Physiological Entomology (1995) **20**, 100–108

Rates of progress up odour plumes by tsetse flies: a mark–release video study of the timing of odour source location by *Glossina pallidipes*

NIGEL GRIFFITHS, QUENTIN PAYNTER and JOHN BRADY
Department of Biology, Imperial College of Science, Technology and Medicine, London

Abstract. The arrival of individually marked *Glossina pallidipes* Austen at a host odour source after their video-timed release from 30–75 m downwind was measured in the field in Zimbabwe. In the absence of odour, the proportion recaptured was <2% (= ≃ random expectation); when synthetic ox odour was released, the probability of recapture at the source increased with proximity of release, from 6% at 75 m to 21% at 30 m (about twice this number arrived within ~2 m of the source). There were two distinct distributions of recaptures: a 'fast' cohort which found the source within 40 s, and a 'slow' cohort which took from one to >20 min, with ~50% of the flies in each cohort. The fastest flies probably reached the source in a single, mainly straight flight from take-off, at an overall average (straight line) displacement speed of 2.8–4.5 m s^{-1} (i.e. close to the preferred flight speed of ~5 m s^{-1}). The flies apparently maintained their ground speed largely independent of the wind speed they headed into. The 'slow' cohort had a constant probability of arrival at the source, presumably after losing and re-contacting the plume, and after having stopped at least once on the way. There were no marked correlations with wind parameters, although the probability of recapture increased slightly with the directness of the wind from the source, and the probability of 'slow' flight increased slightly with wind speed. It is inferred that a repeated sequence of anemotactic 'aim-then-shoot' orientation at take-off plus optomotor-steered in-flight correction of direction is used as a form of biassed random walk to bring the flies close to the odour source, rather than the use of moth-type anemotactic zigzagging.

Key words. *Glossina*, tsetse fly, flight speed, anemotaxis, odour plume, orientation, host-finding, behaviour, video analysis.

Introduction

Having once contacted a plume of host odour, tsetse flies apparently find its source with a high probability during that afternoon's activity peak (Vale, 1980). This high success rate evidently depends on the flies' use of a combination of orientational responses to odour and wind (see Colvin & Gibson, 1992). These include: mechanical upwind anemotaxis at take-off (Bursell, 1987; Torr, 1988a); chemokineses in relation to odour concentration (Warnes, 1990; Gibson *et al.*, 1991; Paynter & Brady, 1993); optomotor-guided turning on entering and leaving host odour (Gibson & Brady, 1985, 1988; Torr, 1988b; Colvin *et al.*, 1989; Paynter & Brady, 1993); in-flight upwind 'anemotaxis' while navigating within an odour plume (Vale, 1974b, 1984; Torr, 1988a, b; Gibson *et al.*, 1991; Brady & Griffiths, 1993); and a final visual homing-in on some obvious, contrasty object (Torr, 1989; Bursell, 1990; Gibson, 1992; Green, 1993).

Correspondence: Dr J. Brady, Imperial College, Silwood Park, Ascot, Berks. SL5 7PY.

It seems that huge plumes of host odour may lure flies from hundreds of metres away downwind (Hargrove & Vale, 1978; Hargrove *et al.*, 1995), but the plume from a single ox may be navigable to its source from only about 90 m (Vale, 1977). What is quite unknown is how long it takes the individual fly to navigate up any kind of odour plume to the source – although Bursell (1988) has suggested net upwind progress rates of around 5 m min^{-1}, inferred from the changing rates of catch at an electric net after an odour source was switched on.

The only direct measurement of flying insects' progress towards distant sources of odour in nature has been the ingenious

experiment of Elkinton *et al.* (1987) on gypsy moths. They employed concentric rings of observers timing the departure of marked males from cages and the hand-netted recapture of successful males at the source. This revealed that the maximum distance from which the males could find a point pheromone source was about 80 m; beyond that the wind's meandering reduced the correlation between wind direction and source direction to unusably low levels (see Brady *et al.*, 1989, for discussion re tsetse flies). Willis *et al.* (1991) developed this idea, using video to record the males' flight tracks between 2 and 20 m downwind of the source, relating flight responses to wind patterns.

A similar approach with tsetse flies has proved impossible, though several attempts have been made (Brady *et al.*, unpubl.). The two chief problems are the confusing presence of human odour in the release procedure, and the flies' strong tendency to escape from a release cage the instant it is opened. Take-off thus occurs in an uncontrolled relationship with the wind and/or the arrival of odour. We report below the results of an attempt to reduce these problems by using video to record marked flies' spontaneous take-off and an electrified target to time their recapture at an odour source, which allowed the first direct measurement of tsetse flies' flight times up odour plumes.

Materials and Methods

Sites. The initial experiments were done at Nguruman, south-western Kenya, in 1990. The results encouraged the experiments reported below, which were done at Rekomitjie Research Station in the Zambezi valley, northern Zimbabwe. The final observations were all carried out in one site in typical 'mopani' woodland (described by Vale, 1974a) during the dry seasons of 1991 and 1992 (July–October) when the trees and understorey were largely leafless and the herbage heavily grazed. The wind at this time of year blows consistently from the east at Rekomitjie, so that the release and recapture sites could be aligned along its mean axis.

