East African Trypanosomiasis Research Organization, Tororo, Uganda
C/o Hope Department of Entomology, University Museum, Oxford,

Comparisons between three small collections of *Glossina morsitans morsitans* (Machado) (Diptera: Glossinidae) from the Kilombero River Valley, Tanzania Part 1. Characteristics of flies exhibiting different patterns of behaviour

J. Ford, I. Maudlin[1] and K. C. Humphryes[2]

[1] Present address: Department of Entomology, London School of Hygiene and Tropical Medicine, London, W.C.1.
[2] Present address: Imperial Cancer Research Fund Laboratories, Lincoln's Inn Fields, London, W.C.2.

Abstract

Five types of behaviour were recognised in *Glossina morsitans morsitans* (Machado) post-teneral males collected on a fly round near Ifakara in central Tanzania. Each could be characterised by differences in nutrition state measured by fat content, by haematin content, indicating frequency of empty or partially full intestines, and also by differences in the age groups predominating in each behaviour type sample. The nutrition and age characteristics of flies which entered a landrover while travelling through the area are also described. Female and teneral data are discussed and shown to be not without significance in relation to behaviour. One object of the study is to present data in a form in which they are most likely to be comparable with those obtained in laboratory studies of tsetse behaviour.

Introduction

The faults of the fly round as a measure of population density of tsetse flies have been much discussed (see, for example, several chapters in MULLIGAN, 1970). What is measured is response to a bait. This varies with time of day, with season, with place, with climate and with the behaviour of the bait. Different baits elicit different responses. Recently, in the Zambezi valley fly belt, PILSON & MacKENZIE (1972) have shown that an ox or a donkey may attract more *Glossina morsitans* and *G. pallidipes* than do tamed, or constrained, natural hosts, kudu, bushpig and bushbuck. Catches from human or domestic animal baits, however, have a use other than density measurement. They contribute to the assessment of infection risks. Regarded from this point of view the fly round (men walking along a path and periodically stopping to catch) is a means of studying situations in which human beings walking through the bush may be subjected to different rates of transmission of trypanosomes from wild hosts. Thus, where teneral or immature flies preponderate among those which bite man the latter's risk of becoming infected must be less than in other situations in which flies of all ages attack him (FORD, 1969).

Long ago JACKSON (1933) demonstrated that *Glossina morsitans* which perched "head-up" on a man were "hungry" as compared with those which perched "head-down" or which followed him and rested on the ground when he halted. The present study revives this approach to the behavioural responses made by tsetse flies to man under natural conditions.

We analyse three small collections of *Glossina morsitans morsitans* (Machado), hitherto commonly called *G. morsitans orientalis* Vanderplank, from the eastern fly belt of Tanzania, made between 11 and 23 September 1969 (that is in the middle of the dry season) near Ifakara (8° 10' S, 36° 38' E). The largest, of 216 *G. morsitans* (and 2 *G. pallidipes* Austen and 1 *G. brevipalpis* Newstead) was gathered during 14 visits to an area 10 miles north-east of Ifakara known as Matalawe. It is drained by a stream of that name running off the Iringa escarpment into the Kilombero River. It was the source of tsetses examined for trypanosome infections by GEIGY, KAUFFMANN & DIEHL (1968) and referred to by them as the Ifakara region. The second collection comprised 61 *G. morsitans* collected inside the landrover at the end of each journey from Ifakara to Matalawe. The third collection, of 90 *G. morsitans,* came from an area called Mau-mau, near the edge of the Selous National Park and about 25 miles south-east of Matalawe. The two areas are separated by the wide treeless flood plain of the Kilombero River.

Field work was based upon the Rural Aid Centre run by the Swiss Tropical Institute at Ifakara and laboratory analyses were carried out at the East African Trypanosomiasis Research Organization Laboratory near Tororo, Uganda.

Methods of collection and analysis

(a) *Field work.* Collections 1 and 3 were made by an observer with two assistants operating a fly round. That at Matalawe had 27 sections of 50 paces each and took from 1 to 1½ hours to work. The first morning's work, on 11 September, was cut short by a grass fire which passed through the area between 10.30 and 11.00 hours. At Mau-mau there were 40 sections, worked once on the evening of 14 and once on the morning of 15 September. The second, landrover, collection comprised only flies that entered the vehicle when it was travelling, at not more than 15 m.p.h., along a track through 2 miles of woodland which had to be traversed before the Matalawe fly round was reached. Flies following or carried on the outside of the landrover were not collected.

Each fly was put in a small, numbered, polythene tube with a hinged lid on which, before closing, one drop of ethyl acetate was allowed to fall from a dropping bottle. Information about each fly was recorded as illustrated in Figure 1. Apart from the first two rows, which show location and number of each specimen, field observations are entered by ticks. Time of day and psychrometer readings are noted at the beginning and end of each traverse of the fly round. On return to base the tubes were opened and stored upright in a desiccator to await the journey to Uganda.

Locality: Matalawe. Date: 23. 9. 1969. Man.
Time begun: 16.08. Dry bulb: 30. Wet bulb: 21.
Time ended: 17.30. Dry bulb: 28. Wet bulb: 22.

