Onderstepoort Journal of Veterinary Science and Animat Industry, Volume II, Number 1, January, 1934.

The Trypanosome Infections of *Glossina pallidipes* in the Umfolosi Game Reserve, Zululand.

(Final Report.)

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INTRODUCTION.

A preliminary report on this subject was published in the 18th Report of the Director of Veterinary Services and Animal Industry Whitnall (1932). This report contained the percentages of proboscis infections found in *Glossina pallidipes* from several trapping sections of the Umfolosi Game Reserve, between November, 1931, and April, 1932.

In the present report the results of dissection during the period October, 1932, to June, 1933, are given, and a comparison is made of the infections obtained during this period, with those previously recorded. Attempts have been made to follow the possible effect of a reduction in the fly population by means of traps upon the trypanosome infections of the tsetse. The infections found among stray flies have also been briefly considered.

Data pertaining to the vertebrate hosts of the trypanosomes is mentioned in connection with the infections found in fly, the invertebrate hosts.

A short discussion is given upon a peculiar anatomical anomaly which has been noted in this fly.

Further, the salient points, resulting from an investigation into the rôle played by *Stomoxys* as a possible transmitter of Nagana, are recorded in an appendix.

GENERAL.

(1) *Material.*—Work of this nature is largely determined by such external factors, as weather conditions and the number of live flies obtainable.

In all cases the flies taken for dissection have been taken from a standard object, the "Harris" trap.

In the preliminary report, the flies were also taken from the "Harris" traps in the following sections of the Reserve: Siyembeni, A.5, Domba, A.4, Dadetu, A.3, Mhluzi, B.5, and Mbuzana, B. 1. In the present report the flies have been taken from the traps in Siyembeni, A.5, Mhluzi, B.5, and Dengeza, B.4 only.

The Sections A.5 and B.4 were selected for the following reasons: -(1) The nearest possible continuous data of infection could be obtained from A.5, a section in which trapping has been continuous. (2) In order that the possible effect of the diminution of the fly population upon infections might be followed. (3) Section B.4 was selected because the fly density was greater and material readily obtainable. As trapping had been abandoned since December, 1931, the flies from this section were used as a control against those from A.5, an operating section.

Harris (1932) mentions the unfortunate break in the trapping operations at this period.

(2) Condition of Flies.—The condition of the flies examined is based upon external observations and "feel". 50 per cent. to 60 per cent. of the females are regarded as pregnant, according to the distension of the abdomen. If, however, the females had been completely dissected and examined for developing larvae, it is felt that the percentage of pregnancies would have been higher.

It will be seen in Tables II and III that young flies pale in colour and soft to touch vary in prevalence from month to month.

(3) Sex Ratio.—The sex ratio in the various sections is given in Tables II, III and IV.

In all cases there is a predominance of females amongst the flies examined. This predominance is due to the fact that the traps have a greater attraction for females than for males.

Where fly is scarce the percentage of females appears to be relatively low (62 per cent. to 64 per cent.), while on the other hand, where fly is denser the percentage of females appears to be relatively high (71 per cent. to 77 per cent.).

DISCUSSION.

(1) A Comparison of the Infectivity of G. pallidipes over a Period of Two Years.

Table 1 gives a comparison between the two periods November, 1931, to April, 1932, and October, 1932, to June, 1933. The infections from the different sections are given in aggregate form. TABLE I.

The Tryanosome Infections Found in G. pallidipes from Various Localities in the Umfolosi Game Reserve.