Flies. Live male and female *Glossina pallidipes* Austen were collected from odour-baited 'epsilon' traps, deployed at ~14.00 hours each experimental day (for trap design, see Fig. 1 of Hargrove & Langley, 1990). The first flies were collected after about 1 h. Each fly was then colour-marked with oil-based paints to indicate day and fly number, and was placed in a compartment of a 'release battery'. No significant behavioural differences were detected between males and females, which are therefore pooled in the results described.

Release of marked flies. The release site was set at 30, 50 or 75 m downwind from the recapture site. The release battery consisted of sixty-three numbered cardboard tubes in a horizontally-aligned, honeycomb array of 7 × 9. Each tube was 4–5 cm in diameter by 15–25 cm long, and was closed at one end with a mosquito-netting flap through which the fly could be inserted; the other end was completely open, but was sealed until the moment of release by a door (of cardboard and foam rubber) that closed all the tubes together. When the door was opened, all sixty-three tubes were simultaneously unsealed at their upwind end. A video camera with a telephoto lens (Cohu/Fujinon) was set up so that the array of tube ends filled its field of view. It recorded with a time-base so that the exact time of departure of each marked fly could be identified on playback and correlated with its recapture time.

Recapture of marked flies. The odour source was at the foot of a vertical, 1-m-square electric net set across the mean wind line (see Griffiths & Brady, 1994). The net sat over a two-sided metal shute that delivered the stunned flies separately from the upwind and downwind sides of the net into an underground chamber in which two observers sat (an 'observation pit' *sensu* Hall & Langley, 1989). One observer maintained an audio-tape commentary on the arrival of flies, synchronized with the time-base of the release site video camera; the other called out the colour code of marked flies as they arrived and the side of the net which a fly hit, putting marked flies into numbered specimen tubes for later verification and treatment.

A 'model warthog' (a black cylinder 50 cm long by 37 cm diameter; see Vale, 1974b, Fig. 1.4) was placed ~20 cm upwind of the electric net to provide a visual focus for the arriving flies. The odour source released CO_2 at ~20 l min^{-1}, and acetone, 1-octen-3-ol, 3-*n*-propylphenol, and 4-methylphenol at ~500, 0.4, 0.1 and 0.8 mg h^{-1}, respectively. This blend provides a reasonable surrogate for ox odour (Torr, 1990; Vale, 1991), with CO_2 released in generous quantities to maximize fly attraction rates.

Control experiments with no odour released were done for the 30 m and 50 m distances, to determine the number of flies which moved upwind in the absence of odour. To estimate random dispersal rates from the release battery, an unbaited 'warthog' and electric screen were placed due downwind of the release point, at the same distance as the recapture site.

Experimental protocol. The odour was released continuously from at least 5 min prior to the release of flies until the end of a trial. A trial commenced when an observer, who stood 10 m downwind and to the side of the release battery, pulled a cord which opened the battery door, thereby initiating airflow through the tubes. All departures (and arrivals at the source) were recorded for the next 22 min (the length of a video cassette). Longer recording would have been pointless, because the flies would have had time to travel hundreds of metres in any direction before recapture.

Wind speed and direction were recorded at the recapture site for the duration of each experiment (though equipment failure prevented wind data collection for the 30-m experiment). The monitor was a Solent 'Standard' ultrasonic anemometer sampling at 1-s intervals. Daily maximum temperatures were taken from a meteorological station 500 m away.

Supplementary control. An additional experiment was done at 50 m with four 1.5 × 1.5 m electric nets placed in an incomplete ring (*sensu* Vale & Hargrove, 1979) around the experimental recapture net and ~2 m from it. Recaptured flies were collected on sticky trays on either side of the nets. This control tested whether significant numbers of marked flies arrived in the clearing around the observation pit but did not get caught by its net. The same protocol was used with and without odour (ten trials each).

Statistics. Recapture rates, flight times and wind data were analysed either by standard regression statistics or, where appropriate, using generalized linear modelling (GLIM; Crawley, 1993).

Results

Take-off behaviour

On average, 92% of the flies took off by the end of a 22-min trial (the total number used ranged from 33 to 63, depending on availability). Take-off distributions were analysed (Fig. 1) for the 50-m experiment only, there being no reason to suppose that it would not be typical of all three distances. There was no significant difference between the departure rates with and without odour.