Sector No.	11	11	11	11	11	12	13	13	14	14
Fly number	910	278	895	942	904	939	879	913	940	886
Species 1	√		√	√	√	√	√	√	√	√
Species 2		√								
Species 3										
Male	√		√	√		√	√	√	√	√
Female		√			√					
Teneral		√								
Head-up		√							√	√
Head-down										
Not probing		√							√	√
Probing										
Mjanja					√					
On ground	√		√	√		√	√			√
On vegetation										
Man – body		√							√	√
Man – legs										
Ox										
Fray	1	1	1	2	1	1	1	1	1	3
Vein	3.7	4.3	3.5	3.8	4.2	3.7	3.7	3.8	3.5	4.0
DW	11.5	7.5	10.6	12.8	11.2	11.9	13.9	6.1	9.6	12.0
RDW	8.8	6.5	7.8	9.1	9.2	8.2	10.9	5.6	7.2	8.3
Fat	3.5	1.0	2.8	3.7	2.0	3.7	3.0	0.5	2.4	3.7
Haematin	0.055	0.012	0.372	0.185	0.004	0.276	0.760	0.008	0.012	0.010

Fig. 1. Type of data recording form to accommodate field observations and laboratory analyses.

(b) *Laboratory analysis.* Results are entered on the same form. It is a simple matter to transcribe data for each fly to a punched card for subsequent computer analysis.

To facilitate handling in the laboratory the flies were transferred to $^1/_2$ ounce bijou bottles in which they retained their identity for all subsequent work. They were dried in batches of about 100 bottles in a vacuum oven at 85 °C for about 16 hours. After drying they were stored in a desiccator from which they were removed singly and weighed to the nearest 0.1 mg on a torsion balance to determine the dry weight (DW). The flies were then treated with three changes of chloroform in 36 hours to remove fat, heated at 56 °C for one hour to drive off excess

chloroform and then re-weighed to give the residual dry weight (RDW). Fat weight equals DW less RDW as in BURSELL (1961a).

One wing was removed from each fly. Scotch tape was used to fix wings to slides, the number of each wing being written on the slide in wax pencil or diamond. Ten wings were fixed to one slide for subsequent assessment, under a low power microscope, of wing fray and measurement of the middle section of the 4th longitudinal vein. (These techniques have most recently been described by POTTS in MULLIGAN, 1970).

Finally the fatless abdomen was removed from the thorax and snipped into thin slices with bow-spring entomological scissors. Haematin from any residual blood meal (RBM) was extracted by addition of 0.1 N sodium hydroxide and 80% ethanol. Sodium hydrosulphite and freshly distilled pyridine were added at the last moment to form the haemochromagen, pyridine ferroporphyrin (PHC). Optical density of PHC was measured spectrophotometrically at 558 m. The whole process was a slight modification of the method adapted by BURSELL (1966) from SUTTON & ARTHUR (1962).

The observed behaviour of *Glossina morsitans*

Behaviour patterns were recorded as in an earlier study in Nigeria (FORD, 1972) but with one addition. Not uncommonly a catching party encounters flies which are very active, making short rapid flights from one person to another or from one part of the body to another. They perch only momentarily and are thus difficult, but not always impossible, to catch. Field assistants in East Africa may speak of them as "mjanja", a swahili word meaning "elusive" or "cunning". We use the same word but without attaching to it any implication of evasiveness. No nuptial flight was seen. From the description in FORD (1972) it might be inferred that this is the more usual method of mating. Males probably more often encounter females when the latter are feeding or about to feed.

Standardization of observation of "probing" was attempted by instructing catchers to count up to 20 before netting a fly which had alighted on themselves and noting that it had not lowered its proboscis. This count of 20 averaged about 17 seconds. It is likely that had a longer interval been allowed more "head-up" flies would have been seen to probe.

Primary analysis

We are here concerned to define the characteristics of flies exhibiting different patterns of behaviour at Matalawe. In Part 2 we shall compare

the Matalawe data with those obtained from Mau-mau and, in both, attempt to relate them to environmental factors.

Table 1 gives the numbers, means and standard deviations of residual dry weights, fat weights and PHC values for all teneral and post-teneral flies recorded at Matalawe on the fly round and in the landrover. Mean vein lengths (eyepiece micrometer units) and wing fray assessments in three instead of the usual six categories are also included.

Only in vein length do teneral females differ significantly from teneral males. BURSELL (1960), using surface area of the thorax as a measure of size, showed that teneral females are about 13 per cent bigger than males of the same RDW. In Table 1 mean female vein length is 17.6 per cent longer than that of males. Both males and females increase in weight as they grow older. BURSELL (1961b) suggested that male *Glossina swynnertoni* need to complete two feeding cycles and females three before their thoracic muscles are fully developed.

Among post-tenerals the greater RDW is due not only to the growth of muscles but also to gut contents representing blood meals in various stages of digestion. In some of the post-teneral females there is also the added weight of a larva in the uterus.

Teneral fat is derived from the pupa and, ultimately, from the mother. In post-tenerals the greater fat weight is the result of accumulation derived from blood meals. In pregnant females it includes uterine larval fat which, however, may not be extractable because the larval integument is not always penetrable by chloroform.