Form.
[ggregate]
in A
Given

	.bəxiM	$\frac{0}{21.8}$	$12 \cdot 1$
	.bəxiM	$0 \cdot 74$	0.62
	T. brucei.	%	12 · 1
ed.	.isourd .T	0.24	0.62
s Infect	. sengolense. T	% 25+5	36.2
of Flie	.эsnэlognos .T	0.89	1.86
Percentage of Flies Infected.	T. vivax.	% 45·4	39.7
Per	.xpaia .T	1.55	$2 \cdot 04$
	Female.	3.46	4.45
	.əlsM	3.31	6.87
	.IstoT	3.42	5.14
ed.	Female.	41	36
Flies Infected.	Male.	14	22
Flie	.IstoT	55	58
	Female.	1,183	809
	Male.	423	320
	No. of Flies Examined.	1,606	1,129
	Date.	Nov., 1931, to April, 1932.	Oct., 1932, to June, 1933

A. B. M. WHITNALL.

The flies were collected over a wide area, extending from the junction of the Black and White Umfolosi Rivers (Section A.5), to the North-western Section of the Reserve (B.1).

Comparing the infections of the first period with those of the latter, the percentage of flies infected appears to have increased from 3.42 per cent. to 5.14 per cent. There also appears to have been an increase in the percentage of each sex infected.

In the first period the percentage of males and females infected was approximately the same $(3 \cdot 31 \text{ per cent.} \text{ males and } 3 \cdot 46 \text{ per cent.}$ females). In the latter period more males than females showed infection (6.87 per cent. males and $4 \cdot 45$ per cent. females).

An increase in the occurrence of each species of trypanosome appears also to have occurred. *T. vivax* was the predominating infection in the first period, representing 45 per cent. of the total infections, or 1.55 per cent. tsetses harboured this parasite. *T. congolense* represented 25 per cent. of the total infections, or 0.89 per cent. flies showed this infection.

T. vivax was still the predominating infection during the latter period, representing 39.7 per cent. of the total infections and occurring in 2.04 per cent. flies. T. congolense occurred in 1.86 per cent. flies.

It will be seen that infections of T. brucei appear also to have increased during the latter period from 7.6 per cent. to 12.1 per cent. of the total infections. The occurrence of this trypanosome in the fly increased from 0.24 per cent. to 0.62 per cent.

In the same period the percentage of mixed infections appears to have slightly decreased, i.e. from 0.74 per cent. to 0.62 per cent. flies infected. They have occurred as a combination of *T. vivax* and *T. congolense*, and *T. vivax* and *T. brucei*.

In considering the significance or interpretation of these results, consideration must be given to the influence of certain external factors. (1) For example, trapping of the flies has been carried on continuously in the eastern sections of the Reserve and has greatly reduced the fly population. (2) In the western sections trapping was abandoned from December, 1931. (3) Game and its movements is also a factor which must have great influence on the trypanosome infections of the fly. Unfortunately this factor cannot be accurately recorded even by continuous observation.

(2) A Comparison of the Infections in the Two Sections A.5 and B.4.

Due to the game reduction and trapping of the tsetse in the Umfolosi Game Reserve, no true infectivity of *G. pallidipes* under normal undisturbed conditions, is obtainable, for comparison.

An attempt can, however, be made to compare the infectivity of fly under the nearest possible normal conditions, with those which may be considered as decidedly abnormal. There was considerable variation in the local conditions in the two Sections A.5 and B.4. Section B.4 was in the abandoned area and consequently fly conditions were returning gradually to normal. Section A.5, on the other hand, was in the area of continuous operation and conditions regarding fly density were abnormal, due to intensive trapping.

The flies from Section B.4 were taken from three of the four check traps, which had been put into operation, for the purpose of gaining knowledge regarding the fly density in this section after abandonment. The fly was found to be relatively dense as compared with the density in the operating area (60.2 flies per trap per day, October, 1932).

Numerous field traps provided the flies from Section A.5. The fly density was very low (1.8 flies per trap per day, October, 1932).

The results of proboscis examinations from these two localities are given in Tables II and III. It is thus possible to make a comparison over the nearest possible similar periods of time.

The percentage of flies infected will be seen to vary in the two localities.

At Section B.4 during October, 1932, 7.6 per cent. flies were infected and at Section A.5, 3.5 per cent. In January, 1933, Section B.4 gave 5.2 per cent. flies infected, while Section A.5 gave 4.4 per cent. The infection for April was 5.8 per cent. and for May 8.5per cent. in Section B.4, while in Section A.5 it was 3.7 per cent. and 5.3 per cent. for the same periods.

