

# A REPORT ON TRYPANOSOMIASIS OF DOMESTIC STOCK IN NORTH-WESTERN RHODESIA

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The Expedition reached Broken Hill, the present head of the Cape to Cairo Railway (approximately  $14^{\circ} 35' S.$ ,  $28^{\circ} 40' E.$ ) on June 17th; on the 23rd the examination of some cattle at a farm in the neighbourhood revealed the presence of trypanosomes.

## LOCAL VIEWS ON THE DISEASE

'Fly disease,' 'fly struck' or 'fly' is well known to the inhabitants of the district, and most deaths in cattle are ascribed to it. The views held locally are those of stock owners South of the Zambesi. The passage of cattle through a known tsetse area is only undertaken under compulsion, and wherever possible is effected during the night,

and it is considered that the bite of more than one fly is usually necessary to give the disease.

Cattle are held to be most susceptible, but cases are recorded of animals which do well and breed in villages situated within fly areas. There is a fairly general impression that animals born in the fly area possess a degree of immunity not enjoyed by others, and this is held to account for the herds which some native chiefs are said to own. We have seen such a village near the River Kafue, and have taken *Glossina morsitans* within a mile and a half of it. The chief of this village regards the freedom of their cattle from the disease as being due to care in herding, and admits that if they were taken away from the old garden clearings except at night that sickness would be expected. Another village was frequently quoted as an instance of this inherited immunity, but on arrival there we found that within the past two years most of the cattle had died of fly disease, and the rest, which had been sold to a European, had also succumbed. We only met with one case which would tend to bear out this suggestion of local insusceptibility.

Sixteen head of yearling stock were purchased from a chief near the Kafue in 1902. These animals had been bred there and *Gl. morsitans* is found within half a mile. For three years they were kept on the purchaser's farm, within a few hundred yards of tsetse flies, during which time one animal died suddenly, and six were sold fat for slaughter. In 1905 the remainder were moved twenty miles down the river, and during the rains of 1906-1907 six deaths occurred. Five of these had given birth to calves, and these, with the rest of the cows, appeared in excellent condition when examined in November, 1907. These cattle had spent their lives within a mile of *Gl. morsitans* which had frequently been caught feeding on them. Game is plentiful around. Bushbuck, puku, waterbuck, and hartebeest have often been seen grazing a few hundred yards from the homestead.

The susceptibility of the mule and donkey is disputed. A team of the former and some donkeys were kept for transport near Broken Hill, and are said to have frequently been in contact with the tsetse, but we were not informed of any deaths which could be directly attributed to this cause, and an examination of eight of these mules did not show trypanosomes. We received the histories of three donkeys which are said to have lived in fly areas for from one to three years without ill effect. One of these was later shown to be susceptible to infection by *T. dimorphon*. A second was taken into a badly infested district in August under the belief that it was immune. Its blood was examined in November by a layman, who, however, had a good deal of experience in blood examinations; and

it was stated to have trypanosomes. Fifteen days later it appeared in good health, and we were unable to verify the finding in the single observation made. Dr. Yale Massey showed us films of *T. dimorphon* made from the blood of a donkey which died of the disease at Ruwe in the Congo Free State.

Goats are considered as immune by Europeans and natives alike. This view is held to such an extent that they will graze a herd of these animals over land intended for occupation in order to drive away the tsetse, which is said to be not only harmless to them, but also to be repelled by the odoriferous nature of the adult males. We were informed of many instances where goats had lived and bred within fly areas. From one such area a European purchased forty head in February, 1907. They commenced to die immediately at the rate of one or two a week, the symptoms being emaciation lasting for one or two months. In November three adults (all that remained), each of which had a kid, were examined with negative result.

Native dogs are regarded as immune, and it is stated that natives will expose their dogs, a valuable asset to them, without fear. We have seen three cases of natural infection in 'essential kaffirs.' English and Colonial dogs are considered susceptible; but if born or bred in the fly district their powers of resistance are said to be increased.

No special symptoms are recognised. Gradual and progressive emaciation; periods of manifest depression followed by others in which the animal appears brighter; lacrimation and nasal discharge; these, taken with a history of passage through a fly district, are held to be sufficient for diagnosis. Oedema, enlargement of the superficial glands or paresis, are not mentioned. On autopsy an oedematous condition of the connective tissues and a paleness of the muscles are said to be constant; the presence of fluid in the body cavities has not always been noted by stock owners.

Deaths are said to be more frequent just after the break of the rains, and it is considered that any undue exposure of an animal to water will 'bring out' the latent disease; so much is this held that we have been informed of cattle being purchased subject to a test of pouring a bucket of water over them. Should they not show evident signs of sickness within a few days, they are considered as free from 'fly.' Our observations at Broken Hill, and since, do not

bear out the interpretation of this seasonal prevalence given by owners, most of whom think that the deaths will occur soon after the commencement of the rains irrespective of the date of infection. The more reasonable explanation would be that as waggon transport is impossible except in the dry season, the infection takes place between May and October; and since the disease is of a rapidly fatal character (one to five months), deaths will normally occur about November at the time rains are expected to break. Further, it is obvious that any debilitating influence, and particularly this sudden climatic change, will act as an exciting agent and hasten a fatal termination.

The ordinary method of prevention consists in avoiding tsetse country; where this is impossible, animals are driven through at night, and are kept on an open grass plain during the day, for owners have noticed that the tsetse (*G. morsitans*) frequents by choice the bush. A native method of prophylaxis exists, and it appears to have also been used as a curative. Previous to entering a fly area the water of the cattle is restricted so as to compel them to drink a bitter decoction made from the bark of a tree (*Kangomba*) in which the body of one tsetse is placed. At night the animals are kept in a hut and a fire made of the young twigs and leaves of a bush (*m'safwa*), in the smoke of which they remain till morning. The effects of this treatment are held to last for three to four days, but daily adoption is recommended. One European expressed some faith in it, having twice taken an animal into a tsetse zone after fumigation. Another put the matter to a more thorough test, and after medication sent three cattle into a fly area; they all died within six months. As a preventative, fumigation may be to a degree efficacious, for the presence of an obnoxious agent would certainly tend to repel the attacks of the fly; but we question the correctness of the diagnosis in those cases, and they are admittedly few, in which a cure is said to have resulted.

With one exception, cattle owners European and native, have incriminated the tsetse to the exclusion of all other biting flies, and the views common in South Africa regarding the association of *Glossina* and game are also held, but to a more limited and disputed extent. The exception was a native of North-Eastern Rhodesia, whose father, one of the greatest chiefs on the Western side of the

territory, had lost some eighty head of cattle within a year. This man caught us a *Tabanus*, closely allied to *T. dorsivitta*, Walk., and stated that his people believed it to be the cause of the disease. While this is no proof, the spontaneity of the act indicated these natives' belief in the statement, which coming from such a source is the more remarkable, as ordinarily they might be expected to blame the recognised enemy, the tsetse.

#### DOMESTIC ANIMALS IN THE TERRITORY

These observations refer to that part of North-Western Rhodesia which lies between 28° 40' East Longitude and the River Kafue, and more particularly to that area in which one of us travelled.

Cattle breeding is extensively carried on by the Mashakalumbwe people, who occupy the North bank of the Kafue as it runs Eastwards to join the Zambesi. Isolated members of the same tribe living to the North of the rest are also cattle owners; the other tribes are not cattle-men, though a few of the largest chiefs whose villages are marked on the map possess a few cows in the kraal. If they own any more, these are quartered out with other chiefs resident in fly-free districts. It would appear that even these few are discontinuing the custom, partly owing to the losses they sustain from the tsetse, and perhaps also because with the presence of Europeans the prices of cattle have risen, and they will no longer replace losses. As practically the whole area North of a line drawn from the South-Western corner of the 'Hook of the Kafue' to the South-Eastern boundary of the Congo Free State is infested by *Glossina morsitans*, it is understood that cattle-raising to any extent would be impossible, and that its absence is not entirely due to the indolence of the native. We are informed that in 'the old days' cattle breeding was extensively carried on here, and that the spread of the tsetse, the epidemic of rinderpest, and the trihal raids that took place before the British entered into possession, have brought about its cessation.

European settlers own one to three spans, each of sixteen to eighteen head, for agricultural work; but the opening up of mines, and the lack of other forms of transport, have caused these animals to be taken from the farms and utilised for this purpose. Cows are scarce, and are now obtained, together with the working bullocks, from the Mashakalumbwe on the Kafue, or from the Barotse Valley



on the Western side of the British sphere of influence, which is said to be free from fly and an excellent cattle country. There are a few animals of the Zebu type coming from German East Africa, or from the Ngoni Country to the East of Rhodesia.

Goats are kept at most of the larger villages, which are relatively few in number. They are of a very inferior strain, small and undersized, and would appear to have been in-bred for generations. Those in the hands of Europeans are somewhat improved, and the rams have been imported from the better stock-raising districts South of the Kafue.

Sheep in North-Western Rhodesia are, so far as we are aware, owned mainly by settlers. In North-Eastern Rhodesia, however, natives own large flocks, mostly small ill-shapen animals, showing but an element of Persian blood. Excepting for the smaller development of their tails, which hang straight at the tip, they are not unlike the Indian *dumbah*.

