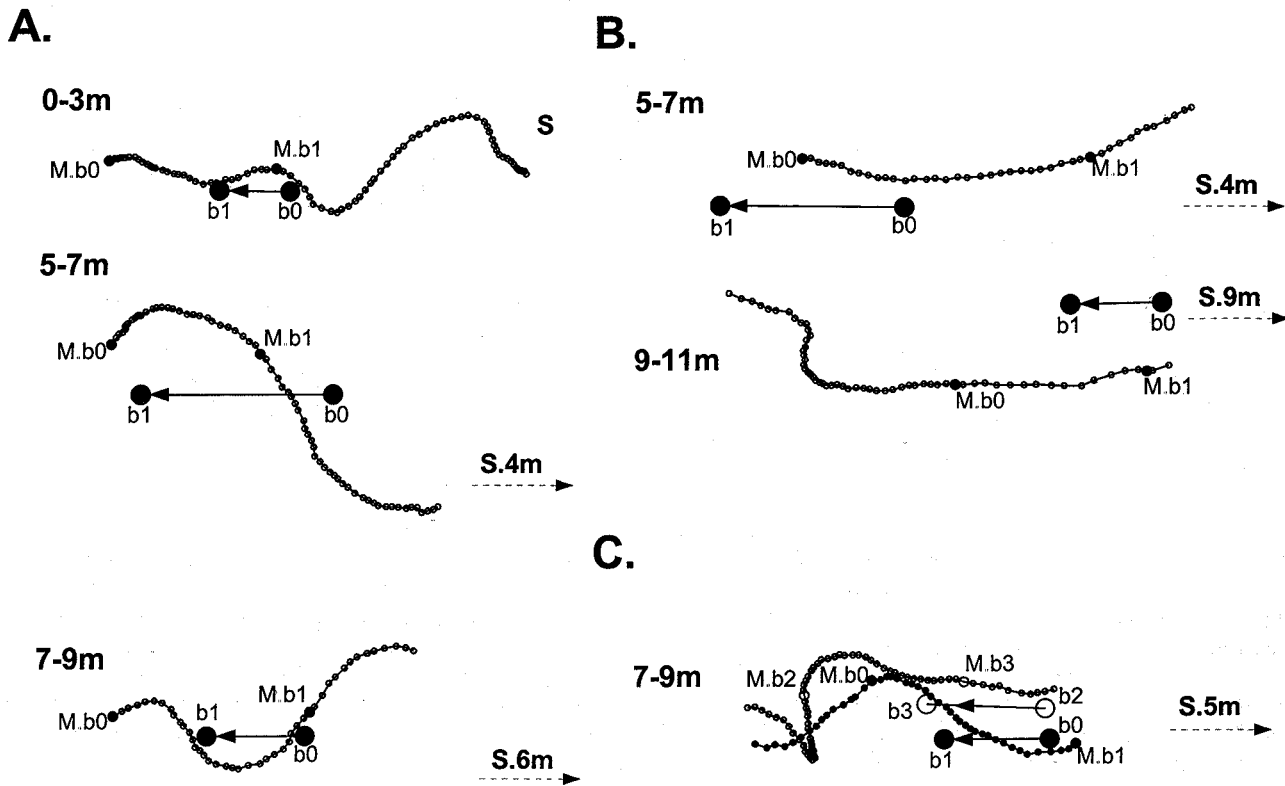


Fig. 1. Some moths that flew through the field of view did so with slow, zigzag tracks (A). Others, within the field of view at almost the same time, flew through very quickly with little deviation from the windline (B). Zigzag tracks occurred at all distances from the source but straight tracks were more likely to occur at the 5–7, 7–9 or 9–11 m distances. The wind direction and speed for each moth is indicated by a solid line connecting two solid circles (bubble path b0–b1). The position of each male at these two points is indicated by M b0 and M b1, respectively. The moth tracks are marked each 1/30 s by small open circles, with positions M b0 and M b1 being enlarged and solid. The approximate position of the pheromone source is indicated by S, with a distance in metres. The distance group (0–3, 5–7, 7–9 or 9–11 m) of each particular track also is given. Two moths flying through the field of view almost simultaneously (C), did so with different track shapes. Moth 1 (track marked by filled circles and corresponding to bubble path b0–b1) had a distinct zigzag track shape. Moth 2 (track marked by open circles corresponding to bubble path b2–b3) flying through the field of view less than 4 s later exhibited an initial cast followed by a direct and straight upwind path. The scale bar is 1 m in length.

In the process of analysing the tracks another interesting phenomenon came to light, that of the moth's ability to relocate the plume in a shifting wind field (Fig. 2A, B). In each case illustrated in Fig. 2, the wind swing was to the males' left. Five out of six males that encountered a windswing such as this appeared to respond by exhibiting their first casting track angle (>60°) toward the correct direction, i.e. toward the displaced plume.

A few tracks were analysed to determine the reactions of individual moths to contact or loss of pheromone. Two representative tracks are shown in Fig. 3. In both tracks the moths' mean airspeeds were faster during the upwind sections of the tracks than during the crosswind sections. This showed that males changed their speed to fly faster over the ground during upwind flight (in pheromone), than during casting flight (in clean air).