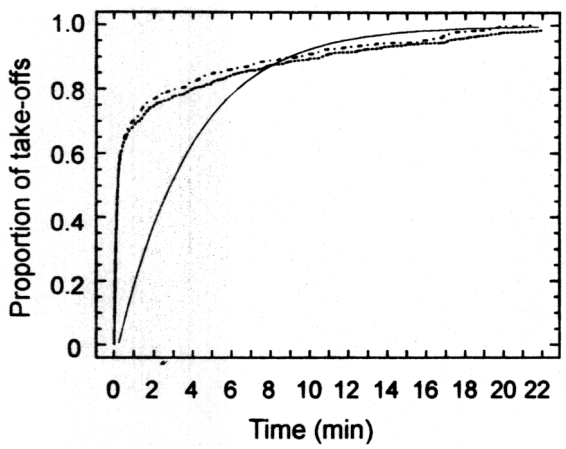

Fig. 1. Cumulative distributions of the rates of departure of flies from the release battery for the 50-m experiment, when odour was released from the recapture site (broken line, n = 710 flies) and when it was not (dotted line, n = 646). There is no significant difference between the two distributions (Kolmogorov-Smirnov DN 0.065, P = 0.11); but both distributions differ from the best fit of a single exponential function (solid line, DN 0.475, P <0.001).

There was a high initial rate of departure, mainly in the first minute, followed by a more constant rate over the next 20 min. This distribution differed highly significantly from the best-fit single exponential (Fig. 1), so take-offs did not occur constantly through time. Presumably the rapid initial take-offs were mainly responses to the opening of the door; the other flies often walked to near the open end of their tube before flying. Even when take-offs within the first minute are excluded, there is no difference between the rates of take-off with and without odour ($P > 0.5$).

Take-off directions were not obviously biassed by the fact that the release battery pointed upwind. Direct observation (from overhead) of the direction of departures of flies indicated that roughly as many doubled back behind the battery, or flew off in other directions, as flew away from it in the direction the tubes were pointing. It might have been interesting to be able to relate individual flies' departure directions to their arrival at the odour source, but this was not feasible technically.

Arrival upwind

There was no detectable difference between the initial, fast departers and the slow departers in the rate at which they found the odour source: time of departure correlated neither with the flies' probability of recapture (t = 0.75, df = 386), nor with the recaptured flies' total flight time to the odour source (r^2 = 0.2%). In odour, the proportions of marked flies that were recaptured decreased significantly with the distance from the release point (Table 1). The vast majority (89±3%) of recaptured flies arrived at the source in upwind flight (i.e. they were caught on the downwind-facing side of the electric net).

Table 1. The mean proportions (% ±SE) and mean numbers of flies recaught each trial at the three release distances, when odour was released at the recapture site and when it was not. For the negative relationship between recapture rate and release distance r^2 = 32% (P <0.01).

Release distance	Odour present		Odour absent	
	Recaught (%)	Mean catch	Recaught (%)	Mean catch
30 m	21.1± 1.9	11.6		1.0
50 m	9.6± 0.8	5.6		0.3
50 m + net ring	18.5± 1.7	10.0		0.4
75 m	6.4± 1.0	4.0		

With odour present n = 8, 24, 10 and 10 trials, respectively; with odour absent n = 5, 12 and 10 trials, respectively.

Without odour. When no odour was released from the recapture site, the numbers recaught were very low – less than 2% of the released flies (Table 1). Even at 30 m, at best only two flies per trial were recaught out of the sixty-odd leaving the release battery. This is slightly less (though not significantly so) than would be expected from random radial dispersal from the release site followed by visual homing-in to the model 'warthog' from the edges of the roughly 5-m radius of the clearing around the odour source (which the flies' visual resolution should comfortably allow; Gibson & Young, 1991). It is also the same as the catch at the un-baited 'warthog' placed a similar distance *down*wind of the release battery (e.g. 1.3–1.5% of recaptures at 50 m with or without odour, and without significant variation between trials).

With odour. When 'host' odour was released from the recapture site, recapture rates were much higher, with a mean of over 6% at even 75 m (Table 1). This is 3 times the ~2% that would be expected with random radial dispersal from the release battery (P < 0.01). It was theoretically possible, however, that these recaught flies had arrived in the vicinity of the recapture site for reasons unrelated to odour, and were only then induced by its presence to head for the visual target of the 'warthog'. The supplementary control experiment with the ring of nets (Methods) was performed to test whether this was the case. For the 50-m experiment, it showed that the four nets placed around the 'warthog' net caused no increase in the total catch in the absence of odour, but doubled it when odour was present (Table 1, row 3).

Thus, many of the flies 'attracted' to the region of the 'warthog' by the odour were *not* then induced to head directly for it by the odour; on the contrary, their presence in the vicinity was revealed only by the more distant nets. Additionally, this experiment indicates that the real number of flies successfully locating the odour source, by arriving within 2 m of it, was in the range of 12–42% of releases – closer to 6 times random expectation than the 3 times inferred above; and if one takes the total efficiency of electric nets as ~45% (Griffiths & Brady, 1994), this figure may be even higher.

Condition of the flies. In order to check the physiological state of the marked–released flies, samples were taken (in the 75-m experiment) of: (i) the recaptured marked flies, (ii) 'unreleased' flies that were trapped at the same time but not used experimentally, and (iii) 'wild' flies 'recaptured' along with the marked flies. These were examined for body weight, residual blood meal, fat content, wing fray (approximate age), and female ovarian age. The only significant difference found between the three groups was that the 'wild' females contained ~22% more fat than the recaptured flies. The marked males were thus indistinguishable from the typical local flies active at the time, and the marked females were closely similar.