Equines are limited to a few Europeans. They came from South of the Zambesi, and some of the donkeys from German East Africa.

#### TRYPANOSOMIASIS IN CATTLE

Our present observations date from June 23rd, between which date and September 30th, 1907, experimental work on the morphology of the parasites and the curative influence of atoxyl and mercury was conducted. Since October we have been travelling continuously.

This work was only rendered possible by the kindness of two agriculturists whose animals were affected Messrs. J. F. F. Johnson and F. C. Miles—who permitted us to make what use we could of their sick spans. Much of the experimental work on pathogenicity is due to the liberality of the Administration, The British South Africa Company, which, in conjunction with Mr. H. U. Moffat, the Superintendent of the Bechuanaland Exploration Company, placed a sum of money at our disposal for the purchase of the more expensive animals. To the Administrator himself, Mr. R. Codrington, and to Mr. Moffat, it is difficult to adequately express our indebtedness for the constant and continuous interest they manifested in the work. Observations of the effects of the trypanosomes met with upon the usual laboratory animals was only possible

through the kindness and generosity of Dr. Arnold Theiler, C.M.G., the Veterinary Bacteriologist to the Transvaal Government at Pretoria, and of Dr. Robertson in charge of the laboratory of the Medical Officer of Health at Cape Town. Finally, our thanks are due to every European in the neighbourhood for the constant courtesy they extended, and for the many demands made on their time in supplying us with the result of their experiences; to the agriculturists who, without exception, placed their animals at our disposal for examination; and to the various Government Officials who did all in their power to forward the research.

We established a temporary camp and laboratory near Broken Hill, and during our stay there, 36 cases of trypanosomiasis in cattle were detected, of which 29 were under continuous observation. This work was discontinued on September 30th, in order to prosecute our enquiries further North in accordance with the scheme drawn up before leaving England.

The history of many of the 36 cases was uncertain, save that at some period within the past six months they had been in a fly district. One herd, however, possessed peculiar interest as indicating in the most convincing circumstantial manner that biting flies other than *Glossina* can transmit infection. This will be discussed under 'Transmission.'

We have already noted that with the opening up of mines, cattle previously and primarily intended for agricultural work have been utilised for transport, and on *primâ facie* grounds this exposes them to greater risks of infection by bringing them into contact with the tsetse's haunts. At the present time one of the four 'roads' from Broken Hill must be used for waggon transport. The two which run North enter a fly district about fourteen miles out; the third and most frequently used one passes through a patch in which tsetse are generally seen, about eight miles South, but being a narrow zone, night marching may avoid attack. The fourth runs for 112 miles South-West to the copper properties there; only within this last year have tsetse been found encroaching at about the 100th mile. This road joins that coming North from Kalomo, which was used prior to the railway. From the neighbourhood of the junction a road runs North to the copper mines, and is in fly-belts most of the way, and a new one has just been cut in a South-Easterly direction to join the

railway some 30 miles North of the Kafue. This runs most of the way on a watershed; *Gl. morsitans* has not yet been seen, and the nature of the country is not indicative of its presence, but on all the other roads the danger of infection is constant, and when the Northern ones are used it is recognised that all cattle must be sacrificed.

In the first herd examined the trypanosomes seen in fresh cover-glass preparations appeared to be of two varieties. One, the prevailing type also found in other herds, was seen to possess the morphological features of *T. dimorphon*, Dutton and Todd. The second was seen in four of these animals, and later in a fifth coming from another herd, and in two cattle which had been exposed to tsetse-flies experimentally. This, by reason of its extraordinary rapidity of motion in cover-glass preparation, is regarded as allied to *T. vivax*, Ziemann.

#### INFECTION WITH *T. DIMORPHON*

(1) The natural disease in Cattle.—There is nothing in the clinical picture of this disease to differentiate it in any way from other forms of trypanosomiasis. At some period there is an appearance of emaciation and dulness, the coat harsh and hide-bound, head drooping, eyes dull and watery, but petechiae on the conjunctival membranes were very rarely noted, and occasionally there is a nasal discharge. Weakness or paresis of the hind limbs is not common, and oedema was not seen. Enlargement of the lymphatic glands, notably the prescapular and precrural, is constant, but is of little diagnostic importance owing to its prevalence in apparently healthy animals in which trypanosomes could not be found on blood examination or gland puncture.

All animals under observation were placed at night into the ordinary cattle kraal of the country—an open enclosure fenced to a height of seven or eight feet to avoid the attacks of wild beasts—and were allowed out to graze all day. Temperatures were taken and blood examinations commenced between 8 and 11 a.m., and the temperature taken in the evening between 4-30 p.m. and sunset. For these operations the animals were brought into a kraal-like enclosure leading to a 'crush,' into which each was in turn driven. After the



first day the cattle accommodated themselves to this method, which cannot be held as interfering with the proper thermometric registration. Under these conditions the normal temperature of healthy cattle was found to be approximately  $100^{\circ}$  to  $102^{\circ}$  F., two degrees representing the normal diurnal variation.

During the course of the disease the temperature was almost constantly elevated; to a slight degree it was paroxysmal, but, as the accompanying charts show, this was not a marked feature.

Parasites could usually be found on direct examination of peripheral blood. For graphic representation a similar system to that adopted by Lingard is used, but the number of trypanosomes present is so much less (12 to a field (Zeiss Oc IV, Obj. D) was the greatest number seen) that lower values are accorded to each mark. When numerous it was customary to count forty fields and take the average; but when scanty sixty to a hundred, according to indications. No animal is marked '*absent*' unless this latter number was counted; and in necessary cases, as in the treatment experiments, a  $\frac{3}{4}$ -inch square cover-glass was searched before placing a minus sign.

There is no close relationship between the temperature and the number of parasites seen; sometimes a temperature of  $104^{\circ}$  or  $105^{\circ}$  F. was unaccompanied by trypanosomes, or only one to twenty fields; and again, a temperature of  $101^{\circ}$  has been seen with four organisms to a field. Two cases were observed in which organisms were not seen for two or three weeks, and in one (No. XIII) which was diagnosed on July 18th, trypanosomes were only seen on August 10th (one to a cover-glass during the ten weeks the animal remained under observation). These might be considered as chronic or latent infections, though both died, apparently within the usual time of the disease.

Gland puncture of the prescapular lymphatics was tried as an additional aid to diagnosis, though its general employment is not so generally necessary owing to the fairly constant presence of trypanosomes in the peripheral blood. The method is essentially that described by Dutton and Todd for the diagnosis of human trypanosomiasis, all specimens being sealed with vaseline and examined immediately. It will be seen from the figures below that it is of less value than blood examination as a means of diagnosis, but its

employment is advisable before any suspected animal be considered free from the disease.

	Animals in which tryps. were found in peripheral blood	Animals not showing tryps. peripherally
Number of glands punctured	... 26	... 33
Tryps. seen in gland juice	... 19	... 1

From a few observations made in India on camels suffering from Surra we considered that this method was a valuable aid to diagnosis, and in this respect we agree with the views of Dutton and Todd,<sup>4</sup> who used it in the detection of trypanosomiasis in cattle of the Congo; but in Rhodesia our results so far lead us to consider this method as of little diagnostic importance.

The duration of the disease cannot be definitely asserted, but from the views held by local stock-owners and our own observations on animals whose histories are fairly complete, and on experimental cattle, from one to five months would appear to represent the normal. In fourteen of our cases the time which elapsed between diagnosis and death averaged thirty days, and many of these animals were in excellent condition and could not be suspected clinically. It seems probable that in a few cattle a chronic form is established; of this we have no direct evidence, but the histories of some stock examined, and the occurrence of two herds in which a large percentage were clinically cases of trypanosomiasis, but which did not show organisms, would indicate the possibility. In one animal of this nature (No. XXVII) every sign of the disease from a clinical standpoint was shown, but trypanosomes only appeared five days previous to death, after an absence of 15 days.

The lesions observable *post-mortem* are those of an emaciating disease—paleness of the visible mucous membranes, flaccidity and pallor of the muscles. The amount of fluid in the body cavities varied. In some cases it was practically *nil*, whilst in one animal approximately 1750 c.c. were obtained from the peritoneal cavity. Petechiae are present on most of the serous surfaces, notably that of the spleen. The blood clots to a considerable degree, and the white clots common in equines with surra were not seen. All lymphatic glands are enlarged, particularly the precrucial, and some of the mesenteric, which are also frequently haemorrhagic. Enlargement

of the spleen is inconstant, but it is usually friable with prominent malpighian bodies, and the capsule studded with petechiae. In only one case, No. XLI, showing marked nervous symptoms, were any gross changes seen in the spinal cord.

Trypanosomes were not constantly seen in the blood at death, nor were films, made direct from the lymphatic glands, invariably positive. Those, however, made from the haemorrhagic lymphatics of the mesentery possessed greater diagnostic value than the others, and owing to the danger of a negative diagnosis being made on a blood film sent in for opinion, we would recommend the forwarding of one made from a haemorrhagic gland in addition, though it must be understood that it is necessary for this to be made as quickly as possible after death in order to avoid undue phagocytosis.