Further, it will be seen in the tables that the occurrence of the different species of trypanosome appears to vary at the two localities.

The infection most commonly found in Section A.5 was T. congolense, while in Section B.4 T. vivax appeared to be most prevalent. In Section B.4 T. brucei frequently occurs, but this infection has only once been recorded from A.5.

A point of interest is the percentage of males and females which showed infection, and this also seems to vary over similar periods of time in the two localities. As the trapping of the fly might have had an influence on this aspect of the trypanosome infections, this variation has been discussed later with reference to one particular section.

It is remarkable that these variations have occurred in localities about five miles apart, and they appear to indicate amongst other things, that the flies do not pass with any regularity from one focus to another.

(3) VARIATION OF INFECTIVITY FROM MONTH TO MONTH.

An analysis of the results presented in Tables II and [1] indicates further that the infectivity varies in one locality from month to month. TABLE II.

The Proboscis Infections of G. pallidipes taken at Different Intervals from the Same Locality—Siyembeni, A.5.

lies		Female.					3.8		4.2	
Percentage Flies Infected.	nfeeted		9.4		$12 \cdot 5$		3-5		8.0	
Perce		.IstoT	ç. ç		4.4		3.7		5.3	
of		.bəxiM		1	[1.4	25
ected w urrence ceics of osome.		T, brucei.		1	i.	25	1]	1	1
% Files Infected with and % Occurrence of each Species of Trypanosome.		.92n9lognos .T	3.5	100	1-1	25	1.9	50	2.7	50
and %		.xosiv .'T	Ţ	1	5.5 75	50	1.9	50	1.4	25
		.IstoT	5		4		4		4	
	ed ions.	T. bruck and T. brucei.								
ions.	Mixed Infections.	T. vivax and T. congolense.	[1					
Infections.		T, brucei.					1		1	
		.əsnəlopnoə ,T	ro.		1		61		61	
	.xpaia .T		!		67		61		-	
	.gunoX		10.6		13.3		0.93		9.6	
		.səilf gano¥	15		12		-		7	
		.enales.	62 62	_	64		73		66	
		Females.	88		58		62		48	<u>.</u>
	.Rales.				32		29		25	
	Zo. of Flics Examined.				90		108		73	
	Locality.			A. 5	Siyembeni,	A. 5	Siyembeni,	A. 5	Siyembeni,	A. 5
	Date.		14/10/32	1/11/32	28/1/33	1/3/33	11/4/33	12/5/33	29/5/33	22/6/33

The Proboscis Infections of G. pallidipes taken at Different Intervals from the same Locality-Dengeza, B.4. TABLE III.

flics		Female,	9.8		4.6		5.6		4.3		6.7	
Percentage Flics Infected.		.9IsM	2.2		2.2		$4 \cdot 0$		10.7		14.3	
Percei		.fstoT	$2 \cdot 6$		4 · 0		5.2		5.8		8.5	
of		Mixed.	$9 \cdot 6$	30 20	9.6	$14 \cdot 3$	2.1	40	I]	3.4	40
% Flies Infected with and % Occurrence of each Species of Trypanosome.		T. brucei.		[9.0	14.3	$1 \cdot 0$	20	10. 10. 10.	13	1.7	20
Pilos Infected v % Occurrence Pach Species of Trypanosome.		. сопдоlепsе.	1.3	16.7	6. 5.	57.1	$1 \cdot 0$	20	-	1	1.7	20
% I %nd		.xviia .T	5.7	75	0.6	14 · 3	$1 \cdot 0$	20	3.3	22	1.7	20
		Total.	12		4		£		1-		1G	
	ed ions.	T. brucei.					1		1			
ions.	Mixed Infections.	T. vivax and T. congolense.	1		1		61		1		ଦ୍ୟ	
Infections.		T. brucei.			-		Ť		ŝ		μĬ	
		. чеполовиоз .Т	61		-		F-1		ļ			
		.xmaia .T	6	_	1				4		1	
.ganoY		$11^{0/3}$		2.9		6-2		0.83		1 . 7		
		.ssilā gauo¥	18		10		0		-			
		Females.	% 71		47		74		22		76	
		Females.	112		130		72		92		45	
Males.			46		45		25		28		14	
Xo. of Flies Examined.			158		175		97		120		59	
Locality.			Dengeza.	B. 4	Dengeza,	B. 4	Dengeza,	B, 4	Dengeza,	B. 4	Dengeza,	ρ
Date.			2/10/32	$\frac{11}{10/32}$	7/11/32	$^{ m to}_{ m 8/12/32}$	23/1/33	26/1/33	7/4/33	13/4/33	15/5/33	to