Flight times

Distribution. The frequency distributions of release-recapture intervals for the three distances are shown in Fig. 2. It is apparent that, as with take-off (Fig. 1), the rate of arrival at the odour source was not constant, but had a large initial peak. Fig. 3 shows the distributions plotted as log-survivorship curves (control data are not shown because total recaptures involved <5 flies per curve; see Table 1).

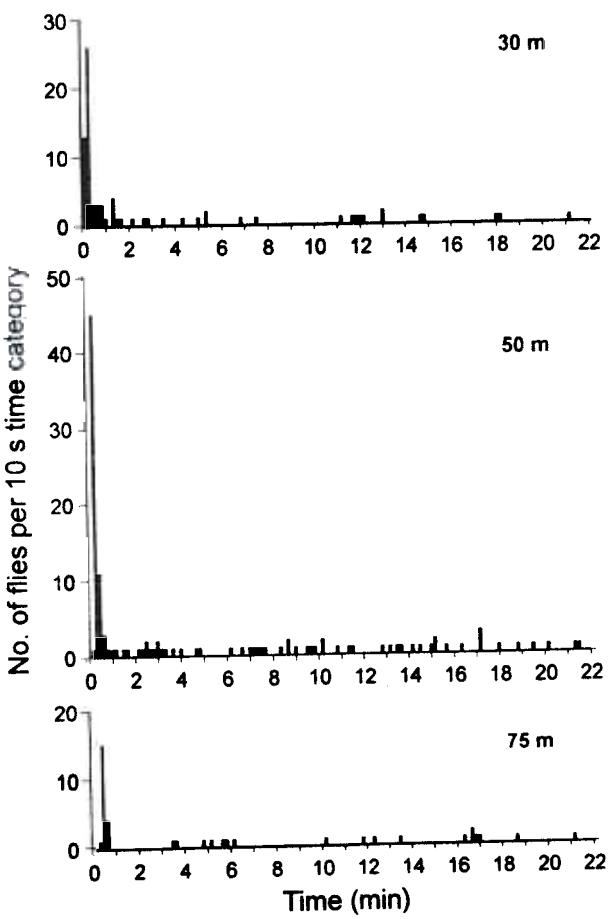

Fig. 2. Frequency distributions of the flight times (delay between take-off and arrival) of all flies recaptured after release from 30, 50 and 75 m downwind.

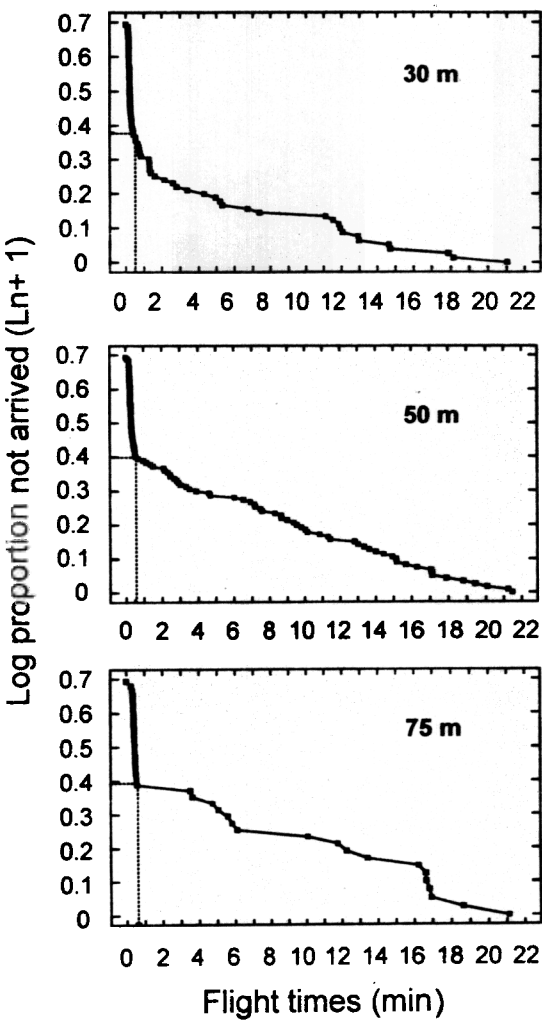

Fig. 3. Log-survivorship profiles of the flight times of all recaptured flies at 30, 50 and 75 m (from 77, 118 and 40 accurately known times). The cut-off points (inflections) between the 'fast' and 'slow' rates of arrival are shown by the cursor lines in each figure (at 26, 32 and 36 s, respectively).

Each curve has a sharp change of slope at approximately the same point, indicating two distinct rates of arrival: a 'fast' rate comprising flies arriving within 40 s of take-off (at speeds that are normally distributed; Fig. 4), and a 'slow' rate comprising flies arriving with a much lower, constant probability over the ensuing 21 min (as is visually obvious from the distribution of flight times beyond 1 min in Fig. 2).