These cattle, compared with those of India, are singularly free from organic changes in any organ, and from other animal parasites. In most (16 in 22) there were a few *Filaria papillosa*, and in many (10 in 22) *Paramphistomum conicum* was seen. *Distomum hepaticum* was not encountered despite the prevalent idea that 'fluke' occurs here. No filarial embryos were seen in the blood, and *Piroplasma bovis* was only seen on one occasion in two animals.

(2) The experimental disease.—The strain of *T. dimorphon* mainly used in these experiments was derived from a naturally infected cow, Case No. XXV. This animal was selected as representative of the disease; as she had been confined to the homestead and had not travelled on the surrounding roads, the danger of a mixed infection being thereby reduced. Though apparently in perfect health, she had, owing to her contact with the sick, been subjected to four examinations between June 26th and July 26th, when she was brought in for daily observation. Trypanosomes were first seen on July 29th, and it is believed that she contracted the disease on the farm. The same strain passed through Case No. XLII served for most of the other inoculations.

1. CATTLE. Both the animals inoculated with virulent blood were later utilised in the experimental work on treatment, which was regarded as of more direct importance. Organisms appeared on the seventh and eighth day following inoculation, and remained constant until the exhibition of atoxyl, in one case, No. XLII, for 24 days. Death in the case of No. XLI was largely due to atoxyl-intoxication.

The other animal is stated to have died suddenly 63 days after treatment was commenced, and 87 days after inoculation.

CASE No. XLI.—August 20th. Inoculated subcutaneously with 2.0 c.c. citrated blood of XXV. Trypanosomes first seen August 27th, one to a cover-glass. Temperature remained but slightly elevated till the morning of the 28th, when it rose to 104° F., and was 105° the same evening. Organisms increased in numbers up to the 30th (three to a field) when 5.0 grm. atoxyl were administered and the normal disease no longer continued.

CASE No. XLII.—August 20th. Inoculated as above. The temperature on the 27th evening was 104° F., and the following morning trypanosomes were seen, 1 in 2 fields. These remained constantly present and in considerable numbers for 24 days. During this period the temperature was between 102° a.m. and 106.2° p.m. the animal rapidly lost condition, and showed evident clinical manifestations of the disease, and we consider that if treatment had not been attempted death would have occurred within two or three weeks at the outside. (vide chart i.)

Two other cattle, Cases Nos. XLIV and XLV, which were infected by exposure to the bites of *Glossina morsitans* are described later under 'Transmission.'

## 2. DONKEY.

CASE No. LV.—This animal is said to have lived in fly districts for upwards of three years and was considered immune. September 3rd, inoculated subcutaneously with 2.0 c.c. citrated blood of XLII. During the four weeks it remained under observation there was no change from normal in its temperature and daily direct examination of blood and three centrifugal examinations did not reveal organisms.

On November 20th it was again examined and *T. dimorphon* found, three to a cover-glass. It did not at this time show any clinical indications of the disease. Since that date we have not received any report as to its condition.

## 3. SHEEP.

The disease in these animals and goats is of considerable interest, for in the quite characteristic temperature chart we have evidence for distinguishing this trypanosome from that which we hold resembles *T. vivax*, Ziemann. Three sheep were inoculated; in two healthy animals the incubation was seven and eight days, in a third, which had passed through a previous infection by the other trypanosome, it was eleven days.

In both healthy animals the temperature assumed at first the type produced by the benign tertian form of malarial parasite in man, later giving place to the quartan form. The regularity of the fever is striking, as also is the fact that each exacerbation was accompanied by an influx of trypanosomes into the peripheral circulation. In order to demonstrate this periodicity of trypanosomes and temperature, these animals were examined every three hours for a period of 56 hours, for as we are unaware of any other form of trypanosomiasis in which this feature is so pronounced, an effort was made to ascertain, if possible, any developmental cycle undergone by the



parasite in the sheep. The time at our disposal, however, was not sufficient for a careful study of the question, and our observations were without definite results.

No special symptoms were noted. During the course of the disease a rapid emaciation took place, but all the animals were alive when the work was concluded at the end of September. Each access of fever was manifested clinically by the weakness, depression and excessive lachrymation shown by the animal. Oedema was not seen in either these animals or goats.

Parasites were never numerous: on one occasion only were as many as six to a field seen. The number, however, depends upon the period in the onset or decline of the paroxysm at which the routine examination happened to be made.

CASE No. XXXVI.—An aged ram, fat-tailed variety, purchased locally, but probably imported from the South. August 7th, inoculated subcutaneously with 5.0 c.c. citrated blood of XXV. The temperature became irregular on the 5th, and organisms were first seen on the eighth day, August 15th. For the first fourteen days the paroxysms were tertian in type, afterwards becoming less regular and approaching the quartan type. Emaciation was rapid, but the animal was still alive on November 20th, and organisms were present. (vide chart iii.)

CASE No. LVI.—Two-year-old female. September 3rd, inoculated subcutaneously with 1.0 c.c. citrated blood of XLII. Organisms first seen on the 7th day, September 10th, and the initial thermal paroxysm occurred during the night of the 11th. On the evenings of September 14th, 16th, 18th, 20th, 22nd, and 24th, the temperature exceeded 106° F., and on the 28th it reached 107°. During the twenty-one days of observed disease the type was essentially the same as in No. XXXVI, the chart of which is reproduced.

CASE No. XXXIII.—Male, aged one year. Between August 19th and September 2nd this animal had shown what we regard as *T. vivax* with which it had been inoculated. After this latter date the temperature remained about normal and trypanosomes were not seen.

September 17th, received subcutaneously 0.5 c.c. heart blood of guinea-pig, Case XLVI, dead with *T. dimorphon*. The temperature was fairly constant until the 27th, when it commenced to rise. On the 28th, the 11th day, *T. dimorphon* was seen, the first paroxysm occurring the same day, the second (107.2°) on the evening of the 30th, the last day of observation. (vide chart ix.)

4. GOATS. These animals, as already stated, are regarded as immune. Three readily took infection with *T. dimorphon* after an incubative period of seven to twelve days. The disease is of essentially the same type as that in sheep, viz., a tertian and quartan febrile reaction and a concomitant influx of organisms into the peripheral circulation. No special symptoms were observed.

CASE No. XXXV.—Male, aged one year. August 7th. Inoculated under the skin with 5.0 c.c. citrated blood of XXV. The temperature became irregular until organisms appeared on the seventh day, August 14th. During the seven weeks



of disease in which this animal was observed, paroxysms occurred with the regularity of those in Sheep, only the thermal reaction being more pronounced— $108.2^{\circ}$  F. on one occasion. Emaciation was rapid, and the animal died in the interval of one and a half months that elapsed previous to November 20th. An exact date was unobtainable as no European had been on the farm. (vide chart iv.)

CASE No. LVII.—Male, aged two years. September 3rd inoculated subcutaneously with 1.0 c.c. citrated blood of XI.II. Organisms were first seen on the ninth day, September 12th, and paroxysms occurred on the 15th, 17th, 18th, 20th, 22nd, 24th, 26th, 28th, and 30th. (vide chart v.)

CASE No. XL.—Young male. Inoculated on August 9th with blood of Goat, No. XI, containing *T. vivax*. Trypanosomes were never seen, and on September 17th it received 0.5 c.c. heart blood of guinea-pig, No. XI.VI, dead of *T. dimorphon*.

On September 29th, the twelfth day, *T. dimorphon* was seen, and again the following day the last of observation. On November 20th parasites were still present, and the animal had lost much of its condition.

5. DOGS. Three native dogs were inoculated with this strain of *T. dimorphon*, and despite the suggested immunity of the race, all took the infection readily, and we shall later note that naturally infected native dogs were met with, in which, however, the parasite did not show the morphological characteristics of *T. dimorphon*.

In all three, the disease was acute; death taking place within two weeks of the appearance of organisms in an adult, and within ten days in young animals of three months old.

Emaciation is rapid; the coat becomes harsh, eyes lachrymose, and in the adult, opacity of the cornea was noted three days before death. Superficial lymphatic glands, notably the prescapular, become enlarged and soft, but not painful; a feature, however, noted in certain apparently healthy dogs. Only one gland puncture was made in the case of an adult, when organisms were not seen in the peripheral blood; but its employment did not reveal trypanosomes. Nervous symptoms were noted in one young dog, No. LIV, which showed partial incoordination of the hind limbs on the day preceding death. The incubation period was sixteen days in adult, and seven and eleven respectively in young animals. The temperature curves of these latter are most irregular; hardly ever elevated, but showing a tendency to become sub-normal. In the older dog the type is that of other trypanosomiases, showing a close resemblance to many charts of canines infected with *T. evansi*. Organisms increased progressively up to death. In the adult the temperature and trypanosome curves synchronised.

CASE No. XXXIV.—A 'kaffir' bitch, aged about two years. August 7th, inoculated subcutaneously with 1.0 c.c. citrated blood of XXV. The temperature

remained normal until the morning of the 21st when it commenced to rise, continuing to do so gradually till the morning of the 26th when it was 105° F. That evening it was 102°, and it kept normal for three days to rise again for the second paroxysm on the 30th. Trypanosomes appeared on the third day after the temperature rose, August 23rd, one to a cover-glass, and increased progressively to the 25th, one to six fields. On the 26th, 27th, 28th, they were not seen; they reappeared on the 29th, and were twelve to a field on September 1st. They gradually fell in number towards death, which took place on the night of the 4th. (vide chart vi.)