Standard deviations are large in relation to means of fat weights. BURSELL (1966) illustrates the frequency distributions of nutrition reserves (mostly fat) to show that participation in the following swarm cannot be regulated by the nutrition state. His histograms show a normal distribution and demonstrate that post-teneral male *G. morsitans* taken on a fly round may be almost completely devoid of fat (under 0.5 mg) or may have fully developed fat bodies (of over 5.0 mg). Post-teneral male fat weights at Matalawe ranged from 0.3 to 5.5 mg.

Variation is most pronounced in the PHC data. Here it is due to a very marked skewness in distribution of individual values. These features are examined below. It may be noted here that females caught on the fly round show a PHC mean almost identical with that of females caught in the landrover. On the other hand, males from the fly round yield a PHC mean significantly higher than that obtained from males in the landrover.

In column 6 of Table 1 the mean vein length from the small sample of teneral males is significantly ($P < 0.001$) less than that obtained from post-teneral males. Size reflects the nutrition state of the mother, but the appearance of these smaller flies at this particular time may have been in some part due to the above-noted grass fire.

Table 1. Teneral and post-teneral *G. morsitans* caught on a fly round or in a landrover, with sex ratios, mean residual dry weight (RDW), fat weight, mean optical density of haemochromagen (PHC) and vein length, with standard deviations. Also numbers and percentages (in brackets) of flies in different age (wing fray) categories

Sample	Sex	1 N	2 Sex ratio %	3 RDW mg	4 Fat mg	5 Mean PHC optical density	6 Vein length units	7 Wing fray 1	2	3
Tenerals	M	5	24	4.4 ± 1.1	0.9 ± 0.3	(0.005)	34.8 ± 1.3	5 (100)	0 (0)	0 (0)
	F	16	76	5.0 ± 1.4	0.8 ± 0.4	(0.007)	40.9 ± 1.9	16 (100)	0 (0)	0 (0)
Post-tenerals	M	180	90	7.5 ± 1.5	2.6 ± 1.2	0.137 ± 0.293	37.3 ± 1.5	86 (48)	56 (31)	37 (21)
Fly round	F	19	10	7.9 ± 1.8	1.3 ± 0.7	0.070 ± 0.169	41.6 ± 2.5	12 (63)	6 (32)	1 (5)
Post-tenerals	M	29	51	6.3 ± 1.1	1.3 ± 1.0	0.017 ± 0.033	37.2 ± 1.9	19 (66)	7 (24)	3 (10)
Landrover	F	28	49	10.1 ± 2.1	3.3 ± 2.0	0.083 ± 0.172	41.3 ± 1.5	10 (36)	13 (46)	5 (18)

Post-teneral differences

Males: RDW Fly round > Landrover 1.2 mg $t = 4.73$ $P < 0.001$
 Fat Fly round > Landrover 1.3 mg $t = 6.31$ $P < 0.001$
 PHC Fly round > Landrover 0.120 mg $t = 5.20$ mg $P < 0.001$

Females: RDW Fly round < Landrover 2.2 mg $t = 3.84$ $P < 0.001$
 Fat Fly round < Landrover 2.0 mg $t = 4.25$ $P < 0.001$
 PHC Fly round = Landrover No difference

PHELPS & JACKSON (1971) have shown that a rise of 1 or 2 °C in an otherwise constant temperature may produce an increase in eclosion rate. However, the difference between female teneral and post-teneral means is not significant. BURSELL & GLASGOW (1960) suggest that hot season stresses act with greater selective force on male tenerals than on females which are larger.

In column 7 teneral flies of both sexes show, as expected, an absence of fraying. Our wing fray 1 includes wing fray 2 of JACKSON's (1945) system but, according to POTTS (in MULLIGAN, 1970) this category is made up of wings showing "very slight or doubtfully genuine damage such as might be caused by the net during capture". No quantitative comparison is possible between wing fray numbers of the two sexes, nor do we attempt to estimate mean ages. We describe our wing fray 1 group as young flies, 3 as old and 2 as flies of intermediate age.

Sex and teneral ratios

Conventional treatment of fly round data excludes tenerals of both sexes as well as post-teneral females from consideration. "This idea", says GLASGOW (1970), "has never been critically examined." It is not our intention to examine it here, but only to compare differences in teneral and female catches on the fly round or in the landrover. It is evident from Table 1 that the sex ratio of post-teneral flies from the landrover does not depart significantly from 1 : 1, whereas on the fly round females form about 10 per cent of the catch, a proportion which experience leads one to expect.

In comparing the teneral ratios and also the comparative ages of flies taken by the two methods it is convenient to treat the two sexes separately. Male data are set out in Table 2. In it, and in Table 3, young flies include tenerals with those having unfrayed wings (WF1); older flies are those in wing fray categories 2 and 3. Within the limitations of our data Table 2 shows that the ratios of teneral to post-teneral and of young to older males do not differ significantly between fly round and landrover. This is not the case among females. Table 3 shows that tenerals predominate on the fly round, comprising 40.6 per cent of the female catch, whereas in the landrover tenerals form only 9.8 per cent of the female catch. When young females are compared with older the predominance of the former on the fly round is much greater (78.1 per cent) than in the landrover where older females form more than half (58.1 per cent) of the catch.

If the inside of the landrover is regarded as a trap, although in motion, these results are in accord with those of SAUNDERS (1962, 1964) and of HARLEY (1967), both of whom worked with *G. pallidipes* and showed that females caught in traps included more old individuals than did samples taken by walking men.