At higher windspeeds, males steered courses that were more directly upwind than at lower windspeeds (Fig. 4A; $P=0.0019$) and they also tended to fly faster at higher windspeeds, developing higher airspeeds (Fig. 4B; $P=0.00027$). The result was that males maintained their groundspeed at $c. 200 \text{ cm s}^{-1}$ regardless of the windspeed (Fig. 4C; $P=0.169$, NS), and their track angles centred about 33° regardless of windspeed (Fig. 4D; $P=0.38$, NS).

Overall, when the tracks were averaged for each distance, the data indicate that the farther from the source that males were flying the more they steered into the wind (Fig. 5; $P=0.01$ for course angles), resulting in track angles that were more directly upwind (Fig. 5; $P=0.003$). Also, males farther from the source flew faster, using higher airspeeds, than when they were closer to the source (Fig. 6; $P=0.008$). Interestingly, males maintained a steady groundspeed of $c. 200 \text{ cm s}^{-1}$ regardless of their distance

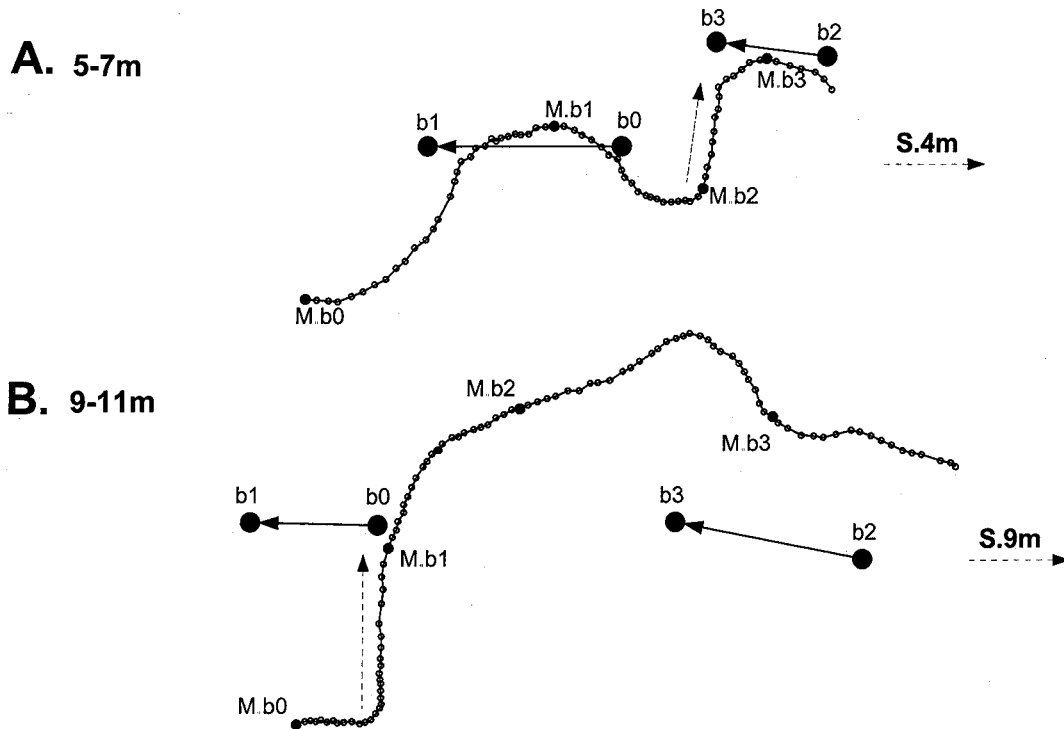


Fig. 2. Males that flew through the field of view during a shift in wind direction seemed to respond to the loss of pheromone, by 'correctly' casting in the right direction (dashed arrows) and hence relocating the shifted plume more quickly than if they had casted in the opposite direction. The initial wind direction and speed are indicated by bubble path b0–b1. In both examples the shifted wind vector is indicated by the second bubble path b2–b3. Tracks are marked each 1/30 s (small open circles) with the position of each male relative to its respective bubbles indicated by M.b0–M.b3 (larger filled circles along the track). The approximate position and distance to the source (S.) in metres is shown by a dashed arrow. One track was grouped in the 5–7 m category (A), the other in the 9–11 m category (B). The scale bar is 1 m in length.