CASE No. XLIII.—A 'kaffir' pup, aged three months. August 20th, inoculated 0.5 c.c. citrated blood of XXV. The temperature was very irregular throughout, varying from 96.4° to 103.8°. Trypanosomes appeared on the seventh day, and remained constantly present till death on the fifteenth day. During the last three days they were very numerous, ten to a field, and the temperature was usually subnormal.

CASE No. LIV.—Brother to the last. September 8th, inoculated subcutaneously with 1.0 c.c. citrated blood of XLII. The temperature showed considerable daily variation. Organisms appeared on the eleventh day, September 14th, one to five fields, and remained present in slightly increasing numbers, till death on the eighteenth day after inoculation, September 21st.

*Post-mortem appearances.*—In all three animals the most noticeable feature was a considerable enlargement of the spleen, which in the adult dog (weight about 30 lbs.) measured 33 cm. in length, 10 cm. in width, and 4.5 cm. in thickness; dark in colour, soft and friable and edges rounded. Lymphatic glands of the mesentery enlarged congested and haemorrhagic. Petechiae studded the serous membranes; the liver pale and friable. The pericardial sac contained from 20 to 75 c.c., and the peritoneal cavity of Case LIV 250 c.c.; the amount in the thoracic cavity was not greatly increased.

6. RABBIT. Only one animal was inoculated. The disease in this was of chronic nature, and organisms were rarely seen in an ordinary cover-glass examination.

CASE No. XXXVII.—August 7th, inoculated intraperitoneally with 1.0 c.c. citrated blood of XXV. During the four weeks during which it was taken, the temperature showed no gross variations. Organisms appeared on the tenth day, one to a cover-glass, and on the following day five to a cover-glass were seen. They were again detected on the fourteenth, fifteenth and thirty-second day after inoculation; from this time they were not seen on the daily examination up to September 30th, nor on those made about every fifth day since. During the first month of disease the rabbit lost condition; hair began to come out at the base of the ears, around the eyes, and on the rump, and sores formed on both tarsi. No oedema was noted, nor did any opacity of the cornea or signs of paralysis occur. During the next three months the rabbit appeared to improve in condition, and the tarsal ulcers dried up. Towards the last fortnight a slightly purulent discharge collected on the nostrils and around the eyes. Death took place in a very emaciated state on January 21st, 1908, 168 days after inoculation.

*Post-mortem.*—The spleen was considerably enlarged and rounded, but firm in consistency, the mesenteric lymphatics swollen but pale. Trypanosomes could not be found on direct examination; no sub-inoculations were made.

7. GUINEA-PIGS. Five guinea-pigs were inoculated with this trypanosome. In all, the disease was of a rapidly fatal character, the

average duration after appearance of organisms being only twelve days. The incubation period in three cases was nine days; in the other two, sixteen and nineteen respectively. Organisms were constantly present after detection, and were numerous up to death, which occurred ten to fourteen days later.

The temperature in the one animal taken varied little from normal until the trypanosomes appeared, when it rose and continued slightly elevated to death. The loss in condition was rapid, and the visible mucous membranes became very pale. Conjunctival discharge was not a constant feature, and in no case was any indication of paralysis noted.

On *post-mortem*, the spleen was much swollen, measuring from 5.5 to 7 cm. by 3.5 to 4 cm., congested, rounded, soft and friable, with an average weight of 14 grammes. In one case rupture of the capsule had accelerated death. The lymphatic glands were enlarged, oedematous and congested.

9. WHITE RATS. Five of these animals were inoculated. The period of incubation varied between six and ten days, and trypanosomes remained constantly present till death, which occurred in from 18 to 29 days after inoculation.

On autopsy, the spleen was enlarged, congested, soft and rounded, and the minute mesenteric lymphatic glands were distinct and haemorrhagic.

#### INFECTION WITH *T. VIVAX*

At the examination of the first herd on June 23rd, we noticed in one bull the presence of an organism, whose extraordinarily rapid passage from edge to edge of the cover-glass, and the transient corpuscular displacement produced, caused us to consider it as a spirochaete. When our camp was established this animal was brought under daily observation, and stained films, and later, cover-glass preparations some hours old, showed the organism to be a trypanosome. In the same herd one other animal was seen to be similarly affected, and two in which *T. dimorphon* had been found showed an occasional parasite whose rapidity of motion approximated to that of the bull. Three days before leaving Broken Hill we examined 14 cattle not previously inspected, one of which showed the same organism.

(1) Natural infection.—It would appear that stock-owners do not recognise more than one form of 'fly disease,' and that the animals infected by this trypanosome are considered as cases of ordinary fly disease. Of these five animals, that first seen was kept under observation for ten days at the original camp, and was later purchased and brought to that at which our experimental work was conducted.

Both animals which showed a mixed infection died within three weeks of diagnosis. The one detected just before leaving Broken Hill was alive, but considerably thinner six weeks later; whilst the fifth was reported to have died in August, seven weeks after parasites were seen, at which time it was in good working condition.

CASE NO. VIII.—Bull, aged about five years. (vide chart vii.) No very reliable history of this case is available, but it appears that this animal, with the rest of the herd, had been grazed away during the rainy season of 1906-1907, and had passed through fly areas on the way home in February, and that since that date it had gradually lost condition.

On examination, it was emaciated and hide-bound, signs of excessive lachrymation were present and the conjunctivae showed a few small petechiae, a condition seldom noted in animals infected with *T. dimorphon*. Prescapular and precaral glands enlarged; abdomen tucked-up; no oedema, but the bull presented all signs of trypanosomiasis, from which it was suspected of suffering by the owner. The case was under observation for 26 days between July 10th and August 15th.

A noticeable feature in the temperature is the great daily variation, as much as 5.4° F. being met with. By joining up the evening or morning registrations the 'curve' produced would not be great; there is no suggestion of a paroxysm, and the mean of morning and evening temperatures would not vary much from normal.

Trypanosomes are always scanty, from one to ten to a cover-glass was the usual number, only exceeded on four occasions, when approximately one in ten fields was seen. The great rapidity of the organism rendered any degree of accuracy in counting impossible, for their presence was generally only ascertainable by the slight displacement of corpuscles which accompanied their hurried passage across the field, a passage it was impossible to follow by any ordinary movement of the slide. With such an organism it is very probable that the one or two organisms present in a preparation would not come within the field of vision. On two occasions when they were not seen, centrifugalisation showed them to be present, and in one case a trypanosome was found in a thick-stained film. We would consequently suggest that the trypanosomes were almost constantly present in the peripheral blood and that with this parasite there is no great paroxysmal increase, and their detection is to a more considerable extent a matter of chance than is usual in the other forms of Trypanosomiasis.

During the 25 days of observation, parasites were seen on 16 occasions, eleven were marked +, one ++, and four +++, three of these latter occurring in the last week of life, and the other after the animal had been driven 22 miles in two days to the camp. Two days previous to death extreme weakness was manifest, the patient falling down several times and showing great difficulty in rising.

*Post-mortem* commenced 15 minutes after death. Rigor mortis distinct, mucous membranes pale, and a few petechiae on one conjunctiva. Skin closely



adherent, subcutaneous tissues firm and dry. Muscles pale but firm. *Thorax*. Only a few c.c. of a straw coloured fluid present. Lungs normal, save for slight emphysema in both apices. Heart normal in size; no fat in auricular-ventricular furrow; muscular tissue pale and streaked with fatty degeneration. Blood dark with half-formed clots in each chamber. *Abdomen*. Approximately 50 c.c. faintly blood-tinged fluid. Serous membranes show a few petechial haemorrhages on both parietal and visceral surfaces. Fat practically absent. *Liver* pale, slightly fatty and friable. Gall-bladder distended, bile of normal colour and viscid. *Spleen*, not enlarged. Capsule firm, somewhat fibrous and studded with small haemorrhages. Malpighian bodies pale, the rest of the splenic pulp pale, lying within the unduly defined trabeculae. *Kidneys* pale, capsule strips readily. Other organs normal. *F. papillosa* and *P. conicum* present.

(2) The experimental disease.—The organism for this work came from the bull No. VIII, whose history has just been given, and from several animals inoculated from it.

### I. CATTLE.

CASE No. IX.—Calf aged nine months. (chart viii.) July 11th, 1907, inoculated subcutaneously with 10.0 c.c. blood direct from VIII. This animal was under observation for ten days after inoculation, during which time the temperature remained about normal and parasites were not seen. Daily examination was recommenced on August 2nd, when it arrived at the new camp.

Trypanosomes were not seen on arrival, but appeared the following day, and as with case VIII were intermittently present until death, which occurred 61 days after inoculation. During the 38 days following detection of organisms, these were marked + on seventeen, ++ on four, and +++ on four, and on thirteen occasions they were not seen. Two days before death, when the animal was very weak, they were extremely numerous, almost swarming across the field, but still retaining their great motility.

The temperature chart is of the same nature as that of the bull; daily variations of 3° to 5° F., without paroxysmal tendencies except during the first week after arrival at the new camp, which might possibly be that accompanying the initial influx of organisms.

