

Optomotor anemotaxis polarizes self-steered zigzagging in flying moths

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ABSTRACT. Experiments with oriental fruit moth males, *Grapholita molesta* (Busck), provide evidence that a pheromone plume in zero wind elicits an endogenous, self-steered programme of counterturning (zigzagging) flight, and that wind experienced in flight establishes the polarity of the counterturns; they become aligned so that displacement occurs toward the source, even after the wind is stopped. In zero wind, males located a pheromone source more frequently when they had experienced a wind after having already taken flight before the wind was stopped (46%) compared with those that took flight later and therefore only experienced wind while they were in contact with the ground (14%). Furthermore, males placed in a stationary pheromone plume in zero wind located the source, eventually, on 21% of occasions. The flight tracks of these males, as well as those having experienced a wind only while on the ground, often exhibited repetitive counterturns (zigzags) of c. 180–200°. However, the counterturns meandered around the flight tunnel, the inter-reversal track angles having no consistent direction. Sometimes the males displaced down-tunnel in the stationary plume, sometimes up, eventually locating the source and performing a courtship display. The inter-reversal track angles of males counterturning in wind, on the other hand, displayed a consistent orientation of c. 60° to either side of the wind line, resulting in consistent upwind displacement toward the source. With no pheromone present, with or without wind, counterturns were not observed.

Key words. Pheromone, anemotaxis, zigzagging flight, orientation, oriental fruit moth, *Grapholita molesta*.

Introduction

Recent evidence suggests that two major mechanisms operate in flying moths to locate a distant pheromone source: a programme of

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counterturns is initiated whose frequency and amplitude are modulated by pheromone concentration (Kennedy *et al.*, 1980, 1981; Kennedy, 1982; Baker & Kuenen, 1982; Kuenen & Baker, 1982, 1983; Willis & Baker, 1984); and optomotor anemotaxis is displayed (Kennedy & Marsh, 1974; Marsh *et al.*, 1978; Kennedy, 1982, 1983). Substantial evidence for the coun-

terturning (Tobin, 1981; Bell & Tobin, 1982) programme can be found in experiments performed in wind (Kennedy *et al.*, 1980, 1981; Kuenen & Baker, 1982, 1983; Cardé & Haganman, 1979; Willis & Baker, 1984) or immediately after wind stoppage when the insects may have already established a movement pattern biased to some degree by the optomotor anemotaxis performed when wind was present (Baker & Kuenen, 1982; Kuenen & Baker, 1983).

In previous experiments involving stopping the wind (Farkas & Shorey, 1972; Baker & Kuenen, 1982; Kuenen & Baker, 1983) moths had been allowed to begin displacing toward a source while a wind was present. The high success rate in locating a source was assumed to be at least partly due to the optomotor anemotaxis performed by the moths before the wind stopped which biased their displacement toward only one direction, the source (Kennedy & Marsh, 1974; Marsh *et al.*, 1978; Baker & Kuenen, 1982; Kennedy, 1983; Kuenen & Baker, 1983). We report here the results of experiments designed to test whether optomotor anemotaxis induced before wind cessation does indeed influence the direction of movement.

Materials and Methods

Insects

Oriental fruit moths, *Grapholita molesta* (Busck)*, were reared on small green thinning apples. Pupae were separated according to sex and males allowed to emerge in isolation from females. Adult males were then separated daily according to age and maintained in an environmental chamber with positive air pressure to eliminate the possibility of exposure to pheromone before they were used in experiments. All males were 2–5 days old and had an 8% sucrose solution available to them at all times. During rearing, all life stages were maintained at approximately 25°C on a LD 16:8 h cycle.

Pheromone

A rubber septum (A. H. Thomas Co. No. 8753-D22, sleeve type, 5 × 9 mm) was impreg-

* Current correct genus name is *Grapholita*, not *Grapholitha* as previously used (Roelofs & Brown, 1982).

nated with the blend of three components used by *G. molesta* for sexual communication: 6% (*E*)-8-dodecenyl acetate and 4% (*Z*)-8-dodecenyl alcohol (Cardé *et al.*, 1979) in (*Z*)-8-dodecenyl acetate (Roelofs *et al.*, 1969). The amount of (*Z*)-8-dodecenyl acetate applied to the septum in its wide end was 30 µg. The formulation of this optimal ratio (Baker & Cardé, 1979a) was verified by gas-liquid chromatography (GLC) on a 10% XF-1150 (50% cyanoethyl, methylsilicone on 100–120 mesh Chromosorb W-AW-DMCS) 2 m × 2 mm i.d. glass column. Nitrogen gas carrier flow was 25 ml/min at 160°C. All three components, obtained from the stock solutions of Baker *et al.* (1981), had <0.5% volatile impurities as determined by GLC analysis on the XF-1150 column.