Recaptured flies are thus categorized hereafter into one or other of these cohorts, referred to as 'fast' and 'slow'. Almost exactly 50% fell into each cohort, at all three distances, and there were also no differences between them as to the proportion arriving in upwind flight (89.1% fast versus 89.4% slow). The nine control flies showed no sign of separating out in this way, their recapture times being evenly distributed throughout the 22 min.

'Fast' flies. The actual numbers of flies that found the odour source in 'fast' flight was small, being on average only 3–10% of all released flies (i.e. 50% of Table 1, column 1), or about six,

Table 2. Comparisons between the fit of normal *versus* exponential functions for the 'fast' and 'slow' release–recapture times at the three distances (significance measured as the Kolmogorov-Smirnov statistic DN for the differences between the data and the model).

	Normal		Exponential		
Distance	DN	*P*	DN	*P*	Verdict
		~0.50	0.43	<0.01	Normal
		~0.10	0.46	<0.01	Normal
		~1.00	0.50	<0.01	Normal
B. 'Slow' flies					
30 m	0.20	~0.10	0.19	~0.10	Either
50 m	0.11	~0.50	0.16	~0.10	Either
75 m	0.20	~0.45	0.19	~0.50	Either

three and two flies per trial from 30, 50 and 75 m. These numbers are, however, double the expectation for random dispersal from the release point (<3, 2 and 1 respectively, see above), and highly significantly different from that expectation ($P<0.001$ at each distance; χ^2).

None of the initial 'fast' peaks at the three distances differ from normal distributions (see Fig. 4), but all differ significantly from the negative exponential distribution which would arise if the flies arrived with a constant probability through time (Table 2A). There was also no relationship between the probability of arriving in fast flight and whether or not a fly was in the fast initial take-off peak shown in Fig. 1. The fast flies thus apparently represent a single behavioural phenomenon centred around mean flight times of 13, 16, and 18 s for the three distances (see Fig. 3 legend).

As would be expected, the mean release–recapture times of the fast flies increased linearly with distance over the 30–75 m range (Fig. 5). The mean straight-line displacement speed for all three distances pooled was 2.8 m s^{-1} (Fig. 4), but this assumes that the rate of upwind displacement was independent of the distance from which the fly started, whereas treating each distance separately gives mean flight speeds of 2.2, 2.8 and 2.9 m s^{-1} from

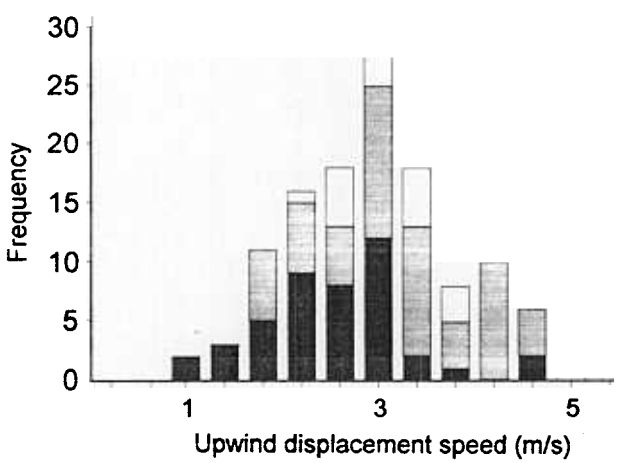

Fig. 4. Frequency histogram of the rates of upwind displacement (i.e. straight-line speed) of 'fast' flies recaptured from 30 m (solid), 50 m (shaded) and 75 m (open). Mean speed from all 'fast' flies pooled is 2.8 (SE ± 0.08) m s^{-1}.

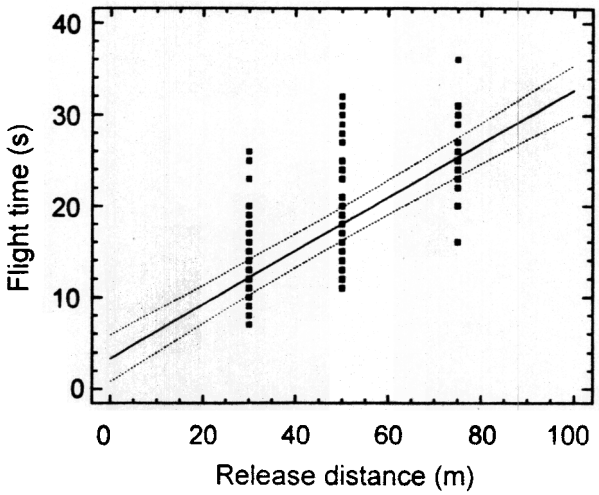

Fig. 5. Regression (with 95% c.i.) of release–recapture times of the 'fast' flies against the three release distances ($y = 3.34 + 0.29x$; $r^2 = 47\%$).