The loss in condition was gradual, but not excessive, and to the end the animal presented a bright appearance despite a progressive weakness, marked during the last few days. There was but little lachrymation, but petechiae on the conjunctival membranes were observed on several occasions.

Prescapular and precrural glands were enlarged at the time of inoculation, and did not show any perceptible increase in size.

The animal was accidentally killed by a leopard.

2. DONKEY. One animal only was inoculated, and during the twenty-four days of observation organisms were not seen.

CASE No. I.VIII.—September 6th, inoculated subcutaneously with 20 c.c. citrated blood of IX. The temperature rose to 103.2° on the evening of the same day and the animal showed slight abdominal distension and pain. It was again 102° on the evening of the sixth day, and the donkey was somewhat dull with a watery discharge from the eyes. It appeared well the following day, and showed no further symptoms up to September 30th. During this time no trypanosomes were seen, nor did they appear present on November 20th, when the animal still retained a healthy look.



3. SHEEP. Four sheep were inoculated with this trypanosome, and in three the organism reappeared. Only one, a small weakly animal, died during the period they were under observation; and from an examination of the temperature charts and of the animals themselves, it would seem that recovery may occur naturally, though, of course, we were unable to prove this in the time. The inoculation period in two cases was seven days. Trypanosomes were always scanty, rarely exceeding four to a cover-glass, their activity being as great as in cattle. This fact may account for the irregularity of their detection and the apparent lack of relationship between their occurrence and a thermal rise. The picture presented by the temperature chart differs absolutely from that following inoculation with *T. dimorphon*. The temperature rose between the fourth and seventh days, and fell again a few days later, and remained irregular for two or three weeks. There was no periodic exacerbation of a tertian or quartan type.

CASE No. X.—A small 'weedy' animal of five months old. July 11th, inoculated subcutaneously with 5.0 c.c. blood direct from Case No. VIII. The temperature rose on the fourth day and continued elevated for the ten days it was under observation at the first camp. Mr. Johnson, on whose farm the second camp was established, kindly visited the animal nine days later and made films in which trypanosomes were detected. It was then weak, and the native in charge said it had been sick for three days. It was carried in, 22 miles, to our new camp and died almost immediately on arrival.

*Post-mortem*.—Trypanosomes were not found in the blood or gland juice. Visible mucous membranes pale, those of the conjunctivae showing a few petechiae. The subcutaneous lymphatic glands were enlarged, those of the mesentery being also hæmorrhagic. There was no excess of fluid in the body cavities. Small petechiae studded the pleurae and pericardium. The spleen was not enlarged, its capsule was firm and showed a few petechiae. The liver and kidneys appeared normal. Neither intestinal parasites, nor *D. hepaticum* were found to account for the debilitated state at the time of inoculation. Two specimens of *Cysticercus tenuicollis* were present in the mesentery.

CASE No. XXXIII.—A healthy sheep aged one year. August 7th, inoculated subcutaneously with 1.0 c.c. citrated blood of IX. The temperature rose to 103.2° on the morning of the sixth day and organisms were seen the next morning. During the succeeding six weeks the temperature was four times above 105°, and trypanosomes were seen on eleven occasions. After September 2nd the temperature remained about normal and parasites were not observed. On September 17th it was inoculated with *T. dimorphon*. (*vide antea* chart ix.)

CASE No. XXXIX.—A healthy sheep aged one year. August 9th, inoculated under the skin with 1.0 c.c. blood direct from goat, Case XI. Trypanosomes appeared on the seventh day, and for the next three weeks were fairly constantly present, but during the last three and a half weeks of observation they were only seen seven times. The temperature rose on the sixth day, and during the ensuing forty days of disease was similar to that of bovine piroplasmiasis in India.

CASE No. LIX.—A companion sheep to the last September 5th, inoculated subcutaneously with 1.5 c.c. citrated blood from IX (parasites present). The temperature remained normal and trypanosomes were not seen during the twenty-four days it continued under observation.

4. GOATS. Three goats were inoculated, one becoming infected. Parasites were intermittently present in scanty numbers in the one positive case.

CASE No. XI.—A goat aged one year, in poor condition and coming from a herd infected with scabies. July 11th, inoculated subcutaneously with 10 c.c. blood from Case VIII. Organisms were not seen during the ensuing ten days. It arrived at the new camp on August 2nd, and trypanosomes were first seen on the 5th of that month. They continued intermittently present in scanty numbers for the six weeks of life remaining.

The temperature, as in sheep, showed no tendency to reproduce the malarial type, but assumed rather that seen in the two infected cattle.

This animal became badly infected with a form of dermatitis, which spread to the lips and around the eyes and ears, causing considerable irritation and probably hastened death, which occurred on September 13th, the sixty-fourth day after inoculation.

*Post-mortem.*—Trypanosomes were not seen in blood or gland juice. Mucous membranes pale, a few petechiae on the conjunctivae. A few small circumscribed ulcers were present on the gums and hard-palate. No fluid in the thoracic and abdominal cavities. Heart pale and flabby, lungs normal. Liver pale but of firm consistence, capsule adherent to the diaphragm, gall bladder distended. In the large biliary canals several tape-worms were found. Spleen showed no gross alterations. Mesenteric glands pale and not greatly enlarged. Small concretions resembling those formed by *Gesophagostomum columbianum* occurred in the large intestine. (vide chart x.)

CASE No. XXXII. Healthy goat aged four months. August 7th, inoculated subcutaneously with 1.0 c.c. citrated blood of Case IX. The temperature was irregular, but trypanosomes were not seen, and it was alive and in good condition on November 20th. Sheep, Case XXXIII, inoculated at the same time became infected.

CASE No. XL. August 9th, inoculated with 1.0 c.c. blood direct from goat. Case II. The temperature showed a daily variation somewhat greater than usual, but trypanosomes were never seen. On September 17th it was inoculated with *T. dimorphon* and became infected on the twelfth day. Sheep, Case No. XXXIX, inoculated on August 9th from the same Case XI became infected.

5. MONKEY. An adult female, probably *Cercopithecus pygerythrus*, was inoculated subcutaneously on September 6th with 1.0 c.c. of blood from Case IX. It remained active and in perfect health until October 1st, when it died suddenly through the maltreatment of the boy in charge.

6. GUINEA-PIGS. Five guinea-pigs were inoculated from, respectively, Cases IX, XI, XXXIII, XXXIX and X (*post-mortem*). They showed no disturbance in health, the temperature remained normal and organisms have never appeared. They are all still alive,

February 18th, 1908, despite the rough usage to which they have been subjected on the march.

7. DOGS. Three 'kaffir' dogs were inoculated from Cases IX and XXXIII. The temperature remained normal, and organisms were never seen. One appeared to lose condition during the two months following inoculation, but later regained it, and with the others is now alive.

8. RABBIT. A rabbit was inoculated on August 3rd with 1.0 c.c. of blood from Case IX, when parasites were present. Organisms were never seen, and it continued in perfect health until November 14th, when it suddenly died. On *post-mortem* no signs of trypanosomiasis could be found.

9. WHITE RATS. Five white rats were inoculated simultaneously with the five guinea-pigs. None have shown any disturbance in health, and four are still alive. The fifth was inoculated in November with a dog trypanosome, and died.

#### INFECTION WITH *T. THEILERI*

During the routine blood examinations of cattle Nos. XVI, XXI and XXII, a large trypanosome was seen on one occasion in each. Further examinations in fresh and centrifuged specimens were negative. In Case XVI this trypanosome was seen three days after *T. dimorphon* had been expelled by means of atoxyl, and four days before its reappearance.

CASE No. XXIII. A bull naturally infected with *T. dimorphon*. August 2nd. Inoculated subcutaneously with 90.0 c.c. citrated blood of Case XXII, which showed *T. theileri* that morning, *T. dimorphon* also present. The animal died of *T. dimorphon* infection three weeks later without ever showing *T. theileri* again.

#### TRYPANOSOMIASIS IN SHEEP (*T. DIMORPHON*)

A European who visited the camp informed us of mortality amongst his sheep, which were kept in a camp on the Lukanda river, 45 miles North of Broken Hill. No specific details were obtainable, but it appears that between 30 and 40 had been sick and had died or been destroyed. *Gl. morsitans* exists all round, but infection by *D. hepaticum* was suspected. A visit was paid to this camp on November 23rd, during the owner's absence, and three sheep found alive. One was *in extremis*, and blood examination revealed the presence of *T. dimorphon*. On *post-mortem*, there was an excess of fluid in all cavities, the lymphatic glands much swollen and oedematous but pale, the spleen was enlarged, soft and rounded.

## NATURAL TRYPANOSOMIASIS IN DOGS

We saw three dogs which were suspected by their owners of having naturally acquired trypanosomiasis. All had, at some recent date, been in the fly country around N'dola, 15° S., 24° 40' E., and on examination showed trypanosomes and the clinical indications of the disease—a haggard appearance, anaemia, loss in condition, and in two, corneal opacity.

Sub-inoculations were made from two of these dogs.

From Dog 'A.'