Wind tunnel

Males were flown in a large sustained-flight wind tunnel constructed after the design of Miller & Roelofs (1978) using clear polycarbonate plastic. The tunnel had a working section 3.6 m long, 1 m high at maximum, and 1 m wide at floor level. A ground pattern consisting of 10 cm diameter solid red circles randomly located (c. 20–30 cm apart) on a white cloth background was positioned 1.5 cm below the 6 mm thick clear acrylic plastic floor. Air flow was provided by a 1 hp voltage-regulated fan. Air turbulence was reduced by passing the air through a wooden 'mixing chamber' housing three particle-board frames over each of which was stretched a fine-mesh metal screen plus a layer of muslin. The frames were spaced 8 cm apart inside the chamber so that the air passed through each of the three layers of muslin and finally through a layer of fine-mesh polyester fabric as it entered the tunnel's working section.

The pheromone septum was positioned 40 cm from the upwind end of the working section in the centre of a 15 × 15 × 0.05 cm galvanized sheet-metal plate placed on a sheet-metal stand 15 cm above the centre of the tunnel floor. Pheromone was removed from the tunnel by a 30 cm diameter exhaust tube (air speed in centre = 2.9 m/s) aligned with the centre of the plume. The remainder of the air passing through the tunnel was recirculated through the room. Light intensity was 250 lux,

and was provided by four voltage-regulated 100 W incandescent bulbs whose light was diffused by reflection from a white expanded-polystyrene ceiling.

Stopping the wind

Flexible vinyl windowshades (rollerblinds), one at each end of the tunnel, were lowered manually to stop the wind. Both were supported on rollers having spring-loaded retraction mechanisms so that they could be easily repositioned for their next lowering. The windowshade at the upwind end was contained within the mixing chamber box and was lowered by means of a cord leaving the bottom of the box and travelling over a pulley to the operator at the upwind end of the tunnel. This shade was located at the downwind-most particle-board frame, and when lowered, the vinyl sheet blocked the windflow and was pressed against the frame, effectively sealing the wind from the tunnel along the edges. The top of the mixing chamber was open and hence when the windowshade blocked windflow into the tunnel, wind escaped into the room out the top of the chamber. Then the fan was switched off and the pressure against the shade allowed to subside slowly.

The windowshade at the downwind end of the tunnel was lowered simultaneously with the upwind one because initial smoke source visualization of the plume in 'zero wind' revealed that without this shade there was a very slight but consistent down-tunnel displacement of *c.* 0.5 m of the down-tunnel end of the plume caused by air movement from outside the tunnel from the room's ventilation and air-conditioning systems. The lowered windowshade, although not perfectly sealing the down-tunnel end, covered the semi-circular Plexiglas firmly enough that no down-tunnel displacement or other movement occurred.

When both shades were abruptly lowered, wind (plume movement) stopped virtually instantaneously. Although some of the plume filaments showed signs of slowly rolling and curling, there was no consistent directionality to this movement. Instead, the filaments slowly expanded and dispersed in all directions either while stationary or slowly curling until after *c.* 60 s they then reached the walls and ceiling of the tunnel. The maximum down-

tunnel velocity of the down-tunnel portion of the plume measured during a series of test-wind-stoppages was 0.5 cm/s. The up-tunnel two-thirds of the plume never exhibited this much displacement and the down-tunnel portion usually exhibited less.

Data recording and analysis

Flights were recorded from above in plan view using a rotary-shutter camera (Sony RSC 1050) positioned on top of the tunnel, connected to a Sony SLO 340 video recorder. In initial recordings only one camera was used, but the majority of recordings were performed using two such cameras and recorders. Due to the large amount of time required for complete analysis of a single track, the flights of only a small fraction of moths were recorded and analysed. The cameras were positioned so that there was *c.* 30 cm overlap in their fields of view, the dimensions of which for each camera at plume level were 110 cm lengthwise, and 81 cm across the tunnel. The lenses were covered by a red filter so that in these black-and-white recordings the red circles on the floor pattern became virtually white against the white cloth background to facilitate viewing the moths during playback (David, 1982). The recordings from the two cameras were synchronized by means of a light-emitting diode in view of both cameras, which flashed to indicate the moment of wind stoppage.

For analysis, individual flight tracks were re-recorded onto a Sony SVM-1010 motion analyser. Frame-by-frame playback from this system allowed the moths' consecutive 1/60 s positions to be marked by an ink dot on a Mylar® (clear plastic) sheet taped to the viewing screen of the analyser.

An X/Y digitizer (Houston Instruments, HIPAD DT 11) serially interfaced with a microcomputer (Radio Shack TRS 80 Model III) was used for analysing the Mylar® sheet tracings. The digitizer's resolution was set at 39 μm with an accuracy of 117 μm . Each tracing was consistently orientated on the digitizer surface using reference marks, and the X,Y coordinates of consecutive points were entered into the computer.

All measurements of velocity were made relative to the stationary tunnel. A computer program analysed the tracks for: (1) overall

velocity (velocity along the actual sinuous track), (2) net velocity (distance along the wind line per unit time), (3) turning frequency (number of turns/s), (4) distance per turn (cm along the actual track), (5) turning magnitude (degrees/turn), (6) angular velocity (degrees/second), and (7) the angular-to-linear velocity ratio (degrees/cm).

A turn was defined as beginning at any point along the track where the direction of movement changed from clockwise to anti-clockwise (or vice-versa), and to end at the start of the next turn. To allow for the angular error inherent in transcribing tracks and entering the X,Y coordinates, a threshold value of 50° change of direction had to be accumulated and exceeded before a new turn was registered, and the turn was deemed to have started (or ended) at the first point on the track where the change in direction had begun. The beginning and end of each tracing were therefore parts of incomplete turns and were not included in the turning data.