Table 2. Numbers of teneral and post-teneral (A) and of young (tenerals + WF1) and older (WF2 + WF3) males (B) caught inside the landrover or on the fly round

(A)	Teneral	Post-teneral	Total	(B)	Young	Older	Total
Landrover	1	29	30		20	10	30
Fly round	4	179	183		90	93	183
Total	5	208	213		110	103	213

$$P = 0.386$$

$$\chi^2 = 2.498$$
$$P < 0.2$$

Table 3. Numbers of teneral and post-teneral (A) and of young (tenerals + WF1) and older (WF2 + WF3) females (B) caught inside the landrover or on the fly round

(A)	Teneral	Post-teneral	Total	(B)	Young	Older	Total
Landrover	3	28	31		13	18	31
Fly round	13	19	32		25	7	32
Total	16	47	63		38	25	63

$$\chi^2 = 6.401$$
$$P < 0.05$$

$$\chi^2 = 7.174$$
$$P < 0.05$$

Females

BURSELL (1966) writes that mean fat weight in males gives a good indication, for comparative purposes, of the nutrition state of the tsetse population. Female fat weights, for reasons already given, cannot be used in this way; nevertheless, they are not entirely without significance.

In Table 1 the mean fat weights of tenerals of both sexes are virtually the same, but post-teneral females from the fly round have a mean fat weight half that of the males. By contrast, females in the landrover are much heavier, both in RDW and in fat, than are the males. They are also much heavier than females taken on the fly round. These differences are significant.

A comparison of frequency distributions of female fat weights is made in Figure 2. in which the wing fray of each individual contributing to the histograms is also shown. Clearly there is no significant difference between tenerals (A) and post-tenerals from the fly round (B), although
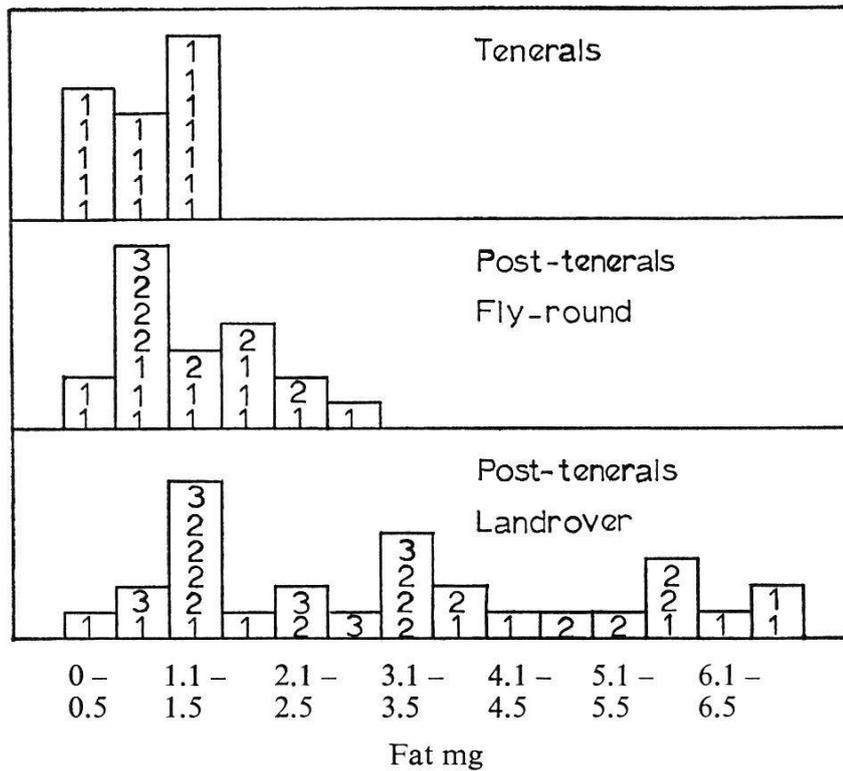
Fig. 2. Frequency distributions of female fat weights. Numbers refer to age groupings of individual insects contributing to histogram columns.

not all the latter are young flies. However, fat weights from landrover females (C) show a very different picture, with a wide range displaying an apparently trimodal distribution. It is likely that the troughs are associated with the presence in the uteri of larvae with skins impermeable to chloroform.

Age also contributes to this variation. Young (WF1) flies predominate among those with extractable fat weights above the median of 3.5 mg (6 above, 4 below), while all the old flies (WF3) have less than the median weight of fat (0 above, 5 below). The exact probability (Fisher's method) for significance of difference of these ratios is 0.042.