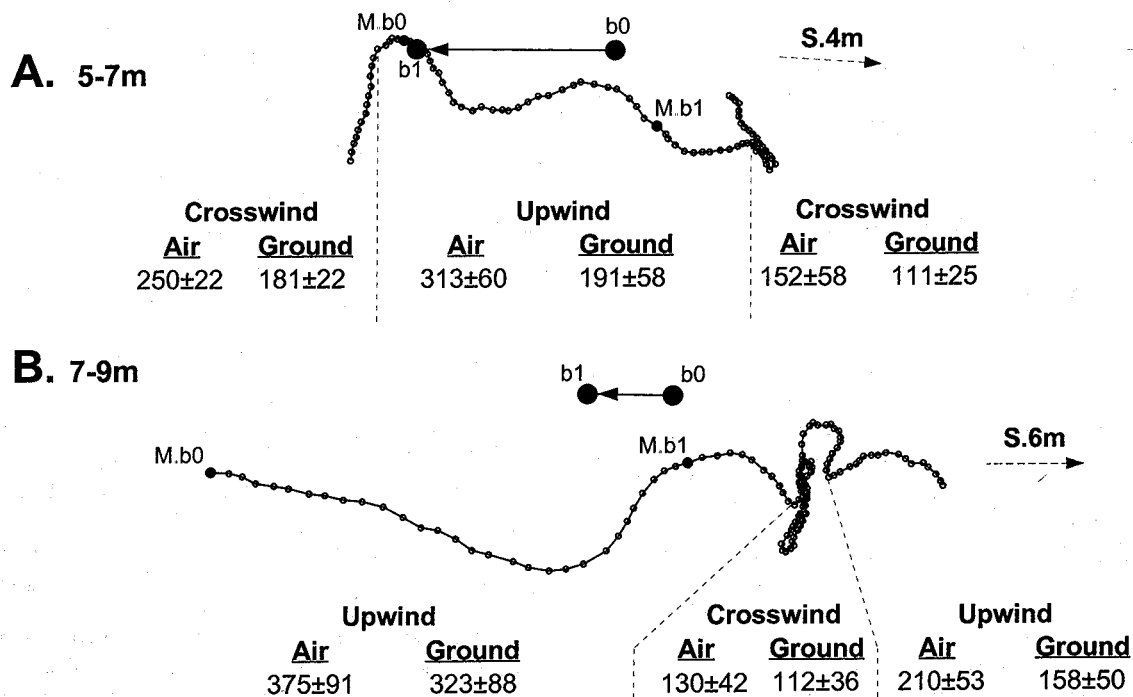


Fig. 3. Tracks of two individual males marked with mean vector airspeeds and groundspeeds in cm/s (\pm SD) show that males increased their airspeeds during pheromone contact as indicated by their performing upwind flight (track angles $<60^\circ$ upwind), and slowed their airspeeds in clean air as indicated by their performing crosswind flight (track angles $>60^\circ$). The transitions between crosswind and upwind flight are indicated by dashed lines. The wind vector is indicated by the bubble vector b0–b1 (large, solid circles) as is the position of the moth relative to the bubbles (M.b0–M.b1, larger solid circles on moth track). The moth track is marked every 1/30 s by small open circles. Windspeeds were calculated as 135 cm/s (A) and 58 cm/s (B). The approximate distance and location of the source (S.) in metres is indicated by a dashed arrow. The scale bar is 1 m in length.