Inter-reversal track angles (Marsh *et al.*, 1978) were measured by hand. The upwind line, or in the case of moths flying in zero wind, the former upwind line, was designated as 0°, and angles to the right varied from 0° to 180°, and to the left from 180° to 359°. For moths flying in wind the relatively straight portion of the track (inter-reversal track) between reversals was more often straight than for moths flying in zero wind. Therefore in both cases a straight line was drawn averaging the direction of the relatively straight portion, and hence for moths in zero wind the slightly

more sinuous 'straight' portion fit this line less precisely (Fig. 3).

Procedure

Males were exposed to several regimes of wind to assess their ability to locate the source. In all cases when wind was present, its velocity was 28 cm/s. The major types of wind regimes (Table 1) were: (i) continuous wind plus pheromone; (ii) zero wind plus pheromone; (iii) zero wind plus no pheromone; and (iv) continuous wind plus no pheromone. Within category (ii) the moths were exposed to wind in several different ways before it was stopped (Table 1). All regimes were tested in random order on the same day during the period of maximum responsiveness to pheromone (Baker & Cardé, 1979b) which begins 1–2 h before lights-off, and extends as much as 1 h into what normally would be the scotophase. For conditions (i) and (ii), the pheromone septum was present in the centre of the platform throughout each male's entire flight, and for (iii) and (iv), no septum was present, only a clean platform.

Males were transported individually in 8 ml vials from their holding cage and transferred to 6.5 cm high cone-shaped narrow-mesh screen release cages, 8 cm diameter at the wide end of the cone. A single male was placed (point of the cone down) in an 8.4 cm diameter ringstand located 20 cm above the floor, 170 cm down-tunnel from the pheromone septum.

For situation (iii), the shades at both ends of the tunnel were pulled down and the fan was

TABLE 1. Procedures for creating experimental wind and pheromone combinations

Procedure	Sequence of actions*	Resulting experimental condition
i	A, C, F, H	Males exposed to wind continuously, take flight in pheromone plume
ii	A, C, H, E	Males briefly exposed to wind after taking flight in pheromone plume, but then fly in zero wind
iib	A, C, G, E, H	Males exposed to wind while on ground in pheromone plume, but take flight in zero wind in plume
iic	A, D, E, C, H	Males exposed to wind while on ground in clean air, take flight in zero wind in pheromone plume
iid	A, E, C, H	Males not exposed to wind, take flight in stationary plume
iii	B, E, D, H	Males not exposed to wind, take flight in clean stationary air
iv	B, F, D, H	Males exposed to wind continuously, take flight in clean wind

* (A) Place pheromone septum in tunnel; (B) remove pheromone septum from tunnel; (C) place cone containing male in ringstand in pheromone plume; (D) place cone containing male in ringstand in clean air; (E) stop wind; (F) allow wind to continue; (G) allow male c. 5 s of exposure to wind while in cone; (H) allow male to take flight.

left off (zero wind) at all times. For (i) and (iv), the wind was left on throughout all flights. The sequences of procedures for introducing males into the tunnel are as listed in Table 1.

Regime (ii) was achieved using four different procedures (Table 1), all resulting in a male flying in zero wind in a stationary pheromone plume in the centre of the tunnel. In all four cases the wind was stopped by pulling down the windowshades at each end of the tunnel. The operator of the up-tunnel shade signalled the moment of wind stoppage (shade down) by flashing the light-emitting diode (LED) in the camera's field of view. Then he quickly turned off the fan. Another operator lowered the downwind shade simultaneously with the upwind one on verbal cue.

All four of these procedures (iia-d, Table 1), were practiced first with a smoke source to visualize the plume, and these sessions confirmed that when they were performed correctly they did not distort the stationary plume. The practice sessions using smoke did point out the importance of completely closing the side door, leaving no crack, before the wind was stopped. The pressure changes upon shade-lowering would force air into the tunnel through such a crack, and from the side, push the centre section of the plume over towards the far wall very quickly. Males in procedure (iia) were allowed to take flight before the wind was stopped. This resulted in some having already begun zigzagging upwind at the moment wind was stopped, while others were still keeping station while 'locking on' to the

plume. Still others had scarcely taken flight when the wind was stopped. Included in this last group were some males from procedure (iib) which had been judged upon viewing the recordings to have taken flight before the wind stopped. All males in (iia) thus had had at least some exposure to wind while flying.