Post-teneral male behaviour

The 180 post-teneral males caught on the Matalawe fly round include 125 caught on the ground; 28 taken "head-up" on one or other of the catching party; 19 similarly caught "head-down"; 6 "mjanja" flies and 2 seen to probe within a period less than that needed to count twenty. Mean nutrition values for each behaviour category are given in the first four columns of Table 4. Flies taken from the ground have more fat and more haematin and have significantly greater RDW than those displaying other types of behaviour. Part of the greater RDW of ground flies is due to the greater weight of the residual blood meal

Table 4. Nutrition and age data for non-teneral male *G. morsitans* exhibiting different types of behaviour on the Matalawe fly round

Behaviour	1 N	2 RDW mg	3 Fat mg	4 Mean PHC Opt. Dens.	5 Log. PHC $\times 10^3$	6 Corrected RDW	7 Fat % of cor. RDW	8 Wing fray 1	2	3
On ground	125	7.9 ± 1.4	3.0 ± 1.0	0.170 ± 0.319	1.61 ± 0.72	7.2	42	56 (45)	40 (32)	29 (23)
"Head-up"	28	6.7 ± 1.6	1.4 ± 0.3	0.079 ± 0.270	1.08 ± 0.61	6.3	22	17 (61)	6 (21)	5 (18)
"Head-down"	19	6.3 ± 0.9	2.1 ± 1.0	0.032 ± 0.077	1.16 ± 0.44	6.2	34	12 (63)	7 (37)	0 (0)
"Mjanja"	6	6.2 ± 0.6	1.1 ± 0.6	0.011 ± 0.008	0.94 ± 0.30	6.2	18	4 (67)	1 (17)	1 (17)
Probing	2	(7.2)	(1.6)	(0.016)	–	7.1	22	1 (50)	1 (50)	0 (0)
Totals	180	7.5 ± 1.5	2.6 ± 1.5	0.137 ± 0.293	1.45 ± 1.01	6.9	35	90 (50)	55 (31)	35 (19)

(RBM) indicated by the higher mean PHC value. Other causes for the lower RDW observed in the other categories may be (a) that flies are immature and have not yet developed their full musculature or (b) that they are mature but starved, having consumed muscle tissue as a last source of energy. Fat weights suggest that "head-down" flies are not starved, but that the latter condition may characterise "head-up" and "mjanja" flies.

Interpretation of PHC values

Figure 3 displays the correlation between PHC values and RDW for all intermediate (WF2) and old (WF3) males, except those with RDW less than 6.0 mg. Omission of young (WF1) flies excludes any with incompletely developed muscles and integument, while removal of underweight but mature flies should exclude those in a starved state. Flies which yield PHC values of 0.012 or less are also left out. The range of
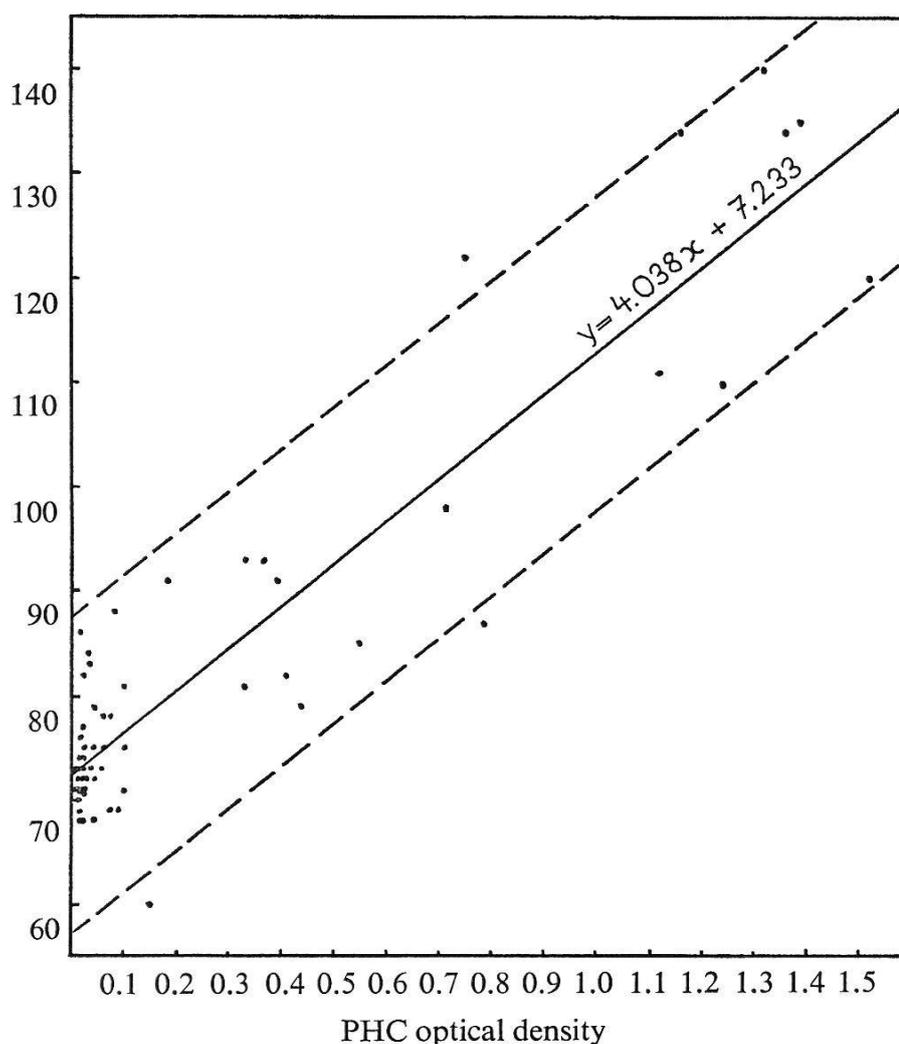

$$y = 4.038x + 7.233$$

Fig. 3. Correlation between PHC optical densities and residual dry weights of male *G. morsitans,* excluding young (wing fray 1) flies, flies with RDW less than 6.0 mg and flies which gave PHC values of 0.012 and under.

readings from 21 teneral flies ran from 0.003 to 0.012. By definition tenerals have not fed and the assumption is made that flies yielding PHC optical densities in this range have empty intestines.