A. B. M. WHITNALL.

For example, at Section B.4 (Table III) for October and November, 1932, and for January, April and May, 1933, the percentages of flies infected were 7.6 per cent., 4.0 per cent., 5.2 per cent., 5.8 per cent., and 8.5 per cent., respectively.

There is further an apparent variation in the occurrence of the several trypanosomes. A predominance of T. vivax appears at certain periods, while at others T. congolense is more frequent. In regard to T. brucei it occurred from November, 1932, to June, 1933, in this section.

Similarly, at Section A.5 (Table II) there were fluctuations of infection from month to month.

From these results and those mentioned in the preliminary report, the proboscis infections of tsetses in Zululand appear to occur throughout the year, while fluctuating in frequency from month to month.

No attempt has been made in this work to determine the percentage of mature and immature infections. However, in 87 per cent. of the infected flies the hypopharynx was found positive, and from this it would seem safe to assume that a large majority of the infected flies harboured trypanosomes of the infective form, and were capable of transmitting the disease.

(4) Possible Effect of Reducing the Fly Population by Trapping upon the Trypanosome Infections.

Table IV gives the total percentage of infections discovered, together with the percentage of males and females infected, and also the percentage of females examined from Section A.5 for eight months.

The daily density per trap is also given for the months where records are obtainable.

In nature the percentage of males and females in *Glossina* is probably 50-50 per cent., yet the "Harris" trap continuously captures more females than males. Further, as the fly population dwindles, the percentage of females captured becomes less.

TABLE IV.

The Trypanosom	e Infections	of G. pal	lidipes fr	om One	Locality
Siyembeni A	.5, showing th	he Possible	Effect of	Reducing	the Fly
Population u	pon these In	fections.			

Date.	Females.	Percer	itage Flies In	fected.	Remarks.
	1 Onlines.	Males.	Females.	Total.	
12/12/31 to 17/12/31	% 68	4.2	2.0	2.7	Severe drought. Fly dense. 18·1 flies per trap per day.
24/1/32 to 3/2/32	82		3.7	3.0	Severe drought. Fly dense. No density records.
8/3/32 to 20/3/32	87	2.3	3.6	$3 \cdot 4$	Heavy rains, 20.–22 Feb. Fly dense. No records.
3/4/32 to 6/4/32	87		3 · 8	3.3	Fly dense. 5.4 flies per trap per day.
14/10/32 to 1/11/32	62	$9 \cdot 4$		3.5	Fly scarce. 1.8 flics per trap per day.
28/1/33 to 1/3/33	64	12.5		$4 \cdot 4$	Fly scarce. 0.4 flies per trap per day.
11/4/33 to 12/5/33	73	3.5	3.8	$3 \cdot 7$	Fly scarce. 0.4 flies per trap per day.
29/5/33 to 22/6/33	66	8.0	4.2	ŏ•3	Fly scarce. 0.4 flies per trap per day.

This is clearly shown in the table. During the first period (December, 1931, to April, 1932), fly was still comparatively dense in Section A.5 (18.1 flies per trap per day December, 1931) and the percentage of females was relatively high. In the latter period fly became scarcer (0.4 flies per trap per day June, 1933) as trapping proceeded and the female percentage dropped.