Results

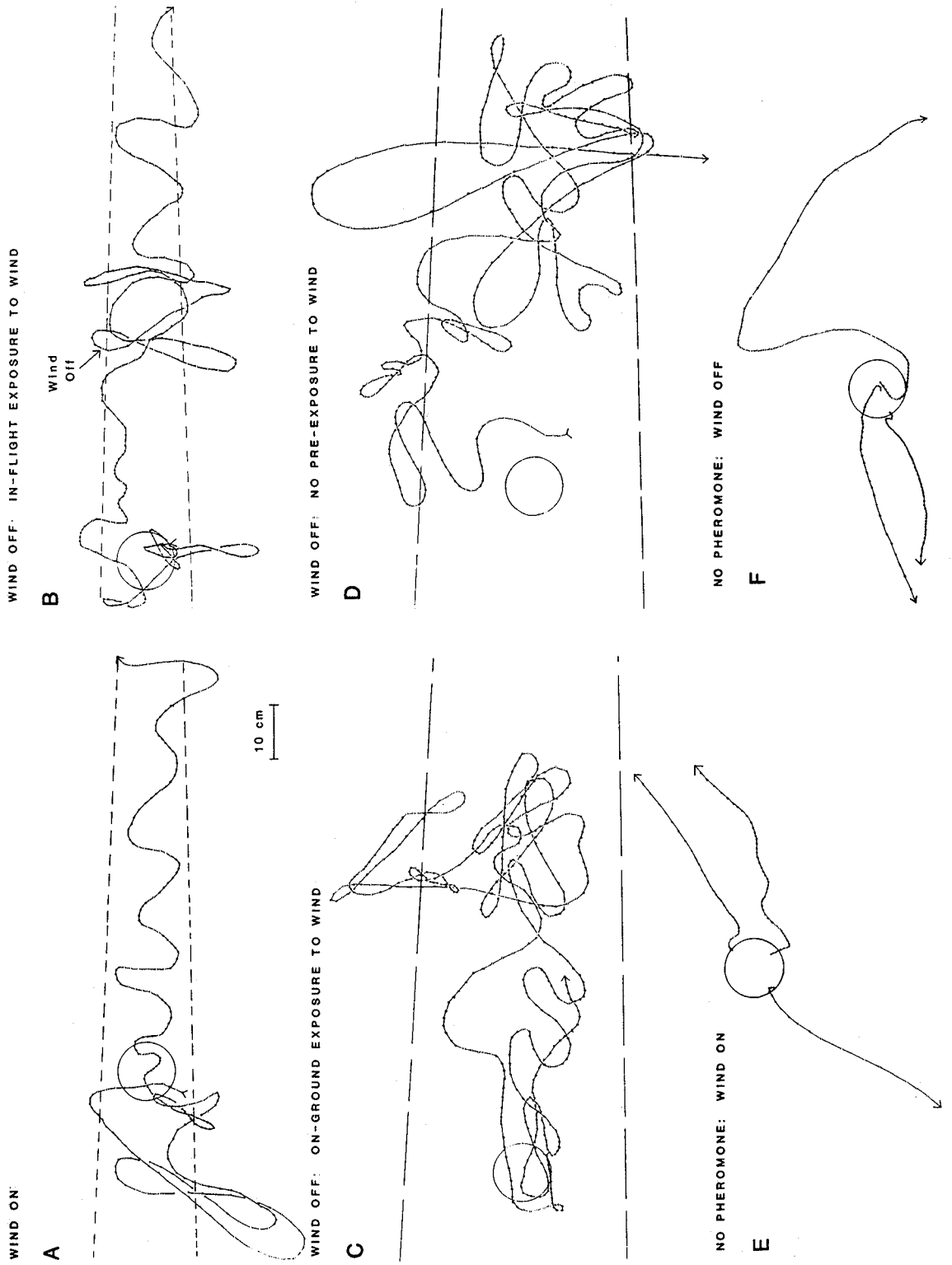
When the wind was stopped, only those males already in flight could locate the pheromone source as frequently as males flying along a pheromone plume in continuous wind (Table 2). Males experiencing wind while still in contact with the ground, regardless of whether the wind did or did not contain pheromone, were not able to locate the source in zero wind any more frequently than males having had no exposure to wind whatsoever. Males having no exposure to wind were placed into the stationary plume in still air, but surprisingly were able to locate the source, eventually, in a significant percentage of cases (Table 2). When no pheromone was present with wind either on or off, males never located the platform and never flew closer than 1 m from it. They usually flew quickly and directly to the ceiling or side walls, rarely with any up-tunnel displacement (Fig. 1).

The tracks of males taking flight in pheromone after wind had already stopped usually exhibited regular zigzags similar to those of males flying in pheromone in wind (Fig. 1). That these zigzags were in fact consistently

TABLE 2. Effect of in-flight exposure to wind upon subsequent *G. molesta* success at locating pheromone source in zero wind.*

	Of males taking flight, percentage locating source*
(i) Continuous wind + pheromone	49% a (68/140)
(ii) Zero wind + pheromone	
(a) In-flight exposure to wind + pheromone	46% a (17/37)
(b) On-ground exposure to wind + pheromone	22% b (28/128)
(c) On-ground exposure to wind	14% b (12/88)
(d) No exposure to wind	21% b (22/106)
(iii) Zero wind + zero pheromone	0% c (0/36)
(iv) Continuous wind + zero pheromone	0% c (0/26)

* Percentages are of males that took flight within 1 min of being placed in position in the centre of the pheromone plume (or tunnel if no plume present); those remaining in release cage are not included here. Males were given 1 min to locate the source after taking flight. Percentages having no letters in common are significantly different according to χ^2 2 \times 2 test of independence ($P < 0.05$).