The 95 per cent limits of error are shown. Contributions to variation other than those mentioned above must include the stage reached in digestion of the blood meal. Haematin split off from haemoglobin is not absorbed during digestion but passes unchanged through the gut (BURSELL, 1970). A fly interrupted soon after beginning to feed into an empty gut might yield a measure of haematin the same as that given by another fly in which the only haematin present was in the last quantum of faeces remaining to be voided after completion of digestion. Some flies may contain haematin from freshly ingested blood as well as from faeces still to be expelled from the previous meal. Haematin forms 2.7 per cent of the dry weight of undigested blood but 3.7 per cent of the dry weight of tsetse faeces (BURSELL, 1970). Variability will also derive from size differences, for large flies will tend to take bigger meals than small flies (BRADY, in press).

Data from 60 flies wer available for the construction of Figure 3. The correlation is good ($r = 0.914$; $P < 0.001$). The regression of y upon x intercepts the y axis at 7.23 mg and this figure may therefore be taken as the mean RDW of mature flies not suffering from starvation but lacking any residual blood meal.

The mean RDW of the 60 flies is 8.32 mg. The difference between this and 7.23 yields a mean RBM of 1.09 mg which corresponds to the mean PHC value of 0.268. A PHC reading of 0.10 is therefore equivalent to an estimated RBM of 0.407 mg. The mean PHC for ground flies of 0.17 in Table 4 thus converts to 0.7 mg RBM to give a corrected mean RDW of 7.2 mg. This could hardly be otherwise as the majority of the 60 flies used in Figure 3 were caught on the ground. Flies in other behaviour categories, save the two taken while probing, all give corrected means for RDW about 1.0 mg less than that of the ground flies (column 6).

In general the PHC means are not informative and logarithmic transformation of the data in column 5 does not assist interpretation. The chief interest of the PHC data lies in the extreme skewness of their frequency distribution. To facilitate construction of histograms the PHC values are arranged, in Figure 4, in groups rising by intervals of 0.01. Ground caught fly data are compared, in this figure, with the combined data for the other behaviour categories (excluding the two probing flies). The majority of flies, whatever their behaviour at the time of capture, have little or no haematin. Their intestines are empty or nearly so. This is in line with the observations of BURSELL (1966) as is also the fact that a few of them, especially among those caught on the ground, yield PHC values large enough to indicate that the blood meal can
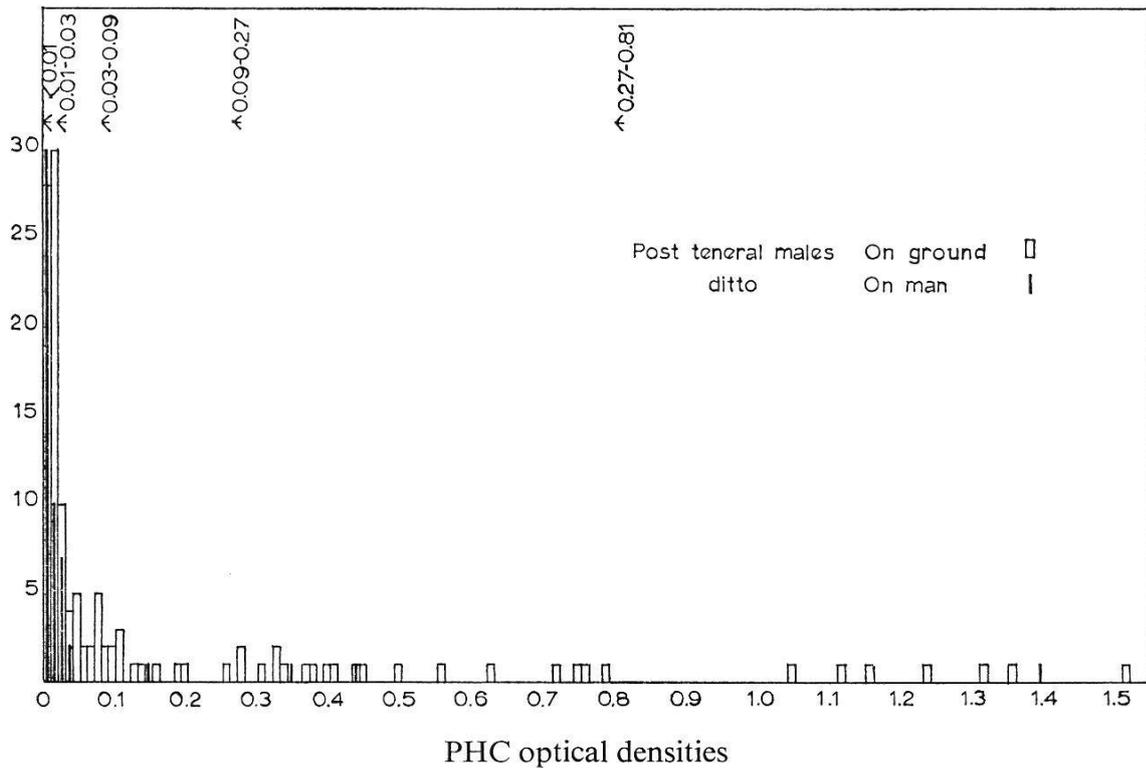
Fig. 4. Distribution of PHC optical densities, rising by intervals of 0.01, to show difference between haematins obtained in post-teneral male *G. morsitans* caught, on a fly round, on the ground or on man. Frequency groupings used in Table 5 are indicated.