30, 50 and 75 m ($P<0.05$ for 2.2 *v* 2.9). Furthermore, although the inflections in the survivorship curves in Fig. 3 show that the separation of the fast and slow cohorts occurred later as distance from the release point increased, this delay is far less than would be expected from the simple increase in flight distance. No physiological factors (nutrition, age, etc.) explained this speed variance, so some sort of change in flight performance is indicated.

It seems that there is a small but significant increase in displacement speed with increasing distance between the point of take-off and the odour source. This seemingly paradoxical result is in fact consistent with previous observations of decreasing flight speed with increasing odour concentration as an odour source is approached (Warnes, 1990; Gibson *et al.*, 1991).

'Slow' flies. The slow flies performed very differently. For them, there are no grounds for dismissing the hypothesis that they had a constant probability of arrival over time (although their distributions do not differ from normal either; Table 2B). The hazard functions of the exponential distributions at the three distances are not significantly different ($\chi^2 = 5.38$, df 2, $P > 0.05$), so we conclude that they all arrived at the source with a constant probability over the ~20-min-period – in marked contrast to the fast flies' arrivals, which were grouped around a central mean. As the mean duration of each flight by a tsetse is less than ~2 min (Brady, 1972, 1988), these slow flights must have involved at least one, and perhaps many rests *en route*. Flight speeds are therefore not calculable.

Effects of wind

We measured (at the odour source) mean wind speed, wind direction, and directional variance, for the 90 s before take-off, during flight, and for the 60 s before recapture, for the 50 and 75-m release distances (equipment failure excluded 30 m). No differences in the wind's speed or direction prior to take-off were

Table 3. Analysis of deviance in the probability that a fly fell into the 'fast' cohort, in relation to mean wind speed during a trial (50 and 75 m data pooled). The probability of fast flight is taken as a binary variable (fast 0, slow 1) against which potential explanatory variables are fitted (parentheses = d.f.).

Parameter	Residual deviance		Change in deviance (χ^2)		P
Total deviance	215	(154)	–	–	
+ Date	167	(132)	-48.3	(22)	<0.001
+ Wind speed	212	(153)	-2.8	(1)	N.S.
+ Date + Wind speed	158	(131)	–	–	
– Wind speed	167	(132)	+9.0	(1)	<0.01
– Date	212	(153)	+54.5	(21)	<0.001

revealed between recaptured and non-recaptured flies in the 50-m experiment, and no effect of speed in the 75-m experiment, but for this distance the mean wind direction prior to take-off was more on line from the odour source for the recaptured flies than for the 'lost' flies ($t = 4.36$; $P < 0.001$).

There was significant variation in the proportion of fast flies from day to day (independent of season; cf. temperature, below). When this was allowed for, the probability of fast flight decreased as the wind speed during flight increased (Table 3). That implies that the flies experienced increasing difficulty in finding the source in a single step in higher winds, a conclusion also suggested by Fig. 6, which implies that the proportion of flies recaptured may correlate negatively with the mean wind speed during flight (though see Discussion).

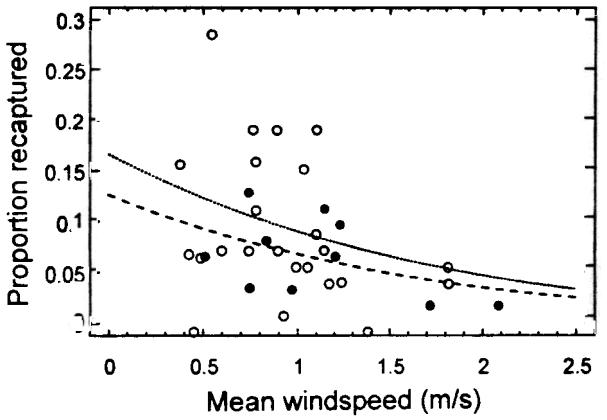

Fig. 6. Fitted slopes for the relationship between the recapture rates of marked flies and the mean wind speeds during the 50-m (dotted line, open circles) and 75-m (broken line, solid circles) experiments; no wind data were available for the 30-m experiments. Lines are curved after detransformation from a logit-link fit ($r^2 = 11\%$ overall).

Effects of temperature

We collected suggestive evidence (not shown) that the proportion of recaptures was affected by the daily maximum temperature, with a positive correlation up to around 32° and a negative correlation above that. However, because of the changing season during the observations, the rising slope of the relationship relates to data from the 50-m experiment only and the falling slope to the 75-m experiment only, so other (unknown) factors may have been involved.

Nevertheless, the implied inverted 'U' agrees qualitatively with the bimodal correlation of activity level with temperature shown by Brady & Crump (1978), and may therefore reflect the increased tendency for tsetse to seek shade above about 32° (Pilson & Pilson, 1967). Temperature had no detectable effect on flight times, so flight speeds did not increase with temperature, as might have been expected from typical Q_{10} effects on flight muscle rates.

Discussion

Natural conditions?