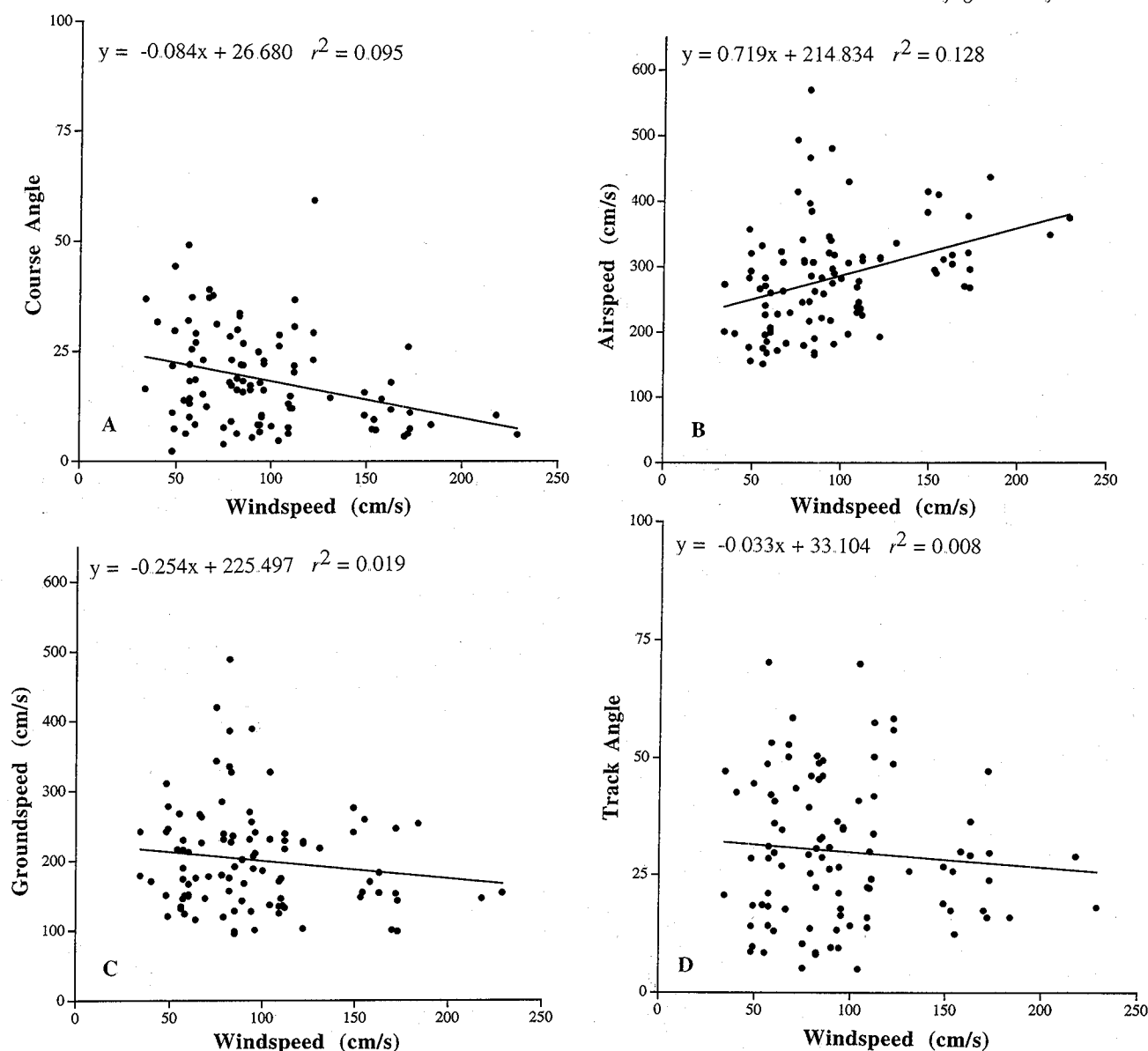


Fig. 4. (A) Mean course angles of individual males as a function of windspeed regardless of distance from the source. This relationship is significant ($P < 0.05$) with $P = 0.0019$. The means of 1/30 s course angle vectors that occurred at 0–1, 1–3, 3–5, 5–7, 7–9 and 9–11 m from the source were calculated and are represented by each black dot in the figure. Angles were positive values whether they were right or left of the windline, and the course angle of a male had a value of 0° if the vector was directly up the windline. (B) Mean airspeeds of these same males, calculated over the same increments as above for course angles. This relationship is significant ($P < 0.05$) with $P = 0.00027$. (C) Mean groundspeeds of these same males, calculated over the same increments as above for course angles. This relationship is not significant ($P > 0.05$) with $P = 0.169$. (D) Mean track angles of these same males, calculated over the same increments as above for course angles. Angles were positive values whether they were right or left of the windline, and the track angle of a male had a value of 0° if the vector was directly up the windline. This relationship is not significant ($P > 0.05$) with $P = 0.38$. In A–D the number of 1/30 s vectors available in each 2 m long track section usually totalled fifteen to thirty, but never was fewer than ten and sometimes reached forty or more.

from the source ($P = 0.38$, NS), except at less than 1 m from the source, where there was obviously a substantial diminution of airspeed, resulting in a diminished groundspeed.

Analysis of counterturning tendencies supported the above data and the impressions from the tracings of the tracks that males fly straighter tracks the farther they are from the source. A lower percentage of males counterturned when flying farther from, compared with closer to, the source (Fig. 7A), and those that did

counterturn farther from the source did so with a lower frequency than males that were closer to the source (Fig. 7B).

Discussion

H. virescens males exhibited a wide variety of manoeuvres when locating the source of their synthetic sex pheromone under field

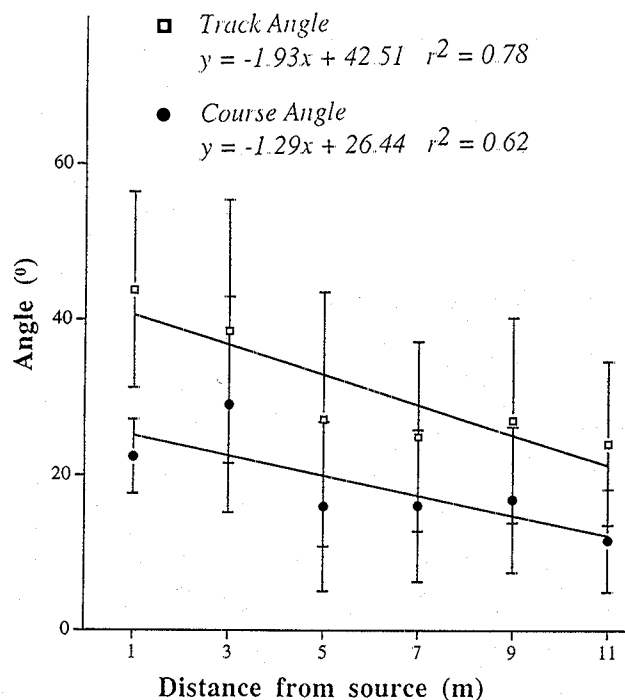


Fig. 5. Mean course and track angles for all moths calculated from each male's mean angle over 2 m increments away from the source, plotted as a function of distance from the source. Bars above and below the means denote standard deviations ($n=10, 15, 20, 30, 15$ and 8 males for 0–1, 1–3, 3–5, 5–7, 7–9 and 9–11 m, respectively). These relationships (course angle and track angle versus distance) are both significant ($P<0.05$) with $P=0.01$ and 0.003 , respectively. The r^2 values in both cases also are both significant.