These facts appear to have an influence on the infections. During November, 1932, to June, 1933, when the fly population had been greatly reduced, those flies captured in Section A.5 showed abnormal conditions in regard to infection, assuming that the findings for the former period can be accepted as normal. When fly was comparatively dense in the Umfolosi Reserve, i.e. between November, 1931, and April, 1932, it was found that 3.31 per cent. males and 3.42 per cent. females were infected in 1,606 flies examined (see Table I).

Duke (1930) has shown that with G. palpalis and T. gambiense and T. rhodesiense there is a slightly higher percentage of females than males infected with the flagellates.

Thus the two sexes of *Glossina* appear to be equally susceptible to infection with Trypanosomes. Males and females live on the same food, and the chances of acquiring infection must therefore be equal.

Bearing in mind the above remarks, some facts of extraordinary interest appear in the table.

In March, 1932, when the fly population was comparatively dense (approximately 5.4 flies per trap per day) and the female percentage of flies dissected was 87 per cent., the infections were 2.3 per cent. males and 3.6 per cent. females. In April, 1932, with approximately similar density, the percentage of females examined was 87 per cent. No males were infected and 3.8 per cent. females.

In November, 1932, when the density had fallen to 1.8 flies per trap per day, the percentage of females examined was 62, and the infections were 9.4 per cent. males, but no females. In February, 1933, when the density had fallen as low as 0.4, 12.5 per cent. males were infected, but no females. Later in May the infections were 8.0 per cent. males and 4.2 per cent. females.

Similarly at Section B.4 as the fly population became less dense (60 flies per trap per day in October, 1933, to 17 in June, 1933, using four check traps as basis) so the percentage of males infected increased from $2 \cdot 2$ per cent. to $14 \cdot 3$ per cent. The percentage of females caught has, however, remained the same.

Minor exceptions occur, but the general tendency is as follows: As the fly population has become less dense, the percentage of flies infected has increased. The increase has occurred in the number of males infected.

The fly population at Section A.5 therefore, when greatly reduced by trapping, has a predominance of males with a high infectivity.

It is possible that this remarkable phenomenon might be due to the greater attraction of the traps for females, and in such cases young females would be picked up more rapidly than males. Thus the male portion of the population in a particular section might show higher infectivity than the female, due to the fact that the males when they happen to be caught, have had a longer period in which to acquire infection.

(5) STRAY FLIES AND THEIR INFECTIONS.

Three traps were erected in the southern buffer zone, within a radius of two miles from camp, to determine the density of fly in this locality. Some of the flies caught have been examined for infections.

It is of interest that the "Harris" trap was capable of catching tsetses where the population was extremely sparse. Doubtless the trap would be a most useful weapon when conducting a fly survey. Fly was present in small numbers in the vicinity of camp. They appeared to be the merest stragglers from the main body of fly and were only trapped at irregular intervals. From 21st November, 1932, to 10th February, 1933, the three traps caught twenty-five flies, fourteen males and eleven females.

Four females were pregnant, two in advanced stages. The occurrence of pregnant females which are about to deposit larvae, probably has some bearing upon the young flies amongst the stragglers. One tsetse, a young female, was caught at the Nseleni drift, eleven miles from the Game Reserve.

Eighteen flies have been dissected and examined from outside the Game Reserve and four were infected. The numbers are small, but in proportion the percentage infected appears high (22.2 per cent.). Once again, where the fly density was low the percentage of males infected was much greater than the percentage of females infected.

The infections found comprise three T. congolense and one T. brucci. It thus appears that both of the most pathogenic infections are being carried from their source, the Game Reserve. Though the numbers are small the fact of infected flies straying far afield is one of importance, and has some bearing upon outbreaks of Nagana.

TRYPANOSOMES OF CAME IN ZULULAND.

This work would hardly be complete without some mention of the available data pertaining to the vertebrate host of the trypanosome.

Four workers, Bruce (1895 and 1903) Mitchell (1914), Curson (1921) and Neitz (1931 and 1933 *) have investigated the reservoirs of Nagana infection amongst the game animals of Zululand. Neitz (1931) gives some details regarding the earlier investigators.