It is impossible to recreate exactly the conditions in which tsetse flies take off naturally, but our release tubes provided a situation that was perhaps as close to this as is practicable to achieve experimentally. This will have been especially true for the flies which did not take off in the first rush when the door was opened. Also, since there was no detectable relationship between the rapidity of take-off and either the probability of finding the odour source or the speed with which flies did so, the take-off conditions seem to have created no behaviourally significant biasses.

Three other factors also imply that we were looking at natural behaviour during the flies' orientation to the odour source. First, our flies were physiologically closely similar to the contemporary local 'wild' flies. Second, negligible numbers of flies arrived at the recapture point unless odour was released there. Third, the ring-of-nets experiment showed that we were not merely measuring an odour-evoked visual response to the model 'warthog' from flies that had arrived near it by some other, irrelevant means.

Efficiency of odour source location

Vale's 1980 results imply that at least 80% of the flies that contact an ox-odour plume respond to it and arrive near its source during the course of an afternoon's activity. Our own recapture rates are much less than half this, even if one takes the ring-of-nets experiment as showing that twice as many flies arrived near the 'warthog' as were actually caught by it. However, we cut off our observations at 22 min, and if we had continued them we would certainly have increased the recapture rate, since the constant rate of 'slow' arrivals continued when we test-recorded a second 22 min.

In any case, a closer look at Vale's modelling suggests that his parameter Z may be a better representation of what we were measuring. His Z is the proportion of the flies that entered a 130-m-wide clearing and were caught by a 'warthog' at the clearing's upwind apex. He estimates Z as about 25%, which falls in the middle of our 10–40% range. Interestingly, these rates coincide with the rates at which gypsy moths are reported to find a

pheromone source under similar circumstances, namely, with 17–45% success from 80 to 20 m away (Elkinton *et al.*, 1987).

The role of the wind

With only one anemometer, sited at the odour source, we could not know the course of the wind between recapture and release sites, and therefore could not be certain whether odour was present at fly's take-off (cf. the Elkinton *et al.* experiment). However, with the mean wind pointing towards the release site and blowing at just less than 1 m s^{-1} (Fig. 6), the plume would have been at least 10 m across by 50 m (since plumes expand at ~10° from their source at this wind speed; Brady & Griffiths, 1993). Thus, looking for correlations between wind and flight by using the wind 90 s before take-off should normally have ensured the presence of odour when the wind was on-line.

Even so, correlations between arrival rates and the wind parameters were weak, the only significant results were: (i) a positive relationship between the 'on-lineness' of the wind before take-off and the flies' success at finding the source from 75 m, and (ii) an indication that, within any one day, low wind speeds promoted the probability of locating the source in 'fast' flight.

The weak negative correlation between recapture rate and wind speed implied in Fig. 6 is not what had been expected from the relationship between wind speed and the straightness of odour plumes (Brady *et al.*, 1989, 1990). However, the significance of this regression depends entirely on the four points above 1.5 m s^{-1}, and we now know that the relationship between wind speed and tsetse flies' success at finding odour sources is more complex than we had originally supposed (a matter pursued in two subsequent papers: Brady *et al.*, 1995; Griffiths & Brady, 1995).

Wind speed was, indeed, not a significant factor when fitted with other variables in a multiple ANCOVA of flight times. Interestingly, however, flight times during 'fast' flight from 75 m did increase significantly with wind speed (r^2 = 14%). This would be expected if flights took longer because of the counter pressure of increasing headwinds, but the slope of the relationship is much less than it should be from such a simple mechanical effect. For 75 m, there was in fact only an additional 3.5 s of flight time for each additional 1 m s^{-1} of wind, whereas about 5–15 s would have been expected for a headwind increase from 0.5 to 1.5 m s^{-1} (and about 10–45 s for the increase from 1.5 to 2.5 m s^{-1}); the precise figures depend on the assumptions made about flight speed in zero wind.

This result thus strongly supports the notion that tsetse flies maintain a preferred groundspeed (probably at around 5 m s^{-1}; Brady, 1991), largely independent of wind speed, just as other insects do (David, 1986), an inference also supported by the lack of any increase in flight speed with increasing temperature (see *Temperature*, above).

Orientation mechanisms

Tsetse flies certainly steer their upwind flight in host odour 'anemotactically' (Introduction); we use this term to cover odour-modulated, optomotor-guided navigation upwind (Kennedy, 1986). Moreover, we know that the majority of our recaptured flies must have responded to the odour in order to find the source, because it was only in the presence of odour that they arrived in numbers greater than random – about 3–6 times greater. At least in part, therefore, they must have located the source anemotactically. However, we do not know when a fly was within the odour plume and when not. Its release–recapture interval is therefore the total time it spent reaching the source, rather than the time it spent actually orienting in the odour, which may be substantially less.

As the 'slow' cohort of flies took from 1 to more than 20 min to reach the source, they must have stopped at least once along the way, since tsetse are rarely active for more than a minute or so at a time (Brady, 1972, 1988). It is therefore impossible to infer anything useful about their flight speeds or orientation mechanisms. At 20 min their upwind progress approaches Bursell's hypothesized rate of ~5 m min^{-1} for a 'series-of-steps' strategy (Bursell, 1988), but it is more parsimonious to assume that their 'slowness' merely represents a more interrupted version of the 'fast' flies' behaviour than something qualitatively different.