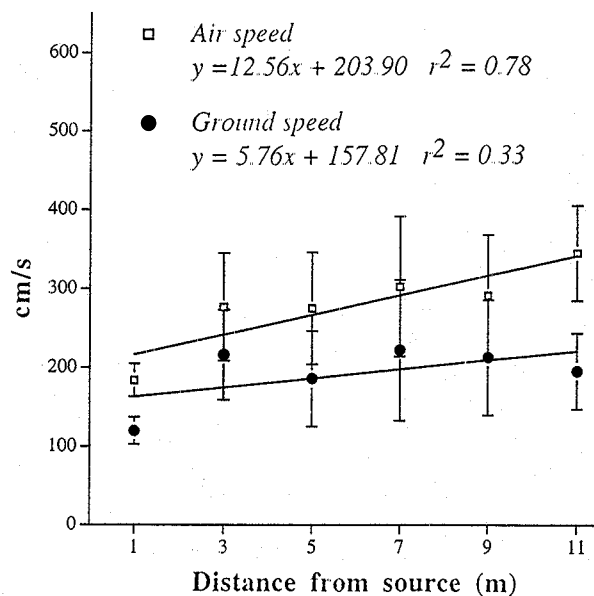


Fig. 6. Mean airspeeds and groundspeeds calculated from each male's mean speed over 2 m increments away from the source, plotted as a function of distance from the source. Bars above and below the means denote standard deviations ($n=10, 15, 20, 30, 15$ and 8 males for 0–1, 1–3, 3–5, 5–7, 7–9 and 9–11 m, respectively). The relationship between airspeed and distance from the source is significant ($P<0.05$) with $P=0.008$ and a significant r^2 value. The relationship between groundspeed and distance from the source is not significant ($P=0.38$) nor is the r^2 value.

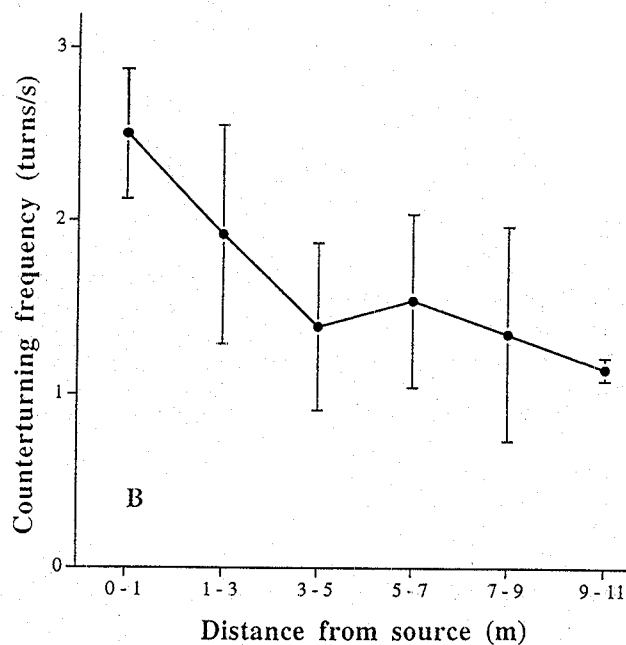
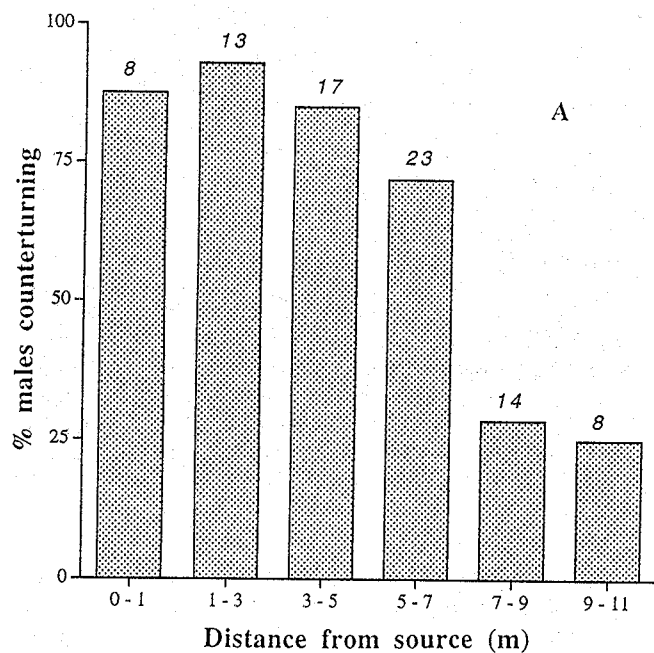


Fig. 7. (A) Percentage of males (with n given above each bar) that exhibited counterturning at different distances from the source as they flew through various 2 m sections toward the source. Percentages at 7–9 and 9–11 m are significantly lower than those closer to the source ($P<0.05$; $\chi^2 2 \times 2$ test of independence). (B) Of males that counterturned at least once over a 2 m section toward the source, the frequency of counterturns that were exhibited at different distances from the source. Bars above and below the means denote standard deviations. Mean counterturning frequencies at 3 m or farther from the source are significantly lower than 0–1 m from the source ($P<0.05$; ANOVA followed by Tukey's w procedure).

conditions of wind that shifted direction and changed speeds. They responded anemotactically by steering more into the wind and flying faster when there were higher windspeeds. They counterturned less frequently when they were farther compared with closer to the source, and they also flew faster and more directly upwind when they were farther from the source. Similar changes in behaviour (except for counterturning frequency, which was not analysed) were observed in gypsy moth males responding to pheromone at different distances from the source (Willis *et al.*, 1991).