## 1. GUINEA PIG.

CASE No. LXXII.—Nov. 8th, 1907, inoculated intraperitoneally with 1 c.cm. citrated blood. Parasites first seen on Nov. 20th and at each succeeding examination, about every fourth or fifth day, up till death on January 18th, 1908. Paralysis of the hind legs was noted on December 25th, but the animal recovered in three days. The same symptom reappeared on January 16th, and continued up till death two days later. Duration of disease 72 days.

## 2. RAT.

CASE No. LXXIII.—Same date. Inoculated subcutaneously with 1.0 c.cm. citrated blood. Organisms appeared on November 18th (tenth day) and were present at each examination up to January 10th, when the animal died. Duration of disease 64 days.

From Dog 'B.'

## 1. GUINEA PIG.

CASE No. LXXVII.—November 15th, 1907, inoculated intraperitoneally with five drops of blood in citrate solution. Organisms appeared between November 22nd and 26th, on which later date they were present in fair numbers. On one examination, made December 12th, they were not found but were present on all other occasions. Rat died January 12th. Duration 68 days.

## 2. RAT.

CASE No. LXXVI.—Same date. Inoculated intraperitoneally with five drops of citrated blood. Parasites appeared between the 26th and 29th of November, and were constantly present on all succeeding examinations up to January 3rd, when it died—49 days. This rat had previously been inoculated with blood containing *T. vivax*, but had not shown infection.

## I. MORPHOLOGY OF CATTLE TRYPANOSOMES

*T. DIMORPHON*, Dutton and Todd

The parasite commonly encountered in the naturally infected and the experimentally inoculated cattle corresponds to the short forms of *T. dimorphon* described by Dutton and Todd as 'tadpole' and 'stumpy.' The former predominated in all animals, except during

the last few days of life in a few cases, when the stumpy type was more frequently seen.

If the distinction between 'stumpy' and 'long' is based upon the possession, by the latter, of a definite flagellum, this was not encountered in cattle. Its appearance, however, in sub-inoculations indicates that the parasite in question belongs to the *dimorphon* group. 'Tadpole' forms were found in every animal, with the exception of one moribund case mentioned later. They measured from 9.75 to 15.3  $\mu$  in length, and up to 1.5  $\mu$  in width. The body protoplasm stains a rather deep blue with the Giemsa stain, granules rarely being present; blepharoplast terminal or sub-terminal, small and rounded; nucleus a short oval, 1.7 to 2.5  $\mu$  in length, staining homogeneously a rather dark purple. Undulating membrane very rudimentary; in some, a small, fin-like, single fold could be seen. Flagellum absent, but an anterior prolongation of the protoplasm occasionally supported a minute extension of the rim of the undulating membrane. The posterior extremity is usually bluntly angular, but every gradation was met with. The greatest width lies posterior to the nucleus. Forms slightly larger than the above, which may be classed as 'stumpy,' were occasionally seen in some animals, particularly in those in which the disease was running a more acute course, and have been constantly observed, but they become relatively more numerous towards death. In one animal, which had not shown organisms on two examinations previously, only this form was found at a third examination when the beast was moribund. It was not constantly to be observed at death; in five animals none but what are termed 'tadpole' forms were seen. This form measured from 17.75 to 21.25  $\mu$ , and is of a stouter build than the 'tadpole.' The protoplasm assumes a pinker tint, the blepharoplast is better defined, the nucleus almost invariably distinctly rounded and staining less deeply, and the widest part of the body is more usually on this level. An undulating membrane could be more easily distinguished, in some rare cases two or three folds being visible, and the bordering rim is commonly produced with the anterior extremity of the body to form an abruptly square-cut bristle-like beak, not exceeding 2  $\mu$  in length, but prominent in a well-stained preparation. Granules were seen in some, and divisional forms of this type were encountered. The distinction between this form and what is described as 'tadpole' is



more marked than between it and the 'long' form. These latter, that is to say, a trypanosome possessing a flagellum of more than a few  $\mu$  in length, were not seen in cattle, and were only found in inoculated guinea-pigs and rats. In these animals they measured from 25 to 31  $\mu$  in length, and could, on the whole, be clearly separated into the two arbitrary classes, 'male' and 'female,' the former being thinner, staining more homogeneously pink, the nucleus elongated, and the protoplasm free from granules; the latter stouter throughout the whole length, the nucleus oval, and the protoplasm taking a blue tint and often containing granules. The flagellum measured from 5 to 11  $\mu$ , and continued from an undulating membrane which was not always so evident as in the larger of the 'stumpy' forms. The blepharoplast is fairly prominent, rounded or oval, and commonly situated some little distance from the posterior extremity, which itself assumes most frequently a rather finely tapering point. The width of these forms varied between 1.5 and 2.5  $\mu$ , dividing forms being as much as 3.5  $\mu$  across. While we refrain from the use of the expression 'free flagellum,' there appears little doubt that in the larger of the 'long' forms, such a structure, free of cytoplasm, does exist. In the large 'stumpy' and the short 'long,' which cannot be clearly separated, the cytoplasm is unquestionably continued for a certain distance anterior to what we would regard as the normal extremity of the body, the place where body, undulating membrane and flagellum meet, and it is often impossible to determine the point at which this prolongation ceases.

In fresh cover-glass preparations the corpuscular displacement is local and the organism does not readily pass out of the field of the microscope. The smaller forms cause, naturally, less commotion amongst the corpuscles. They are more readily retarded by these bodies, and the slow rolling displacement they produce is markedly different from the furious lashing of those with a long flagellum and the resulting scattering of the corpuscles. Neither form was ever noted to produce the peculiar 'catherine-wheel' effect seen in blood containing *T. vivax*.

We were unable to make permanent slides from our donkey during the visit of November 20th, but Dr. Yale Massey kindly allowed us to examine the slides he had made on the day previous to the death of a naturally-infected donkey at Ruwe, in the Congo to the North of the

Rhodesian district we have under review. Short and long forms were present in this animal, the classification of 200 being 'tadpole' 10 per cent., 'stumpy' 70 per cent., and 'long' forms 20 per cent. The first-named measured between  $10.5$  and  $14.75\mu$ , the 'stumpy' from  $15$  to  $18.25\mu$ , and the 'long' forms between  $18$  and  $27.25\mu$ . In some of these latter, a flagellum measuring from  $3$  to  $5\mu$ , which appeared quite free of cytoplasm, was seen. The structure of these several forms was essentially the same as that of those already given.

In sheep and goats only the 'tadpole' forms were seen. These measured from  $9.75$  to  $14.5\mu$  in length and from  $1$  to  $1.75\mu$  in width. In no particular could they be held to differ from those in bovine blood, and in the films examined only one form was seen which might possibly be regarded as 'stumpy.' It has been noted that none of these animals died whilst under observation, and this may account for the non-detection of any but 'tadpole' forms. It is also to be remembered that in sheep, Case No. XXXIII, and goat, Case No. XL, the organism which appeared was of the 'tadpole' variety, whereas the guinea-pig from which they were inoculated had shown 'long' forms, and 'stumpy' ones were present at the time of inoculation. In none of our inoculated dogs were forms longer than  $16\mu$  seen. Most were of the 'tadpole' variety, but forms corresponding to 'stumpy' were encountered towards the end of the disease. We reserve the description of the organisms encountered in the naturally infected animals till later.

In rabbits, in the films made from our single case, only 'tadpole' forms were seen.

Guinea-pigs are the most satisfactory animals for revealing the dimorphic variations of this trypanosome. It has been noted that the average period of duration of organisms was ten days. During the first four days, 'tadpole' forms are almost exclusively present, giving place between the fifth and eighth days to 'stumpy' and 'long'; whilst at death, and on the two or three days previous, 'stumpy' forms predominate.

The 'tadpole' and 'stumpy' forms correspond to the descriptions given, while that of the 'long' form is based on its appearance in these animals. We do not hesitate to say that in many of these, particularly the so-called male forms, a flagellum whose length sometimes exceeded  $10\mu$  was seen free from cytoplasm. These forms

were present, though less numerous than the 'stumpy,' in blood inoculated into the goat, Case No. XXXIII, sheep XI., and rat LXXI, in which the 'tadpole' forms were reproduced.

Rats were infected by blood containing all forms. 'Tadpole' forms predominated throughout the first few days of the disease, while the 'stumpy' became more numerous towards death. 'Long' forms were very rare, but were seen in those inoculated from an ox, a goat, and a guinea-pig; but they were not seen after inoculation from a dog or a second goat infection. It is to be remarked, however, that a thorough examination of all slides has not been possible.

We have here, then, a trypanosome whose prevailing type in naturally and experimentally infected animals is short, measuring only  $9.75$  to  $15.3\mu$ , and from which there is a relative absence of a flagellum. This type under the influence of a different host, or under natural conditions in the same animal, assumes a distinct form which measures from  $25$  to  $31\mu$  in length, and possesses a flagellum which may be upwards of  $10\mu$  long. With the knowledge at present available there can be no hesitation in ascribing the name *Trypanosoma dimorphou*, Dutton and Todd, 1904, to such a dimorphic organism.