It is recent investigations which alone will be considered here, amount briefly to the following:----

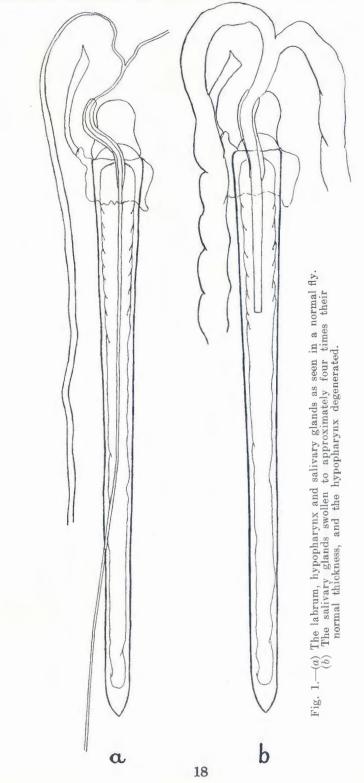
Two blood, two spleen, and two gland smears from each of 616 game animals have been examined microscopically for blood parasites.

These 616 animals, including zebra, bushbuck, duiker, reedbuck, kudu, warthog, steenbuck, klipspringer, blue wildebeest, waterbuck, and inyala, represent some of the 26,000 head which were shot during the period from May, 1929, to November, 1930, in the Umfolosi buffer zones (Harris, 1932).

Infection was found in seven instances. These comprised four T. vivax and three T. congolense, and were recorded from four bushbuck, two kudu, and one zebra.

Before considering these results emphasis must be laid on the following factors:—(1) The periodicity with which trypanosomes might appear in the peripheral blood. (2) The technique employed. (3) The time that expired after death before the slides were taken. These three factors have tremendous importance in regard to the frequency with which trypanosomes are encountered in stained preparations.

^{*} This paper is now in press.



The percentage of game infected, determined from the above fragmentory data appears to be 1.14 per cent., while the tsetse fly population on the average appears to be infected to the extent of 4.13 per cent.

The complicating factors are so numerous that no attempt will be made to correlate these happenings. Nevertheless it is of interest to note that T. vivax and T. congolense have been recorded from game, and the former appears to be more common. T. brucei has not been recorded since Bruce found it in 1895 and 1903, at which time the three species of trypanosomes had not been differentiated.

In the fly T. vivax and T. congolense frequently occur and the former is the most common. T. brucei, on the other hand, is a fairly rare parasite.

AN ANATOMICAL ANOMALY IN "C. PALLIDIPES".

While engaged on fly dissection a peculiar anomaly in the structure of the salivary glands and hypopharynx of this tsetse was noted. In 1,129 flies examined, 32, that is 2.8 per cent., showed a distinct thickening of the salivary glands. It was most noticeable and was detected as soon as the salivary glands were displayed. Furthermore, in each case where the salivary glands were thickened the hypopharynx was invariably degenerated.

Normally, the hypopharynx, a continuation of the salivary duct, is a long slender tube extending beyond the tip of the labrum, while the salivary glands are slender thread-like organs. Fig. 1 (a).

In the cases now under discussion the glands may be swollen to more than four times their normal thickness, and show convulutions, without sign of infection. The hypopharynx is broad and stumpy, being approximately three or four times the normal breadth, and sometimes less than one-tenth of the normal length. Fig. 1 (b).

The anomaly has been found in males, females and young flies. That such flies can feed, and live for some time, is borne out by the fact that mature trypanosome infections of T. brucei have been found in some of them, indicating that the flies have lived at least three weeks.

The point of interest arises in connection with the trypanosome infections.

Lloyd and Johnson have stated that in the case of T. brucei the hypopharynx is used only as a passage for mature trypanosomes from the salivary glands. With T. congolense and T. vivax pre-infective forms are said to enter the hypopharynx and there become infective. The infective forms accumulate.