Previous studies in the laboratory showed that track straightness in *H. virescens* males responding to pheromone is a result of the frequency of contact with individual filaments of pheromone in the finely structured plume (Vickers & Baker, 1994, 1996). Contact with a single filament causes males to surge more directly upwind with higher airspeed, and exposure to the clean wind following the filament causes the surge to wane with crosswind steering and counterturning now being expressed (Vickers & Baker, 1994, 1996). During the surge, males regulate their altitude more narrowly than they do during their casting and counterturning in clean air, resulting in flutter flight, which probably contributes to the higher airspeeds (Vickers & Baker, 1996). The shape of a male's flight track in response to a pheromone plume from a point source of pheromone is thus a series of reiterative surging and casting responses to the filaments, with more frequent contact with filaments causing straighter, flatter, and more directly upwind flight (Vickers & Baker, 1994, 1996). The quality of the pheromone in the filaments also determines the straightness and length of the surge in response to a single filament (Vickers & Baker, 1997). Each single surge can become stunted, for instance, when the pheromone blend is tainted with the inclusion of a small amount of antagonist, and in a continuous point-source plume in which the antagonist is present, this results in less upwind progress, more casting, and finally arrestment of upwind progress and failure to reach the source (Vickers & Baker, 1997).

Under the field conditions of the current study, the quality of pheromone was such that no antagonist was present, and optimal upwind flight could be elicited. The tracks of many males were extremely straight and directly up the windline, implying optimal and frequent contact with filaments, producing reiterative upwind surges with no time for counterturn-dominated casting flight to be expressed in between filament contact. Other males, often flying simultaneously in the same plumes as the straight-flying males, exhibited more zigzagging flight, still aimed upwind but alternating left and right across the windline. Presumably these males were not contacting filaments as frequently as the straight-flying males. Individual variation in flight track shape is known in *H. virescens* males to result from the exposure of a male to a particular plume structure, creating a certain probability of filament contact by the male (Vickers & Baker, 1991). For instance, if a male enters the plume and immediately positions himself directly within the central axis of a plume, this can produce nearly perfectly straight upwind flight. However, if the male enters the plume off to one side and he maintains this off-axis positioning while progressing upwind, his track will zigzag due to the less frequent contact with filaments that occur only off to his one side (clean air side) and not the other (Vickers & Baker, 1991).

The structure of pheromone plumes at different distances from the source has been measured using simulated pheromone mol-

ecules such as ionized air coupled with ion detectors (Murlis & Jones, 1981; Murlis, 1986; Murlis *et al.*, 1990), as well as with pheromone itself by using antennae as detectors of the filaments in the plume (Baker & Haynes, 1989; Willis *et al.*, 1991). The general trends are that as the distance from the source increases, the frequency of filament contact by a detector exposed to the finely structured plume does not decrease (excluding periods when large-scale wind-swings bring large pockets of clean air over the detector) (Murlis & Jones, 1981; Murlis, 1986; Baker & Haynes, 1989), but the duration of each contact increases, probably due to the increased diameter of each filament at greater distances caused by turbulent spreading (Murlis & Jones, 1981; Murlis, 1986). Higher windspeeds do not necessarily increase the frequency of antennal contact with pheromone filaments (Baker & Haynes, 1989; Baker & Vickers, 1994), unless the higher airspeeds that occur in these conditions arise predominantly from the males' own groundspeed, given a certain filament generation rate (Baker & Vickers, 1994).

From what is known about the fine structure of pheromone plumes and the dependency of the males' behavioural responses on their response to individual filaments, we can assume that in the current study, the straighter and more directly upwind tracks that were observed in some males, and especially the trend for such tracks to occur more frequently at greater distances from the source, will be due to a higher probability of contact with filaments. Such a higher frequency of contact is to be expected at greater distances, as shown by measurements of plume structure, which indicate that the filaments are broader and of longer duration at these distances due to turbulent spreading (Murlis *et al.*, 1982). Although the overall average concentration of filaments is diminished at greater distances from the source, some relatively undiluted and highly concentrated filaments still occur out there, although less often than they do closer to the source (Murlis & Jones, 1981; Murlis, 1986; Murlis *et al.*, 1990). Thus, the higher airspeeds, reduced course angles and less frequent counterturning observed farther from the source may be due to the lower average concentrations in these filaments, the higher peak-to-mean concentrations, or longer duration encounters with each filament. However, there is thus far no behavioural data measuring males' responses to single filaments that have been experimentally altered in these ways, so we do not know which of these factors might be responsible. We know for *H. virescens* that the frequency of contact with filaments significantly determines the shape of the track due to elicitation of reiterative surges (Vickers & Baker, 1994, 1996a), and that the quality of the filaments also determines the shape of the surge and the overall track (Vickers & Baker, 1997). In another species, *Cadra cautella*, the frequency of contact with filaments also is known to significantly affect track shape due to elicitation of surges from single filaments (Mafrá-Neto & Cardé, 1994).

Our moths were able to stay in contact with the pheromone plume filaments such that only one or two successive casts were observed. Pheromone was quickly recontacted usually after one straight, long leg oriented more crosswind, and the moth then continued his more directly-upwind progress. Interestingly, five out of six moths that casted during a wind-swing appeared to direct their initial, more crosswind, leg toward the 'correct' direction, i.e. toward where the wind had displaced the pheromone. Laboratory studies with *H. virescens* have indicated that there is

a time-lag in the male's response to a change in image flow of c. 0.3 s, a change of flow that would of course occur if there was a sudden change in wind direction. Likewise there is a lag of c. 0.3 s in male's responses (by casting) to the loss of pheromone, which would occur when males flew into the large pocket of clean air in the windswing area (Baker & Vickers, 1994). From these studies a prediction was made (Baker & Vickers, 1994) that the interaction of these visual and chemical lag times should result in a male entering a pocket of clean air in the windswing area and be, in effect, 'deflected' toward the side of the windline where the plume had swung away from the male (Baker & Vickers, 1994). It remains to be seen whether this is in fact what was occurring in the current field studies, but at least this is one possible explanation for the males' tendencies to move in the correct direction. In other field studies in which wind swings were observed, gypsy moth males also seemed to more often than not cast correctly toward the displaced plume (David *et al.*, 1983; David & Birch, 1989).