## II. *T. VIVAX*, Ziemann

We have already remarked that the movement of this organism in fresh cover-glass preparation is of extraordinary rapidity, and the effect produced by the passage across any one field closely resembles that of a spirochaete, the corpuscular displacement being transitory and of no greater magnitude than that produced by drawing a floating hair across the surface of still water. It is impossible to retain any one organism in the field by movement of the slide, even when the mechanical stage is not employed, the parasite crossing and altering its direction with bewildering swiftness. In a preparation which has been kept for an hour or two the movements of some become more sluggish, and in fields where corpuscles are scanty, progression is seen to be due to a rotatory motion of the whole body, to the exclusion of a wavy undulating membrane and the vibrations or lashings of a free flagellum.

When one of these parasites is obstructed by corpuscles or fibrin threads, the action again differs from that of other trypanosomes. Sometimes the ordinary lashings are produced, but more frequently the posterior end becoming a fixed point, the anterior circles round with regular sweeps, forcing the corpuscles away as the arc-like radiations pass from a burning catherine-wheel. The trypanosomes measure from 20 to 26 $\mu$  in length, and up to 3.4 $\mu$  at the widest part, posterior to the nucleus. The body tapers anteriorly from this in a rather regular fashion, while the posterior end is distinctly rounded. The blepharoplast is large, round or oval, measuring up to 1 $\mu$  in diameter, and is usually terminal. The nucleus is commonly an elongate oval, up to 3.75 $\mu$  in length and 2.5 $\mu$  in breadth (average 3.4 by 2), and occupies nearly the whole transverse diameter at this level. The undulating membrane is very narrow, about 1 $\mu$ , the bordering rim arises from the neighbourhood of the blepharoplast and is continued as the flagellum after running parallel to the body, so giving an aspect of stiffness to the whole structure. For the greater part of its length this 'free' flagellum is accompanied by a continuation of the periplast, which may or may not contain cytoplasm. The actual free extremity does not appear more than 3.5 $\mu$  in length, commonly less; whilst the total length of the tapering end anterior to the cessation of the membrane is upwards of 8.5 $\mu$ . The length of this whip was somewhat greater in the later stages of the disease, thus our figures in the inoculated calf, case IX, on August 4th (twenty-fourth day after inoculation) give an average of 3.4 $\mu$ ; those on September 8th (fifty-eighth day) vary between 3.4 and 8.5, with an average of 5.9 $\mu$ .

The cytoplasm stains homogeneously; vacuoles are seldom seen and granules are not very common. Dividing forms are rare; in the peripheral blood we have only seen those with a double blepharoplast. We have never noted the alveolar arrangement of the cytoplasm described and figured by Lühe.<sup>5</sup> This trypanosome has retained its quite characteristic movement in fresh preparations and the same appearance in stained films, excepting only the slight differences in the length of the anterior extremity, in all animals which took the experimental infection.

### III. THE TRYPANOSOMES OF NATURALLY-INFECTED DOGS

The forms seen in all three animals are the same. The trypanosomes measure from  $20\cdot25$  to  $28\cdot9\mu$  in length, and from  $1\cdot5$  to  $2\cdot5\mu$  in width. The protoplasm stains pink with Giemsa, vacuoles and granules were inconstant; the posterior extremity is most commonly pointed. The nucleus is situated towards the anterior part of the body, and varies in shape from a round to an elongate oval, the former taking a deeper stain than the latter. The blepharoplast is rounded, usually terminal, but may be removed from the posterior extremity by as much as  $2\cdot5\mu$ . A well-developed undulating membrane is present, and the rim is continued as a flagellum, which varies from 3 to  $10\mu$  in length, and is accompanied, in the short forms at least, by a prolongation of the cytoplasm. Those carrying a short flagellum correspond in type to the so-called 'female' forms, and the long flagellar forms to the 'male' of other trypanosomes. It would be impossible to assert that these 'females' are not the 'stumpy' forms of *T. dimorphon*, which are somewhat larger than normal. Trypanosomes which could in any way be considered as having any relation to 'tadpoles' were not encountered; the smallest measurement made was  $20\cdot25\mu$ . In the two guinea-pigs, the trypanosomes which reappeared measured from 21 to  $30\mu$ ; in the smaller forms the projecting flagellum is short. The variations in length noted depend mainly upon the size of this flagellum; the body itself being fairly constant at 18 to  $20\mu$ . Tadpole-like forms were not seen at any stage of the infection.

In rats no forms except 'long' ones were met with. These measured from 21 to  $30\mu$ , and from  $1\cdot5$  to  $2\mu$  in width. There is a distinct flagellar appendage in all, which differ from those seen in guinea-pigs only in being more granular and staining less deeply.

The natural dog trypanosomes are monomorphic in so far as no variations comparable to those existing between 'tadpole' and 'long' forms were seen. Being monomorphic and of large size, a parasite of a domestic animal and occurring in Africa, this trypanosome must be placed in that heterogeneous collection whose type is *T. brucei*.



## ARGUMENT

It must be admitted that our present knowledge of trypanosomes does not permit of any satisfactory classification. In Africa the confusion in nomenclature is appalling, and the number of specific or suggested names, based largely upon the country of origin or the first found host rather than upon morphological or biological characteristics, renders absolute diagnosis of an individual form almost impossible without a typical living strain for comparison.

There is perhaps no great difficulty in asserting that the body structure of certain trypanosomes shows dimorphic variations under certain conditions. In all trypanosomes morphological differences between individuals occur to a greater or lesser extent, but only in the one species, *T. dimorphon*, Dutton and Todd, are they considerable. In this, unless the 'tadpole' and the 'long' be seen, they are not striking; and between the larger of the 'stumpy' and the shorter of the 'long' we do not consider the variations greater than between some of the so-called 'male' and 'female,' or the smaller or the larger forms of *T. evansi*. An additional difficulty is that there is no one structural point that can be seized upon as a basis for classification; most gradations can be met with in the shape of the posterior extremity and the length of the anterior and the amount of the flagellum that is free. As we have found forms recognisable as 'tadpoles' and 'long' both under natural conditions and those of ordinary animal experimentation, we have no hesitation in asserting that the cattle trypanosome first described is dimorphic according to the interpretation placed upon that word by workers on trypanosomiasis.

The first dimorphic trypanosome was that described in the Gambia by Dutton and Todd,<sup>1</sup> who at the same time described the pathological reactions produced by it there, which reactions were later confirmed in Europe by Thomas and Breinl,<sup>2</sup> and by Laveran and Mesnil.<sup>3</sup> They showed that rats, guinea-pigs and, with one exception, dogs are susceptible, and die within one or two months. The trypanosome encountered in the cattle of the Congo Free State was also dimorphic in type, but the animal reactions in that country differ from those obtained in the Gambia. At one post, Romee, laboratory animals were shown to be highly susceptible; at the other

posts twenty-nine inoculations were made from cattle whose blood showed trypanosomes of this type. One rat showed organisms twice, after the original and after a re-inoculation, but for one day only in each case; a second rat only became infected after several months' incubation. Dutton, Todd and Kinghorn<sup>4</sup> consider this Congo trypanosome as *T. dimorphon* on the grounds that the morphological characteristics of this species are peculiar in the genus *Trypanosoma*, and identical with those of the form they describe; and that the variation in virulence is not sufficient proof that more than one species of trypanosomes was present. The trypanosome which we describe shows animal reactions approaching those of the Gambian strain; all our dogs, rats and guinea-pigs have succumbed to an acute infection within two months, and, as in Gambia, the rabbit has a more chronic disease and the cattle an acute form. With similar morphological appearances and similar animal reactions, we consider the Rhodesian form to be *T. dimorphon*, Dutton and Todd, 1904.

We have much more hesitation in ascribing the second of the Rhodesian cattle trypanosomes to a specific class. The morphological characteristics seen in animals which were susceptible are not sufficient to consider it dimorphic; 'tadpole' forms, or any approach to these, were not encountered; the variations seen were not greater than have been found in monomorphic trypanosomes, and to this group it is assigned. We distinguish it from *T. dimorphon* which occurred in cattle of the same herds and also in some of the same animals by:—

1. The morphological appearances; the fairly constant size and the extreme rapidity.
2. The clinical type of disease induced; the great daily variation of temperature as opposed to the 'curve' in cattle suffering from *T. dimorphon*, and the absence of the striking febrile reactions seen in sheep and goats.
3. The animal reactions: Five rats, five guinea-pigs and three dogs were inoculated without effect. One goat and three sheep did not become infected, and in those which did there were indications of recovery. In one goat and one sheep which had been inoculated with this trypanosome, and were not showing parasites, a re-inoculation with

*T. dimorphon* brought about a febrile reaction and the appearance of this organism in the blood.

4. The *post-mortem* appearances: Splenic enlargement was not noted in the three cases upon which autopsies were possible, whereas in *T. dimorphon* infection it was a common feature.

Sander and Hennig<sup>6</sup> state that according to Ziemann *T. vivax* occurs spontaneously in cattle, sheep and goats; the incubation period is 5-8 days; and on *post-mortem*, enlargement of the liver or spleen is seen. Experimentally, grey rats died in 8-11 days; in donkeys the disease is chronic, and in a German dog the reaction was apparently doubtful. A negative result was obtained in a white rat. In Rhodesia white rats and native dogs were negative; the one donkey inoculated did not show organisms, but it has not been examined or reported on for three months and we are ignorant of its present state. A point of difference we note is the enlargement of the spleen, but our observations are based on three *post-mortems* only.