For the 'fast' flies, two possible orientation mechanisms suggest themselves.The fastest flights were at around 4.5 m s^{-1}, with the 'winner' a fly that did 50 m in 10.6 s, equivalent to a straight line speed of 4.7 m s^{-1}. With a cruising speed of ~5 m s^{-1} (Brady, 1991), this implies progress up the plume in a single, almost straight flight. Indeed, the average deviation of the upper quartile of recaptured flies (those above 2.8 m s^{-1}) from a direct line to the odour source would only add some 20 m to a 50-m flight. This amount of deviation leaves room for *some* in-flight anemotactic correction of flight direction, but contrasts sharply with moths' zigzagging across the wind line as they progress up pheromone plumes (David, 1986; see Kuenen & Cardé, 1994, for later references).

The obvious alternative interpretation is that, for the fastest flights, the speed and the lack of any clear correlation between wind direction and the probability of finding the source imply little or no in-flight anemotactic direction correcting. Such flights might then be best explained by an 'aim-then-shoot' process (Kennedy, 1986), an open-loop control system in which the fly follows the upwind direction set by mechanical anemotaxis at take-off for a considerable distance, before it finally changes course or lands.

This occurs in other Diptera, such as cabbage root flies (Hawkes & Coaker, 1979) and onion flies (Dindonis & Miller, 1980), and has been proposed for tsetse flies (Bursell, 1984, 1987) in which upwind take-off in host odour undoubtedly does occur (Bursell, 1984, 1987; Torr, 1988a). Whether these flights remain more or less straight in some 'ballistic' manner or involve minor in-flight anemotactic corrections is unclear, however. It is not known how flight oriented directly upwind may be steered (see David, 1986; Brady *et al.*, 1995), but there is evidence that even moths head almost due upwind in their upwind 'surges', at least for a second or two (Haynes & Baker, 1989; Mafra-Neto & Cardé, 1994).

In fact, unless our aim-then-shoot flies 'took aim' very accurately (to within ~±5°), they would probably have missed the 'warthog' if they had not made some kind of correcting manoeuvre in flight in order to pass sufficiently close to see it. Some in-flight anemotactic responses are therefore implied, even

for the fastest flies. If they did not do this, their only other means of direction correcting would have been to land to reassess the wind direction. At least for the upper quartile of flies, however, that seems unlikely, because it could only have been for a few seconds, and such brief wind assessments would be particularly susceptible to misleading turbulence at the landing site (Brady *et al.*, 1989).

The slower 'fast' flies which arrived towards the end of the fast cohort had the time to have flown between 2 and 4 times the straight-line distance to the source. They therefore had ample opportunity to make anemotactic corrections while in flight, although they might also have landed briefly (for <~20 s) on losing contact with the plume, and then responded on re-contacting it with an aim-then-shoot response.

As tsetse travel at around 5 m s^{-1} for up to 10 min a day, they can cover some 3 km during one afternoon's activity peak. They therefore have the possibility to 'search' a large area of bush for any sources of host odour they encounter. It seems likely that they progress towards a particular source by a form of biassed random walk (Brady *et al.*, 1990; Williams, 1994), in which the bias is provided by a combination of upwind take-off in odour of the 'aim-then-shoot' kind, and in-flight corrections of direction whenever they enter or leave host odour plumes. If take-off aim is lucky, they will typically find a host in less than a minute. If it is not, they may then enter an anemotactically-guided flight, which can last the minute or two of a single flight. If that, too, is unsuccessful and they lose contact with the odour, then the combined strategies must be repeated, for perhaps tens of minutes and covering large areas of bush.

The fact that tsetse use what appears to be a rather crude form of source location, rather than the more precise methods employed by moths, presumably relates to the nature of their respective targets. The calling female moth is a point fixed in space that 'wants' to be found, whereas the tsetse's host is large (very large if in a herd), certainly does not want to be found, and may well move off before the tsetse can find it, at a speed faster than the tsetse can fly. It may thus be that a biassed random walk which covers a large area of bush is, on average, a surer form of host location than any version of a moth's precision tactics.

Acknowledgments

We thank the Assistant Director, Department of Veterinary Services, Tsetse and Trypanosomiasis Control, Zimbabwe, for hospitality and much material help at Rekomitjie Research Station. Dr G. A. Vale and Dr S. J. Torr were particularly helpful with advice during the running of the experiments; Dr Vale and Mr K. Groenendijk commented most usefully on the manuscript. We also thank Mr P. Madungaza, Mr S. Madungaza, Mr B. Mbumbhuru and Mr M. Ngazana for invaluable assistance in the field. The work was financed by the Overseas Development Administration of the Foreign and Commonwealth Office, London, under contract No. X0138.

We dedicate this paper to the late Einar Bursell, who led the study of behavioural physiology in tsetse for so long.

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Accepted 24 January 1995