H. virescens males in the current study flew with much higher airspeeds than those observed in laboratory wind tunnels (cf. Vickers & Baker, 1994, 1996, 1997). The rest of the males' behaviour appeared similar to that observed in the laboratory, such as their course angles and counterturning frequencies. One obvious reason for the higher airspeeds would be the anemotactic response of the males to the 2 or 3 times higher windspeeds than they were exposed to in the field in the current study compared with in the laboratory. *H. virescens* males are powerful fliers, and they were able to elevate their airspeeds to maintain an average groundspeed centred around 200 cm s⁻¹. Although this groundspeed is still somewhat higher than what has usually been observed in laboratory studies, this may be due to the fact that in the field the males were flying 5 or 6 times higher above the ground pattern than they were in the laboratory, in which the plume was routinely placed 15 cm above the floor. Higher flight above the ground will produce a higher groundspeed in insects responding optomotor anemotactically to attain the same velocity of image movement (Kennedy, 1951), and this effect has been shown to occur in *H. virescens* males flying in a wind tunnel in response to pheromone (Kuenen & Baker, 1982). Higher airspeeds and groundspeeds in *Grapholita molesta* males flying in the field compared with the laboratory also have been observed (Baker & Haynes, 1996), and height of flight and windspeed were similarly thought to be the cause of these increased velocities. Higher airspeeds and groundspeeds also were observed in gypsy moth males flying in the field compared with their usual speeds in the laboratory (Willis & Cardé, 1990; Willis *et al.*, 1991); these males also were flying higher above the ground pattern than in the laboratory.

Overall, the data from these analyses of flight tracks of *H. virescens* males responding to pheromone in the field are consistent with the current models for pheromone-mediated upwind flight (Baker, 1990; Kaissling & Kramer, 1990; Willis & Arbas, 1991). Males were observed to fly both extremely straight upwind or with a zigzaggy track, and this was probably dependent upon the frequency of exposure to filaments that they were experiencing. They were able to shift direction when the plume shifted, and also to use crosswind casting to recontact pheromone that had apparently been lost. Loss of pheromone, as indicated by casting tracks, did occur even in wind that had not shifted, similar to behaviour

observed in laboratory wind tunnels. The entirety of the observed behaviours in the field is explicable by what is known about plume structure and the dependency of *H. virescens* male moth behaviour on the frequency of contact with filaments in such finely structured plumes and on the latencies of their responses to such contacts. These field analyses have provided us with more insight as to what these powerful moths are capable of. However, the same mechanisms that we know to be operating under the more constrained conditions in the wind tunnel can be satisfactorily invoked to explain the behaviours we observed of these males flying in the open field.

Acknowledgments

We thank R. S. Vetter for rearing the moths used in this study, and Drs H. Fadamiro and A. Corsé for helping with the statistical analyses and with Figs 4–6. Data collection was conducted while the authors were at the University of California, Riverside, and we thank this institution for the use of their facilities. Data analyses were conducted at Iowa State University. This work was initiated under U.S. Department of Agriculture Competitive Grant 87-CR-CR-1-2577 and complete with support from USDA National Research Initiative Competitive Grant 92-37302-7636, both awards to T.C.B.

References

- Baker, T.C. (1986) Pheromone-modulated movements of flying moths. *Mechanisms in Insect Olfaction (NSF-NATO Symposium)* (ed. by T. L. Payne, C. Kennedy and M. C. Birch), pp. 39–48. Clarendon Press, Oxford.
- Baker, T.C. (1990) Upwind flight and casting flight: complementary phasic and tonic systems used for location of sex pheromone sources by male moths. *Proceedings of the 10th International Symposium on Olfaction and Taste* (ed. by K. B. Døving), pp. 18–25. Graphic Communication System A/S, Oslo, Oslo.
- Baker T.C. & Haynes, K.F. (1989) Field and laboratory electroantennographic measurements of pheromone plume structure correlated with oriental fruit moth behaviour. *Physiological Entomology*, **14**, 1–12.
- Baker, T.C. & Haynes, K.F. (1996) Pheromone-mediated optomotor anemotaxis and altitude control exhibited by male oriental fruit moths in the field. *Physiological Entomology*, **21**, 20–32.
- Baker, T.C., Willis, M.A. & Phelan, P.L. (1984) Optomotor anemotaxis polarizes self-steered zigzagging in flying moths. *Physiological Entomology*, **9**, 365–376.
- Baker, T.C. & Vickers, N.J. (1994) Behavioral reaction times of male moths to pheromone filaments and visual stimuli: determinants of flight track shape and direction. *Olfaction and Taste XI* (ed. by K. Kurihara, N. Suzuki and H. Ogawa), pp. 838–841. Springer-Verlag, Tokyo.
- David, C.T. & Birch, M.C. (1989) Pheromones and insect behaviour. *Insect Pheromones in Plant Protection* (ed. by A. R. Jutsum and R. F. S. Gordon), pp. 17–35. John Wiley and Sons, New York.
- David, C.T., Kennedy, J.S. & Ludlow, A.R. (1983) Finding of a sex pheromone source by gypsy moths released in the field. *Nature*, **303**, 804–806.
- Kaissling, K.-E. & Kramer, E. (1990) Sensory basis of pheromone-mediated orientation in moths. *Verhandlungen des Deutschen Zoologischen Gesellschaft*, **83**, 109–131.