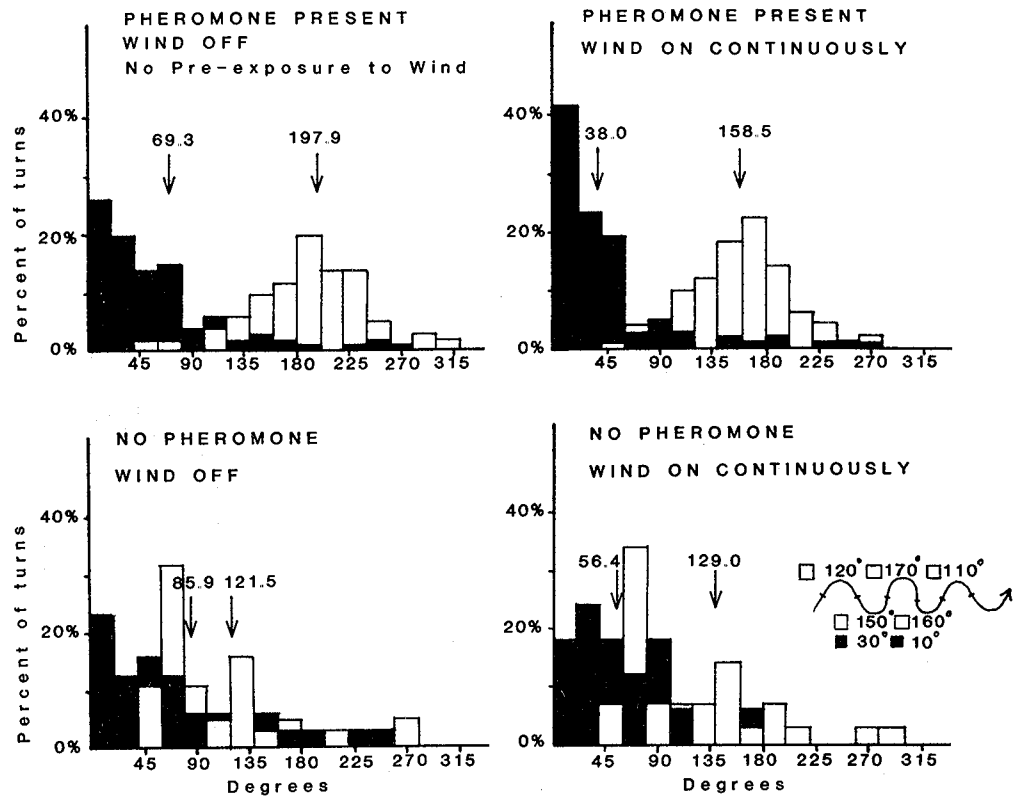


FIG. 2. Distributions of turn magnitudes (\square) and differences between consecutive turns (\blacksquare), demonstrating that counterturns are performed by males with pheromone present, wind on or off. Consecutive turns are very similar in magnitude with pheromone present, despite the fact that their magnitudes are greater than with pheromone absent. Numbers above arrows are the means of the distributions. Bars are at 20° intervals. In contrast to data in Table 3, turn magnitudes greater than 360° were not included here because of the difficulty in wind of knowing whether such large turns are due to a course reversal or not. The following is a listing of the above means \pm SD and n . 69.3 ± 80.2 , $n=184$ (15 males); 197.9 ± 58.5 , $n=194$ (15 males); 38.0 ± 38.6 , $n=173$ (14 males); 158.5 ± 48.1 , $n=204$ (14 males); 85.9 ± 85.3 , $n=30$ (15 males); 121.5 ± 73.8 , $n=39$ (15 males); 56.4 ± 36.7 , $n=16$ (12 males); 129.0 ± 78.3 , $n=29$ (12 males). With no pheromone present, most of the turn magnitudes were at 40 – 80° because the threshold was set at 50° in our computer program. Thus, due to the straight tracks of these moths, turns of the minimal magnitudes were registered, hence the large bars near the threshold value but not below it.

FIG. 1. Tracks of male *G. molesta*, flying in the wind tunnel, recorded from above in plan view. Wind, when present, is from the right. Dots along track indicate males' positions every $1/60$ s. Large open circles represent the screen release cones in which males were placed in position in the tunnel. Dashed straight lines in A and B indicate time-averaged pheromone plume's position while wind was on, and those in C and D when wind had been stopped for 5 s. In A, C and D, portions of the complete track have been omitted for the sake of clarity. (A) Wind was on continuously, male located the source. (B) Wind stopped at point indicated by arrow, after male had experienced wind while flying; male located the source. (C) Male flying in zero wind. Male experienced wind while in the release cone for c. 5 s, then wind was stopped before take-off. Note the many counterturns of c. 180° , and their lack of consistent orientation with respect to the source, which the male eventually located. (D) Male flying in zero wind. Male was placed into the stationary plume with wind off, and had therefore never experienced wind. Again note the regular counterturns and their lack of consistent direction with respect to the source, which was not located. (E), (F) Tracks of six males taking flight with no pheromone present, wind on continuously and wind off continuously, respectively. Note relatively straight tracks with few regular turns. Males flew fairly directly up toward the ceiling and landed, never coming near the source.