The morphology, as given by Lühe, differs from that of the form under discussion in possessing an alveolar protoplasm and a somewhat pointed end. These are minor points, negligible so far as the morphology of other trypanosomes is concerned, in which similar variations are commonly seen. The experimental work on both sides is limited, but the animals used are similar, and if Ziemann's dog was negative, and such is a possible interpretation, the results coincide, though we were unable to use grey rats and pigs in which Ziemann obtained positive reactions.

We know of no other trypanosome whose activity approaches that of this form. On morphological grounds, then, and animal reactions this trypanosome coincides more closely to that of Ziemann than to any other, and with all reserve, we feel justified in considering the second parasite of Rhodesian cattle as sufficiently closely allied to *T. vivax*, Ziemann, to bear that name until the classification of the genus be put on a more satisfactory basis.

## TRANSMISSION OF CATTLE TRYPANOSOMES

European and native unite in incriminating the tsetse-fly common throughout the Northern part of the area of North-Western Rhodesia. The examinations of our specimens of these has so far only shown *Glossina morsitans*, the approximate distribution of which is marked on the attached map; but it must be understood that the lack of signs in certain areas does not imply freedom from the fly, but only that we have no positive knowledge of its occurrence.

*Tabanidae* have only been incriminated in the one instance already recorded. During June, July and August, 1907, we did not see any, but during the latter end of September they were occasionally seen at our camp, and they were very numerous in Broken Hill on the first few days of October. On our line of march they were constantly encountered, and may be held as having an almost universal distribution. The larger members of this family are locally known as 'hippo flies'; *Haematopota*, also common in November and December, are usually spoken of as 'blind flies.'

*Stomoxys* were taken in the cattle kraal of the farm where our camp was established in July and August, and they were caught on the River Kafue in November and on the River Luapula in December. They were most frequently met with in villages, but on two occasions were taken from recently shot game.

*Lyperosia* were caught in the same cattle kraal in July and the first week in August, and again during the latter part of September. They were not seen in the interval, nor have they been taken anywhere on the route followed.

*Hippoboscidae* are, in comparison with India, rare. One specimen of *H. rufipes* (?) was shown us as coming from near the Kafue. We have thrice taken *Lipoptena* on dead buck.

Owing to the relative frequency of *Glossina* and their association with game and domestic animals, there can be no question that, assuming game to be the pre-existing reservoir, they are the most capable flies for transmitting the trypanosome to cattle or other domestic animals taken into their haunts. We may mention that we have seen trypanosomes in fresh preparations of blood made from a recently shot Hartebeest (*Bubalis lichstensteini*) and a bush buck (*Tragelaphus scriptus*), but our examination of the slides and inoculated animals is incomplete.



Nearly all the cases of trypanosomiasis in cattle examined could be given a history, often very imperfect, of having at some recent date been exposed to the tsetse; but in one herd, where the history is reliable, the evidence is suggestive that *Stomoxys* and *Lyperosia* had acted as transmitting agents.

Forty-four animals from this herd had lived at 'Kapopo, which itself is free from fly, for upwards of three years, during which time they had always been in good health and deaths were rare. In August, 1906, they were brought to Broken Hill, a distance of about 97 miles, following as far as possible a route where tsetse were scanty or not known to exist, and adopting the usual precautions, such as marching at night. They remained in perfect health until June, 1907, when they came under observation. During these eleven months two animals had died, apparently of some acute inflammatory disease, and one had been destroyed as the result of an accident, and six others which had not been recently in tsetse areas were added. The farm where these cattle were kept is at least two and a half miles from the nearest known fly area, and they were all employed on this farm in agricultural work; the three cows and three bulls grazing close to the buildings. On April 18th, six bullocks were sent on a journey of thirty miles Southwards to Mwomboshi, and returned four days later. *G. morsitans* occurs on the road travelled about eight miles from the farm, but it is limited to a narrow patch. On June 25th, three of these animals and one which had not been away showed trypanosomes, and they were all dead within a month; one died within ten days, and this at the height of the dry season, when deaths are said not to take place. Four other animals were suspected, but did not then show organisms. The rest of the herd were all in good condition, looking bright and doing the hard agricultural work well. On July 18th, all remaining animals were examined, and eleven, some of them straight from the plough, showed trypanosomes. The owner, who possessed a small microscope, picked out another on the 22nd, one on the 27th, and we found two more on the 29th. Three of these were cows which had not been exposed to *Glossina* for twelve months at least.

All these animals were segregated, and those not showing organisms were placed by themselves in a kraal and grazing area which appeared free of all biting flies, such as *Stomoxys* and

*Lyperosia*. Further examination of these apparently healthy animals weeded out five more cases in August. The balance, fourteen, continued healthy, and of these thirteen were taken for transport work in September. They did the work well, and showed no greater death-rate on the 400-mile march than did the other ninety cattle taken.

After the first examination, at which only suspected cattle were presented, all animals inspected were, without exception, free of any clinical signs of the disease. These became manifest in about two weeks time, and death took place within an average of thirty days of diagnosis. This is the average between these dates in fourteen apparently healthy cattle, and excludes all those upon which experimental work on treatment could be held as influencing the course of the disease in either direction.

With a disease of such rapidity and virulence, we consider it highly improbable for the infection to have lain dormant in the animals since August, 1906. From an examination of all conditions, we think it probable that one or more of the six cattle which went to Mwomboshi in April contracted the disease on the road and brought it to the farm, where, in the presence of *Stomoxys* and *Lyperosia* in the kraals, these animals, including cows and bulls, which did not leave the place, became infected, and that the segregation from these flies checked its spread to the fourteen cattle which remained healthy. The one animal which did not go on trek with the thirteen cited above was brought to live with the sick on September 10th. It was then in excellent condition, but on a visit to the farm on November 20th it showed *T. dimorphon*, and would in all probability succumb within two weeks. Owing to the lack of facilities, and the pressure of other work, we were unable to conduct any transmission experiments with these two flies, but as affording corroborative evidence we examined the road on which the outbreak was suspected of having originated. We were informed by the owners concerned that they had lost animals during the present year after travelling through this area with their spans, and this despite the usual precautions. We, therefore, exposed two healthy bullocks to *Glossina morsitans* there.

CASES No. XLIV, XLV.—Two animals from small herd at Mwomboshi, where they had lived for between two and three years, and where stock apparently does well. August 19th, 1907. These two were driven between 10 a.m. and 1 p.m. over the road, through the area inhabited by *Gl. morsitans*. Only three flies were

seen to feed, two on No. XLV, and one on No. XLIV. On arrival at our camp they were segregated and carefully kept from all association with other cattle. The temperature remained normal until September 3rd, the fifteenth day after being bitten. On September 6th, the eighteenth day, trypanosomes were seen in the blood of both. This trypanosome was morphologically identical with that we have called *T. vivax*. On September 13th (XLV) and September 16th (XLIV) *T. dimorphon* appeared, and both organisms were present until September 30th, when No. XLV died. Both had been submitted to treatment by Atoxyl and Mercury, and No. XLIV has been detailed elsewhere. (vide chart ii.)

This double infection by such a small number of flies as were seen, and on a road where game is scarce, caused further enquiries to be made, and we found that a span of oxen had travelled that road on August 14th, five days prior to the date our animals passed. On September 21st we examined this span, and found two animals infected with *T. dimorphon*, and a third showing both *T. dimorphon* and *T. vivax*.

If the *Glossina morsitans* which bit Cases Nos. XLIV and XLV had not derived these organisms from game, it would appear that they have the power of transmitting five days after the infecting feed, which would have been taken from this span, whose owner reported that he had lost several animals during the months of June to September from what he regarded as 'fly' infection.

### CONCLUSIONS

1. That trypanosomiasis of domestic stock is very prevalent in the Northern area of North-Western Rhodesia, and that it is due to *T. dimorphon* (Dutton and Todd), *T. vivax* (Ziemann), and one morphologically allied to *T. brucei* (Plimmer and Bradford). *T. theileri* also occurs, but does not appear to cause serious damage.

2. That these trypanosomes may be transmitted by *Glossina morsitans*, *Stomoxys calcitrans*, and a species of *Lyperosia*. In nature it will depend upon the conditions under which cattle are maintained, to which of these genera special attention must be paid in prophylaxis.

3. That treatment, as detailed in a previous report, shows certain indications of success, and we urge facilities for a continuation of the work on this disease, which tends to stagnate the proper development of a wealthy mineral and agricultural country.

## REFERENCES

1. DUTTON AND TODD. The Liverpool School of Tropical Medicine, Memoir XI.
2. THOMAS H. WOLFERSTAN AND BREINL, A. The Liverpool School of Tropical Medicine, Memoir XVI.
3. LAVERAN AND MESNIL. Trypanosomes et Trypanosomiases, Paris, 1904.
4. DUTTON, TODD, AND KINGHORN. The Annals of Tropical Medicine and Parasitology. Vol. 1, No. 2, pp. 233-271.
5. LÜHE. Mense, Handbuch der Tropenkrankheiten, Leipzig 1906, Band III, I, p. 125.
6. SANDER AND HENNIG, idem, Band III, II, p. 721.