Seven infections have been found in the thirty-two cases of this anomaly; three of the proboscis group (T. vivax); one of the proboscis and gut group (T. congolense) and three of the proboscis, gut and salivary gland group (T. brucei). In no case, however, were trypanosomes seen in the degenerated hypopharynx, and the question appears to arise as to whether such flies are capable of transmitting infection.

There also appears to be some doubt as to the passage taken by the trypanosomes in passing from the labial cavity to the hypopharynx.

Some workers maintain that the trypanosomes enter the hypopharynx through a slit in its wall at the proximal end. Authorities, however, are by no means agreed that such an opening exists. If the hypopharynx has no such opening, then it would seem that the trypanosomes enter at the opening at the extremity of the proboscis.

When the hypopharynx is degenerated as above described, the passage of the trypanosomes of the T. *brucei* group from the gut to the labial cavity, and thence through the tip of the hypopharynx to the salivary glands is easily conceivable. That the trypanosomes of the T. *brucei* group enter the salivary glands through the tip of the hypopharynx seems to be further indicated by the appearance of the long slender proventricular forms at its tip and middle, in normal flies infected with T. *brucei*.

With regard to the transmission of the trypanosomes by tsetses with a degenerated hypopharynx, it is suggested that in the case of T. brucei (where this organ is used as a passage only) that such flies could transmit trypanosomes. The normal passage through the hypopharynx would be replaced by the tube formed by the labium and labrum in apposition. Along this both saliva and the mature trypanosomes would pass into the wound when the proboscis punctures the skin.

The infections of the swollen glands were phenomenal, as both glands, throughout their lengths were swarming with trypanosomes, among which were infective forms.

In the case of T. vivax and T. congolense, where a definite phase of development of the trypanosome takes place in the hypopharynx, it would seem that in flies in which this organ is degenerated, the infection could not mature, and therefore could not be transmitted.

APPENDIX.

STOMOXYS-A POSSIBLE TRANSMITTER OF NAGANA.

A severe outbreak of Nagana occurred at the Ntambanana Settlement during the winter of 1932, and complaints were made by the farmers that biting flies were a great pest to the cattle.

Acting on the instructions of the Director of Veterinary Services, the possible rôle played by these biting flies as a transmitting agent of Nagana, was investigated during July, August and September.

The results obtained amount briefly to the following: -

(1) With the aid of the "Harris" trap, *Stomoxys* was shown to be present in enormous numbers at different farms in the Settlement. The species of *Stomoxys* captured were identified as *S. calcitrans*, *S. nigra*, *S. brunnipes* and two species which have not been identified.

The flies were persistent in their attacks and were a great pest to the cattle.

- (2) Positive cases of Nagana (T. congolense and T. brucei)were found among the cattle in the Settlement. It is possible that these constituted a focus from which infection was spreading.
- (3) Stomoxys, dissected soon after feeding on a donkey, whose blood showed very few trypanosomes, were negative in both proboscis and gut.
- (4) When there were very numerous trypanosomes in the peripheral blood of the donkey, the flies readily took up the infection.
- (5) The longest period that trypanosomes (T. congolense) remained active in the gut of *Stomoxys* was twenty hours.
- (6) Eight per cent. of *Stomoxys* interrupted while feeding on a donkey, heavily infected with *T. brucei*, were positive in both gut and proboscis.
- (7) From this it seems possible that trypanosomes may be conveyed from infected to healthy animals in a mechanical manner by *Stomoxys*, provided that: (a) *Stomoxys* is present in large numbers. (b) Infected and clean animals are in close contact. (c) Very numerous trypanosomes are present in the peripheral blood of the infected animals.
- (8) No Glossinae were encountered, but no definite search was made for this fly

ACKNOWLEDGMENTS.

In conclusion, I wish to thank Dr. P. J. du Toit for all facilities and for his encouragement and interest in the work. Mr. R. H. T. P. Harris has at all times given advice and assistance, and to him I am particularly grateful. To Mr. W. Foster, with his knowledge and experience of the game country, I am obliged for much help and many kindnesses.

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