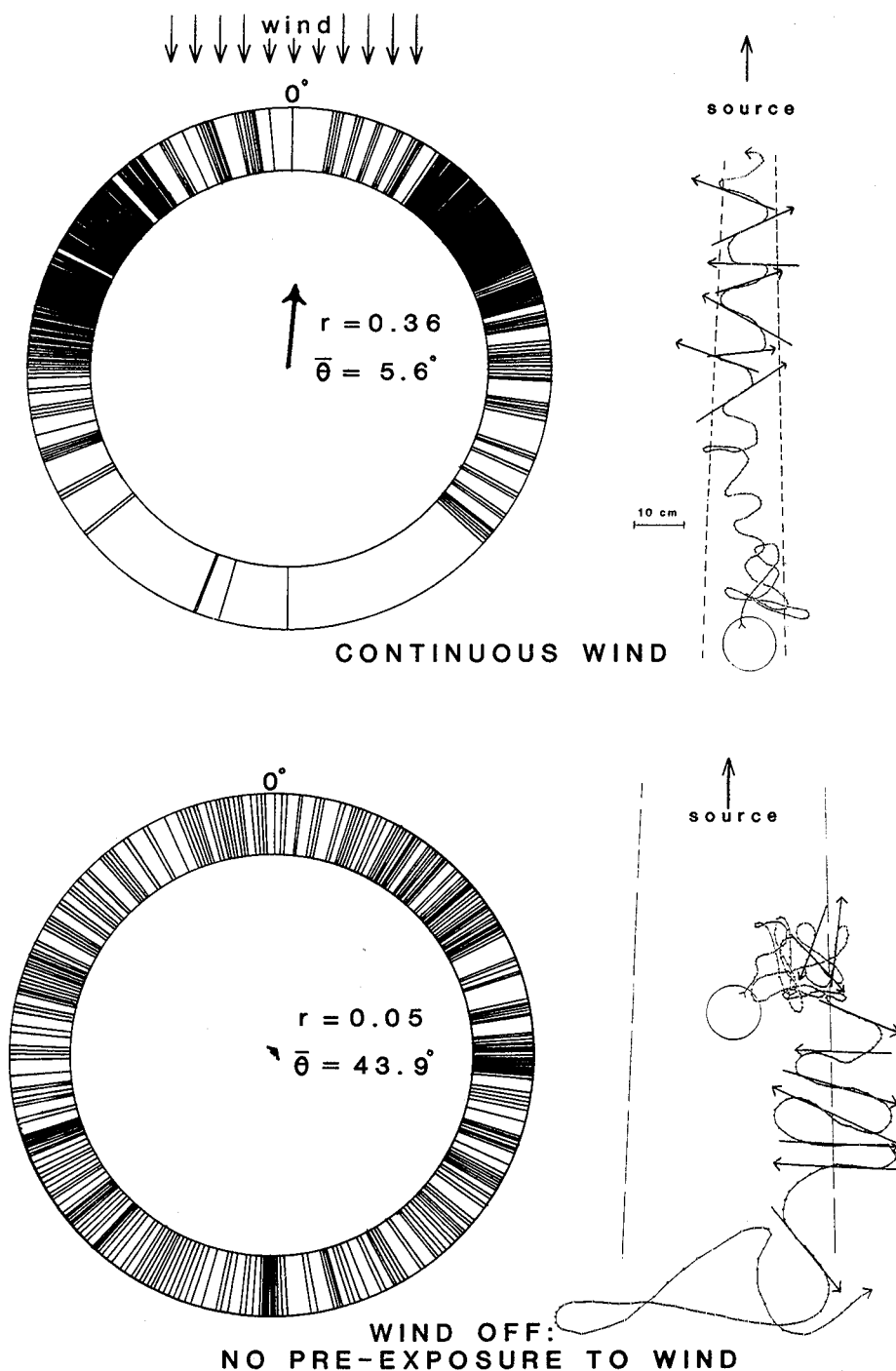


FIG. 3. Distribution of inter-reversal track angles for flying males counterturning in wind (top) and after taking off in zero wind (bottom). Sample tracks of moths from both conditions are at right, and superimposed on them are arrows showing how some of the inter-reversal track angles were measured (for clarity, only some of the arrows are drawn). $n=279$ (14 males) for continuous wind, and $n=333$ (15 males) for wind off.

executed counterturns is illustrated by plotting the turn magnitudes and the differences of the magnitudes of consecutively executed turns (Fig. 2). With pheromone present, wind on or off, it is clear that differences between consecutive turns were small, on average. The consistency of these repetitive turns, with or without the wind's extra polarizing influence is even more important when considering their high mean magnitude, 159° and 198° , respectively (Fig. 2). Contrast these values to those of turns executed with no pheromone present. The mean differences between consecutively executed turns were higher when pheromone was absent, wind on or off, and this is more meaningful considering that the turns executed in clean air were of smaller magnitude. Thus despite performing shallower turns in clean air than in pheromone, they were of less consistent magnitude.