only have been taken a short while before. The highest PHC reading of 1.520 converts to an estimated RBM of 6.2 mg. The water content of whole blood is about 79 per cent and therefore the fly from which this reading was taken contained blood which would have weighed about 30 mg, an acceptable figure for an average meal.

The PHC data are further arranged arbitrarily in Table 5 into 6 groups, which are indicated in Figure 4, each group having 3 times the range of its predecessor. This table may be considered as a contingency table in which the ratio, one to another, of the 6 groups among the ground flies is taken as the standard. χ^2 can then be used to determine if the ratios displayed among flies caught on man deviate significantly from that predicted by the null hypothesis. The main contributions to the very significant χ^2 value of 25.908 are made by flies with PHC values below 0.01, which are more numerous on man and those with high PHC values which are fewer on man than would be predicted by the ground fly ratio. Flies with PHC values between 0.01 and 0.03 do not differ from expectation and comprise about one third of the catch. They may be regarded as having had at least one blood meal, of which a trace is still detectable after complete defaecation. A similar result is obtained by comparing the "head-up" flies alone with the ground flies ($\chi^2 = 19.536$), but samples of other behaviour types are too small for comparison in this way.

Table 5. Arbitrarily grouped frequencies of PHC values of post-teneral males exhibiting different behaviour patterns, each group having three times the range of its predecessor

Behaviour		PHC value frequencies						Totals
		< 0.01	0.01– 0.03	0.03– 0.09	0.09– 0.27	0.27– 0.81	> 0.81	
Caught on ground		28	40	20	11	19	7	125
On man	"Head-up"	17	8	0	1	1	1	28
	"Head-down"	9	7	2	0	1	0	19
	"Mjanja"	4	2	0	0	0	0	6
Total on man		30	17	2	1	2	1	53
Total on ground + man		58	57	22	12	21	8	178

On ground v "head-up": $\chi^2 = 19.536$; P < 0.001
On ground v on man: $\chi^2 = 25.908$; P < 0.001

Nutrition state and behaviour

While we accept BURSELL's (1966) view that the concept of behaviour as regulated by the state of food reserves is untenable, it is clear that JACKSON's (1933) demonstration of a relationship between perching behaviour on a fly round and nutrition state is borne out and extended in our observations. Flies caught off the ground tend to be heavier and to contain more fat than those caught on man. Fewer of them have completely empty intestines and more of them contain partly digested blood meals than do the latter. "Head-up" flies tend to weigh about 1 mg less and to have only about half as much fat as ground flies. "Head-down" flies, on the other hand, although also deficient in RDW, have more fat than "head-up" flies but less than ground flies, both differences being significant. "Mjanja" flies are too few for any definite statement, but may be less well supplied with fat and include a still higher proportion with completely empty intestines than do the "head-up" flies.

Age and behaviour

The weight of fat in "head-down" flies suggests that they cannot be starving: their low RDW may thus be a consequence of immaturity. On the other hand, if the low RDW of "head-up" flies is due to starvation,

then there is no reason to suppose that they include more or fewer young or old individuals than do the well-nourished majority which are caught off the ground.

Again using the latter as the standard of comparison, the null hypothesis may be tested with "head-up" and "head-down" flies in 2×2 tables, omitting flies of intermediate age. This is done in Table 6. The "head-down" males, lacking any old flies, are evidently drawn from a younger section of the population than are the ground flies. Equally clearly there is no such distinction between the latter and the "head-up" flies. The samples are small but they support the interpretation put upon fat weights and RDW.

Table 6. Numbers of young and old males perched "head-up" or "head-down" on man, compared with numbers caught on the ground

	Young	Old	Total			Young	Old	Total
On ground				On ground				
	56	29	85			56	29	85
"Head-up"	17	5	22			12	0	12
				"Head down"				
Total	73	34	107			68	29	97

$$\chi^2 = 0.593$$
$$P < 0.5$$

$$P = 0.010$$
(Fisher's method)

Behaviour of females and tenerals

It remains to examine behavioural responses to the fly round party of post-teneral females and of tenerals of both sexes. They are shown in Table 7. Teneral males are too few for comment but it is notable that "head-up" flies predominate among females, both post-teneral and

Table 7. Frequencies of appearance of different behaviour types among post-teneral females and tenerals of both sexes caught on the fly round

Behaviour	Post-teneral females	Teneral females	Teneral males	Totals
On ground	2	–	–	2
"Head-up"	9	11	1	21
"Head-down"	3	–	2	5
"Mjanja"	3	–	–	3
Probing	2	2	1	5
Totals	19	13	4	36

teneral. This posture is normally taken up by feeding flies of both sexes and the conventional view of females and tenerals which come to a fly round party is that they come to feed. Table 7 gives 5 flies seen to probe out of a total of 36. In Table 4, which displays only post-teneral male data, the corresponding ratio is 2 out of 180. The discrepancy between these ratios is highly significant ($\chi^2 = 11.761$; P $<$ 0.001) and our data therefore support the classical interpretation of the female and teneral catch.