- Kennedy, J.S. (1940) The visual responses of flying mosquitoes. *Proceedings of the Zoological Society of London*, **109**, 221–242.
- Kennedy, J.S. (1951) The migration of the desert locust (*Schistocerca gregaria* Forsk.) *Philosophical Transactions of the Royal Society, B*, **235**, 163–290.
- Kennedy, J.S. (1983) Zigzagging and casting as a programmed response to wind-borne odour: a review. *Physiological Entomology*, **8**, 109–120.
- 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.
- Kuenen, L.P.S. & Baker T.C. (1982) Optomotor regulation of ground velocity in moths during flight to sex pheromone at different heights. *Physiological Entomology*, **7**, 193–202.
- Kuenen, L.P.S. & Baker, T.C. (1983) A non-anemotactic mechanism used in pheromone source location by flying moths. *Physiological Entomology*, **8**, 277–289.
- Mafrá-Neto, A. & Cardé, R.T. (1994) Fine-scale structure of pheromone plumes modulates upwind orientation of flying moths. *Nature*, **369**, 142–144.
- Murlis, J. (1986) The structure of odour plumes. *Mechanisms in Insect Olfaction* (ed. by T. L. Payne, M. C. Birch and C. E. J. Kennedy), pp. 27–38. Clarendon Press, Oxford.
- Murlis, J. & Bettany, B.W. (1977) The night-flight towards a sex pheromone source by male *Spodoptera littoralis* (Boisd.) (Lepidoptera, Noctuidae). *Nature*, **268**, 433–434.
- Murlis, J. & Jones, C.D. (1981) Fine-scale structure of odour plumes in relation to insect orientation to distant pheromone and other attractant sources. *Physiological Entomology*, **6**, 71–86.
- Murlis, J., Bettany, B.W., Kelley, J. & Martin, L. (1982) The analysis of flight paths of male Egyptian cotton leafworm moths, *Spodoptera littoralis*, to a sex pheromone source in the field. *Physiological Entomology*, **7**, 435–441.
- Murlis, J.M., Willis, M.A. & Cardé, R.T. (1990) Odour signals: patterns in time and space. *Proceedings of the 10th International Symposium on Olfaction and Taste* (ed. by K. B. Døving), pp. 6–17. Graphic Communication System A/S, Oslo.
- Shorey, H.H. & Hale, R.L. (1965) Mass-rearing of the larvae of nine noctuid species on an artificial medium. *Journal of Economic Entomology*, **58**, 55–68.
- Vetter, R.S. & Baker, T.C. (1983) Behavioral responses of male *Heliothis virescens* in a sustained-flight tunnel to combinations of seven compounds identified from female sex pheromone glands. *Journal of Chemical Ecology*, **9**, 747–759.
- Vickers, N.J. & Baker, T.C. (1991) The effects of unilateral antenectomy on the flight behaviour of male *Heliothis virescens* in a pheromone plume. *Physiological Entomology*, **16**, 497–506.
- Vickers, N.J. & Baker, T.C. (1992) Male *Heliothis virescens* sustain upwind flight in response to experimentally pulsed filaments of their sex-pheromone. *Journal of Insect Behavior*, **5**, 669–687.
- Vickers, N.J. & Baker, T.C. (1994) Reiterative responses to single strands of odor promote sustained upwind flight and odor source location by moths. *Proceedings of the National Academy of Sciences of the United States of America*, **91**, 5756–5760.
- Vickers, N.J. & Baker, T.C. (1996) Latencies of behavioural response to interception of filaments of sex pheromone and clean air influence flight track shape in *Heliothis virescens* (F.) males. *Journal of Comparative Physiology A*, **178**, 821–847.
- Vickers, N.J. & Baker, T.C. (1997) Chemical communication in heliothine moths. VII. Correlation between diminished responses to point source plumes and single filaments similarly trained with a behavioural antagonist. *Journal of Comparative Physiology A*, **180**, 523–536.
- Willis, M.A. & Arbas, E.A. (1991) Odor-modulated upwind flight of the sphinx moth, *Manduca sexta* L. *Journal of Comparative Physiology A*, **169**, 427–440.
- Willis, M.A. & Cardé, R.T. (1990) Pheromone-modulated optomotor response in male gypsy moths, *Lymantria dispar* L.: upwind flight in a pheromone plume in different wind velocities. *Journal of Comparative Physiology A*, **167**, 699–706.
- Willis, M.A., Murlis, J. & Cardé, R.T. (1991) Pheromone-mediated upwind flight of male gypsy moths, *Lymantria dispar*, in a forest. *Physiological Entomology*, **16**, 507–521.

Accepted 13 May 1997

1000

1000