Unlike the counterturns in wind with pheromone present, those performed with pheromone in zero wind meandered about the tunnel, sometimes resulting in displacement down-tunnel in the plume, sometimes up-tunnel, and sometimes toward the sides of the tunnel (Fig. 1). Those males zigzagging in the generally up-tunnel direction in zero wind sometimes came within *c.* 30 cm of the source (out of view of the camera) and when they did, the zigzags seemed to narrow and become more frequent before the males landed and displayed their hairpencils at the septum.

The 'meandering' of the zigzags of males in zero wind having no in-flight wind experience is illustrated by the directions of the inter-reversal track angles (Fig. 3) of males placed in the stationary plume in zero wind. There was no consistent directionality to these angles compared with those of males flying in wind, which exhibited a bi-modality of *c.* 60° to either side of the upwind line that contributed to their up-tunnel displacement. Although such bi-modality prevents the mean vector from being tested for a significant single 'preferred' direction (Batschelet, 1972), it is clear that the relative length and direction of this mean inter-reversal angle vector reflects a pronounced displacement up-tunnel along the windline (Fig. 3). It should be pointed out that the inter-reversal angles of males in zero wind are the angles of both their track and course (Marsh *et al.*, 1978), whereas in wind these are only the inter-reversal track angles.

The tracks of males already in flight toward the source when the wind was stopped were very similar to those of males flying along a plume in continuous wind. Occasionally, though, the zigzags of some of these males began to meander away from the generally up-tunnel direction and the moths sometimes then failed to regain their up-tunnel displacement and locate the source (Fig. 4).

Other measurements of the moths' movements in pheromone were quite similar across all treatments in zero wind but often different

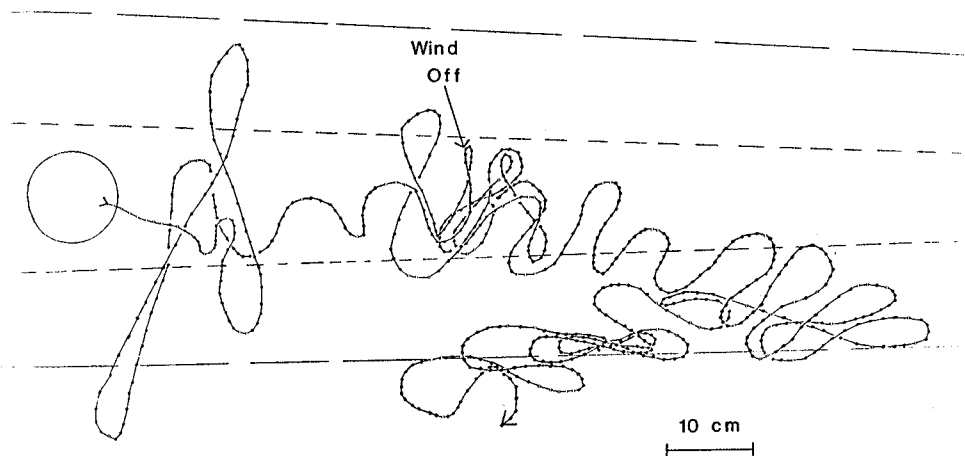


FIG. 4. Track of male that began flight in wind, showing how counterturns continue when wind velocity drops to zero, and how they sometimes lose their up-tunnel polarity after a while. This male eventually located the source (complete track not shown for sake of clarity)

TABLE 3 Movement parameters (mean \pm SD) of *G. molesta* males in pheromone in continuous wind or in zero wind after receiving different types of pre-exposure to wind.

	Linear velocity (cm/s)	Angular velocity ($^{\circ}$ /s)	Turn frequency (turns/s)	Turn magnitude ($^{\circ}$ /turn)
(i) Continuous wind + pheromone	61.8 b \pm 9.2	673.8 a \pm 130.5	4.48 a \pm 1.26	179.8 a \pm 26.5
(ii) Zero wind + pheromone				
(a) In-flight exposure to wind + pheromone	74.3 a \pm 11.7	653.4 ab \pm 119.7	3.83 ab \pm 0.77	194.5 a \pm 33.3
(b) On-ground exposure to wind + pheromone	84.5 a \pm 10.9	579.9 b \pm 125.0	3.55 b \pm 0.74	197.4 a \pm 30.3
(d) No exposure to wind	80.4 a \pm 26.8	618.6 ab \pm 116.4	3.88 ab \pm 0.74	184.2 a \pm 21.8

Means in same column having no letters in common are significantly different according to Duncan's multiple range test ($P < 0.05$). $n=25$ males for (i), and 14 each for (iia), (iib) and (iid).

from those in continuous wind (Table 3). These included differences in overall linear velocity, angular velocity, turning frequency, and turning magnitudes. The major difference between the tracks of those able to locate the source in zero wind with a high rather than low success rate was again the more consistent polarization of the counterturns in one direction.