Some interest attaches to the two post-teneral females caught on the ground, a pattern of behaviour usually ascribed to mature males only. Both had unfrayed wings and were therefore young. Both yielded high nutrition reserve values (fat 2.4 and 2.7 mg and PHC readings 0.202 and 0.427) while in both of them a heavy RDW (9.1 and 9.4 mg) suggested that both were pregnant.

Discussion

It has always been a simple matter to suggest teleological reasons for commonly observed patterns of tsetse fly behaviour. Flies which perch "head-up" on man do so because they are about to feed and the posture adopted ensures maximum stability when the abdomen is gorged. Males follow a moving object because they are in a sexually appetitive state and their behaviour puts them in an advantageous position for meeting females that come to feed. The same argument is said to explain the paucity of females because they approach a moving bait only to feed and not to meet males. MORRIS (1960) attributed the success of his trap to its deceptive resemblance to a living animal and thought the flies it caught had come to feed; but SMITH & RENNISON (1961) concluded from their study of Morris trap samples that no such generalisation was possible.

Such ideas have generally been used to formulate explanations for differences between population samples taken under different environmental conditions. Thus a high female percentage is thought to indicate a hungry population and long ago FISKE (1920) produced evidence that removal of the fauna from a Lake Victoria island was followed by an increase in the proportion of females in samples of *G. fuscipes* caught by his assistants. Apart from JACKSON's (1933) studies little attention was given to the behaviour and characteristics of the individual insects making up a population sample. BURSELL & GLASGOW (1960) indeed considered separately *G. fuscipes* which were caught on man and those which were caught on the ground or vegetation, but did not record the perching positions of the former. However, FORD (1972) has shown that in this species the nutrition states differ between males perched "head-up" or "head-down" in the same way as with *G. morsitans*.

There are three reasons why an approach based upon observation and analysis of individual insects might be useful. One was noted in our introduction. The epidemiology of trypanosomiasis must be influenced by the biting rates to which the various hosts of the parasites are subjected. Secondly, such an analysis might confirm, or not, the conventional interpretations used to assess data from field surveys. Thirdly, there is a growing need to relate laboratory studies of tsetse behaviour to the pattern of behaviour observed in nature. Although, for example, GATEHOUSE (1972) is unable to perceive any relation between his observations on the different responses made by male and female teneral *G. morsitans* to olfactory stimuli in the laboratory and the departures from unity of the sex ratios observed in the field, the understanding of such relationships may not be very remote. What may be needed to achieve the connection is increased precision in field observation of (a) behaviour of individual insects and (b) behaviour of the bait or target. In this way data may be presented in terms similar to those used in the laboratory, where the responses of flies carefully standardized as to age and nutrition state are observed in relation to precisely defined stimuli.

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Zusammenfassung

Fünf Verhaltenstypen wurden bei Männchen von *Glossina morsitans morsitans* (Machado) (nach der ersten Blutmahlzeit) beobachtet. Die Glossinen wurden auf Fliegen-Runden («fly rounds») bei Ifakara in Zentral-Tanzania gesammelt.

Jede der verschiedenen Verhaltensweisen konnte durch Unterschiede im Ernährungszustand definiert werden. Letzterer ließ sich durch den Fettgehalt, den Haematingehalt, der auf die Häufigkeit leerer oder teilweise voller Därme hinwies, und die Unterschiede in der Zusammensetzung der Altersgruppen innerhalb der einzelnen Verhaltenskategorien bestimmen.

Der Ernährungszustand und das Alter von Fliegen, die während der Fahrt durch das Untersuchungsgebiet in den Landrover gelangten, werden ebenfalls beschrieben.

Ergebnisse mit Weibchen und frisch geschlüpften Fliegen ohne Blutmahlzeit werden diskutiert; sie scheinen nicht unwichtig zu sein für die Beurteilung des Verhaltens.

Eine Absicht der vorliegenden Arbeit ist es, Informationen zu liefern, die möglicherweise mit Ergebnissen aus Laborexperimenten über das Verhalten von Tsetsefliegen verglichen werden können.

Résumé

Cinq types de comportement ont été identifiés chez des mâles (ayant déjà pris leur premier repas sanguin) de *Glossina morsitans morsitans* (Machado) capturés dans la région d'Ifakara (Tanzania). Chaque type est caractérisé par des différences d'état nutritionnel (aprécié par mesure du contenu intestinal en liquide et en hémine) et par des différences dans les groupes d'âge prédominants. Ces caractéristiques ont aussi été étudiées chez les glossines capturées dans la Landrover lorsqu'elle traversait la région.

Des résultats obtenus chez les femelles et avant le premier repas sanguin sont également discutés; ils se revèlent ne pas être sans relation avec le comportement.

Un des objectifs de cette étude est de présenter des résultats sous une forme qui permette vraisemblablement d'effectuer des comparaisons avec ceux obtenus au laboratoire dans l'étude du comportement des glossines.