Discussion

The results of this study are consistent with recent hypotheses concerning orientation mechanisms employed by flying moths for pheromone source location (Kennedy, 1982, 1983; Kuenen & Baker, 1983). Recent evidence points to a pheromone-modulated programme of counterturning (Kennedy *et al.*, 1980, 1981; Baker & Kuenen, 1982; Kennedy, 1982, 1983) integrated with optomotor anemotaxis (Kennedy, 1982; Kuenen & Baker, 1983). Our results indicate that in *G. molesta*, counterturning (Tobin, 1981; Bell & Tobin, 1982) of *c.* 180° is performed in response to pheromone in zero wind, and moreover by males having had no experience whatsoever of wind. This is the first time such a programme has been reported, uncomplicated by wind, in flying moths. Similar results have also been obtained by J. S. Kennedy, C. T. David and A. R. Ludlow (personal communication) who found that male gypsy moths beginning flight in zero wind exhibit counterturning in pheromone but not in clean air.

For *G. molesta*, the counterturns differ from those in wind in that they meander around the tunnel, the interreversal angles having no consistent orientation as they have in wind. Thus although a source is located on a surprising 21% of occasions by males placed in a plume in zero wind, the males' displacement is usually very indirect.

The addition of wind produces counterturning which is polarized in the up-tunnel direction, presumably by means of optomotor anemotactic compensation for drift. This conclusion is supported by the fact that males having at least a brief flight in wind in pheromone are more likely to locate a source after the wind stops than those experiencing wind while still on the ground and then taking flight in zero wind. Thus, not just any exposure to wind can polarize the counterturns, but specifically it must be exposure while in flight and involve the optomotor response. As previously suggested (Kennedy & Marsh, 1974; Baker & Kuenen, 1982; Kuenen & Baker, 1983) displacement continues toward a source in zero wind in such moths because the drift compensation has already effectively channeled the movement in that direction. The counterturning continues along a plume in the established direction in zero wind unless the concentration drops, whereupon the frequency of the turns decreases and the inter-reversal angles change so that displacement toward the source is slowed or stopped (Baker & Kuenen, 1982; Kuenen & Baker, 1983).

Males experiencing wind while on the

ground behave like those never having experienced wind and placed directly into the centre of a stationary plume. They do locate the source a significant number of times, but they also meander around the tunnel as they counterturn. One reason why such males are able to locate a source appears to be that they control their height as they counterturn, and thus stay close to the ground near the level of the plume. Like the tethered gypsy moths described by Preiss & Kramer (1983), the lift, and hence changes in height by our moths, appeared to be controlled tightly while in pheromone, whether in wind or not. As in their gypsy moths, lift is less regulated when no pheromone is present, resulting in the moths quickly reaching the ceiling and then sitting, never having a chance to displace toward the platform area. Again, this occurs in wind or without it.

Whether the counterturns in zero wind are performed with or without the aid of visual feedback remains an open question. With continued pheromone contact, males should remain sensitive to drift and compensate quickly if drift is sensed. With wind present the drift is usually alternated accurately right-to-left with each inter-reversal leg of the counterturn (Fig. 3), but with increasing time after wind cessation the lack of drift seems to allow the counterturns to occasionally stray from the former wind line. In this study, longer segments of track were analysed than in previous reports (Baker & Kuenen, 1982; Kuenen & Baker, 1983), and it is clear that even when a male is already zigzagging toward a source before the wind ceases, sometimes its subsequent path in zero wind eventually wanders, even while still in contact with pheromone.

Other recent findings indicate that for some species counterturning continues in its narrow, high frequency form only if the male receives discontinuous pheromone stimulation (Kennedy *et al.*, 1980, 1981; Kennedy, 1982; Willis & Baker, 1984). Continuous pheromone stimulation results in a rapid return to the wide cross-wind casting characteristic of that in clean air, perhaps due to adaptation (Kennedy *et al.*, 1981; Kennedy, 1982).

In our experiments, a male flying in pheromone in zero wind should be receiving discontinuous stimulation as it counterturns because the plume (as visualized by smoke)

retains its filamentous structure for quite a long time after the wind ceases. With increasing time, though, the lateral and vertical spread and dispersion of the filaments could result in a more homogeneous cloud in the tunnel that causes more continuous exposure and a waning of the response.

Olberg (1983) recently identified from the ventral nerve cords of *Bombyx mori* males 'flip-flopping' interneurons whose firing rate was switched from a constant high to a constant low frequency, or vice-versa, with each onset of pheromone stimulation. If interneurons behaving similar to *B. mori*'s exist in *G. molesta*, tonic receptor output, not adaptation, might decrease counterturning frequency by reducing state-switching. In filamentous plumes that are too concentrated or unbalanced with too much of one pheromone component, receptors might be incapable of recovering rapidly enough between filament-induced bursts of activity to register the next filament's onset, resulting in effectively tonic output.

It is clear from this study and the other recent findings discussed above that pheromone-modulated, self-steered counterturning in flying moths is a component distinct from anemotaxis, but unless it is integrated with anemotaxis the efficiency of reaching the pheromone source is much reduced. In *Bombyx mori*, which has lost its ability to fly, movements in the visual field in addition to repeated onsets of pheromone are registered as state changes in other classes of flip-flopping interneurons that descend from the brain along with the pheromone-triggered types (Olberg, 1983). If these types of interneurons could be shown to exist in *G. molesta* (or in other species that fly) and proven to have similar responses to pheromone and visual field movements, a neuronal basis for the integration of the self-steered counterturning programme with optomotor anemotaxis would be at hand.

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